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Analytical Probing of Complex Systems

ABSTRACT BOOK

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PS2-16 Studying the entropic pulling of Hsp70/DnaK at the single-molecule level using a biological nanopore
Verena Rukes, *Mathieu E. Rebeaud, Paolo De Los Rios, Chan Cao*

PS2-17 DFT and dynamics simulation studies to understand probing of folic acid using β-cyclodextrin functionalized copper nanoclusters and vitamin B6 cofactor pyridoxal by displacement approach
Suban Sahoo

PS2-18 Direct and Selective Quantification of Cr(VI) in Waste Waters using Raffinose Capped Silver Nanoparticles as Sensitive Optical Sensor
Penka Vasileva, *Irina Karadjova*

PS2-19 Quality by Design Approach for a Multicomponent Quantification Using HPLC-PDA and HPLC-MS: Application to Dosage Form and Biological Body Fluids

***Naser Al-tannak**, Bashayer Al-Shatti, Abdullah Al Ali, Ahmed Hemdan*

PS2-20 Expanding the exposomics toolbox towards metals
***Simone Braeuer**, Max L. Feuerstein, Elisabeth Foels, Tina Buerki-Thurnherr, Raimund Widhalm, Claudia Gundacker, Benedikt Warth, Gunda Koellensperger*

PS2-21 3D stochastic microsensor based on graphene for the simultaneous determination of p53, HER-3, and HER-4
***Catalina Cioates Negut**, Hbil Raluca Ioana Stefan-van Staden, Ruxandra-Maria Ilie-Mihai, Maria Coros*

PS2-22 Stochastic sensors as new tools for the assay of CA72-4, CA19-9, CA12-5 and CEA in biological samples
***Ruxandra-Maria Ilie-Mihai**, Habil Raluca Ioana Stefan-van Staden, Alexandru Adrian Bratei, Damaris-Cristina Gheorghe*

PS2-23 Comparison of different sample preparation techniques for degradation products of nerve agents in biological fluids
***Engin Koçak**, Sermet Sezigen, Nurgül Bakırhan Karadas, S. Irem Kaya, Sibel A. Ozkan*

PS2-24 Enzyme-based platform immunoassay for the simultaneous quantification of drug and anti-drug antibodies.
***Frans Kokojka**, Sigal Pressman, Yehuda Chowers, Robert Marks*

PS2-26 Fast screening of biological and food samples using miniplatforms based on 3D stochastic microsensors
***Andreea-Roxana Niculae**, Raluca Ioana Stefan-van Staden*

PS2-27 Harnessing programmable zwitterionic coacervates as versatile sensing platforms
***Francesca Torrini**, Philippe Lenzen, Karl Normak, Carolina Paganini, Alessandro Gori, Marina Cretich, Paolo Arosio*

PS2-28 The development of a MIP-based electrochemical sensors for antiviral drug detection using different electroanalytical techniques
***Ahmet Cetinkaya**, Altay Unal, Hasan Nazır, Emin Çorman, Lokman Uzun, Sibel A. Ozkan*

PS2-29 Online biomass monitoring of Chlorella vulgaris cultures by dielectric spectroscopy
***Juan Limon Petersen**, Caspar Demuth, Lukas Neutsch, Manuel Maurer, Nicolas Pirolet*

PS2-30 Electrochemical classification of benzodiazepines: a comprehensive approach combining insights from voltammetry and liquid chromatography − mass spectrometry
***Jonas Schram**, Nick Sleegers, Filip Van Durme, Alexander van Nuijs, Karolien De Wael*

PS2-31 A label-free insight into the molecular aspects of electrochemical DNA sensors for mercury ion detection
***Anna Szymczyk**, Marcin Drozd, Marcin Olszewski, Robert Ziolkowski, Elzbieta Malinowska*

PS2-32 Paper-based Device for Point-of-care Nucleic Acid Quantification Combining CRISPR/Cas System and Personal Glucose Meter
***Yohei Tanifuji**, Guodong Tong, Yuki Hiruta, Daniel Citterio*

PS2-33 Platform for verification of electrochemical sensors for biomedical applications
***Alwin Verschueren**, Jos F.M. Oudenhoven, Yawar Abbas, Thijl Boonen, Dimitrios Koutsouras, Greja Brom-Verheyden, Esra Kamer, Chloe Baldasseroni, Sneha, Marcel A.G. Zevenbergen*

PS2-34 Self-powered optical potentiometric sensors array based on electronic paper
***Yaotian Wu**, Riqileng Ao, Eric Bakker*

PS2-35 Hierarchical architectures of graphene as sensitive membranes for electrochemical sensors
***Volodymyr Zaitsev**, Albina Mikhraliieva, Olena Artiushenko, Michael Nazarkovskyi*

PS2-36 In-depth Study of Tyrosine Oxidation Using Electrochemistry, Capillary Electrophoresis, and Mass Spectrometry
***Seyedehelahe Bagherimetkazini**, Frank-Michael Matysik*

PS2-37 Modification-free boron-doped diamond as a sensing material for direct and reliable detection of the anti-HIV drug nevirapine
Simona Baluchová*, Antigoni Mamaloukou, Rombert H.J.M. Koldenhof, Josephus G. Buijnsters*

PS2-38 Simultaneous voltammetric determination of prothioconazole and bixafen on a boron-doped diamond electrode
Mariola Brycht*, Andrzej Leniart, Barbara Burnat, Sławomira Skrzypek*

PS2-39 The development of molecularly imprinted polymer-based electrochemical sensor for the selective and sensitive determination of tolvaptan
Fatma Budak*, Leyla Karadurmus, Ahmet Cetinkaya, Esen Bellur Atici, Sibel A. Ozkan*

PS2-40 Molecularly imprinted sensor based on CNFs for voltammetric detection of dasatinib
Emin Çorman*, Emrecan Yıldız, Ahmet Cetinkaya, Lokman Uzun, Sibel A. Ozkan*

PS2-41 The application of the modified carbon paste electrode in voltammetric sensing of ibuprofen
Ana Đurović*, Zorica Stojanović, Sanja Panić, Snežana Kravić*

PS2-42 All-solid-state potentiometric sensors based on graphene oxide as novel ion-to-electron transducer for nitrate and nitrite detection in environmental waters
Renato Gil*, Begoña Espiña, Raquel Queirós*

PS2-43 Spectroelectrochemical approaches for the qualitative and quantitative analysis of acetaldehyde in wine, fentanyl in drug of abuse and pesticide detection.
David Ibáñez, ***Laura del Carmen Garcia Alcalde****, María Begoña González-García, David Hernández-Santos, Pablo Fanjul-Bolado*

PS2-44 On-site simultaneous determination of calcipotriol and betamethasone in topical pharmaceutical formulations and surface water samples using an intelligent mini platform based on carbon nanotubes-gold nanoparticles screen-printed electrode modified with calix[6]arene
Bianca-Maria Tuchiu*, Raluca Ioana Stefan-van Staden, Jacobus (Koos) Frederick van Staden*

PS2-45 An electrochemical sensor for trace analysis of morphine in human serum and saliva
Hamideh Imanzadeh, Leila Hazrati, ***Mandana Amiri****, Alireza Khataee*

PS2-46 Uncovering the multiple adsorption mechanisms of heavy metals by eggshells
Yair Amar

PS2-47 Assessment of metal content in agricultural soils and vegetables and their risk to human health in rural Roma communities in Transylvania, Romania
Mihail Simion Beldean-Galea*, Ioana Perhaița, Adrian Cadiș, Vlad Alexandru Pănescu, Victor Bocoș-Binținţan, Maria-Virginia Coman, Vidar Berg*

PS2-48 Conception of a test gas system for simulating complex air mixtures of biogenic volatile organic compounds in the ppt range
Jennifer Braun

PS2-49 Development of a multiresidue method including organotins, based on liquid chromatography coupled to tandem mass spectrometry, for the quantification of emerging micropollutants in Gammarus fossarum
Mathilde Duny*, Aurélie Cortéjade, Laure Wiest, Mickael Nicolas, Emmanuelle Vulliet*

PS2-50 Strategies for on-site determination of trace elements in officinal plants by stripping voltammetry
Ornella Abollino, Mery Malandrino, Valentina Isaja, Paolo Inaudi, Lorenza Bertaina, Alessia Pirra, ***Laura Favilli****, Agnese Giacomino*

PS2-51 Determination of Benzo(a)pyrene adsorbed onto plant pollen samples by microwave extraction and HPLC-FLD
Juan Jesús Hidalgo-Barquero*, Selena Carretero-Peña, Eduardo Pinilla-Gil*

PS2-52 Fingerprinting of Chlorinated Paraffins and Olefins in Sewage Sludge of a Swiss Wastewater Treatment Plant

***Jules Hutter**, Oscar Mendo Diaz, Marco Knobloch, Markus Zennegg, Andreas Buser, Jean Claude Vogel, Urs Stalder, Laurent Bigler, Susanne Kern, Davide Bleiner, Norbert Heeb*

PS2-53 Tyre wear ingredients: Markers and environmental behaviour in soil
*Nadja Plüss, Luca Meyer, Basilius Thalmann, **Susanne Kern***

PS2-54 Analysis of Per- and Polyfluoroalkyl Substances in Aqueous Samples by SPE and LC-MS/MS according to EPA Draft Method 1633
***Hans Rainer Wollseifen**, Lukas Emmerich*

PS2-56 Determination of 2-chloroethanol as a marker of fumigant ethylene oxide in sesame seeds by HS-SPME-GC-MS
***Frank Michel**, Deyny Mendivelso-Perez, Olga I. Shimelis*

PS2-57 Development and analysis of flavonoids and phenolic acids from mandarin fruits by LC-DAD/MS
***Luna Maslov Bandic**, Kristina Vlahovicek-Kahlina, Marija Sigurnjak Bureš*

PS2-58 Evaluating the potential of Irish Faba Beans as a protein alternative using multiple analytical techniques (FAAS, GFAAS & Kjeldahl Method)
***Laura Mcdaid**, Denis McCrudden, Sheila Alves*

PS2-59 A novel HPLC-DAD method for determination of hydrogen peroxide in milk
***Liudmila Istomina**, Veronika Mishina, Sergey Andreev, Konstantin Sakharov, Andrey Kuzovlev*

PS2-60 Continuous monitoring of Lactoferrin for real-time process control
***Claire Michielsen**, Junhong Yan, Arthur de Jong, Menno W. J. Prins*

PS2-61 Classification of Soybean Paste Products Using Laser-Induced Breakdown Spectroscopy, Inductively-Coupled Plasma Optical Emission Spectroscopy, and Inductively-Coupled Plasma Mass Spectrometry
***Sang Ho Nam**, Yonghoon Lee, Hyang Kim*

PS2-62 Developing analytical method for the determination of Inpyrfluxam and its metabolites residues in agricultural products
Inju Park

PS2-63 Stability of water-soluble vitamins in enteral food
***Kristina Pregiban**, Lidija Brkljačić, Ivanka Jerić*

PS2-64 GC/MS/MS AS A THE BEST TECHNIQUE FOR DETECTION AND IDENTIFICATION OF LONG-TERM STEROID MARKERS IN DOPING CONTROL
***Anna Jarek**, Marzena Wojtowicz-Zawadka, Agnieszka Urbaniak-Zbyszynska, Katarzyna Chajewska, Malgorzata Pasik, Dorota Kwiatkowska*

PS2-66 Investigation of bioactive metabolites in 36 Iris species and cultivars grown under different cultivation conditions
***Tereza Jaegerová**, Marie Zlechovcová, Jitka Viktorová, Petr Kaštánek, Miroslav Vosátka, František Beneš, Lenka Fišarová, Jana Hajšlová*

PS2-67 Adaptative response to tetraconazole-based fungicides shapes the proteome of Saccharomyces cerevisiae Lalvin EC1118™
*Noelia Briz-Cid, Javier Alonso del Real, Elena Martínez-Carballo, Amparo Querol, **Raquel Rial-Otero***

PS2-68 Development of a HILIC-MS/MS method for covering short, medium and long chain acyl-CoA in one analytical run
Madhulika Singh

PS2-69 SCORE-metabolite-ID – Identification of metabolites from complex mixtures by correlation of 1D-1H or 2D-HSQC NMR, MS and LC data
***Stephanie Watermann**, Thomas Hackl*

PS2-70 Application of microdialysis combined with UHPLC-QTOF/MS to screen for endogenous metabolites in aquatic organisms as biomarkers of exposure to an emerging contaminant, triclosan
***Yu He**, Bo-wen Huang, Adabelle Ong, Wen-li Wang, Yong-jun Xiao*

PS2-71 Untargeted urinary metabolomics for identification of bladder cancer biomarkers using HPLC-MS
***Anastasiia Frolova**, Ivan Plushchenko, Mikhail Vokuev, Igor Rodin*

PS2-72 Electrochemical Approach on Interaction of Nerve Agent Metabolite and Albumin
***Nurgul Karadas Bakirhan**, Sermet Sezigen, Irem Kaya, Sibel A. Ozkan*

PS3-01 The study of topotecan sorption/desorption kinetics for poly(2-hydroxyethyl methacrylate) gels by UHPLC-MS/MS
*Barbora Mudrova, **Zuzana Bosakova***

PS3-02 Synthesis, Characterization, and Anticancer Evaluation of Phenanthroline-Based Macrocyclic Ligand and Nickel Complex: DNA Binding and Thermal Stability Studies
***Emmanuel Ohaekenyem**, Ikenna Onyido*

PS3-03 A technique to analyze and measure the amount of tar generated from the pyrolysis of waste tyres
***Sergejs Osipovs**, Aleksandrs Puckins*

PS3-04 Development of a high-throughput screening assay to identify glutathione S-transferase (GST P1) inhibitors for potential use in cancer treatment
***Sarah A. P. Pereira**, Jonathan Vesin, Marc Chambon, Lúcia M. F. S. Saraiva, Paul J. Dyson*

PS3-06 Polyampholite hydrogels organized by dynamic bonds
***Esra Su**, Gaukhar Toleutay*

PS3-07 Pregnancy as a factor influencing the change of the steroid profile in terms of assessment of athlete's biological passport
Dorota Kwiatkowska, Arkadiusz Kaplinski, Agnieszka Urbaniak-Zbyszynska, Anna Jarek, Monika Tarka, Katarzyna Chajewska, Paulina Siek, Malgorzata Pasik, Zuzanna Szczepanska, ***Marzena Wojtowicz-Zawadka***

PS3-08 Development of a multi-targeted UHPLC-MS/MS method for steroid profiling in biological samples
***Mathieu Galmiche**, Gioele Visconti, Oriane Strassel, Isabel Meister, Serge Rudaz*

PS3-09 Construction of a generalized interaction model for molecular pattern-recognition of pectic heteropolysaccharides by TLR4
***Gyuhwan Hyun**, IN HO CHO, SUNG WON KWON*

PS3-10 Trace Determination of Silicones in Pharmaceutical Devices Using Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES)
***Peter Franzmann**, Basil Bösch, Rene Frankfurter, Gisela Fontaine*

PS3-11 Bio-based antimicrobial peptides for smart response self-disinfected surfaces
***Sutida Jansod**, Amira Ben Mansoura, Justine Horner, Rudy Koopmans, Roger Marti*

PS3-12 Residual enzyme limit test by UHPLC-MS
Naomi Lagarde

PS3-13 Systematic assessment of feature selection methods with PLS-DA model for photonic in vitro detection of lung cancer
***Harun Hano**, Andreas Seifert*

PS3-15 Unified Approach to Univariate Analytical Calbration
Paweł Kościelniak

PS3-16 A model for the identification of wood-derived mordant dyes in cultural heritage objects using mass spectrometry and chemometric tools
***Katarzyna Lech**, Izabela Nasiłowska*

PS3-17 Leveraging physics-informed machine learning to expand use of electronic tongues for environmental applications
***Amy Mueller**, Ben Eck*

PS3-18 Sensitive detection and electrochemical evaluation of the anticancer drug tofacitinib in pharmaceutical and biological samples using two different electrodes
***Fatma Budak**, Ahmet Cetinkaya, S. Irem Kaya, Esen Bellur Atici, Sibel A. Ozkan*

PS3-20 ELECTRODE DESIGN AND ANALYSIS OF Cr DOPING INTO NASICON-STRUCTURED Na3V2(PO4)3 CATHODE WITH SELF-CARBON-COATING
Jaekook Kim

PS3-21 Combining Electroanalysis with Photocatalysis: Moving Beyond Remediation
***Padraig McDonagh**, Wesley McCormick, Denis McCrudden, Nathan Skillen, Peter Robertson*

PS3-22 Biosensor development: Employing Self-Assembled Monolayers and Electrochemical Transducers
***Jennifer McLeod**, Dianne Lee, Ishwar Singh, Stephen Brown, Cathleen Crudden, Zhe She*

PS3-23 Fabrication of cobalt oxide-supported carbon paste electrode for sensitive and selective Levofloxacin sensing
***Tijana Mutić**, Miloš Ognjanović, Dalibor Stanković, Slavica Ražić*

PS3-24 Ultrasensitive fluoride detection in aquatic environments.
***Andrea Nonis**, Robin Nussbaum, Stéphane Jeanneret, Thomas Cherubini, Eric Bakker*

PS3-25 MERCURY: FROM ATMOSPHERIC POLLUTION INTO BLOOD. ULTRASONIC MICROEXTRACTION AND DISPOSABLE SCREEN-PRINTED GOLD ELECTRODES FOR VOLTAMMETRIC MONITORING OF Hg IN BLOOD SAMPLES
***María Del Rosario Palomo Marín**, Eduardo Pinilla Gil, Pedro Suero Luna*

PS3-26 Development of a Novel Molecularly Imprinted Polymer-Based Electrochemical Sensor for the Selective Determination of Ethyl Methylphosphonic Acid
***Sermet Sezigen**, S. Irem Kaya, Nurgul Karadas Bakirhan, Sibel A. Ozkan*

PS3-27 Self-referencing Pulstrode: Further Optimization and New Electrode Designs
Ayian Speck, Tara Forrest, Elena Zdrachek, Eric Bakker, Silvia Generelli, Davide Migliorelli, Loïc Burr

PS3-28 Sensitive simultaneous electrochemical determination of reduced and oxidized glutathione in urine sample using modified carbon paste electrode
***Zorica Stojanović**, Ana Đurović, Snežana Kravić, Amir Ashrafi, Zuzana Koudelková, Ondřej Zítka, Lukáš Richtera*

PS3-29 Electrochemical determination of phenolic antioxidant BHT in cosmetic and food samples
***Ruxandra-Maria Ilie-Mihai**, Raluca Ioana Stefan-van Staden, Jacobus (Koos) Frederick van Staden*

PS3-30 Antimony remediation using a new magnetic system in potable aqueous samples.
*Maria Mar Lopez Guerrero, **Irene Sanchez Trujillo**, Irene Morales Benitez, Juan Carlos Garcia-Mesa, Carlos Vereda Alonso*

PS3-31 In situ seasonal monitoring of the potentially bioavailable Nickel dissolved fraction in Lake Geneva
***Nicolas Layglon**, Sébastien Creffield, Tanguy Gressard, Laureline Gorse, Eric Bakker, Mary-Lou Tercier-Waeber*

PS3-32 Photodegradation of Textile Pollutants in Wastewater by Nanocomposite Membranes
***Hafiza Hifza Nawaz**, Muhammad Umar, Hugh Gong*

PS3-33 Pollution assessment and source apportionment of Persistent Organic Pollutants in soil of Rural Roma Communities in Transylvania
***Vlad Alexandru Pănescu**, Mihail Simion Beldean-Galea, Mihaela Cătălina Herghelegiu, Victor Bocoș-Binținţan, Vidar Berg*

PS3-34 Direct measurement of organic micropollutants in natural water and wastewater using fluorescence spectroscopy
***Lesly Paradina Fernández**, Urban Wünsch, Kathleen Murphy*

PS3-35 Analysing sorption Kd of fluoroquinolone antibiotics in soils and soil components
***Anna Rigol**, Miquel Vidal, Joel Fabregat-Palau*

PS3-36 DIRECT MERCURY SPECIATION IN SOLID SAMPLES USING THERMAL RELEASE COUPLED TO ELECTROTHERMAL ATOMIC ABSORPTION SPECTROMETRY
***Olga Shuvaeva**, Dmitrii Troitskii, Ivan Bekesha*

PS3-38 EXTREMADURAN CHARCOAL: QUALITY AND POSSIBILITIES AS A BIOCHAR
***Francisco Javier Yuste-Córdoba**, Belén Godoy-Cancho, Delia Omenat-Morán*

PS3-39 Ion-Selective Electrodes as Companion Diagnostics for Personalized Treatment of Mental Health Disorders
Maral Mousavi

PS3-40 Point-of-care testing of LDL cholesterol using molecularly imprinted polymers
Gian Luca de Gregori[1], Denis Prim, Marc Emil Pfeifer, ***Jean-Manuel Segura***

PS3-41 Smart Portable Device Based on the Utilization of a 2D Disposable Paper Stochastic Sensor for Fast Ultrasensitive Screening of Food Samples
***Raluca Ioana Stefan-van Staden**, Irina-Alina Chera-Anghel, Damaris-Cristina Gheorghe, Jacobus (Koos) Frederick van Staden, Marius Badulescu*

PS3-42 IDENTIFICATION OF SELECTED NONSTEROIDAL ANTI-INFLAMMATORY DRUGS IN HORSES' URINE AND BLOOD – GC/MS/MS METHODS VALIDATION.
***Anna Jarek**, Marzena Wojtowicz-Zawadka, Agnieszka Urbaniak-Zbyszynska, Katarzyna Chajewska, Danuta Stanczyk, Dorota Kwiatkowska*

PS3-43 A chemometric approach to discrimination of isobaric β- and γ- isoforms of tocopherol and tocotrienol using RPLC-ESI-MS
***Katarzyna Pawlak**, Zuzanna Jopek, Elżbieta Święcicka-Fuchsel, Jesse Namo Ombugadu, Alicja Kutyła, Kamil Wojciechowski*

PS3-44 New method to increase the efficiency of DNA extraction using dielectrophoresis
***Camila Campos**, Ying Ting Set, Liesbet Lagae*

PS3-45 A lateral flow smartphone-based biosensor for rapid on-site assay of carcinoembryonic antigen
***Varvara Pagkali**, Ioanna Tsogka, Electra Mermiga, Christos Kokkinos, Prfessor Anastasios Economou*

PS3-46 Multiplexed LC - MS permeation analysis in artificial cell systems
***Robert Strutt**, Simon F. Berlanda, Petra S. Dittrich*

PS3-47 Electromembrane extraction of peptides based on hydrogen bond interactions
***Samira Dowlatshsh**, Frederik André Hansen, Chen Zhou, María Ramos-Payán, Trine Grønhaug Halvorsen, Stig Pedersen Bjergaard*

PS3-48 VIRTUAL INSTRUMENTS FOR FILLING A GAP IN PEAK EVALUATION SOFTWARE FOR FLOW-BASED METHODS
Nataša Gros

PS3-51 Multiple Critical Quality Attributes Assessment of mAbs for Process Control - Agilent InfinityLab Online LC Solution for automated heart-cutting 2D-LC experiment
Susanne Stephan, Edgar Naegele, ***Jens Trafkowski***

PS3-52 Calix[6]arene and TiO2 modified reduced graphene oxide electrode-based portable stochastic platform for the determination of nonivamide from topical pharmaceutical dosage forms and water samples
***Bianca-Maria Tuchiu**, Raluca Ioana Stefan-van Staden, Jacobus (Koos) Frederick van Staden*

PS3-53 Efficient sol-gel immobilization of microporous polymer on silica-based adsorbent for the enrichment of non-steroidal anti-inflammatory drugs
Abdullah Alhendal*, Mohammad Rashad, Ali Husain, Fouzi Mouffouk, Saad Makhseed*

PS3-54 Application of microextraction in a packed syringe approach for determination of phosphate in natural water samples
Serhii Zaruba, Vivien Oltmanová, ***Vasil Andruch***

PS3-55 Assessment of Hollow Fiber and Dispersive Solid Phase Microextraction combined with Total Reflection X-ray Spectrometry (TXRF) for Inorganic Arsenic Speciation Analysis in Water
Manuela Hidalgo*, Eva Marguí, Santanu Majumder, Debashis Chatterjee*

PS3-57 Polymer nanofibrous disks for preconcentration of environmental contaminants prior to HPLC determination
Ivona Lhotská*, Martina Háková, Slavomíra Zatrochová, Jana Nastoupilová, Dalibor Šatínský*

PS3-58 New zwitterionic materials for the selective extraction of analytes from environmental samples
Alberto Moral, Francesc Borrull, Núria Fontanals, ***Rosa M. Marcé***

PS3-59 Vacuum Assisted Sorbent Extraction: VASE, a qualitative and green approach for VOCs to SVOCs analysis using GCMS
Dalel Raclot

PS3-60 A new environmentally friendly procedures for preconcentration and online monitoring of selected analytes
Andrea Gajdošová, Kristína Terebesiová, Diana Kendiová, Vasil Andruch, ***Jana Šandrejová***

PS3-61 In Situ Rapid Electrochemical Fabrication of Porphyrin-based Covalent Organic Frameworks Fibers for Electro-enhanced Solid-phase Microextraction
Wenmin Zhang*, Hui Chen, Lan Zhang*

PS3-62 Nanoparticle-directed metal organic framework and ionic liquids@metal organic framework nanocomposites hybrid monolith for efficient capillary microextraction
Ting Huang, Shuqiang Wang, ***Xiaoping Wu***

PS3-63 A new concept for the control of functional food creation methods by speciation analysis of various elements present in microalgae
Lena Ruzik

PS3-64 Honey characterization and classification based on chromatographic profiles and antioxidant capacity
Victor García-Seval, Monica Fernández-Estellé, Javier Saurina, Oscar Núñez, ***Sonia Sentellas***

PS3-65 Dietary fatty acids as a new binding partner of C - phycocyanin: a fluorimetric study
Miloš Šunderić*, Luka Velickovic, Nikola Gligorijevic, Ljubodrag Aleksic, Milan Nikolic, Marija Takic, Simeon Minic*

PS3-66 Determination of Veterinary Drug Residues in Foods of Animal Origin Using QuEChERS methology by LC–MS/MS
Hans Rainer Wollseifen*, Rebecca Nuessgen*

PS3-67 Simultaneous determination of vitamins B5, B7 and B9 using stochastic sensors as tools
Damaris-Cristina Gheorghe*, Raluca Ioana Stefan-van Staden, Ergün Rasit, Jacobus (Koos) Frederick van Staden*

PS3-69 Extra-virgin olive oil phenolic compounds as modulators of the gut microbiota in diabetics: unravelling their colonic metabolism
Carmen González-Barreiro*, Patricia Reboredo-Rodrígueza, María Figueiredo-González, Lucía Olmo-García, Alegría Carrasco-Pancorbo, Sonia Pérez-Castro, Beatriz Cancho-Grande*

PS3-70 Development of ultrafast PCR assays to detect Artemisia annua and Ambrosia artemisiifolia
Lim Ho Soo*, Hong Yewon, Lee Ja Hyun, Kim Bora, Kim Hyung-Soo, Mun Jae-Eun*

PS3-71 Simplified LC-MS/MS method for glyphosate and related compounds in oat cereals using a new Carbon HPLC column
Frank Michel, *Olga I. Shimelis, Clinton Corman, Martin J. Ross, Michael Ye, Cory Muraco*

PS3-72 Characterization of the diurnal pattern of exhaled fatty acids and enteric methane emissions in dairy cows
Stamatios Giannoukos, *Zakirul Islam, Susanna Räisänen, Yang Li, Fabian Wahl, Renato Zenobi, Mutian Niu*

PS3-73 Methodological and kinetic aspects of Oxygen Radical Absorbance Capacity assay for evaluation of radical scavenging capacity
Andreia N. Meireles, Joana R. B. Carvalho, Sara S. Marques, Bruno J. R. Gregório, Inês I. Ramos, Eduarda M. P. Silva, Luisa Barreiros, ***Marcela Segundo***[1]

PLENARY LECTURES

PL-1 Historical View on Analytical Sciences in Switzerland

Detlef Günther[1]
[1]*ETH Zurich, Zurich, Switzerland*

Analytical Sciences have a long tradition and history and major and important developments applied and accepted today have their roots within Switzerland. The analytical history starts, beside many other contributions, with Jean Charles Galissard de Marignac (Geneva) who discovered the two elements gadolinium and ytterbium and determined the exact atomic weight of 29 elements with highest precision. Almost at the same time, Francois Alphonso Forel studied sediments and water which finally led to the foundation of the research direction of Limnology. A pioneer in quantitative analysis of inorganic compounds has been Frederick Pearson Treadwell, who became Ordinarius at ETH Zurich in 1993. He provided textbooks and tables for quantitative analysis, which have been translated into 6 different languages and have been used for teaching at universities. Unusually, due to his sudden death followed his son William Dupré as professor for Analytical Chemistry at ETH.

Another well-known Analytical Scientist with a significant impact in Switzerland has been Karl Gerold Schwarzenbach (1904-1978) who became professor for "Analytische und Spezielle Anorganische Chemie" at the University of Zurich and 1955 ordinarius at ETH. He was involved in the research on EDTA and developed 1945 the analytical procedure for detection of metal-ions by chelatometry. The pioneering work has been summarized in a standard text book about titration.

Out of his school followed Werner Stumm (Harvard), Walter Schneider (ETH), and Hans Michael Widmer (Ciba-Gigy Ltd., Novartis). Walter Schneider became the first chair of environmental science (Umweltnaturwissenschaften) and has shaped this research at the interface of chemistry and environment. Hans Michael Widmer pushed the miniaturization and is pioneer together with his student and later colleague Andreas Manz, which finally found his way into micro fluidics. Wilhelm Simon worked in the Laboratorium Ruzicka developed the field of automated determination of carbon, hydrogen and nitrogen. He studied the ion transport through artificial membranes, which finally resulted in the potassium-selective electrode. From his research group a major impact on analytical and environmental sciences has been created in a large number of professors who contributed in Analytical Sciences and industry (e.g. Andreas Manz, René Schwarzenbach, Walter Giger, Ernö Pretsch, Ursula Spichiger-Keller).

The list of achievements and contributions from Swiss scientists is long and will never be complete. However, some major milestones will be presented and some evolution trees creating major impact for Analytical Sciences in Switzerland will be given.

PL-2 Identifying Metabolic Regulation through Metabolomics

Uwe Sauer[1]
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In the cellular context, metabolomics is possibly the most difficult Omics to make sense of because of extremely rapid metabolite responses and the absence of a direct link to the genome. Delineating

confounding metabolome responses that arise as indirect consequences of a perturbation is pivotal for mechanistic interpretation and a challenge for experimental design. Exploiting the potential of high-throughput metabolomics by flow-injection TOF, I will highlight experimental designs that allow to infer metabolic regulation (1-4). A particular focus will be on short-term regulation through allosteric metabolite-protein interactions. In metabolism, the regulatory logic of these interactions is apparently conserved across enormous phylogenetic distance, suggesting their relevance for maintaining cellular homeostasis in ancient biological processes that are common to all cells (5).

1. *Christodoulou D, Link H, Fuhrer T, Kochanowski K, Gerosa L & U Sauer. Cell Systems 6: 569-578 (2018).*
2. *Zampieri M, Hörl M, Hotz F, Müller N & U. Sauer. Regulatory mechanisms underlying coordination of amino acid and glucose catabolism in E. coli. Nature Comm. 10:3354 (2019).*
3. *Basan M, Honda T, Christodoulou, Hörl M, Chang y-F, Leoncini E, Mukherjee A, Okano H, Taylor BR, Silverman JM, Sanchez C, Williamson JR, Paulsson J, Hwa T & U Sauer. A universal trade-off for bacterial growth in fluctuating environments. Nature 584: 470-474 (2020).*
4. *Schastnaya E, Raguz Nakic Z, Gruber CH, Doubleday PF, Krishnan A, John NI, Park J, Harris HH & U Sauer. Extensive regulation of enzyme activity by phosphorylation in E. coli. Nature Comm. 12:5650 (2021).*
5. *Gruber CH, Diether M & U Sauer. Conservation of metabolic regulation by phosphorylation and non-covalent small-molecule interactions. Cell Systems 12: 538-546 (2021).*

PL-3 Luminescent sensors: making the invisible visible

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Optical sensors since long belong to arsenal of modern analytical tools. Among them, luminescent sensors not only allow for reliable quantification of many analytes but literary can enable “seeing” the distribution of them with help of dedicated equipment or even a human eye [1]. Luminescent sensors can be miniaturized rather easily that makes it possible to measure in very small objects including cells and even organelles. Another attractive feature is the property of light to guide multiple information that is utilized to simultaneously measure several parameters with the same material.

In the talk luminescent sensors for gases (oxygen [2], carbon dioxide [3] and ammonia [4]), pH and alkali metal ions [5] will be highlighted. It will provide an overview of indicator dyes and materials, show selected applications in biology, oceanography [6], medical research and other fields and briefly discuss commercialization of optical sensing technology. Recent progress in design of new generation of molecular thermometers [7] making use of thermally activated delayed fluorescence will also be highlighted.

[1] M. Schäferling, The art of fluorescence imaging, Ang. Chem. Int. Ed. 51 (2012) 3532-3554. DOI: 10.1002/anie.201105459
[2] R. I. Dmitriev, S. M. Borisov, H. Düssmann, S. Sun, B. J. Müller, J. Prehn, V. P. Baklaushev, I. Klimant, D. B. Papkovsky, Versatile Conjugated Polymer Nanoparticles for High-Resolution O2 Imaging in Cells and 3D Tissue Models, ACS Nano 9 (2015) 5275-5288. DOI: 10.1021/acsnano.5b00771.
[3] S. Schutting, T. Jokic, M. Strobl, S. M. Borisov, D. de Beer, I. Klimant, NIR optical carbon dioxide sensors based on highly photostable dihydroxy-aza-BODIPY dyes, J. Mater. Chem. C 3 (2015) 5474-5483. DOI: 10.1039/C5TC00346F
[4] M. Strobl, A. Walcher, T. Mayr, I. Klimant, S.M. Borisov, Trace ammonia sensors based on fluorescent NIR-emitting aza-BODIPY dyes, Anal. Chem. 89 (2017) 2859-2865. DOI: 10.1021/acs.analchem.6b04045
[5] B. J. Müller, S. M. Borisov, I. Klimant, Red-to NIR-Emitting, BODIPY-Based, K+-Selective Fluoroionophores and Sensing Materials, Adv. Funct. Mater. 26 (2016) 7697-7707, DOI: 10.1002/adfm.201603822
[6] C. Staudinger et al. A versatile optode system for oxygen, carbon dioxide, and pH measurements in seawater with integrated battery and logger, Limnol. Oceanogr.Meth. 16 (2018) 459-473. DOI: 10.1002/lom3.10260

[7] A. Russegger, A. C. Debruyne, D. Carvajal Berrio, S. Fuchs, J. Marzi, K. Schenke-Layland, R.I. Dmitriev, S.M. Borisov Bright and Photostable TADF-Emitting Zirconium(IV) Pyridinedipyrrolide Complexes: Efficient Dyes for Decay Time-Based Temperature Sensing and Imaging, Adv. Opt. Mater. (2023) in press. DOI: 10.1002/adom.202202720

PL-4 Spectroscopy with Quantum Cascade Lasers for High-Precision Gas Analysis

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Infrared (IR) laser spectroscopy is a powerful tool for gas sensing. Especially attractive is the mid-IR spectral region, where the molecules of interest have their fundamental absorption bands with cross sections that are up to five orders of magnitude larger than in the near-IR. With the advent of room-temperature semiconductor lasers, mainly quantum cascade lasers (QCL) and interband cascade lasers (ICL), mid-IR light sources have become available that allow the development of highly sensitive, selective and compact gas sensors.

This paper highlights some illustrative examples from our laboratory based on QCL absorption spectroscopy. It includes (i) long-term, high-precision monitoring of CO2 and its stable isotopes at the high-altitude station Jungfraujoch, (ii) the detection of nitrogen dioxide (NO2) with sub-pptv precision in ambient air (iii) the selective quantification of N2O and CO2 isotopomers and isotopologues, (iv) drone-based measurements of methane for leak detection in the oil and gas industry, (v) water vapour measurements on-board of a balloon up to the lower stratosphere, (vi) multi-component, high-precision measurements of air-quality and greenhouse gases, and (vii) high-precision measurements of volatile organic compounds and their stable isotopes.

S.M. Pieber et al. Analysis of regional CO2 contributions at the high Alpine observatory Jungfraujoch by means of atmospheric transport simulations and δ13C. Atmos. Chem. Phys. (2022). doi.org/10.5194/acp-22-10721-2022
B. Tuzson et al. High precision and continuous field measurements of δ13C and δ18O in carbon dioxide with a cryogen-free QCLAS. Appl. Phys. B (2008). doi.org/10.1007/s00340-008-3085-4
B. Tuzson et al. Selective measurements of NO, NO2 and NOy in the free troposphere using quantum cascade laser spectroscopy. Atmos. Meas. Tech. (2013) doi.org/10.5194/amt-6-927-2013
N. Sobanski et al. Advances in high-precision NO2 measurement by quantum cascade laser absorption spectroscopy. Appl. Sci. (2021). doi.org/10.3390/app11031222
K. Kantnerová et al. First investigation and absolute calibration of clumped isotopes in N2O by mid-infrared laser spectroscopy. Rapid Commun. Mass Spectrom (2020). doi.org/10.1002/rcm.8836
A. Nataraj et al. Quantum cascade laser absorption spectrometer with a low temperature multipass cell for precision clumped CO2 measurement. Opt. Express (2023). doi.org/10.1364/OE.44717
B. Tuzson et al. A compact QCL spectrometer for mobile, high-precision methane sensing aboard drones. Atmos. Meas. Tech. (2020). doi.org/10.5194/amt-13-4715-2020
M. Graf et al. Compact, circular, and optically stable multipass cell for mobile laser absorption spectroscopy. Opt. Lett. (2018). doi.org/10.1364/OL.43.002434
M. Graf et al. Compact and lightweight mid-infrared laser spectrometer for balloon-borne water vapor measurements in the UTLS. Atmos. Meas. Tech. (2021). doi.org/10.5194/amt-14-1365-2021
M. Hundt et al. Multi-species trace gas sensing with dual-wavelength QCLs. Appl. Phys. B. (2018). doi.org/10.1007/s00340-018-6977-y
O. Aseev et al. High-precision ethanol measurement by mid-IR laser absorption spectroscopy for metrological applications. Opt. Express. (2019). doi.org/10.1364/OE.27.005314
R. Brechbühler et al. Rapid detection of volatile organic compounds by switch-scan tuning of Vernier quantum-cascade lasers. Anal. Chem. (2023). doi.org/10.1021/acs.analchem.2c04352
A. Nataraj et al. Position-specific isotope analysis of propane by mid-IR laser absorption spectroscopy. Anal. Chem. (2022). doi.org/10.1021/acs.analchem.2c05489

PL-5 Decoding the protein dance: probing the proteome-wide choreography of protein conformational changes

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Significant progress has been recently achieved in the prediction of protein structures, due to advances such as AlphaFold, RoseTTAFold and in general deep-learning based methods. These tools are revolutionizing the field of structural biology by providing reliable predictions of protein 3D structures. Despite these developments, predicting protein dynamics remains a formidable challenge due to the inherent complexity of dynamic motions of proteins and the scarcity of experimental data on alternative protein conformational states. Detecting and characterizing protein conformational changes on a large scale would be however very relevant as they can profoundly alter protein function and activity. In my talk, I will present a mass spectrometric method that enables in situ analyses of protein conformational changes for thousands of proteins simultaneously and across conditions. The approach probes dynamic alterations of protein structures due to post-translational modifications, binding of other molecules, cleavages and protein aggregation events. Such a structural 'omics readout, combined with predicted or experimental protein structural data, enables screening for pathological or physiological protein conformations from cells and tissues, supports the generation of mechanistic hypotheses and bridges system and molecular views. I will present applications of this approach to the in situ detection of drug-target interactions and the identification of a novel class of structural disease biomarkers.

PL-6 Counting molecules, dodging blood cells: continuous, real-time molecular measurements directly in the living body

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The availability of technologies capable of tracking the levels of drugs, metabolites, and biomarkers in real time in the living body would revolutionize our understanding of health and our ability to detect and treat disease. Imagine, for example, a dosing regimen that, rather than relying on your watch ("take two pills twice a day"), is instead guided by second-to-second measurements of plasma drug levels wirelessly communicated to your smartphone. Such a technology would provide researchers and clinicians an unprecedented window into physiology and pharmacology, and could even support ultra-high-precision personalized medicine in which drug dosing is optimized minute-by-minute using closed-loop feedback control. Towards this goal, we have developed a biomimetic, electrochemical sensing platform that supports the high frequency, real-time measurement of specific molecules (irrespective of their chemical reactivity) in situ in the blood and solid tissues of awake, freely moving subjects.

PL-7 Environmental Mass Spectrometry: the long road from sensitive target to comprehensive non-target screening

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For decades, triple quadrupole mass spectrometers coupled to liquid chromatography (LC) or gas chromatography (GC) have been the gold standard for the ultra-sensitive detection and quantification of toxic environmental compounds such as pesticides or estrogens in environmental samples. Only for very complex mixtures of pollutants, such as dibenzodioxins, sector field instruments with high mass resolving power were used at a very early stage. With the advent of high resolution mass spectrometers (HRMS) like Time-of-Flight and Orbitrap instruments coupled with liquid chromatographs (LC) using electron spray ionization (ESI), the field of environmental mass spectrometry evolved rapidly toward the analysis of suspect and non-target compounds. Further enhancements of the instrumental hardware over the past decade in terms of improved mass accuracy, mass resolution, and sensitivity have established LC-HRMS as the standard screening method when it comes to the detection and identification of unknown persistent, mobile and toxic (PMT) compounds in environmental samples.

With the combination of separation technics such as ion mobility separation, ion chromatography or supercritical fluid chromatography, the use of HRMS instruments could even be extended to the detection of isobaric and very polar mobile organic compounds. The next revolution in environmental mass spectrometry is likely to be triggered by the development of robust, fully automated, transportable mass spectrometers with high sample throughput as these mobile devices will enable the on-site detection of highly dynamic concentration courses directly in the field in almost real time (1). In parallel with the tremendous progress in instrumentation, comprehensive software workflows and new data processing algorithms have been developed to obtain full detection and identification power from acquired HRMS and MS/MS datasets. Still, a complete elucidation of all non-target HRMS features detected in an environmental sample is not feasible. Therefore, prioritization strategies based on chemical or experimental characteristics need to be further developed to direct identification efforts toward environmentally relevant compounds (2).

This presentation will highlight the current status and emerging trends in (in-situ) sampling, instrumentation, and software workflows for the detection of target, suspect, and non-target compounds in environmental samples. Illustrative examples, primarily from the aquatic environment, will be used to discuss the challenges and limitations for detecting PMT compounds by suspect and non-target HRMS analysis. A balance of sensitive target methods and comprehensive non-target screening methods remains essential for a holistic assessment of water quality. Environmental mass spectrometry has not yet reached the end of the road

(1) M. Stravs, et al., Environ. Sci. Technol. Lett. 8 (2021) 373−380. DOI: 10.1021/acs.estlett.1c00066
(2) J. Hollender et al., Environ. Sci. Technol. 51 (2017), 11505. DOI: 10.1021/acs.est.7b02184

PL-8 Single cell metallomics

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We will demonstrate the power of spatial single cell metallomics unraveling the biological/toxicological function of metals within tissues. We developed an bioimaging platform relying on high speed laser ablation in combination with inductively coupled plasma time of flight mass spectrometry integrating a standardization concept and a dedicated data evaluation pipeline for pixel-based or single cell based image analysis. Highly precise gelatin microdroplets, perfectly mimicking the tissue matrix, serve as multi-point external calibrants [1]. Our strategy allows the assessment of metal accumulation upon exposure or perturbed homeostasis in single cells [2]. At the same time, cell type, state and function are characterized by integrating immunohistochemistry utilizing metal labelled antibodies [3]. Data evaluation is key for the biological interpretation of the single cell data. Our pipe-line involves the basic steps as established in state-of-the-art imaging mass cytometry (preprocessing, cell segmentation, and downstream statistical analysis), adding the aspect of absolute quantification of the tissue single cell metallome. We will showcase in vitro and in vivo models on perturbed iron homeostasis and toxicity studies of metals in human skin.

[1] A. Schweikert, et al., Anal. Chim. Acta, (2022) 1223 DOI: 10.1016/j.aca.2022.340200
[2] M. Schaier, et al., JACS Au, 3 (2), (2023) 419 DOI: 10.1021/jacsau.2c00571
[3] D. Bandura et a., Anal. Chem. 81, 16 (2009), DOI: 10.1021/ac901049w

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PL-9 Microfluidic devices for analytical and pharmaceutical applications

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Over the past 30 years, the "microchip format" has provided a wealth of innovations to the field of analytical chemistry, with either vastly improved or completely new applications in biochemistry, medicine, pharmaceutical research and the life sciences in general. The combination of microfluidics - the underlying enabling technology - and channel networks on planar substrates, has made it possible to exploit intriguing physics and design highly versatile systems offering a range of functionalities on a single device. Beyond the obvious advantages of reduced consumption of sample and reagents, the true strength of microfluidic devices often lies in providing very controlled chemical environments, where flows are well behaved, diffusion distances are minimized and adverse effects, such as dispersion, can be held at bay more efficiently. Additionally, the almost endless combinations of different functions that can be integrated on a single substrate and the enormous versatility this design freedom offers have also contributed to the continued interest and research efforts invested in such devices. Resulting systems have become known as micro-Total Analysis Systems (uTAS) or lab-on-a-chip devices.

With respect to the above mentioned design freedom alone, planar microfluidic devices seem to have an edge over more traditional formats used in analytical chemistry, such as the (steel) column or the (fused silica) capillary. However, because there is no commonly agreed upon standard for microfluidic devices (despite several attempts), the possibilities for choosing suitable substrate materials has led to an almost bewildering variety of conventional and novel materials and fabrication methods, which can

make it hard for newcomers to enter the field, or for commercial vendors to provide the desired solutions from all possible combinations. It is not unthinkable that this aspect alone has so far hindered a wider acceptance and penetration of microfluidic devices, especially also in industry.

I will attempt to highlight some of the more successful realizations of microfluidic devices from the last 30 years, and also current ones from my own lab, to pinpoint and discuss the advantages, shortcomings and possible ways forward for such systems. Examples will mainly be taken from analytical chemistry and pharmaceutical application, e.g., for sample preparation in tiny volumes, for providing venues for novel drug delivery routes, or for protein structure elucidation. In this context, a still often overlooked strong point of microfluidic devices, namely to act as versatile front-ends for large detection infrastructure will be re-visited as well.

PL-10 Microplastics in the Aquatic Environment : Green Analytical Protocols, Vectors of Pharmaceuticals and Risk to Biota

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Plastic pollution is nowadays a global and ubiquitous problem being detected everywhere: marine environment , sand beaches, wastewaters, surface waters, soils, sludges, sediments, biota, food and air .Microplastics (MPs) are directly released into the water or formed by degradation of Macro-plastics. Having said that , this presentation will cover different aspects of MPs and Macro-Plastic litter pollution in coastal waters , rivers ,sediments and lakes . Case studies of MP pollution in several coastal environments, sediments and catchments of China,. Saudi Arabia, India, Europe and Australia will be reported Another bullet point of this lecture will discuss green analytical chemistry (GAC) protocols for the analysis of MPs in water . Within the last years green, eco-friendly and sustainable solutions are making their way into analytical chemistry. Information on the consumption of toxic solvents and energy is now a part of everyday life.

This lecture will describe as well MPs as vectors of pharmaceuticals as well as its increasing use under Covid-19 outbreak and risk to the aquatic environment. As regards to the increasing concern of MPs as vectors of pharmaceuticals being adsorption as key element that will depend on different factors such as the hydrophobicity of the compounds having a crucial role on the sorption affinity of pharmaceuticals on MPs . MPs contamination and risks substantially increased due to the COVID-19 pandemic due to the excessive use and consumption of single-use plastics (including personal protective equipment such as masks and gloves) . Risks to the aquatic environment affect communities, biological diversity, and ecosystem processes . As regards to toxicity of MPs that is not only related to the toxicity they release into the environment, but also to their size and shape in particular, smaller MPs and nanoplastic particles are more toXic to organisms. Bioindicators and biomonitoring using bivalves such as Mytilus galloprovincialis- “mussel watch”- have been found to be the most suitable organisms for biomonitoring of MPs in the marine environment. Lastly, discussions on the excessive use of plastic should start soon with the involvement of the scientific community, plastic producers, politicians , NGOs and the general public in order to find a sustainable solution.

PL-11 Where nanomaterials can be a unique tool for the improvement of biosensors

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Liposomes in the form of unilamellar nanovesicles with diameters around a hundred to a few hundred nanometers have long been used as cell mimics and as superior enhancement tool in bioanalytical assays and biosensors. Signaling marker molecules can be entrapped in their interior volume and recognition molecules can be coupled to their outer membrane. As they present a soft matter biological surface to their environment and can easily be modified to provide such dual functionality, they are unique among other nanoparticles and vesicles used in bioanalysis. Recent effort in our lab focused on the possibility to study functional lysis of liposomes by bacterial toxins and by the complement system. In both instances, erythrocytes are typically used to monitor cell lysis behavior, a method that is difficult to standardize as it is always dependent on varying erythrocytes sources. The detection of toxin producing pathogens can be accomplished within a few hours rather than the current multiple day analysis.

The complement system is a part of the immune system that plays a crucial role in fighting infections, whereas abnormalities in this system cause a variety of diseases. This includes age-related macular degeneration, Alzheimer's or autoimmune diseases. Moreover, in recent years, a new class of immunosuppressive drugs that target the complement system, have been approved for clinical use and others are in the pipeline in clinical studies. At the same time, surprisingly, much is unknown about the functional roles of its participating proteins. Current technology available to assess the functionality of the complement system is based on protein quantification with an ELISA or fresh erythrocyte lysis. Here, we discuss the development of highly serum-stable liposomes for targeted, specific and reliable functional assays, designed to provide quantitative, standardized data analyses. They are applied to study the total complement activity and pathway-specific analysis while providing simplified handling and avoiding the use of sheep erythrocytes. Furthermore, such liposomes allow fluorescent, visual, luminescence or electrochemical detection strategies and therefore easily enable biosensor and point-of-care strategies in addition to high-throughput studies. Such assays and sensors will become a reliable diagnostic technology accompanying complement-based research efforts and complement-based therapies.

KEYNOTE LECTURES

KN1-1 Mid-Infrared Photonics: From Emerging Technology to Enabling Tool

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Mid- infrared spectroscopy plays an increasingly important role in modern biodiagnostics. This has led to the evolution of mid-infrared photonics from an emerging technology in the clinical/medical domain to an enabling tool that finds its way into daily (bio)medical and clinical use.

With applications ranging from non-invasive exhaled breath analysis to the in-vivo assessment of cartilage damage, mid-infrared (MIR; 3-20 µm) photonics ranges among the most flexible molecular sensing platforms nowadays available. This development has been catalyzed by the emergence of quantum and interband cascade laser (QCL, ICL) technology providing miniaturized laser light sources based on quantum heterostructures that lend themselves to the on-chip hybridization and/or integration of entire MIR sensing systems with the perspective of IR-lab-on-chip devices. The inherent molecular selectivity of MIR signatures enables studying small molecules (e.g., volatile organic compounds; VOCs) in the gas phase, as well as biomacromolecules (e.g., proteins) or entire biological specimen (e.g., cells, viruses, microbes) in the liquid phase at yet unprecedented detail in a label-free and non-destructive fashion.

Finally, the combination with advanced multivariate data evaluation and deep learning algorithms facilitates analyses in real-world complex matrices of biomedical and clinical relevance. The discussion of latest MIR photonic technologies in this presentation we will be augmented by highlight applications underlining the utility of next-generation MIR photonic technologies.

[1] V. Schorer, J. Haas, R. Stach, V. Kokoric, R. Groß, J. Muench, T. Hummel, H. Sobek, J. Mennig, B. Mizaikoff, Towards the direct detection of viral materials at the surface of protective face masks via infrared spectroscopy, Scientific Reports, 12:2309 (2022), DOI: 10.1038/s41598-022-06335-z
[2] I. Vasilikos, J. Haas, G. Q. Teixeira, J. Nothelfer, C. Neidlinger-Wilke, H. Wilke, A. Seitz, D. G. Vavvas, J. Zentner, J. Beck, U. Hubbe, B. Mizaikoff, Infrared attenuated total reflection spectroscopic surface analysis of bovine-tail intervertebral discs after UV-light-activated riboflavin-induced collagen cross-linking, Journal of Biophotonics 13, e202000110 (2020), DOI: https://doi.org/10.1002/jbio.202000110
[3] J. Glöckler, C. Jaeschke, Y. Kocaöz, V. Kokoric, E. Tütüncü, J. Mitrovics, B. Mizaikoff, iHWG-MOX: A Hybrid Breath Analysis System via the Combination of Substrate-Integrated Hollow Waveguide Infrared Spectroscopy with Metal Oxide Gas Sensors, ACS Sensors 5, 4, 1033–1039 (2020), DOI: 10.1021/acssensors.9b02554
[4] A. Lopez-Lorente, P. Wang, B. Mizaikoff, S. Stein, J. Balko, L. Durselen, R. Lu, Surface analysis of sheep menisci after meniscectomy via infrared attenuated total reflection spectroscopy, Journal of Biophotonics e201800429 (2019), DOI: 10.1002/jbio.201800429
[5] E. Tütüncü, M. Naegele, S. Becker, M. Fischer, J. Köth, C. Wolf, S. Köstler, V. Ribitsch, A. Teuber, M. Gröger, S.Kress, M. Wepler, U. Wachter, J. Vogt, P. Radermacher, B. Mizaikoff, Advanced Photonics Sensors Based on Interband Cascade Lasers for Real-Time Mouse Breath Analysis, ACS Sensors, 3, 1743-1749 (2018), DOI: 10.1021/acssensors.8b00477
[6] M. Sieger, B. Mizaikoff, Toward On-Chip Mid-Infrared Sensors, Analytical Chemistry, 88 (11), 5562–5573 (2016), DOI: 10.1021/acs.analchem.5b04143
[7] Á.I. López-Lorente, B. Mizaikoff, Mid-infrared spectroscopy for protein analysis: potential and challenges, Analytical and Bioanalytical Chemistry, 408 (11), 2875–2889 (2016), DOI: 10.1007/s00216-016-9375-5
[8] M. Sieger, J. Haas, M. Jetter, P. Michler, M. Godejohann, B. Mizaikoff, Mid-Infrared Spectroscopy Platform Based on GaAs/AlGaAs Thin-Film Waveguides and Quantum Cascade Lasers, Analytical Chemistry, 88 (5), 2558–2562 (2016), DOI: 10.1021/acs.analchem.5b04144

[9] A. Wilk, J.C. Carter, M. Chrisp, A.M. Manuel, P. Mirkarimi, J.B. Alameda, B. Mizaikoff, Substrate-Integrated Hollow Waveguides: A New Level of Integration in Mid-Infrared Gas Sensing, Analytical Chemistry, 85, 11205–11210 (2013), DOI: 10.1021/ac402391m
[10] B. Mizaikoff, Waveguide-Enhanced Mid-Infrared Chem/Bio Sensors, Chemical Society Reviews, 42 (22), 8683–8699 (2013),
DOI: 10.1039/C3CS60173K

KN1-2 New Ways to Prepare More Performant Stationary Phase Supports for Liquid Chromatography

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Liquid chromatography has significantly evolved over the last century and is today a mainstream working horse for chemical analysis in the (bio)pharmaceutical, chemical, food and other industries. However, the chromatographic separation performance still suffers from randomness of the particle packing. This issue can be resolved by changing to another column format, such as multi-capillary columns. In practice however, the polydispersity effect, which emanates from the inevitable small differences in capillary diameter, completely ruins the potential gain in separation performance.

In this contribution, we prove that the polydispersity effect can be countered by introducing a diffusional crosstalk between the different channels of a multi-capillary column [1]. This has been performed by a fluorescence microscopy based dispersion study of silicon-micromachined microfluidic chips, mimicking a total of 8 different multi-capillary chromatography systems. In the considered examples, diffusional bridging improved the dispersion with a factor of 1330 and 210 for a bidisperse and a polydisperse random multi-capillary systems respectively. The experimental results have moreover been used to validate analytical expressions regarding dispersion in multi-capillary flow systems.

Another way to perfectly ordered columns consists of using micromachined and electrochemically anodized micro-pillar array columns. A first generation of these µPAC's was introduced a couple of years ago and found adoption in proteomics research. Recently, a second µPAC generation has been introduced where the characteristic dimensions (pillar diameter and interpillar distance) have been halved. We provide an on-chip comparison of this new generation compared to the first generation. The observed reduction of H with a factor of 2 around the uopt-velocity and with a factor of 4 in the C-term dominated regime of the van Deemter-curve is in full agreement with the theoretically expected gain. Compared to Gen 1, Gen 2 offers a 4-fold reduction of the required analysis time around the optimal velocity and about a 16-fold reduction in the C-term-dominated range [2].

A downside of the pillar array columns is that they do at present not provide the diverse range of core shell and fully porous spherical particles used in classical columns. Therefore, we recently introduced a new column concept where the existing chromatographic particles are not longer randomly packed, but perfectly ordered in a 2D array of interconnected microgrooves [3]. Computational work shows that this way, the minimal reduced plate height hmin can be reduced from 1.9 to 1.0 for an analyte with a zone retention factor k" = 2.

KN1-3 Analysis of complex biological samples with Confocal Raman Imaging and Chemometrics. A case study: Microplastics in Tissues.

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In this talk, a comprehensive guideline regarding the analysis of complex biological samples is presented with specific emphasis on detecting distinctive features and studying transfer processes of microplastics (MPs) in tissue by Confocal Raman Imaging and Chemometrics. Raman Imaging has demonstrated its high utility but also challenges in detecting minor compounds in samples composed of a high variety of other substances, with uncontrolled baseline drifts, peak overlapping and spikes of different intensities and origin, which makes the Raman signal less selective. All these artifacts have a strong impact on any predictive model approach applied to the spectra. Consequently, comprehensive but concise studies are needed for a most adequate analytical pipeline. This presentation will put forward an analytical methodology composed of a meticulous sample processing, accurate spectral imaging, and a precise chemometric strategy to detect unequivocally 1 µm size polystyrene (PS) microplastics (MPs) in mussels. The strategy combining Principal Component Analysis (PCA) and Multivariate Curve Resolution–Alternating Least Squares (MCR-ALS) in a concise manner to detect the MPs inside the volumetric sample unequivocally.

KN1-4 Sizing and counting particles by high-resolution native charge detection mass spectrometry

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Native mass spectrometry (MS) is a powerful tool for the characterization of non-covalent biomacromolecular assemblies, enabling analysis of proteins and their complexes. Nowadays, for high-resolution, high-mass analysis into the megadalton range, the Orbitrap mass analyzer is frequently employed. In electrospray-based MS experiments, masses are not measured directly, but inferred from the mass-to-charge (m/z) ratios of millions of recorded charge-resolved ions. This method renders impossible for large heterogeneous analytes, because of broad unresolved charge state distributions. Charge detection MS (CDMS) allows circumvention of this issue by simultaneously measuring the charge and m/z ratio of single particles. In this talk I will describe the feasibility of CDMS also an Orbitrap mass analyzers (Orbitrap-based CDMS), successfully enabling the characterization of large polydisperse assemblies, ranging from highly glycosylated antibodies to viruses. We so far demonstrated the power of Orbitrap-based CDMS applied to a variety of fascinating systems, assessing for instance the cargo load of recombinant AAV-based gene delivery vectors, the build-up of immune-complexes involved in complement activation, and quite accurate masses of highly glycosylated proteins, such as the SARS-CoV-2 spike trimer proteins. Moreover, the single molecule nature of the analyses allow us to track the behavior of individual macromolecular ions inside the Orbitrap, which provides unique, fundamental insights into mechanisms of ion dephasing and demonstrated the

(astonishingly high) stability of high mass ions. For standard Orbitrap analyzers, the accessible transient times are maximally ~1 second. By modifying an Orbitrap UHMR we trapped and monitored individual ions for up to 25 seconds, resulting in a unprecedented improvement in signal-to-noise, mass resolving power, and accuracy in charge and mass determination.

KN2-1 High affinity synthetic ligands for protein and virus sensing

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While antibodies are the gold standards for high affinity and selective recognition of molecular targets, they have important limitations. Many of these have been addressed by advances in antibody engineering. However, even if the translation of antibodies from their natural milieu to that required by an analytical task (device) is seamless, which is often not the case, the lack of a full control over their production and properties is a strong enough motivation for the development of synthetic ligands. Therefore, the replacement of natural ligands with synthetic alternatives received outstanding attention and led to new classes of fully synthetic ligands with unprecedented performance and properties. In this respect synthetic ionophores that replaced or complemented the natural macrocyclic compounds (e.g., valinomycin) are some of the most convincing early examples.

For protein recognition aptamers and molecularly imprinted polymers (MIPs) can be highlighted among the most promising fully synthetic ligands, largely because of the general concepts they involve, i.e., evolutionary in vitro screening for aptamers and use of templates to generate materials with molecular memory for MIPs. Therefore, we become interested to develop such ligands for proteins, and through viral proteins to intact viruses[1-2]. Certainly, the affinity and selectivity of such synthetic ligands vary within large limits for the same target, but a general concept if well proven, shows always hope for improvement by refining the building blocks and synthetic methodology. Hence, in case of MIPs, we have developed chip-based high throughput methodologies for both their electrochemical synthesis and label-free screening, which ensured outstanding reliability to produce MIPs recognizing the receptor binding domain of the spike protein of SARS-CoV-2.[3]
Here we are going to present the core enabling technologies and new general approaches to improve the affinity of the MIPs and aptamer ligands:
- by synthesizing and using new amino acid conjugated monomers as buildings blocks [4] for protein MIPs, i.e., “synthetic proteins” that mimic more closely the structure and range of target-ligand interactions of antibodies;
- designing new flexible aptamer structures by catalyst-free click chemistry capable of cooperative binding that we termed superaptamers.

[1] K. Percze et. al, Sci Rep 7 (2017) 42794.
DOI: 10.1038/srep42794
[2] Z. Szakács et al., Nanoscale 10 (2018) 13942.
DOI: 10.1039/c8nr01310a
[3] Z. Bognár, et al. Chem Sci 13 (2022) 1263.
DOI: 10.1039/D1SC04502D
[4] Z. Bognár, et al., Electroanal, (2023), e202300025.
DOI: 10.1002/elan.202300025

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The therapeutic success of monoclonal antibodies (mAbs) in the treatment of various diseases has contributed to their rise, ranking six mAbs and derived products among the 10 best-selling drugs in 2020. From production to patient administration, mAbs are subject to numerous chemical and enzymatic modifications that can alter their biological activity and pharmacological profile.

A complete characterisation of therapeutic proteins and their variants must therefore be carried out by analytical methods, to ensure the safety of the products.

The aim of this presentation will be to detail some innovative approaches recently developed in our laboratory to improve the possibilities offered by chromatographic techniques for the analysis of therapeutic proteins.

A first innovative approach is based on the use of ultra-short columns (only a few millimetres) in reverse phase liquid chromatography (RPLC), to obtain separations as efficient as with standard size columns, but with significantly reduced analysis times. This behaviour is due to the fact that large proteins have an on-off retention mechanism (also known as bind-elute). Analyses of mAbs and immunoconjugates (ADCs) could thus be performed in only a few tens of seconds.

The second approach is based on the use of special gradient conditions, which greatly improve the selectivity between different protein isoforms. Based on the on-off retention mechanism, it is possible to add one or more isocratic steps during the RPLC analysis to increase the distance between the chromatographic peaks. Using this principle, we were able to achieve infinite selectivity between protein chromatographic peaks simply by adding a suitable isocratic step. We have also tested the possibility of using negative gradients instead of an isocratic step to achieve even greater selectivity between protein isoforms.

KN2-3 Novel Printing Strategies to Underpin Quantitative Imaging

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The availability of suitable calibration standards for quantitative imaging remains a challenge. For LA-ICP-MS in particular, whilst several methods for manual preparation of in-house calibration standards have been reported, there is still a lack of stable, bio-compatible and well characterised calibration standards or reference materials for use by the bio-imaging community. This makes the achievement of comparable data between different laboratories a very challenging task. The feasibility of gelatin-based standards has been demonstrated for matrix-matching calibration in tissue and cell elemental imaging. In this vein, cryosectioning of calibration standards remains a classical preparation method which is well described in the literature and has been applied to a variety of laser ablation methods and biological systems. However, the reproducible preparation of gelatin standards using cryosectioning requires careful optimisation and sophisticated technical skills that cannot be easily

standardised or transferred between laboratories. More recently, the automated production of calibration standards as micro-droplets has been reported for the quantification of multiple elements, added as ions, via full standard ablation [1]. The promising features of this approach in terms of throughput, reproducibility flexibility of standard preparation and application to biological samples has opened a new door towards innovative ways of standardisation for quantitative elemental bio-imaging.

This lecture will discuss the feasibility of a novel strategy based on the combination of nanoparticle characterisation workflows, bioprinting nano-doping technology and LA-ICP-ToF-MS analysis for the automated preparation and characterisation of gelatin-based calibration standards spiked with nanoparticles of potential use in biomedicine. The features of the bio-printing based approach will be compared against conventional manual cryo-sectioning standard preparation, in terms of throughput, between batch repeatability and standard homogeneity (at 5 µm spatial resolution). The use of NPs rather than elemental ions helped increase the potential use of the bioprinted standards with bio-imaging techniques other than LA-ICP-MS to obtain quantitative data of relevance to clinical diagnosis. An example of application of the developed strategy to underpin development of a model for the quantification of Au nanoparticles combined with surface-enhanced Raman scattering (SERS) reporters [2], as used for bio-marker imaging of single cancer cells, will be presented.

[1] Schweikert, A, Theiner, S., Wernitznig, D., Schoeberl, A., Schaier, M., Neumayer, S., Keppler, B. K. and Koellensperger, G.; Anal. Bioanal. Chem. 2022, 414, 485-495
[2] A. A. Leventi, K. Billimoria, D. Bartczak, S. Laing, H. Goenaga-Infante, K. Faulds, and D. Graham, Anal. Chem., 2023, 95, 5, 2757–2764

KN2-4 Structure Elucidation of Iron Chelators Produced by Microorganisms

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Microorganisms produce iron chelators called siderophores that are a rich source for drug discovery or plant protective agents. Pyoverdines are a class of siderophores from fluorescent Pseudomonas members and consist of different peptide chains specific to each bacterial species. The structural elucidation and characterization of pyoverdines require comprehensive analytical methods as bacterial extracts are complex mixtures. Here, we present a high-throughput UHPLC-MS/MS pipeline and the application of ion mobility spectrometry to facilitate research in the field of medicine and agriculture. In a second part, we present the purification and full structure elucidation by NMR and MS/MS of chryseochelin A, a novel citrate-based siderophore secreted by three Chryseobacterium strains involved in plant protection.

K. Rehm, et al., Anal. and Bioanal Chem. 414 (2022) 2671. doi:10.1007/s00216-022-03907-w
K. Rehm, et al., Biometals 36 (2023) 19. doi:10.1007/s10534-022-00454-w
K. Rehm, et al., Metallomics 15 (2023) mfad008. doi:10.1093/mtomcs/mfad008

KN3-1 Modern designs of molecularly imprinted polymers for electrochemical sensing and analysis: Recent developments and future prospects

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Molecular imprinting technology, which forms molecularly imprinted polymers (MIPs), is a creative method that enables synthetic biorecognition gaps to imitate real biological derivatives like antibodies, receptors, enzymes, etc. [1]. After removing the target analyte, synthetic cavities enable the recognition and selective rebinding of the template. In this case, molecular imprinting technology offers biosimilar receptors with higher specific affinities and better stability than natural receptors and biomolecules [2]. Although stable and durable MIPs seem relatively easy to create to achieve maximum efficiency, some optimization parameters should be considered, such as appropriate functional monomer and crosslinker and optimal ratios between functional monomer, template, and crosslinker [3]. The optimization process can vary based on the polymerization technique (electropolymerization, photopolymerization, and thermal polymerization). In addition, the structure of the polymeric matrices and the type of bond contact between the template and the polymer are two important factors in MIPs [4]. It was reported that template monomer interactions are realized through non-covalent interactions such as van der Waals forces, hydrogen bonds, and dipolar interactions [5]. The unique feature of superior selectivity of MIPs enables them to be used in various fields. Among them, MIP-based electrochemical sensors have a significant place because, with MIPs, it is possible to overcome the lack of selectivity issue in electrochemical sensors.

MIP-based electrochemical sensors and miniature electrochemical transducers can detect target analytes in situ. Thanks to superior chemical and physical stability, low-cost manufacturing, high selectivity, and fast response, MIPs have become an interesting field recently [6]. The studies on electrochemical MIP-based sensors to identify pharmaceuticals, heavy metals, hormones, enzymes, and biomarkers have grown. Moreover, without requiring time-consuming preparation procedures, these sensors have been successfully used in biological fluids (serum and urine samples) and pharmaceutical samples. In addition, various parameters such as detection limit, linear concentration range, and excellent recovery results showing its applicability to real samples to be analyzed are also indicators of the superiority of the developed sensor [7-8]. Therefore, there has been an increasing number of recent studies on determining different analytes using MIP-based electrochemical sensors.

[1] J. Pan, W. Chen, Y. Ma, G. Pan, Molecularly imprinted polymers as receptor mimics for selective cell recognition, Chem. Soc. Rev. 47 (2018) 5574e5587. DOI: https://doi.org/10.1039/C7CS00854F
[2] L. Uzun, A.P.F. Turner, Molecularly-imprinted polymer sensors: realising their potential, Biosens. Bioelectron. 76 (2016) 131e144. DOI: 10.1016/j.bios.2015.07.013
[3] K. Graniczkowska, M. Pütz, F.M. Hauser, S. De Saeger, N.V. Beloglazova, Capacitive sensing of N-formylamphetamine based on immobilized molecular imprinted polymers, Biosens. Bioelectron. 92 (2017) 741e747. DOI: 10.1016/j.bios.2016.09.083
[4] A.M. Mostafa, S.J. Barton, S.P. Wren, J. Barker, Review on molecularly imprinted polymers with a focus on their application to the analysis of protein biomarkers, TrAC Trends Anal. Chem. 144 (2021) 116431. DOI:10.1016/j.trac.2021.116431
[5] B. Sellergren, M. Lepistoe, K. Mosbach, Highly enantioselective and substrate-selective polymers obtained by molecular imprinting utilizing noncovalent interactions. NMR and chromatographic studies on the nature of recognition, J. Am. Chem. Soc. 110 (1988) 5853e5860. DOI:10.1021/JA00225A041
[6] S. Ramanavicius, A. Jagminas, A. Ramanavicius, Advances in molecularly imprinted polymers based affinity sensors (review), Polymers (Basel) 13 (2021). DOI: 10.3390/polym13060974

[7] A Cetinkaya, SI Kaya, EB Atici, ME Çorman, L Uzun, SA Ozkan, A semi-covalent molecularly imprinted electrochemical sensor for rapid and selective detection of tiotropium bromide, Analytical and Bioanalytical Chemistry 414 (28), 8023-8033. DOI: 10.1007/s00216-022-04335-6
[8] I Sulym, A Cetinkaya, M Yence, ME Çorman, L Uzun, SA Ozkan, Novel electrochemical sensor based on molecularly imprinted polymer combined with L-His-MWCNTs@ PDMS-5 nanocomposite for selective and sensitive assay of tetracycline, Electrochimica Acta 430, 141102. DOI:10.1016/j.electacta.2022.141102

KN3-2 Emerging mycotoxins in the food chain: challenges and perspectives

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Since the discovery of aflatoxins in the 1960s, more than 300 fungal metabolites have been characterized as so-called „mycotoxins“. The respective structures are as divers as the modes of action, comprising cytotoxic, genotoxic, immunomodulatory as well as endocrine disruptive compounds. In 1985, the FAO estimated that about 25% of the global food crop are contaminated with mycotoxins. Since then, global climate change has impacted the occurrence spectrum of respective fungi and, at the same time, improvement in analytics has enabled milestones in sensitivity. Nowadays, estimations speculate up to a contamination rate of 90%, at least in trace amounts. In addition to the few regulated mycotoxins such as aflatoxins or ochratoxin A, now numerous additional secondary metabolites can be detected. For most of these potentially „emerging“ mycotoxins data on occurrence and/or toxicity are not sufficient yet for comprehensive risk assessment. Prominent representatives are Alternaria toxin, formed by black molds of the genus Alternaria, which occur ubiquitously and are able to grow under varying temperature and moisture condition as well as on a large diversity of substrates. Reports on the occurrence of Alternaria toxins comprise a broad spectrum of plant-based food commodities including grain and grain-based products, apples, oilseeds, sun flower oil and tomato products. Although Alternaria species are known to generate a spectrum of secondary metabolites, toxicological studies have focused so far predominantly on the commercially available toxins e.g. alternariol (AOH), its monomethyl ether AME, tenuazonic acid (TeA) and tentoxin (TEN) as single compounds. In 2011, EFSA performed the first risk assessment on Alternaria toxins, but could only evaluate these four toxins due to the limited amount of data. In April 2022, the European Commission published a recommendation of indicative values for AOH, AME and TeA in certain food commodities. However, Alternaria alternata is able to generate a broad spectrum of secondary metabolites with different activity profile. Thus, in native toxin mixtures, a complex overlay of biological activities might occur including immunosuppressive, endocrine disruptive and genotoxic properties. Besides TeA , TEN and the two major dibenzo-α-pyrones AOH and AME, Alternaria spp. may produce significant amounts of perylene quinone derivatives, potent DNA-damaging components in complex mixtures. So far, the occurrence of perylene quinones in food is still unknown. Taken together, Alternaria toxins comprise a spectrum of mycotoxins with different molecular targets. Data are accumulating, arguing for relevance of these compounds in the food chain.

KN3-3 Expanding the droplet microfluidic toolkit: Electrokinetic manipulation of droplet composition

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Introduction: Droplet-based techniques have had a profound impact in biotechnology, owing to an ability to perform rapid and massively parallel reactions in minute fluid volumes. However, once droplets are formed, their composition can be altered through limited functions including the addition of reagents through droplet merging, which increases droplet volume, and through in-droplet mixing. Further, while droplet contents can be measured through, there remains a need for more versatile methods to probe droplets without significantly altering their contents. In this presentation, we describe a suite of in-droplet electrokinetic methods including de-mixing, mobility-based separations, desalting, and “salting”. Finally, we will share initial results for the measurement of the ionic content of droplets.

Experimental Approach: In this study, a stream of water-in-oil droplets in a microfluidic channel is flowed between ion exchange membranes that provide ionic communication between the droplets and two electrolyte-filled auxiliary microchannels. Inorganic and small molecular ions are transported through the membranes in response to voltage bias applied across the auxiliary channels. The selectivity of the membranes for cations or anions and the polarity of the applied voltage are tailored to accomplish each of the desired functions, which differ in terms of their extraction or injection of anions and/or cations out of or into droplets. These processes are characterized by several methods including fluorescence imaging of the distribution of a charged fluorescent tracer, brightfield imaging during formation of an insoluble salt, and measurement of ionic current.

Results and Discussion: Our results indicate that charged fluorescent tracers are enriched and separated within the droplet when both membranes are cation exchange membranes (CEMs). When both a CEM and anion exchange membrane (AEM) are employed, a fluorescent tracer is extracted during "desalting". "Salting" is demonstrated by injection of silver (I) ions into droplets containing calcium chloride, which leads to the formation of silver chloride precipitate within the droplets. Both desalting and salting rates are quantified through monitoring of the ionic current that is passed during these processes. Finally, we leverage ionic current as a means of quantifying the ionic content of droplets.

Conclusions: These results demonstrate the versatility of ion exchange to manipulate and measure the contents of water-in-oil droplets. This work lays a foundation for understanding and controlling the rate and extent of desalting and salting. Ongoing efforts on this project seek to extend the capabilities of these techniques.

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KN3-4 Engineering biology to bring diagnostics to low resource areas

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Biosensor-diagnostics used in low and middle income countries (LMICs) are often imported from high income countries as a finished device or assembled locally from kits produced elsewhere. The result is that the devices are high cost when taken in the local context. The required biological reagents for a diagnostic are often the largest proportion of its total cost (typically 50-85%). Without requiring changes to the end measurement we have revisited the ‘unaffordable’ diagnostics and considered the challenge of end-to-end local manufacturing of the biosensor, redesigning the biological reagents for

direct integration into the final device or reagent kit. We will report on the bio-design that can be manufactured locally, with basic infrastructure. We have used synthetic biology and incorporated locally resourced materials targeted to easy local production in resource poor areas.

Taking a 'gene to diagnostic' approach, the engineered protein design will be discussed for a diagnostic platform that uses low cost multifunctional fusion enzymes for point-of-care diagnostics. The platform is demonstrated for engineered BST DNA polymerase fusion proteins which can be isolated on silica via a fused R5 silica-affinity peptide and used in nucleic acid diagnostics. Data from a clinical study of malaria are presented raising some new questions about primer base sequence in loop-mediated isothermal amplification (LAMP) performance, possibly as a result of different binding of phosphates of the dNTPs. In malaria testing, the limit of detection depended on Plasmodium species and primer set. For example, 1000 copies of P. knowlesi 18s rRNA could be detected with the P.KNO-LAU primer set, but even 10 copies of P. ovale 18s rRNA could be detected with the P.OVA-HAN primer set. The results are discussed in comparison with qPCR and sampling protocol and show that the Si-BST polymerase can be optimised to meet the WHO recommended guidelines. The principles have been applied to similar diagnostic challenges in other diseases (eg dengue, leptospirosis, covid-19).

Enzyme engineering has also been applied to a device for sarcosine determination in urine (as a marker of early-stage prostate cancer), while other enzyme fusions have been engineered with luciferase and, in a structure+function fusion, using silk-like proteins. Applied in distributed low cost green manufacturing, self-labelling single chain antibody (scFv) fusions have been produce for lateral flow devices.
We have the first step towards providing low cost diagnostics in resource poor areas, which could deliver a sustained improvement in healthcare, while also developing the local economy.

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https://doi.org/10.1002/elan.202000032
https://doi.org/10.1016/j.biomaterials.2018.12.003

KN4-1 Precision medicine: The rise of electrochemical biosensing at the molecular level

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Today's society pursues personalisation in material things as much as in medicine and nutrition, being aware that these latter can certainly have a decisive influence on our life expectancy and quality. This, together with the particular context in which we find ourselves, shaped by the experience of an unprecedented pandemic, Brexit, the Russian invasion of Ukraine and rising inflation, makes the dream of re-imagining research and the implementation of precision medicine more important than ever. Advancing in these goals will guarantee all of us a longer and better quality of life, a more rational use of society's resources and the right and opportunity to participate in our own care.

And this will undoubtedly be fuelled by the development and application of disruptive technologies, such as electrochemical bioplatforms, which after the unprecedented evolution experienced in recent

years, have demonstrated tremendous versatility of design and use for the identification, determination and evaluation of the individual or collective clinical potential of new molecular markers of different omic profile, in an affordable, simple, fast, sustainable and point-of-care manner.

With all this in mind, this lecture will critically and concisely discuss the most remarkable features and opportunities of some selected electroanalytical biotools, recently proposed by our research group and collaborators, to advance the research and implementation of precision medicine in cancer, Alzheimer's and COVID-19. Two paths have been followed, On the one hand, the reliable interrogation of candidate molecular markers at different omics levels in clinical samples and, on the other hand, identify and confirm the diagnostic value of new molecular signatures of autoantibodies.

In particular, disposable bioplatforms that exploit ingenious assay formats and amplification strategies that can be applied in any environment for the multiplexed determination of miRNAs, methylations and single point mutations in nucleic acids, and antibodies or autoantibodies against antigens of different nature (viral, circulating or exosomal tumour antigens, peptides and proteoforms) identified by directed proteomics and produced by attractive technologies (HaloTag, phage display and targeted mutation) will be highlighted. All these bioplatforms demonstrate translational compatibility with the clinic and society in terms of simplicity, sensitivity and reliability, competitiveness with available methodologies in terms of sustainability, multiplexed and/or multi-omics character, affordability and applicability in any environment and by any user, and have faced with very promising results the analysis of selected patient cohort samples.

KN4-2 Analytical advancements in speciation analysis to explore trace element cycling in the environment

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Trace elements (metals, metalloids, non-metals) play a major role in human and animal health. Certain trace elements are toxic, even at very low concentrations. Other trace elements, referred to as micronutrients, are essential to humans and other organisms, however only in specific concentration ranges. There exists a close link between the abundance of a given trace element in the environment, e.g., its concentration in the soil and plant system as well as in natural waters (ground- and surface waters), and elemental exposure/ biological uptake of these elements. However, for uptake by humans and other organisms, not only elemental concentrations are relevant, also the form in which a given element occurs, which is referred to as its chemical speciation, is important know. With respect to trace element speciation, in past research in environmental geochemistry, identifying and quantifying inorganic speciation has been a main focus; however, in many environmental systems, trace elements occur in organic forms and these remain largely unexplored due to the challenges posed by their analysis. Furthermore, it is important to not only focus on speciation in specific compartments but to also understand processes affecting speciation across compartments, e.g., at and across their interfaces, to better understand biogeochemical element cycling on a broader scale.

In this talk, I will present how advancements in analytical chemistry help elucidate the inorganic and organic speciation of health-relevant trace elements and therefore improve knowledge of their environmental processes and pathways. Examples will be given on how trace element analysis via liquid and gas-phase chromatography coupled to inductively coupled plasma mass spectrometry, in combination with further analytical methods gives insight into trace element cycling in and across

terrestrial, marine and atmospheric environments. Finally, it will be discussed how modelling efforts can be combined with laboratory and field-based biogeochemical analyses to obtain a more complete picture of cycling of trace elements and their environmental distributions.

KN4-3 High-Throughput Quantification and Classification of Nanoparticles and Microparticles with Single Particle ICP-TOFMS

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Single-particle inductively plasma time-of-flight mass spectrometry (spICP-TOFMS) is used to analyze mixtures of nano- and micro-particles from a wide range of environmental sample types. With spICP-TOFMS, researchers aim to classify anthropogenic particle fractions based on multi-element signatures and to record the particle-mass (i.e. size) distributions and number concentrations of diverse particle types. However, the accuracy of these measurements requires an understanding of the fundamental structure of spICP-TOFMS data and the development of consistent approaches to detect, classify, and quantify particles in natural samples. In this regard, improvement to the accuracy and robustness of calibration methods for spICP-MS and the development of statistics-based methods to interpret recorded single-particle signals are critical.

In this presentation, I will discuss how the use of an online microdroplet calibration approach improves the accuracy of quantification of element masses in particles and particle-number concentrations. I will demonstrate that online microdroplet calibration offers high-throughput, matrix-independent calibration of metal-containing nanoparticles and microplastics. With accurate determinations of the mass amounts of elements in individual particles, spICP-TOFMS data can be used to classify particle types (i.e. from anthropogenic or natural sources) at the single-particle level based on multi-element compositions. To accomplish this classification, we have developed a suite of data analysis tools, including particle-type detection limits (1), hierarchical clustering analysis (2), and semi-supervised machine-learning classification approaches for the identification and classification of particles with heterogeneous elemental compositions. I will discuss our classification methods based on two case studies involving the analysis of cerium-rich and titanium-rich anthropogenic and natural particles. Through the lens of these studies, I will examine the current state of the art of spICP-TOFMS as a tool to classify and quantify diverse nanoparticle types.

(1) S. E. Szakas, et al. Environ. Sci.: Nano 9 (2022) 1627. DOI: /10.1039/D1EN01039E
(2) Mehrabi, K., et al. Environ. Sci.: Nano 8 (2021) 1211. DOI: 10.1039/D0EN01066A

KN4-4 Environmental metabolomics for unraveling the toxicity mechanisms of metals and nanoparticles in phytoplankton species

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The important advances the omics- techniques, together with chemometric statistical tools opened novel avenues in uncovering the possible contaminant-induced effects in complex environmental settings. Metabolomics, the youngest among -omics technologies, characterizes low-molecular-weight

molecules involved in different biochemical reactions and provides an integrated assessment of the physiological state of an organism. In the present talk, the potential of the liquid chromatography - based targeted metabolomics to uncover the contaminant-induced effects in the aquatic microorganisms will be illustrated at the specific case of the phytoplankton species exposure to nanomaterials (nanosilver and nanotitania) and mercury species. The green alga Chlamydomonas reinhardtii, diatom Cyclotella meneghiniana and brown yellow alga Poterioochromonas malhamensis were selected as representatives of freshwater phytoplankton species. The results revealed that in all cases the abundance of metabolites involved in various pathways corresponding to amino acid, nucleotides, fatty acids, tricarboxylic acid cycle (TCA), and antioxidant metabolism was altered in various treatments. The metabolomics results correlate well with the physiological results and confirmed that (i) oxidative stress is a major toxicity mechanism for nTiO2 exposure [1]; (ii) dissolved Ag released by nAg seems to be a major toxicity driver, even though nAg is internalized in the food vacuoles of P. malhamansis. However, nAg play an important role in the perturbation of amino acid metabolism, TCA cycle and oxidative stress [2]; (iii) metabolic perturbations in the green alga [3] and diatom were common for inorganic and monomethylmercury treatments, however the intensity of the perturbations was mercury species and phytoplankton species dependent. These results demonstrate the value of environmental metabolomics as a tool for understanding the molecular basis for these metabolic and physiological changes, and to detect early on metabolic changes that can later express themselves physiologically.

[1] Liu, W., Li, M., Li, W., Keller, A.A., and Slaveykova, V., 2022, Metabolic alterations in alga Chlamydomonas reinhardtii exposed to nTiO2 materials: Environmental science. Nano, v. 9, no. 8, p. 2922-2938.
[2] Liu, W., Majumdar, S., Li, W., Keller, A.A., and Slaveykova, V., 2020, Metabolomics for early detection of stress in freshwater alga Poterioochromonas malhamensis exposed to silver nanoparticles: Scientific Reports, v. 10, no. 20563.
[3] Slaveykova, V., Majumdar, S., Regier, N., Li, W., and Keller, A.A., 2021, Metabolomic Responses of Green Alga Chlamydomonas reinhardtii Exposed to Sublethal Concentrations of Inorganic and Methylmercury: Environmental Science & Technology, v. 55, no. 6, p. 3876-3887.

KN5-1 Single Molecule Electrochemistry: From electrochemically modulating single molecule fluorescence to counting single proteins for quantitative analysis

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The electrochemistry of single molecules is one of the frontiers in electrochemistry. The challenge in exploring single molecules is getting a signal from that molecule. There are three common approaches to doing this which are 1) monitoring single or a few electrons, 2) converting electrons into photons or 3) using a single molecule to modulate the flow of charge in an electrochemical system as performed with nanopore sensors [1]. In this talk we will present our findings using the latter two strategies.

In the first part of the presentation we will discuss how we can use total internal fluorescence microscopy (TIRF) to follow the fluorescence of single Alexa Fluor-647 labelled bovine serum albumin molecules adsorbed onto indium tin oxide (ITO) electrode surfaces. What was observed was the fluorescence of the Alexa-647 could be reversibly modulated as a function of the potential applied to the ITO. The fluorescence intensity of the Alexa Fluor 647 decreased, or even disappeared, at negative potentials but returned to similar levels to open circuit potential when the potential was swept back positive [2]. An observed pH dependence in the fluorescence strongly suggested the involvement of

electron and proton transfer in the switching of the fluorescence. A mechanism for the potential modulating of fluorescence is shown. We then surveyed a variety of other fluorescent dyes and the switching behavior is correlated with molecular structure. The importance of this electrochemical control over the fluorescence of single molecules for the super-resolution light microscopy method, single molecule localisation microscopy will be discussed.

The presentation will then switch to using nanopores to detect single protein molecules in a blood [3,4]. This is achieved using a solid state nanopore that was modified for the protein, prostate specific antigen (PSA). Rather than using the traditional approach of having a protein translocate through the nanopore, an antibody-modified magnetic nanoparticles ((anti-PSA)-MNPs) is used that captures the PSA and brings it rapidly to the nanopore which is then blocked. Reversing the magnetic field removes (anti-PSA)-MNPs that have not captured PSA but not those that have captured PSA. This selection of (anti-PSA)-MNPs that have captured the analyte alleviates limiting non-specific effects and allows the sensor to work in whole blood where the detection limit is as low as 0.8 fM detection limit. We finish by showing how this nanopore blockade sensor could be converted to an optical readout [4].

[1]. J.J. Gooding, Angew. Chem. Int. Ed. 55 12958-12958 (2016).
[2]. S.J. Fan, J.E.A. Webb, Y. Yang, D.J. Nieves, V.R. Gonçales, J. Tran, G. Hilzenrat, M. Kahram, R.D. Tilley, K. Gaus, J.J. Gooding, Angew. Chem. Int. Ed. 58 14495-14498 (2019).
[3]. K. Chuah, Y. Wu, S.R.C. Vivekchand, K. Gaus, P.J. Reece, A.P. Micolich, J.J. Gooding, Nature Comm 10 2109 (2019).
[4]. Y.F. Wu, Y. Yao, S. Cheong, R.D. Tilley, J.J. Gooding, Chem. Sci. 11 12570-12579 (2020).

KN5-2 Trace metal monitoring in aquatic systems: emphasis on the development and application of in situ metal bioavailability-oriented sensing tools

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The aquatic ecosystems are vital to the livelihood and food security of billions of people, and to the economic prosperity of most countries. Identification at appropriate temporal and spatial scale of chemicals from natural or anthropic sources that may have adverse effects on the ecosystem equilibrium and their living resources is thus crucial. Current research and developments focus on robust, easily usable, cost effective autonomous sensing tools that provide reliable in-situ measurements of key compounds. To reach these objectives, involved institutions have to tackle technical and analytical challenges.

This will be illustrated here by the development of innovative sensors and submersible sensing probes that enable autonomous in situ monitoring of trace metal species that are available for uptake by the microorganisms. Examples of in situ applications will be presented to reflect (i) the accuracy of these tools to record short-term subtle variations of the potentially bioavailable metal species, (ii) the potentiality they offer to identify abiotic and biotic processes that control the concentrations and cycling of the bioavailable trace metal species, and iii) assess the impact of climate change on trace metal biogeochemistry and (eco)toxicity. Submersible metal bioavailability-oriented sensing systems are also required to support the revision of European and International environmental quality standards (EQS) and guidelines (EQG) for metals that respect the chemical forms accessible to the biota to better manage metal impacts on aquatic ecosystems and ultimately human health.

Abdou, M., Tercier-Waeber, M.-L. New insights into trace metal speciation and interaction with phytoplankton in estuarine coastal waters. Mar. Pollut.Bull. 181, 2022, 113845. https://doi.org/10.1016/j.marpolbul.2022.113845.
Tercier-Waeber, M.-L., Confalonieri, F., Abdou, M., Dutruch, L., Fighera, M., Bakker, E., Schäfer, J. Advanced multichannel submersible probe for autonomous high-resolution in situ monitoring of the cycling of the potentially bioavailable fraction of a range of trace metals. Chemosphere 282, 2021, 131014. https://doi.org/10.1016/j.chemosphere.2021.131014.
Tercier-Waeber, M.-L., Stoll, S., Slaveykova, V. Trace metal behavior in surface waters: emphasis on dynamic speciation, sorption processes and bioavailability. Arch. Sci. 65, 2012, 119-142.

KN5-3 Clinical assays with paper, naked eye or camera: simplicity versus sensitivity?

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Since the introduction of the concept of microfluidic paper-based analytical devices (µPADs) by Whitesides and coworkers in 2007 [1], research targeting the development of paper-based analytical platforms has experienced continuous growth. Systems providing an optical signal output are particularly attractive since results can be obtained by the naked eye or the use of ubiquitous smartphone cameras. Nevertheless, the number of reports successfully demonstrating the application of such devices to practical clinical assays is still lacking behind. Different causes can be identified. First, paper-based analytical devices are generally praised for their simplicity and hence, their suitability for point-of-care applications. But in many instances, they still lack the user-friendliness expected from such type of systems [2]. Second, compared to clinical assays performed on large scale benchtop equipment, limits of detection and sensitivity are often inferior. For selected clinical applications, this is not an issue since targeted analytes are found in sufficiently high concentrations. But for others, this is not necessarily the case.

This talk will begin with shortly discussing some examples of paper-based clinical assays with optical/colorimetric signaling developed by our group over the past few years from aspects of simplicity and sensitivity. It will be shown that there are situations where assay operation and signal interpretation simplicity can be prioritized over sensitivity, since the latter is not an issue due to relatively high target concentrations in examples of urine analysis [3-5]. In another example, it is demonstrated that assay sensitivity and limit of detection of a known paper-based approach to urinary sarcosine analysis can be significantly enhanced by simple modifications without any reduction of assay simplicity [6]. The final part of the talk will introduce one of our latest efforts towards boosting assay sensitivity by adapting the CRISPR/Cas system to a paper-based analytical platform in the form of a simple, rapid, and highly sensitive detection method for non-nucleic acid targets by integrating CRISPR/Cas12a and an enzyme-linked immuno-sorbent assay (ELISA). For this purpose, secondary antibodies are labelled with DNA activating the trans-cleavage capability of the CRISPR/Cas12a complex. An origami paper-based device has been designed, where the detection of targets on the paper substrate is based on the monitoring of the fluorescence signal caused by the activated enzyme cleaving a probe ssDNA, which has been labeled by a fluorescent dye. The purpose of the origami approach is to keep assay operation as simple as possible.

[1] A.W. Martinez, et al., Angew. Chem. Int. Edit. 46 (2007) 1318. DOI:10.1002/Anie.200603817
[2] K. Yamada, et al., Lab Chip 17 (2017) 1206. DOI:10.1039/C6LC01577H
[3] R. Hiraoka, et al., ACS Sens. 5 (2020) 1110. DOI:10.1021/acssensors.0c00050
[4] K. Misawa, et al., ACS Sens. 5 (2020) 2076. DOI:10.1021/acssensors.0c00637
[5] S. Ohta, et al., Lab Chip 22 (2022) 717. DOI:10.1039/D1LC01060C
[6] M. Masumoto, et al., Anal. Bioanal. Chem. 414 (2021) 691. DOI:10.1007/s00216-021-03682-0

KN5-4 Vibrational Spectroscopy for Process Understanding

Katherine Bakeev

Vibrational spectroscopy has been an important analytical tool for chemical and structural analysis for decades, often used to confirm a newly synthesized compound, or otherwise identify unknown materials. Adoption of NIR, Raman and IR spectroscopy for process analysis gives a window into what is being created during the process in real-time. This use of Process analytical technology (PAT) opens the window for increased process understanding in terms of kinetics, formation of intermediates and also when a process may go off course. Increased process understanding through the development process can result in improved products and processes, as the details in real-time are revealed with in situ vibrational spectroscopy. Recent advances including time-gated Raman spectroscopy, enable rapid Raman spectral measurements with a minimized interference of fluorescence, while also providing fluorescence lifetime data.

Important aspects of PAT implantation will be discussed along with examples of NIR and Raman for polymorphic form conversion and biofermentation.

KN6-1 Conducting vial electromembrane extraction and development of generic methods

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Electromembrane extraction (EME) is a microextraction technique for ionic compounds, acids and bases [1]. EME involves mass transfer from an aqueous sample, across a liquid membrane and into an aqueous acceptor facilitated by an external electric field. The liquid membrane comprises 2-10 µL of organic solvent (membrane solvent) immobilized by capillary forces in the pores of a polymeric support membrane. The membrane solvent is immiscible with water, and therefore the liquid membrane is stable during extraction. The acceptor is a pure aqueous buffer solution.

The mass transfer in EME is controlled by the external electrical field. The negative electrode is located in the acceptor for extraction of bases, and the positive electrode is located in the sample. The sample and the acceptor are neutral or acidic, and in this way the bases are extracted as protonated species. For acids, the polarity is reversed, and the sample and the acceptor are neutral or alkaline. In such way, acids are extracted as deprotonated species.

The selectivity in EME is controlled by the chemical composition of the liquid membrane, the direction and magnitude of the electrical field, and pH in sample and acceptor. Sample volumes are typically from 0.05-10 mL, and EME may provide substantial pre-concentration. The acceptors are aqueous, and they can be injected directly in LC-MS. Thus, there is no need for evaporation and reconstitution.

EME was commercialized recently [2], based on vials produced in conducting polymer. In this system, the vials are used as electrodes and as containers for the sample and acceptor. For this technology to

be attractive there is a need for generic extraction conditions, and this is now under development. In this presentation, the operational principles of EME with conducting vials will be discussed, as well as the experiences and work with generic conditions.

[1] S. Pedersen-Bjergaard, K. E. Rasmussen, Journal of Chromatography A, 2006, 1109, 291-307.
[2] M. Drobnjak, F. Hansen, E. Leere Øiestad, T. Løvli, R. Trones, Ø. G. Martinsen, S. Pedersen-Bjergaard, LCGC North America, 2020, 38, 435-439.

KN6-2 Chemical uptake and potential health risks of using treated wastewater in agriculture: An analytical perspective

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Interest in reusing treated wastewater (TWW) is increasing, with the main driving forces being water scarcity, circular economy and environmental protection. To offset this problem, many scientists and professionals are looking towards moving from a traditional linear approach, where wastewater is an end-of-pipe issue, to a circular one, where TWW is a valued resource. However, such practices may create problems by introducing potentially hazardous chemicals into the food chain.

This presentation will address the uptake of chemicals of emerging concern (CEC), their effects on crop quality attributes, and potential health risks from consuming tomatoes produced using TWW. Accordingly, the uptake of 14 CEC and 27 elements was studied in tomatoes grown in soil-less (hydroponically) and soil-media (lysimeters) irrigated with potable and treated wastewater using GC-MS/MS, LC-MS/MS and ICP-MS. The quality of tomato fruits was evaluated by analysing their amino acid (AA) and volatile organic compound (VOC) profiles using GC-MS and HS-SPME GC-MS. In addition, essential nutritious elements were also assessed, along with the stable isotopic composition of light elements ($\delta^{13}C$, $\delta^{15}N$, $\delta^{34}S$) by IRMS.

Results showed that bisphenol S (BPS), 2,4 bisphenol F, and naproxen were present in fruits irrigated with spiked potable water and wastewater under both conditions, with BPS having the highest levels (0.034–0.134 µg kg−1 f.w.). The amounts of all three compounds were statistically more significant in tomatoes grown hydroponically than in soil. At the same time, elemental composition varied between tomatoes grown hydroponically and in the soil and tomatoes irrigated with wastewater and potable water. No health risks were estimated from consuming tomatoes grown hydroponically and in soil, based on CEC uptake and elemental composition alone. Hydroponically grown tomatoes in potable water had significantly higher concentrations of five AAs than those produced in spiked potable water, four of which were essential, suggesting that added contaminants might negatively influence the quality of tomatoes. Also, the aroma profile of over 140 VOCs and stable isotopic compositions ($\delta^{13}C$ and $\delta^{15}N$) differed between hydroponically and soil-grown tomatoes.

In summary, while our data highlight the influence of different irrigation regimes on chemical uptake and tomato quality attributes, additional studies are required to provide a conclusive answer regarding the safety of wastewater reuse for irrigation. These studies should also take into account potential CEC synergistic effects and the formation and effects of their metabolites and transformation products.

Kovačič, A. et al. (2022) Contaminant Uptake in Wastewater Irrigated Tomatoes, Journal of Hazardous Materials, 448, 130964. https://doi.org/10.1016/j.jhazmat.2023.130964.

KN6-4 Commercializing cell and gene therapies: A perspective from the analytical quality control function

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Cell and gene therapies can be highly curative. The response rate for advanced therapy medicinal products is unprecedented. The Lonza New Product Introduction and Lifecycle process for cell and gene therapies enables to achieve a robust commercially viable manufacturing and control process. The key modalities in cell and gene therapies each present unique challenges and opportunities also leading to a need for different analytical method requirements. Challenges in cell and gene include accelerated clinical development which requires faster timelines for chemistry, manufacturing and controls thus for analytical development. Manufacturing comparability evidence must be supported by fit for purpose analytical methods and linked to clinical evidence, respectively tailored potency assay methods allowing for mimicking clinical outcomes. Short shelf live of the products impose specific challenges to analytical methods. In this presentation, Lonza`s approach to development, industrialization, and delivery to cGMP is outlined from an analytical quality control perspective.

KN7-1 Universal electrochemical biosensor for all HIV types

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Human immunodeficiency virus (HIV) of human retroviruses are the causing agents of acquired immunodeficiency syndrome (AIDS).1 Since the start of the AIDS epidemic, according to the United Nations Joint Programme on HIV/AIDS, a total of 75.7 million people have been infected with HIV, and of which 32.7 million have died due to AIDS-related illnesses.2 In 2016, UNAIDS defined several targets for HIV testing and treatment to be achieved by 2020 to end the AIDS epidemic by 2030. Such milestones include the reduction of the yearly infection rate to 500,000 new infections/year, the reduction of yearly AIDS-related deaths to 5,000,000 deaths/year, and the 90-90-90 goal, requiring that 90% of HIV-infected individuals be aware of their status, of which 90% are receiving treatment, and of which 90% have undetectable viral loads.3 Although the targets were reached in a handful of countries, most low- and middle-income countries continue to fall behind the global goal in part due to decreases in funding and income disparities.4 To achieve these goals, extensive HIV testing is required, not only to diagnose new cases but in order to monitor the viral loads of those living with HIV.4, 5

When patients are placed on HIV treatment, termed anti-retroviral therapy, their viral loads are periodically monitored, using a quantitative nucleic acid test (NAT).5, 6 A limitation of current quantitative NATs is that there is no commercially available NAT assay that can accurately quantitate all subtypes and groups of HIV-1 and HIV-2,7-11 and it has also been reported that some assays underestimate the viral loads of patients infected with certain HIV subtypes due to a large number of primer-template mismatches.12 In regions such as Africa, where there is a wide diversity of circulating

HIV-1 subtypes, or West Africa where HIV-1 subtypes are present alongside HIV-2, the lack of a 'universal' HIV quantitative assay poses a major challenge to the implementation of viral load testing in those areas

Thus, we proposed the development of a cost-effective, sequence-independent, robust assay for the detection and quantitation of HIV, utilizing a modified nucleic acid sequence-based amplification (NASBA) protocol coupled to an electrochemical biosensor. Our modified NASBA reaction involves the addition of a tag on the 5' end of each amplicon, therefore the detection becomes independent of the viral RNA sequence itself. This protocol coupled with electrochemical four-way junction biosensors, which already proved a high selectivity even for a single base mismatch, becomes a promising technology.

1. Douek, D. C.; Roederer, M.; Koup, R. A., Emerging concepts in the immunopathogenesis of AIDS. Annu Rev Med 2009, 60, 471-84.

2. Global HIV & AIDS Statistics — 2020 Fact Sheet. https://www.unaids.org/en/resources/fact-sheet (accessed 09/22/2020).

3. United Nations Joint Programme on HIV/AIDS. 90–90–90 - An ambitious treatment target to help end the AIDS epidemic; 2017.

4. United Nations Joint Programme on HIV/AIDS. Executive summary — 2020 Global AIDS Update — Seizing the moment — Tackling entrenched inequalities to end epidemics; 2020.

5. United Nations Joint Programme on HIV/AIDS. The need for routine viral load testing - Questions and answers; 2016.

6. Clinical Guidelines Program: New York State Department of Health AIDS Institute. Viral and Immunologic Monitoring. 2019.

7. bioMeriéux; Organon Teknika NucliSensÒ HIV-1 QT; 2001.

8. Bayer Corporation VERSANT HIV-1 RNA 3.0 Assay (bDNA). https://wayback.archive-it.org/7993/20170111144019/http://www.fda.gov/BiologicsBloodVaccines/BloodBloodProducts/ApprovedProducts/PremarketApprovalsPMAs/ucm091214.htm (accessed 09/27/2020).

9. Abbott Laboratories Realtime HIV-1 Viral Load. https://www.molecular.abbott/us/en/products/infectious-disease/realtime-hiv-1-viral-load (accessed 9/27/2020).

10. Roche Amplicor HIV-1 Monitor Test. https://www.fda.gov/vaccines-blood-biologics/approved-blood-products/roche-amplicor-hiv-1-monitor-test (accessed 9/27/2020).

11. COBAS AmpliPrep/COBAS TaqMan HIV-1 Test, 48 Tests; COBAS AmpliPrep/COBAS TaqMan Wash Reagent, 5.1 L. https://www.fda.gov/vaccines-blood-biologics/approved-blood-products/cobas-ampliprepcobas-taqman-hiv-1-test-48-tests-cobas-ampliprepcobas-taqman-wash-reagent-51-l (accessed 9/27/2020).

12. Christopherson, C.; Sninsky, J.; Kwok, S., The effects of internal primer-template mismatches on RT-PCR: HIV-1 model studies. Nucleic Acids Res 1997, 25 (3), 654-658.

KN7-2 Do Biomolecules Retain their Native Conformation in the Gas Phase?

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Electrospray ionization (ESI) is capable of producing intact gas-phase ions from complex biomolecules, and using "native ESI", even noncovalently bound complexes survive, which would imply that biomolecular ions produced by native ESI are still in their native form in the gas phase. However, structural information of biomolecular ions in the gas phase is difficult to obtain: many methods give either only global information (e.g., ion mobility spectroscopy), or have serious limitations in terms of molecular size (e.g., cryogenic ion spectroscopy). The question whether biomolecular ions produced by native ESI really assume a native-like structure in the gas phase thus has not been answered conclusively.

We present a new, synergistic approach which utilizes Förster resonance energy transfer (FRET) of trapped gas-phase ions, ion mobility-mass spectrometry, and differential ion mobility spectrometry that employs microsolvation by an auxiliary gas, to provide multiple constraints for molecular modeling of gas-phase ion structures. In this context, we also developed a novel transition metal FRET method for measuring relatively short distances, 10-40 Å, between a donor dye and a noncovalently bound Cu2+ ion serving as a quencher, by measuring fluorescence lifetimes. Molecular dynamics simulations complement the experimental work.

Multiple systems are being studied with some or all of the methods mentioned above, including Ala-rich α helical polypeptides, peptides containing β hairpins, so-called "stapled" peptides, and cyclic peptides with -S–S- bonds; both of the latter fix the gas-phase conformation to some degree. The influence of the length of the molecular linker between the peptide scaffold and the dye moiety is also subject of ongoing research. In some cases, seemingly contradictory results were obtained that highlight the problem of relying on only a single method for deriving gas-phase structures. These apparent contradictions could be resolved by an extensive search of the conformational space of simulated structures, which identified those that satisfied all the experimentally determined constraints as the most probable gas-phase conformations.

P. Tiwari et al., J. Am. Chem. Soc. 143 (2021) 11291.
DOI: 10.1021/jacs.1c01915
R. Wu et al., J. Am. Chem. Soc. 144 (2022) 14441.
DOI: 10.1021/jacs.2c02776
R. Wu et al., Nature Commun. (in press)
DOI: 10.1038/s41467-023-38463-z

KN7-3 Glimpses into an Analytical Chemistry Textbook of the Future

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Daniel Harris's Quantitative Chemical Analysis textbook has been the "gold standard" in the field for four decades a balance of rigor and humor in a discussion that delivers the fundamentals while keeping abreast of current developments in the field.

But textbooks are evolving. The next edition of Harris [and some other guy] will be "digital first". This presentation overviews some of the features that a digital-first textbook offers, such as flow formatting, hyperlinked solutions and videos, and interactives.

But the field of analytical chemistry is also evolving, with biochemical reagents and applications becoming more prevalent, and vibrational spectroscopy and chemometrics [to name only two techniques] becoming increasingly common. This presentation will share plans for incorporating some of these developments, and my regrets and reasons for not incorporating others.

Finally, who does analytical chemistry is also evolving. Strategies, tactics and challenges for incorporating diversity, equity and inclusion within the textbook will also be shared.

KN7-4 Open droplet arrays for multimodal analysis at high throughput

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Droplet microfluidics is a powerful method to encapsulate, manipulate and analyze individual cells at high throughput. Most assays in nL droplets, however, are based on fluorescence spectroscopy, which limits the choice of assays and multiplexing capability. Mass spectrometry, on the other hand, allows for label-free detection and identification of multiple components. Recently, we have interfaced droplet microfluidics with matrix-assisted laser desorption ionization mass spectrometry (MALDI-MS). Instead of the standard MALDI targets to deposit the analytes in 384 wells, we have fabricated custom-made, transparent, indium-tin oxide coated targets, on which thousands of aqueous nL-droplets reside on the surface, covered by fluorinated oil. After spotting droplets on the target, the droplets are stable for several days, and compounds can be added by further spotting runs. Analysis of the droplets is performed by optical and fluorescence microscopy as well as by means of a MALDI-MS imaging system (Bruker rapifleX).

In this presentation, the use and versatility of the method for various applications will be discussed. After validation of the platform, we first confirmed the use of the platform for protein analysis and determination of posttranslational modifications (1, 2). Next, we adapted the platform for cell analysis. For example, the biosynthesis of the enzyme phytase in yeast could be monitored by both fluorogenic assay as well as label-free by mass spectrometry (3).

Further advancements of the platform include the analysis of supernatant by droplet splitting, the production of chemical gradients and the use of hydrogels for embedding cells.
Finally, a small change in the design of the plates enables depositing the droplets in proximity. In these experiments, the interface of aqueous droplets and oil is covered by a lipid monolayer. Adjacent droplets connect and form a droplet interface bilayer, which is a simple model for cell and organelle membranes used for studies of molecule permeation (4) and studying reactions across lipid membrane-separated compartments (5).

(1) Küster et al. Anal. Chem. 85 (2013), 1285. DOI: 10.1021/ac3033189
(2) Küster et al. Angew. Chem. 54 (2015) 1671. DOI: 10.1002/anie.201409440
(3) Haidas et al. Anal. Chem. 91 (2018) 2066. DOI: 10.1021/acs.analchem.8b04506
(4) Bachler et al. Anal. Chem. 93 (2021) 5137. DOI: 10.1021/acs.analchem.0c04939
(5) Bachler et al. Comm. Biol. 3 (2020) 769. DOI: 10.1038/s42003-020-01489-w

INVITED TALKS

IT1-1 Assessing and minimising measurement artefacts in phosphorescence lifetime based sensing

Dmitri Papkovsky[1]
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Phosphorescence based optochemical sensors, mainly O2 sensors and respirometric assays, are actively used in biomedical research and industrial applications including food packaging and microbial safety, environmental monitoring, bioprocessing and live cell analysis. These sensors usually rely on phosphorescence lifetime measurements, which provide good analytical performance, ruggedness and stability, and the possibility of calibration-free operation. However, in certain conditions such sensors can fail and produce erroneous readings and measurement artefacts. Understanding the causes and mechanisms of such effects can help reduce their impact and optimise the sensor system and corresponding (bio)assays.

Recently, we have conducted several studies, in which we have assessed and compared:
a) phosphorescence lifetime measurements of solid-state Pt-porphyrin based O2 sensors on four different detection platforms [1];
b) performance of commercial solid-state sensor coatings and soluble probes in respirometric assays with enumeration of bacterial cells [2];
c) six different types of Pt-porphyrin based soluble O2 probes in respirometric bacterial cell assays with complex samples and media.
In the last two cases the sensors/probes were challenged with harsh conditions using complex samples and assay media [3].

In each study, a number of interfering effects and factors have been revealed, linked to the format of the sensor, type of sample, medium and additives used, detector type, its settings, method of lifetime determination and/or algorithm used. This knowledge was used to optimise these sensor systems for reliable operation with complex samples, such as crude homogenates of food samples, colored media and environmental samples, dense cell cultures, with maximal accuracy and precision.

References:
[1] Sen R et al., Sens Actuat B., 2022, 132496.
[2] Elisseeva S, et al., Sens Actuat. B. 2023, 383, 133582.
[3] Papkovsky DB, Kerry JP, Sensors, 2023, 23: 4519.

IT1-2 HPLC and cylindrical PAGE purification of RNA aptamers with single nucleotide resolution

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Separation of RNAs is generally required for not only using RNAs as probes and therapeutics, but also for studying RNA structure and function. This is because either enzymatic transcription or solid-state synthesis of RNA generally produces a heterogeneous mixture. RNA heterogeneity refers to difference

in length or size (i.e., the number of nucleotides or nt), sequence, and/or alternative but coexisting conformations. Ion-pair, reverse-phase high performance liquid chromatography (HPLC) is a standard analytical technique for separating, purifying and analyzing RNAs. However, a single-nucleotide resolution by using HPLC is currently limited to RNAs shorter than 25 nt. Here we describe two methods for RNA detection and a large-scale purification. First, we have established a method of separating three RNA aptamers with 57, 58 and 59 nt on ion-pair, reverse-phase HPLC by a single-nucleotide resolution. Furthermore, our method allows us to resolve two structurally different, yet sequence or mass identical 59-nt RNA aptamers. These RNAs are isolated from a large RNA library (~1014) and for inhibiting AMPA receptor activity. These RNA inhibitors or aptamers are potential drug candidates for treatment of neurological diseases, such as amyotrophic lateral sclerosis (ALS). We have also established that the optimal condition to achieve single-nucleotide resolution correlates to 50 ºC and zero magnesium concentration in mobile phases. The ion-pairing agent, the buffer and the solvent we use are also compatible for post-HPLC analysis such as mass spectrometry. The second method we have developed is electrophoresis on a cylindrical polyacrylamide gel (PAGE) for a large-scale RNA purification without the use of any RNA staining dye. To demonstrate the utility of this method, we have used two size-identical (i.e., 59-nt in length), yet structurally different RNAs. Using this cylindrical PAGE, we show that even two closely running, structurally different but sequence identical RNA species can be cleanly resolved and separately collected. An overall recovery yield of ~80% in our experiment is achieved, comparable with the value by the use of conventional slab PAGE. However, a cylindrical polyacrylamide gel has a larger loading capacity or higher throughput (weight/time). Once a gel is properly cast, it can also be used 3-4 times. The second method we have established provides a practical way to prepare a pure RNA sample in large quantity. This may be especially useful when the RNA is intended for in vivo use as a potential therapeutic.

IT1-3 Exploring the Versatility of X-ray Techniques for Nanoparticles Characterization and Quantification

Laura Torrent Fàbrega[1], Hang Xiang[1], Kevin Kilchhofer[1], K.Y. Cheung[1], P.A. Alpert[1], David Baudouin[1], Mònica Iglesias[2], Eva Margui[2], Manuela Hidalgo[2], Frédéric Vogel[1], Markus Ammann[2], Christian Ludwig[1,3]

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X-ray techniques have emerged as versatile tools with the ability to reveal concealed aspects across diverse scientific domains. By leveraging X-ray techniques, valuable insights can be obtained regarding material behavior, elemental composition, and particle dynamics. Providing a glimpse into the potential of X-ray methods, this contribution highlights their diverse applications in two distinct areas: the fate of nanoparticles (NPs) in the environment, and in hydrothermal processes.

Investigating the interaction of silver nanoparticles (AgNPs) with soils by kinetic adsorption experiments, total reflection X-ray fluorescence spectrometry (TXRF) demonstrated to be a valuable alternative to commonly used techniques (e.g. inductively coupled plasma optical emission spectroscopy). Through the evaluation of sample preparation and calibration strategies, the quantification of AgNPs in solid and aqueous samples was obtained. In addition, the retention of AgNPs on soils was quickly achieved, providing valuable insights into their environmental behavior. Furthermore, a methodology combining cloud point extraction (CPE) and TXRF analysis allowed improving the limits of detection and offered a complementary approach to distinguish dissolved and particulate silver, ensuring accurate quantification [1,2].

In the realm of hydrothermal processes, scanning transmission X-ray microscopy coupled with near-edge X-ray absorption fine structure (STXM/NEXAFS) unravels hidden complexities. By studying carbonaceous particles produced during a waste-to-fuel conversion process, valuable information regarding their composition and properties could be obtained. The analysis of C K-edge spectra revealed the formation of carbonaceous "coke" through distinct carbon functional groups. The investigation of O K-edge spectra unveiled heterogeneity and provided insights into the presence of salts. Additionally, the identification of calcium phosphates and oxides through Ca K-edge spectra consolidated the findings from elemental analysis.

These case studies exemplify the broad utility of X-ray characterization techniques in diverse applications. By leveraging the power of X-rays, scientific community can gain a deeper understanding of the fate of nanoparticles in environmental systems and uncover crucial insights into hydrothermal processes. These findings highlight the versatility of X-ray based methods, which contribute to optimizing energy production, safeguarding the environment, and advancing scientific knowledge.

[1] L. Torrent, et al., Spectrochim. Acta Part B 126 (2016) 71-78. DOI: 10.1016/j.sab.2016.10.019.
[2] L. Torrent, et al., J. Anal. At. Spectrom. 33 (2018) 383-394. DOI: 10.1039/C7JA00335H.

IT1-4 Localizing N-glycan Changes in Aging Skin by MALDI FTICR MS Imaging

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In skin the observable effects of aging are characterized by changes in the dermal structure and its cellular composition. Underlying effects can be considered manifold. The contribution of protein glycosylation has so far not been studied in detail. Available data from total N-glycan analysis hint to the fact that in aging skin tissue, decreased levels of mannose and an increased presence of sialylated N-glycans are observed. Quantitative changes were reported for isome complex and hybrid N-glycan structures in the stratum corneum, whereas there seems to be no to only moderate changes in high mannose structures. We present the first spatially resolved study of N-glycan distributions in skin biopsies from both young and old human donors using MALDI FT-ICR MS imaging to get more detailed information on the availability of different glycan structures in the different skin layers.

Formalin-fixed, paraffin-embedded (FFPE) human skin-tissue longitudinal sections were cryosectioned, dewaxed by washing with various organic solvents, and antigen retrieval was performed in an automated heat-induced epitope retrieval device (2100 Antigen Retriever) with a citric buffer solution (pH 6). A linkage-specific sialic acid derivatization step was included to stabilze this sensitive glycan end group. PNGase F was sprayed on the tissue, followed by overnight digestion before covering the tissue with CHCA as MALDI matrix. MALDI MSI measurements were performed on a Bruker 7T scimaX FT ICR instrument at lateral resolutions of 10 and 40µm.

We present the identification and distribution of various N-glycans in skin sections and their relative variation between young and old human donors. Adaptation and fine tuning of available protocols for in-situ imaging of N-glycans in skin by MALDI MSI were essential due to the fragile nature of the samples. In detail, the choice of supporting materials, cryostat sectioning thickness settings, enzyme incubation periods, and matrix deposition parameters were optimized to ensure successful and reproducible results. Skin sections were analysed at 40 µm lateral resolution (survey) while smaller areas were investigated at 10 µm to distinguish fine details of in-situ N-glycan distribution. N-glycan

identification was confirmed by high mass accuracy measurements wherein the achieved mass error was below 3 ppm.

N-glycan distribution reveals noticeable differences between young and aged skin biopsies. Specifically, the former show an increased presence of specific N glycans in upper levels of the skin compartment (i.e., towards the stratum corneum), whereas in the latter the same glycans are distributed in deeper dermal layers.

IT2-1 POLYMERIC NANOFIBERS AS SENSORS – TOWARDS LAB ON A MAT

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Electrospun polymer nanofibers offer significant advantages as analyte sensitive layers. The nanofibers format offers the possibility of a significant extension of the surface area of the probe while minimizing the thickness of the receptor layer. This opens new possibilities to tailor performance of sensors requiring analyte transfer through solution/ probe interface and within the probe bulk. Thus nanofibers mats used a receptors in e.g. ion-selective sensors offer improvements in terms of analytical performance of devices based on classical analyte recognition principles. Moreover application of nanofibers opens a possibility to propose new devices to follow concentration changes of analytes for which ionophores are not available. Nanofibers format receptors were applied to improve optical as well as electrochemical sensors.

For ion-selective optical sensors close to 2D optode configuration obtained due to surface coverage of inert nanofibers with liquid receptor [1], results in a sensor free from limitations related to analyte transport in the receptor phase. The unique properties of polymeric nanofibers related to high surface to volume ratio allow application of these systems to prepare sensors for e.g. organic liquids dispersed within the aqueous phase. Interactions of organic solvents with nanofibers results in solubilization of dyes giving rise to optical signal [2]. For electrochemical conducting polymers based sensors nanofibers allow bulk transformation of the polymer upon contact with analyte – the effect that is not achievable for the same material in film format.

1 Baranowska-Korczyc at al. Analyst, 2019, 144, 4667
2 Kaczmarczyk et al. Electroanalysis https://doi.org/10.1002/elan.202200496

IT2-2 How to optimize SFC-MS methods effectively using current

state-of-the art instrumentation

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The popularity of SFC-MS is steadily growing due to important advances in instrumentation and complementarity to other chromatographic techniques. Despite its popularity, there are still several

issues that need to be taken in account. Indeed, the coupling of supercritical fluid chromatography and mass spectrometry is not straightforward compared to LC-MS due to inherent properties of CO2 used as a main component of SFC mobile phase. Although CO2 enhances evaporation during atmospheric pressure ionization (API), a density drop responsible for the decrease in solvating power and precipitation of analytes is observed after decompression in API ion sources. As a consequence, the chromatographic performance may be lost, and sensitivity may be compromised. To prevent these issues, dedicated interfaces are needed in SFC-MS, such as pre-BPR splitter with sheath pump or BPR with sheath pump without splitter. A sheath pump delivering a make-up solvent is an inherent component of both interfaces. Indeed, both the make-up solvent and the mobile phase composition have an important effect on MS response and thus require careful optimization. Despite numerous published studies on SFC-MS, this coupling usually relies on tedious and time-cosuming experimental step-by-step optimization.

In our study, we evaluated the effect of the make-up solvent composition using different types of current state-of-the-art instrumentation including different ionization techniques and mass analysers. A large set of compounds with different physicochemical properties was employed to cover a wide range of applications and to evaluate different ionization techniques. Among them, electrospray ionization (ESI), atmospheric pressure chemical ionization, and Unispray, all in the positive and negative modes, were evaluated. Using ESI, the differences needed to take in account among various MS platforms including single quadrupole, triple quadrupole, and quadrupole time-of-flight analysers were evaluated.

24 solvents, including methanol, ethanol, and isopropanol as neat make-up solvents, and methanol with additives, including ammonia, ammonium formate, ammonium acetate, formic acid, acetic acid, and water, all at different concentrations, were examined. All experimental data were used for the statistical calculations and creation of the prediction models using multilinear regression analysis. In the next step, the developed prediction models were tested on a set of blind probes not contained in the initial study set to prove the correctness of prediction of the optimal make-up solvent composition for target analytes.

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IT2-3 Biosynthetic trifluoromethyl (CF3) methionine labelling to probe structures and dynamics of virus coat proteins and molecular chaperone oligomers by 19F NMR spectroscopy

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This study explores the use of genetic incorporation of fluorinated chemical groups to high molecular weight protein complexes and novel fluorine-19 (19F) nuclear magnetic resonance (NMR) techniques to probe the behaviours of these biomacromolecules.

Since the initial development originated by Richard R. Ernst and improved by Kurt Wüthrich, NMR methodologies have been widely used to study chemical structures of biomolecules in atomic resolution. Nevertheless, detecting the structural changes of large macromolecules in complex

biological mixtures remains challenging for NMR. The increase in molecular size and structural complexity in a target substrate causes significant signal broadening and spectral crowding.

Here, to address these challenges, the author and his team used a site-specific trifluoromethyl (CF3) incorporation technique to produce CF3-labelled protein oligomers and 19F NMR techniques to study these biomacromolecules in high-resolution. Since fluorine is almost absent in natural biological systems, effective CF3 incorporation into a protein of interest produces 19F NMR spectra with clean backgrounds even if the target is in heterogeneous mixtures. Furthermore, the CF3-group has attractive properties as an NMR probe. For instance, it has three chemically equivalent fluorine atoms to enhance the signal intensity and also rapid rotation around the sigma bond, giving sharp 19F NMR signals.

To take advantage of these benefits, we produced recombinant proteins labelled with the CF3-methionine using genetically engineered bacterial cells, and successfully obtained the 19F NMR signals from the target sites in these fluorinated proteins. In particular, we targeted methionine residues in protein sequences to incorporate the CF3 NMR probe because the swap of a natural CH3 methyl group in a methionine side chain to a CF3 group would not massively alter the native protein structures. The series of solution-state 19F NMR measurements, including 19F chemical exchange saturation transfer (CEST) experiments, detected a very fast and complicated dynamic equilibrium in virus coat proteins. The retention of the large oligomeric self-assembly of chaperone protein complexes after the unnatural fluorinated amino acid incorporation was confirmed by comparing proton (1H) and 19F diffusion NMR data.

By using these unique fluorine labelling techniques and 19F NMR methodologies, we succeeded in analysing the macromolecular structures of high molecular weight protein complexes (virus coat proteins and molecular chaperone oligomers) with unprecedented accuracy and clarity. Overall, our work paves the way to apply the biomolecular NMR methodologies in the analytical probing of complex biological systems by combining state-of-art techniques in chemical biology and biophysics.

R. R. Ernst, et al., Rev Sci Instrum. 37 (1966) 93.
DOI:https://doi.org/10.1063/1.1719961
K. Wüthrich, et al., Nat Rev Drug Discovery. 1 (2002) 211.
DOI:https://doi.org/10.1038/nrd748
L. E. Kay, et al., Cell. 184 (2021) 577.
DOI:https://doi.org/10.1016/j.cell.2020.12.034
K. Wüthrich, et al., Science. 335 (2012) 1106.
DOI:https://doi.org/10.1126/science.1215802

IT3-1 APTAMER-BASED DETECTION OF EMERGING CANCER BIOMARKERS TO GUIDE CANCER DIAGNOSIS AND MANAGEMENT

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Early detection of cancer is a major clinical challenge, requiring coordinated efforts to achieve substantial progress. Much research has been directed at deciphering the molecular events that contribute to cancer initiation and progression. Consequently, new biomarkers are increasingly identified, serving as valuable diagnostic indicators that can help clinicians to detect, better understand

and predict the treatment effect heterogeneity in different patients. Among them, glycoproteins containing aberrant glycosylation patterns and extracellular matrix (ECM) components are key structural players in cancer pathogenesis. However, these emerging biomarkers are not easily translated to clinical practice due to the lack of analytical methods with the required sensitivity and selectivity for their detection in biological fluids. The development of new technologies capable of detecting these low abundant components, released by tumor cells and with subtle changes with respect to those released by normal cells, would provide researchers and clinicians new tools for early cancer diagnosis, supporting precision personalized medicine.

Towards this goal, we have obtained aptamers against the glycosylation site of glycoproteins [1] and ECM components [2], targeting specific sites on these molecules that are expected to be altered during tumorigenesis to increase the clinical specificity. Using these aptamers as specific receptors, we have developed aptamer-based electrochemical sensors for the rapid and convenient detection of these biomarkers, showing that they can be useful for diagnostic purposes in biological fluids [3,4]. Combined with traditional biomarkers, these tools would provide clinicians with reliable options for cancer diagnosis.

References:
[1] A. Díaz Fernández, R. Miranda-Castro, N. Díaz, D. Suárez, N. de-los-Santos-Álvarez, M. J. Lobo-Castañón. Chem. Sci. 2020, 11, 9402-9413.
[2] R. Lorenzo-Gómez, R. Miranda-Castro, J.R. de los Toyos, N. de-los-Santos-Álvarez, M.J. Lobo-Castañón, Anal. Chim. Acta 2022, 1189, 339206.
[3] A. Díaz Fernández, R. Miranda-Castro, N. de-los-Santos-Álvarez, M. J. Lobo-Castañón, P. Estrela. Biosens. Bioelectron. 2021, 175, 112872.
[4] R. Lorenzo-Gómez, A. Casero-Álvarez, R. Miranda-Castro, M. García-Ocaña, J.R. de los Toyos, N. de-los-Santos-Álvarez, M.J. Lobo-Castañón, Talanta 2022, 240, 123196.
Acknowledgments
The work has been financially supported by Spanish Government (projects PID2021-123183OB-I00 and PDC2022-133435-I00).

[1] https://doi.org/10.1039/D0SC00209G
[2] https://doi.org/10.1016/j.aca.2021.339206
[3] https://doi.org/10.1016/j.bios.2020.112872
[4] https://doi.org/10.1016/j.talanta.2021.123196

IT3-2 LC-MS Analysis of Antibiotics in Fermentation Medium

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Liquid Chromatography-Mass Spectrometry (LC-MS) is a powerful tool for the analysis of antibiotics in complex matrices. Antibiotics play a crucial role in various industries, including pharmaceuticals, agriculture, and biotechnology. However, in biotechnology, their effective quantification within fermentation media presents challenges due to the intricate matrix effects that can influence the accuracy and reliability of analytical results.

The presentation will focus on LC-MS method development and optimization for the analysis of Spectinomicyn in the fermentation medium. Various sample clean-up procedures, chromatographic column selection, and internal standardization approaches will be discussed in the context of matrix effect management.

By understanding and addressing matrix effects, analytical chemists and researchers can confidently achieve reliable and reproducible results, ensuring the safety and efficacy of antibiotics in various applications. A selection of appropriate LC-MS techniques and effective management of matrix effects will be essential for ensuring the success and sustainability of a fermentation process.

IT3-3 Laser Induced XUV Spectrometry (LIXS): Even Better Than the Real LIBS

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Laser ablation (LA) is a spatially resolved technique enabling fast sampling of any kind of matrix without sample preparation. The ability to measure important elements such as H, C, N, O or Li, Be, B, F, P, Cl, makes laser-induced breakdown spectroscopy (LIBS) complementary to established laboratory techniques such as X-ray Fluorescence spectroscopy (XRF) or Laser Ablation ICP-MS (LA-ICP-MS). LA-ICP-MS is very sensitive for quantitative analysis, while XRF is extremely specific and precise with calibration-free quantification. While poorer on the target analysis, LIBS offers a substantial potential for non-target qualitative analysis, if precision and specificity would improve consistency. Furthermore, although LIBS has the unique advantage to be operable in situ, i.e. in the field and/or in a low-pressure environment for space exploration, its susceptibility to the conditions limits its impact for heterogeneous materials.

Laser-Induced XUV Spectroscopy (LIXS) is similar to LIBS [1-4], but at a much shorter wavelength domain, the soft X-ray (10-100 nm) [3]. LIXS happens when the early laser-plasma is extremely hot and dense, giving selective prevalence to ion lines. These make the spectrum cleaner, stable and intense, with modest noise. The generation of a LIXS spectrum requires a high-fluence laser pulse, and a vacuum spectrometer for the short wavelength. The degraded resolving power at shorter wavelengths makes it generally difficult to collect a non-distorted (stigmatic) spectrum below 100 nm. We have addressed this technical challenge. The application of LIXS to characterize the heterogeneity of energy and valuable materials is discussed

Keywords LIBS; LIXS; precision; heterogeneity; energy materials, gemstones

References
[1] Qu, D., Bleiner, D. (2022). High-precision micro/macro-analysis with laser-induced XUV spectroscopy (LIXS) Chimia,76(1-2), 153.
[2] Qu, D., Ohannessian, N., Wyder, C., Trottmann, M., Wichser, A., Lippert, T., & Bleiner, D. (2021). High-precision mapping of fluorine and lithium in energy materials by means of laser-induced XUV spectroscopy (LIXS).Spectrochimica Acta B: Atomic Spectroscopy,181, 106214
[3] Bleiner, D., Juha, L., & Qu, D. (2020). Soft X-ray laser ablation for nano-scale chemical mapping microanalysis.Journal of Analytical Atomic Spectrometry,35(6), 1051-1070.
[4] Bleiner D, Qu D, Kraft K, Shlyakhtun O. Laser-induced XUV spectroscopy (LIXS): From fundamentals to application for high-precision LIBS. Spectrochimica Acta Part B: Atomic Spectroscopy. 2023 Mar 31:106668.

IT3-4 "Direct" Thorium-Lead dating of gem quality corundum by laser ablation ICP-TOF-MS

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The geographical origin of a natural gem quality corundum, i.e. sapphire or ruby, has an impact on its value, often quite significantly. Trace element content is one of the methods to help identifying the origin of a gemstone and laser ablation inductively coupled plasma time of flight mass spectrometry (LA-ICP-TOF-MS) is used in our laboratory to do it (Wang 2016). Acquiring the whole element mass spectra with an ICP-TOF-MS eliminates the chance to overlook a rarely occurring element. Getting an age of a gemstone by dating a surface exposed mineral inclusion like zircon (Link 2015) provides other evidence of the origin as sapphires and rubies from the Himalayan orogeny (Kashmir and Myanmar) have ages <50 million years (Ma) whereas those from the East African orogeny (Sri Lanka, Madagascar and East Africa) show an age of 500 to 800 Ma. Unfortunately, direct dating of corundum is very difficult (Krebs 2019).

However, our analyses over the years revealed that about 2% of the analyzed corundum gems have a seemingly homogenous Thorium concentration of >1 µg/g. Thorium is decaying with a half-live of 14.05 billion years to 208Pb providing the opportunity to date these gemstones. Although having relative uncertainties of tens of percent, vastly improving with higher Thorium concentrations, it is still possible to distinguish corundum with an age of e.g. 50 Ma and 500 Ma.

Closer inspection of the gemstones in question indicates that Thorium is likely present in very small inclusion of just a few micrometers in diameter or smaller. Elevated concentrations of light rare earth elements are found while Thorium is present but no correlation with other elements reported to form nano-inclusion in corundum (Baldwin 2017) like Nb, Ta, W and Ti were found. Indicating that Thorium is one of the major elements in the inclusion similar to the brockite inclusions reported by Guo et al.
A very rare case where a sapphire with ~3 µg/g Thorium resulting in an age of 571 +/- 30 Ma had also an exposed zircon inclusion with gave a 206Pb/238U age of 536 +/- 11 Ma (uncertainties given as 1s) indicating the accuracy of the method.

H. A. O. Wang, et al., J. Gemmol. 35 (2016) 212. DOI : 10.15506/JoG.2016.35.3.212
K. Link, J. Gemmol. 34 (2015) 692. DOI : 10.15506/JoG.2015.34.8.692
M. Y. Krebs, et al., Chem. Geol. 523 (2019) 42. DOI: 10.1016/j.chemgeo.2019.05.035
L. C. Baldwin, et al., Contrib. Mineral. Petrol. 172 (2017) 43. DOI : 10.1007/s00410-017-1362-0
J. Guo, et al., Contrib. Mineral. Petrol. 122 (1996) 368. DOI: 10.1007/s004100050134

IT4-1 A Disassembly Approach for Analyte Detection

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This contribution presents a stimulus-induced disassembly approach for fluorometric analyte detection. In particular, the selective fluorometric detection of pyrophosphate (PPi) and homocysteine (Hcy) with metal-imine complexes are described.

In the disassembly approach, the target analyte selectively sequesters a metal ion from a metal-chelate complex. The “unlocked“ ligand subsequently hydrolyses into its molecular subunits generating a detectable signal. Mechanistic studies yielded deep insights into the mode-of-action of these type of chemodosimeters and structural modifications alter significantly their selectivity, reactivity and stability. Therefore, discrimination between structurally related analytes is possible with optimized probes as demonstrated for the selective detection of PPi over other polyoxophosphates with an iron-

salen complex. Applications of the disassembly strategy for analyte detection (PPi, Hcy) in biological media and foodstuff are presented.

N. Kumari, F. Zelder, Chem. Commun. 51 (2015) 17170.
DOI: 10.1039/C5CC07413D
P. Yadav et. al. Inorg. Chem. Front. 8 (2021), 4343.
DOI: 10.1039/d1qi00209k
X. Li et. al. ChemistryOpen 11 (2022) e202200106.
DOI: 10.1002/open.202200106

IT4-2 Development of dipstick-type DNA biosensors for visual identification of olive cultivar origin

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Cultivar verification of olives and olive oil is of particular importance because their sensorial characteristics and nutritional properties depend on the cultivar. Monovarietal oils have become the center of interest for producers and consumers. Various DNA markers have been used for olive cultivar identification including genomic microsatellites, RAPD, ISSR, SCAR, AFLP and single-nucleotide polymorphisms (SNP) in combination with electrophoresis, microarrays or fluorescent microspheres and flow-cytometry. In this context, we have developed dipstick-type biosensors, for visual identification of olive cultivar. The method consists of the following steps: (a) DNA extraction, (b) A quadruplex PCR for simultaneous exponential amplification of four DNA sequences, two from the cycloartenol synthase (Cyclo1 and Cyclo2) and two from the lupeol synthase (Lup1 and Lup2) genes. The sizes of the amplification products, as confirmed by agarose gel electrophoresis, are 179, 170, 162 and 145 bp, respectively. (c) A multiplex primer extension reaction using 8 allele-specific primers. A primer is extended only if perfectly complementary to the interrogated sequence. During extension, biotin-modified dUTP and dCTP are introduced in the new strands. Characteristic oligonucleotide sequences (tags) are attached to the primers for subsequent capture and detection. (d) Aliquots of the extension reaction mixture are applied to the conjugate pad of two dipstick-type DNA biosensors, each enabling visual detection of 4 alleles of the cycloartenol synthase gene and 4 alleles of the lupeol synthase gene. Each biosensor consists of an absorbing pad, for immersion in the appropriate buffer, a conjugate pad, on which antibiotin antibody-conjugated gold nanoparticles are deposited, and a biosensing membrane containing 4 spots of immobilized anti-tag oligonucleotide sequences. As the running buffer flows by capillary action, the extension reaction products are captured via tag/anti-tag hybridization and detected through biotin/antibiotin interaction. A red spot is obtained only if the primer has been extended, thus denoting the presence of the corresponding allele. The combination of the red spots in the 8 positions of the two biosensors provides the characteristic genotype of the cultural variety of the olive or olive oil sample. The method was evaluated by using the following Greek olive varieties: Adramytini, Chondrolia, Kalamon, Gaidourelia, Tsounati, Koroneiki, Lianolia, Throumbolia and Kothreiki.

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IT4-3 Advancing measurements at nanoscale: analytical strategies to evaluate encapsulation efficiency, drug release and nanoparticles concentration

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The therapeutic index of drug-delivery nanoparticles (NPs) depends on the number of NPs reaching the target site, on the amount of drug carried by these NPs, and on their ability to release the loaded cargo when at the target site (S. Đorđević, 2022). The characterization of these attributes is, therefore, crucial during formulation development to determine whether it can proceed to clinical trials. Nevertheless, standard procedures to complete these evaluations have not been established yet. Moreover, the available methods for characterizing these attributes present limitations that often preclude accurate results. Thus, alternative analytical strategies are required for evaluating the encapsulation efficiency, drug release and concentration of drug-delivery NPs, for advancing NPs characterization and/or providing a means for result validation.

In this work, several contributions will be addressed, focusing in both polymeric and lipidic NPs. First, an automated strategy for monitoring in real-time NPs´ drug release and drug permeation through a skin model, implemented in a flow-based setup (A. Alves, 2016), will be shown and its potentialities discussed. The impact of ultrafiltration conditions when separating loaded NPs from unloaded molecules for encapsulation efficiency measurements (S. Marques, 2020), and the importance of adjusting the procedure in accordance with NPs properties, will also be discussed, along with an alternative strategy to ultrafiltration to separate loaded NPs from unloaded molecules and simultaneously quantify both fractions. Finally, an analytical methodology under the lab-on-valve platform for the automated measurement of NPs concentration in approximately 2 min will be presented, showing advantageous features, such as strict control of nanoparticles handling procedures and avoiding drug leaching as NPs dilution is limited (S. Marques, 2023).

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A. Alves, et al., Talanta 146 (2016) 369. DOI: 10.1016/j.talanta.2015.08.070
S. Đorđević, et al., Drug Deliv. Transl. Res. 12 (2022) 500. DOI: 10.1007/s13346-021-01024-2
S. Marques, et al., Molecules 25 (2020) 1879. DOI: 10.3390/molecules25081879
S. Marques, et al., Anal. Chem. 95 (2023) 4619. DOI: 10.1021/acs.analchem.2c04631

IT4-4 Development of Dried Milk Spots Sampling Method for Comprehensive Human Milk Composition Analysis: A Novel Analytical Approach for Global Health Studies

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Breast milk is acknowledged as vital for infant health and is recommended as the exclusive food source for the first six months of life. However, the associations between specific milk compounds and infant health remain understudied. Current studies are limited by small number of samples, which in turn is limited by conventional sampling methods. Regular sampling approaches are burdensome for participants which makes it difficult to have high sampling frequency; the necessity of cool chain logistics also restricts the geographical diversity of sample collection.

Paper-based sample collection techniques have been developed for blood sampling and have demonstrated significant advantages. These methods typically involve depositing a sample onto a piece of paper, allowing it to dry in open air for several hours, and storing it in a plastic bag. This drying process removes most of the water, reducing the risk of degradation and permitting samples to be stored at room temperature for weeks. Moreover, these samples occupy limited space and can be easily transported.

In this study, we evaluated the performance of commercial papers for human milk collection as dried milk spots (DMS) and determined they were unsuitable for storage and representative recovery of proteins. Consequently, we developed a wax-coated paper to collect breast milk as DMS. The milk would be dried on the wax coating, and during extraction with water/hexane, the wax layer would dissolve, liberating the milk proteins and allowing their dissolution in the water phase with limited retention in the paper itself. We thereby successfully maintained a consistent β-casein to α-lactalbumin ratio, the two most abundant human milk proteins, over four weeks of storage, based on analysis by HPLC-UV and HPLC-MS. Moreover, our preliminary data suggest the hexane fraction could be used to study the fatty acid composition (as methyl esters) using GC-MS. Finally, the aqueous fraction, after further clean-up, could reveal the oligosaccharide composition, when analyzed by MALDI-TOF. We are currently investigating whether less abundant proteins can also be detected, allowing for a more comprehensive relative protein composition analysis.

In summary, the DMS sample collection method offers a simple, safe, and reproducible alternative to current breast milk sampling techniques for clinical and epidemiological research. This novel approach aligns with the themes of analytical science and global health by enabling more extensive and accessible studies of breast milk composition and its impact on infant health.

[1] Alimoradi, F., Javadi, M., Barikani, A., Kalantari, N., & Ahmadi, M. (2014). An Overview of Importance of Breastfeeding. Journal of Comprehensive Pediatrics, 5(2). https://doi.org/10.17795/compreped-14028
[2] McDade, T. W., Williams, S., & Snodgrass, J. J. (2007). What a drop can do: dried blood spots as a minimally invasive method for integrating biomarkers into population-based research. Demography, 44(4), 899-925. https://doi.org/10.1353/dem.2007.0038
[3] Bravi, F., Wiens, F., Decarli, A., Dal Pont, A., Agostoni, C., & Ferraroni, M. (2016). Impact of maternal nutrition on breast-milk composition: a systematic review. The American journal of clinical nutrition, 104(3), 646-662. https://doi.org/10.3945/ajcn.115.120881

IT5-1 Low-cost Flexible Laminated Graphene Paper Solid-contact Ion-selective Electrodes

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We show in this work that flexible laminated graphene paper electrodes are a cost-effective alternative to rigid glassy carbon and platinum electrodes. Several identical graphene paper electrodes, which are suitable planar substrate materials for wearable and single-use ISEs, can be easily fabricated with a standard pouch laminator in a single lamination step in ca. 40 min to a price less than €1.[1]
Our results show that the flexible laminated graphene paper potassium-selective solid-contact ion-selective electrodes (K-SCISE) have a performance on par with the state-of-the-art SCISEs. The K-SCISEs had a close to Nernstian slope of 56.9 ± 0.1 mV and a reproducibility of the standard potential of ±4.4 mV that is typical for the SCISEs (n=6).

We have used polyaniline doped with with dinonylnaphthalene sulfonic acid as a model compound for the solid contact (SC) because of its solubility in organic solvents allowing for drop casting of the SC on the graphene paper substrate. The K-SCISEs had high selectivity against sodium, lithium, hydrogen, magnesium and calcium ions, and the electrodes showed neither water layer formation, light nor oxygen gas sensitivity. However, we observed a minor response to carbon dioxide (pH) because of the pH response of PANI.

As the lamination concept is universal, the conducting polymer and the PVC-ISM can be easily substituted with other materials that are compatible with the lamination process. Especially in wearable applications it is important that no delamination of the ISM or the SC occur that can cause electrode failure when they are repeatedly exposed to bending. We did not experience any delamination in this work with stationary K-SCISEs, but this must be certainly studied in more detail if the electrodes are used in practical applications.

The presentation also discusses shortly the lamination of Ag/AgCl pseudo-reference electrodes and fully laminated coated-wire electrodes.

1. M. Rutkowska, T. Lindfors, Z. Boeva, M. Strawski, Sens. Actuators B Chem., 337 (2021) 129808; doi.org/10.1016/j.snb.2021.129808

IT5-2 Testing the Chalcogenide Fe3+ Electrode in Seawater

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The lack of available iron (Fe) in seawater is widely accepted as a factor that limits phytoplankton growth in extensive high nutrient, low chlorophyll (HNLC) ocean areas, as well as some coastal upwelling regions. More recently, iron has been shown to impact the ecology and species distribution of stratified oligotrophic (ocean “desert”) regimes as well. Productivity (or lack thereof) at the scale of these iron limited areas is significant, as the microscopic plants involved support all marine life. The availability of a reliable Fe sensor would permit high resolution datasets to be obtained on a routine basis, resulting in improved understanding of global marine productivity. In this work we present initial tests of a chalcogenide Fe3+ ion selective electrode (Fe-ISE) in natural and artificial seawater, operated in a classic potentiometric mode and over a range of pH. Voltages are interpreted in the context of a chemical model of the system including a known dissolved organic ligand background in the case of

artificial seawater and a crudely characterized ligand background in the case of natural seawater. One objective of this work is to enhance our understanding of the complex chemistry that exists at the surface of the chalcogenide ISE in the presence of a heterogeneous dissolved organic matter pool, such as seawater. We also aim to develop an improved Fe-ISE methodology that is suitable for long term autonomous use in the marine environment.

IT5-3 Exploring the potential of laser ablation as a means of sample introduction for microplastics characterization via inductively coupled plasma-mass spectrometry operated in single-particle mode

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As early as the 1970s, scientists raised concerns about the gradual degradation of plastics to micro- and nano-sized particles in the environment and their potential impact on biota and human health. These tiny particles can easily be taken up by biological organisms and potentially cross biological barriers and bioaccumulate. Over the last years, there has been growing concern about the plastic soup our oceans are turning into. As a result, the scientific community is developing tools to estimate and map the distribution of plastics in our ecosystems and to investigate their potentially toxic effects on human health. However, so far, little is known about the occurrence and toxicity of low µm-size microplastics (MPs), thus requiring the development of novel and robust analytical methodologies to detect MPs in environmental matrices and to accurately determine their particle size distributions, number-based concentrations and chemical composition. A pioneering work carried out at A&MS-UGent, demonstrated the potential of inductively coupled plasma-mass spectrometry (ICP-MS) operated in single-event mode to characterize MPs with a diameter down to approximately 1 µm.[1] The approach relied on the monitoring of the transient $^{13}C+$ signals produced upon introduction of suspensions of polystyrene microparticles (1 and 2.5 µm diameter). However, the efficient introduction of larger µm-sized particles, especially up to 10-20 µm diameter, can be hampered by the limited transport efficiencies provided by liquid sample introduction systems. In this presentation, the potential of laser ablation as a means of sample introduction for MPs characterization will be discussed. Attention will be paid to the characterization of a broader MP size range (1-20 µm) and the monitoring of different polymer types (PS, PMMA and PVC). The figures-of-merit of this new approach will be presented and compared to those attainable in previous works and the advantages and disadvantages of this novel strategy will be highlighted into detail.

1. E. Bolea-Fernandez, et al., J. Anal. Atom. Spectrom. 35 (2020) 455-460. DOI: 10.1039/C9JA00379G

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The mTOR pathway is a highly conserved signaling pathway that plays a crucial role in regulating various cellular processes contributing to growth, through reversible protein phosphorylation.

Understanding how the pathway responds to different environmental cues, including not only the nutrient availability and energy status but also chemical exposure, could have potential applications in predictive ecotoxicology. Commonly, protein phosphorylation within signaling networks has been studied by antibody-based methods. However, these assays are time-consuming and can be expensive, especially when multiple protein targets need to be analysed. Moreover, suitable antibodies may be lacking for non-mammalian model organisms. To address this issue, we developed a mass spectrometry-based targeted (phospho)proteomics workflow allowing to simultaneously quantify phosphorylation and abundance of multiple protein targets in zebrafish (Danio rerio) PAC2 cell line, which is an important model species in environmental toxicology and human health fields. Our workflow starts with the optimized sample preparation, which includes a fast cell lysis (1 min) by 5% SDS, in-column protein trapping and digestion (S-Trap™), followed by desalting and finally enrichment of the phosphopeptides. The enrichment step substantially removed interferences and enabled the detection of target phosphopeptides. Fast and sensitive detection was achieved via the multiple reaction monitoring (MRM) that we developed using synthetic peptides, which allowed us to monitor changes in protein abundance and phosphorylation of 19 protein targets along the mTOR pathway. We applied our workflow to study the mTOR pathway dynamics at (i) different growth stages in cell culture, (ii) after nutrient deprivation, and (iii) after exposure to chemicals, including mTOR inhibitors and chemicals known to affect fish growth in vivo. Based on these analyses, we will present time-resolved protein phosphorylation responses within the zebrafish mTOR pathway, including several checkpoints involved in the regulation of cell growth and proliferation.

IT6-1 Smart Wound Dressings for the Real-Time Monitoring of the Healing Status

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The increasing demand for wearable technologies is giving rise to a strong push for the design of innovative chemical sensors targeting the real-time acquisition of vital parameters. Among the most challenging applications, nonhealing wounds monitoring is a scarcely explored medical field that still lacks quantitative and minimally invasive tools for the management of the healing process. This contribution deals with the development of smart bandages for the real-time and quantitative tracking of wound pH, uric acid (UA) concentration and moisture, all of which correlate with the healing stages and can potentially give access to the wound status, avoiding invasive procedures and unnecessary dressing changes that perturb the wound bed.

Fully textile sensors were obtained by screen-printing an optimised ink formulation containing a biocompatible organic semiconductor, i.e., poly(3,4-ethylenedioxythiophene):polystyrene sulfonate (PEDOT:PSS), in the desired sensing pattern. In particular, exploiting the intrinsic electrochemical properties of PEDOT:PSS, the moisture sensor operates by wirelessly monitoring in real-time impedance variations that span over several orders of magnitude and allows to discriminate between dry and wet states [1]. Differently, an organic electrochemical transistor (OECT) configuration was exploited for the design of the uric acid sensor, which proved to selectively and reversibly detect UA concentration within the biologically relevant range of 220−750 μM [2]. Finally, a two-terminal pH sensor was realised by functionalising the screen-printed polymeric ink with IrOx particles, which

spontaneously exert an electrochemical gating on the polymer and reversibly modulate its conductivity in a pH-dependent fashion [3] within the medically relevant range for wound monitoring (pH 6−9). The three textile sensors were combined with other medical gauzes with different absorption properties, thus leading to a final smart dressing ensuring the delivery of a continuous wound exudate flow across the sensor area. This setup allowed us to assess the analytical performances in flow conditions for better mimicking the final use of these wearable devices. Thanks to the careful selection of the textile materials and to the compactness of the final assembly, as well as the robustness of the sensing elements and transduction mechanisms, the smart dressings showed excellent resiliency to mechanical deformations and temperature variations.

[1] Tessarolo, M.; Possanzini, L.; Gualandi, I.; Mariani, F.; Torchia, L. D.; Arcangeli, D.; Melandri, F.; Scavetta, E.; Fraboni, B. Wireless Textile Moisture Sensor for Wound Care. Front Phys 2021, 9, 722173. https://doi.org/10.3389/fphy.2021.722173.
[2] Arcangeli, D.; Gualandi, I.; Mariani, F.; Tessarolo, M.; Ceccardi, F.; Decataldo, F.; Melandri, F.; Tonelli, D.; Fraboni, B.; Scavetta, E. Smart Bandaid Integrated with Fully Textile OECT for Uric Acid Real-Time Monitoring in Wound Exudate. ACS Sens 2023. https://doi.org/10.1021/acssensors.2c02728.
[3] Mariani, F.; Serafini, M.; Gualandi, I.; Arcangeli, D.; Decataldo, F.; Possanzini, L.; Tessarolo, M.; Tonelli, D.; Fraboni, B.; Scavetta, E. Advanced Wound Dressing for Real-Time PH Monitoring. ACS Sens 2021, 6 (6), 2366–2377. https://doi.org/10.1021/acssensors.1c00552.

IT6-2 Greener Approach to Determination of Free Tryptophan in Cold-pressed Oils by Reversed-Phase Dispersive Liquid-Liquid Microextraction and High-Performance Liquid Chromatography

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Deep eutectic solvent (DES), consisting of choline chloride and urea, was used in reversed-phase dispersive liquid-liquid microextraction (RP-DLLME) and high-performance liquid chromatography (HPLC) for the determination of free tryptophan in vegetable oils. The influence of eight variables affecting the efficiency of RP-DLLME was investigated using a multivariate approach. A Plackett-Burman design to screen the most influential variables, followed by a central composite response surface methodology, resulted in an optimal RP-DLLME setup for 1 g oil sample: 9 mL hexane as dilution solvent, vortex extraction with 0.45 mL DES (choline chloride urea) at 40 ^{o}C, without salt addition, and centrifugation at 6000 rpm for 4.0 min. The reconstituted extract was directly injected into an HPLC system operating in diode array mode. At the concentrations tested, the limit of detection (MDL) was 11 mg/kg, linearity for matrix-matched standards was R2 ≥ 0.997, relative standard deviation (RSD) was 7.8%, and average recovery was 93%. The method was used to analyze cold-pressed oils from nine vegetables.

Greenness assessment of DES-RP-DLLME-HPLC for the determination of tryptophan in oils was performed using three approaches to evaluate the environmental impact of this method: Analytical Eco-Scale, Green Analytical Procedure Index (GAPI) and Analytical Greenness metric (AGREE).
The findings of this study suggest that the use of DES-based methods for the extraction and analysis of target analytes in complex food matrices can contribute to sustainable development in analytical chemistry in general and could serve as a model for the development of more environmentally friendly and sustainable food analytical methods in the future.

IT6-4 A “Hot” Date with Capsaicinoids: Molecular Networking meets TRPV1

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Chili plants are known for their production of capsaicinoids, specialized metabolites defined by their vanilloid head and the C9-C11 carbon long tail (acyl moiety). The most well studied capsaicinoid, capsaicin, is a well-characterized agonist of the human pain and heat receptor, TRPV1, and has been used in a wide range of medicinal applications1. Despite capsaicin's significant role in medicine, other capsaicinoids remain relatively uncharacterized, with about only 20 capsaicinoid structures reported throughout literature2. Furthermore, most studies exploring capsaicin’s biological relevance have not been expanded to other capsaicinoids. Utilizing UPLC-HRMS,MZmine3 data processing3, feature-based molecular networking4, and SIRIUS5, two powerful in-silico MS/MS bioinformatic tools, we analyzed 40 different Capsicum sp. varieties and identified potentially novel capsaicinoids. Of the 40 varieties, the spicier varieties contained the richest chemical spaces, i.e. the most total feature coverage. Furthermore, 5 varieties – Chocolate Habanero, Chiluacle, Fatalii White, Lemon Drop and Longum Sahara – covered almost 90% of the entire chemical space detected amongst all 40 varieties. These 5 varieties were selected, plus Carolina Reaper due to its notoriety, for further deep-scanning MS/MS analysis. The deep scans of the 6 varieties are being used to build the most complete natural capsaicinoid library. Current in-silico estimation is 65 capsaicinoids, with another 24 features not predicted to be capsaicinoids but clustered together due to similar MS/MS fragmentation. In addition, capsaicinoids’ biological effects are being investigated by using a stably transfected cell line that expresses rTRPV1 and an established fura-4 calcium-influx assay. The goal is to create the most complete library of capsaicinoids with corresponding biological information on their activation of TRPV1.

1. Caterina, Michael, et at., Nature. 389 (1997) 816-824.
DOI: 10.1038/39807
2. Maria de Lourdes Reyes-Escogido, et al, Molecules. 16(2) (2011) 1253–1270.
DOI: 10.3390/molecules16021253
3. Schmid, Robin, et al., Nat. Bio. Tech. 41 (2023) 447–449.
DOI: 10.1038/s41587-023-01690-2
4. Nothias, Louis-Félix, et al., Nat. Meth. 17 (2020) 905-908.
DOI: 10.1038/s41592-020-0933-6
5. Dührkop, Kai, et al., Nat. Meth. 16 (2019) 299-302.
DOI: 10.1038/s41592-019-0344-8

IT7-1 Electrochemical performance of nitrogen doped carbon films and their application for electroanalysis for biological fluid

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Carbon films have been used for electroanalysis since they have wide potential window and low capacitive current. The films can be fabricated into the electrodes with any sizes and shapes. We have been studying carbon film electrodes formed by sputtering processes for detecting various analytes including biomolecules[1]. In such films, nitrogen doped carbon films show excellent electrochemical

performance including improved electron transfer for various redox species[2] More recently, we reported that electrocatalytic property of Ni nanoparticles for sugar oxidation were improved by modifying on the nitrogen doped carbon film compared with Ni particles on the pure carbon film[3] Here, we report improvement of biocompatibility by introducing nitrogen atoms on the carbon film electrodes because biochemical samples such as blood and saliva contain various interfering molecules such as protein, which rapidly fouls the electrode surface. We studied the electrochemical performances of carbon film electrodes before and after water vapor (H_2O) and ammonia gas (NH_3) plasma treatments[4]. The H_2O plasma treatment increased the surface oxygen concentration of the carbon films and decreased their water contact angle. The NH_3 plasma treatment increased the surface nitrogen content to about 5 at %, but a similar amount of oxygen remained on the surface. The sp^2 bond amounts decreased and the sp^3bond amounts increased after the H_2O and NH_3 plasma treatments. The NH_3 plasma treated carbon film indicated almost unchanged a peak separation (ΔE) of 1 mM ferricyanide after containing 100 mg/mL Bovine Serum Albumin (BSA) in the measurement solution, while ΔE of an untreated carbon film increased about 600 mV after BSA addition. However the sensitivity in the protein containing solution decreased by increasing protein concentration. We studied this sensitivity decrease by comparing diffusion coefficient of ferricyanide.

in protein containing solution and ethylene glycol containing solution by adjusting same solution viscosity, and found that the sensitivity decrease is due to two factors, one is diffusion coefficient decrease by increasing viscosity, the other is interaction between serum protein and ferricyanide. The obtained results enable us to evaluate the diffusion coefficient of biomolecules in the biological fluids such as serum.

1. Z. Zhang, S. Ohta, S. Shiba, O. Niwa, Nanocarbon film electrodes for electro-analysis and electrochemical sensors, Current Opinion in Electrochemistry, 2022, 35, 101045. doi.org/10.1016/j.coelec.2022.101045

2. T. Kamata, D. Kata, O. Niwa, Electrochemical performance at sputter-deposited nanocarbon film with different surface nitrogen containing groups, Nanoscale, 2019, 11, 10239. doi.org/10.1039/C9NR01569H

3. S. Shiba, S. Ohta, K. Ohtani, S. Takahashi, D. Kato, O. Niwa, Supporting effects of a N-doped carbon film electrode on an electrodeposited Ni@Ni(OH)2 core–shell nanocatalyst in accelerating electrocatalytic oxidation of oligosaccharides, RSC Advances2021, 11, 13311-13315. doi.org/10.1039/D1RA01157J

4. S. Ohta, S. Shiba, T. Yajima, T. Kamata, D. Kato, O. Niwa, Gas-phase Treatment Methods for Chemical Termination of Sputtered Nanocarbon Film Electrodes to Suppress Surface Fouling by Proteins, J. Photopolym. Sci. Tech., 2019, 32, 523-528. doi.org/10.2494/photopolymer.32.523

IT7-2 Does “push-pull” agriculture, as practiced by farmers, alter the composition of plant volatiles in fields to promote biological pest control?

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“Push-pull” technology is a form of mixed cropping developed to control insect pests of a focal crop using volatile chemical properties of plants. An intercrop is selected for its ability to repel pests of the focal crop. Around field perimeters, a trap crop attractive to pests, but on which they will not complete their life cycle, is meant to pull them out of fields and reduce pest populations. In addition, either or both companion crops may attract predators and parasitoids of pests (beneficials). These effects are thought to be driven primarily by emission of specific blends of bioactive plant volatiles from inter- and

trap crops. Implementations of push-pull for cereal crops, especially maize and sorghum, have been developed collaboratively with smallholder farmers in sub-Saharan East Africa, especially in Kenya. Agronomic considerations have driven these developments, with only a handful of studies assessing how changes may affect mechanisms underlying push-pull. As the technology is adopted by a growing number of farmers across sub-Saharan East Africa, improved mechanistic understanding of current systems may help to adapt and optimize functions for a variety of locations and needs. The project UPSCALE seeks to understand and remove barriers to push-pull expansion while supporting its continued development.

As part of UPSCALE, we synthesized existing literature on the chemistry which may underlie push-pull effects in different systems. We then characterized plant volatiles from current push-pull companion crops in comparison to focal crops, as well as ambient samples, in and near push-pull and paired non-push-pull fields run by farmers in Kenya. We used a simple and field-robust passive sampling approach on polydimethylsiloxane (PDMS) for presence-absence analyses, combined with a selection of more sensitive and semi-quantitative collections by actively pumping air through Tenax or Poropak Q filters. Additional sampling of potted plants under semi-field conditions was performed for a subset of common or recently implemented intercrops. Sampling was conducted across different times of day with the greatest number of replicates capturing the time frame expected to be most relevant to the activity of pests and beneficials in fields. We will correlate our results to insect distribution data for pests and beneficials collected by UPSCALE partners. We are furthermore experimentally testing the influence of companion plant volatiles on colonization of focal crops by pests. We describe our findings in terms of evidence for the difference in bioactive plant volatile composition in push-pull fields of different configurations, versus non-push-pull fields.

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https://upscale-h2020.eu/

IT7-3 Drug Quantification in Whole Blood using a Paper-Analytical Device for Point-Of-Care Therapeutic Drug Monitoring

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Therapeutic Drug Monitoring (TDM) allows for personalized dosage during therapeutic treatments and is often mandatory for modern potent drugs against cancer, infections or in organ transplantation cases [1]. A prototypical example is the antibiotic tobramycin, which is often prescribed to neonates in case of bacterial infection and requires TDM to ensure efficacy while avoiding oto- and nephrotoxicity. Currently, the process of TDM is demanding for the patient as several milliliters of blood are required, is slow and costly due to the transfer of sample to a central laboratory, and suffers of limited efficacy owing to the difficulty to interpret the results for a non-specialist. To circumvent these problems, we aimed at developing a point-of-care device enabling the quantification of therapeutic drugs in blood [2].

Our strategy is based on the use of fluorescence-polarization immunoassay (FPIA), a simple and rapid assay that may however suffer from interferences caused by the micro-environment. Here, we show

that FPIA can be downsized with reduced requirements in blood amounts (1 µL) and number of steps, without compromising reliability, and can be integrated within paper-like microstructures.
For Tobramycin, the integrated assay enabled quantification in serum with satisfactory performance in terms of precision and recoveries. Furthermore, whole-blood measurements were made possible by using the same paper-like microchamber as a filtering device and a measurement chamber. The final TDM point-of-care test requires minute amounts of blood and minimal handling steps.

[1] A. Taddeo, D. Prim, E.-D. Bojescu, J.-M. Segura, M. E. Pfeifer, J Appl Lab Med 5 (2020) 738. DOI: 10.1093/jalm/jfaa067
[2] E.-D. Bojescu, D. Prim, M. E. Pfeifer, J.-M. Segura, Anal Chim Acta 1225 (2022) 340240. DOI: 10.1016/j.aca.2022.340240

IT7-4 Application of digitalisation tools for efficient data processing, electronic lab notetaking, and population and use of databases in UHPLC method development of peptide and protein-based pharmaceuticals.

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The FAIR (Findable, Accessible, Interoperable and Reusable) principles are a concise and measurable set of values in describing the flow of data and efficient transfer of knowledge within and between academia, industry, funding agencies, and scholarly publishers for use by both humans and machines. In an industrial pharmaceutical setting, these principles are key to the more efficient use of data, faster process development, more automated workflows, and higher quality data. In addition, data management is vital for documentation in a regulated industrial context as well as reproducibility in scholarly settings. Transparency of data within these ecosystems allows for automation, combatting human error as well as releasing time for creativity and innovation [1].
This work describes the application of automated results display, data processing strategies and database storage in the development of ultra-high pressure liquid chromatography (UHPLC) purity determination and content methods in drug development. Python scripts are coupled with a Streamlit user interface and a centralised digital architecture to offer a simple and clear user experience allowing for the fast development of user-specific apps. One example shows the development of a method toolbox database with semi-automated entry, restricted removal, and querying tool, with an option to write to an electronic lab notebook, allowing for a fast overview of previously designed methods and easy documentation. With this tool, the eventual goal would be to apply machine learning to be able to predict method parameters for faster and more robust method development.

[1] M. Wilkinson, et al. Sci Data 3, 160018 (2016). https://doi.org/10.1038/sdata.2016.18

IT8-1 New trends in the development of boron-doped diamond electrodes: Approaches based on heteroepitaxy and additive manufacturing

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Conductive boron-doped diamond (BDD) is a remarkably versatile electrode material, which has been extensively employed in electroanalysis and sensors development [1]. Notably, a majority of BDD

electrodes employed for sensing applications are of polycrystalline nature. Consequently, they show heterogeneous character stemming from diamond grains composed of various crystal facets and grain boundaries, where sp^2 carbon impurities often reside and can deteriorate electrochemical performance. The broader use of single-crystal diamond electrodes, on the other hand, possessing a well-defined surface composition and orientation, is prevented in electrochemical applications predominantly due to the size limitation. The heteroepitaxy approach, however, allows to synthesize large-area and heavily-doped (100)-oriented single-crystal BDD (SC-BDD) electrodes, as we proved in [2]. This recently introduced and scalable heteroepitaxial SC-BDD represents a highly attractive electrode material, manifested by wide potential window (~3.3 V) and low double-layer capacitance (<4 $\mu F/cm^2$). Further, more satisfactory analytical parameters were recognized for dopamine on heteroepitaxial SC-BDD, compared to the polycrystalline electrode. Moreover, the excellent anti-fouling property of SC-BDD electrodes, resulting in improved response stability, was clearly demonstrated during experiments with dopamine and anthraquinone-2,6-disulfonate probe. Therefore, SC-BDD certainly manifested the potential to replace and even outperform conventional polycrystalline BDD, particularly in applications where low susceptibility towards (bio)fouling is desired.

Another topic gaining attention is the fabrication of patterned BDD in a straightforward and inexpensive way, which is required for a variety of practical applications, including the development of BDD-based electrochemical sensors. We describe for the first time a simplified, bottom-up fabrication approach for the BDD-based three-electrode sensor chip resulting from selective-area seeding via direct inkjet printing of nanodiamond particles. Subsequently, the inkjet-seeded substrate was subjected to a chemical vapor deposition growth step to obtain miniaturized thin-film BDD working and counter electrodes. Following, the chip was completed with the inkjet-printed silver reference electrode. The electrochemical performance of novel chip-based BDD electrodes towards redox markers and structurally different organic molecules was examined and compared to the performance of commercial 'screen-printed' BDD electrodes. It was concluded that sensing chips fabricated via inkjet printing route do not suffer any apparent drawbacks which would limit their application.
Finally, new potential routes based on 3D printing techniques for removing technological barriers hindering integration of conductive diamond with flexible platforms are currently explored in our team.

Acknowledgements
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[1] S. Baluchová, et al., Anal. Chim. Acta, 1077 (2019) 30. DOI: 10.1016/j.aca.2019.05.041.
[2] Z. Liu, et al., Carbon, 201 (2023) 1229. DOI: 10.1016/j.carbon.2022.10.023.

IT8-2 Potential of Electron Microscopy for Micro – Nanoplastic analysis

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Quantifying the number of small (1-10µm) microplastic particles (MP) and nanoplastic particles (<1µm, NP) in complex matrices is challenging. Most frequently used single particle based methods include µ-Fourier Transform – Infrared (FT-IR) and µ-RAMAN spectroscopy. However, both methods run into diffraction limitation when addressing MP/NP and/or suffer from limited automation algorithms. With its excellent spatial resolution in combination with analytical capabilities at the nanoscale, electron microscopy (EM) offers great potential to address the challenges related to MP/NP analysis.

To assess small MP, an approach based on automated scanning electron microscopy (SEM) is presented. Polyethylene (PE), polyvinylchloride (PVC) and Lufa 2.4. soil -all sieved to 1-10µm - were mixed in different proportions to simulate contaminated soils. Suspensions of these mixtures were filtered through gold-coated membrane filters (Nuclepore, Whatman), which were imaged using an SEM operated at low acceleration voltage (3kV). The experimental conditions were derived from Monte Carlo simulations of the interaction of the electron beam with solid materials using the software code CASINO (v. 3.3.0.4) [1]. Individual particles are detected based on their backscattered electron signal. PE and PVC are successfully identified based on characteristic elemental ratios (using a windowless energy dispersive x-ray (EDX) analysis system (X-TREME, Oxford Inst.)), also in the presence of an overwhelming amount of soil particles.

To detect NP, a scanning transmission electron microscope (STEM) in combination with EDX and electron energy loss spectroscopy (EELS) is used. Polystyrene (PS) NP with size range from 30 – 100 nm were mixed with silica particles of similar sizes and deposited on Si3N4 membranes, which provide an almost carbon free substrate. Based on the elemental signature of individual particles, PS and silica particles can be identified using both, EDX and EELS. Compared to the more frequently used EDX analysis, EELS offers a considerably higher collection efficiency translating into reduced measurement times. Furthermore, the carbon edge recorded in EELS spectra contains a wealth of information about the bonding stated of the respective materials, which eventually may even allow the identification of individual polymer types of NP particles. Furthermore, the low loss region (plasmon peaks) may be used to distinguish between different polymer types. As these features are associated with a very limited energy loss of up to ~50eV, measurement times are further reduced.

[1] Demers, H., et.al. (2011): Three-dimensional electron microscopy simulation with the CASINO Monte Carlo software. Scanning, 33: 135-146

IT8-3 Discovery of Antimicrobials Against Multidrug-Resistant Pathogens from Unexplored Natural Sources

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In recent years, antibiotic resistance has evolved dramatically and is now classified as the next pandemic.[1] As life-threatening multi-resistant (MDR) pathogens are rising, the discovery of novel molecules is key to contribute at solving this pressing issue.[2] However, the diversity of natural sources explored by scientists in the last century limits the prospects of finding novel molecular scaffolds. Our research process combines a new, powerful, and open source dereplication application (OctoChemDB) with a classic bio-assay-guided fractionation applied on extremophiles as well as fungi, from different ecosystems, that were not previously investigated for their activity against MDR pathogens. Briefly, extracts of these organisms were fractionated by solid-phase extraction and reversed-phase HPLC. Active fractions against MDR pathogens were purified to single products and characterized by High-

Resolution Mass Spectrometry (HRMS) analysis. OctoChemDB, which is a browser application that compiles main chemistry databases, is able to sort out bioactive molecules corresponding to the purified analyte. This allows to rapidly identify known antibiotics and dereplicate them to maximize chances of finding novel molecules. Altogether, the natural products discovery process was significantly accelerated: six known antibiotics with activity against MDR isolates were successfully dereplicated from 4 organisms not known to produce these compounds, including Cephalochromin,[3] and additional fractions active against MDR pathogens are being investigated.

[1]D. J. Chinemerem Nwobodo, et al., Clin. Lab. Anal. 36 (2022). DOI:10.1002/jcla.24655
[2]S. E. Rossiter, et al., Chem. Rev. 117 (2017) 12415–12474. DOI:10.1021/acs.chemrev.7b00283.
[3] C. J. Zheng, et al., Biochem. Biophys. Res. Commun. 362 (2007) 1107–1112. DOI:10.1021/acs.chemrev.7b00283.

IT9-1 Label-free detection of protein post-translational modifications with a biological nanopore

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Protein post-translational modifications (PTMs) play a crucial role in countless biological processes, profoundly modulating protein properties on both the spatial and temporal scales. Protein PTMs have also emerged as reliable biomarkers for several diseases. However, only a handful of techniques are available to accurately measure their levels, capture their complexity at a single molecule level and characterize their multifaceted roles in health and disease. Nanopore sensing provides high sensitivity for the detection of low-abundance proteins, holding the potential to impact single-molecule proteomics and PTM detection in particular. Here, we demonstrate the ability of a biological nanopore, the pore-forming toxin aerolysin, to detect and distinguish peptides bearing single or multiple PTMs. The characteristic current signatures of the wild-type peptide and its PTM variants could be confidently identified using a deep learning model for signal processing. We further demonstrate that this framework can quantify peptides at picomolar concentration. Collectively, our work highlights the unique advantage of using nanopore as a tool for the simultaneous detection of multiple PTMs and paves the way for their use in biomarker discovery and diagnostics.

IT9-2 Development of multi-residue methods for the determination of high production volume chemicals in muscle, skin and liver of seafood

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The Organisation for Economic Cooperation and Development (OECD) has compiled a list of high production volume chemicals (HPVC) that are produced at levels above 1,000 tons per year in at least one country in the member region. The production of these compounds has increased in recent years, and many of them have toxicological characteristics for humans and the ecosystem [1,2]. These compounds include synthetic musk fragrances, organophosphate esters, benzothiazoles, benzosulfonamides, and phthalates. Due to their high production, these compounds are found in the

environment, such as seas and oceans, and consequently in the organisms that live in them. Therefore, there is a need for a simple, fast, and robust method for the determination of HPVC in fish samples.
In the present study, thirty compounds were determined in fish muscle, skin and liver samples by gas chromatography-tandem mass spectrometry (GC-QqQ-MS/MS). A QuEChERS salt extraction method was developed to extract HVPC from muscle samples. Two extraction methods based on QuEChERS extraction ultrasonic assisted extraction (USAE) with acetonitrile, were optimized and compared for the extraction of HVPC from liver and skin samples, being the USAE the one which provided higher recoveries.

Due to the high content of lipids in all the samples, a clean-up procedure was successfully applied using LipiFiltr® cartridges. Muscle samples were monitored every three months for one year to estimate the risk and exposure rate of consumer ingestion, while skin and liver samples were analyzed to investigate the bioaccumulation processes of HVPC in fish.

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1. *L. Trabalón et al., Environ Res. 143 (2015) 116-122. doi:10.1016/j.envres.2015.04.007*
2. *Ó. Castro et al., J Chromatogr A. 1626 (2020) 461356. doi:10.1016/j.chroma.2020.461356*

IT9-3 Understanding mental health from single hair by nanoparticle-assisted laser desorption/ionization mass spectrometry imaging

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To demonstrate new methodology for understanding our mental health from single hair sample, in the present study, we visualized stress level from a vertical hair-slice section by nanoparticle-assisted laser desorption/ionization (Nano-PALDI) imaging mass spectrometry (IMS)(1).

Hair contains an enormous amount of molecular information that is sensitive to chemical and physiologic influences. Because hair grows, it is possible to evaluate biological changes over time by analyzing hair samples from the root to the shaft. Thus, hair samples can be used to obtain daily information non-invasively.

Conventionally, we should divide hair as segment from tip to root and analyze the extracts of hair sample by liquid or gas chromatography technique. However, it is complicated to understand location (time-varying) of target molecules.

Thus, using a vertical hair-slice section, we compared the components of hair using two ionization methods, matrix-assisted laser desorption/ionization (MALDI: conventional) and Nano-PALDI IMS (2,3), respectively.

Nano-PALDI IMS can also be used to acquire high-resolution images (5 μm) due to the absence of crystallization effects observed with MALDI. Nanoparticles are only physically adsorbed onto the sample surface, so even if the particles aggregate to form a secondary particle, the size is limited to approximately 100 nm in diameter(4). Therefore, IMS using the Nano-PALDI method can be easily performed with higher spatial resolution than that possible with conventional MALDI methods.

In Nano-PALDI high spatial image, localization of cystine and 18-methyleicosanoic acid as endogenous hair components were confirmed at the cuticle and cortex and cuticle, respectively.

In addition, we image stress markers from depression mice hair. Corticosterone as known marker for stress was imaged when symptoms of depression have advanced. However, new marker that we newly found by omics analysis was imaged from early symptoms. This is the result because Nano-PALDI high spatial image from thin hair samples. In future, hair sample will use to understand own mental health and physical examination by non-invasively.

(1) S. Taira (corresponding author) et al. 'Analysis of hair components by nanoparticle-assisted laser desorption/ionization mass spectrometry imaging' Arabian Journal of Chemistry 15, 104205 (2022)
(2) S. Taira (corresponding author), K. Shiono "Imaging of Plant Hormones with Nanoparticle-Assisted Laser Desorption/Ionization Mass Spectrometry" Methods in Molecular Biology 'Mass Spectrometry Imaging of Small Molecules Chap 9, 127-139 (2022)
(3) S. Taira (corresponding author) et al. 'Nanoparticle-assisted laser desorption/ionization using sinapic acid-modified iron oxide-nanoparticles for mass spectrometry analysis' Analyst 140, 8134-8137 (2015) (Cover article)
(4) S. Taira et al. "Nanoparticle-Assisted Laser Desorption/Ionization Based Mass Imaging with Cellular Resolution" Analytical Chemistry 80, 4761-4766 (2008)

IT9-4 Sequence confirmation and impurity characterization of therapeutic oligonucleotides – A quality by design approach

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Introduction
Detailed characterization of therapeutic oligonucleotides and their impurities is crucial for process development and quality control of drug substances(1). This includes the confirmation of the sequence with all their synthetic modifications and a detailed structural elucidation of chromatographically separated and co-eluting impurities. Common approaches use liquid chromatography coupled to low-resolution mass spectrometry, which makes characterization unprecise, when compared to the use of ultrahigh resolution mass spectrometry. In a GMP regulated environment the use of ultrahigh resolution mass spectrometry in oligonucleotide analysis is set to become standard but there is still a lack in regulatory guidance and systematic workflows for method development. CpG1018, a short (22-mer) oligonucleotide used in preclinical testing as an adjuvant for immunization against hepatitis B virus, is used exemplarily to show the procedure for method development for sequence confirmation using collision induced dissociation and ultra-high resolution mass spectrometry. Offline desalting prior to direct infusion ESI-MS results in high precursor signal intensity, which is essential for complete sequence coverage. Fragments resulting from CID by varying the precursor charge state and the laboratory frame collision energy are evaluated statistically with the goal to find the optimal experimental conditions that are suitable for validation and performance under GMP conditions.
Optimized collision induced dissociation parameters are also applied for direct localization and quantification of product related such as PS-PO conversions, methylations and truncated sequences by liquid chromatography coupled to the mass spectrometer. Due to low specificity of the chromatography, the localization of modifications on impurities is performed by calculating the fractions of modified and unmodified fragments. This allows for precise characterization of related impurities, which is crucial in a GMP environment and highly demanded by authorities in late phase pharmaceuticals.

All measurements are acquired with in house produced 22-mer oligonucleotide CpG1018 using a Maxis II qTof (Bruker Daltonics, Bremen, Germany) coupled with a Vanquish UHPLC System (Thermo Fisher Scientific, Bremen, Germany). Offline desalting prior to direct infusion ESI-MS is performed by using reversed phase Solid SPE alkylamine containing eluents through an inhouse developed workflow. Sample introduction for collision energy optimization is performed via a syringe pump. Data acquisition and processing were performed by Bruker oTof Control and Bruker Data Analysis. Fragment assignment is performed by OMAOPA software package (2).

(1) Investigation of the Influence of Charge State and Collision Energy on Oligonucleotide Fragmentation by Tandem Mass Spectrometry. Molecules 2023, 28, 1169
(2) A. Nyakas, L. C. Blum, S. R. Stucki, J.-L. Reymond, S. Schürch. OMA and OPA - software-supported mass spectra analysis of native and modified nucleic acids. J. Am. Soc. Mass Spectrom. 2013, 24, 249-256

IT10-1 Purpose-Made Capillary Electrophoresis Instrumentation

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In contrast to chromatography the separation method of capillary electrophoresis (CE) does not require ultra-high-pressure manifolds. It is therefore possible to construct CE instruments relatively easily in the laboratory. In the spirit of open-source hardware this allows cost savings, but there are also applications of CE for which commercial solutions are not readily available. This includes field-portable and battery-powered instruments, automated on-site monitoring systems (process analysis), or laboratory instruments dedicated to special tasks.

While sample injection into the capillary may be done by manual syphoning this is not always easy to carry out, especially in the field, and it is preferable to automate this step as well as flushing operations by employing a flow-injection approach. The CE part may then be considered as a powerful multi-analyte detector in a flow-injection system. Pumping can be carried out by pneumatic pressurization and miniature valves allow the sequencing of operations with the help of a microcontroller. By making use of available miniature components compact instruments can be assembled in a highly flexible and adaptable microfluidic breadboard approach. The most demanding part of these instruments is the detector, but capacitively-coupled contactless-conductivity detectors (C4D) may be constructed in the lab if electronic expertise is available or be bought at relatively low cost. Also possible is the in-house building of absorbance or fluorescence detectors based on light-emitting diodes (LED) or laser diodes. The presentation will highlight recent developments in our laboratory.

I.J. Koenka, J. Sáiz, P. Rempel, P.C. Hauser, Anal. Chem. 88 (2016) 3761.
J.S. Furter, P.C. Hauser, Anal. Chim. Acta 1058 (2019) 18.
J.S. Furter, M.-A. Boillat, P.C. Hauser, Electrophoresis 41 (2020) 2075.
J.S. Furter, P.C. Hauser, J. Chromatogr. A, 1656 (2021) 462533.

IT10-2 Holistic analysis of a Swiss karst spring using on-site, in-situ RPLC-HRMS/MS and laboratory based IC-HRMS/MS

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Karst groundwater resources are susceptible towards contamination due to the often highly weathered bedrock, exhibiting voids such as pipes and caves leading to fast groundwater flow velocities and little time for attenuation. Especially, agriculturally used catchments pose a risk to karst groundwater quality as pesticides and their transformation products might reach the groundwater rapidly after precipitation events. To investigate the concentration dynamics of 295 pesticides and their transformation products we used a transportable RPLC-HRMS/MS monitoring station, which collected and analyzed in situ and automatically one sample every 20 minutes at a karst spring in the Swiss Jura, in May and June 2021.

To account for persistent and mobile, very polar, anionic compounds with low log DOW values that could not be analyzed with the online RPLC platform, we additionally collected 42h-composite samples with an automatic sampler throughout the growing season 2021. To detect such compounds we developed an anion-chromatography-based method, coupled to HRMS/MS. The method comprises the precipitation of interfering matrix components (chloride and sulfate) and subsequent enrichment via vacuum assisted evaporative concentration.

In this study, we could show that active substances as well as their transformation products can be mobilized at the surface and can reach the studied karst spring after precipitation events. Concentrations of several pesticides exceeded the drinking water limit of 100 ng/L up to ten times for time periods of several hours to days. The developed ion-chromatography method with a median matrix LOQ of 30 ng/L for 64 substances enabled detection of two transformation products of pesticides for the first time: chlorothalonil TP SYN548008 and dimethenamid TP M31. Overall, using high sampling frequencies and two complementary, highly sensitive analytical methods, we could characterize the concentration dynamics of a broad range of compounds in a karst spring in the Swiss Jura and hence deepen our understanding of such valuable drinking water resources.

IT10-3 Support for understanding analytical chemistry by questions and videos

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Active learning involves students directly in the process, is contrasted to a traditional lecture, and is often advocated as more effective than passive approaches.[1] However, engaging students in a lecture-based course can be challenging.[2] Among active learning modes and means are problem-based learning, collaborative learning, inquiry-based learning, classroom response systems and peer instruction. Yet, key and so far understudied ingredients for active learning are appropriate questions, problems, and tasks. Due to the lack of tangible design models, the development of questions requires much effort and is mostly left to individual instructors.[3] Likewise, and not just since distance learning during the COVID-19 pandemic, videos have been widely used for educational purposes. Technological advances have made their production and distribution more affordable, enabling utilization in face-to-face and online learning environments. Nevertheless, videos in a purely lecture-style format seldomly make use of their full potential and advantages, e.g. providing connections with otherwise difficult to access procedures, instrumentation, places, or people.[4]

Here, the use of questions and videos within the context of an undergraduate course in quantitative instrumental element analysis is explored. Emphasis is placed on the benefits and limitations of multiple-choice questions inside and outside the classroom and how they can provide a beneficial link for aligning teaching objectives, activities, and assessment in analytical chemistry. In addition, examples are given of how short video clips combined with questions can successfully engage students in classroom discussions. Details on planning, production, and use will be provided for video-supported case studies at the end of the semester for course revisions.

[1] (a) J. Michael, Adv. Physiol. Educ. 30 (2006) 159. DOI:10.1152/advan.00053.2006, (b) M. Prince, J. Eng. Educ. 93 (2004) 223. DOI:10.1002/j.2168-9830.2004.tb00809.x, (c) M. Cooper and R.L. Stowe, Chem. Rev. 118 (2018) 6053. DOI:10.1021/acs.chemrev.8b00020.
[2] G. Schwarz, J. Chem. Educ. 98 (2021) 2122. DOI:10.1021/acs.jchemed.0c01339
[3] G. Schwarz, Chimia 75 (2021) 33. DOI:10.2533/chimia.2021.33
[4] (a) M. Fyfield et al., Australas. J. Educ. Technol. 35 (2019) 1. DOI:10.14742/ajet.5930, (b) G. Winslett, Australas. J. Educ. Technol. 30 (2014). DOI:10.14742/ajet.458 (c) G. Schwarz et al., Anal. Bioanal. Chem. 414 (2022) 3301. DOI:10.1007/s00216-022-03983-y.

IT10-4 Digital Microfluidic Analytical Systems with Integrated Chemical Sensor and Antimicrobial Surfaces

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Digital microfluidics (DMF) are an exciting variant of microfluidic platforms owing to their intrinsic capability for process automation whereby all required procedures can be programmed and executed without user intervention. However, in order to work efficiently, these systems require very accurately defined surfaces to allow for precise droplet movement and minimize detrimental effects such as biofouling.

In order to extend the capabilities of these DMF systems it is useful to integrate further functionalities such as sensing features for real-time monitoring of biological systems. We have previously shown a DMF-integrated optical temperature sensor that allowed imaging of microscopic temperature distributions of DMF operations [1] as well as a DMF system with an oxygen sensor that was employed for a miniaturized antimicrobial susceptibility assay [2].

We now demonstrate the live monitoring of extracellular acidification on DMF using a chip-integrated fluorescent pH sensor array. The metabolism of various types of cells were investigated through recording the extracellular pH (pHe) change. An optical pH sensor array with spots of around 2 mm diameter was integrated onto a DMF interface using a fluorescein probe covalently bound to poly-2-hydroxyethylmethacrylate and immobilized on an indium tin oxide interface on a DMF top plate.

Label-free and non-invasive monitoring of extracellular acidosis was realized within a pH range of ca. 5.0-8.0. The platform was used to monitor the pHe decrease during MCF-7 and A549 cancer cell proliferation due to abnormal glycolysis. A rapid pH decrease in the presence of cancer cells could be detected within two minutes while no signficant pHe change was observed with HUVEC healthy cells. Real-time detection of cell acidification and cellular response to different metabolic conditions such as glucose levels or administered anti-cancer drugs could be demonstrated.

Also we present a sandwich-structured polydopamine and silver hybrid material. This film with a thickness of around 30 nm was integrated on the bottom plate of a DMF platform with an oxygen sensor film on the top plate. The coating showed a 99.9% reduction in E. coli population after one-hour contact under non-nutritive conditions and effectively inhibited E. coli growth in nutrient LB broth for 8 hours. Furthermore, the platform maintained a very low cytotoxicity to human cells. After 24 hours, 82 % HEK 293 and 86 % HeLa cells remained viable, respectively. The coating provides a hydrophilic area of adjustable size with a high antimicrobial effect and a low cytotoxicity in DMF and potentially other bioanalytical platforms.

1) Z. Lu, et al., ACS Appl. Mater. Interfaces 12 (2020) 19805
DOI:10.1021/acsami.0c02128
2) W. Qiu, et al. ACS Sens. 6 (2021) 1147
DOI: 10.1021/acssensors.0c02399

ORAL PRESENTATIONS

OP1-1-1 Real-time continuous monitoring of dynamic concentration profiles with biosensing by particle motion

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Real-time continuous biosensing is important for enabling the monitoring of the dynamics of biological systems and biotechnological processes. Biosensing by Particle Motion (BPM) is a continuous biosensing method with single-molecule resolution that relies on the tracking of the motion of biofunctionalized particles that interact with a biofunctionalized substrate [1,2,3]. The particles switch between bound and unbound states due to reversible single-molecule interactions, influenced by the presence of analyte molecules. To achieve real-time measurements with a high precision and a high time resolution, 1,000 to 10,000 particles need to be simultaneously tracked and analyzed in real time. This poses a computational challenge, because data streams of several gigabytes per minute should be analyzed in a short time (seconds to minutes). In recent work, we developed a signal processing approach that enables real-time, high-precision, continuous measurements [4] and applied it for real-time sensing of dynamic cortisol concentration profiles, studied for step functions and sinusoidal oscillations of analyte concentration [5]. The experiments allow the quantification of time delays in real-time continuous biosensing, that originate from physicochemical processes (advection, diffusion and reaction) and signal processing. The total time delay of the studied real-time cortisol sensor was below 2 minutes. Monitoring of sinusoidal cortisol concentration profiles showed that the sensor has a low pass frequency response with a cutoff frequency of 4 mHz and a lag time of approximately 60 seconds.

[1] Visser, et al., Nature Communications 2018, 9, 2541. DOI:10.1038/s41467-018-04802-8
[2] Buskermolen, et al., Nature Communications 2022, 13, 6052. DOI:10.1038/s41467-022-33487-3
[3] Van Smeden, et al., Reversible immunosensor for the continuous monitoring of cortisol in blood plasma sampled with microdialysis. ACS Sensors 2022, 7, 10, 3041–3048. DOI:10.1021/acssensors.2c01358
[4] Bergkamp, et al., High-throughput single-molecule sensors: how can the signals be analyzed in real time for achieving real-time continuous biosensing? Under review.
[5] Bergkamp, et al., Real-time continuous monitoring of dynamic concentration profiles with biosensing by particle motion. In preparation.

OP1-1-2 Continuous blood typing within capillary via packing-enhanced nanoscattering of gold nanoparticles

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Blood typing is a critical issue not only for transfusion medicine but also in the emergency room of a hospital. In the past few years, some groups proposed novel analytical methods for rapid blood typing based on the microfluid device[1], paper-based screening[2] as well as nanoparticle packing-enhanced

nanoscattering on individual erythrocytes followed by observation by objective-type dark-field microscopy (OTDFM)[3]. The single erythrocyte blood typing by OTDFM is different from traditional hemagglutination, thus, a prozone effect caused by overloaded antibodies or cells is negligible. In other words, the relative quantities of antibodies and erythrocyte is no longer a critical factor for hemagglutination. In this work, we extend the principle of OTDFM on a capillary column followed by collecting light scattering coaxially through a low magnification (20X) with a high N.A. objective (0.75). Most scattering from the objective and wall of the capillary column was rejected by a set of field stops while scattering from erythrocytes and gold nanoparticles (AuNPs) can be collected and focused on a photomultiplier. Both the scattering from erythrocytes and antibodies-coated AuNPs produce a significant and distinguishable signal once scatters flow through the capillary window. Furthermore, the aggregation of AuNPs on erythrocytes after the recognition by blood group-specific antibodies may produce a dominant scattering leading to blood group typing in an easier manner. Because all the scattering measurements are performed by a simple hydrodynamic injection from a capillary inlet, thus allowing for continuous blood typing for different specimens is no longer impossible.

References:
1. Ding, S., Duan, S., Chen, Y., Xie, J., Tian, J., Li, Y., & Wang, H. Centrifugal microfluidic platform with digital image analysis for parallel red cell antigen typing. Talanta, (2023) 252, 123856. DOI: 10.1016/j.talanta.2022.123856
2. Chomean, S., Bunnun, P., Auttapong, J., & Kaset, C. Phenotyping of minor blood groups (C, c, E, e, and Mia) using a paper-based device and image-based high-throughput detection. Analytica Chimica Acta, (2023) 1237, 340573. DOI: 10.1016/j.aca.2022.340573
3. Chen, I. S., Davis, S. J., Chang, M. L., Hung, C. H., Radenovic, A., & Chang, P. L. Rhesus blood typing within a few seconds by packing-enhanced nanoscattering on individual erythrocytes. Analytical Chemistry, (2021) 93, 15142-15149. DOI: 10.1021/acs.analchem.1c03590

OP1-1-3 Machine Learning-Assisted Biothiols Detection using Multicolor Plasmonic Patterns Enabled by Controlled Growth of Silver on Gold Nanorods

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Colorimetric sensor arrays have emerged as promising tools for multi-sensing, utilizing cross-reactive semi-selective sensor elements to achieve multi-sensing instead of individual lock-and-key sensors that rely on a univariate response from a highly selective sensor for the detection of a single analyte [1-3]. Array-based sensors generate a unique fingerprint pattern for each analyte, enabling simultaneous detection of multiple analytes. This high-throughput approach has found applications in the identification and quantification of a wide range of analytes, from ions [4-5], and pesticides [6] to biomolecules [7-8], cells [9], and pathogenic bacteria [10]. In this study, we present a colorimetric assay for rapid identification and quantification of four important biothiols: cysteine (Cys), homocysteine (Hcy), glutathione (GSH) and glutathione disulfide (GSSG). Our approach makes use of the varying inhibitory effects of different biothiols on the growth of a silver shell on the surface of gold nanorods. This creates a unique colorimetric signature that can be used to identify and detect each biothiol. Principal component analysis was coupled with linear discriminant analysis (PCA-LDA) to identify the fingerprint pattern of each biothiol and recognize different mixtures of biothiols. Partial least squares regression (PLSR) was used to quantify the concentration of biothiols over a wide concentration range (0.07 – 25 µmol L−1). The practical usability of our sensor was successfully validated by detecting and discriminating biothiols in human serum samples. By offering a sensitive and selective detection

method for monitoring biothiol levels in biological samples, our sensor holds significant promise for early disease diagnosis and treatment monitoring.

[1] https://pubs.acs.org/doi/abs/10.1021/acs.chemrev.8b00226
[2] https://pubs.rsc.org/en/content/articlelanding/2017/nr/c7nr03311g
[3] https://www.sciencedirect.com/science/article/abs/pii/S0165993619305059
[4] https://pubs.acs.org/doi/abs/10.1021/ac501953z
[5]https://www.sciencedirect.com/science/article/abs/pii/S0925400518309675
[6] https://pubs.acs.org/doi/10.1021/acs.analchem.6b01616
[7]https://pubs.acs.org/doi/abs/10.1021/acsami.0c17200
[8]https://www.sciencedirect.com/science/article/abs/pii/S092540052200497X
[9] https://www.sciencedirect.com/science/article/abs/pii/S0304383512005629
[10] https://pubs.acs.org/doi/10.1021/acs.analchem.1c05006

OP1-1-4 Reversible Thermochromic Polydiacetylene/Zinc(II)/Cadmium Selenide Quantum Dots Nanocomposites for Optical Sensing Applications

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Polydiacetylene (PDA) is a class of stimuli-responsive polymers with unique optical properties that has been utilized in advanced sensing applications. A visible blue-to-red color transition can be triggered from the environment stimuli, including temperature, pH, chemicals, and ligand-receptor interactions. The commercially available PDA exhibits irreversible thermochromism and a “turn-on” fluorescence signal in the red phase [1]. Recently, our previous work has successfully developed the reversible PDA/Zinc (Zn) (II) thermochromic materials which is facile preparation, multiple usage, and low in cost. Tuning sensitivity could be achieved by varying the zinc precursors, synthesis methods, and alkyl side chain lengths of diacetylene monomer. From In-situ Raman spectroscopy, we found that PDA(8,12)/$Zn(CN)_2$ demonstrated gradual peak shift to higher wavenumbers upon heating and fully reversible pattern after cooling down from 100°C to room temperature [2]. To improve the fluorescent properties of PDA, the fresh synthesized CdSe/CdZnS core-shell quantum dots (QDs) was used to mixed with PDA-based materials. Uniform mixing films were prepared by spin coating technique. Herein, red phase of PDA played a role as the doner molecules to transfer the energy to the acceptors that is the QDs. The UV/vis spectroscopy showed the overlap between the broad emission spectrum of the polymer (λmax= 563 nm) with the absorption spectrum of the QDs (λmax= 597 nm). In this work, the resultant red-PDA/Zn(II)/CdSe nanocomposites exhibited the maximum wavelength at 608 nm. We also measured the photoluminescence (PL) lifetime of the pure red PDA, pure QDs, blue-PDA/Zn(II)/CdSe nanocomposites and red-PDA/Zn(II)/CdSe nanocomposites to analyze and confirm the energy transfer. The lower PL lifetime of the red composite system indicated some intermolecular interactions compared to the blue one that no spectral overlapping. Moreover, different emission QDs and other measurements were carried out to explore the molecular origins of the behaviors. Our finding extends the potential of PDA as optical sensors, smart materials, switchable turn-on fluorescent probes.

[1] R. Pasquino, et al., J. Colloid Interface Sci. 394 (2013) 49-54.
DOI:10.1016/j.jcis.2012.11.039
[2] J. Rueangsuwan, et al., Colloids Surf. A Physicochem. Eng. Asp. 656 (2023) 130490.
DOI:10.1016/j.colsurfa.2022.130490

OP1-2-1 Investigation of the Retention Mechanisms of Porous Graphitic Carbon as Stationary Phase in HPLC

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For decades, high performance liquid chromatography (HPLC) separations of both small and large molecules have been carried out predominantly using silica-based particles. Since the introduction of HPLC, these materials provided the required mechanical stability and batch-to-batch reproducibility, as well as the flexibility, to modify and change the properties of the stationary phase by many silylation reagents. Alternative HPLC packing materials, such as carbon, alumina, zirconia, and titanium dioxide, are available as well, but have never gained popularity like silica-based materials, likely due to the different, and often not fully understood, retention mechanisms and how to control them.

Porous graphitic carbon (PGC) as a stationary phase in HPLC has been known for many years [1,2]. Advantages of PGC, in comparison to silica-based stationary phases, are stability over the complete pH range (1-14) and for temperatures up to 250 °C. In the beginning, it was assumed that this entirely carbonaceous stationary phase would act solely as a highly hydrophobic material, but results from experiments with polar compounds contradicted this hypothesis. Often, PGC retained polar compounds much better than C18 materials. Based on these observations, the term PREG (Polar Retention Effect on Graphite) was generated. Pioneers in the development and chromatographic characterization of PGC, Knox and Ross, attempted to explain this unexpected polar retention behaviour on PGC [3].

This work will focus on a new PGC material with smaller particle size (2.7 µm), a narrower particle size distribution and improved mechanical stability. The mechanisms on how the stationary phase is interacting with various nonpolar and polar analytes of different size and geometry is investigated. Especially for the polar compounds, analytes have been chosen that are known to be weakly retained on silica-based reversed-phase columns. Conclusions out of the resulting chromatograms will be drawn to explain the fundamental retention mechanisms on this carbon stationary phase. The comparison with silica-based columns will provide further insights into these mechanisms. This work will be completed by application examples showcasing how PGC columns add additional selectivity options to the classical silica-based columns for easier HPLC method development.

[1] J.H. Knox, et al.. J. Liq. Chromatogr. 6 (1983) 1. DOI: https://doi.org/10.1080/01483918308067647
[2] J.H. Knox, et al., J. Chromatogr. 352 (1986) 3. DOI: https://doi.org/10.1016/S0021-9673(01)83368-9
[3] J.H. Knox, P. Ross, Carbon-based packing materials for liquid chromatography: structure, performance, and retention mechanisms, In: Brown PR, Grushka E, editors. Advances in Chromatography, 37, New York: M. Dekker, 1997, 73-119.

OP1-2-2 PEGDA-BASED IONIC IMPRINTED POLYMERS FOR SELECTIVE BINDING OF LITHIUM

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Lithium is one of the main components of lithium-ion batteries (LIBs) involved in many fields such as electric vehicles, mobile phones, and laptops. Due to the environmental regulations and technological revolution, the global demand of lithium is growing exponentially, and it has been estimated that the demand will triple by 2025 so it will exceed the global production that currently sits on sourcing from mineral resources [1]. This high rate of demand has made Li part of the list of critical raw materials for the energy transition [2]. In this context, efficient recovery of lithium from industrial wastewater of spent LIBs is a mandatory alternative to primary geological sources to meet the global demand.

State of the art on lithium recovery shows that currently there are many methodologies available such as chemical precipitation, leaching, solvent extraction, and membrane separation. These methods consume large amounts of freshwater and chemicals, are expensive and inefficient [3]. Consequently, there is a need for selective absorption materials. For this purpose, the ion imprinting technique has been proposed as an innovative approach to separative issues to provide polymers with 3D nanocavities characterized by receptor-like properties, able to specifically recognize metal ions with a reversible binding behavior. This technology involves preparation by bulk-polymerization of macroporous and highly cross-linking polymers characterized by selective binding properties towards the target ion, which is previously introduced during polymerization and then accurately removed [4]. In this work, we present an original approach to obtain ionic imprinted polymers (IIPs) based on different diacrylate polyethylene glycol monomers (PEGDAs) known to form pseudo-crown ethers in polymer mixture capable of coordinating alkali ions by virtue of the macrocyclic effect [5]. In this way, the use of expensive polymerizable crown ethers is avoided. Polymerization mixture was optimized evaluating the behavior of different PEGDAs, synthesis conditions, monomer: ion molar ratio and leaching solution. The binding parameters were obtained through equilibrium partition isotherms by ion chromatographic analysis, also evaluating the selectivity factors towards binding competing ions (Na+, K+).

Experimental data show that the optimized polymerization mixture allows to produce an IIP able to recover lithium. IIPs have good binding capacities included within 51.7 ± 3.6 µmol/g and 4.7 ± 0.5 µmol/g according to different PEGDA:Li(I) molar ratios, with a good Na/Li and K/Li selectivity at least of 0.38 ± 0.1 These preliminary data show that ionic imprinted polymers have great potential for application as substrates for solid-phase extraction (SPE).

[1] A. Amato, et al., J. Clean. Prod., 300, (2021), 126954.
DOI:10.1016/j.jclepro.2021.126954
[2] Ministero della transizione ecologica e dipartimento per l'energia e il clima. La situazione energetica nazionale nel 2021. (2022)
[3] K.Garcia, et all, Processes, 11, (2023), 418
DOI:10.3390/pr11020418
[4] C. Branger, et al, React. Funct. Polym., 73, (2013),859
DOI:10.1016/j.reactfunctpolym.
[5] L. Xubiao, et al, ACS Sustain. 3, (2015), 460-467
DOI:10.1021/sc500659h

OP1-2-3 Hyphenated thermogravimetry–gas chromatography–mass spectrometry: a successful technique for the analysis of complex materials and thin films.

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Reliable and convenient analytical methods were developed exploiting the hyphenated TGA-GC-MS. This technique, applied to the study of complex polymeric materials, allowed obtaining quantitative analysis from complex systems with high sensitivity and without any sample preparation. The TGA stage operates a separation according to the temperature of degradation providing the weight loss of the sample, while the synchronized GC stage separates all the different compounds emitted at the same temperature allowing the resolution of the TGA peaks in terms of composition. At last, the MS stage provides the identification of each compound contributing to the weight loss. Polymeric materials were used as test-case to explore the possibility of developing quantitative methods, and the technique was also applied to the study of layered hybrid materials such as hydrotalcites and saponites intercalated with organic guests. Specific methods were developed to analyze ultrathin polymer films,[1] highlighting the high sensitivity of the technique, allowing the detection of small differences in composition, also with minimal amounts of sample. Diethylphosphate-end capped polymers with varying chain length, in bulk and as ultra-thin film were also studied,[2] developing, calibrating, and validating a method for the quantification of phosphorus in polymeric samples and even the determination of repeating units per chain end. The technique proved to be very sensitive, allowing the determination of phosphorous even in films of a few tens of nanometer thickness, and to be very flexible and reliable, maintaining its accuracy over a wide range of sample amount, going from bulk to thin layer samples, without the need of a re-calibration. In the analysis of grafted polymers,[3] an important advantage of TGA-GC-MS is that the sample can be introduced without preparation and together with its substrate, avoiding inducing structural changes. Moreover, this technique allowed to shed light on the interlayer composition of hybrid lamellar host-guest materials, being able to discern the between the weight loss due to the organic guest intercalated inside an inorganic host versus that due to the same molecule adsorbed on the surface, allowing not only chemical selectivity but also “topological” selectivity. [4-6]

1. *V. Gianotti, et al., Journal of Chromatography A, 1368 (2014) 204–210. https://doi.org/10.1016/j.chroma.2014.09.073*
2. *V. Gianotti, et al., Journal of Analytical and Applied Pyrolysis, 128 (2017) 238–245. https://doi.org/10.1016/j.jaap.2017.10.005*
3. *D. Antonioli, et al., Analytical and Bioanalytical Chemistry, 408 (2016) 3155–3163. https://link.springer.com/article/10.1007/s00216-016-9380-8*
4. *E. Conterosito, et al,. Chem. A Eur. J., 21 (2015) 14975–14986 https://doi.org/10.1002/chem.201500450*
5. *V. Toson, et al., Molecules, 27 (2022), 3048. https://doi.org/10.3390/molecules27103048*
6. *E. Conterosito, et al., Submitted to Chemistry a European Journal, (2023)*

OP1-2-4 Actual developments in HPLC modeling

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Modeling HPLC separations has a long tradition. It started 1986 as Lloyd Snyder started to model isocratic separations. Later modeling was extended for gradient methods in Reversed Phase Chromatography (RPC) and for Ion-Exchange Chromatography (IEC). Recent developments include HILIC, HIC, SFC and Robustness modeling. All techniques need experimental support to measure peak positioning, depending on pH, temperature, gradient time, salt- or additive concentration, etc. From

the measured data a software (f.Ex. DryLab) can model a separation under a number of variable condition in seconds instead of hours.
The results are reliable, robust methods, which enable the fast development and market-entry of new and more efficient drug products. The lecture will present several case studies on modeling from actual developments.

OP1-3-1 Analytical spectroscopical assessment of the interaction between metal nanoantimicrobials and lipid membranes

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Metal nanostructures have been proposed worldwide as antimicrobial agents, providing a strategic approach to fight antimicrobial resistance and biofilm formation. However, significant concerns about their potential toxicity are arising. Particularly, the well-known health risks related to the NP size-controlled unprecedented reactivity should be considered. In this regard, metal nanoparticles should be investigated not only in terms of their direct biological effects, but also for the actual processes dictating their human bioavailability. The first step in investigating the toxicity mechanisms of nanoantimicrobials is the exploration of permeability and distribution of NPs in cell membranes. To this end, lipid nanoparticles, also known as liposomes, have been consolidated as an artificial biomembrane model to explore the potential in vitro supramolecular interactions of contaminants (e.g., inorganic nanostructures) with the phospholipids composing the membrane. In particular, NP-membrane interaction can induce membrane deformation, NP absorption or wrapping, depending on the size, composition and properties of the NP-membrane system [1]. This contribution presents an analytical chemistry holistic approach for the assessment of the interactions between ultra-small metal nanoparticles and phospholipid membranes. In particular, copper nanoparticles (CuNPs) have been taken into account, because of their increasing use in several real-life goods. Large unilamellar vesicles (LUVs) made of soybean phosphatidylcholine were used as biomimetic eukaryotic cell membranes. A fluorescence spectroscopic investigation of the interaction of CuNPs with liposomes was carried out to study their potential membranotropic effects. The individual incorporation of fluorescent membrane probes bearing naphthalene moieties (namely, Laurdan and Prodan) was used to investigate the interactions with the biomembranes [2,3]. In particular, such probes are able to penetrate into the phospholipid bilayer at different depths. Laurdan, due to the lauryl acyl chain, is located inside the membrane at the level of glycerol backbone, whereas Prodan is placed closer to the lipid-aqueous interface, at the polar moieties. Their fluorescent properties, which depend on the polarity of the microenvironment they are surrounded by, allow low resolution monitoring of changes in bilayer organization by generalized polarization (GP) measurements, thereby providing specific information about alterations in lipid order. Specifically, the results demonstrated a fluorescence hypsochromic shift, which is attributed to the progressive membrane dehydration at the hydrophobic-hydrophilic interface region of the membrane in which the probe's fluorophore is aligned. Furthermore, such results suggested an enhancement of lipid packing in the presence of CuNPs. The methodology will be extended to screen the potential risk of different antimicrobial MeNP systems.

[1] C. Contini, et al. J. Exp. Nanosci., 2018, 13, 62–81.
[2] M. Oliver, et al. Environ. Sci. Technol., 2016, 50, 7135–7143.
[3] M. Oliver, et al. Sci. Total Environ., 2020, 738, 140096.

OP1-3-2 Polarization-Modulation InfraRed Reflection Absorption Spectroscopy (PM-IRRAS) : an innovative tool for "in situ" characterization of polymer coatings.

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Polymer coating deposition on a wide variety of engineering substrates has gained significant attention. Coatings are tailored to provide and improve specific characteristics, such as corrosion, chemical, wear and weathering resistance. Characterization of polymer coatings requires the use of specific characterization techniques. Polarization-Modulation InfraRed Reflection Absorption Spectroscopy (PM-IRRAS) is an innovative and original vibrational spectroscopy that is used for "in situ" reflectivity experiments to characterize organic[1] or polymer[2] coatings deposited as thin films on reflective metallic substrates and access the mechanism of thin film formation and further structuration of polymer chains at interfaces. Due to the polarization modulation of the incident IR wave, its reflection at the interface according to surface selection rules increases the sensitivity of the spectral response, allowing determination of molecular orientation, organization, structuration or crystallization effects after polymer chains adsorption. Atomic force microscopy (AFM) analyses were also performed on the same polymer coatings in order to access the surface topology and to evidence amorphous and crystalline phases if any. Two case studies will be more precisely highlighted. First, the study of the kinetic of a film-forming process of an adhesive emulsion will be described on the basis of IR surface reflectivity measurements. The diffusion process of the different species (water, polymer, additives) during the film-drying is evidenced and the specific interactions responsible for the thermodynamic miscibility and for the film-forming are quantified. Second, the directed adsorption, structuration and crystallization of thin polymer films will be described. Various polymers were adsorbed by spin-coating on metallic substrates. The surface chemistry (hydrophilic/hydrophobic) of the substrates was controlled by chemical grafting and its influence on the organization/structuration of adsorbed polymer chains was then studied. Results show first that the competition between polymer/polymer and polymer/substrate interactions has a direct effect on the chains orientations and conformations and thus surface morphologies, and second that substrate surface chemistry alters the balance between these interactions significantly.

[1] https://doi.org/10.1080/00218464.2013.757521
[2] https://doi.org/10.1016/j.apsusc.2022.154428

OP1-3-3 Combining high sensitivity laser infrared spectroscopy with gas chromatography

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Infrared laser spectroscopy is a widely used method for the sensitive detection of small gas molecules in various applications, including atmospheric research and process engineering. The technique's chemical speciation power is based on direct probing of the chemical structure of molecules by vibrational transitions. However, selective and sensitive analysis of larger and more complex molecules

using infrared spectroscopy is currently challenging because of their complex and broadband absorption features.

At the University of Helsinki, we are developing a novel optical measurement methodology that can simultaneously detect small and large molecules with high sensitivity and chemical selectivity. This is achieved by combining high sensitivity laser spectroscopy with pre-separation of the analytes using gas chromatography (GC). A combination of GC with cantilever-enhanced photoacoustic spectroscopy (CEPAS) and an EC-QCL laser source has already been demonstrated, enabling the sensitive and selective analysis of a mixture of alcohols that would have been impossible to scrutinize with only optical spectroscopy [1].

The main obstacle in combining laser spectroscopy with GC is the inherent mismatch in sample volumes. Sensitive laser spectrometers typically employ gas cells with sample volumes of 50 - 500 mL, which is not suitable for connection to a GC column that typically has an eluted analyte peak volume of less than 1 mL. The current CEPAS cell has a sample volume of 7 mL, but further reduction of the cell volume would be beneficial for ultimate sensitivity. To address this issue, the plan is to develop the methodology further and build a miniature-volume (< 1 mL) cavity-enhanced (CE) laser spectrometer utilizing a mid-infrared optical frequency comb (OFC) as a light source. The broadband mid-IR OFC spectrum combined with GC separation will enable the simultaneous detection of a large number of analytes, and CE spectroscopy will provide high sensitivity, with projected detection limits of < 1 ppb for many analytes.

We will present the features and performance of the current GC-CEPAS setup as well as outline the projected capabilities of the planned GC-CE-OFC spectrometer.

1. Tomberg et al. Anal. Chem. 92 (2020) 14582-14588. DOI: 10.1021/acs.analchem.0c02887

OP1-3-4 On the Measurement of the Mutual Diffusivity of Binary Gas Mixtures with FTIR Spectroscopy

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Gas diffusion plays a crucial role in many applications. For instance, in membrane separation processes, concentration polarization at the material surface generates a concentration gradient in the gas phase controlled by the mutual diffusivity of the gas pair [1,2]. The latter is usually measured with a Loschmidt cell or a two bulb system by resorting to mass spectrometry or gas chromatography ex situ or to holographic interferometry in situ [3].

A novel approach based on FTIR Spectroscopy in the transmission mode is implemented to measure in situ the mutual diffusion coefficient of binary gas mixtures constituted of at least one heteronuclear chemical species. The equimolar counter-diffusion of carbon dioxide in either methane, ethane or propane is studied at ambient temperature to validate the method. The IR spectrum is collected at one end of the closed volume system and the diffusion kinetics is monitored until thermodynamic equilibrium is reached. First, the IR signals of each species are calibrated according to the Beer-Lambert relation and the protocol is described in detail [4]. Optical interferences are observed which may affect the diffusion kinetics measurement. Second, the diffusion process is modelled as in a two bulb system. The characteristic geometry of the so-defined apparatus is evaluated by modelling the diffusion of

propane in carbon dioxide with Fick's law of binary diffusion and by following the method of Arora et al. [5]. Last, the diffusion of either methane or ethane in CO_2 is investigated and modelled analogously to retrieve the mutual diffusivity. The technique also returns the concentration of each species and, as such, it allows a straightforward comparison of the experimental result with the Chapman-Enskog theory. The observed deviation is lower than 9% when the molecular interactions are modelled with the Lennard-Jones potential.

The advantages of the method over the mentioned classical approaches are multiple. The measurement is conducted in situ and the whole diffusion kinetics is analysed without treating it at steady state and without assuming a time invariant concentration gradient within system. Future perspectives refer to the extension of the method to ternary non-reacting gases and to the possibility of tuning the temperature, the pressure and the composition of the gas mixture.

[1] V. Loianno Chem. Eng. Sci. 270 (2023) 118546
DOI: 10.1016/j.ces.2023.118546
[2] V. Loianno, et al., J. Mem. Sci. 652 (2022) 120445
DOI: 10.1016/j.memsci.2022.120445
[3] P. Zangi, et al., Int. J. Thermophysics 40 (2019) 18
DOI: 10.1007/s10765-019-2484-6
[4] V. Loianno and G. Mensitieri, Meas. Sci. Technol. 33 (2022) 065017
DOI: 10.1088/1361-6501/ac5a2f
[5] Arora, et al., Rev. Sci. Instrum. 48 (1977) 673
DOI: 10.1063/1.1135105

OP1-4-1 Pyrylium based derivatization imaging mass spectrometer revealed the localization of L-DOPA

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Due its rapid metabolism, little is known about the localization of L-DOPA in the brain. We firstly visually reported the localization and function of L-dihydroxyphenylalanine (L-DOPA) which is the direct precursor of dopamine (DA) at brain by derivatized imaging mass spectrometry(1). Furthermore, to recognize whether the detected molecules are endogenous or exogenous L-DOPA, 3-place deuterated L-DOPA (D3-L-DOPA) that indicated M+3 as mass shift 3Da was injected to mouse.

Imaging mass spectrometry (IMS) can easily recognize and provide such spatial information. Following two-dimensional MS measurements on sample sections at regular intervals, reconstruction of target signals is obtained as an ion image. Thus, IMS enables simultaneous detection of multiple analytes even in the absence of target-specific markers, such as antibodies in a single experiment.

In this study, we found that 2,4,6-trimethylpyrillium tetrafluoroborate (TMPy) can selectively and efficiently react and derivatize with the target molecules. Especially, simultaneous visualization of DA and NE of metabolite from L-DOPA with high steric hinderance was archived by derivatized- IMS. TMPy-labeled-L-DOPA, DA and NE, and D3-them were detected at m/z 302.1, 258.1 and 274.1, 305.0, 261.1 and 277.1 in mice brain, respectively. Localized region of -L-DOPA and D3-L-DOPA is coincident at the brainstem (BS), a different pattern from DA and NE, which co-localize with tyrosine hydroxylase.

Interestingly, the terminal region of neuronal projections differed for the metabolites of D3-L-DOPA such as D3-DA and D3-NE compared with that of L-DOPA. Normal DA localized at caudate putamen (CPu) and nucleus accumbens; (NAcc) that are correlated with movement and cognition, and reward system. On the other hand, D3-DA preferentially located at CPu not NAcc. These findings suggest that there is a mechanism in the brainstem that allows LDOPA to accumulate without being metabolized to monoamines downstream of the metabolic pathway and deuterated DA may works better for movement and cognition. Parkinson's disease patients cannot produce DA in brain and have a reduced accumulation of DA at CPu. We may discover good material to improve symptoms of cranial nerve disease.

(1) S. Taira,et al. "Pyrylium based derivatization imaging mass spectrometer revealed the localization of L-DOPA" Plos One 17 e0271697 (2022)*

OP1-4-2 Optimization of the use of Py-Tag for next generation derivatization reagents in imaging mass spectrometry

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Imaging mass spectrometry (IMS) is well known as a usable visualization method, recently. To date, several derivatization reagents have been applied to IMS to enhance ionization efficiency and provide clear imaging of target molecules. However, it is difficult to reproduce derivatized-IMS due to the requirement for the development of individual target-specific techniques. This research reports the accurate analysis of catecholamines and amino acid using derivatization reagents (2,4,6-triethyl-3,5-dimethyl pyrylium trifluoromethanesulfonate (Py-Tag)) and discovered the difference between normal and cranial nerve disease, visually.

To reproduce this reaction on tissue sections, we constructed a reaction container to maintain humidity levels and facilitate the derivatization reaction(1). Ten different Py-Tag reaction conditions with the targets were considered. The optimal condition for the Py-Tag reaction with the targets was identified as a 70% methanol with 5% trimethylamine (v/v) solution at 60 ℃ under homogenous conditions. Resulting, visualization of dopamine (DA) and γ-aminobutyric acid (GABA) was succussed d by derivatized-IMS. Specifically, brain sections of unilateral 6-OHDA lesioned Parkinson's disease (PD) model rats showed Py-Tag DA (m/z 328.3) in the unilateral striatum and Py-Tag GABA (m/z 278.3) in the cerebral cortex, striatum, hippocampus and hypothalamus. Since 6-OHDA selectively impaired DA neurons in the striatum, this would indicate that DA was not produced in the striatum and did not project to the caudate putamen (CPu). In the midbrain, GABA showed localization in the cerebral cortex, CPu and nucleus accumbens; (NAcc), hippocampus and hypothalamus. Interestingly, 1.2 times higher intensity signal was observed in the right region of the CPu compared to the left (PD side). Thus, it is possible that GABAergic interneurons act in a compensatory capacity for DA in PD, although this hypothesis remains to be proven. Our IMS data provide indirect support for the compensatory ability of GABAergic interneurons.

These findings indicate that it is possible to achieve accurate and selective reaction, and high reaction efficiency between target molecules, and Py-Tag as well as high quality imaging of sections.

(1) H. Shikano, A. Ikeda, Y. Maejima, S. Kobayashi, T. Terauchi, Jun Yokoyama, K. Shimomura, S. Taira 'Optimization of the use of Py-Tag for next generation derivatization reagents in imaging mass spectrometry' Journal of Bioscience and Bioengineering 134, 264-268 (2022)*

OP1-4-3 Transition metal identification and speciation in cultural heritage samples by MALDI FT-ICR MS as salen complexes

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Metal complex analysis is usually performed by ICP-MS hyphenated with liquid chromatography or electrophoresis. Recently, ESI-MS has been shown to be an alternative as sensitive as ICP-MS. This work reports on a versatile and timesaving analytical protocol utilizing solely MALDI FT-ICR MS for transition metal detection and speciation after complexation with salen ligands.

The salen binding behaviour has been optimized by altering the substituent as rationalized by theoretical chemistry calculations. A synthetic original route of modified salen was found to enhance complex stability in function of the metal oxidation state to allow speciation. Analytical grade metal solutions including Co(II), Cu(I), Cu(II), Fe(II), Fe(III), Mn(II), Pb(II), Ti(IV), Zn(II), Zr(IV) pure or in mixture at different relative concentrations were treated with different Salen chelating agents, proving the complex formation. Ab-initio calculations at DFT and MP2 levels and EPR analysis were used as complementary techniques to assign the metal oxidation number in the oxidized Ligand-M^(n+) or protonated Ligand-M^(n-1)+ +H^+ species observed in the mass spectra. Cu(II), Fe(II) and Fe(III) complexes were analysed by distinct ionisation sources such as LDI, MALDI, nano-ESI and ESI ionisation. Each ionisation method can influence the oxidation state of the metal. For this reason, a deep understanding firstly of the complexation and secondly of the ionisation mechanism is necessary for speciation analysis.

Mass spectra were acquired with optimized conditions and parameters on a Bruker Solarix XR 9.4 Tesla MALDI FT-ICR instrument that presents a resolution of 1 million at 400 m/z which allows separate peaks which differs by less than a millimass unit. Starting from metal at different oxidation state we observed complexes of different formula for example for iron [salen-2H, Fe(III)]^+ and [salen-H, Fe(II)]^+ as the first isotope of [salen-2H, ^{56}Fe(III)]^+ is cleanly separated from the monoisotopic peak of [salen-H, ^{56}Fe(II)]^+.

The analytical protocol was applied to the identification and speciation of transition metal in cultural heritage samples for examination and restoration purposes. We develop for pigments a mild dissolution with a mixture of a hydrochloric and hydrofluoric acid followed by a reverse phase SPE tip purification. We then applied this workflow to pigments extracted from modern and historical paint tubes, paintings and dryers, such as Kortrijk dryer samples.

OP1-4-4 Determination of hydrolysis products of organophosphorus nerve agents in soil and plant materials using liquid chromatography and tandem mass spectrometry

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Organophosphorus nerve agents (OPNAs) were originally developed as a type of chemical warfare agent in the early 20th century. Their high toxicity is related to the irreversible inhibition of the enzyme acetylcholinesterase, resulting in cholinergic syndrome (mental confusion, convulsions or tremors, and in some cases, death). Despite the presence of the Chemical Weapons Convention (CWC), there is still a threat of incidents of CWs illegal use due to current terrorist attacks and regional conflicts.

The most toxic representatives of OPNA's include VX, CVX and VR, which are highly reactive. Once in the organism, the processes of their rapid degradation and metabolism occur. Therefore, it is relevant to identify their long-lived degradation products or so-called OPNAs markers.

To date, there is a large number of publications devoted to the determination of OPNA's metabolites. Biological objects (blood plasma, urine) and environmental objects (water, soil) are the most studied. Plants, in contrast, are poorly examined for verifying CWs misuse. However, plant material is still a promising object due to the ability to accumulate metabolites for a long time. The latter makes it possible to develop a more reliable and retrospective approach for the determination of CWs.
In the present work the following stable transformation products of organophosphorus compounds - methylphosphonic acid (MPA) and some of its alkyl esters or AMPAs (ethyl-, isopropyl-, isobutyl-, cyclohexyl-, pinacolyl-) are studied as OPNAs in their native form undergo structure changes during transformation processes. The presence of these metabolites in samples can reliably confirm the use of nerve agents.

The aim of this research was to study the process of MPA and AMPAs accumulation in the widespread plant Hedera Helix which grew in contaminated soil, by using a developed and validated high-performance liquid chromatography-tandem mass spectrometry (HPLC-MS/MS) analytical approach. Selected markers were applied once to the soil, and their content was monitored for four weeks.
A fast and simple method of sample homogenization with liquid nitrogen followed by ultrasonic-assisted liquid extraction was used to extract the analytes. Determination was performed using HPLC-MS/MS. The developed approach for HPLC-MS/MS-identification and quantitative analysis of toxic substances' metabolites using deuterated internal standards allows to detect all studied markers in both soil and plant objects for at least one month. Therefore, in addition to well-studied environmental objects such as soil and water, plants with their further analysis can become a powerful tool to verify the alleged use of chemical weapons.

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OP2-1-1 Continuous biomarker monitoring with single molecule resolution by measuring free particle motion

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Sensing technologies that can continuously monitor concentration levels of biomolecules are needed for applications such as patient care, fundamental biological research, biotechnology, food industry, as well as the environment. However, it is fundamentally difficult to develop measurement technologies that are not only sensitive and specific, but also allow monitoring over broad concentration ranges and over long timespans. [1-3]

Here we present a continuous biomolecular sensing methodology based on the free diffusion of biofunctionalized particles hovering over a biofunctionalized sensor substrate [4]. The method records digital events of particles switching between bound and unbound states caused by reversible single-molecule interactions. The sensor enables continuous biomarker monitoring at picomolar to micromolar concentrations without consuming any reagents. Measured bound and unbound state lifetimes give insight into molecular interaction dynamics and analyte concentration in the sample.
The affinity-based sensing methodology is demonstrated for oligonucleotide sandwich and competition assays, and for an antibody-based cortisol assay. We present results on the monitoring of an inflammatory protein (the cytokine TNF-α) and on long-term dry storage of sensor cartridges. Finally, we discuss how the continuous monitoring sensor can be applied for studies on organs-on-a-chip, for the monitoring of patients in critical care, and for the monitoring of industrial processes and bioreactors as well as ecological systems.

[1] Steinmetz, L.M. et al. Mol. Syst. Biol. 12, (2016). https://doi.org/10.15252/msb.20166873
[2] Wang, B. et al. Front. Bioeng. Biotechnol. 8, (2020). https://doi.org/10.3389/fbioe.2020.00007
[3] Lubken, R.M., et al. ACS Sensors 6, (2021). https://doi.org/10.1021/acssensors.1c01991
[4] Buskermolen, A.D., et al. Nat. Commun. 13, (2022). https://doi.org/10.1038/s41467-022-33487-3

OP2-1-2 Using a 3D printer for low-cost construction of the sensing areas of self/rapid tests

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The great usefulness of self/rapid tests has been established since the outbreak of SARS-CoV-2 pandemic. Scientific community has turned towards alternative approaches of low-cost, rapid and easy-to-use that do not require trained personnel. To this concept, we have integrated, for the first time, an affordable 3D printer with inexpensive tools such as rapidograph, disposable pipette tips and 3D-printed accessories for the construction of the control and the test areas (zones and spots) of self/rapid tests. The produced tests were applied for the detection of nucleic acids, both single and double-stranded DNA and microRNA sequences corresponding to the latter, and for proteins/antibodies. We also used this developed system for the construction of multiple test spots on the membrane of the tests for multiplex analysis. Various optimizations studies were performed regarding the concentration of the dispensed reagents, the numbers of the passes of the dispenser across the membrane and the printing speed (i.e. deposition speed). The analytical performance, by terms of sensitivity to different target concentrations and repeatability, of the proposed system was also evaluated and compared to a commercially available dispensing system. Our proposed low-cost

system showed similar results to the commercial one with very good repeatability (%CV values ranging from 0.3 to 7.6%), without the need of gas or expensive automated pumps for the dispensing of the reagents onto the membrane of the tests. In conclusion, the developed 3D printer-based system is a simple, inexpensive and easy alternative for self/rapid tests production that can be easily adopted by any research laboratory for various analytes and applications.

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OP2-1-3 Gold Decorated Polyaniline toward Glucose Oxidation

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Along with the rise of diabetes mellitus issue, glucose sensor has become an imperative tool for healthcare. Among glucose sensors, electrochemical glucose sensors have attracted attention because of their simplicity, sensitivity, short response time, and selectivity. Electrodes used in electrochemical glucose sensors are composed of a support matrix and a catalytic material capable of catalyzing the oxidation of glucose. Conducting polymers are ideal supporting materials because of the ease in the preparation by electro-polymerization and manipulation of the properties by the electrochemical conditions. In addition, a supporting material with a high specific surface area is beneficial for dispersion of the catalytic materials on the surface. Precious metals are known to have high catalytic activity in electro-oxidation of glucose [1, 2]. Metal oxide catalyst, on the other hand, has also been widely investigated for non-enzymatic glucose sensors. In this study, polyaniline (PANI) was used as the support. Gold, TiO_2 and MnO_2 nanoparticles (NPs) were the catalytic materials. The performance as the cataltyic electrode in non-enzymatic glucose sensor was evaluated.

The PANI film was prepared by electro-polymerization in 2 M tetrafluoroboric acid (HBF4) containing 0.1 M aniline monomer [2]. The TiO_2 particles were incorporated into the PANI by co-electrodeposition method. The MnO_2 were decorated by direct-dropping $KMnO_4$ solution onto the PANI coated electrode. The gold NPs were decorated by direct-dropping $KAuCl_4$ solution onto surfaces of the MnO_2-PANI composite. No enzymes were used in preparation of the catalytic electrode. The amount and size of the gold NPs and MnO_2 NPs were both found to be dependent on amount of the $KAuCl_4$ and $KMnO_4$ solutions dropped onto the PANI electrode. The performance in electro-oxidation of glucose was evaluated by cyclic voltammetry (CV) in 0.1 M KOH containing 0~50 mM of glucose.

The result showed that the decoration of gold NPs was beneficial to improve the catalytic activity, and the performance further improved after blending with TiO_2 [1] and decoration of MnO_2 [2]. The Au-MnO_2/PANI and Au-TiO_2/PANI composites showed current responses in the CV of solutions containing comparable glucose concentrations as the normal physiological levels of glucose concentration in human blood. The result confirmed the applicability of the Au-MnO_2/PANI and Au-TiO_2/PANI composites as the catalytic electrode in non-enzymetic glucose sensors.

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[2] 10.1016/j.mne.2023.100175

OP2-2-1 Time Efficiency: A Wonderful but Little-known Performance Indicator in Separation Sciences

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While the concept of 'separation efficiency' (expressed by the number of theoretical plates, N) is well known and widely adopted as performance indicator in separation sciences, the now well defined 'time efficiency' [1] indicator remains mostly unknown and underexplored. It is defined as the ratio between the number of theoretical plates (N) divided by separation time squared (N/t2). Separation time is known as elution time in chromatography and migration time in electrophoresis. In this presentation, the concepts of Separation Efficiency (N) and Time Efficiency (N/t2) will be presented in a very didactic and illustrated manner. Moreover, it will be shown that the use of these two definitions, defined above, produces dozens of very compact and beautiful performance indicator equations in separation sciences. These include, but are not limited to: 1) height equivalent of a theoretical plate, 2) resolution between two neighboring peaks or bands, 3) resolution per unit time, 4) band capacity, 5) band capacity per unit time, 6) peak capacity, and 7) peak capacity per unit time.

The equations of N, N/t2, and the above-mentioned seven performance indicators, expressed as a function of the operation parameters (V, E, μ, D, and separation length), will be presented for both the common (Open) Electrophoresis layout and for the Toroidal Electrophoresis layout [1,2]. Toroidal electrophoresis uses a separation medium with a closed loop geometry, i.e., toroidal slabs in gel electrophoresis, a closed loop channel in toroidal microchip electrophoresis or a torus made of fused silica in toroidal capillary electrophoresis. Toroidal capillary electrophoresis, despite being more complicated to operate, has already been shown to produce a separation efficiency of more than 100 million theoretical plates [3,4], the highest separation efficiency reported in the literature so far. Considering this, what is the world record for time efficiency (N/t2)? Why is time efficiency so important, and how can it be optimized? All of these questions will be didactically answered in this presentation.

References
[1] Kist, T.B.L. Open and Toroidal Electrophoresis. 2020, Chichester: Wiley. See tables 5.4 and 5.5. ISBN-10: 1119539404
[2] Kist, T.B.L. Number of theoretical plates achievable by a toroidal capillary electrophoresis system. Journal of Separation Science 40: 4619-4627, (2017).
[3] Zhao J., Hooker T., Jorgenson J. W. Journal of Microcolumn Separations 11: 431-437, (1999).
[4] Zhao J., Jorgenson J. W. Journal of Microcolumn Separations 11: 439-449, (1999).

OP2-2-2 Continuous manufacturing of monoclonal antibodies: Dynamic control of multiple integrated polishing chromatography steps using BioSMB

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We propose a strategy for automation and control of multi-step polishing chromatography in integrated continuous manufacturing of monoclonal antibodies. The strategy is demonstrated for a multi-step polishing process consisting of cation exchange chromatography in bind-and-elute mode followed by mixed-mode chromatography in flowthrough mode. A BioSMB system with a customized

Python control layer is used for automation and scheduling of both the chromatography steps. Further, the BioSMB valve manifold is leveraged for in-line conditioning between the two steps, as tight control of pH and conductivity is essential when operating with multimodal resins because even slight fluctuations in load conditions adversely affect the chromatography performance. The pH and conductivity of the load to the multimodal chromatography columns is consistent, despite the elution gradient of the preceding cation exchange chromatography step. Inputs from the BioSMB pH and conductivity sensors are used for real-time control of the 7 pumps and 240 valves to achieve in-line conditioning inside the BioSMB manifold in a fully automated manner. This is confirmed by showcasing different elution strategies in cation exchange chromatography, including linear gradient, step gradient and process deviations like tubing leakage. In all the above cases, the model was able to maintain the pH and conductivity of multimodal chromatography load within the range of 6 ± 0.1 pH and 7 ± 0.3 mS/cm conductivity. The strategy eliminates the need for using multiple BioSMB units or integrating external pumps, valves, mixers, surge tanks, or sensors between the two steps as is currently the standard approach, thus offering a simple and robust structure for integrating multiple polishing chromatography steps in continuous downstream monoclonal antibody purification trains.

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OP2-2-3 A native multi-dimensional monitoring workflow for at-line characterization of mAb titer, size, charge, and glycoform heterogeneities in cell culture supernatant

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With growing maturity of the biopharmaceutical industry, new modalities entering the therapeutic design space and increasing complexity of formulations such as combination therapy, the demands and requirements on analytical workflows have also increased. A recent evolution in newer analytical workflows is that of multi-attribute monitoring workflows designed on chromatography-mass spectrometry (LC-MS) platform. In comparison to traditional one attribute per workflow paradigm, multi-attribute workflows are designed to monitor multiple critical quality attributes through a single workflow, thus reducing the overall time to information and increasing efficiency and throughput. While the 1st generation multi-attribute workflows focused on bottom-up characterization following peptide digestion, the more recent workflows have been focussing on characterization of intact biologics, preferably in native state. So far intact multi-attribute monitoring workflows suitable for comparability, utilizing single dimension chromatography coupled with MS have been published. In this study, we describe a native multi-dimensional multi-attribute monitoring workflow for at-line characterization of monoclonal antibody (mAb) titer, size, charge, and glycoform heterogeneities directly in cell culture supernatant. This has been achieved through coupling ProA in series with size exclusion chromatography in 1st dimension followed by cation exchange chromatography in the 2nd dimension. Intact paired glycoform characterization has been achieved through coupling 2D-LC with q-ToF-MS. The workflow with a single heart cut can be completed in 25 mins and utilizes 2D-liquid chromatography (2D-LC) to maximize separation and monitoring of titer, size as well as charge variants.

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OP2-3-1 Absolute quantification of pure free radical reagents by combination of effective magnetic moment method and quantitative electron paramagnetic resonance method

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An absolute quantitative analysis of free radicals in a diamagnetic matrix by combining "effective magnetic moment method" and the quantitative electron paramagnetic resonance [qEPR] method is proposed. This combined method utilizes the advantages of both the analytical methods and compensates for their disadvantages.

The principle of the effective magnetic moment method is based on both Curie-Weiss law and EPR fundamental equation. The temperature dependence of magnetic moment under a magnetic field is measured using a superconducting quantum interference device [SQUID]. Resonant magnetic field was measured using a EPR spectrometer. The number of free radicals in sample can be obtained by both results of SQUID and EPR measurements. However, the effective magnetic moment method has some disadvantages;(1) the measurement time of SQUID in a wide temperature region from 4 K to room temperature is long (more than 24 h). (2) If the interaction between the radicals becomes stronger with a decrease in the sample temperature, the magnetic moment deviates from the Curie−Weiss law in the lower temperature region.

On the other hand, the qEPR method compares a “primary standard sample” and “secondary standard sample”, with some advantages; (1) short-time measurement at room temperature, typically, the measurement time of one sample is within 10 min, and no cryogen is necessary for room-temperature measurements; (2) even if a free-radical chemical has a significant interaction with the radicals in the low-temperature region, it is expected that the chemical obeys the Curie−Weiss law at room temperature; (3) only the paramagnetic moment of the free radicals is detected. However, the qEPR method needs the primary standard sample with known purity.

The qEPR method using the primary standard sample with purity determined by the effective magnetic moment method realizes a simple purity analysis of free radical reagents with traceability to the International System of Units (SI). The purity of the free radicals by the qEPR method for pure 4-hydroxy-2,2,6,6-tetramethylpiperidine 1-oxyl benzoate [4HTB], 1-Oxyl-2,2,6,6-tetramethyl-4-hydroxypiperidine [TEMPOL], and di(phenyl)-(2,4,6-trinitrophenyl)iminoazanium [DPPH] reagents was obtained with a relative expanded uncertainty of 0.7% for the 4HTB to 1.5% for the DPPH. Some purity values of the free radicals for these reagents differed from those stated by the manufacturers. This combined method enables short-time quality control of pure radical reagents, instead of quality control by separation analytical methods or titrations.

Ref.) Matsumoto, N.; Itoh, N; Yamasaki, K., Anal. Chem. 94, 2022, 12595-12603
10.1021/acs.analchem.2c01005

OP2-3-2 Vibrational spectroscopy of blood plasma glycoproteins

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Glycosylation is the most common and most complex post-translational modification: more than half of human proteins contain at least one glycan, while the total number of glycan structures reaches several thousands.[1] Protein glycosylation carries a wealth of information about the health state of an individual, and analytical science only starts to tap into this direction.[2] Even glycoforms of classical plasma proteins are informative for disease detection,[3–5] nevertheless their glycosylation patterns are rarely evaluated.

This situation calls for a robust, simple analytical workflow that could flag relevant changes in the glycosylation of a wide range of proteins. We put forward infrared (IR) spectroscopy: it is sensitive to alternations in the glycan structures and covers both N- and O-glycans in a label-free manner.[6] Vibrational spectroscopy is particularly suited for quantitative analysis of glycosylation, since the signals originating from glycans arise at a different spectral range than the major protein backbone features.[7] Therefore, intact proteins can be investigated, eliminating the need for glycan release. Previously, IR spectroscopy has been proposed, for example, as a tool to compare glycosylation in therapeutic monoclonal antibodies.[8,9] Albeit promising, it has not yet, to our knowledge, been applied to proteins derived from human blood plasma.

Typically, purification of plasma proteins is a multi-step analyte-specific procedure.[10] Fortunately, advances in sample handling make analysis of intact proteins from crude samples possible.[11] In line with that, we developed an analytical workflow for measuring vibrational spectra of plasma proteins: using ion exchange (IEX) chromatography[12,13] we separate a blood plasma sample into several fractions, de-salt and concentrate them with centrifugal filters and perform an IR absorption measurement.

Our results demonstrate that infrared spectroscopy can identify different patterns and global levels of glycosylation of intact proteins. We confirm our data interpretation using a well-established glycomic workflow: glycan release, fluorescent labeling and UHPLC-MS analysis of the same samples that were investigated spectroscopically.

Next, we perform spiking experiments that model a biomarking scenario: supposedly, glycosylation of a particular protein is affected by certain disease and this change needs to be spectroscopically detected. We confirm that chromatographic separation significantly improves the detection capabilities. Stepping away from the model system, we discuss the potential applications of infrared spectroscopy of blood glycoproteins to disease detection using the example of breast cancer.
Generally, we will discuss how the simplicity of a spectroscopic measurement helps uncover the richness of the biological information contained in protein glycosylation.

(1) Reily, C.; Stewart, T. J.; Renfrow, M. B.; Novak, J. Glycosylation in Health and Disease. Nat. Rev. Nephrol. 2019, 15 (6), 346–366. https://doi.org/10.1038/s41581-019-0129-4.
(2) de Haan, N.; Wuhrer, M.; Ruhaak, L. R. Mass Spectrometry in Clinical Glycomics: The Path from Biomarker Identification to Clinical Implementation. Clin. Mass Spectrom. 2020, 18, 1–12. https://doi.org/10.1016/j.clinms.2020.08.001.
(3) Clerc, F.; Reiding, K. R.; Jansen, B. C.; Kammeijer, G. S. M.; Bondt, A.; Wuhrer, M. Human Plasma Protein N-Glycosylation. Glycoconj. J. 2016, 33 (3), 309–343. https://doi.org/10.1007/s10719-015-9626-2.
(4) Bones, J.; Mittermayr, S.; O'Donoghue, N.; Guttman, A.; Rudd, P. M. Ultra Performance Liquid Chromatographic Profiling of Serum N -Glycans for Fast and Efficient Identification of Cancer Associated Alterations in Glycosylation. Anal. Chem. 2010, 82 (24), 10208–10215. https://doi.org/10.1021/ac102860w.
(5) Gudelj, I.; Lauc, G.; Pezer, M. Immunoglobulin G Glycosylation in Aging and Diseases. Cell. Immunol. 2018, 333 (January), 65–79. https://doi.org/10.1016/j.cellimm.2018.07.009.

(6) Derenne, A.; Derfoufi, K.-M.; Cowper, B.; Delporte, C.; Claire I. Butre; Goormaghtigh, E. Mass Spectrometry of Glycoproteins. In Mass Spectrometry of Glycoproteins; Delobel, A., Ed.; Humana Press, 2021; p 401. https://doi.org/https://doi.org/10.1007/978-1-0716-1241-5.
(7) Khajehpour, M.; Dashnau, J. L.; Vanderkooi, J. M. Infrared Spectroscopy Used to Evaluate Glycosylation of Proteins. Anal. Biochem. 2006, 348 (1), 40–48. https://doi.org/10.1016/j.ab.2005.10.009.
(8) McAvan, B. S.; France, A. P.; Bellina, B.; Barran, P. E.; Goodacre, R.; Doig, A. J. Quantification of Protein Glycation Using Vibrational Spectroscopy. Analyst 2020, 145 (10), 3686–3696. https://doi.org/10.1039/c9an02318f.
(9) Derenne, A.; Derfoufi, K. M.; Cowper, B.; Delporte, C.; Goormaghtigh, E. FTIR Spectroscopy as an Analytical Tool to Compare Glycosylation in Therapeutic Monoclonal Antibodies. Anal. Chim. Acta 2020, 1112, 62–71. https://doi.org/10.1016/j.aca.2020.03.038.
(10) Nickerson, J. L.; Baghalabadi, V.; Rajendran, S. R. C. K.; Jakubec, P. J.; Said, H.; McMillen, T. S.; Dang, Z.; Doucette, A. A. Recent Advances in Top-down Proteome Sample Processing Ahead of MS Analysis. Mass Spectrom. Rev. 2021, 1–39. https://doi.org/10.1002/mas.21706.
(11) Vimer, S.; Ben-Nissan, G.; Sharon, M. Mass Spectrometry Analysis of Intact Proteins from Crude Samples. Anal. Chem. 2020, 92 (19), 12741–12749. https://doi.org/10.1021/acs.analchem.0c02162.
(12) Ahamed, T.; Nfor, B. K.; Verhaert, P. D. E. M.; van Dedem, G. W. K.; van der Wielen, L. A. M.; Eppink, M. H. M.; van de Sandt, E. J. A. X.; Ottens, M. PH-Gradient Ion-Exchange Chromatography: An Analytical Tool for Design and Optimization of Protein Separations. J. Chromatogr. A 2007, 1164 (1–2), 181–188. https://doi.org/10.1016/j.chroma.2007.07.010.
(13) Tomono, Tsljgikazu; Ikeda, H.; Tokunaga, E. High Performance Ion Exchange Chromatography of Proteins. J. Chromatogr. 1983, 266, 39–47. https://doi.org/10.2116/bunsekikagaku.33.7_E287.

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Driven by increasing quality requirements for peptide drug substances, their increasing complexity and due to Bachem's expansion into oligonucleotides, we introduced NMR spectroscopy as a new analytical technique in our QC department. In here, we would like to give an overview about the implementation process and challenges during evaluation and qualification as an example for complex analysis instruments in an GMP environment.
The hardware requirements were mainly defined by our focus on quality control of starting materials especially for oligonucleotide syntheses and the option for a high sample throughput. As it is the first NMR spectrometer at Bachem, a room fulfilling the hardware specifications regarding temperature stability, fringe fields and interfering vibrations had to be found and rebuild.

The demands on cGMP compliance and data integrity as laid out within 21 CFR Part 11 are increasing rapidly and the software for acquisition and evaluation is hence in focus of the authorities. This can well be seen on the amount of current FDA warning letters dealing with data integrity and is underlined by the fact, that Swissmedic decided to review the qualification documents and our first method validation. Thus, we had a detailed view on data integrity. A new software called Mdrive was offered, which promises to close the gaps present in standard acquisition/processing software. However, several issues were identified and resolved during our internal qualification.

Although NMR spectroscopy is routinely used in analytical laboratories, standard software packages are not necessarily fulfilling the needs defined by cGMP requirements. NMR Software generally needs to improve to satisfy the increasing demands from the authorities as well as those of a routine lab. Currently, various gaps remain to be solved using individual workarounds which led in our case to several compliance events and tedious adjustments on operating system level.

OP2-4-1 Rapid profiling the glycosylation effects on cellular entry of SARS-CoV-2 using MALDI-MS with high mass detection

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Introduction

The entry of SARS-CoV-2 viruses is mediated by the binding of the spike (S) protein of SARS-CoV-2 to the angiotensin-converting enzyme 2 (ACE2) receptor on cell surfaces. Both the S protein and ACE2 are glycosylated, which could affect the binding of S with ACE2. In recent years, high mass matrix assistant laser ionization/desorption mass spectrometry (HM-MALDI-MS) has become a robust technique for the quantitative analysis of protein-protein interactions (PPIs). Here, we utilized HM-MALDI-MS for profiling the effects of glycosylation on the binding of the S protein receptor binding domain (RBD) to ACE2.

Methods
RBD and ACE2 purchased from Sino Biological were reconstituted in Milli Q water. Enzymatic glycosylation modifications were carried out to obtain proteins with differences in their glycosylation pattern. RBD and ACE2 samples were mixed with different molar ratios. The binding was carried out at 25 °C for 1h and then crosslinked with BS(PEG)9 for 1h. Sample spots were prepared by the sandwich method (using saturated sinapinic acid (SA) + sample + SA) and analyzed using an Applied Biosystems 4800 MALDI TOF/TOF instrument with a high mass detector (CovalX). The area ratios between the RBD•ACE2 complex and ACE2 peaks were calculated and plotted against the RBD/ACE2 molar ratios to obtain a titration curve.

Results
Partially deglycosylated ACE2 (96 kDa) showed much better binding compared to ACE2 carrying the full glycosylation (100 kDa). Removing the terminal sialic acids also enhanced the RBD•ACE2 binding, while removing galactose of ACE2 resulted in decreased binding. RBD without terminal sialic acids also showed improved binding to ACE2. In the human body, ACE2 is a homodimer; it can also form dimers in solution. Dimerization of ACE2 was also affected by its glycosylation pattern. Deglycosylation and desialylation could improve the dimerization, while degalactosylation reduced the dimerization. The mass spectra of a high concentration RBD / ACE2 mixture (5 µM in solution) showed multiple binding possibilities between the proteins and protein aggregates: the interaction was dominated by the binding of RBD monomer to ACE2 monomer and dimer. An ACE2 dimer can bind two RBD monomers. We can also study the binding of RBD to ACE2 monomer and of the dimer at the same time, results showed that an ACE2 dimer exhibited higher binding affinity to RBD than ACE2 monomer.

Novel Aspect
Rapid profiling the glycosylation effects on protein-protein interaction and revealing the complex interactions between proteins and protein aggregates.

OP2-4-2 Comparative Analysis of Haemoglobin Solution and Gas Phase Stability Using Mass Spectrometry

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Native ion mobility-mass spectrometry (IM-MS) can be used to determine the structural characteristics of biomolecules. This technique is used to measure the time an analyte takes to transverse a gas-filled cell when pushed by an electric field (drift time). A biomolecule's area can be determined from its drift time, charge state, as well as IM-MS instrument parameters. Since the combination of IM and MS allows for ions to be distinguished based on mass and area, multiple analytes can be measured simultaneously. This allows for the elucidation of structural information not readily obtainable by other techniques.

The structural stability of an analyte can also be investigated by IM-MS. This is achieved by subjecting ions to collisional energy prior to the drift time measurement. By plotting the change in the analyte drift time as a function of collisional voltage, it becomes possible to map its gas-phase unfolding pathway. This provides a fast method for gaining detailed insights into analyte stability with low sample consumption. However, questions remain about the comparability of gas phase and solution stabilities, as the experiments are performed in different mediums. To the best of our knowledge, only one study has compared these stabilities in detail, which showed that gas phase stability correlates with protein melting temperature.[1] The paucity of data for comparing gas and solution stabilities warrants further studies. This could be achieved by comparing the stabilities of haemoglobin in solution and the gas phase. Haemoglobin is a well-characterized protein, the stability of which varies depending on which animal it is obtained from as well as solution conditions. The ability to generate multiple, structurally similar analytes of different stabilities makes haemoglobin ideal for this comparative analysis.
Most techniques used for melting experiments only provide a low-resolution picture of what is happening in the solution. One MS technique that aims to overcome this is temperature-controlled nanoelectrospray (TC-nESI).[2] Briefly, this technique allows for the continuous analysis of several biomolecular classes and has been shown to provide comparable data to other solution-based techniques. Also, TC-nESI is uniquely positioned for this type of comparative analysis. Since both are MS-based methods, they can be coupled together to better understand the nature of the interactions responsible for stability differences in biomolecules. In this work, the solution, and gas-phase stabilities of haemoglobin from different sources were compared.

L. Nouchikian, et al., JASMS, 31, 2020, https://doi.org/10.1021/jasms.9b00130
J. A. Harrison, et al., Chem. Eur. J. 27, 2021, https://doi.org/10.1002/chem.202102474

OP2-4-3 In vitro and in vivo assessments of metabolic stability, pharmacokinetic and pharmacodynamic properties of a potent dual inhibitor of 5-lipoxygenase and soluble epoxide hydrolase by mass spectrometry-based approaches

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The design of multitarget drugs represents a promising strategy in medicinal chemistry and seems particularly suitable for the discovery of anti-inflammatory drugs. Here, we describe the identification of an indoline-based compound inhibiting both 5-lipoxygenase (5-LOX) and soluble epoxide hydrolase (sEH).

In silico analysis of an in-house library identified nine promising compounds but enzymatic and cellular assays revealed 73 compound as potential dual 5-LOX/sEH inhibitor. Considering the data collected from the in vitro activity screening, we decided to perform an in vitro pharmacokinetic evaluation of 73 compound.

The chemical stability of the compound was first studied in phosphate buffer at pH 7.4, then in mouse plasma and finally, after incubation with liver microsomes. In this assay the metabolism of the parent compound over time was measured by LC-MS in the presence of CYP450 and UGTs microsomal systems and the metabolites were tentatively characterized by their accurate mass, fragmentation pattern and retention times. Finally, pharmacokinetic assessment demonstrated a suitable profile of 73 in vivo (t1/2: 1.6h; Cmax: 15.4 ng mL-1; AUC0–∞: 42.7 ng/mL h).

Finally, compound 73 was challenged in murine model of zymosan-induced peritonitis. Compound 73 displays in vivo anti-inflammatory effects in decreasing the leukotrienes levels as well as cell infiltration and the levels of proinflammatory mediators in the peritonitis model. Finally, to confirm in vivo sEH inhibition, we measured the levels of several epoxy- and dihydroxy-unsaturated fatty acids in peritoneal exudate upon compound 73 administration, using AUDA as a positive control. LC-MS/MS analysis showed that the epoxy-unsaturated fatty acid levels were about 8 times higher than the corresponding dihydroxy-unsaturated fatty acids in both 73- and AUDA-treated mice.

In conclusion, this evidence suggests that compound 73 can be taken into consideration for further development as a therapeutic tool in inflammatory diseases.

I. Cerqua, et al., J. Med. Chem., 65, (2022) 14456
https://doi.org/10.1021/acs.jmedchem.2c00817
S. Codony et al., J. Med. Chem. 2022, 65, 6, 4909–4925
https://doi.org/10.1021/acs.jmedchem.1c02150

OP2-4-4 Considerations for developing an analytical strategy for fast small molecule MS-based screening in complex samples in industrial biotechnology

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DSM is a company active in Health, Nutrition and Bioscience. In our bioprocess and bioproducts R&D, mass spectrometry is one of the core technologies used in our labs for small molecule analysis to generate insight for microbial strain development and screening.

In this presentation we will show an overview of different technologies investigated for fast MS analysis for various classes of small molecules. An extra challenge in this investigation is the fact that the targeted metabolites can be of very different chemical composition for different innovations and the same is true for the microbial matrix of interest. As a result, the analytical technologies should be widely applicable for various classes of compounds. We will show examples for amino acids, organic acids and peptides of fast LC-MS analysis, application of dual LC/multiplexing LC-MS and flow injection analysis (FIA)-MS. In addition, we also investigated external technologies like fast solid-phase extraction (SPE)-MS and acoustic droplet injection (ADE)-MS as high-throughput alternatives.

Normal LC-MS runs last between 5-10 min. Pushing the limits of UHPLC-MS typical can reduce runtimes to 2-3 minutes with still sufficient separation. The next step is tackling the inefficient use of the MS due to washing and equilibration steps by performing LC-MS in multiplexing mode with which the efficient runtime can be reduced by roughly a factor of 2. Separation however takes time, so leaving the separation out will result in a further significant decrease in runtime. Flow injection analysis by MS (FIA-MS) is an example of this with typical runtimes of 30-60 sec, but with no separation and therefore more matrix interference. If even more speed is necessary, more dedicated equipment can be used, like fast SPE-MS with typical runtimes of 10-20 seconds or ADE-MS with runtime of only 1-3 seconds.

Each of the technologies presented have their specific advantages and the specific bioproduct development requirements will determine which approach for small molecule analysis is the most appropriate, there is no one-size-fits-all solution. The subtle balance between speed, separation, quantification, robustness and costs and the expected (future) needs will determine which of these technologies are the right analytical strategy for each small molecule MS-based screening.

OP3-1-1 Application of aptamer-based biosensors for electrochemical detection of heavy metal cations

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Concerning the constant development of industry, it may yield not only some new advancements or technologies but also require the usage of chemicals that can be harmful to environment and human health. As a result, this might cause the damage to environment as well as occurrence of diseases in plant, animal, and human organisms. One of the major threats is the possibility of accumulation of heavy metal ions for example in human organism which might pause a harmful effect even at low concentrations [1]. Hence, it is necessary to elaborate techniques which could provide early, sensitive, and selective detection of heavy metal cations. Traditionally, atomic absorption spectroscopy (AAS), inductively coupled plasma/ atomic emission spectrometry (ICP – AES) or inductively coupled plasma mass spectrometry (ICP-MS) were applied for that purpose [2]. One of the major disadvantages of above-mentioned techniques is the necessity of laborious sample preparation, the use of sophisticated equipment as well as limited portability.

An interesting alternative could be application of electrochemical biosensors utilizing aptamers as receptor elements. Aptamers are known as short, single-stranded DNA or RNA strands which change their conformation upon binding with a specific analyte including cations, proteins, or cells [3]. Further introduction of modifications into the sequence allows for their surface immobilization, conjugation with fluorescence or redox active labels or enhancement of stability. Therefore, there has been a dozen examples of usage of aptasensors also for determination of heavy metal ions.

Herein we present studies on the detection of heavy metal ions using aptamer-based layers concerning metal cations. For the development of sensing layer DNA aptamers were utilized and the research

concerned the choice of the content of receptor layer as well as experimental conditions for electrochemical studies including the selection of redox active indicator. Based on conducted experiments the working parameters of proposed aptasensors were defined including the lower limit of detection, range of linear response as well as selectivity.

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[1] X. Zheng, et al., Rev. Anal. Chem. 39 (2020) 231.
DOI:10.1515/revac-2020-0118
[2] B. Feist, et al., J. Hazard. Mater. 152 (3) (2008) 1122.
DOI:10.1016/j.jhazmat.2007.07.095
[3] S. Song, et al., TrAC Trends Anal. Chem. 27 (2008) 108.
DOI:10.1016/j.trac.2007.12.004

OP3-1-2 Electrochemical bioplatform for interrogating the most common and carcinogenic human papillomavirus DNA

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Cervical cancer is one of the most common types of cancer among women worldwide. Although it is a theoretically preventable cancer among female genital cancers, it is an important cause of death, especially in developing and underdeveloped countries. Human papilloma viruses (HPV) are the most common sexually transmitted disease, and most cervical cancers (99.7%) are related to HPV. Among the identified HPV genotypes, HPV 16 and 18 cause 70% of cervical cancers thus, they are considered as high-risk genotypes(1). Electrochemical bioplatforms, continue to unfold their competitiveness versus conventional strategies as powerful tools for in situ, affordable, rapid, portable, sensitive, and specific assays of target nucleic acids.

This work reports a new bio tool for the determination of HPV16 DNA. The developed bioplatform combines the advantages of using MBs as scaffolds for the implementation of the selected bioassay format by exploiting two different affinity reactions: DNA/RNA hybridization and the recognition of the formed heteroduplexes with an antibody able to recognize an epitope of only 6 bp. Finally, the prepared magnetic bioconjugates were trapped on screen-printed carbon electrodes (SPCE) for amperometric transduction (2).

The biotool achieved a low limit of detection (0.5 pM) for the synthetic HPV16 target DNA and it was able to discriminate between HPV16 positive and negative human cancer cells using only 25 ng of amplified DNA in just 45 min.

The bioplatform exhibits some advantages over other available analytical methods utilized in routine clinical practice and in clinical laboratories in terms of cost and applicability at the point of care, it being also advantageous with respect to other reported electrochemical methodologies in terms of simplicity and required assay time.

Moreover, the versatility of the strategy allows determining RNAs instead of DNAs by simply changing the capture probe and, the multiplexing compatibility of the developed strategy, make it easily extrapolated to develop platforms for the simultaneous detection of hrHPV DNAs and the mRNAs encoding E6 and E7 proteins, which significantly affect the oncogenic properties of hrHPVs, thus allowing to know whether the virus is active and to specifically discriminate between transient or potentially progressive lesions.

1 Q. Lv, Y. Wang, C. Su, T. Lakshmipriya, S. C. B. Gopinath, K. Pandian, V. Perumal and Y. Liu, Int. J. Biol. Macromol., 2019, 134, 354–360.DOI: 10.1016/j.ijbiomac.2019.09.012
2 G. Ozcelikay, M Gamella, G Solís-Fernández, R Barderas, JM Pingarrón, S Campuzano, SA Ozkan. J. Pharm. Biomed. Anal. 2023, 231, 115411. DOI: 10.1016/j.jpba.2023.115411

OP3-2-1 Targeted quantification of odour-active thiols in wine by LC-MS/MS using in situ on-line derivatization

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Volatile thiols are an important family of wine aroma compounds with low odour threshold including 3-mercaptohexan-1-ol (3MH), 3-mercaptohexyl acetate (3MHA), and 4-mercapto-4-methylpentane-2-one (4MMP) amongst others. They significantly contribute to the typical aroma profile of several wine varieties, such as Sauvignon blanc, Petite Arvine or Divona, reminiscent of citrus and tropical fruits. The content and relative distribution of these thiols in wine provide valuable information to winemakers on the effect of different viticultural or oenological practices on the typicity and quality of the wines produced.
HPLC-MS/MS determination of these thiols present at trace levels is usually done after derivatisation to protect these highly reactive compounds and is generally followed by an extraction on SPE or QuEChERS and an additional concentration step to reach the required instrumental sensitivity [1].

The present work was aimed at avoiding sample preparation and to develop a quantification method for the routine analysis of 3MH, 3MHA and 4MMP by allowing the direct injection of wine samples into the HPLC-MS/MS system. In the proposed method, derivatisation of thiols is performed on-line and in situ at the head of the chromatographic column after injection of 100 µl of wine sample and by adding the derivatisation agent 4,4-dithiodipyridine (DTDP) into the aqueous mobile phase A (0.1 mM DTDP in 100 mM ammonium format buffer at pH 4.2). The three thiols are concentrated at the top of the column, allowing to react for 12 minutes to ensure complete derivatisation and are then eluted and separated by a linear gradient of acetonitrile. MS/MS detection was performed in the positive ionisation mode using Multiple Reaction Monitoring of specific mass transitions.

Validation of the method confirmed that the calibration is linear ($R2 > 0.99$) in the quantification range needed for wine samples i.e., between 10–2000 ng/L for 3MH and 1–200 ng/L for 3MHA and 4MMP. Recovery at low and high concentration of analytes was excellent for 3MH and 4MMP (96 – 110%) and acceptable for 3MHA (80 – 95%). Overall, the repeatability values determined in white wines were below 10% RSD.

This method was successfully applied to the analysis of white wines produced from new promising resistant grape varieties developed at Agroscope.

1) Unraveling the Mystery of 3-Sulfanylhexan-1-ol: The Evolution of Methodology for the Analysis of Precursors to 3-Sulfanylhexan-1-ol in Wine; J. R. Muhl, L. I. Pilkington, B. Fedrizzi and R. C. Deed; Foods 2022 Vol. 11 Issue 14 Pages 2050; https://www.mdpi.com/2304-8158/11/14/2050

OP3-2-2 Separation of e-waste metals using green aqueous two-phase systems based on functionalized ionic liquids and deep eutectic solvents

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Electrical and electronic waste is one of the fastest-growing waste categories in the world containing a large amount of technology-critical elements (TCE). Recently, new, more environmentally friendly technologies using less toxic solvents such as ionic liquids (ILs) and deep eutectic solvents (DESs) have been used to separate metals from e-waste. In this work, aqueous biphasic systems (ABS) are used for the extraction and separation of the targeted TCE (Co^{2}+, Ni^{2}+, In^{3}+, La^{3}+, Ce^{3}+, Nd^{3}+, and Dy^{3}+). ABS consists of two immiscible phases that form when certain specific water-soluble compounds are combined in a certain concentration range. ABSs were prepared using either functionalized ionic liquids (ILs) ($[Ch]_3$[DTPA] and ([Ch][Lac]) or deep eutectic solvents (DES) ($(ChCl)_3$:DTPA and Ch: Lac) with polypropylene glycol 400 (PPG400). The extraction points were selected based on the binodal curve of the studied ABSs, and the concentrations of the metals were determined using ICP-OES. The anions of ILs and the acids of DES are responsible for the formation of complexes with metals. The highest partition coefficients (K) were determined for Ce^{3}+ (KCe=2807) and In^{2}+ (KIn=1580) using ABSs based on IL [Ch][Lac] and [Ch]3[DTPA], respectively.). It was also found that the metals Co, Ni and In can be separated from the lanthanides, depending on the anion type of IL. DTPA forms more stable complexes with indium (KIn=1580), cobalt (KCo=793), and nickel (KNi= 1030), and the lactate anion with the lanthanides (KLn=1344, KCe=2807, KDy=1959, KNd=1355). This method can be used as a sample preparation method for the successful separation and quantification of TCE from aqueous samples.

OP3-2-3 Electrospray Ionization Drift Tube Ion Mobility Spectrometer with Ultra-High Resolving Power: Design and Optimization

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Improving the separation performance is one of the main challenges that drift tube ion mobility spectrometry (DTIMS) is facing. Commercial devices typically reach a resolving power of 80 (FWHM). A resolving power higher than 200, which has been classified as ultra-high resolving power, has only been reported in a handful of cases. Electrospray ionization (ESI) allows the direct analysis of liquid samples. With the aim of overcoming this ultra-high performance threshold for an ESI-DTIMS, an improved instrument was carefully designed. The electrodes composing the drift tube were constructed using the new flexible printed circuit board approach, which greatly simplifies the assembly. Based on theoretical background equations, the main causes of peak broadening were

identified, experimentally investigated, and finally minimized. For this purpose, the drift length was increased, and the components were chosen to tolerate higher voltage levels, up to 40 kV. Such voltages were necessary to maintain short drift times, thus limiting ion losses. The supporting frame of the DTIMS electrodes was machined with high precision in order to create a highly homogeneous electric field along the drift path. In addition, the influence of the response time of the transimpedance amplifier processing the ion current was evaluated. Using ESI, a series of four tetraalkyl ammonium ions with different chain lengths was separated with an ultra-high resolving power of up to 228. The ideality of the setup with respect to the theoretical resolving power limit was determined for three voltages and drift lengths. The ideality was found to range from 87 to 97%. It confirmed the excellent electric field homogeneity achieved with the flexible electrodes and validated the optimization steps. The device was tested with two ion injection schemes at a three-grid shutter, namely a traditional Tyndall-Powell voltage pulse and a more recently described so-called tristate pulse. The separation performances were similar in both cases after optimization of the injection parameters. Finally, three benzalkonium chlorides, used as desinfectants, were separated, with resolving powers of over 210.

OP3-3-1 Improvement of fuel-cells based on data from multiple analytical techniques

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In the global effort of curbing carbon dioxide emissions, hydrogen production is one of the promising options for energy storage. Combustion is a low efficiency process for energy conversion, except for heating. Fuel-cells are more promising. One of their critical components in terms of stability and lifetime is the ion-exchange membrane separating anode and cathode. Currently, per-fluorinated materials are preferably used because of their stability against oxidative damage. Such «forever chemicals» pose ecological and toxicological problems. With politics taking this class of chemicals under scrutiny, widespread future application in technology appears uncertain. Therefore, alternative materials on the basis of hydrocarbons are under development.

Current perception blames radicals like HO•, produced in the operating fuel-cell, as dominant initiators for degradation. Hydrocarbons on the whole exhibit unsatisfactory resistance against such oxidants: there is a need for improved membranes and for additives that increase their lifetime. The chemical reactions invoked in the community oftentimes have not been properly characterized or are at odds with published data from other fields. We therefore aimed to get reliable physical chemical data that would allow, at least, a qualitative understanding of the major processes and informed strategies for membrane improvement.

Radiolysis allows for the production of the radicals of interest while also providing mass-balance. Irradiation of model compounds in water with nanosecond pulses of ionizing radiation initiates radical reactions that were followed time-resolved. Supporting data were gathered by flash-photolysis and stopped-flow experiments. Hypotheses on the reaction scheme were first verified on the basis of sample degradation caused by gamma-irradiation, quantified by HPLC. On the basis of these ex-situ investigations, we came up with a chemical mechanistic framework that, in principle, would allow for catalytic repair of oxidative damage.[1,2] One of the potential catalysts is Ce. However, in trials, we ran into discouraging results. The problem could be identified with the help of metals analysis by LA-ICP-MS. These investigations allowed for an adapted strategy which resulted in a proof-of-concept that

fuel cell membranes can be repaired catalytically, in-situ, under the extensive chemical stress of an operating fuel cell.[3]

(1) L. Gubler, T. Nolte, and T. Nauser. Antioxidant Strategies for Hydrocarbon-Based Membranes. ECS Transactions 2018, 86 (13):369-379; DOI: 10.1149/08613.0369ecst
(2) T. de Wild, T. Nemeth, T. M. Nolte, T. J. Schmidt, T. Nauser, and L. Gubler. Possible Repair Mechanism for Hydrocarbon-Based Ionomers Following Damage by Radical Attack. J.Electrochem.Soc. 2021, 168 (5):054514; DOI: 10.1149/1945-7111/abf9be
(3) T. de Wild, T. Nemeth, P. Becker, D. Günther, T. Nauser, T. J. Schmidt, and L. Gubler. Repair of aromatic hydrocarbon-based membranes tested under accelerated fuel cell conditions. Journal of Power Sources 2023, 560:232525; DOI: 10.1016/j.jpowsour.2022.232525

OP3-3-2 Cross-validation of ID ICP/MS, RBS, and MEIS for determination of Absolute Mole Fractions of Elements in Nanometer-Thick Metal Alloy Films

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Rutherford backscattering spectroscopy (RBS) or medium energy ion scattering spectroscopy (MEIS) were used to determine the mole fractions of constituent elements in silver copper (AgCu) and indium gallium zinc oxide (IGZO) alloy films. The results were compared them with those determined by isotope dilution inductively coupled plasma mass spectrometry (ID ICP/MS). Nanometer-thick AgCu alloy films were selected as a model alloy system to demonstrate the equivalence of measurement results from the different analytical methods. The AgCu alloy films with two different thicknesses and five mole fractions were grown on Si (100) wafers by ion beam sputter deposition. The mole fractions of AgCu alloy films with 100 nm thickness measured by RBS and ID ICP/MS showed a great agreement within 0.4 % difference, while those of 10 nm thickness AgCu alloy films measured with MEIS and ID ICP/MS also showed a negligible difference of about 1.0 %.[1] In addition, the same techniques were applied to determine the accurate mole fractions of In, Ga, and Zn in 40 nm-thick IGZO film on glass. The mole fractions of IGZO thin film determined by ID ICP/MS were also consistent with those measured by MEIS within the associated uncertainties. These results indicate that ID ICP/MS, RBS and MEIS can be used as complementary methods to certify the mole fractions for thin alloy film reference materials.

1. T. G. Kim, S. W. Heo, W. J. Min, T.-H. Han, Y.-H. Yim, H. Yu, and K. J. Kim, Metrologia 47 (2021) 253. https://doi.org/10.1088/1681-7575/ac28e2

OP3-3-3 Capabilities of LA-N_2-MICAP-MS for Direct Solid Analysis

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Laser ablation (LA) coupled to an Inductively Coupled Plasma Mass Spectrometer (ICP-MS) has been applied in elemental analysis for more than 30 years in forensic sciences, material sciences or for

geological applications among others.[1] All commercially available ICP-MS instruments are based on argon as plasma gas and helium has been used as carrier gas to transport the laser-generated aerosol into the ICP.[2] Beside a lot of effort to circumvent Ar-based interferences using either high resolution sector field ICP-MS instruments[3] or the use of reaction cell technology,[4] using a different plasma source gas would make some elements better accessible. Furthermore, the use of an alternative plasma gas could also contribute to a more cost-effective ICP-MS operation.[5]

Recently, a novel high-power nitrogen microwave inductively coupled atmospheric-pressure plasma (N_2-MICAP) has been developed and our prototype showed similar performances when compared to a commercially available argon based ICP-MS.[5] Given the increased gas tolerance of the nitrogen plasma compared to an Ar-ICP, different carrier gases for laser-generated aerosols could be used. Our studies focused on the comparison of helium, nitrogen and air as laser aerosol carrier for LA-N_2-MICAP-MS. The achievable results related to ablation rate, sensitivities, limits of detection and quantification capabilities will be shown and discussed.

1. *D.B. Aeschliman, et al., J. Anal. At. Spectrom., 18, (2003), 872. DOI:10.1039/b205683f*
2. *S.M. Eggins, et al. Appl. Surf. Sci., 127-129, (1998), 278. DOI:10.1016/S0169-4332(97)00643-0*
3. *M. Moldovan, et al. Anal. At. Spectrom., 19, (2004), 815. DOI:10.1039/B403128H*
4. *S.D. Tanner, et al., Spectrochim. Acta B, 57, (2002), 1361. DOI:10.1016/S0584-8547(02)00069-1*
5. *M. Schild, et al., Anal. Chem., 90, (2018), 13443. DOI:10.1021/acs.analchem.8b03251*

OP3-4-1 Signal beat on quantification accuracy of spodumene by LA-ICPMS

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Laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS) has become a widely accepted technology for in situ major and trace element analysis particularly in geochemical science. However, the occurrence of aliasing (or spectral skew) due to the inherently pulsed aerosol introduction [1] is a potential obstacle in quantification with sequential ICP-MS (e.g. quadrupole or single collector sector-field MS) combined with LA sampling. This effect is specifically pronounced when low dispersion aerosol transport systems are used, resulting in transient signal oscillations of the ICPMS and biased quantification results. Aerosol dispersion devices downstream the LA cell were frequently installed to reduce the oscillations. But this aerosol smoothing by stretching the individual aerosol pulses may lead to the mixing of ablated material across tens of laser pulses and thus impairing the identification of inclusions or heterogeneous sections in a sample.

Recently, Hattendorf et al. proposed an averaging signal intensity method based on the least common multiple (LCM) duration of mass scan and laser pulse period, which can efficiently eliminate systematic bias in the quantification from aliasing effects in particular for aerosol transport systems with low dispersion [2]. Here, we presented our work on the quantification accuracy of spodumene samples by LA-ICPMS for both synchronized and non-synchronized acquisition with and without aliasing in the transient signals [3]. It was found that synchronized acquisition settings could attenuate the oscillations in the transient signals substantially but showed large variability (up to 18% RSD for Li2O) among individual acquisitions. While non-synchronized acquisition, with and without visible aliasing was producing reproducible results without systematic biases when averaging ion signals from a sufficient number of scans (>100) for quantification. For the latter it was found that non-aliasing methods allowed for a direct identification of heterogeneities or inclusions in the transient signals and these were thus preferred for analysis. Besides, the carrier gas flow rate was also observed to have a

substantial effect on both transient signal structure and LA-ICPMS quantification. This was ascribed to differences in ionization efficiency within the ICP caused by the higher abundance of easily ionized elements in the spodumenes compared to the calibration standard. In conclusion, by selecting appropriate acquisition settings for sequential MS detection and robust operating conditions, the repeatability of the analyses could be improved to values less than 1% for the Li2O mass fractions determined.

1. D. Günther, et al., Fresen. J. Anal. Chem. 368 (2000) 4. Doi: 10.1007/s002160000495.
2. B. Hattendorf, et al., Anal. Bioanal. Chem. 411(2019) 591. Doi: 10.1007/s00216-018-1314-1.
3. X.J. Tan, et al., Chem. Geol. 583(2021) 120463. Doi: 10.1016/j.chemgeo.2021.120463.

OP3-4-2 Single-cell analysis using a downward-pointing vertical ICP-TOFMS

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The inductively coupled plasma time-of-flight mass spectrometry (ICP-TOFMS) is widely used for the analysis of single particles and cells due to its wide linear dynamic range, low limit of detection (LOD), high sensitivity and the simultaneous detection of elements in a single entity. However, one major limitation is to introduce the single cell/particle into the horizontal oriented ICP at a high transport efficiency due to the gravity. A transport efficiency of 100% has already been reported for cells with average sizes of 3-4 µm but not for larger cells yet [1-2].

To overcome this limitation, a downward-pointing vertical ICP-TOFMS has been developed in our group, which allows to achieve 100% transport efficiency regardless of the sample's mass, size and shape due to the gravitational-assisted sample introduction. Recent studies have already shown that microparticles and cells (PBMC, mouse spleen cell and CHO) have been successfully introduced using microdroplet generator systems (MDG) into the ICP [3-4]. Furthermore, the vertical oriented ICP allows to introduce samples at a higher throughput of up to 1000 Hz whereas for the horizontal oriented ICP it is limited to 400 Hz [3].

The downward-pointing vertical ICP-TOFMS has been recently used for the analysis of glass microspheres and different algae cells. Additionally, the coupling to a fluorescence-activated cell sorter (FACS) is still under development.

[1] S. Miyashita, et al., J. Anal. At. Spectrom. 29 (2014) 1598
DOI: 10.1039/c4ja00040d
[2] K. Shigeta, et al., J. Anal. At. Spectrom. 28 (2013) 637
DOI: 10.1039/c3ja30370e
[3] T. Vonderach, et al., Anal. Chem. 93 (2021) 1001
DOI:10.1021/acs.analchem.0c03831
[4] T. Vonderach, et al., J. Anal. At. Spectrom. 36 (2021) 2617
DOI: 10.1039/d1ja00243k

OP3-4-3 Compound Specific Radiocarbon (14C) Dating of Our Colourful Past: from Theory to Practice

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Any observer of an artwork speculates about two details: who is the artist and when was the object created. Answering these queries is not an easy task, however the second question may be addressed by radiocarbon (^{14}C) dating, which affords a time frame of its constitutive materials.

A recurring keyword in describing cultural heritage objects is complexity, as these items are composed of a wide array of materials combined in an infinite number of ways. Many components are organic compounds isolated from natural sources and so are potential chronological markers for the object's life.

The main weakness of the majority of studies that attend to date art objects is the failure to address how the ^{14}C age is estimated on the average ^{14}C content of all carbon bearing materials present. Today, state of the art equipment enables the analysis of samples in the range of tens of micrograms of carbon but it is the measurement of samples as bulk samples that limits the accuracy of the results. Despite recognizing the potential impact of exogenous carbon source on data interpretation, there is no systematic research addressing this question or the development of a separation strategy to purify samples.

The diversity of materials in cultural heritage objects represents one of the biggest challenges for ^{14}C applications, but at the same time this can also represent a chance. The present research focusses on the coloured materials used within an artwork, specifically on natural organic dyes. These compounds are carbon rich and their carbon isotopic ratio represents a snapshot of the atmospheric CO_2 during the years of growth of the organism (plant or insect) from which it was isolated. As such, these compounds represent ideal material for ^{14}C analysis and can serve as proxies for dating the creation of a coloured object, yet until today no such analysis has ever been conducted. The separation of intermingled carbon sources is without question the most difficult problem, yet feasible with the help of compound specific radiocarbon analysis (CSRA). With the range of natural organic dyes and possible matrices being extremely diverse, the proof of principle experiment scope was narrowed to red dyed textiles. Here, we discuss preliminary results and highlight the potential of radiocarbon dating the compounds responsible for the object's colour, which in turn will allow to open new routes to date cultural heritage objects (Hendriks et al. 2022).

L. Hendriks, et al., Helvetica Chimica Acta. 106 (2022)
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OP4-1-1 All Covalently Bound Ion-Selective Membranes for Increased Stability in Potentiometric Sensing

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Solid-contact ion-selective electrodes have gained significant interest over the last decade due to their ease-of-use, miniaturisation possibilities and low maintenance. They can now be routinely found in the bioanalytical field where they are used to measure a wide range of blood electrolytes or in environmental monitoring where they enable the continuous measurement of a large range of relevant ions, such as nitrate, pH or carbonate. Solid contact ion-selective electrodes include an

electron conducting material, such as glassy carbon or gold, covered by a transducing material that is known to improve the stability of the signal and suppress undesired ion transport. The last component is a polyvinyl chloride (PVC)-based plasticised membrane loaded with ion-exchanger and ionophore that enable the selective and sensitive sensing of the target analyte. Unfortunately, this system suffers from leaching of membrane components that over time causes drift and loss of sensitivity [1]. To minimise components leaching, different strategies were envisioned such as enhancement of lipophilicity [2] or covalent binding [3]. Increasing the lipophilicity only slows down the leaching and can also be detrimental in terms of synthetic modification and solubility in organic media. The second approach was based on a plasticiser-free cross-linked poly(decyl methacrylate) matrix that was functional if a single membrane component (either ion-exchanger or ionophore) was covalently attached. Although some studies on the topic exist, reports of attempting the covalent linking of all membrane components [4-5] are scarce.

We present here a new strategy for creating a leak-free ion-selective plasticised membrane, where we decided here to take advantage of "Click" chemistry to safely anchor membrane components. Chlorine groups naturally present on PVC can be easily replaced by azide groups, thus generating an ideal platform to perform a "Click" reaction, also known as azide alkyne cycloaddition. Membrane components can in a second step be modified to include an alkyne group, needed for the final covalent attachment. Taking advantage of the high yield of "Click" reactions, alkyne-modified membrane components can be covalently attached in a quantitative manner by controlling the stoichiometry to prevent any leaching. The new electrodes will be tested using thin-layer membranes [6] to accelerate the leaching process and confirm their improved performances compared to conventional membrane that only rely on lipophilicity.

[1] E. Bakker, P. Bühlmann, E. Pretsch, Chemical Reviews 1997, 97, 3083-3132 10.1021/cr940394a.
[2] B. Néel, M. Ghahraman Asfhar, G. A. Crespo, M. Pawlak, D. Dorokhin, E. Bakker, Electroanalysis 2014, 26, 473-480 10.1002/elan.201300607.
[3] K. R. Choi, X. V. Chen, J. Hu, P. Bühlmann, Analytical Chemistry 2021, 93, 16899-16905 10.1021/acs.analchem.1c03985.
[4] Y. Qin, S. Peper, E. Bakker, Electroanalysis 2002, 14, 1375-1381 10.1002/1521-4109(200211)14:19/20<1375::Aid-elan1375>3.0.Co;2-8.
[5] Y. Qin, E. Bakker, Analytical Chemistry 2003, 75, 6002-6010 10.1021/ac034447c.
[6] T. Forrest, E. Zdrachek, E. Bakker, Electroanalysis 2020, 32, 799-804 10.1002/elan.201900674.

OP4-1-2 SAM/AgCl mixed phase modification of silver surface for functionalization with biomolecules and stabilization of electromotive force

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Quantitative information of nucleic acids such as microRNA and circulating tumour DNA in body fluids is a useful biomarker for the clinical diagnosis of cancer. Liquid biopsies based on such information are now frequently incorporated into clinical trials. There remains room for improvement in the reproducibility and reliability of chip-based assays in clinical settings. Solid-state potentiometric sensors are well-suited for achieving simple, multiplexed, and miniaturized point-of-care testing. To functionalize the surface of the electrode for biosensing, the strong affinity between sulphur and transition metal surfaces is usually used. Organosulfur compounds such as alkanethiols coordinate strongly to gold, silver, copper, and platinum. Biomolecules or probe molecules are then immobilized

using functional groups at the terminal of the self-assembled-monolayer (SAM) for molecular recognition. Transition metals are not usually used as materials for potentiometric measurements. This is because the surfaces of transition metals such as gold, platinum, or silver are polarized and the electromotive force (EMF) at the metal/aqueous solution interface is not electrochemically defined. Drifts in the EMFs of transition metal electrodes are usually observed in potentiometric measurements because of the lack of electrochemical equilibrium.

A solid-state potentiometric biosensor based on self-assembled monolayer (SAM) and silver chloride (AgCl) mixed phase modification of a silver surface has been proposed1. Stabilization of the electromotive force and functionalization with biomolecules on the sensing surface were simultaneously achieved using silver chloride chemically deposited with 1,3-diaminopropanetetraacetic acid ferric ammonium salt monohydrate and a SAM with oligonucleotide probes, respectively. The formation of silver chloride and adsorption of alkanethiol on the silver surface were confirmed with X-ray photoelectron spectroscopy. For the hybridization assay, a mixed SAM of oligonucleotide- and sulfobetaine (SB)-terminated alkanethiols was formed together with silver chloride on the surface of the sputtered Ag thin film. The resulting modified surface reduced the nonspecific binding of interfering biomolecules and achieved a high signal to noise ratio. The lower detection limit of the target microRNA 146 was 0.1 pM, at which concentration the signal to noise ratio was 9.1. The achieved detection limit was approximately two order of magnitude higher than that of the SAM/Au momdified electrode2, because the proposed sensor with the organic and inorganic mixed structure could be operated with a stable electromotive force (EMF). The proposed biosensor could be useful as a disposable single-use sensor in medical fields such as liquid biopsies.

1. Miyuki Tabata, Chiho Kataoka-Hamai, Kozue Nogami, Daiju Tsuya, Tatsuro Goda, Akira Matsumoto, Yuji Miyahara "Organic and inorganic mixed phase modification of silver surface for functionalization with biomolecules and stabilization of electromotive force", RSC Advances, 2021, 11, 24958–24967. doi.org/10.1039/D1RA03449A
2. Tatsuro Goda, Kozue Masuno, Junko Nishida, Nobuyoshi Kosaka, Takahiro Ochiya, Akira Matsumoto, Yuji Miyahara, "A Label-free Electrical Detection of Exosomal microRNAs using Microelectrode Array", Chem. Commun., 2012, 48(98), 11942-11944. doi.org/10.1039/C2CC36111F

OP4-1-3 Determination of benzoate in cranberry and lingonberry using a solid-contact ion-selective electrode

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Benzoic acid is widely used as a preservative in processed foods and benzoic acid is also a naturally occurring compound in berries such as cranberry and lingonberry [1].

In the present work, a solid-contact benzoate-selective electrode was developed based on an acyclic derivative of 1,3-bis(carbazolyl)urea [2] as ionophore in a solvent polymeric membrane of the following composition (wt-%): 1 % ionophore, 0.3 % TDMACl, 65.6 % o-NPOE, and 33.1 % PVC. A control membrane without ionophore was used for comparison (0.3 % TDMACl, 66.7 % o-NPOE, 33.0 % PVC). Electrosynthesized poly(3,4-ethylenedioxythiophene) (PEDOT) was used as solid contact for the benzoate-selective electrodes (benzoate-ISEs) and the control electrodes (control-ISEs). Six benzoate-ISEs and six control-ISEs were prepared and studied in parallel. The ISEs were characterized by electrochemical impedance spectroscopy and potentiometry. The benzoate-ISE showed a near-Nernstian slope (-56.7 ± 0.7 mV/dec) to benzoate in a background solution of 0.01 M phosphate buffer.

On the contrary, the slope for the control-ISE was sub-Nernstian (-49.1 ± 0.8 mV/dec) under the same experimental conditions.

The benzoate content of cranberries and lingonberries were determined potentiometrically with the ISEs using the standard addition method. The results obtained with the benzoate-ISEs and control-ISEs were compared to results obtained with ion chromatography (IC).

The benzoate concentration in cranberry (0.23 ± 0.04 g/kg) and lingonberry (1.08 ± 0.20 g/kg) obtained with the benzoate-ISEs were in good agreement with the concentrations obtained by IC, i.e. cranberry (0.17 ± 0.03 g/kg) and lingonberry (0.88 ± 0.07 g/kg). However, the results obtained with the control-ISE were significantly higher, i.e. cranberry (0.92 ± 0.07 g/kg) and lingonberry (1.50 ± 0.13 g/kg), indicating contribution from interfering ions on the ionophore-free control-ISE. These results show that the 1,3-bis(carbazolyl)urea-based ionophore provides sufficient selectivity for the determination of benzoate in cranberry and lingonberry.

1. S. Viljakainen, et al., Acta Agric. Scand., Sect. B, Soil and Plant Sci. 52 (2002) 101. DOI:10.1080/090647102321089846
2. A. Rüütel, et al., Beilstein J. Org. Chem. 16 (2020) 1901. DOI:10.3762/bjoc.16.157

OP4-1-4 Sensing of cancer related-cell membrane proteins using ion-sensitive field-effect transistors for liquid biopsy

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Liquid biopsy has got attention by clinicians to diagnose or control disease through minimally-invasive body fluid testing. There are several biomarkers in liquid biopsy for cancer detection such as circulating small nucleic acids, circulating tumor cells (CTCs) and extra cellular vesicles (EVs). In particular, detection of CTCs in the blood of patients provides important information based on the analysis of proteins and sugar chains expressed on the membrane and the nucleic acids contained therein. Detection of CTCs remains technically challenging, because CTCs are rare compared with the background of millions of blood cells.

For detection of CTCs, we propose a new concept that is a cell membrane protein detection with ion-sensitive field-effect transistors (ISFETs) combined with chemical signal amplification based on enzyme reactions[1, 2]. Breast cancer cell lines were used as models of CTCs, and human epidermal growth factor receptors (HER2 and EGFR) expressed in their membranes were detected using the proposed cell-based FETs. After capturing breast cancer cells onto the ISFETs, they were exposed to enzyme-conjugated antibodies targeting HER2 or EGFR. Addition of the substrate for the enzyme caused an immediate pH change, and that was monitored by the ISFET. This pH shift depends on the expression level of HER2 or EGFR. The concept for detecting HER2 via an enzyme reaction using the cell-based FET was successfully demonstrated using the BT474 breast cancer cell line. After proof-of-concept, EGFR expression level was compared among four types of breast cancer cell lines. The order of the expression levels of EGFR among the cell lines, determined with the cell-based FETs, was consistent with the results of fluorescence detection determined by fluorescence-activated cell sorting (FACS).

In that no laser excitation or detector is required, the cell-based FETs are advantageous for miniaturization and in massive parallel analyses of target molecules expressed on the membranes of not only CTCs but also EVs. In order to realize more quantitative measurement, we are currently developing a digital sensing device. It could be a useful tool for detection of epithelial-mesenchymal transition (EMT). Their small size and cost effectiveness for cancer testing could enable their realization in a future liquid biopsy.

[1] Miyuki Tabata, Chattarika Khamhanglit, Sayo Kotaki, Yuji Miyahara, "Detection of cell membrane proteins using ion sensitive field effect transistors combined with chemical signal amplification", Chemical Communications, 2022, 58, 7368–7371
DIO: 10.1039/D2CC02159E
[2] Miyuki Tabata, Xinyue Liu, Chattarika Khamhanglit, Sayo Kotaki, Yuji Miyahara, "Detection of epidermal growth factor receptor expression in breast cancer cell lines using an ion-sensitive field-effect transistor in combination with enzymatic chemical signal amplification", Journal of the American Chemical Society, 2022, 144, 36, 16545–16552
DIO: 10.1021/jacs.2c06122

OP4-2-1 A rapid strip test for molecular identification of the European sardine, Sardina pilchardus, Walbaum, 1792 (Osteicthyes)

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Fish adulteration has been observed worldwide and is a global growing concern. Adulteration denotes the substitution of one fish with another fish of lower price and nutritional quality. Sardines are among the most vulnerable fish to adulteration, as they are widely consumed in many countries. Therefore, methods of fish adulteration detection are in great demand. Most methods entail DNA-based identification of fish species in fresh, processed, and canned products. We have developed a simple and rapid strip test for the detection of fish adulteration and particularly the adulteration of Sardina pilchardus from Sardinella aurita, the most common species used for substitution. The method involves (i) DNA isolation from fresh tissue and processed samples, (ii) amplification by PCR using a common pair of primers, and (iii) detection of amplified sequences with the strip test via hybridization with species-specific DNA probes. Gold nanoparticles were used as reporters for visual detection by the naked eye. The detection is completed within 10 min and the results are displayed in the form of a red zone. We were able to detect as low as 3 fmol of pure Sardinella aurita, 1% of adulteration in mixtures of PCR products, and 5% of adulteration in adulterated mixtures of processed fish samples. The proposed strip test is simple, rapid, and has an excellent analytical performance. Finally, this test can be applied to detect other fish species.

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OP4-2-2 Development of a new method for determination of total antioxidant capacity of the macroalgae using fiber optic reflectance spectrophotometer

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Increasing demands for additive-free natural products and innovative production techniques resulted an increase in the interest in natural additives. One of the most important groups, antioxidants, can prevent or delay oxidation even with trace amounts. Natural antioxidants such as tocopherols, ascorbic acid, carotenoids, flavonoids, amino acids, phospholipids, and sterols are used, especially in foods and cosmetic products. Antioxidants inhibit free radicals and convert them into non-toxic products. For this reason, there are many methods in the literature to determine the antioxidant capacity of foods, plants and similar products (Apak et al., 2007; Ozyurt et al., 2012). Macroalgae, which play an important role in the food chain, are used in various fields such as cosmetics, food, agriculture, medicine (Thiyagarasaiyar et al., 2020). They have seconder metabolites which are antioxidants, phenolics, flavonoids and fatty acids. In most of the products, antioxidant activity is trying to be enhanced by synergistic effects of multi-substances.

In this study, a simple, fast, inexpensive, a new method was developed using the fiber optic reflectance spectrophotometer (FORS) for determining the total antioxidant capacity of antioxidant-poor samples such as marine macroalgae extracts. The developed optical sensor enables to determine the total antioxidant capacity of macroalgae without any pre-concentration process. The chemical optical sensor was developed using fiber optic reflection spectroscopy due to the quantitative absorption of the magenta-colored anionic Fe(II)-FZ complex on Sephadex QAE A-25 resin. The increase in sensitivity using resin allowed the measurement of the total antioxidant capacity (TAC) of antioxidant-poor samples and complex systems. The reflectance change associated with the formation of the highly colored Fe(II)−FZ chelate on the Sephadex QAE A-25 resin as a result of reaction with antioxidants was measured at 562 nm by using a reflectance spectrometer. The molar absorptivity, linear concentration range and Trolox equivalent antioxidant capacity (TEAC) of some antioxidants were found. The trolox equivalent antioxidant capacities (TEAC) of various antioxidant compounds using the proposed method were comparable to those of the spectrophotometric Ferrozine assay (Berker et al., 2017). Calibration curves (lines) of Trolox, quercetin, and vitamin e individually and in macroalgae infusions were paralleled using the standard addition method, confirming that the added antioxidants did not interact with macroalgae components to cause chemical deviations from Beer's law.

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R. Apak, et al., Molecules, 12 (2007), 1496.
DOI: 10.3390/1207149
K.I. Berker, et al., Anal Sci. 33 (2017), 683.
DOI: 10.2116/analsci.33.683
D. Ozyurt, et al., J. Fluoresc., 21 (2012), 2069.
DOI: 10.1007/s10895-011-0905-4
K. Thiyagarasaiyar, et al., Mar. Drugs, 18 (2020),323.
DOI: 10.3390/md18060323

OP4-2-3 Tracking transformations of dietary metabolites through gut microbial metabolism

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Transformation of food chemicals by the gut microbiota profoundly impact human health. We present here an approach to measure the rate and fate of dietary metabolites through gut microbial metabolism for application to physiological based kinetic models, and to identify yet unknown modifications performed by gut microbes. Using in vitro fermentations of human and rat gut microbial communities coupled with merged targeted/untargeted LC-MS/MS metabolomics analyses, supported by in silico tools, we calculated rates of chemical reactions and uncovered predicted reaction products. We will present metabolic alterations resulting from consuming food-relevant compounds such as natural phenolics, nanoparticles, sweeteners, or administration of antibiotic drugs. Further potential applications of this platform include addressing how complex food matrices are transformed in the gut and predicting diet/microbiota relationships that impact human health.

OP4-2-4 RECOVERY OF PHENOLIC COMPOUNDS FROM OLIVE TREE LEAVES: CHARACTERIZATION OF DEEP EUTECTIC SOLVENT EXTRACTS

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Agri-food industries generate a large amount of waste that offers great revalorization opportunities within the circular economy framework. In this line, waste from the olive industry is a noticeable source of bioactive compounds, mostly phenolic compounds, that can be extracted and reused for the production of raw materials related to the chemical, cosmetic, and pharmaceutical sectors. Traditionally, solvents for the extraction of bioactive compounds are based on hydroalcoholic mixtures but, due to toxicological and environmental reasons, they are mainly restricted to ethanolic solutions. Hence, these conventional solvents are being replaced with greener alternatives, such as natural deep eutectic solvents (NADESs). NADESs are biodegradable, non-toxic, easy to prepare, and relatively inexpensive solvents and have been demonstrated to have similar, or even higher, extraction efficiencies than conventional solvents. In addition, they can be combined with more advanced extraction technologies, such as microwave-assisted extraction (MAE), to reduce solvent waste, extraction time, and energy requirements while increasing extraction efficiency.

In this study, a methodology for extracting phenolic compounds from olive tree leaves was optimized based on NADES. In addition, the effect of MAE was also evaluated. The resulting extracts were characterized in terms of total polyphenolic content, determined by high-performance liquid chromatography with ultraviolet detection (HPLC-UV), and the antioxidant capacity based on the ferric-reducing antioxidant power (FRAP) and the 2,2-diphenyl-1-picrylhydrazylhydrate free radical (DPPH) assays. In general, 2- to 10-fold higher recovery yields were obtained with NADES compared to conventional hydro-organic solvents. Additionally, HPLC coupled to high-resolution mass spectrometry

(HRMS) using data-dependent acquisition mode with an Orbitrap HRMS instrument was used for identification purposes. As a result, more than 50 different phenolic compounds were annotated tentatively, of which about 20 were confirmed from the corresponding standards. Because of their remarkable content in phenolic compounds and high antioxidant capacity, olive tree leaves and other olive waste have been identified as an especially suited source for polyphenol recovery.

OP4-3-1 Capillary electrophoresis coupled to ICP-MS: a new promising analytical tool for separation and detection of nanoplastic particles

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Microplastics and nanoplastics are released throughout the whole life cycle of plastic consumer products. As part of the EU's Plastics Strategy and Circular Economy Action Plan, there is a strong push to ban single-use plastics and develop plastic-free and recyclable materials. Nonetheless, plastic disposal and its fragmentation to smaller particles remains a threat to the natural environment and organisms.

In the framework of the EU-funded CE4Plastics project, state-of-the-art analytical methods are explored to identify and quantify nanoplastics released into drinking water from single-use and reusable plastic bottles. Capillary electrophoresis is the main pillar for the nanometrological approach herein described, in which separation of differently sized nanoplastic particles is envisaged. Amongst different electrolyte components (tris(hydroxymethyl)aminomethane, borate, ammonium acetate), sodium phosphate in conjunction with sodium dodecyl sulphate in alkaline medium (pH=8.9) has been selected as the optimum buffer for an effective separation method of polystyrene particles, size ranging from 30 to 300 nm, through a bare fused silica capillary and ultraviolet detection at 220 nm. Detection limit was found to be at the mg/L level, whilst repeatability obtained for migration time and peak area was approximately 4 and 8%, respectively. The behaviour of these differently sized particles has been investigated in terms of electrophoretic mobility, figures for this parameter varying from -3.53 to -5.76 µm×cm/V×s amongst the smaller and larger particles, respectively. Surface-charge ratio also exhibited the same trend, as it is proportional to electrophoretic mobility. This phenomenon is in accordance with lower migration times revealed by the smaller particles, as their peaks are closer to the electroosmotic flow, whilst the larger particles show a more negative surface-charge ratio and electrophoretic mobility, and consequently a longer migration time.

To further extend and improve the method performance, CE was hyphenated to inductively-coupled plasma-mass spectrometry. Europium-doped polystyrene particles (100-300 nm) were used as a tracer to optimise the analytical workflow, including a particle preconcentration strategy, and were detected using an innovative ICP-MS calibration method. The particle size resolution power of CE, together with the high sensitivity of ICP-MS, can be further assessed as an analytical tool for separation and detection of nanoplastics in drinking water released from plastic bottles, thus providing a response to society awareness on this concern.

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OP4-3-2 Speciation of Nanoparticles by Imprinting

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Nanotoxicity deals with the adverse effects of nanomaterials on human health and the environment. It is evident that the nanotoxicity of nanoparticles (NPs) depends on different parameters such as the material they are made of as well as their size, shape, and surface chemistry. This means that we have to treat the toxicity of NPs similarly to that of heavy metals, namely, by speciation. In other words, it is insufficient to determine the total concentration of NPs in either the liquid or the gas phase but it is essential to develop tools that will be able to determine selectively the concentration of NPs as a function of their above-mentioned characteristics.

We have developed a new concept for the selective recognition and detection of NPs termed nanoparticle imprinted matrices (NAIM).[1-5] It is analogous to the well-known concept of molecularly imprinted polymers (MIP) in which the molecular analyte is imprinted in a polymer by polymerization of proper monomers with which it chemically associates. The removal of the template forms complementary cavities capable of selective recognition of the analyte. Instead of molecular species, we imprint NPs in various matrices. The NPs are then removed to form nanometric voids that can selectively recognize the originally imprinted NPs. The NAIM approach works so well that we can detect NPs that are stabilized by different carboxylic acid short molecules.

We will present a few new systems by which we show how NPs can be imprinted inside a matrix. Approaches for studying the NP-matrix interactions, the imprinting of non-spherical NPs, and the detection of NPs from the gaseous phase, will be discussed.

[1] L. Dery, et al. ACS Sensors 7(1) (2022) 296-303.
[2] D. Zelikovich et al., ACS Appl. Nano Mater. 4(10) (2021) 10819-10827.
[3] N. Bruchiel-Spanier et al., Nano Res. 12(2) (2019) 265-271.
[4] M. Hitrik et al. Nanoscale 8(29) (2016) 13934-13943.
[5] S. Kraus-Ophir et al., Angew. Chem.-Int. Edit. 53(1) (2014) 294-298.

OP4-3-3 Nanoscale Investigation of Heterogenous Catalytic Processes using Tip-Enhanced Raman Spectroscopy

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During the last two decades, Tip-Enhanced Optical Spectroscopy (TEOS) has emerged as a powerful analytical tool for studying surface chemistry with nanoscale spatial resolution [1-3]. In this talk, I will cover several key aspects of the application of TEOS in studying heterogenous catalytic reactions in two parts. In the first part, I will first discuss the application of Tip-enhanced Raman Spectroscopy (TERS) to study plasmon-driven photocatalytic reactions. I will highlight the ability of TERS to map catalytic activity at the nanoscale, providing insights into the spatial distribution photocatalytic reaction hotspots on a nanostructured Ag surface [4]. Then, I will discuss the exploration of photocatalytic processes in liquid phase using TERS, showcasing the capability of TERS to observe

dynamic changes during heterogenous catalytic reactions [5]. Finally, I will delve into the investigation of reactive arrangement in on-surface photocatalytic coupling reactions using TERS [6]. By combining TERS with molecular-level insights, we can gain a deeper understanding of the role of reactive arrangement in the efficiency of these reactions.

In the second part of the talk, I will discuss the use of a different but related technique called Tip-Enhanced Fluorescence (TEFL) imaging for nanoscale chemical imaging of zeolite acidity in fluid cracking catalyst particles [7] and the characterization of coke formation on ZSM-5 zeolite catalysts during methanol-to-hydrocarbon reaction [8]. These studies demonstrate the unique capabilities of hyperspectral TEFL imaging in providing spatial and chemical information at the nanoscale.

Overall, this talk will highlight the significant contributions and potential of Tip-enhanced Optical Spectroscopy (TEOS) in the nanoscale investigation of surface catalytic processes. By elucidating the spatial distribution, dynamics, and reactive arrangement of catalytic reactions, TEOS offers valuable insights for advancing our understanding of surface chemistry and guiding the development of efficient catalysts.

[1] N. Kumar, S. Mignuzzi, W. Su, D. Roy, European Physical Journal Techniques and Instrumentation, 2015, 2(1), 9. https://doi.org/10.1140/epjti/s40485-015-0019-5
[2] D. Mrđenović, Z.-F. Cai, Y. Pandey, G. L. Bartolomeo, R. Zenobi, N. Kumar*, Nanoscale, 2023, 15, 963-974. https://doi.org/10.1140/epjti/s40485-015-0019-5*
[3] Z.-F. Cai, N. Kumar, R. Zenobi, CCS Chemistry, 2023, 5, 55-71. https://doi.org/10.31635/ccschem.022.202202287
[4] N. Kumar, B. Stephanidis, R. Zenobi, A. J. Wain, D. Roy, Nanoscale 2015, 7, 7133–7137. DOI https://doi.org/10.1039/C4NR07441F
[5] N. Kumar, C. S. Wondergem, A. J. Wain, B. M. Weckhuysen, The Journal of Physical Chemistry Letters 2019,10, 1669-1675. https://doi.org/10.1021/acs.jpclett.8b02496
[6] Z.-F. Cai, J. P. Merino, W. Fang, N. Kumar, J. O. Richardson, S. D. Feyter, R. Zenobi, Journal of the American Chemical Society, 2022, 144, 538–546. https://doi.org/10.1021/jacs.1c11263
[7] N. Kumar, S. Kalirai, A. J. Wain and B. M. Weckhuysen, ChemCatChem 2019, 11, 417-423. https://doi.org/10.1002/cctc.201801023
[8] S. Bienz, S. H. van Vreeswijk, Y. Pandey, G. Bartolomeo, B. M. Weckhuysen, R. Zenobi, N. Kumar, Catalysis Science & Technology 2022, 12, 5795-5801. https://doi.org/10.1039/D2CY01348G

OP4-4-1 Development of a novel dynamic headspace Vacuum In-Tube Extraction (VITEX) method for volatile compounds

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Headspace in-tube extraction (HS-ITEX) and solid phase microextraction (HS-SPME) sampling, followed by gas chromatography-mass spectrometry (GC-MS), are widely used to analyze volatile compounds in various food matrices. The extraction efficiency of volatile compounds from foodstuffs is crucial for obtaining relevant results. However, the efficiency of extraction methods is often limited by long extraction times and requirements for large sample quantity. This study reports on the development and application of a new extraction technique based on the ITEX-DHS hardware (In-Tube Extraction - Dynamic Headspace). In this study we show significant enhancement in extraction rate and capacity by operating ITEX-DHS under reduced pressure, called Vacuum-ITEX-DHS. The results of the study indicate that VITEX-DHS improves the extraction of the target compounds. The area of the mass spectrometer signal for each compound can be up to 450 times more intense than the HS-SPME and

HS-ITEX techniques performed in similar conditions of extraction temperature and time. In addition, the lifetime of an ITEX trap is up to 100 times longer than an SPME fibre.

VITEX-DHS runs in automated mode, making it possible to work with smaller sample quantity and at the same time to favor the HS extraction of all volatile compounds. The VITEX development is supported by CTC Analytics in Zwingen and is compatible with the PAL-RSI and PAL RTC autosamplers.

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OP4-4-2 LC-MS characterization and stability assessment elucidates role of charge variants in the degradation of monoclonal antibody therapeutics

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Monoclonal antibodies therapeutics encounter many stresses during their entire production lifetime. Many critical quality attributes (CQA) such as charge and hydrophobic variants are believed to impact degradation propensity in addition to environmental factors. However, there is a lacuna in our understanding on the direct impact of charge on mAb aggregation. This study therefore aims to elucidate any potential underlying correlation between charge heterogeneity and stability of mAbs. In this regard, three acidic and one basic variant of trastuzumab were isolated using semi-preparative cation exchange chromatography. Separated variants were subjected to a battery of stresses commonly encountered during downstream purification of mAbs, such as thermal, mechanical, high salt and low pH stress. Size-based and charge-based heterogeneities were monitored using analytical size exclusion chromatography and cation exchange chromatography, respectively. Dynamic light scattering was used to assess changes in hydrodynamic size upon stress application. LC-MS analysis and peptide mapping identified these variants as deamidation of light chain Asn30 residue (acidic 1), 2x deamidation at light chain Asn30 and heavy chain Asn55 (acidic 2), isomerization variant (acidic 3), and oxidation of Met83 (basic variant), respectively. Stability assessment indicates that charge variants behave differently vis-à-vis the control mAb samples. We observe that thermal and low pH stress impact acidic 2 more than others causing maximum aggregation. Whereas mechanical and salt stress caused highest insoluble degradation in acidic 3 variant vis-à-vis main specie. Thermal and mechanical stress caused higher visible degradation as observed by DLS. Thermal stress causes maximum soluble and insoluble aggregation due to increased deamidation propensity over and above the existing modification in any charge variant. This increases accumulation of acidic variants in the sample significantly in all cases. In summary, altered charged profile of the variants increases their instability and overall degradation propensity of the final product cocktail. This knowledge is essential to carry out pooling decisions and warrants a case-by-case assessment of variants to ensure complete knowledge of the product in the Quality by Design paradigm.

OP4-4-3 Fast screening of biological fluids for VSIG1 – a diagnostic tool for gastric cancer

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Gastric cancer is a commonly occurring silent type of cancer. The detection of incipient gastric carcinoma continues to pose a significant challenge in the field of medicine. The current research explores the potential utility of V-set and immunoglobulin domain containing 1 (VSIG1), a relatively new biomarker, member of the JAM family, as diagnostic tool for a specific type of cancer. The expression of the VSIG1 gene has been noted to be prevalent in the epithelial cells situated in the gastric region [1]. Therefore, a fast screening test of biological fluids such as whole blood, saliva, urine, and tumoral tissue is needed for its reliable detection and quantification. A 3D sensor (NS-co-doped graphene modified paste) and a 2D sensor (SPE based on gold) were designed and characterized for the assay of VSIG1. To obtain the stochastic signal, calix[4]arene-25,26,27,28-tetrol was used as modifier for the paste and for the surface of the 2D sensor. Stochastic sensors and the stochastic mode were selected for the screening of the biological samples due to the complexity of the matrix, and taking into account that the stochastic sensors can reliably perform both qualitative and quantitative analysis. The signature of VSIG1 was determined, and used for the identification of its signal in the diagrams obtained after the fast screening of biological samples, followed by its quantitative determination. High sensitivities and low determination limits were obtained using both sensors. The validation was done using the real biological samples. The proposed sensors are cost-effective; they can be used for the fast screening tests (3 minutes) of biological samples for more than 100 measurements, and as long as one month.

[1] Y. Chen et al., J Surg. Oncol. 106 (2011) 286. DOI: 10.1002/jso.22150

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With more than 1.9 million new diagnoses per year [1], colorectal cancer is among the leading global causes of death in cancer patients. While the definitive diagnosis usually involves biopsy, in vivo Raman spectroscopy, a less invasive examination method, has shown great potential to discriminate between normal and cancerous tissue [2] . However, the absence of a suitable classifier as well as the time consuming and expertise demanding pre-processing of such in vivo Raman spectra are the main obstacles to the adoption of this minimally invasive technique in clinical practice. By developing a real-time classification pipeline coupled with a user-friendly utility for non-spectroscopists, we look to remedy these obstacles. In addition to routine colonoscopy, in vivo Raman spectra of healthy and cancerous colorectal tissue were acquired using a custom-made microprobe. The spectra were then loaded into the pipeline and pre-processed in several steps, including normalisation and finite impulse response filtration. The quality of the pre-processed spectra was checked using signal-to-noise ratio before the suitable spectra were decomposed and classified using principal component analysis and random forest, respectively. Additionally, a utility with a graphical user interface was developed to facilitate the use of our data pipeline by non-spectroscopist in a clinical environment. Overall, the combination of algorithmic preprocessing of in vivo measured Raman spectra with supervised and unsupervised machine learning appears to be a viable way of reducing the relatively large number of biopsies currently needed to definitively diagnose colorectal cancer.

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[1] H. Sung, et al., CA: Cancer J. Clin. 71 (2021) 209-249.

DOI: 10.3322/caac.21660

[2] W. Wang, et al., J. Biophotonics 8 (2015) 527.
DOI: 10.1002/jbio.201400026

OP5-1-1 Inkjet Printing in the Development of Solid-State Potentiometric Sensors

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Inkjet printing of miniaturized planar electrodes presents a greener, scalable, and cost-effective way of fabricating solid-state potentiometric sensors. Such devices are needed to enable emerging applications of (bio)chemical sensing in aqueous solutions. A key challenge in the process of inkjet printing electrodes is the formulation of conductive inks with suitable fluid dynamic and surface properties, which are required for adequate jetting, wetting and adhesion of the ink to the substrate. While chemical modification of the substrate is a viable option to promote adhesion, such printed electrodes may not be compatible with measurements in solution. We have thus developed conductive nanoparticle-based inks based on modification of the nanoparticle itself. In the first case, an amphiphilic silver nanoparticle ink was obtained by modifying poly(acrylic acid) capped nanoparticles with 3-morpholynopropylamine (MPA) [1]. The nanoparticles were inkjet printed on different substrates, including glass, PET and polyimide and they exhibited sheet resistances below 1 Ω/sq. A second conductive ink was formulated based on mechanically exfoliated graphene nanosheets stabilized by melamine [2]. The two inks were used in the development of a solid contact ion selective electrode (SC-ISE). Due to its high conductivity, the silver ink was used for printing the electrical contact, while the graphene ink was printed over to provide an inert surface of the working electrode and a high capacitance solid contact. Both thermal processing and intense pulsed light were evaluated as a means of increasing electrode conductivity. Lastly, a plasticized PVC membrane, containing nonactin as ionophore, was deposited on the solid contact and an ammonium-selective electrode was thus obtained. The fabricated SC-ISE demonstrated good sensitivity, reproducibility, linearity (R^2 = 0.9980) and a near-Nernstian response of 64.3 mV/dec in the linear range (4 - 1 pNH_4), along with no observable water layer formation.

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[1] I. Ivanišević et al., Nanomaterials 12 (2022) 4252. DOI: 10.3390/nano12234252
[2] M. Kralj et al. Nanomaterials 12 (2022) 2936. DOI: 10.3390/nano12172936

OP5-1-2 Peculiarities of the potentiometric response of ion-selective membranes containing two neutral ionophores

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Ion-selective electrodes (ISEs) with membranes containing neutral ionophores comprise a well-established analytical tool widely used under zero-current (potentiometric) mode. It was also shown that linear sweep or cyclic voltammetry measurements can be performed with ISEs equipped with

membranes containing several neutral ionophores [1-5]. In this way it is possible to detect several ions with the same sensor. The use of membranes with several ionophores, per se, is not new [6]. However, the composition of the membranes for multianalyte detection may differ significantly from those traditionally used in potentiometry: the ion-exchanger is in excess over the ionophores [3-5]. This may result in an unusual potentiometric response.

We have studied the potentiometric response of membranes containing neutral ionophores valinomycin and Li-ionophore VIII, and potassium tetrakis(p-chlorophenyl)borate as ion-exchanger. We have found that the potentiometric response of membranes combining two ionophores and ion-exchanger in excess over the ionophores is very different from those of membranes with only one ionophore, and of membranes with excess of ionophores over ion-exchanger. The excess of ion-exchanger results in a sub-Nernstian response to the respective cation in pure solutions (KCl or LiCl) whereas in mixed solutions with equal concentrations of the two electrolytes the response is close to Nernstian.

The origin of the aforementioned peculiarities will be explained by means of computer simulations.

1. G. Crespo, et al., Anal. Chem. 87 (2015) 7729. DOI: 10.1021/acs.analchem.5b01459
2. P. Greenawalt, et al., Anal. Chem. 88 (2016) 5827. DOI:10.1021/acs.analchem.6b00397
3. M. Cuartero, et al., Anal. Chem. 88 (2016) 1654. DOI: 10.1021/acs.analchem.5b03611
4. M. Cuartero, et al., Anal. Chem. 88 (2016) 5649. DOI: 10.1021/acs.analchem.6b01085
5. C. Mao, et al., J. Electroanal. Chem. 880 (2021) 114800 DOI: 10.1016/j.jelechem.2020.114800
6. M. Erdosy, et al., Electroanalysis 14 (2002) 1433.
DOI: 10.1002/1521-4109(200211)14:19/20<1433::AID-ELAN1433>3.0.CO;2-M

OP5-1-3 Long-term continuous monitoring of biomarkers with single-molecule resolution: which molecular mechanisms are limiting?

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Biosensing by Particle Motion (BPM) is a sensing method with single-molecule resolution that has been specifically designed to enable the continuous monitoring of biomolecules at low concentrations, such as nucleic acids, metabolites, proteins, and hormones1,2,3,4.The method relies on optically tracking of the motion of individual biofunctionalized particles (1 µm in diameter) that interact with a biofunctionalized sensor surface. The particles switch between bound and unbound states due to reversible single-molecule interactions caused by analyte molecules and affinity molecules on particles and sensing surface. To enable long-term continuous sensor operation over days and weeks, detailed understandings of the functionality and stability of the molecules and their molecular mechanisms are required. We performed long-term experiments with tethered and with free particles in order to quantify changes in specific as well as non-specific interactions. We observe that particles can be categorized in different classes with distinct time-dependent behaviors. The experimental results are supported by Monte-Carlo modelling with loss rates of molecular components. We will present the research methodology, show experimental results and simulations, and discuss strategies that enable long-term use of continuous biosensors with single-molecule resolution.

1. Visser, E. W. A.; Yan, J.; van Ijzendoorn, L. J.; Prins, M. W. J.; Continuous biomarker monitoring by particle mobility sensing with single molecule resolution. Nat. Commun. 2018, 9, 2541. DOI: https://doi.org/10.1038/s41467-018-04802-8

2. Yan, J.; van Smeden, L.; Merkx, M.; Zijlstra, P.; Prins, M. W. J. Continuous Small-Molecule Monitoring with a Digital Single-Particle Switch. ACS Sens. 2020, 5 (4), 1168–1176. DOI: https://doi.org/10.1021/acssensors.0c00220
3. Van Smeden, L; Saris, A.; Sergelen, K.; de Jong, A. M.; Yan, J.; Prins, M. W. J. Reversible Immunosensor for the Continuous Monitoring of Cortisol in Blood Plasma Sampled with Microdialysis. ACS Sens. 2022, 7 (10), 3041–3048. DOI: https://doi.org/10.1021/acssensors.2c01358
4. Buskermolen, A. D.; Lin, Y. T.; van Smeden, L; van Haaften, R. B.; Yan, J.; Sergelen, K.; de Jong, A. M.; Prins, M. W. J. Continuous biomarker monitoring with single molecule resolution by measuring free particle motion. Nat. Commun. 2022, 13, 6052, 1–12. DOI: https://doi.org/10.1038/s41467-022-33487-3

OP5-2-1 3D-Printed microreactor for "in-situ" detection of ammonia in natural water

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Chemicals monitoring in natural water environment has become an essential observation due to human activity in the last decade (1). Indeed, pollution from chemicals can significantly affect aquatic conditions and thus the flora and fauna (i.e. Eutrophication) (2). It's also known that sampling and lab analysis is time consuming and costly over a prolonged period (maintenance, Quality Control & Quality Assurance…). Therefore, an inexpensive and automated "In-Situ" instrument will be an ideal solution to monitoring analytes in real time and to reduce cost. Here, we present a 3D-printed microreactor chip using a high resolution polyjet 3D-printer. With the help of a small heating device coupled to this microfluidic chip, we are now able to realize a 2 min colorimetric reaction for the detection of ammonia in a continuous flow reactor. With a printed channel reaction length of 1.5m on a total area of 4 square centimeters this chip provides an ideal portable heated reaction device for ammonia detection "in-situ".

(1). https://doi.org/10.3390/w13192660
(2). https://doi.org/10.1016/j.scitotenv.2009.10.020

OP5-2-2 Robust and portable ion chromatography-based nutrient analyser for in-field nitrite and nitrate monitoring in water

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Nitrate (NO3-) and nitrite (NO2-) are naturally occurring anions that play a crucial role in promoting the growth of aquatic plants and algae in environmental waters. However, anthropogenic activities, such as the overuse of fertilizers and the discharge of industrial wastewater and domestic sewage, can contribute to excessive levels of these anions, causing severe detrimental effects on both the aquatic environment, through processes such as eutrophication.[1] Therefore, regular monitoring the nitrate and nitrite levels in natural waters is essential to assess their concentration levels, and help identify and control any input sources to minimize their impact on both the ecosystem and human health. However, whilst traditional grab sampling and lab-based analysis are often used for monitoring nitrite and nitrate levels, they have a well-known tendency to produce significant errors due to the loss of anion nutrient species during transport and storage. This loss is largely caused by on-going biological activity, even during sample refrigeration.[2] Further, such approaches are manually intensive, have

limited detailed spatial coverage when applied to whole catchments, and typically miss isolated (and otherwise unknown) temporal point source inputs.

In this presentation, we will introduce the quantitative advantages of real-time, on-site analysis and address the problem of substantial underreporting that can occur with current "grab and lab" environmental monitoring methods. This study used a portable ion chromatography-based nutrient analyser (Aquamonitrix®) to measure the nitrite and nitrate concentrations in real-time, both from individual riverside locations, and during river catchment boat cruises, sampling from freshwater and semi-saline environments. All samples were collected and analysed immediately at each site, and then also analysed for seven days post-sampling to monitor nitrite and nitrate level changes under different transport and storage conditions. The real-time field analysis results were compared with daily laboratory analyses to assess nitrate and nitrite conversion rates as storage time progressed up to the seventh day.

[1] Wang, Q.H., Yu, L.J., Liu, Y., Lin, L., Lu, R.G., Zhu, J.P., He, L. and Lu, Z.L., 2017. Methods for the detection and determination of nitrite and nitrate: A review. Talanta, 165, pp.709-720. https://doi.org/10.1016/j.talanta.2016.12.044

[2] Avanzino, R.J. and Kennedy, V.C., 1993. Long-term frozen storage of stream water samples for dissolved orthophosphate, nitrate plus nitrite, and ammonia analysis. Water Resources Research, 29(10), pp.3357-3362. https://doi.org/10.1029/93WR01684

OP5-3-1 Monitoring lag-phase α-synuclein aggregation in various conditions using RT-fast

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Parkinson's disease, the second most widespread neurodegenerative disease, has over six million patients today. That number is expected to double by 2050. α-Synuclein is a biomarker for Parkinson's disease, it is a highly disordered protein whose aggregation is thought of as the cause of Parkinson's disease. The aggregation of α-Synuclein follows several phases that starts with a lag phase. This phase is characterized with the formation of toxic oligomers that are difficult to detect using the real time quick (RT-QUICK) method. RT-QUICK is the most widespread technique used for detecting α-Synuclein aggregation [1,2]. Nanopores are a tool used for single molecule detection that measures the quantity and the size of said particles through an electrochemical technique called Resistive pulse. Nanopipettes are a type of solid state nanopores that are cost-effective and require small volumes (in the order of a tenth of µL) [3]. Previous studies have used nanopipettes to follow the aggregation of α-Synuclein using a technique known as real-time fast amyloid seeding and translocation (RT-FAST). RT-FAST is a technique for measuring amyloid aggregation during the lag phase using a nanopipette. It consists of aggregating the amyloid inside the nanopipette then measuring the resulting aggregates in intervals over a period of time. [4]. In this study, we used RT-FAST to study the aggregation of α-Synuclein during the lag phase using nanopipettes with various sizes. α-Synuclein monomers were incubated inside several different nanopipettes of various pore diameters over a period of time. Then resistive pulse measurements were taken over a set interval of time during said period. The resulting current trace is then analyzed to determine the presence and size of the α-Synuclein aggregates. We followed this study by introducing α-Synuclein fibers and CuII that are known to affect α-Synuclein aggregation. The results show that the α-Synuclein fibers and CuII increase the size of α-Synuclein oligomers generated

during aggregation, thus showing their promoter effect on α-Synuclein aggregation during the lag phase.

[1] P. Magalhães, H.A. Lashuel, npj Parkinsons Dis. 8 (2022) 93. doi:10.1038/s41531-022-00357-0
[2] G. Musteikytė, et al., Biochimica et biophysica acta. Biomembranes 1863 (2021) 183536. doi:10.1016/j.bbamem.2020.183536
[3] N. Meyer, et al., Advances in Colloid and Interface Science 298 (2021) 102561. doi:10.1016/j.cis.2021.102561
[4] N. Meyer, et al., ACS central science 8 (2022) 441–448. doi:10.1021/acscentsci.1c01404

OP5-3-2 A generic approach based on long-lifetime fluorophores for the assessment of protein binding to polymer nanoparticles by fluorescence anisotropy

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The use of synthetic nanoparticles (NPs) has become indispensable in medical diagnostics, regenerative medicine, drug delivery and in bioassays as novel affinity materials and signal enhancers. When NPs are introduced into living biological systems, a protein layer, the so-called protein corona builds up on their surface, influencing their biological impact and functionality. [1] The protein-NP interactions, either specific or non-specific, are also determinant when using fully synthetic nanoparticle-based affinity reagents for in vitro assays in biofluids.

Currently, most approaches to quantify protein-NP interactions rely on the separation of the free and NP-bound proteins after equilibration [2]. However, low-affinity interactions, which exhibit high dissociation rates, cannot be studied in this way. The separation-free methods are more powerful in this respect, but their use is generally limited to particular types of nanoparticles, to high reagent concentrations, or involves the immobilization of one of the interacting partners [3].

Therefore, here we put forward a novel general and separation-free approach to quantitate protein-nanoparticle interactions based on measuring the fluorescence anisotropy changes of long-lifetime fluorophore (τ > several hundred ns) labelled proteins. With conventional fluorophore labels that feature only a few ns fluorescence lifetimes, this is not possible due to the inherently high anisotropy of the labelled proteins that would not change significantly upon binding to a NP. However, with long-lifetime fluorophores, the rotational correlation time of the labelled protein becomes insignificant compared to the fluorescence lifetime and low anisotropy is expected. Since NPs are several orders of magnitude larger than antibodies, the protein binding to the NPs would result in much higher rotational correlation times, and as a consequence, significantly higher anisotropy values. Thus, the protein-nanoparticle interaction could be sensitively detected and quantified in this way.

As a proof-of-concept, the interaction of lysozyme with engineered poly(N-isopropylacrylamide-co-N-tert-butylacrylamide-co-acrylic acid) NPs [4] was studied by using the long-lifetime, ruthenium-bis(2,2'-bipyridine)-4'-methyl-4-carboxy-pyridine complex as the protein label [5]. Fluorescence anisotropy measurements were performed to study the binding kinetics and affinity of the labelled protein to the NPs. From the binding isotherm the equilibrium dissociation constant and the cooperativity of the protein-NP binding were determined. The binding isotherm obtained from the anisotropy measurements was validated with an independent separation-based method. The bioanalytical use of the proposed approach was also shown through the quantitative assessment of lysozyme by competitive binding to the synthetic NP affinity ligand as a highly relevant model for further molecularly imprinted polymer nanoparticle (nanoMIP)-based assays.

[1] I. Lynch, et al., Nano Today 3 (2008) 40.
DOI: 10.1016/S1748-0132(08)70014-8
[2] R. García-Álvarez, et al., Nanomaterials 11 (2021) 888.
DOI: 10.3390/nano11040888
[3] L. Li, et al., Analyst 135 (2010) 1519.
DOI: 10.1039/c0an00075b
[4] K. Yoshimatsu et al., Angew. Chem. Int. Ed. 51 (2012) 2405.
DOI: 10.1002/anie.201107797
[5] H. Szmacinski et al., Biophys. Chem. 62 (1996) 109.
DOI: 10.1016/S0301-4622(96)02199-0

OP5-3-3 Application of capillary electrophoresis coupled to ICP-MS/MS for examination of cisplatin encapsulation in liposome nanocarriers

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Although cisplatin is an anticancer drug often used in the chemotherapy of various malignancies, its non-selective transport into cells results in serious side effects of treatment. Non-toxic nanomaterials such as liposomes can be used to provide targeted delivery of this drug. Features such as biocompatibility (similarity to cell membranes), biodegradability, ease of formation and surface modification, and the ability to adjust the degree of drug encapsulation distinguish them from other nanomaterials used as drug nanocarriers. Although 10 liposome–anticancer drug systems have already been approved for marketing, none contain cisplatin in their composition. This situation may result from i.e., the use of ineffective analytical tools for their characterization. Therefore, the aim of the work was, on the one hand, to elaborate a simple procedure for the formation of liposomes and effective encapsulation of the drug inside them and, on the other hand, to develop the method for their monitoring using capillary electrophoresis (CE) combined with inductively coupled plasma tandem mass spectrometry (ICP-MS/MS). Although the CE-ICP-MS technique has already been employed to study liposome-cisplatin systems, the ICP tandem mass spectrometer utilization is the analytical novum [1]. Thanks to the application of CE with ICP-MS/MS hyphenation, the optimized method can be used not only for the qualitative and quantitative monitoring of changes in the cisplatin–liposome systems (based on platinum and phosphorus isotopes detection) but also for their interactions with proteins (sulfur detection), which opens new analytical possibilities in the study of the transport mechanisms of these chemical individuals.

[1] A. Wróblewska, et. al, J. Anal. At. Spectrom. 37 (2022) 37, 1442. DOI: 10.1039/D1JA00459J

OP5-4-1 Optimization of MSI technologies for environmental toxicology: A case study with Zebrafish eleutheroembryos

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The Zebrafish (Danio rerio) has become a powerful model organism in a wide range of scientific fields, including ecotoxicology. The Zebrafish model presents various advantages concerning other common model organisms such as its effortless manipulation or large offspring. Strikingly, toxicological data from zebrafish can be extrapolated not only to aquatic species but also to other vertebrates, including humans. Moreover, zebrafish embryos are considered an excellent alternative animal model with fewer ethical restrictions, ensuring the fulfillment of the 3R's principle (Replacement, Reduction, and Refinement) in animal research. For that reason, their use in ecotoxicological studies and, particularly, in endocrine-disrupting chemicals (EDCs) assessment is broadly boosted.

Bulk omic technologies have contributed to a better understanding of how organisms respond to pollutants at the molecular level. However, bulk analysis disregards the heterogeneity of individual cell types and their spatial organization. In heterogeneous samples such as whole animals or embryos, it is critical to improve the knowledge of the effects of pollutants at the tissue or cellular level. To overcome these challenges, breakthrough technologies have emerged to encompass single-cell and spatially resolved omics, including mass spectrometry imaging (MSI).

Particularly relevant is the use of Matrix-Assisted Laser Desorption/Ionization (MALDI-MSI) owing to providing a favorable balance between sample preparation, chemical sensitivity, and spatial resolution [1]. Despite its outstanding features, MALDI approaches have some limitations in lipidomics studies. For instance, the conditions for the optimal ionization of certain lipid classes (i.e., sterols) or the spatial resolution compared to other MSI techniques [2]. For that reason, different analytical techniques (e.g., on-tissue-derivatization, and metal deposition) have recently been developed to improve MALDI capabilities. More recently, laser-post ionization coupled with the MALDI (MALDI-2) tool has emerged as a potential new game-changer in the MSI field [3]. MALDI-2 improve sensitivity for a high number of suppressed lipid classes leading to a decrease in pixel size.

In this study, we developed and optimized a spatial lipidomics protocol to analyse zebrafish embryo sections using both MALDI and MALDI-2-MSI with a lateral spatial resolution of up to 5 µm. Our results revealed the presence of different lipid clusters corresponding to different sections of the zebrafish embryo. Therefore, these results demonstrate the usefulness of spatial omics studies in this biological model, particularly underlining possible lipid biomarkers for relevant tissues such as the eye or brain.

[1] T.C.Baker, et al., Curr. Opin. Biotechnol. 43 (2017) 62-69. DOI: 10.1016/j.copbio.2016.09.003
[2] C.Harkin, et al., Mass Spec. Rev. 41 (2021) 662-694. DOI:10.1002/mas.21680
[3] M.Niehaus, et al., Nature Methods 16 (2019) 925-931. DOI: 10.1038/s41592-019-0536-2

OP5-4-2 The histone code of pancreatic cancer stem cells by nanoLC-MS/MS based epiproteomics

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Pancreatic ductal adenocarcinoma (PDAC) is an aggressive malignancy characterised by the presence of pancreatic cancer stem cells (PCSCs), which are pluripotent and self-renewable cells, capable of driving tumorigenesis, metastasis and chemoresistance. Notably, the epigenetic changes, including histone PTMs, are responsible for reprogramming, transformation and de-differentiation of cancer stem cells [1], and the pharmacologic inhibition of erasers and writers of histone PTMs could represent a strategic therapy against PDAC [2]. Despite this, a characterization of histone PTMs in PCSC is still missing, hindering the identification of new targets and, as a consequence, the development of effective drug treatments for PDAC.

In this study, parental and cancer stem cells of two PDAC cell lines (i.e., PaCa3 and Panc-1) were subjected to an epiproteomic analysis of histone acetylation and methylation. Extracted histones were mixed with super-SILAC mix [3], separated on a SDS-PAGE, H3 and H4 bands were excised and in-gel digested with trypsin. Peptide mixtures were then separated by a nanoLC-MS/MS. The epiproteomic data were then integrated with total proteome and immunoblotting analyses.

A total of 55 and 48 modified histone peptides were identified in PaCa3 and Panc-1 cell lines, respectively. The epiproteomics revealed altered histone PTMs distinguishing PCSCs from their relative differentiated cells, which are related to quiescence, apoptosis, chemoresistance and epithelial-mesenchymal transition. For example H4K20me3 (a repressive mark of key drivers of the epithelial state [4]) and H3K9me3 (a mark correlated to pro-apoptotic gene silencing in chemoresistant PDAC cells [5]) were found to be abundant in PCSCs of both cell lines as compared to parental cells. Other modified histone peptides dysregulated in PCSCs included H3K27me3, H3K4me3, acetylated H4K5K8K12K16 and acetylated H3K9K14. The induction in PCSCs of H4K20me3 was also confirmed by immunoblotting, whilst for the other histone marks the immunodetected band pattern was not easily interpretable. Interestingly, the comparative proteome analysis of Panc-1 cells allowed the detection in PCSCs of dysregulated proteins (including some histone methyltransferases) which influence specific histone modifications, as well as of epigenetic regulators, and of enzymes involved in 1C- metabolism, which also affects histone methylation.

In conclusion, this research paves the way for identifying the global epiproteomic signatures of pancreatic cancer stem cells, leading to the detection of new therapeutic targets, and the subsequent development of an improved PDAC anticancer therapy, which currently not effective on PCSCs.

[1] "Epigenetic regulation of cancer stem cell formation and maintenance" Int J Cancer 2021; 148: 2884-2897.
[2] "Epidrugs: targeting epigenetic marks in cancer treatment" Epigenetics 2019; 14: 1164-1176.
[3] "A Super-SILAC Strategy for the Accurate and Multiplexed Profiling of Histone Posttranslational Modifications" Methods Enzymol 2017; 586:311-332.
[4] "SUV420H2 is an epigenetic regulator of epithelial/mesenchymal states in pancreatic cancer" J Cell Biol 2018; 217: 763-777.
[5] "Contrasting roles of H3K4me3 and H3K9me3 in regulation of apoptosis and gemcitabine resistance in human pancreatic cancer cells" BMC Cancer 2018; 18: 149.

OP5-4-3 Development and validation of an untargeted LC-MS metabolomics method with post-column infusion for matrix effect monitoring in plasma and feces

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Untargeted metabolomics with RPLC-MS is a powerful tool that has demonstrated great potential in exploring metabolic changes in health and disease conditions. However, standardizing the development process requires attention to critical factors that are still under discussion or easily overlooked, such as injection solvent and sample injection amount optimization, performance validation, and matrix effect monitoring. In this study, we developed and validated an untargeted metabolomics method for plasma and fecal samples and implemented a post-column-infusion (PCI) approach for real-time matrix effect monitoring. Our results showed that optimizing the reconstitution solvent and injection amount was critical for balancing metabolite coverage and signal saturation in the RPLC-MS method. Method validation with stable-isotopically labeled standards (SILs) demonstrated good linearity, precision, accuracy, and acceptable recovery repeatability of our method. To tackle the issue of matrix effect, the PCI approach was applied to monitor the real-time absolute matrix effect (AME) and relative matrix effect (RME). The monitoring showed different AME and RME profiles between plasma and feces. The comparison of the REM data acquired by post-extraction spiking to that monitored with PCI compounds showed that these two methods were comparable in terms of REM assessment. Therefore, we applied the PCI approach to predict the RME of targets in our in-house library and found that targets detected in negative polarity were more vulnerable to RME, regardless of the sample matrix. Given the value of the PCI approach in identifying the strengths and weaknesses of our method in terms of matrix effect, we recommend implementing the PCI approach in method development and applying it routinely in untargeted metabolomics.

OP6-1-1 Electrochemical bioplatforms for sensing food derived nucleic acids: Aiding personalized nutrition

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Food allergies management has undergone a major transformation in the era of precision nutrition, which involves tailored nutritional recommendations to the sensitized patient. Since the avoidance diet must be tailored according to the patient, detailed information including advice on food labels and labeling laws, hidden allergens, and suitable food replacement must be provided[1]. Therefore, sensing food allergens and adulterations is of great importance to ensure consumers' protection and health. As an alternative to conventional allergen protein-based detecting techniques, since DNA preserves its integrity better than proteins during food processing[2], its determination represents a great potential for the sensitive and selective detection of food allergens.

We outline here the development of disposable nucleic acid-based amperometric bioplatforms, built on the surface of micro-size magnetic beads, relying on the formation of DNA/RNA heterohybrids which are recognized and tagged with specific antibodies and enzyme bioconjugates for the sensitive and selective interrogation of relevant DNA targets derived from animals or plants. The success of their application in the identification of tomato[3] and mustard[4] genomic DNA led us to explore the interrogation of other organelle-derived genome targets such as chloroplast and mitochondrial DNA, which contain a higher number of copies per gene, thus improving the throughput of gene extraction processes and considerably simplify the entire determination protocol. In this context the pioneering determination of meat adulteration directly in raw mitochondrial lysates was demonstrated, reaching the legislation required limits, without any extraction or amplification methods[5] and the interrogation of specific peanut chloroplast DNA sequences is currently being evaluated. The unique features in terms of simplicity, sensitivity, disposability, affordability, and compatibility with multiplexed determinations make these bioplatforms a very interesting complement to established conventional methodologies for identifying any type of nucleic acid, regardless of its nature (DNA or RNA) and origin (plant or animal), which make them a very promising alternative to ensure food safety and labeling regulations to guarantee personalized nutrition.

1. E. D'Auria, et al., Nutrients 11 (2019) 359. DOI: doi:10.3390/nu11020359.
2. R. Linacero, et al., Crit. Rev. Food Sci. Nut. 60 (2020) 1077.DOI:10.1080/10408398.2018.1557103.
3. M. Pereira-Barros, et al., Biosens. Bioelectron. 137 (2019) 171. DOI: 10.1016/j.bios.2019.05.011.
4. M. Gamella, et al., 150 (2023) 108357. DOI: 10.1016/j.bioelechem.2022.108357.
5. V. Ruiz-Valdepeñas Montiel, et al., Anal. Chem. 89 (2017 9474. DOI:10.1021/acs.analchem.7b02412

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Identifying and quantifying 3,4-methylenedioxymethamphetamine (MDMA) on-site in suspected illicit drug samples, whether it be at recreational settings or manufacturing sites, is a major challenge for law enforcement agencies (LEAs). Various analytical techniques exist to fulfil this goal, e.g. colourimetry and portable spectroscopic techniques, each having its specific limitations (e.g. low accuracy, fluorescence, no quantification) and strengths (e.g. fast, easy to use). Here, for the first time, an electrochemical MDMA sensor is presented to become a detection tool that can realistically be used on-site. More specifically, the use of a single buffer solution and an unmodified screen-printed electrode, along with the integration of a data analysis algorithm and mobile application permits the straightforward on-site identification and quantification of MDMA in suspicious samples. Multiple studies investigating different parameters, including pH, concentration, reproducibility, temperature and binary mixture analyses, were executed. To fully understand all the occurring redox processes, liquid chromatography coupled with high-resolution mass spectrometry analysis of partially electrolyzed MDMA samples was performed unravelling oxidation of the methylenedioxy group. Validation of the methodology was executed on 15 MDMA street samples analysed by gas chromatography coupled with mass spectrometry and compared with the performance of a commercial portable Raman and Attenuated Total Reflection Fourier Transform Infrared Spectroscopy (ATR-FTIR) device. The novel methodology outperformed the spectroscopic techniques, correctly

identifying all 15 street samples. Additionally, the electrochemical sensor predicted the purity of the tablets with a mean absolute error of 2.3%. Furthermore, the MDMA sensor is involved in multiple projects with partners in supply reduction and harm reduction organizations (Police Amsterdam, Sciensano,...), on which will be reported during the conference. Overall, this new, electrochemical detection strategy provides LEAs the rapid, low-cost, on-site detection and quantification of MDMA in suspicious samples, without requiring specialized training.

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OP6-1-3 Electrochemical biosensing platforms in molecular oncology for clinical sample analysis

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Electrochemical detection of nucleic acids, i.e., DNA and RNA, is a promising analytical tool for determination of these biomacromolecules as important cancer biomarkers. The main advantages of electrochemistry are low instrumentation cost, speed, simplicity, low sample consumption and the possibility of miniaturization, making it suitable for personalized decentralized medicine at the point-of-care. When coupled to novel, PCR-free isothermal amplification techniques, such as loop-mediated amplification (LAMP), rolling circle amplification (RCA) or recombinase polymerase amplification (RPA), excellent sensitivites and selectivities can be achieved. We show here application of these techniques into analysis of clinical samples from oncological patients, targeting diverse DNA/RNA biomarkers (1-4). For instance, we developed bioassays for analysis of HPV oncoviruses in cervical cancer, long non-coding RNAs in prostate cancer, or DNA point mutations in BRAF gene in colorectal cancer or melanoma. Electrochemistry could thus be an interesting alternative in current molecular cancer diagnostics.

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1) https://doi.org/10.1016/j.aca.2018.08.020
2) https://doi.org/10.1039/C9AY02383F
3) https://doi.org/10.1016/j.aca.2021.339145
4) https://doi.org/10.1016/j.talanta.2021.123064

OP6-2-1 Phytosomes use to enhance the anti-ageing effectivness of nutraceutics and cosmeceutics

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Phytosomes are structures mimicking cells obtained by a stoichiometric reaction between active compounds from an extract and phospholipids (phosphatidylcholine-PC, phosphatidylserine-PS, lecithin-LE).

Mainly, the reaction consists of bounding the choline moiety to hydrophilic compounds while the lipid soluble phosphatidyl moiety covers the choline -bound complex, thus enhancing lipid solubility. Phytosomes preparation was performed via antisolvent precipitation process using an aqueous solution as a contra-solvent. PC/PS or LE were used as phospholipid, standardized extracts in quercetin, rutin and silymarin, respectively, from extracts obtained using corresponding vegetal by-products: rosehip, sea buckthorn and respectively milk thistle.

The FTIR, SEM and DLS measurements were performed to assess the phytosomes and phytosomes-active compounds complexes. Phytosomes nanoparticles show values of hydrodynamic diameter between 266.1 ± 8.8 nm and 317.4 ± 86.7 nm and negative zeta potential, ranging between -6.79 and 0 mV.

FTIR spectroscopy confirmed the presence of physical and chemical interactions between active compounds and phospholipid, the band intensity at 3550 (O-H), 2940 (-C-H), and 1752 (-C=O) cm^{-1} decreasing or even disappearing in the phyto-formulation, while shifting of the frequency was notices. Phytochemical content, encapsulation efficiency, and release percentage were assessed by the HPLC-DAD-MS method. Encapsulation efficiency ranged between 81 and 95%. Loading capacity was around 25% for all tested extracts. In vitro active compounds release studies were performed using two media simulating the gastrointestinal conditions and one for trans-dermal simulation since obtained phytosomes addressed nutraceutical and cosmeceutical formulation. For silymarin it was proved that a following the gastrointestinal transit more than 10 % of silymarin remains encapsulated, allowing greater intestinal absorption. Results were similar in the case of phytosomes formulated with the active principle in pure state (standard silymarin complex).

Release profile for phytosomes complexes with rosehip extracts and, respectively, sea buckthorn proved a maximum release at the gastric level.

It was shown that phytosomes are able to enhance the active compounds availability/bioavailability when doze-effect response is compared to that obtained using classical formulation.

OP6-2-2 Analysis of PFAS from food samples

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According to the OECD, over 4730 per- and polyfluoroalkyl substances (PFAS) are currently known [1]. Due to their physico-chemical properties, they are used in a wide range of industries worldwide (e.g. textiles, household products, firefighting, automotive, food processing, construction, electronics). The exposure to PFAS may lead to adverse health effects. To protect human health, the exposure of the levels of PFAS along the food chain must be investigated intensively. Therefore, there is need for more sensitive analytical methods for PFAS in food of animal and plant-based origin. This work compares the analysis of PFAS from food according to FDA Method C-010.02 and the Guidance of the network of the European Union Reference Laboratory (EURL) on Analytical Parameters for the Determination of Per- and Polyfluoroalkyl Substances (PFAS) in Food and Feed [2, 3].

The most important step in the successful analysis of PFAS at very low concentrations is sample preparation. This work presents 2 method modules that may be selected depending to laboratories requirements. Modul 1, including a sample preparation with a solid-phase extraction (SPE) method, is

based on the interaction of PFAS with a mixed-mode, weak anion-exchange sorbent. Further clean-up with graphitized carbon black (GCB) material could be done before or after SPE. Modul 2 consist a dispersive solid-phase extraction (dSPE). For the dSPE, a mixture of different salts (e.g. anhydrous MgSO4, NaCl) and sorbents (e.g. primary secondary amine, GCB, C18) can be used.

By using speacially designed SPE columns and dSPE salt mixtures for the tested method modules, it was possible to obtain high recovery rates for 40 PFAS between 80% and 100% with good reproducibility. The method also lead although to an effective matrix reduction and improves the analysis and save time and laboratory cost. The identification and the quantification of PFAS in food were finally carried out by ESI mass spectrometry on a NUCLEODUR® PFAS column.

[1] Toward a New Comprehensive Global Database of Per- and Polyfluoroalkyl Substances (PFASs): Summary Report on Updating the OECD 2007 List of Per- and Polyfluoroalkyl Substances (PFASs), Series on Risk Management, No. 39, ENV/JM/MONO(2018)7.
[2] FDA Method C-010.02 - Determination of 16 Perfluoroalkyl and Polyfluoroalkyl Substances (PFAS) in Food using Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS)
[3] Guidance Document on Analytical Parameters for the Determination of Per- and Polyfluoroalkyl Substances (PFAS) in Food and Feed, 2022, ANEX Version 1.0

OP6-2-3 The Chocolate Benchmark: Evaluating latest PTR-MS Advancements

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25 years ago, one of the first applications used for demonstrating the advantages of the then new Proton-Transfer-Reaction–Mass Spectrometry (PTR-MS) technology was food and flavor analysis [1]. Nowadays, PTR-MS is well-established in this field with hundreds of publications on in vitro and in vivo studies [2]. One particularly complex matrix has frequently been reported on from the very beginnings [3] up to today [4]: chocolate and chocolate products. Hence, chocolate analysis is an ideal benchmark for evaluating instrumental improvements in PTR-MS.

First, we present results of a nosespace study during chocolate consumption utilizing a common PTR-MS instrument equipped with a DC drift tube followed by an RF+DC ion funnel and a TOF analyzer tuned to a mass resolution of about 5,000 m/dm. The study was repeated using a second setup with an identical PTR region, but a novel TOF analyzer capable of mass resolutions up to 15,000 m/dm. A heated interface with disposable nosepieces was connected to the devices for direct sampling of exhaled nosespace air of the test subjects.

The considerably increased selectivity of the high-resolution device is crucial for separating the key aroma compound of cocoa - trimethylpyrazine (C7H10N2) - from the highly abundant isobar C9H14 in room air. With 5,000 m/dm the two molecules can hardly be distinguished, while at >10,000 m/dm two clearly separated peaks are visible in the mass spectrum. That is, with 5,000 m/dm during the blank exhalations the measured concentration (sum of isobars) oscillates around several hundred pptv. After ingestion and starting to chew a piece of chocolate the concentration does not change significantly. It is not clear if trimethylpyrazine is released into the nosespace at all. With 10,000 m/dm the concentration of trimethylpyrazine in blank nosespace is only at about 20 pptv, i.e. the signal is not masked by C9H14 from room air. During chocolate consumption the release of trimethylpyrazine into the nosespace can be monitored with 200 ms time resolution.

For a second study we utilized a PTR-MS setup with a novel RF+DC reaction region which results in a considerable increase of sensitivity up to 80,000 cps/ppbv at 10,000 m/dm. The headspace above solid pieces of chocolate (raspberry, caramel, grappa) was analyzed and the respective flavor was identified via marker compounds, such as maltol, methyl butyrate, ethyl octanoate, etc. Remarkably, the relative errors of marker compound quantification is well below 1% at only 1 s measurement time per sample.

[1] Lindinger et al. https://doi.org/10.1016/S0168-1176(97)00281-4
[2] Biasioli et al. https://doi.org/10.1016/j.trac.2011.03.009
[3] Lindinger et al. https://doi.org/10.1088/0963-0252/6/2/004
[4] Gonzalez-Estanol et al. https://doi.org/10.1016/j.foodres.2023.112726

OP6-3-1 Extension of LC-MS Stability Studies of Eltrombopag Olamine to In-silico Simulations: An Effort to Exploit Drug Related Substances in Drug Discovery

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The recent pandemic has refocused the impact of diseases, their diagnosis, treatment, and influence, on the worldwide health and economy. The rapid screening/discovery of new lead molecules for drug discovery is still a tough challenge. Pharmaceutical impurity profiling and drug discovery can be linked effectively for the discovery of new leads, thus assisting in modern drug discovery approaches. The present study demonstrates such linking by extension of the impurity (forced degradation) profiling for eltrombopag olamine (ELT-O). The drug was exposed to standard degradation, considering its intrinsic stability. The degradation products were primarily resolved by HPLC. This was followed by UPLC-TOF-MS analyses, which led to the identification of five forced degradation products (FDP). The other 33 known related substances (RS) of ELT-O were also considered and molecular similarity checks were performed using Tanimoto similarity searches. A set of structurally and topologically similar molecules was established (ELT-O and 16 RS) and subjected to in-silico ADME and toxicity studies. The RS showing similar or less toxicity than ELT-O and a comparable ADME profile were subjected to molecular docking to trace changes in thrombopoietin receptor affinity. The results indicated that three RS (MEI, I28, I3) had a Jaccard's similarity with the parent drug and higher docking scores, while another RS (EEI) had comparable docking scores with ELT-O. This justifies their entry as new chemical entities (NCE) and potential novel leads as thrombopoietin receptor agonists in drug discovery approaches, with an alternative possibility to explore them for other therapeutic targets.

1. S.B. Ganorkar and Y. Vander Heyden, TrAC-Trends Anal Chem 157 (2022) 116747.
DOI: 10.1016/j.trac.2022.116747
2. S.B. Ganorkar, et. al. J. Pharm. Biomed. Anal. 179 (2020)112982.
DOI: 10.1016/j.jpba.2019.112982
3. David A, J Mole Biol. 434 (2022) 167336.
DOI: 10.1016/j.jmb.2021.167336
4. Jaghoori MM, et al. J. Comput-Aided. Mol. Des. 2016;30(3):237-249.
DOI: 10.1007/s10822-016-9900-9
5. D. Das, et al., J. Chem. Info. Model. 63 (2023)1882-1893.
DOI: 10.1021/acs.jcim.2c01301

OP6-3-2 Development of an analytical method for a fast and accurate determination of elemental impurities in drug products by ICP-MS with a quantification based on isotopic dilution

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To protect public health, the pharmaceutical sector requires a high level of quality assurance in the control of organic and inorganic contaminants. The International Council for Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) established new guidelines to determine elemental impurities in pharmaceutical products. Since 2017, 24 elemental impurities have been restricted, and their concentration is limited based on their toxicity and the drug product's administration route. Simultaneously, the USP released general chapters (<232> and <233>) outlining analytical limits and procedures for the control of elemental contaminants in such samples, and atomic spectrometry techniques such as ICP-OES and ICP-MS are highly recommended. This study focuses on the development of a method for the rapid and precise determination of elemental impurities in pharmaceuticals using ICP-MS and an isotope dilution quantification strategy. The approach was developed and optimized taking into account different analytical steps such as sample preparation, instrument settings, and analytical conditions. To obtain spike recoveries between 70 and 150 % as recommended by the USP, different acid mixtures were tested for the acid digestion step. First, we examined a mixture of nitric acid, which is recognized for its oxidizing action, and hydrochloric acid, which allows elements such as mercury and platinoids to be stabilized in solution. Furthermore, several of the medications used for this study (drugs for cardiovascular and gastrointestinal diseases) include silica and/or titanium dioxide, requiring the addition of hydrofluoric acid. In addition, to select between two heating devices (microwave and hotblocks), screening experimental designs were applied. Better recovery rates (85-103 %) were obtained for all the elements with hotblocks with an optimized digestion duration of 130 min and a temperature of 92°C. Monoisotopic elements were quantified with standard additions to avoid important matrix effects during ICP-MS analysis. For non-monoisotopic elements (Li, Cr, Ni, Cu, Se, Mo, Ag, Cd, Sn, Sb, Ba, Pt, Hg, Tl, Pb), the Isotope Dilution Mass Spectrometry (IDMS) method was applied in the optimized acid digestion conditions with a comparison between a before and after digestion spiking procedures. Better recovery rates (82-110 %) were obtained with a spiking procedure before digestion allowing the compensation of the losses of the elements during digestion and matrix effects. Results demonstrated that the application of IDMS for non-monoisotopic elements analysis in drug samples is a fast quantification method extremely precise and accurate and meets the requirements of the pharmacopeia.

OP6-3-3 How to Overcome Analytical Challenges Commonly Encountered in the Analysis of Cr and Cr(VI) in Environmental and Biological Matrices Using (µLC-) ICP-MS

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Chromium (Cr) mainly exists in the environment as trivalent Cr(III) and hexavalent chromium Cr(VI). Cr(III) is an important micronutrient, while Cr(VI) is an occupational lung carcinogen. The chemistry of Cr plays a major role in its cellular entry and toxic effects. Only Cr(VI) can easily pass the cell membrane. Once inside the cell, Cr(VI) is rapidly reduced to Cr(III). This reduction process can cause damage to cellular components. A sensitive and robust method for the simultaneous determination of Cr(III) and Cr(VI) has been developed. The method uses a hyphenated micro liquid chromatography system coupled to inductively coupled plasma mass spectrometry (µLC-ICP-MS). This presentation will highlight the analytical challenges (including pH dependency, contamination and soot deposit in ICP-MS) encountered during method development. The method has been applied to environmental and biological samples collected within a European human biomonitoring study. The study aimed to harmonize procedures for human biomonitoring. Human biomonitoring indicates exposure to chemicals by measuring either chemicals or markers of subsequent health effects in body fluids or tissues. This presentation will highlight the harmonization challenges (including interlaboratory comparison and availability of certified reference materials [CRM]). The human biomonitoring study evaluated the occupational exposure to Cr(VI). Samples were collected from 299 workers and 103 controls. The principal biomarker used for biomonitoring of Cr(VI) exposure at the workplace is total amount of Cr in urine. The main limitation of this biomarker is that it is not specific for Cr(VI) since it reflects exposure to both Cr(III) and Cr(VI). We studied the use of potential more specific biomarkers, such as Cr in red blood cells (RBC) and Cr(VI) in exhaled breath condensate (EBC). Cr in RBC reflects the exposure specifically to Cr(VI) since only Cr(VI) is able to pass though the red cell membrane. Cr(VI) in EBC can give specific information on the Cr(VI) levels in the lungs (main target tissue). This presentation will highlight the main findings of this study related to the analytical challenges (including low levels and stability). As indicated in this study, the analysis of Cr or Cr(VI) in environmental and biological samples is subject to challenges. Precautionary procedures needs to be taken during method development, analysis, sampling and storage. For the future success of chromium speciation in EBC, CRMs in water or EBC needs to be made available.

Goldoni et al., J Environ Monit, 2010, 12(2), 442–447. DOI: 10.1039/b914673c.
Leese et al., Int. J. Hyg. Environ. Health, 2017, 220(2 Pt B), 415–423. DOI: 10.1016/j.ijheh.2016.12.003.
Santonen et al., 2019, Environ. Res., 177, 108583. DOI: 10.1016/j.envres.2019.108583

OP6-3-4 Interaction between Gemcitabine and divalent metal cations: a speciation study with implication in nanomedicine

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Gemcitabine (GMT) is a nucleoside analog approved by Food and Drug Administration (FDA) for the treatment of various types of solid tumors. However, the rapid metabolism of GMT has greatly limited its potential as a chemotherapeutic agent [1]. Liposomal GMT formulations, aimed at improving its metabolic stability, have led to poor encapsulation efficacy (EE) and drug loading (DL), making the

administration less effective. The interaction between drugs and divalent metal cations has been exploited as driving force for drug loading, allowing to obtain higher DL and EE values [2]. Starting from this premise, a speciation study was conducted to determine the extent of the interaction between GMT and Mn(II) and Zn(II) as a function of parameters such as pH, temperature and metal/drug ratio. This allows to identify the optimal conditions that may be employed in the development of alternative GMT liposomal formulations. A preliminary acid-base study was thus conducted by potentiometric titrations at different values of temperature (t = 15, 25, 37, 45°C) and ionic strength (I = 0.15, 0.5, 1 mol L^{-1}) on NaCl aqueous solutions containing GMT. Potentiometric titrations were carried out on solutions containing various metal to GMT ratios in order to select the best speciation model and to determine the formation constants values of the species formed in solution. Spectrophotometric titrations were also performed under the same conditions to confirm the speciation models and the constant values, as well as to investigate the spectrophotometric behavior of the species. Furthermore, the results of ^{1}H-NMR titrations, employed on solutions containing GMT and Zn(II)-GMT at T = 25°C and I = 0.15 mol L^{-1}, are in full agreement with the results gained by potentiometry. Enthalpy change values were determined by the dependence of the formation constants on the temperature. The sequestering ability of GMT towards Zn(II) and Mn(II) was evaluated under different conditions of pH, ionic strength and temperature by an empirical parameter known as $pL_{o.5}$, i.e., cologarithm of the ligand concentration required to sequester 50% of the metal ion present in traces [3]. The results of this thermodynamic study will provide critical information for the development of new liposomal formulations by exploiting specific interactions between the biocompatible divalent metal cations Mn(II) and Zn(II) with GMT with the aim to achieve higher EE and DL values and the ability to further control release rates of such nanoformulations that may find use in the treatment of various cancers.

[1] T. Matsumoto, et al., Pharm Res. 38 (2021) 1093. DOI: 10.1007/s11095-021-03045-5
[2] G. N. C. Chiu, et al., J Control Release, 104 (2005) 271. DOI:10.1016/j.jconrel.2005.02.009
[3] A. Gianguzza, et al., Coord. Chem. Rev. 256 (2012) 222. DOI:10.1007/s00216-012-6336-5

OP6-4-1 Fast semi-quantification of plasticizer metabolites in urine by the use of a guard column coupled to mass spectrometry

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Phthalates (diesters of the 1,2-benzenedicarboxylic acid), their isomers terephthalates (diesters of the 1,4-benzenedicarboxylic acid), and DINCH (di-iso-nonyl cyclohexane-1,2-dicarboxylate) have been widely used as plasticizer additives [1]. Quantifying human exposure to these compounds is usually performed through the determination of their major metabolites in urine. In most cases, a solid-phase extraction procedure followed by a high- or ultrahigh-performance liquid chromatography tandem mass spectrometry (HPLC-MS/MS or UHPLC-MS/MS) analysis is applied, resulting in very sensitive but expensive, and time-consuming, methods [2].

In this work, we propose a simple and fast method of only 2 min (3 min including the injection cycle) for the determination of 19 plasticizer metabolites in urine. The sample treatment consists of an enzymatic hydrolysis of only 10 min (although larger hydrolysis times have been described-not optimized, in literature), filtration, and subsequent dilution 1/10 v/v with ultrapure water. Diluted samples are injected into a guard column directly coupled to an MS/MS system (guard column-MS/MS). Enzymatic hydrolysis, filtering material, and guard column-MS/MS conditions were optimized

in detail. Limits of quantification ranged from 2.8 to 60 ng/mL, far below the biomonitoring equivalents derived from tolerable daily intakes and reference doses set by EFSA and EPA for some phthalates. Trueness values, calculated as apparent recoveries, ranged from 70 to 135%; intra-day precision at the low concentration level, expressed as relative standard deviation, varied between 11 and 20%. To correct for matrix effects, analyte concentrations in urine were quantified by the standard addition method.

To confirm the results obtained by guard column-MS/MS, an UHPLC-MS/MS method was also applied (17 min of run time). A good agreement was achieved between the concentrations measured with both methods, enhancing the use of the guard column-MS/MS procedure to analyze a large number of samples in a very short time (semi-quantification), and limiting the application of UHPLC-MS/MS to those samples with levels close to/higher than their biomonitoring equivalents (confirmation). This double strategy (semi-quantification and confirmation-when needed) implies significant savings in time and money, since both the cost of the guard column, compared to the chromatographic column, and the run time are reduced considerably.

[1] US Environmental Protection Agency, Phthalates Action Plan (2012) 1–16
[2] A. Ramesh Kumar, P. Sivaperumal, Trends Anal. Chem. 75 (2016) 151-161. https://doi.org/10.1016/j.trac.2015.06.008

OP6-4-2 Propose 'NO' to heart disease! Tracer-based metabolomics: Profiling Nitric Oxide (NO) metabolites in a 3D cell culture model

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Introduction:
The measurement of nitric oxide (NO) metabolites is important in the diagnosis and management of cardiovascular disease. NO is generated by the enzyme endothelial nitric oxide synthase (eNOS/NOS3) and exerts multiple key roles in vascular homeostasis including vasodilation, and inflammation. In endothelial dysfunction (ED) conditions (uncoupling eNOS, inflammation, lack of fluid shear stress, etc.,) the reduced availability of NO converts the endothelial cell phenotype to a pro-inflammatory state with increased oxidative stress leads and a loss of its vasodilatory capacity1–3.

Aim:
To profile nitric oxide metabolites in a 3D micro-vessel-on-a-chip model using a tracer-based metabolomics strategy. This work is to highlight the importance of flow in eNOS activation in terms of NO metabolites.

Methods:
Human Coronary Artery Endothelial Cells (HCAECs) were cultured in a 2D well plate and the 3D platform that was attached to the microfluidic pump for a unidirectional flow system in a controllable manner. The NO substrate – L-Arginine (13C6,15N4-L-Arginine) was treated in the cells along with the stimulatory and inhibitory compounds based on eNOS and arginase enzyme for pathway analysis. The samples were derivatized using the AccQTag reagent and measured using the UPLC-modified MS/MS method.

Results:

We investigated the level of stable-isotope labeled metabolites in the NO mechanism to determine the eNOS activity by tracking the conversion of L-Arginine to L-Citrulline and L-Ornithine. The critical evaluation of marker metabolite levels at extracellular and intracellular levels provided insights to understand the cell-cell interactions and cellular processes leaving a strong scope for metabolic flux analysis. Compared to the 2D culture, the augmented effects of the NO-specific metabolite L-citrulline in 3D blood vessels were reported due to the presence of hemodynamic shear stress. We also studied the impact of oxygen on endothelial dysfunction and NO metabolism with an in-line oxygen measurement system.

Conclusions:
Our detection method and 3D model with a unidirectional fluid flow provide a more representative physiological environment that exhibits a perfect model to study endothelial dysfunction (ED).

References:
(1) Junaid, A.; et al, Metabolic Response of Blood Vessels to TNFα. Elife. 9 (2020).
DOI: 10.7554/eLife.54754.
(2) Tovar-Lopez, F.; et al, A Microfluidic System for Studying the Effects of Disturbed Flow on Endothelial Cells. Frontiers in Bioengineering and Biotechnology. 7 (2019).
DOI: 10.3389/fbioe.2019.00081
(3) Williams, D.; et al., Stable Flow-Induced Expression of KLK10 Inhibits Endothelial Inflammation and Atherosclerosis. eLife. 11 (2022).
DOI: 10.7554/eLife.72579.

OP7-1-1 Electrochemical study of recombinant manganese peroxidase from maize along with nanocomposite materials for glucose detection.

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Diabetes is a chronic disease that affects about 150 million people worldwide and is one of the leading causes of death and disability. Diagnosis of diabetes requires close monitoring of blood glucose levels for millions of people with diabetes, making glucose the most common analyte tested. Electrochemical sensors are one of the prominent devices that can be easily used for practical applications and bioassays, moreover applied for making economical, portable, and disposable electrode systems. Biosensors are essential in clinical, biological analysis, and environmental monitoring. Due to critical clinical and industrial applications, glucose sensors, one of the most popular biosensors, have been comprehensively studied. The enzyme immobilization composite is crucial in biosensor modification. This can profoundly affect the electrocatalytic activity of the enzyme on biosensors. Immobilizing a biomolecule, such as an enzyme, using electropolymerized film is a simple one-step procedure. Flexible biosensors using conductive polymers(CPs) have attracted much attention due to their potential for use in various fields. CPs can quickly obtain by electrochemical oxidation of aromatic monomers on the electrode surface to create a thin homogeneous layer. Conductive polymer (CP) nanomaterials have beneficial properties and a high potential for cost-effective, large-scale, lightweight, and flexible biosensors. Using nanoparticles (NPs) such as AuNPs to modify sensors shows significant advantages in increasing the mass transfer rate, electrocatalytic activity, and a higher reactive surface. In this work, we study the effect of co-immobilized PPMP, AuNPs, and GOx onto SPEs by electropolymerization of CPs using the electrochemical properties and parameters of an amperometric glucose biosensor.
Corn extracted enzyme, Enzyme-Based Biosensors, Gold Nanoparticles, conductive polymers, Glucose.

OP7-1-2 Biochar - nontraditional and green electrode material for miniaturized electrochemical sensors

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In the last decade, research focused on recycled carbonaceous materials prepared from renewable energy sources or waste shows that, apart from the environmental aspect, they have considerable application potential as more sustainable carbon materials. Biochar is a typical member of this group representing carbonaceous material prepared by the thermal decomposition of biomass in an oxygen-limited environment. Since biochar contains mainly stable aromatic forms of organic carbon, which cannot be easily returned to the atmosphere as CO2, its use is compatible with green analytical chemistry. Due to its low cost with a prominent carbon concentration, high specific surface area and large porosity, chemical stability, tunable functionalization, electrical conductivity and availability, biochar has been intensively studied in the last decade in energy storage and wastewater treatment as well as in electrochemical sensors. Despite tremendous progress in the development of miniaturized and portable systems, electrochemical sensors prepared from green, alternative, low-cost materials remain a key challenge in analytical chemistry, electrochemistry, nanotechnology, and materials chemistry.

This contribution reports the characterization and analytical application of biochar/ethylcellulose-modified carbonaceous electrodes as a new generation of fully printable miniaturized electrochemical sensors. The environmental-friendly screen-printable inks containing exclusively non-toxic and biodegradable components (biochar and ethylcellulose) were prepared and analyzed by rheological and thermal stability measurements. The viability of these electrochemical sensors was demonstrated by the development and full validation of a new and sensitive square-wave voltammetric method for the fast and reliable determination of the selected drug – paracetamol - in pharmaceutical formulations. Within the method development, all necessary aspects, such as a study of the effect of ethylcellulose concentration, pH study of supporting electrolyte, mechanism of oxidation reaction of the analyte, selection of pulse parameters and analytical performance, were investigated. The obtained results allow us to predict the production and subsequent use of new, printable, cheap, and environmentally friendly sensor platforms which will exhibit good analytical performance with the possibility of commercialization.

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D. Spanu, et al. Microchem. J. 159 (2020) 105506. DOI:10.1016/j.microc.2020.105506

OP7-1-3 Promotion and inhibition of electrochemical reaction for electroactive small molecules on monolayer graphene surface

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Monolayer graphene is attracting attention as a biosensor electrode material because of its excellent electrical conductivity and high biocompatibility. So far, theoretical studies predicted that the graphene surface exhibits a high adsorption interaction for potassium ions (K⁺) [1]. However, there are few experimental reports on how the adsorption interaction of K⁺ on the graphene surface affects electrochemical reactions.

In this study, we fabricated monolayer graphene electrodes (MGE) and investigated the dependence of the K⁺ concentration on redox reactions of small electrochemical species. We measured cyclic voltammograms (CV) of anionic, neutral, and cationic metal complex ions (specifically, [$IrCl_6$]^{2-/3-}, water-soluble ferrocene [FcTAB]^{0/1+}, and [$Ru(NH_3)_6$]$^{3+/2+}$), and two bio-related redox species that behaves as anionic ions in neutral pH solution, namely, ascorbic acid [AA] and uric acid [UA] by varying K⁺ concentration from 0 to 100 mM. We found that the CV of anionic [$IrCl_6$]^{2-/3-} shows significant increase in the peak current for both oxidation and reduction with increasing K⁺ concentration (Promotion effect). It is considered that electrostatic attraction between anionic [$IrCl_6$]^{2-/3-} and K⁺ adsorbed on the monolayer graphene surface causes increase in the density of [$IrCl_6$]^{2-/3-} near the electrode and resulted the increase in the reaction current. Furthermore, the oxidation reactions involving [$IrCl_6$]^{2-/3-}. It indicates that the higher the valence of the anion, the stronger the attractive force acting between K⁺ enhanced the promotion effect. In contrast, as regards the reaction involving cationic species, both oxidation and reduction of [$Ru(NH_3)_6$]^{3+/2+} and the reduction reaction of [FcTAB]⁺ decreased in peak current with increasing K⁺ concentration (Inhibition effect). As for the charge-neutral specie as [FcTAB]^{o}, K⁺ concentration had almost no effect on the peak current. The similar promotion effect was also observed in the reaction of two bio-related anionic species, ascorbic acid [AA] and uric acid [UA]. We then measured CV of above redox species by using a commercially available graphite electrode (with the exposed surface oriented to the basal plane, PGBE) and found that neither promotion nor inhibition effect depending on K⁺ concentration was observed, regardless of the charge of the electrochemically active species.

In summary, a unique electrochemical phenomenon of MGE was found that the redox reaction of anions is promoted while that of cations is inhibited with increasing the potassium ion concentration [2]. The mechanism is explained by the electrostatic interaction between anions/cations and K⁺ adsorbed on the graphene surface.

[1] A. Lugo-Solis et al., Phys. Rev. 76, 235431(2007). https://doi.org/10.1103/PhysRevB.76.235431
[2] S. Kojima et al., Chem. Sens., 38(A), 55-57 (2022).
https://chemsens.electrochem.jp/journal/2022/2022a-abt.html#17

OP7-1-4 A Physically Small, Antifouling Sensor for Selective Detection of Dopamine

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In this work, we will focus on our efforts in developing structurally small carbon electrodes with considerable antifouling capabilities against both biofouling and electrochemical fouling, making them suitable for in vivo dopamine detection in a complex biological matrix. Very often, a challenge during

in vivo dopamine detection is biofouling of electrodes arising from impeded electron transfer of dopamine on an electrode by a nearly impermeable layer formed by non-specifically adsorbed amphiphilic biological molecules (proteins, peptides, lipids, etc.) present in extracellular fluid. Similarly, oxidation of dopamine is known to yield an adsorbed dopamine-o-quinone layer on an electrode surface that leads to electrochemical fouling. Diminishing transient dopamine signals in such work have generated compromising results in time-dependent in vivo dopamine detection experiments. In this work, we have systematically investigated an organic silane reduction strategy on structurally small carbon electrodes (~2 µm tip diameters and ~9 µm axial length) to develop a hydrogenated carbon sensor with a hydrophobic surface that deters adsorption of amphiphilic species and dopamine-o-quinone, while favouring the dopamine electron transfer reaction. Results obtained using triethylsilane, n-butylsilane, phenylsilane, and diphenylsilane will be presented. The antifouling properties of these carbon electrodes will be compared by evaluating the analytical detection of dopamine at electrodes that were deliberately incubated in a laboratory synthetic fouling solution containing bovine serum albumin (a protein), cytochrome c (a protein), caproic acid (a lipid) and human fibrinopeptide (a peptide), before being applied to real-life biological samples.

OP7-2-1 An on-site sample preparation approach for plant eco-metabolomics and its application to agroecosystems in East Africa

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Mass spectrometry-based plant metabolomics is frequently used to identify novel naturally occurring molecules or study the effect of specific treatments on a plant's metabolism. Reliable sample handling is required to avoid artefacts, which is why most protocols mandate immediate shock freezing of plant tissue in liquid nitrogen and an uninterrupted cooling chain to preserve labile molecules. However, the logistical challenges of acquiring liquid nitrogen and establishing an uninterrupted cooling chain make this approach infeasible for some studies. Especially for research focussing on tropical ecosystems, permanent cooling poses a challenge, which is why many of those studies use dried leaf tissue instead. While this approach works for stable molecules, the drying process has a significant impact on the total metabolite profile.

Motivated by the need to profile metabolomes of crops in tropical mixed cropping systems, we screened ten extraction and storage approaches for plant metabolites retrieved from maize leaf tissue across two cropping seasons to find a method which can be used for studies under logistically challenging conditions and on an ecological scale. All methods were evaluated based on changes in the metabolite profile across a two-month storage period at different temperatures. The goal was to reproduce the metabolite profile of shock-frozen leaf tissue as closely as possible. We show that our on-site liquid-liquid extraction protocol provides a good compromise between sample replicability, extraction efficiency, material logistics, and metabolite profile stability.

Our on-site sample preparation was then used to study neighbourhood effects in a push-pull intercropping system. The samples were collected from farmer fields in Kenya, Rwanda, Ethiopia, and

Uganda, and extracted on-site before shipment to Europe for mass spectrometry measurements. Our approach could differentiate maize plants grown under push-pull systems from plants grown with conventional agricultural practices. Molecular identification is currently ongoing.

We demonstrated the feasibility of using an on-site liquid-liquid extraction protocol for plant metabolites from maize leaf tissue. We conclude that our method provides a reliable alternative for logistically challenging conditions regarding sample quality and stability as well as ecological scalability. This protocol allowed us to identify differences in the metabolite profiles of maize plants grown under different agricultural practices, highlighting the potential of this method for future agroecological studies.

M. Macel, et al., Mol. Ecol. Resour. 10, (2010), 583–593, DOI: 10.1111/j.1755-0998.2010.02854.x
Q. Yu, et al., J. Agric. Food Chem. 67, (2019), 1537–1545, DOI: 10.1021/acs.jafc.8b05993

OP7-2-2 Preparation and application of low-cost adsorbents for the removal of antiretroviral drugs in wastewater

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Antiretroviral drugs (ARVDs) are extensively employed for the treatment of human immune virus to improve the quality of life and lifecycle longevity. However, the partial digestion of ARVDs in the human body results in their large amounts entering the wastewater treatment plants where they are incompletely removed. This leads to their continuous introduction to water sources which is a concern as a result of their possible alterations of the ecosystem. Also, the antiviral resistance may develop upon their continuous unintentional consumption resulting in health effects. Therefore, this work assessed the presence of selected ARVDs in water and explored the low-cost adsorbent for their removal from water. The characterization of the adsorbents showed the presence of functional groups on the surface of the adsorbents which are responsible for binding and removing the ARVDs. Also, they have been observed to have high surface area, pore diameter, and pore volume which resulted in the removal efficiency above 80%. The equilibrium behavior of the ARVDs adsorption was described better by the Langmuir isotherm. The pseudo-second-order model well predicted the kinetic behavior. The results indicated that the explored low-cost adsorbents are effective for the removal of the selected ARVDs in water, and thus can be of benefit especially to low and medium income countries like the African countries.

OP7-2-3 Antibiotics invading South African waters: Analytical perspectives from a developing country with limited laboratory infrastructure

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The contamination of water resources with emerging contaminants in South Africa remains a concerning matter. In recent years, large amounts of antibiotics have been detected in South African water resources. This intrigued South African analytical chemists with interests in the environmental

issues. Thus far, analytical approaches are being developed taking into account the limited availability of the most suitable and sensitive analytical instruments such as liquid chromatography-mass spectroscopic tools. Notably, our research group focused on the synthesis of nanocomposite materials which can be used in the solid-phase extraction (SPE) of selected antibiotics prior to analysis with liquid chromatography-diode array detector. When applied for the analysis of tetracycline in wastewater, this analytical approach yielded the limits of detection and quantitation of 0.21 and 0.63 µg/L, respectively. Tetracycline had the highest concentration of 2.92 µg/L in wastewater, while the nanocomposite was proved to be a reusable material during the extraction of environmental samples. In anticipation of the expanding laboratories, we also applied a different approach utilizing the SPE and ultra-high-performance liquid chromatography-quadrupole time-of-flight-mass spectrometry analysis for the screening of 52 antibiotics in a stream receiving effluents and leachates from the dumpsite. In this study, 15 antibiotics were detected with concentrations of sulfamethizole, sulfamethazine, sulfamethoxazole ranging from not detected to 0.133 µg/L, flumequine ranged from 0.222 to 0.686 µg/L, while trimethoprim was up to 0.0618 µg/L. Overall, in addition to the environmental risk assessment which has been conducted, these research strides mean South Africa is advancing its research activities to fully understand the extent of water pollution caused by the antibiotics and their impact on the environment.

OP7-2-4 Ultrasensitive pH Sensing in Natural Waters towards in situ Measurements

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The release of carbon dioxide (CO2) in the atmosphere by the industries results in CO2 dissolution in the marine environment, which leads to ocean acidification. As this process occurs only gradually, established pH glass electrodes do not have sufficient precision (0.02 pH) to monitor these small changes. It is therefore necessary to develop ultrasensitive pH measurement techniques.

Ion-selective electrodes (ISE) are used for ion measurement in complex media. They are typically operated at zero current but their precision suffers from background drift. Bobacka and coworkers put forward a new readout method for solid contact ISE called “constant potential coulometry” which uses the capacitive properties of the ion-to-electron transducing layer [1]. A constant potential is applied between the reference electrode (RE) and the working electrode (WE). Whenever the sample activity changes, the phase boundary potential at the ISE is altered, resulting in an opposite side potential change at the capacitive layer and in a transient current. As these layers tend to deviate from ideality, our group replaced the capacitive layer by an electronic capacitor, achieving automated electronic control and high precision ion measurements [2].

Flowing current across the ISM was recently demonstrated to induce polarization and signal drift with current pulses. This process remains a significant drawback to achieve high precision. A new approach was put forward by using the ISE as RE and a SC-ISE with a dummy membrane as WE, avoiding the polarization at the ISE [3]. However, this approach suffers from the drawbacks of using conducting polymers that were discussed previously.

In this work, a silver/silver chloride element was used as WE and a pH electrode as RE. This allows one to avoid current flow through the membrane when doing coulometry and to drastically improve precision. As this approach required a constant chloride activity in the sample, the setup was not yet

ideal for freshwater analysis. The principle was therefore implemented in a dedicated flow cell to separate the silver chloride electrode from the sample to allow the determination of pH in undiluted natural water samples. This approach will later be implemented in a submersible probe and will allow the direct in situ determination of small pH variation enabling to understand the influence of these variations on the biogeochemical cycles of the elements.

[1] E. Hupa, et al., Electroanalysis 27 (3) (2015) 591–594.
DOI: 10.1002/elan.201400596.
[2] P. Kraikaew, et al., Sens. Actuators B Chem. 344 (2021) 130282.
DOI: 10.1016/j.snb.2021.130282.
[3] N. Delmo, et al., Membranes 12 (12) (2022) 1221.
DOI: 10.3390/membranes12121221.

OP7-3-1 FABRICATION OF ELECTROCHEMICAL PAPER-BASED DEVICES BY PROGRAMMABLE DRAWING

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Although the first emergence of patterned paper in the field of analytical chemistry was in the 1940s, the birth of the modern field of paper-based analytical devices (PADs) was marked by the pioneering work by the Whitesides group which demonstrated that it was possible to perform complex manipulation of liquids on hydrophilic paper channels using hydrophobic barriers [1]. The key features of paper as a substrate for the fabrication of PADs are: a) flexibility; b) low thickness and lightness; c) absorbency; d) high surface-to-volume ratio; e) hydrophilicity and capillary action; f) chemical and biological inertness; g) disposability and biodegradability, and; h) low cost and wide worldwide availability. Combined with low-cost and portable instrumentation, electrochemical PADs (ePADs) are well suited to on-site assays and point-of-care testing and relevant applications have been developed in various fields such as clinical diagnostics, environmental monitoring and food quality control [2-4]. The patterning of the hydrophobic barriers and electrodes is a critical step in the fabrication of ePADs and several approaches have been proposed in the literature [4]. Drawing strategies involve the use of a suitable pen or pencil to deposit functional materials on paper substrates with the view to create either hydrophobic patterns or conductive areas (electrodes) on paper [5]. Compared to the traditional methods for fabricating ePADs, the advantages of PoP approaches include simplicity, low cost, scope for rapid prototyping, flexibility in the design and wide selection of the functional materials to be deposited. The aim of this work was the development of an extremely simple and fast programmable PoP approach for the fabrication of ePADs. For this purpose, PADs were initially fabricated by drawing hydrophobic barriers via x-y plotting with commercial water-repellent marker pens. Electrodes were formed by further depositing electrodes on the PADs via x-y plotting using commercial writing pencils or conductive inks formulated in-house. The type of the paper substrate and the type of the marker pen were studied and different fabrication parameters were optimized. Finally, proof-of-principle applicability of the fully PoP-drawn PADs was demonstrated for electrochemical detection of organic and inorganic analytes.

[1] A.W. Martinez, et al., Whitesides, Angew. Chem. Int. Ed. 46 (2007) 1318. DOI: 10.1002/ange.200603817
[2] E. Noviana, et al., Lab Chip 20 (2020) 9. DOI: 10.1039/c9lc00903e
[3] V.N. Ataide, et al., Anal. Methods 12 (2020) 1030. DOI: 10.1039/c9ay02350j
[4] W. Mazurkiewicz, et al., 7 (2020) 2939. DOI: 10.1002/celc.202000512
[5] H. Lim, et al., Molecules 24 (2019) 2869. DOI: 10.3390/molecules24162869
[6] Z. Li, et al., TrAC 108 (2018) 50. DOI: 10.1016/j.trac.2018.08.010

OP7-3-2 Development of a screening method for total sulfonamides in environmental waters using pipette tip solid-phase extraction with smartphone-based fluorimetric detection.

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Recent significant advances in the miniaturization of spectroscopic analytical instruments have enabled the development of screening methods of contaminants of emerging concern (CECs) in water samples [1], which are rapid, low-cost, portable, easy-to-use analytical techniques before using more powerful and expensive reference analytical methods [2]. A sensitive, miniaturized, and low-cost method combining pipette tip solid-phase microextraction and smartphone-based fluorescent detection has been developed for determination of total sulfonamides in water samples. Sulfonamides antibiotics (SAs) are contaminants commonly found in water matrices, leading to antibiotic-resistant bacteria and risks to human health and the environment. Among SAs detected, sulfamethoxazole (SMX) is the most common found in environmental samples analyzed and has also been included in the 3rd Watch List of substances recommended for monitoring in the European Union in the Framework Directive of the water [4]. For this reason, its real-time monitoring is essential in WWTP effluents for subsequent risk assessment. Sample preparation consisted of preconcentration of SAs using graphene nanoplatelets packed inside a pipette tip, followed by fluorescent derivatization using fluorescamine inside the microplate reader, both 3D printed. Subsequently, a 3D-printed detection platform that houses monochromatic LED strips as radiation source and a smartphone as detector have been used for determination total SAs. Digital image processing was based on the RGB color model using ImageJ software with its readplate plugin and the green intensity channel was used as analytical signal due to its higher sensitivity. Several factors that affect the extraction efficiency and the detection system used have been optimized. Under the optimized conditions, good linearity for SAs studied (SMX, SDX, SMR and SMZ) were obtained in a range of 10-60 µg L-1 with R2≥0.990 and limits of detection between 2.5-3.1 µg L-1 for a sample volume of 10 mL. The recoveries of SMX (as a model compound to express total SAs) spiked in the samples tested at two different levels showed good recoveries from 94% to 102% with RSD ≤ 7.6 % and the results obtained with proposed system were compared with a conventional spectrofluorometer without showing significant differences (p≥0.13).
Keywords: Total sulfonamides, pipette tip solid-phase extraction, graphene nanoplatelets and 3D detection platform.

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[1] R. S. Lamarca, et al., Microchem.J.152(2020) 104297. DOI: 10.1016/j.microc.2019.104297
[2] F. Pena-Pereira, et al., Anal. Chim. Acta. 1158 (2021). DOI: 10.1016/j.aca.2020.11.040
[3] P.S. Peixoto, et al., Chemosensors. 10(2022) 25. DOI: 10.3390/chemosensors10010025/s1

OP7-3-3 Standard Addition for Immunoassays

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The standard addition method offers an advantage when analysing samples with a complex matrix. But this method is only applicable if the calibration curve is linear and intercepts the origin. Thus, it cannot be used for immunoassays which typically show sigmoid calibration curves in semi-log plots. For the use of lateral flow assays in several applications especially in therapeutic drug monitoring a quantitative test result is required and a method for internal calibration would be very useful.
To use the standard addition method for immunoassays the evaluation procedure needs to be adapted. An evaluation procedure for sigmoid calibration curves was suggested by Pang and Cowen [1], but simulations show the limitations of their evaluation procedure. It can only determine concentrations in a subrange of the calibration curve. We adapted the evaluation method to increase the working range of the standard addition method.

The logit function is used to transform the measured signals of the spiked sample aliquots. These are plotted against the logarithmised estimated total concentrations. The estimated total concentration is the sum of the known spike concentration and an estimated sample concentration. A linear regression is performed and as an indicator of linearity the residual sum of squares is calculated. After calculating the residual sum of squares for varied estimated sample concentrations, the sample concentration is found at the best linearity at the minimum of the residual sum of squares.

This method for evaluating signals after standard addition is verified in simulations. To show its applicability in real samples, an established immunoassay is used. A binding inhibition test for testosterone with the label-free method reflectometric interference spectroscopy is performed in buffer and milk samples [2]. Besides, the standard addition method allows quantitative measurements with lateral flow assays. To perform the standard addition directly on the test strip, standards are added to the sample using a structured conjugate pad. The spiked samples flow in separated laser-structured channels on the nitrocellulose membrane and give a decreasing signal according to the spiked analyte concentrations.

The standard addition method in the testosterone assay in buffer and milk samples gave a recovery rate between 70 and 120 %. This shows the applicability of the new evaluation procedure which will increase the field of application for immunoassays and especially lateral flow tests.

[1] S. Pang, S. Cowen (2017), Sci. Rep. 7, 17542, doi: 10.1038/s41598-017-17823-y.
[2] S. Rau, G. Gauglitz (2012), Anal. Bioanal. Chem. 402 (1), 529, doi: 10.1007/s00216-011-5470-9.

OP7-3-4 Effect of substrate porosity in the analysis of residues using Surface Enhanced Raman Spectroscopy (SERS)

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Agrochemicals are engineered to destroy insects, weeds, and fungi that could spoil crop yields. They are toxic by design, and at levels above MRLs (maximum residue limits), they pose risks to human health and the environment. (In order) To monitor and regulate the MRLs, rapid and routine detection of these molecules is essential. Surface Enhanced Raman Spectroscopy (SERS) is an analytical technique suited for field-based chemical analysis with molecular level detection capability. Paper-based SERS

substrates are often preferred for their surface roughness (3D structure), disposability, and use as swabs. In this presentation we will show that paper-based silver SERS substrates, prepared using the Print-Expose-Develop method invented by our group, result in a loss of signal owing to two primary reasons. Firstly, the interaction between the target molecules which are present across the paper depth, and the top nanostructure layer, is limited. This is attributed to the fact that not all molecules are readily chemisorbed on nanostructures, as evident from the calculated adsorption energy of ~32 KJ/mol for an adulterant dye. Secondly, the sub-micron depth of field in typical spectroscopic systems, the presence of nanostructures and the target molecules across the cross-section of the paper substrate (≃100 µm thick) is also not optimum. To overcome this, we prepared SERS active silver dendritic structures on non-porous metal foil via galvanic displacement. These substrates are easy to prepare and show good reproducibility with RSD ≃ 0.10. For the detection of malachite green dye (used as an antimicrobial in aquaculture), the most intense peak can be resolved at concentrations with molarity as low as 1 nM. Here, results from studies for the detection of thiabendazole (a fungicide), separation studies of mixture of dyes, and the signal collection efficiencies of porous and non-porous SERS substrates will be compared in the context of field-detection using a portable Raman Spectrometer.

S.M. Restaino, et al., Anal. Chim. Acta. 1060 (2019) 17.
DOI: 10.1016/j.aca.2018.11.057
S.A. Ogundare, et al., Cellulose. 26 (2019) 6489.
DOI: 10.1007/s10570-019-02580-0
P. Joshi, et al., RSC Adv. 6 (2016) 68545.
DOI: 10.1039/C6RA07280A
A. Kumar, et al., Anal. Chim. Act. 1090 (2019) 106.
DOI: 10.1016/j.aca.2019.08.073
W.A. Tegegne, et al., Microchem. J. 168 (2021) 106349.
DOI: 10.1016/j.microc.2021.106349
W. Li, et al., Sci. Rep. 3 (2013) 1.
DOI: 10.1038/srep01229

OP7-4-1 Rapid, automated Characterization of Microplastics and various other Samples from Materials to bio using Laser Direct Infrared Imaging and Spectroscopy

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Over the last 3.5 years, QCL technology has become increasingly important in IR microscopy. The advantages over FTIR are that large areas of a few square centimeters can be measured in minutes and that the light intensive QCL makes it possible to obtain spectra with excellent S/N even with just one scan. A meanwhile firmly established solution of the LDIR 8700 is the analysis of microplastics. The presence of microplastics in the environment, drinking water, and food chains is gaining significant public interest. To study their presence, rapid and reliable characterization of microplastic particles is essential. Significant technical hurdles in microplastics analysis stem from the sheer number of particles to be analyzed in each sample. Total particle counts of several thousand are not uncommon in environmental samples, while well treated bottled drinking water may contain relatively few.

While visual microscopy has been used extensively, it is prone to operator error and bias and is limited to particles larger than 300 µm. As a result, vibrational spectroscopic techniques such as Raman and FTIR microscopy have become more popular, however they are time-consuming. There is a demand for rapid and highly automated techniques to measure particle count, size, and provide high-quality polymer identification. Analysis directly on the filter that often forms the last stage in sample preparation is highly desirable as, by removing a sample preparation step, it can both improve laboratory efficiency and decease opportunities for error.
Recent advances in infrared micro-spectroscopy combining a Quantum Cascade Laser (QCL) with scanning optics has created a new paradigm, laser direct infrared imaging (LDIR). It offers improved speed of analysis as well as high levels of automation. Its mode of operation however requires an infrared (IR) reflective background, and this has, to date, limited the ability to perform direct "on-filter" analysis. This study explores the potential to combine the filter with an infrared reflective surface filter. By combining an IR reflective material or coating on a filter membrane, with advanced image analysis and detection algorithms, it is demonstrated that such filters can indeed be used in this way. Vibrational spectroscopic techniques play a vital role in the investigation and understanding of microplastics in the environment and food chain. While vibrational spectroscopy is widely deployed, improvements and novel innovations in these techniques that can increase the speed of analysis and ease of use can provide pathways to higher testing rates and hence improved understanding of the impacts of microplastics in the environment.

Due to its capability to measure large areas in minutes, its speed; degree of automation and excellent S/N the LDIR could also implemented for various other samples like food adulteration, coatings, laminates, fabrics, textiles and tissues. This presentation will highlight a few a few of them and focus on the benefits of the LDIR vs classical techniques.

OP7-4-1 New GC-MS source for solving the current Helium shortage

Andreas Kerstan

Gas chromatograph/mass spectrometer (GC/MS) instruments are used for a wide variety of critical analyses in areas such as food safety, environmental, forensics, and petrochemicals. Historically, helium is the preferred carrier gas for GC/MS. Helium is an inert gas with favorable chromatographic characteristics for high resolution separations that minimize unwanted reactions of analytes in the chromatographic process. Helium is also optimal for use with electron impact (EI) MS in terms of sensitivity. The vast majority of spectra in reference libraries such as NIST are acquired with helium carrier gas. In recent years, recurring difficulties with the availability and price of helium have resulted in users of GC/MS considering changing to alternative carrier gases. In GC without MS, several different carrier gases have been used successfully. Hydrogen, nitrogen, and argon (with or without methane) have all been used. However, for EI GC/MS, the only practical alternative is hydrogen. Hydrogen has superior chromatographic properties in terms of speed of analysis compared with helium but can have some limitations due to its reactivity. Hydrogen is also compatible with EI MS in particular with the recently developed HydroInert source. The HydroInert source addresses in-source reaction problems; for the metal source parts, it uses a proprietary material that greatly reduces catalytic activity and tailing. The HydroInert source is installed in place of the inert plus (extractor) source using all the same connections. This makes it possible to be used in the very same instrument as the conventional extractor source. In this presentation it will be shown how the method parameter must be adapted. The effects of the source are demonstrated and explained by some examples.

OP7-4-2 Advanced MS and NMR technologies for deep insights into plant-based food

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DSM is a company active in Health, Nutrition and Bioscience. For the development and biotechnological production of ingredients in food such as enzymes, cultures, hydrocolloids, plant-based proteins, yeast extracts, analytical science is an indispensable driver to provide insights. For supporting the protein transition towards more sustainable food production, development of novel ingredients requires deep molecular and physical-chemical insights into the food matrix to create plant-based foods while maintaining good taste, texture and health.

In this presentation, we will show how we develop our advanced MS and NMR toolbox to characterize ingredients and their interactions in food (related) matrices to steer the food properties.

Texturized Vegetable Proteins (TVPs) are used in meat alternative products and prior to application they are hydrated. We developed a Time Domain NMR method including robust python data-fitting to follow and optimize hydration times of these TVPs and correlated those to their composition (i.e. type of protein source) and processing conditions. To assure the quality of our plant and/or fermentative proteins during product development and shelf-life, characterization the quality and stability of the protein 3D structure is very relevant. Making use of methyl groups (CH3) as sensitive reporters for overall protein structural quality, we implemented natural abundance methyl 1H-13C HSQC NMR fingerprinting [1] of some of our protein products. Comparison to standards allowed us to determine the weight % of well-folded protein.

Furthermore, we have set-up and applied (un)targeted metabolomics workflows using SPME-GC-MS, LC-MS/MS and NMR including sample prep to map flavors from flavor systems, meat- and plant-based burgers to identify off-flavors and find leads that can close the flavor gap.

In conclusion, the protein transition brings complex analytical challenges to create deep insights to improve plant-based food.

[1] L.W. Arbogast, et al., J.Biomol. NMR 72 (2018) 149
DOI: https://doi.org/10.1007/s10858-018-0214-1

OP7-4-3 Direct Phospholipid Speciation of Lipid Feedstock Using A New THF-Based HILIC-ICPMS Approach.

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Phospholipids (PLs) are essential biomolecules in various sample sources due to their structural function in cell membrane walls. In the food industry, PLs are considered valuable supplements. However, in the energy industry, their presence in lipid biodiesel feedstock, such as waste fats and non-edible vegetable oils, can negatively impact production processes, including hydrodeoxygenation (HDO).[1] Consequently, developing a technique that enables direct speciation of PLs in these samples is crucial. Existing literature highlights a common procedure for PL analysis involving pretreatment with solid-phase extraction (SPE), followed by liquid chromatography separation and universal detection systems like evaporative light scattering detectors (ELSD) or mass spectrometry (MS).[2] PLs have a particular chemical structure, with a non-polar tail component and a polar head containing phosphate groups that can be attached to different molecules. These varying molecules give rise to distinct PL families, such as PG, PI, PE, etc. Considering the goal of separating PLs by family, hydrophilic interaction liquid chromatography (HILIC) is a promising alternative. Specific phosphorus detection can be conducted using elemental systems like inductively coupled plasma mass spectrometry (ICP-MS), where other lipids, such as triglycerides, are not observed.[2] This study proposes PL speciation in lipid samples without any pretreatment step, streamlining the process by dissolving the sample in tetrahydrofuran (THF) and analyzing it using HILIC ICP-MS with a THF-water mobile phase system.

In this study, a Dionex HPLC system coupled with a BEH HILIC 100A, 2.5 µm, 2.1 mm x 50 mm column was hyphenated to a Thermo Scientific Element XR double-focusing sector field ICP-MS. The mobile phase comprised a gradient of phase A (H_2O/THF 95/5) and phase B (THF), using stabilized HPLC-grade THF with MilliQ water, along with a buffer of 35 mM ammonium formate and 0.1% formic acid. Oxygen gas was mixed with the argon carrier gas to prevent carbon formation in the platinum cone and skimmer of the ICP-MS. Pure phospholipid standards and Asolectin (soybean extract) were purchased from Sigma-Aldrich. The quantification was performed using CONOSTAN oily-based phosphorus 5,000 ppm standard. The analyzed samples consisted of non-raffinate vegetable and animal oils.

The proposed method allows direct analysis of phospholipids in the samples without pretreatment. Quantification is possible more efficiently and without the need to use phospholipid standards but using the CONOSTAN oil standard. In conclusion, this method enables fast, economical, and reproducible analysis of phospholipids present in a lipid matrix.

(1) Verdier, S.; Alkilde, O. F.; Chopra, R.; Gabrielsen, J.; Grubb, M. Hydroprocessing of Renewable Feedstocks - Challenges and Solutions (v.1). Topsoe white Pap. 2018.
(2) Vosse, C.; Hayen, H.; Thyssen, G. M.; Sperling, M.; Karst, U. Complementary Approach for Analysis of Phospholipids by Liquid Chromatography Hyphenated to Elemental and Molecular Mass Spectrometry. 2020, No. December 2019, 46–55. https://doi.org/10.1002/ansa.20190009.

OP7-4-4 Thermal decomposition of lithium-ion-battery electrolyte and the influence on the cell performance

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The Green Deal defines the climate goals for 2050, an essential part is the decision that only emission-free vehicles will be registered in the EU from 2035. More electrically powered vehicles are being built each year, which use a lithium-ion battery as an energy storage system. One factor that is becoming

more important is the battery lifetime which is closely linked to the composition and stability of the electrolyte in the battery cell. During the charging and discharging of a battery cell, side reactions in the electrolyte occur, resulting in electrolyte degradation products. These reactions can reduce the cell performance as well as the lifetime of the cell.[1] The aim of this research project is to gain more insights into the degradation processes of the electrolyte and draw correlation to the cell performance. For this study different electrolytes are aged in a drying chamber to investigate the effect of the solvent components in the thermal decomposition. The specific conductivity of the electrolyte is being analyzed and the impact of the thermal decomposition products on the cell performance analyzed. Furthermore the impact of the vialmaterial in which the electrolyte is thermaly decomposed is analyzed.[2]

By means of various chemical analysis methods decomposition products are analyzed in the electrolytes. The conductive salt content in the electrolyte is determined by ion chromatography mass spectrometry (IC-MS) and analyzed for changes as the electrolyte ages.[3] Electrolyte solvents are analyzed using headspace gas chromatography-mass spectrometry (HS-GC-MS). The thermal decomposition process leads to the transesterification of the solvent molecules which can be detected by HS-GC-MS. [1] High-performance liquid chromatography coupled to Orbitap-MS (HPLC-Orbitrap-MS) is used to identify traces of degradation products formed by reactions of the conductive salt with the solvent.

References
[1] K. Xu, Chem. Rev., 2004, 104, 4303 – 4417. DOI: 10.1021/cr030203g
[2] M. Winter and S. Nowak, RSC Adv., 2015, 5, 80150 – 80157. DOI: 10.1039/C5RA16679A
[3] M. Winter and S. Nowak, J. Chromatogr. A, 2015, 1409, 201 – 209. DOI: 10.1016/j.chroma.2015.07.054

OP8-1-1 The influence of the surface pretreatment of a boron-doped diamond electrode on the determination of selected pesticides

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The use of the boron-doped diamond electrode (BDDE) for sensing purposes is widely recognized due to its extraordinary electrochemical properties including a wide potential window, a low background current, reduced surface fouling properties, and the ability to withstand extremely high cathodic or anodic potentials [1,2]. Unique properties of BDDE arise from the sp^3 nature of its surface, however, obtaining a pure sp^3 carbon electrode material during diamond synthesis without the existence of sp^2 carbon impurities is almost impossible [3]. The fouling of the BDDE surface, although reduced on the BDDE surface when compared to sp^2 carbon-based electrodes, is connected with the presence of sp^2 carbon impurities in the synthesized BDD material. Up to now, various pretreatment strategies have been developed to eliminate the fouling of the BDDE surface, restore the activity of the aged BDDE surface, enhance the voltammetric signals, and ensure repeatable responses of the analytes [4]. The most frequently used strategy is the electrochemical pretreatment of the BDDE surface, i.e., cathodic pretreatment (CPT) or anodic pretreatment (APT) by applying highly negative or highly positive potentials, respectively, in acidic solutions for a few seconds to minutes [2]. The other strategy is based on the pretreatment by polishing (PPT) of the BDDE surface [2], similarly as for other carbon-based electrodes.

Within this study, special attention was given to the effect of the BDDE surface pretreatment on the voltammetric response of fungicide fenhexamid (FH) and plant growth regulator forchlorfenuron (CPPU). For the sake of comparison, a PPT of the BDDE surface on a polishing pad using alumina (0.05 µm) between each recorded scan and anodic and cathodic pretreatments of the BDDE surface by applying potentials of +2.4 V and −2.4 V, respectively, for 900 s with simultaneous stirring of a 0.1 mol L–1 H2SO4 solution at the beginning of each working day, were tested. In situ APT of the BDDE surface was found to be the most favorable strategy to remediate the passivated BDDE surface by the oxidation (by)product of FH [5] and CPPU. The best validation parameters towards FH and CPPU sensing were achieved on the APT–BDDE. In addition, the FH and CPPU determinations on the APT–BDDE can be conducted without the necessity of time-consuming manual renewal of the electrode surface between individual scans as in the case of sp2 carbon-based electrodes [6].

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[1] N. Yang, S. Yu, J.V. Macpherson, Y. Einaga, H. Zhao, G. Zhao, G.M. Swain, X. Jiang, Chemical Society Reviews 48 (2021) 157-204.
[2] S. Baluchova, A. Danhel, H. Dejmkova, V. Ostatna, M. Fojta, K. Schwarzova-Peckova, Analytica Chimica Acta 1077 (2019) 30-66.
[3] O. Sarakhman, Ľ. Švorc, Crit. Rev. Anal. Chem. 52 (2022) 791–813.
[4] K. Muzyka, J. Sun, T.H. Fereja, Y. Lan, W. Zhang, G. Xu, Anal. Methods. 11 (2019) 397–414.
[5] M. Brycht, S. Skrzypek, V. Mirceski, Analytica Chimica Acta 1249 (2023) 340936.
[6] M. Brycht, A. Łukawska, M. Frühbauerová, K. Pravcová, R. Metelka, S. Skrzypek, M. Sýs, Food Chemistry 338 (2021) 127975.

OP8-1-2 Paper-based electrochemical biosensors for the detection of circulating miRNA signature: a tool towards decentralized management of Lung Cancer

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In the era of liquid biopsy, electrochemical biosensors can offer highly performing, user-friendly, cost-affordable, and potentially scalable solutions for portable diagnostics of cancer biomarkers, ranging from exosomes down to circulating micro-RNA (miRNA). The interest towards miRNAs as novel diagnostic and prognostic biomarkers is growing, and highly sensitive detection of miRNAs can be achieved with various methods (nucleic acid amplification-based or fluorescence imaging methods, sensing strategies) [1]. However, the complexity of biofluids, the time-consuming sampling/treatments and the equipment are limiting the development of devices for cancer management. Among the existing biosensing architectures, electrochemical paper-based sensing platforms are showing promising results in the detection of miRNA signatures in complex biological matrices [2].

Herein, three miRNAs sequences diagnostic for immunotherapy efficacy in advanced stages (III and IV) of non-small-cell lung cancer (NSCLC) were considered according to recent clinical trials [3]. These target sequences, namely miR-2115-3p, miR-224-5p, miR-6503-5p, were determined with an aptasensor based on anti-miRNAs ssDNA sequences labelled with a redox or electrochemiluminescent mediator, the methylene blue or tris(2,2'-bipyridyl)ruthenium(II), respectively. First, miRNAs levels were monitored following the decrease or increase of the redox mediator electrochemical signal, as previously described [4] in spiked plasma samples. These preliminary data allowed verifying the possibility of selectively determine the sequences of interest within a disposable assay.

Afterwards, a fully portable, printed electrode arrays were developed applying different analytical techniques, from square wave voltammetry (SWV) to electrochemiluminescence (ECL). Both electrochemical techniques provided an enhancement of the sensing platform sensitivity with limit of detection for the targets in low nanomolar range. This performance was achieved after optimising the probe density, the target-probe hybridization time as well as other analytical parameters using a one-variable at a time approach. During the study the electrodes arrays were printed on different supports (polyester, paper, etc.) comparing the final multiplex sensors in terms of reproducibility, reliability, and sustainability according to White Analytical Chemistry principles [5].
The detection of these three miRNA sequences is the starting point for the detection of the full miRNA signature [3]. With such platforms, both voltametric and ECL-based ones, we aim at contributing to a more effective overall management of NSCLC immunotherapies. This biosensor array represents the first integrated prototype to be applied on different cohorts of NSCLC patients, opening to opportunities towards other type of cancer prognosis based on miRNA signature.

[1] Cacheux J., Front. Chem. 2019, 7, 815. https://doi.org/10.3389/fchem.2019.00815
[2] Cinti S., Nat Protoc. 2019, 14, 2437. https://doi.org/10.1038/s41596-019-0186-y
[3] Rajakumar T., npj Precis. Onc. 2022, 6, 19. https://doi.org/10.1038/s41698-022-00262-y
[4] Miglione A. et al., Anal. Chem. 2022, 94, 45, 15558. https://doi.org/10.1021/acs.analchem.2c03542
[5] Nowak P.M. et al., TrAC. 2021, 138, 116223.
https://doi.org/10.1016/j.trac.2021.116223

OP8-2-1 Low-cost and miniaturised determination of atmospheric gaseous elemental mercury by passive sampling and voltammetric detection on screen-printed gold electrodes

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Atmospheric mercury is a significant global threat to human health and ecosystems, so there is a strong demand for rapid and decentralized methods for measuring mercury species levels in ambient air. GEM monitoring is a challenging analytical task considering that typical levels of gaseous elemental mercury (GEM) in ambient air are 5 - 10 ng m-3. Passive or active sampling of GEM on gold surfaces is a consolidated technique combined with different detection techniques but has not been coupled with voltammetry on printed electrodes. In this work, we have developed an original methodology based on passive sampling of Hg on screen-printed gold electrodes (SPGEs), followed by determining amalgamated mercury by square wave anodic stripping voltammetry (SWASV) on a portable potentiostat. After the generation of standard GEM concentrations employing a homemade "bell-jar" device, we explored in detail the behaviour of the surface of the electrode SPGE during the sampling process by TOF-SIMS, the stability of the voltammetric signals and the reproducibility between electrodes, with acceptable results. The linearity of mercury adsorption onto the SPGE was evaluated for different mercury concentrations, selecting a sampling time of 30 minutes as optimal. The applicability of two calibration protocols has been verified. The first is based on measuring the area of the stripping voltammetric peak with the following optimized parameters: anodic sweep (potential sweep from 0.1 to 0.65 V, 6 mV step potential, 40 mV amplitude, and 10 Hz frequency). The second is measuring the mercury concentration by standard additions, using the following parameters: 20 s of conditioning time at 0.7 V, 60 s of deposition time at −0.1 V with 300 rpm stirring rate, 10 s of equilibrium time at −0.1 V, potential sweep ranging from 0.1 to 0.65 V, step potential of 6 mV,

amplitude of 40 mV, and frequency of 10 Hz. Suitable calibration parameters were found for both procedures, with detection limits of 5.32 and 5.22 ng dm-3, respectively. These detection limits restrict the applicability of the method in its present stage to air highly contaminated by GEM, but our results open a new line of low-cost electroanalytical strategies for determining gaseous elemental mercury in atmospheric samples.

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OP8-2-2 Factors controlling the mercury entry and bottom-up transfer in aquatic trophic webs

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Mercury (Hg) is regarded as one of the most toxic elements impacting on human and ecosystem health. It is a naturally occurring element, however, since the industrial revolution anthropogenic Hg inputs exceed natural inputs by at least a factor of five. Humans are exposed to Hg mainly through their diet, especially through the consumption of fish and other products from the aquatic environment. Phytoplankton are at the base of the aquatic food webs and serve as an entry point of Hg into organisms of higher tropic levels.

In waters, Hg is commonly found to be complexed with ligands: dissolved organic matter and inorganic nanoparticles which formed the natural nano-colloidal pool (NNC). The functional groups and size of the complexes, may drive Hg bioavailability for phytoplankton. Previous studies have shown that surface-complexes formed by Hg with nanoparticles (> 100 nm) and stable nanoparticles made of Hg sulfide (10 nm), were bioavailable for the microorganisms despite their large sizes. It is, therefore, important to study in more detail the interactions between Hg bioavailability and different sizes of inorganic and organic colloids.

In this study we present the first step towards the development of a methodology able to determine Hg size-distribution, and more specifically to characterize the abundances of components present in the NNC pool and the preferential association of Hg within the NNCs components in ambient water. To this end we used a centrifugal-ultrafiltration technique (for the determination of Hg-bound to NNC pool and to concentrate NNCs) combined to asymmetrical-flow fractionation link to multi-detectors. The latter provides an in-depth characterization of the NNC components in terms of size and nature (organic/inorganic) and can be used either on-line with ICP-MS or off-line with CV-AASF measurements to determine the Hg associated to each NNC components individually. Our first results obtained from Seine River (Paris, FR) for which Hg loading was relatively low (1-10 pM range) will be critically evaluated considering (i) the artefacts cause by filtration process (e.g., membrane fouling/rejection)

which can occur for both techniques, (ii) limit of detection for each elemental analyser used is also discussed.

By optimizing this methodology for water settings with contrasting chemistries (e.g. wetland, hydrothermal vents), and by determining both the content and subcellular distribution of Hg species in local phytoplankton, we expect to refine existing models with the aim to better predict the fate of Hg and other trace metals in aquatic food webs.

OP8-2-3 Improved target, suspect- and non-target analysis of environmental contaminants using a GC-EI&CI-TOF-MS system

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Exogenous exposures to environmental contaminants, such as from diet, lifestyle, or material or urban emissions, can have harmful effects for humans. Especially toxic substances are important to identify and monitor. These can be found in various environmental sources (e.g. water or air) and may not only affect human health but also quality of life or of manufacturing goods, for example. GC-MS is often employed to evaluate potential hazard profiles and to identify compounds, but difficulties arise due to the complexity of these samples. However, conventional EI-MS is limited in its identification confidence for some compounds due to unspecific fragmentation, absent molecular ion signals or by not being available in reference libraries, leading to a lack of compound identification.

The ecTOF (TOFWERK, Switzerland) is a newly developed dual ionization source time-of-flight (TOF) mass spectrometer [1]. The instrument consists of a standard TOF analyzer operating two ionization sources quasi-simultaneously: a standard 70 eV EI source and a medium pressure CI source [2] (HRP; TOFWERK, Switzerland). The ecTOF is coupled to a GC using a Y-splitter and custom designed heated transfer line to connect to the two different ionization sources. Depending on the sample, liquid, gas phase or thermo-desorption is chosen as sample introduction method. For sample preparation, various preconcentration methods such as SPME, tenax tubes are applied.

This talk will highlight the advantages of the combined EI and CI mass spectra for contaminant identification of complex environmental samples. Samples analyzed can include river water taken from industrial sites, air from Swiss and German industrial sides as well as emissions from different building materials and car interior parts. The tentative identification processes and statistical approaches that can be employed using the ecTOF data will be shown. The reduced false positive rates using the quasi-simultaneously generated EI and CI mass spectra as well as the increased identification yields compared to standard GC-MS methods will be presented. Looking at different environmental samples shows the advantage of a flexible CI setup used for various compound classes, for example pesticides, CFCs, PCBs and other persistent organic pollutants. The simple selection of different CI reagents ions (e.g., [NH4]+, [N2H]+, [H3O]+) between chromatographic runs, which enables the adjustment of reactant selectivity and the degree of fragmentation, is proven to be highly valuable for the compound identification process. Various methods of using the generated EI- and CI- mass spectral information within library searches and other tools will be presented.

[1] Bräkling, S. et al. Parallel Operation of Electron Ionization and Chemical Ionization for GC-MS Using a Single TOF Mass Analyzer. Anal Chem., 2022, 94 (15), 6057-6064.
[2] Bräkling, S. et al. Hydrogen Plasma-Based Medium Pressure Chemical Ionization Source for GC-TOFMS. J. Am.

OP8-3-1 Novel RP-HPLC based assay for selective and sensitive endotoxin quantification

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Endotoxin (ET) testing in pharmaceuticals is a crucial requirement for patient safety. This paper presents a novel instrumental analytical ET quantification assay [1]. The Kdo-DMB-LC assay uses common analytical laboratory equipment ((U)HPLC-FLD) and allows the quantification of ETs in complex matrices from about 10e7 EU / mL down to about 30 EU / mL (RSE based). Test results are obtained in concentration units (e.g., ng ET / mL), which can then be converted to commonly used ET activity units (EU / mL). During mild acidic hydrolysis, the rare ET specific 3-deoxy-D-manno-oct-2-ulsonic sugar acid (KDO) is obtained quantitatively. After that, KDO is stoichiometrically reacted with DMB, which results in a highly fluorescent derivative. The mixture is separated using RP-(U)HPLC followed by KDO-DMB quantification by fluorescence detection. From the KDO content the ET content in a sample is calculated. The applicability of the Kdo-DMB-LC in applied research is demonstrated. ETs were quantified in partially purified bacterial biopolymers, which were produced by Gram-negative bacteria. Results were compared to LAL results of the same samples. A high correlation was found between the results of both methods. Further, the new assay was successfully utilized for the development of novel ET specific depth filters, which allow efficient, economic, and sustainable ET removal e.g., during DSP. In addition, the ET content was monitored in the supernatants of Escherichia coli K12 and Pseudomonas putida KT2440 cultivations from inoculation until harvest [2]. The Kdo-DMB-LC assay is an easy to install tool to optimize reactor settings with respect to the ET content in dependence on cultivation time and conditions, a task difficult to achieve using the common LAL assay. It is economic, has a small error and it has the potential to complement the animal-based biological LAL pyrogenic quantification tests, which are accepted today by the health authorities worldwide for the release of commercial pharmaceutical products. The new Kdo-DMB-LC assay brings ET testing to the 21st century.

[1] B. Bucsella, A. Hoffmann, M. Zollinger, F. Stephan, M.Pattky, R. Daumke, F. Heiligtag, B. Frank, M. Bassas-Galia, M. Zinn and F. Kalman, Analytical Methods, 12(38) (2020) 4621-4634.
[2] A. Hoffmann, K. Pacios, R. Mühlemann, R. Daumke, B. Frank and F. Kalman, in preparation.

OP8-3-2 Identification of wine markers in ancient pottery using liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS)

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Between the second BC and second AD centuries, the Roman empire spread throughout the Mediterranean area influencing the lives of all its people. Today much of this legacy can be found buried in archaeological sites and offers a great opportunity to study the lifestyle of these nations. Indeed, the characterization of the organic residues that ancient pottery contains will be of great help to ascertain the type of food they contained and, as a result, to understand those societies.

Wine was very appreciated in the Roman empire, thus it was produced in many regions and became a key product in the Mediterranean trade. Pottery that contained this beverage is often found in the Roman villas, however, as these kinds of vessels may not differ from others storage containers, reliable methods for its content confirmation are needed in order to broaden the knowledge of these villas.
In this work, a liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS) method was established for the determination of organic acids, mainly tartaric acid, and other compounds, which are known to be important components of wine, in fragments of dolia from Roman sites. However, tartaric acid, as well as other wine components, can be extensively found in the plant rhenium, and thus the presence of these compounds in the studied pottery fragments cannot be considered unequivocal evidence of their use. Appropriate blank samples, exposed to the same environmental factors, are then required to be able of drawing the correct conclusions. The strategy proposed here deals with using the outside part of the studied fragments for comparison purposes.

The optimized approach was then used for analyzing fragments of dolia from the Roman sites of Empúries, Collet de Calonge I Sant Antoni, Olivet d'en Pujol, and Vila de casa del Racó in the province of Girona. The analysis of the samples studied has shown that the fragments of dolia from Empúries, Olivet d'en Pujol and Vila de casa del Racó contain tartaric and malic acid in a significantly higher concentration on the interior side than on the exterior side, suggesting that they may have contained wine in the past. On the contrary, in the fragments of Collet de Calonge these indicators have not been found, so the possibility that they contained wine can be ruled out.

OP8-3-3 Towards Continuous Cytokine Monitoring in Organ-based Platforms

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Investigations on organ-based platforms such as organoids, organ-on-a-chip, and transplantation organs, require monitoring strategies in order to optimally control the biological systems. However, there is a lack of tools to continuously measure specific low-concentration biomarkers with minimal perturbation [1]. Biosensing by Particle Motion (BPM) is a sensing method with single-molecule resolution that has been specifically designed to enable the continuous monitoring of biomolecules at low concentrations, such as nucleic acids, metabolites, proteins and hormones [2]. The method relies on tracking the motion of individual biofunctionalized particles (1 µm in diameter) that interact with a biofunctionalized substrate. The particles switch between bound and unbound states due to reversible single-molecule interactions, influenced by the presence of analyte molecules. Recently, sampling by microdialysis has been investigated [3]. In this paper we present the development of a BPM sensor to measure cytokines, exemplified with the detection of Interleukin-6. We will present a study of molecular binders and coupling strategies, focusing on sensitivity and reversibility of the sensor.

Furthermore, measurements with microdialysis will be shown. Finally, we will discuss the prospects of using BPM and microdialysis for the continuous monitoring of low-concentration biomarkers in organ-based platforms.

[1] L. Mou et al. "Integrated Biosensors for Monitoring Microphysiological Systems", Lab on a Chip (2022). https://doi.org/10.1021/acssensors.2c00333
[2] A. Buskermolen, Y. Lin, L. van Smeden et al. "Continuous biomarker monitoring with single molecule resolution by measuring free particle motion", Nature Communications, 13(1) (2022). https://doi.org/10.1038/s41467-022-33487-3
[3] L. van Smeden et al. "Reversible Immunosensor for the Continuous Monitoring of Cortisol in Blood Plasma Sampled with Microdialysis", ACS Sensors (2022). https://doi.org/10.1021/acssensors.2c01358

OP8-4-1 Lean Approach to Analytical Procedure Development for Therapeutic Synthetic Peptides

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In line with the regulatory requirements as laid out e.g., in ICH Q14 and USP 1220, the goal of any analytical procedure development activity is to find procedure conditions that ensure accuracy and precision of the results generated with the procedure, i.e., the analytical procedure must meet the analytical target profile (ATP). Therapeutic synthetic peptides are typically complex molecules and their synthetic impurities/degradation products only differ marginally in physico-chemical properties, which renders the development of chromatographic methods for determination of purity, related impurities, and assay of those materials particularly challenging. This talk outlines the approach used at BACHEM to develop such analytical procedures for (U)HPLC in a resource-efficient manner, which are an integral part of CMC development as well as releases of drug substance for in-human use. To that end, the chromatographic methods must provide sufficient selectivity and sensitivity, must be stability indicating and robust and ideally should be MS-compatible. The talk details how previously gained experience is systematically incorporated through knowledge management and how risk assessments are applied to identify (potentially) critical procedure parameters early on. Procedure development at BACHEM entails screening of a wide range of stationary and mobile phases (in terms of selectivity), which is accelerated with the aid of automation. Design of Experiments (DoE) as well as other mathematical tools are used to optimize procedure parameters and understand their effect procedure performance. Further, the talk addresses how the robustness of analytical procedures is assessed and how DoE is used to consider multivariate interactions of procedure parameters and how ultimately a method operable design region (MODR) might be designed. In summary, through the presented procedure development concept, the generation of analytical procedures, which are fit for the intended purpose and can eventually be validated and applied successfully is ensured.

OP8-4-2 Simulation of Intraluminal Performance of Lipophilic Weak Bases in Fasted Healthy Adults Using DDDPlusTM

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The majority of drug candidates exhibit weakly basic characteristics with high lipophilicity. The risk of intraluminal compound precipitation has been studied in vivo and extensively in vitro using advanced dissolution transfer setups mimicking drug transfer from the stomach to the small intestine. The present investigation aims to evaluate the usefulness of the recently introduced Artificial Stomach-Duodenum in silico tool in the DDDPlus™ platform (ASD-D+) to simulate intraluminal drug behavior. The weakly basic drugs ketoconazole and dipyridamole were used as model drugs within the ASD-D+ model at two dose levels. The simulated amounts per volume were compared to intraluminal data collected from fasted healthy adults. Four different in silico transfer models running on a continuous or a stepwise mode were utilized for the simulations.

Each transfer model exhibited different capabilities to simulate observed intraluminal drug presence. Three out of the four in silico models overestimated the total drug amount measured in vivo (dissolved and precipitated drug), while only two of the four models matched the intraluminal drug concentrations. The stepwise model enabled adequate simulations of both drug concentration and total drug amount. The present investigation highlighted the importance of simulating drug transfer appropriately within the applied methodology prior to estimating precipitation kinetics. As a future step, optimization of ASD-D+ model would enable evaluations on the continuous simulations of solid/semi-solid dosage forms. Lastly, prediction of drug precipitation kinetics following simulation of gastrointestinal transfer may provide mechanistic understanding of drug absorption and appropriate justification of drug formulated parameters within physiologically-based pharmacokinetic models.

M. Statelova, et al., AAPS J. 24 (2022) 89.
https://doi.org/10.1208/s12248-022-00737-7

OP8-4-3 Selected Highlights in Analytical Chemistry at the ZHAW Wädenswil

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At the Institute of Chemistry and Biotechnology (Zurich University of Applied Sciences) in Wädenswil, we are actively engaged in various research and educational activities in the field of analytical chemistry. We see chemistry and biotechnology as converging fields that link discoveries in the natural sciences with technological knowledge. In this context, chemical and biological analysis plays a fundamental role in understanding and improving natural as well as industrial processes. In this brief overview, selected examples of research groups who apply advanced analytical tools to pursue their activities in applied research are presented.

In a project run by the group of Environmental Analytics, particles entering the environment from tyre wear are investigated. They contain additives, such as antioxidants or vulcanization accelerators, which could serve as marker substances for tyre abrasion. After extraction with accelerated solvent extraction, soil and tyre samples were analyzed with gas chromatography coupled to time-of-flight mass spectrometry (GC-QTOF). Mass fractions of these additives in different soil samples were in the µg·kg-1range, depending on how close from roads the samples were taken.

The early stage of development of biopharmaceuticals such as monoclonal therapeutic antibodies involves the screening of candidate molecules in terms of specificity, affinity, solubility and stability.

The group of Bioanalytics characterizes the candidate molecules by means of a developability assessment using a variety of orthogonal bioanalytical methods. Clients are small biotech companies as well as large international pharmaceutical companies with whom we develop mono- and bi-specific antibodies and antibody drug conjugates.

Research of the Coffee Excellence Center includes investigation of coffee along the complete value chain from the seed to the cup. To analyze the relevant volatile compounds in green coffee, roasted whole bean coffee, roast and ground coffee, and coffee brew, advanced analytical techniques such as proton-transfer-reaction mass spectrometry (PTR-ToF-MS) are used and optimized. In correlation with sensory data, these measurements fundamentally improve our understanding of aroma development in coffee extraction.

Different research groups are active in improving process analytical tools for real-time and inline monitoring and control in chemical and biological processes. These include spectroscopic techniques such as inline-Raman and near infrared spectroscopy, and methods related to particle size measurements. As an important but not yet well-exploited parameter in biotechnological processes, biomass concentration is measured using different methods. Models based on data from dielectric spectroscopy in correlation with flow cytometry measurements help to increase the reliability of biomass concentration and – ideally – cellular viability.

OP9-1-1 Fabrication of ZnO Nanoparticles Assisted Molecularly Imprinted Polymer-Based Electrochemical Sensor for the Selective Determination of Sorafenib

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Sorafenib tosylate (SOR) is an orally administered anticancer agent that shows its effect by inhibiting the vascular endothelial growth factor receptor (VEGFR), platelet-derived growth factor receptor, and several other pathways such as RAF-MEK-ERK pathways [1]. The United States Food and Drug Administration (FDA) approved the use of SOR in the treatment of hepatocellular carcinoma, renal cell carcinoma, and thyroid carcinoma [2]. When the literature is evaluated, there are a limited number of studies for the determination of SOR in biological samples, and they are all chromatographic methods [3,4]. This study offers a highly selective, sensitive, and stable analysis option for determining SOR using ZnO nanoparticles assisted molecularly imprinted polymer (MIP)-based electrochemical sensor. During the development of the sensor, the photopolymerization (PP) method was performed on the glassy carbon electrode (GCE) surface using 2-acrylamido-2-methylpropane sulfonic acid (AMPS) as the functional monomer, SOR as the template, 2-hydroxyethyl methacrylate (HEMA) as the basic monomer, ethylene glycol dimethacrylate (EGDMA) as the crosslinker, and 2-hydroxy-2-methylpropiophenone as the initiator. Additionally, ZnO nanoparticles were added to the PP mixture to improve sensor performance and sensitivity. Significant parameters related to MIP (nanoparticle amount, monomer ratio, dropping volume, PP time, removal solution and time, and rebinding time) were optimized. Morphological and electrochemical characterization of the ZnO/AMPS@MIP-GCE sensor was performed using scanning electron microscopy (SEM), cyclic voltammetry (CV), and

electrochemical impedance spectroscopy (EIS). Differential pulse voltammetry (DPV) was used for other electrochemical measurements, and the sensor gave a linear response in the concentration range between $1x10^{-12}$ M and $1x10^{-11}$ M, with the limit of detection (LOD) and quantification (LOQ) values of $2.37x10^{-13}$ M and $7.88x10^{-13}$ M, respectively. The applicability of the ZnO/AMPS@MIP-GCE sensor was tested using a commercial human serum sample and resulted in excellent recovery values (99.57% and 100.66%). Interference-free and selective performance of the sensor was confirmed using common interfering agents (Na+, K+, NO_3–, dopamine, ascorbic acid, etc.) and similarly structured compounds (regorafenib, leflunomide, axitinib, dasatinib, etc.). Finally, the sensor's performance was verified using a non-imprinted polymer-based GCE.

[1] I. Khan, et al., J. Chromatogr. B Anal. Technol. Biomed. Life Sci. 1033–1034 (2016) 261–270. DOI: 10.1016/j.jchromb.2016.08.029
[2] L. Jain, et al., J. Pharm. Biomed. Anal. 46 (2008) 362–367. DOI: 10.1016/j.jpba.2007.10.027
[3] B. Blanchet, et al., J. Pharm. Biomed. Anal. 49 (2009) 1109–1114. DOI: 10.1016/j.jpba.2009.02.008
[4] M. Zhao, et al., J. Chromatogr. B Anal. Technol. Biomed. Life Sci. 846 (2007) 1–7. DOI: 10.1016/j.jchromb.2006.06.005

OP9-1-2 Voltammetry and Amperometry of Biologically Active Organic Compounds - Where We Are Heading 100 Years After the Discovery of Polarography

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More than 100 years ago Prof. Jaroslav Heyrovsky published his paper on electrolysis on dropping mercury electrode [1] for which he later coined the term polarography and for which he received in 1959 Nobel Prize. This presentation will remind that polarography was the first analytical method with automatic registration of dependence of a signal on certain parameter enabling to extract both qualitative (based on position of a signal on registered curve) and quantitative information (based on height of a recorded signal) and paved the way for many approaches used nowadays in all instrumental analytical techniques (e.g. standard addition method). However, the main focus of the lecture will be on the search for novel electrode materials with lower noise, broader potential window and higher resistance to passivation [2]. Practical examples of novel electrode materials successfully used in our laboratory will include renewable surface amalgam electrodes [3], porous silver electrodes [4], carbon composite electrodes [5], chromatographic sorbent modified electrodes [6], polystyrene-based composite electrodes [7], etc. Advantages and disadvantages of different electrode materials and arrangements for both batch analysis and for measuring in flowing systems (HPLC, flow injection analysis, batch injection analysis etc. with electrochemical detection) will be critically compared together with further perspectives of modern electroanalytical methods for high throughput large scale monitoring of biologically active organic compounds in various environmental or biological matrices including their combinations with novel methods of preliminary separation and preconcentration using e.g. hollow fibres.

1. Heyrovský J.: Chem. Listy 16, 256 (1922)
2. Barek J.: How to Improve the Performance of Electrochemical Sensors via Minimization of Electrode Passivation. Chemosensors 9, 12 (2021).
3. Gajdar J., Stafurova K., Barek J., Fischer J.: Retractable Pen-based Renewable Silver Amalgam Film Electrode for Microliter Sample Analysis of Electrochemically Reducible Environmental Pollutants". Sensors and Actuators 329, 129057 (2021).

4. Bountas G., Vyskocil V., Kratochvil B., Michalcova A., Economou A., Barek J.>Differential pulse voltammetric determination of selected nitrophenols on novel type of porous silver working electrode prepared by powder metallurgy. electrode prepared by powder metallurgy. Electroanalysis 35, e202200437 (2023).
5. Dorozhko E., Kazachinskaia E., Kononova Y., Zaikovskaya A., Barek J., Korotkova. E., Kolobova E., Sheveleva P., Saqib M.: Electrochemical immunoassay of antibodies using freshly prepared and aged conjugates of silver nanoparticles. Talanta 253, 124028 (2023).
6. Solomonenko A., Dorozhko E., Barek J., Korotkova E., Vyskocil V., Shabalina A.: Adsorptive stripping voltammetric determination of carbofuran in herbs on chromatographic sorbent modified electrode. J. Electroanal. Chem. 900, 119692 (2021).
7. Jiranek J., Barek J.: The use of non-traditional carbon film electrode based on microcrystalline natural graphite-polystyrene composite film for amperometric determination of 5-aminoquinoline using flow injection analysis minimising electrode fouling. J. Electroanal. Chem. 885, 115085 (2021).

OP9-1-3 Electrochemical detection of enzymatic assay in microfluidic channels

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In recent years, the field of metabolomics has gained interest, as it can help in drug discovery and disease study. To support throughput, the use of microfluidics is very interesting to measure metabolites in small volumes as fast mixing, shorter reaction times and less reagent consumption can be achieved. The metabolites are optimally measured with enzymatic assays. Until now, most experiments have been performed with fluorescence read-out. The need for labelling and sensitivity to quenching and interferences are some important drawbacks of this approach. The use of an electrochemical read-out has the advantage of smaller and easier integration. Hence, we designed a new microfluidic device with integrated microelectrodes to enable electrical read out of these enzymatic assays.

As a case study, we looked at the enzymatic detection of glucose. This is done by measuring the concentration of hydrogen peroxide generated during the enzymatic conversion of glucose by glucose oxidase at the surface of platinum microelectrodes. To control the reaction time of the assay, the enzyme and the analyte are brought together on the microfluidic chip via two separate inlets and allowed to react along the microfluidic flow channel.

The effect of the reaction time was quantified by the electrodes integrated along the channel to enable spatially resolved measurement, and by varying the flow speed. A linear relationship between the current measured at the sensor surface and the distance along the reaction channel (i.e. reaction time) was observed, in agreement with the increasing concentration of hydrogen peroxide. The dependence on the flow speed (v) matched well with the combined effect of a reduced hydrogen peroxide concentration at the electrodes (1/v) and an increased convective mass transport of the peroxide to the electrodes (v^1/3), resulting in an overall v^-2/3 dependence. Furthermore, the dose-response curves correspond to the Michaelis-Menten kinetics. Lastly, as the enzyme concentration increased, the current increased until it was limited by the concentration of oxygen dissolved in the buffer.
These results show that the combination of electrochemical detection and microfluidics is very promising to perform enzymatic assays in the field of metabolomics.

OP9-2-1 Origami-paper devices for rapid diagnosis and wastewater surveillance

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Pathogen detection is of significant importance for both biomedical diagnostics (e.g., infectious disease) and environmental analysis, e.g., pathogen contamination in drinking water), SARS-CoV-2 in wastewater for early warning of pandemic. Here we present a low-cost, deployable paper-based biosensor device for rapid analysis of pathogens for a wide range of application. We will show the capability of paper-origami device for field-testing for veterinary diagnosis in India, and for malaria testing in a local primary school in Africa, as well as a recent clinical sample testing of Hepatitis C virus (HCV). This device is currently developing to trace the source of SARS-CoV-2 for wastewater-based epidemiology for early warning of pandemic, within a UK national wastewater epidemiology surveillance programme (N-WESP) for COVID-19, which was demonstrated for the field-testing in local quarantine hotel in London.

OP9-2-2 Study of variations in polymer inclusion membranes for antibiotic separation from milk

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As antibiotic resistant bacteria pose an increasing threat to general health, more efficient detection methods are in need, especially in foodstuffs. However, complex food samples usually require laborious sample preparations prior to analysis due to the interfering components, such as fat, protein, sugar etc. [1]. For liquid foodstuffs, polymer inclusion membranes (PIM) hold considerable promise as simple, yet effective separators of pharmaceuticals, like antibiotics.

PIMs are liquid membranes made up of a polymer matrix (usually cellulose acetate or polyvinyl chloride) that provides mechanical stability and include a liquid phase, which serves as a plasticizer. These membranes contain a carrier/extractant, which binds to the target analyte either by complexation or by ion-pair formation, and through this, the target is carried across the membrane facilitating its extraction from the sample [2]. PIMs are prepared by dissolving the polymer, the plasticizer and the carrier in a suitable solvent and cast onto a flat surface. Solvent evaporation leaves behind the thin polymer membrane.

In this work, we have developed variations of PIMs for the separation of a chosen antibiotic – oxytetracycline – from milk samples. The experiments were performed using a transport cell comprised of two compartments separated by the polymer membrane: the feed phase, containing the sample, and the stripping phase with ions necessary for the extraction. Transport of oxytetracycline through the PIMs was monitored in time by determining the oxytetracycline concentration in the feed and stripping phase by a newly established and validated HPLC method. The membranes were optimised regarding thickness, solvent type, carrier, plasticizer and base polymer content. The optimal pH and ionic composition of the stripping phase was also determined. Performance of the PIMs was

characterized by their initial flux values [3]. Different types of spiked milk samples were pre-treated this way before HPLC analysis. The recovery and efficiency of the PIM-based sample clean-up process were compared with that of the traditional solid phase extraction procedure.

The use of PIMs in food sample pre-treatment holds the promise of easy separation of the antibiotics from interfering matrix components, making it a useful preparatory method in bioanalytical sensor devices. With the help of such membranes target pre-concentration is also feasible, which lowers the achievable limit of detection.

[1] M. Pérez-Rodríguez, R.G. Pellerano, L. Pezza, H.R. Pezza, Talanta. 182 (2018) 1–21. DOI: 10.1016/j.talanta.2018.01.058
[2] M.I.G.S. Almeida, R.W. Cattrall, S.D. Kolev, Anal. Chim. Acta. 987 (2017) 1–14. DOI: 10.1016/j.aca.2017.07.032
[3] A.M. St John, R.W. Cattrall, S.D. Kolev, Sep. Purif. Technol. 116 (2013) 41–45. DOI: 10.1016/j.seppur.2013.05.021

OP9-2-3 Comprehensive Investigation of different Coatings and Adsorbents for SPME and their Influence on Analytical Performance

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Since its introduction in 1989 [1], Solid Phase Microextraction (SPME) has gained a broad popularity in environmental and food analysis as well as many other application areas. SPME is a fast, sensitive, easy-to-automate and solvent-free microextraction technique which can be applied for various analytes in gaseous, liquid, or solid samples.

The stationary phases in SPME use either a polymer or a solid porous adsorbent embedded in a polymer. The various adsorbents have different ratios of macropores (diameters > 500 Å), mesopores (21-500 Å) and micropores (1-20 Å) which significantly impact their extraction capabilities. The fiber coating extracts analytes from the sample either by absorption for the pure polymer coatings or a combination of absorption and adsorption for the coatings with solid particles in the polymer. Over the years the technology evolved with the development of new coatings that increased extraction efficiency. Variables for the coatings comprise different adsorbents with their different properties, different polymers, the ratio of particles and polymer as well as simply the coating thickness.

In this work, different adsorbent materials, coating lengths and thicknesses have been comprehensively investigated to elucidate the impact of particle structure and properties and the coating dimensions on the extraction and desorption of a diverse set of analytes covering a broad range of molecular weights and polarities. Divinylbenzene particle coatings contain predominantly macro- and mesopores and are slightly less retentive compared to carbon adsorbents containing a higher share of micropores, which are providing better retention of small polar and, to a lesser degree, non-polar analytes. Larger dimensions (length, thickness) of the coating improve extraction capacity, but compromise desorption efficiency. The results of this work support SPME users in the selection of the best suited SPME phase for their analytical task.

[1] J.B. Pawliszyn, et al., Water Quality Research Journal (1989) 24 (1), 179–191. DOI: https://doi.org/10.2166/wqrj.1989.010

OP9-3-1 Fast determination of total malondialdehyde in urine by HPLC-MS/MS

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The development of rapid analytical methods is crucial for the research, discovery, and confirmation of sensitive biomarkers. Furthermore, the implementation of rapid analytical processes contributes to efficient and time-effective procedures. Malondialdehyde (MDA) is an important biomarker of oxidative stress in biological systems and its accumulation has been linked to numerous pathologies. We show here the development of a fast and robust liquid chromatography-mass spectrometry method to quantify MDA in human urine without any associated derivatization reaction. MDA was separated in 3 minutes on an LC Accucore Urea-HILIC column (150x2.1mm, 2.6 µm) and was analyzed using a triple quadrupole mass spectrometer in negative electrospray ionization mode. With a 50-fold dilution as the only sample pretreatment, no matrix effect was present, which allowed a fast and simple external standard calibration with a limit of detection of 0.5 µg/L. The relative recoveries of MDA were found to be satisfactory, ranging from 99% to 107%. The proposed method also demonstrated good repeatability and reproducibility (RSD<15%) for four quality control levels. The whole methodology was validated by analyzing unspiked and spiked urine samples from eleven healthy individuals and comparing with the results obtained by the standard addition method. MDA was detected in all of cases, with natural concentrations varying from 1.2 to 4.6 mg/g creatinine. Our method demonstrated excellent reproducibility, accuracy, stability, and sensitivity, making it highly suitable for routine use in clinical laboratories. One notable advantage of this technique, when comparing with previous bibliography, is that a derivatizing reaction is not needed, resulting in a total analysis time of less than five minutes per sample. This makes this method a fast, non-invasive, effective and suitable process for population studies and subsequent future research.

- K. Kartavenka et al., "LC-MS quantification of malondialdehyde-dansylhydrazine derivatives in urine and serum samples," J. Anal. Toxicol., vol. 44, no. 5, pp. 470–481, 2021, doi: 10.1093/JAT/BKZ112
- M. Giera, H. Lingeman, and W. M. A. Niessen, "Recent advancements in the LC- and GC-based analysis of malondialdehyde (MDA): A brief overview," Chromatographia, vol. 75, no. 9–10, pp. 433–440, 2012, doi: 10.1007/s10337-012-2237-1
- H.-S. Oh, J.-A., Shin, "Simple and sensitive determination of malondialdehyde in human urine and saliva using UHPLC–MS/MS after derivatization with 3,4-diaminobenzophenone," J. Sep. Sci., vol. 40, no. 20, pp. 3958–3968, 2017, doi: 10.1002/jssc.201700490

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Chronic Kidney Disease (CKD) is a common condition characterized by the accumulation of harmful metabolic substances, known as uremic toxins, in the bloodstream. Recent metabolomics studies suggested that metabolites of tryptophan and mineralocorticoid pathways along with several modified

amino acids could indicate the disease's progression [1,2]. Therefore, multitargeted approaches are desired to obtain a quantitative picture based on this data-driven hypothesis. We have thus developed a one-point internal calibration (IC) technique for the simultaneous quantification of 16 relevant analytes using a microsampling device (20 µL of plasma). This novel strategy relied on a response of stable isotope-labeled (SIL) standards added to the study samples to translate the endogenous metabolite concentrations [3].

First, an LC–MS/MS method for separation and analysis of all CKD-related metabolites, including mineralocorticoid isomers, was developed using a biphenyl column and linear gradient of acidified H2O and MeOH (0.1% formic acid). A solution of ammonium fluoride (20mM) was infused post-column to increase ionization efficiency. Chemical purities and concentrations of the respective SIL standards were confirmed to avoid quantitative bias due to the influence of impurities, isotopic interference, and ionization competition. Plasma metabolites were collected from EDTA-K tubes using a volumetric absorptive microsampling device and extracted by sonication in a H2O-ACN mixture (10/90 v/v). After evaporation and reconstitution in H2O-MeOH (95/5 v/v) samples were injected into the LC–MS/MS system. The dynamic range of the method covered three orders of magnitude, with metabolite concentrations ranging from 10 pg/mL to 20 µg/mL. Since a single concentration of internal calibrant is necessary to calculate study sample concentration, the instrument response function was carefully investigated to determine the best SIL concentration. After validation, the trueness of 16 endogenous analytes in authentic human serum ranged from 72.2% to 116.0%, the repeatability from 1.9% to 11.3%, and the overall intermediate precision ranged from 2.1% to 15.4%. The proposed approach was applied to plasma samples collected from healthy control participants and two patient groups diagnosed with CKD, highlighting substantial concentration differences for several monitored analytes. The use of IC eliminates the need for blank matrix and the extensive preparation of multilevel calibration curves, as well as the need for matrix effect studies because calibrants are not spiked into a separate matrix. This method is currently used for longitudinal monitoring of relevant biomarkers in CKD patients to develop individualized and adaptive concentration reference ranges.

[1] Y. Gagnebin, et al. Sci. Rep., 10 (2020). DOI:10.1038/s41598-020-76524-1
[2] M. Breit and K.M. Weinberger, Arch. Biochem. Biophys. 589 (2016). DOI:10.1016/j.abb.2015.07.018
[3] G. Visconti, et al. Anal. Chim. Acta, 1240 (2023). DOI:10.1016/j.aca.2022.340711

OP9-4-1 Green solvents and reagents selection with multi-criteria decision analysis

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Multi-criteria decision analysis (MCDA) is the tool supporting managerial processes that allows to make the decisions in scientifically sound manner. MCDA allows to select the best alternative from many available that are described by many, often contradictory criteria. As a result of MCDA application these alternatives are ranked, from the most appropriate to the least appropriate.

By applying typical greenness factors as criteria, MCDA can be treated as greenness assessment or greenness metric tool. The typical criteria are toxicities of compounds referring to different types of organisms and exposure pathways, environmental persistence criteria, such as hydrolysis half-life, biodegradability or parameters describing safety of handling, such as flash point, flammability or formation of toxic degradation byproducts. With incorporation of these criteria the ranking of

alternatives is made by their greenness. As greenness is multi-dimensional, multi-criteria assessment approach seems to be very appropriate.

MCDA has been used for the assessment of greenness of organic solvents, ionic liquids, deep eutectic solvents, derivatization agents. In each case full rankings were obtained with greenness degree obtained for all alternatives. This allowed to make recommendations on the application of green solvents and reagents and indicate those that should be avoided. MCDA also allows to be applied in combination with design of experiment, bringing greenness optimization into optimization of analytical processes efficiency.

OP9-4-2 Dealing with Moving 1D-Targets in Purity Analyses of Biopharmaceuticals Using 2D-LC Coupled to Mass Spectrometry

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For the analysis of related impurities in biopharmaceuticals in our work we use heart-cutting (HC) 2D-LC-MS [1–3]. The sampling of aliquots from 1D peaks that are targeted for subsequent 2D separations usually occurs in relation to a previously acquired 1D chromatogram. However, retention of large molecular weight compounds is strongly affected by small changes in chromatographic conditions, which renders a challenge in HC 2D-LC, if the 1D target one tries to sample keeps moving [3].

The first part of this contribution will discuss concepts that help address the moving 1D target issue. These include instrumental precautions and/or the observation of specific features in the online 1D signal utilized to correct retention time-based sampling events. The functionality will be demonstrated on data obtained from the analysis of forcibly degraded insulin samples.
The second part will deal with lengthy 2D-LC analysis times. A procedure is introduced that injects multiple cuts from the 1D separation into the 2D column before a single 2D elution program elutes all of the injected material at once for a single 2D chromatogram. Data show this approach to have significantly decreased the overall 2D-LC analysis time from ~250 to ~70 minutes.

[1] P. Petersson, K. Haselmann, and S. Buckenmaier, J. Chromatogr. A. 1468, 95–101 (2016).
[2] D.R. Stoll, K. Shoykhet, P. Petersson, and S. Buckenmaier, Anal. Chem. 89, 9260–9267 (2017).
[3] P. Petersson, LCGC North Am. 37(10), 740–746 (2019).

OP9-4-3 Characterizing nanoparticles: Determining size distribution and elemental composition simultaneously, using SMPS-ICPMS

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Nanoparticles (NPs) have a wide range of industrial applications, and the precise characterization of their size and composition is crucial for understanding their properties and behavior. [1] In this study, the use of scanning mobility particle sizer (SMPS) in combination with inductively coupled plasma mass spectrometer (ICP-MS) to simultaneously determine the size distribution and elemental composition of nanoparticles in suspensions and aerosols is demonstrated. This technique enables size resolved

elemental analysis of nanoparticles, making it a powerful tool for a variety of applications including air quality control, combustion processes, and the production of engineered nanoparticles etc.

Normally, SMPS operates in air, and ICP-MS runs in argon, which is very susceptible to oxygen. The SMPS flow concept was therefore redesigned to operate it in argon mode, and a rotating disc diluter (RDD) is used as a sample introduction interface for the coupled SMPS-ICP-MS. Both commercial and in-house synthesized nanoparticles were used for method development and validation. Particles of different shapes, sizes and compositions were used. The SMPS and ICP-MS signals showed good correlation and the particles containing specific elements could be efficiently resolved from the overall SMPS distribution. This quantification concept allows correcting the calculated PSD of the SMPS and quantifying complex mixtures containing different metal compounds. In order to verify the results, the model NPs were also analyzed with single-particle inductively coupled plasma mass-spectrometry (sp-ICP-MS), low-resolution transmission electron microcopy (LR-TEM), and Scanning TEM energy-dispersive X-ray spectroscopy. The results were in agreement with the hyphenated setup. [2]

In conclusion, SMPS-ICP-MS shows a potential to be used as a complementary analytical tool for the analysis of nanomaterials for different applications.

[1] https://doi.org/10.1002/9781444307504.ch6
[2] https://doi.org/10.1021/acsnano.2c01840

OP10-1-1 Electrochemical screening of lipase activity in pancreatic preparations

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Exocrine pancreatic insufficiency (EPI) is defined as decreased synthesis or secretion of pancreatic enzymes (in particular, lipase) and bicarbonate, due to preexisting pancreatic diseases, leading to the maldigestion of food and subsequently malabsorption of nutrients. Pancreatic enzyme replacement therapy (PERT) is the standard treatment for PEI. Plenty of pancreatic enzyme preparations is already available in the European market, and besides, every year significant number of new species appears on the world market. Hence, fast growth of the global pancreatic enzymes market, in particular the emergence of new manufactures or new raw materials, stimulates considerable interest in search of simple and cost-effective methods for evaluating efficacy and safety of produced pancreatic enzyme preparations. This contribution offers a state-of-the-art strategy for the determination of lipase activity in pancreatic preparations using flow injection analysis (FIA) with electrochemical detection (FIA-ED). The procedure is based on the enzymatic reaction of a specific substrate (1,3-dilinoleoyl-glycerol) with lipase from porcine pancreas and determination of enzymatically formed linoleic acid (LA) at a cobalt (II) phthalocyanine-multiwalled carbon-nanotubes modified carbon paste electrode (Co(II)PC/MWCNT/CPE)) [1, 2]. With respect to previously described conventional assays the electrochemical one presents the number of improvements in the analysis such as high reaction efficiency, less requirement of sample volume and simple preparation of stable emulsion without adding of emulsifier agents only simple stirring. Furthermore, the following implementation of FIA enabled fast, high-throughput and automated screening of enzyme activity. Such characteristics make

this tandem very promising for routine applications. Hence, the practical feasibility of the suggested assay was confirmed by the in-vitro determination of lipase activity in a set of pancreatic preparations with different activities. We strongly believe that the as-developed strategy is very favorable for rapid point-of-care testing and could be applied in near future as an alternative tool for the determination of lipase activity in routine pharmaceutical analysis or even replace the existing methods.

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OP10-1-2 Application of capillary electrophoresis in controlled drug release studies

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HPLC is considered the method of choice for monitoring drug release from polymer nanocarriers and other nanomedicines. Nevertheless, polymer carriers usually cannot be injected into the HPLC system as they can irreversibly adsorb to stationary phase or other components of the system, causing deterioration of separation efficiency or even column clogging. A liquid-liquid or solid-phase extraction step is thus necessary. On the contrary, the open-tubular columns used in capillary electrophoresis can tolerate more problematic sample matrices and eventually adsorbed polymers can be washed out using a strong base, acid, or organic solvent. In this work, we report on a successful application of capillary electrophoresis to the separation and determination of hydrophilic drugs released from polymer carriers.

We have developed a method for monitoring the release of 5-aminolevulinic acid and its hexyl ester from a hydrophilic N-(2-hydroxypropyl)methacrylamide-based copolymer. The separation was performed in 1M formic acid using capacitively coupled contactless conductivity detection as the analytes do not exhibit significant UV absorption. Using Tris cation as an internal standard and flushing the capillary with 1M NaOH, water, and 1M HCOOH before each analysis provided very good linearity and repeatability. The application of pressure to the inlet end of the capillary helped to stabilize the baseline and to shorten the separation time to 5 minutes. The total analysis time including the flushing procedure was 14 minutes. Samples of the copolymer loaded with the drug were incubated at 37 °C in buffers of different pH and directly injected to the analysis without any treatment. Significant differences in release kinetics of both drugs at pH 5.0, 6.5, and 7.4 were observed.

The second method developed within this work for monitoring the release of another hydrophilic drug, acetylsalicylic acid, from the same polymer carrier used direct UV detection. In this case, the background electrolyte was 20 mM sodium tetraborate, allowing an efficient separation of acetylsalicylic acid, salicylic acid, and salicyl hydrazide from the polymer carrier. Benzenesulfonate was used as an internal standard. Separation was completed within 4 minutes. Together with the flushing procedure, the analysis took 9 minutes. Excellent linearity and repeatability were reached without any sample treatment steps.

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OP10-1-3 An ECL Sensor based on N-CQDs as Homogenous Luminophore and Copper (II) Picrate as Electrode Modifier for Determination of Creatinine

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Creatinine (Crn) is a mammal's body metabolite that is produced from creatine during muscles activities. As a waste, Crn is continuously removed from the body by kidney and its amount in blood and urine is clinically determined for diagnosis of diseases related to renal, muscular, and cardiovascular dysfunctions [1]. The most common clinical method for determination of Crn is the Jaffe method. However, this colorimetric method is fraught by problems like low selectivity and time-consuming sample preparation [2]. Therefore finding a new, fast and cost-effective analytical methods for detection of Crn is very important and meets a great necessity. Electrochemiluminescence (ECL) as a developing analytical technique has attracted attentions because of its numerous advantages like high sensitivity and cheap instrumentation [3]. Finding novel ECL systems due to developing of new luminophores, construction of new devices, and application of new current/potential styles are important issues in this area. Nitrogen doped carbon quantum dots (N-CQDs) are recently used as new luminophores in ECL systems. Biological compatibility, cheap precursors, and short route and short time synthesis methods are rendering this quantum dots attractive as common luminophores in ECL analyses.

In our present work N-CQDs were synthesized by one-pot synthesis method and characterized by methods like high resolution transmission electron microscopy (HR-TEM), X-ray photoelectron spectroscopy (XPS), field emission scanning electron microscopy (FESEM), X-ray diffraction (XRD), fluorescence spectroscopy, dynamic light scattering (DLS), Fourier transform infrared (FT-IR) spectroscopy and UV–vis spectroscopy. Multi pulsed amperometric method was used for driving a cathodic ECL. Unlike usual conditions of application of CQDs in ECL systems, synthesized N-CQDs were used for a homogenous ECL. Namely, the N-CQDs were dissolved in the electrolyte of the cell beside of the $S_2O_8^{2-}$ coreactant. Glassy carbon electrode was modified with synthesized copper (II) picrate to improve the selectivity of the creatinine. ECL signals of this system were enhanced linearly by increasing of the creatinine concentration. Experiments showed that this sensor could be applied for analysis of creatinine in human blood serum and urine.

1. N. N. Turner, et al, Oxford Textbook of Clinical Nephrology, fourth ed., Oxford University Press, Oxford 2015. DOI: 10.1093/med/9780199592548.001.0001
2. R. Kumar, et al, Analytica Chimica Acta 1183 (2021) 338748. DOI: 10.1016/j.aca.2021.338748
3. W. Miao, Chemical Reviews 108 (2008) 2506-2553. DOI: 10.1021/cr068083a

OP10-2-1 LC-MS/MS-based strategy for studying the influence of environmental conditions on saponin content in plant organs Saponaria officinalis, L.

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Biologically derived surfactants biosurfactants) are attracting increasing interest from basic research and industry. This is mainly due to the compliance of the biotechnological methods used to obtain them with the principles of sustainable development. Moreover, the biological activity of biosurfactants has accompanied humanity since primitive herbal medicine. In particular, this applies to biosurfactants obtained from plants belonging to the group of saponins. Saponins are compounds of plant origin with amphiphilic properties, in which triterpenes or sterols are the hydrophobic part (aglycone). The hydrophilic part comprises 1-3 multi-membered chains (mono-, bi-, tridesmosides). Depending on the plant species and variety, the organ, stage of development, or environmental conditions, each plant can produce up to 100 types of saponins, differing in aglycone and sugar moiety [2]. The most stable and abundant derivatives have been primarily isolated by chromatographic and characterized by spectroscopic methods (mainly NMR spectroscopy). Still, many saponins have not been characterized. Their routine semi-quantitation would significantly facilitate detailed research on the mechanisms of saponin synthesis and distribution depending on environmental conditions.

The project aimed to create an analytical procedure for the qualitative and quantitative analysis of saponins in extracts from soapwort (Saponaria officinalis, L.), a model plant found worldwide, capable of producing and accumulating significant amounts of triterpene saponins in the organs.

In the first step, the fragmentation mechanism of selected saponins was investigated using ESI-MS/MS. Next to the standard release of sugar residues, products of the retro-Diels-Alder reaction were also observed. The identity of saponins found in soapwort was confirmed by establishing the identity of aglycones obtained via hydrolysis. Finally, the precursor ion monitoring method for selected fragmentation ions was developed to detect unknown saponins, and the multi-reaction monitoring (MRM) method was built to track changes in the content of compounds extracted from plants. The established method allowed us to discover up to 30 different glycosides composed of 8 aglycones. MRM-based results obtained for 150 transitions and chemometric analysis (principle-component and cluster analysis) confirmed that the extract's composition depends on the extraction method, plant cultivation conditions, and the content of metal ions in the soil.

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1. *J. Zhang, at al., Molecular Plant 13 (2020) P1358, DOI:10.1016/j.molp.2020.09.007*
2. *R. Gevrenova, at al., Phytochem. Anal. 29 (2018) 250, DOI:10.1002/pca.2739*

OP10-2-2 OctoChemDB: A Web Service for Efficient Dereplication of Natural Products using High-Resolution Mass Spectra

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The identification of unknown compounds in complex mixtures is a challenging task in mass spectrometry (MS) analysis. Commercial MS software tools often require installation, upgrades, and have proprietary data file formats, which limit their use with instruments from different vendors. To overcome these limitations, we have developed mass-tools [1], a suite of JavaScript open-source libraries that allows web-based analysis of mass spectra [2–4].

Our tools have been enriched with a new feature named OctoChemDB [5]. OctoChemDB allows the identification of unknown compounds using public databases of MS/MS spectra, publications, and patents, and enables filtering by bioactivity, natural origin, and molecular sub-structures. The OctoChemDB web service is created in two phases, namely synchronization and aggregation. During synchronization, a plugin system creates local copies of open databases like PubChem, PubMed, Lotus, etcetera. Each plugin is responsible to create and update the local copy of its own database. In the aggregation phase, these databases are joined based on their normalized 2D structures that omits stereochemistry and tautomeric isomers. The resulting database can be queried by monoisotopic mass as well as other criteria like taxonomy, bioactivity. The results are returned as a JSON object for integration into web applications.

To evaluate the efficiency of our advanced tools, we analyzed extracts from fungi and extremophile organisms. By combining the search by monoisotopic mass/isotopic profiling similarity, MS/MS fragments similarity, the compound PubChem search, and filtering by sub-structure, taxonomy, and bioactivity, we successfully and quickly identified six known antibiotics compounds with high confidence from organisms that were not previously known to produce them, Cephalochromin being one of several successful outcomes to be showcased.

In summary, our suite of web-based MS analysis tools, enriched with the advanced feature OctoChemDB, provides an efficient and cost-effective solution for the processing and identification of unknown compounds. These tools are readily accessible and open source without software installation and can be tested on-line on https://octo.cheminfo.org, making them easy to use for the research community. This approach to compound identification has proven its strength and efficiency in dereplication of natural products, with several interesting hits released from the extracts in a very short time.

(1) Patiny, Luc; Zasso, Michaël; Silvestre, Ricardo. Javascript Libraries Dealing with Mass Spectra, 2023. https://doi.org/10.5281/ZENODO.5091564.
(2) Ortiz, D.; Gasilova, N.; Sepulveda, F.; Patiny, L.; Dyson, P. J.; Menin, L. Aom 2 S: A New Web-based Application for DNA/RNA Tandem Mass Spectrometry Data Interpretation. Rapid Commun Mass Spectrom 2020, 34 (23). https://doi.org/10.1002/rcm.8927.
(3) Patiny, L.; Borel, A. ChemCalc: A Building Block for Tomorrow's Chemical Infrastructure. J. Chem. Inf. Model. 2013, 53 (5), 1223–1228. https://doi.org/10.1021/ci300563h.
(4) Desport, J. S.; Frache, G.; Patiny, L. MSPolyCalc: A Web-based App for Polymer Mass Spectrometry Data Interpretation. The Case Study of a Pharmaceutical Excipient. Rapid Commun Mass Spectrom 2020, 34 (S2). https://doi.org/10.1002/rcm.8652.
(5) Silvestre, Ricardo; Patiny, Luc; Zasso, Michaël. OctoChemDB: A Comprehensive Web Service for Chemical Data Integration, 2023. https://doi.org/10.5281/ZENODO.5091586.

OP10-2-3 COMPREHENSIVE GCXGC HIGH RESOLUTION MS AND SELECTIVE ISOLATION OF CHEMICALS IN THE INVESTIGATION OF HUMAN CHEMOSIGNALS ELICITED FROM EMOTIONAL STIMULATION

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An update from the Potion project, which is investigating compositional changes of sweat volatiles in 40 volunteers undergoing fear stimulation to demonstrate the existence of human chemosignals affecting the emotional state of a receiver [1]. Upon collection from apocrine sweat glands in the armpit by pre-treated pads, chemicals were collected and then extracted, enriched, and trapped into Tenax GR tubes with dynamic headspace (DHS at 60°C). Analyses were carried out by comprehensive two-dimensional gas chromatography (GCxGC) and time-of-flight mass spectrometry (TOF), whereas emotions were induced by immersing individuals wearing mobile sensors (EDA, ECG, respiratory rate) in virtual reality scenarios. Emotional rankings showed the successful induction of fear. Out of 364 detected compounds (311 identified, 38 assigned to class, 15 true unknowns), a subset of 287 sweat volatiles showed significantly different concentration values compared to field blanks (fold change FC < 3.0, p.FDR < 0.05) [2] and 24 were significantly increased during fear vs. relaxed condition (FC: 2.21±0.78, p.FDR.BH < 0.05, with paired t-tests). Lab-made modifications of the GCxGC setup allowed to selectively isolate a number of these chemicals for tests on human volunteers [3]. Real time measurements by proton transfer reaction time of flight confirmed that the concentrations of these chemicals change when volunteers experience fear in the virtual scenario.

[1] N. Gomes, F. Silva, G. Semin, Behav. Res. 52 (2020) 2438–2451.
[2] M. Ripszam et al. Talanta 257 (2023) 124333.
[3] F. Di Francesco, M. Ripszam, T. Bruderer, F. M. Vivaldi, EP patent request: 23163823.0, filed 23/3/2023.

OP10-3-1 Remote teaching in Analytical Chemistry – Lessons learned during COVID-19 pandemic

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In spring 2020, teaching at universities all over the world changed dramatically within a very short period of time. Lecturers as well as students had to get used to new forms of teaching, learning and communication, all of which were mainly based on remote approaches, thus avoiding any personal contact and slowing down the spreading of COVID-19.

While giving lectures and keeping in touch with students could often be rapidly transferred into commercially available web-based tools, the transfer of laboratory lessons and education into a form being adequate for pandemic conditions was even more challenging. Additionally, one should keep in mind that every web-based solution is strongly dependent on infrastructural boundary conditions (e.g., availability of computers, internet access, stability of internet connection, personal working space for lecturers and students – just to mention a few).

In this presentation, the different ways of remote teaching will be reflected and summarised. A focus shall be directed to those forms of remote teaching in Analytical Chemistry that have been evolved during or even due to the pandemic situation – especially considering the transfer of lab courses into new formats. Remote teaching approaches and forms in Analytical Chemistry being also valuable tools in the post-pandemic era shall be identified and discussed.

The presentation will finally give an estimation in how far remote approaches affect communication between lecturers and students as well as between students and students.

OP10-3-2 Case-based active learning in BSc and MSc subjects of analytical chemistry for the improvement of soft skills

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Nowadays, employers demand BSc and MSc graduates holding soft skills essential to develop their professional activity. This fact calls for a paradigm shift in university teaching to implement teaching methodologies that focus on the acquisition of skills, in addition to the transmission of knowledge of the discipline. These skills must include inter- and intrapersonal and communicative aspects, such as analytical capacity, critical thinking, teamwork, data interpretation, effective time management, decision making, and tackling new and open problems.

To achieve this goal, the case-based learning (CBL) methodology is an active learning strategy that poses real or close-to-real problems (cases), which require individual learning, data evaluation and information search, to reach one of the possible 'correct' solutions. This helps the students see the value and meaning of their learning, increase their interest, and improves their skills through active and collaborative tasks and a rich final debate between students and teachers.

As in other disciplines, the history surrounding a given case allows contextualizing the learning objectives of analytical chemistry through a possible real-life problem. Thus, this work describes the implementation of the CBL methodology in an experimental subject of the BSc degree in Chemistry and in two theoretical subjects of the MSc degree in Analytical Chemistry, in the framework of the Institutional Project to Promote Teaching Quality at the Faculty of Chemistry of the Universitat de Barcelona (RIMDA-Chemistry project).

In the experimental subject, cases are worked in small groups throughout the subject and focus on the analysis of a water sample (to ensure its potability), a potentially contaminated solid waste (to categorize its final use), and an animal feed (to determine nutritional parameters). The information provided to the students is minimal (target analytes) and they have to decide the methodology to be applied, the instrumental techniques to be used, and plan who, how and when the determinations will be carried out. In the theoretical subjects, some class sessions are devoted to solving cases within groups of students (e.g. design of a sampling strategy; environmental problem in a highly industrialized river), which allows the students to consolidate the knowledge already acquired through active and collaborative learning.

The CBL approach allows for a systematic improvement in the student acquisition of skills, especially decision making, oral communication, information search and critical interpretation of data and information. After the CBL implementation, the feedback provided by the students evidences a high satisfaction with the learning experience.

OP10-3-3 A modern curriculum for educating industry-oriented specialists in analytical and bioanalytical chemistry

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Swiss Universities of Applied Sciences offer industry-oriented bachelor curricula that are designed to allow students to perform successfully in industry right after their bachelor studies. A new and unique study program was introduced since Fall 2022 at the School of Engineering in Sion (Analytical Chemistry major of the Life Technologies degree program) for students interested in becoming specialists in industrial analytical and bioanalytical chemistry.

With 40 ECTS in analytical and bioanalytical chemistry (excluding the bachelor thesis), the curriculum addresses all current key instrumental analytical chemistry techniques in theory and practice for small and large molecules and biomolecules in simple and complex matrices. Furthermore, practical test method development and validation are integral parts of the curriculum. Acquisition of skills is ensured via industry-oriented practical work topics and projects using a variety of analytical instruments and sample preparation technologies. The curriculum provides a strong background education in basic chemical, engineering, and biochemical sciences, in particular the chemistry of biomolecules. The new curriculum also addresses current topics such as green analytical chemistry and digitalization. Teaching activities are connected to our Life Technologies Institute R&D activities and our ISO 17025-accredited analytical platform service activities, which supports the industry-oriented analytical education.

We believe that this 3y industry-oriented bachelor curriculum in analytical and bioanalytical chemistry is a timely, unique program at a University of Applied Sciences.

OP10-4-1 Addressing some challenges on metal ions determination in dynamic water systems using flow-based approaches

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As water bodies are dynamic systems, the presence of metal ions must be a target of spatial-temporal monitoring. The real-time monitoring is rather cumbersome as current methods rely on transport to off-site laboratories, causing the disruption of the sample characteristics, due to pH and redox potential change and exposure to oxygen, light or temperature shifts, leading to diverse chemical equilibria shifts. This could be potentially overcome by devising new smart sampling procedures and flow-based monitoring. Monitoring water bodies pose some important analytical challenges: coping with a wide range of analyte concentrations; the possible need for analyte enrichment; minimization of interferences; achieving speciation; search for more sustainable chemistries; reducing sample and reagents consumption. To tackle the above-mentioned challenges, the use of in-line separation processes like membrane separation processes and solid phase extraction, have emerged as powerful tools to increase the selectivity and sensitivity of the flow methods, and yet to maintain the major advantages of their use, namely the relative simplicity and good repeatability. In this scenario, some recent contributions of the group in this line of work will be presented.

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OP10-4-2 Monitoring dynamic water systems with microfluidic paper-based devices for in-situ analysis

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To attain an immediate, on-hand response, the use of self-readable, easy-to-use devices is a natural choice namely the concept of microfluidic paper-based analytical devices (µPADs). The portability and low consumption of both reagents and sample associated with this technique, make these devices ideally suited for unskilled operators and frequent analysis. The concept is based upon the microfluidics through the cellulose fibers and the setting of hydrophobic/hydrophilic interfaces for microflow manipulation. Using colorimetric reactions, the analyte concentration can be correlated to the colour intensity, which can be measured with a flatbed scanner and computer software. The use of digital scanning as detection process has enabled to maintain the accuracy and reliability of the analysis in opposition to other paper-based visual indication techniques, with a positive/negative or concentration range response.

The downscaling of the analytical procedures results in a size limitation for in-line pretreatments as well as speciation and interferences minimization processes. Targeting metal ions quantification in dynamic water systems emphasizes these challenges. The adverse effects of metal ions, namely zinc, iron, and copper, are well documented, for displaying toxic, carcinogenic, and mutagenic effects for living organisms if present in high concentrations. The presence of these ions must be a spatial-temporal monitoring and current methods rely on transport to off-site laboratories, causing the disruption of the sample characteristics, due to pH and redox potential change and exposure to oxygen, light or temperature shifts, leading to diverse chemical equilibria shifts.

In this context, we developed a series of µPADs for metal ions quantification for on-site, real-time monitoring of natural waters. To be an effective alternative, all the developed devices were validated by comparing the results obtained in analyzing different water samples with reference standard procedures. Several devices will be presented and the advantages and disadvantages discussed.

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OP10-4-3 Automated solid phase extraction and fluorimetric detection with a flow-based method for the determination of tetracyclines in wastewater

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Tetracyclines are used as antibiotics in human, veterinary, agricultural and livestock medicine. The widespread application of these pharmaceuticals leads to an accumulation of their residues in the environment. This contributes to bacterial resistance and harms human and environmental health [1]. They are the most common antibiotics found in wastewater and can cause inhibition of the biological treatment of wastewater in treatment plants [2]. For this reason, a fast, sensitive and precise method is needed for its detection in wastewater treatment plants.

This work presents a multisyringe flow injection analysis (MSFIA) system for the automatic extraction and determination of tetracyclines in wastewater samples.

The sample was adjusted with McIlvaine-Na_2EDTA buffer before solid phase extraction with an Oasis HLB column carried out for the preconcentration of tetracyclines. The Europium (Eu (III))-based and citrate-mediated method (using Tris-HCl buffer) was selected for the fluorimetric analysis (λexc= 400 nm, λem= 612 nm) [3]. For fluorescence detection, a low-cost system consisting of an USB 2000 CCD detector and a 3D-printed support that stores an LED light source was used. The extraction and detection parameters, including elution solvent, elution volume, eluent loading rate, europium/citrate concentration and others, were systematically optimized. The proposed method provided low limits of detection (9.4 µg L-1) and quantification (31 µg L-1), and good values for intra-day (< 4 %) and inter-day precisions (< 6 %). Recoveries of spiked tetracycline in wastewater samples ranged from 87 to 106%. Finally, the results of this work were compared with those obtained by the reference method commonly used for tetracyclines detection, namely liquid chromatography coupled to a fluorescence detector (UPLC-FLD).

Keywords: Tetracyclines, MSFIA, solid-phase extraction, fluorimetric detection, wastewater analysis

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[1] R. Daghrir & P. Drogui., Environ. Chem. Lett. 11 (2013) 209-227. DOI: 10.1007/s10311-013-0404-8
[2] M. Czatzkowska, et al., Energy. Sci. Eng. 8 (2020) 1880-1897. DOI: 10.1002/ese3.609
[3] F. Hijaz, et al., Antibiotics., 10 (2021) 566. DOI: 10.3390/antibiotics10050566

POSTERS

PS-01 Development of a micro-sampling SPE method for drug separation from human serum coupled to a SERS sensing assay for molecular quantification of relevant drugs in TDM

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Surface-enhanced Raman scattering (SERS) is a simple analytical technique with the advantage of being fast, cost-effective, highly sensitive and requires the use of low amounts of sample for measurement [1]. Label-free SERS detection of small molecules, like pharmaceuticals relevant in therapeutic drug monitoring (TDM), can be performed with high sensitivity in biological matrix samples, such as in blood or serum [2, 3]. However, to perform an efficient label-free approach with high sensitivity detection, the biological matrix should be reduced in complexity, allowing an efficient drug separation. Solid-phase extraction (SPE) is a very efficient method for analyte separation from high complex samples, that has been widely used as a sample pre-treatment prior to analytical analysis, such as in liquid chromatography [4]. Nevertheless, most conventional SPE assays are expensive, time consuming and require the use of high-volume samples and large amount of volumes of organic solvents to ensure the analyte separation [5]. Therefore, the development of SPE-based approaches for minimum volume sample use, less time consuming and cost effective, are needed without compromising the performance of conventional SPE. In this work we developed and implemented an in-house built miniaturized SPE system (µ-SPE) for efficiently separation of methotrexate (MTX) and lamotrigine (LTG), two widely used drugs for treatment in oncology and epilepsy, respectively, that require the current performance of TDM [6]. The µ-SPE method was in-line combined with a SERS assay, based on molecular migration and adsorption through ordered nanopillar structures in a SERS substrate, to enable label-free detection of MTX and LTG in human serum. Furthermore, we implemented a multivariate spectral analysis model, based on partial-least squares regression (PLS-R), for accurate drug quantification within clinically relevant levels. The here developed method showed to be highly efficient (> 80%), within an average recovery of 73%, for MTX and LTG separation, allowing to reach a LoD of 0.15 µM and LoQ of 0.55 µM for MTX, while for LTG it was of 3.2 µM and 9.5 µM for LoD and LoQ, respectively. Moreover, this method led to reduce both the use of high, volume samples and organic solvents, less handling and time consuming in sampling and cost-effective for in-line implementation with SERS measurements. The herein fully developed sensing assay showed the robustness and sensitivity of label-free SERS for LTG quantification and highlights the possibility for point-of-need TDM in clinics.

[1] S.E.J. Bell, et al., Angew. Chemie. 59 (2020) 5454–5462. https://doi.org/10.1002/anie.201908154
[2] O. Durucan, et al., ACS Sensors. 3 (2018) 2592–2598. https://doi.org/10.1021/acssensors.8b00887
[3] S. Rostami, et al., Food Control. 113 (2020). https://doi.org/10.1016/j.foodcont.2020.107183
[4] C. Deriu, et al., Anal. Chem. 91 (2019) 4780–4789. https://doi.org/10.1021/acs.analchem.9b00335
[5] T. Hyötyläinen, Analytical Bioanal. Chem. 394 (2009) 743-758. https://doi.org/10.1007/s00216-009-2772-2.
[6] S.H. Preskorn, et al., Psychiatr. Clin. North Am. 16 (1993) 611–645. https://doi.org/10.1016/S0193-953X(18)30167-9.

PS1-02 INTERACTION OF THE ALKALOID FAGARONINE AND OTHER BENZO[C]PHENANTHRIDINE ALKALOIDS WITH G-QUADRUPLEXES

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G-quadruplexes (G4) are lesser-known DNA structures, which are formed on guanine-rich sequences of nucleic acids. These structures may play an important role in common cellular processes, such as transcription and replication of DNA. Stabilization of these structures can lead to the slowing down of DNA replication and the associated cell division. This phenomenon can be used in cancer treatment to inhibit tumor growth. We are trying to achieve this goal by forming a complex of G4 with plant alkaloids, which have already been proven to have the ability to bond to DNA and stabilize it. Some members of the benzo[c]phenanthridine group of alkaloids are already used as anticancer drugs (such as sanguirubine and sanguinarine), so we focused on the lesser-researched members of this group of alkaloids, especially fagaronine.

In this study, we researched several benzo[c]phenanthridine alkaloids and their interaction with G4s. We measured the properties of the interaction between G4s and alkaloids, determined their stabilizing effect, and compared the lesser-known members with those already in use. We found out that some other representatives from this group of alkaloids have a similar, if not better, stabilizing effect on the investigated G4 structures than those already used in practice, especially the already mentioned fagaronine. These results may lead to the use of these alkaloids as potential anticancer drugs.

Jarosova P, Paroulek P, Rajecky M, et al. Naturally occurring quaternary benzo c phenanthridine alkaloids selectively stabilize G-quadruplexes. Physical Chemistry Chemical Physics. 2018;20(33):21772-21782. , DOI10.1039/c8cp02681e

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PS1-03 Enhancement of luminescence signal by deuterated water

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Luminescence detectors are widely used in analytical instrumentations for the sensitive detection of molecules. Native fluorescence, as well as extrinsic fluorescence (probes, labels, etc.), is very dependent on the conditions of the medium (solvent, pH, ionic strength, etc.). Many experiments are very often provided in normal water as a medium. When water is merely substituted for deuterated water, an increase of both fluorescence intensities and lifetimes can reach units to hundreds of percent. The rise can be seen even by the naked eye. The higher fluorescence intensity would enable the lowering of LOD or decrease the concentration of the sample. The molecules potentially profiting from such an enhancement include many dyes, biomolecules, or pharmaceuticals. Among emission spectra measurements, we have successfully applied deuterated water in HPLC with fluorescence detection, CE-LIF, flow cytometry, fluorescence microscopy, fluorescence lifetime imaging, and other methods to enhance the fluorescence signal.

J. Kucera, O. Pes, T. Janovic, C. Hofr, L. Kubinyiova, J. Toth, S. Kana and P. Taborsky, Sensors and Actuators B-Chemical, 2022, 352. https://doi.org/10.1016/j.snb.2021.131029.

PS1-04 Encapsulation of Vecuronium Bromide by Sugammadex Studied by SERS

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Vecuronium bromide is a drug used during anesthesia to relax skeletal muscles by competing with acetylcholine for acetylcholine receptors. After surgery, recovery of neuromuscular system is accomplished by administration of sugammadex, a γ-cyclodextrin which encapsulates vecuronium preventing it from further binding to receptors.[1]

To study complexation of vecuronium bromide (VecBr) with sugammadex (SDX) surface-enhanced Raman spectroscopy (SERS) was applied. Silver nanoparticles prepared by reduction of silver ions with hydroxylamine hydrochloride were used as SERS active substrates, additionally aggregated with calcium nitrate as needed. Concentration dependent SERS spectra of VecBr and SDX were obtained in the 5×10^{-7}–1×10^{-4} M range, though SERS spectra of cyclodextrin were observed only in the presence of the aggregating agent. VecBr/SDX complexes were prepared by mixing the drug and cyclodextrin in 1/1 molar ratio, but differing in the order of addition of the components into the silver colloid as well as the incubation time.

The SERS spectra indicated that adsorption mechanism of the studied compounds was based on electrostatic interactions with the layer of ions on the silver nanoparticle surface. The positively charged piperidinium moiety allowed adsorption of vecuronium on the silver nanoparticles covered by negatively charged chloride ions, whereas negatively charged carboxylate groups prevented efficient adsorption of sugammadex on the metallic nanoparticles, which was nevertheless facilitated in the presence of the neutralizing Ca^{2}+ ions. Significant spectral changes upon complexation included vibrational modes of SDX glucose rings and VecBr steroid rings, implying that the entry of the VecBr molecule into the SDX cavity was not only driven by the attractive electrostatic interactions between the positive piperidinium and negative carboxylate groups, but also supported by hydrophobic interactions between the host and drug molecule.

1. A. Bom, M. Bradley, K. Cameron, J.K. Clark, J. van Egmond, H. Feilden, E.J. MacLean, A.W. Muir, R. Palin, D.C. Rees, M.-Q. Zhang, Angew. Chem. 2002, 41(2):266-70.
https://doi.org/10.1002/1521-3773(20020118)41:2<265::AID-ANIE265>3.0.CO;2-Q

PS1-05 Real-Time Monitoring of Hydrogenation Reaction at the Nanoscale using Tip-Enhanced Raman Spectroscopy

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Hydrogenation, an essential process in organic synthesis, encompasses a wide range of industrial applications, including the denitrification of carcinogenic compounds, hydrocarbon cracking, and petroleum refining, among others.

The traditional approach to monitoring hydrogenation involves cooling the system, depressurizing it, and collecting a sample, followed by subsequent analysis. Typically, analytical techniques such as HPLC, TLC, and NMR spectroscopy require a considerable period of time to provide an analytical response.[1]

Other conventional spectroscopic techniques for the study of chemical reactions, including infrared, ultraviolet–visible, Raman and fluorescent spectroscopy are restricted to the micrometer scale in terms of their spatial resolution because of the diffraction limit of light. On top of that, Raman scattering is a very weak effect giving relatively low signal intensity.[2] Nevertheless, the challenges posed by the low sensitivity and spatial resolution of Raman spectroscopy can be addressed by leveraging the localized surface plasmon resonance effect in tip-enhanced Raman spectroscopy (TERS), which generates a highly intense and localized field (near-field) at the apex of a metallic scanning microscopy probe.[3]

This study demonstrates the efficacy of TERS as a sensitive analytical tool for dynamic monitoring of hydrogenation reaction under ambient conditions. Remarkably, our findings indicate that hydrogenation can be safely performed in a TERS microscope, obviating the need for a pressure reactor. By investigating the chemical fingerprint change by TERS imaging of chloro-nitrobenzenethiol (CNBT) on Pt(111) surface, we show that conversion to chloro-aminobenzenethiol (CABT) is a near-instantaneous process. Notably, the reduction begins immediately, as evidenced by the Raman map, which shows clear disappearance of NO_2 vibrations at 1336 cm-1 within 5 seconds after introducing hydrogen flow to the system. Along with the disappearance of the NO_2 band, we observed a shift in the vibrational frequencies of C-N and C=C bonds, from 1127 cm-1 to 1108 cm-1 and from 1576 cm-1 to 1595 cm-1, respectively, confirming the formation of the $–NH_2$ group. The reduction process typically achieves complete conversion within 6 seconds, resulting in consistent peak positions in the spectra described above. In summary, this study demonstrates the potential of operando TERS imaging to study the kinetics of hydrogenation reactions at the nanometer length scales.

(1) Marziano, I. et al., Org. Process Res. Dev. 2000, 4 (5), 357–361. DOI:10.1021/op000030m.
(2) Van Schrojenstein Lantman, E. M., et al., Nat. Nanotechnol. 2012, 7 (9), 583–586. DOI:10.1038/nnano.2012.131.
(3) Cai, Z. F., et al., CCS Chem. 2023, 5 (1), 55–71. DOI:10.31635/ccschem.022.202202287.

PS1-06 Multi-elemental analysis of hair by energy dispersive x-ray spectroscopy without sample grinding and mineralization

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Humans are continuously exposed to various toxic elements, which can appear in increased quantities in the atmosphere, water, soil, plants, and animals. These differences may be due to natural causes (volcanic activity, soils, and waters with locally increased metal content) and anthropogenic causes (intensive agriculture, animal husbandry, mining, chemical metallurgy, transport, and households). Therefore, the human chemical environment is very complex and variable over time, leading to diseases with difficult-to-detect causes. Often, chronic and/or acute poisoning by a toxic element through ingestion, inhalation, or dermal contact cannot be ruled out as one of the causes. The result is most often an accumulation of metals in the body and changes in the functioning of organs and glands, such as the heart, brain, kidneys, bones, and liver. Therefore, there is an increasing demand for cheap and easily accessible diagnostic methods to prevent and control the patient's condition during therapy [1-2].

The work aimed to develop a simple screening method for determining metals in human hair using energy-dispersive X-ray fluorescence spectrometry (ED-XRF). Many problems related to the low

amount of material taken for analysis needed to be solved during method development (such as hair homogenization and the preparation of a representative pellet). Grinding was replaced by the dissolution of hair with TMAH, solvent evaporation, and pellet formation. New vessels were also designed, allowing metal determination in twice as small pellets as commercially available ones. Finally, less than 70 mg of hair sample was required to determine metals without loss of sensitivity. The method was validated using certified reference material and comparing amounts of metals in hair by ED-XRF and ICP-MS/MS. Amounts of 13 elements were in agreement with certified values. The developed sample preparation method can be implemented for metal analysis using other XRF systems.

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1. *M. Kosanovic, at al., Environ Monit Assess 174 (2011) 635, DOI:10.1007/s10661-010-1484-6*
2. *A. Taylor, at. Al., J. Anal. At. Spectrom. (2020), DOI: 10.1039/d0ja90005b*

PS1-07 Application of Vibrational Spectroscopy Coupled with Chemometrics for the Discrimination of Organic vs. Conventional Culture Systems for Red Grape Extracts

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The beneficial health effects of fruits have been attributed to the presence of fibers, minerals, vitamins, and phytochemicals, including phenolic acids, flavonoids, and anthocyanins. In grape berries, the phenolic compounds, flavonoids, and non-flavonoids are distributed in different parts of the fruit. This study aims to investigate the applicability of FTIR and Raman screening spectroscopic techniques combined with multivariate statistical tools to find patterns in red grape berry parts (skin, seeds, and pulp) according to grape variety (Merlot, Feteasca Neagra, Pinot Noir, and Muscat Hamburg) and vineyard type (organic and conventional). Spectral data were acquired and processed using the same pattern for each different berry part (skin, seeds, and pulp). In this respect, two vibrational spectroscopic techniques were used during experiments, infrared (IR) light absorption and Raman scattering, both aiming at investigating the chemical functional groups of organic compounds in studied grape samples, and potential changes occurring while applying extraction procedures. Gathering information on differences between grapes sampled from organic and conventional vineyards was also in the scope of this study. Through principal components analysis (PCA), the results of the decomposition of the spectral data have shown that with the first three PCs, over 91% of the total variability of the studied data was included. PCA was able to differentiate organic and conventional culture systems for red grape extracts (skin, seeds, and pulp) for the studied variety; overall differences derived from both score and loading plots emphasize the need to elucidate which key compounds/classes of compounds possess the discriminant ability. For skin and seed extracts, FTIR data processing using AHC has revealed a better classification compared with Raman data, at a lower dissimilarity level subclusters division, allowing a classification based on vineyard type (organic vs. conventional). Classification and cross-validation by PC-DA have shown that a chemometric approach was able to discriminate the two culture systems for skin (87.5%—FTIR data, 100%—Raman data), seeds (100%—FTIR data, 100%—Raman data) and pulp (100%—FTIR data, 100%—Raman data) hydroalcoholic extracts. The innovative approach presented in this work is low-cost and feasible,

expected to have applications in studies referring to the authenticity and traceability of foods. This study's findings are also useful in solving a great challenge that producers are confronting, namely the consumers' distrust of the organic origin of food products.

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PS1-08 Analytical spectroscopic characterization of green copper nanoparticles for antimicrobial applications

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Analytical spectroscopy plays a leading role in the characterization of nanomaterials. An exhaustive nanomaterials characterization and the appropriate choice of methodologies for nano-toxicological risk assessment promise nowadays a reliable and accurate data output to guarantee their safe application in real-life products. In this communication, analytical spectroscopy was used to guide and support the production of human-safe copper nanoparticles (CuNPs). Cu is an essential component of many enzymes involved in chemical reactions and also possesses good antimicrobial activity [1]. This study demonstrates a simple approach to synthesize green core-shell CuNPs stabilized by poly(n-vinyl)pyrrolidone (PVP). CuNPs were prepared using copper sulfate as precursor and glucose as reducing agent. The synthesis of small CuNPs employing glucose as reductant is known since 2006 [2], but the whole process was carried out in nitrogen atmosphere to prevent CuNP oxidation. The presence of PVP in the synthetic medium eliminates the need for an inert atmosphere during the process [3]. PVP can act as stabilizer, dispersant, and reducing agent, its role depending on synthetic conditions. This is possible thanks to the amphiphilic nature of PVP, along with its stabilizing capability. The influence of glucose and PVP concentrations, along with pH of the synthetic medium, were here investigated, considering colloidal stability and NP average size as discriminating issues. Synthetic parameters were properly tuned to obtain NPs with diameters above 100 nm. This way, the dangerous nano-cytotoxicity associated to the use of smaller NPs was waived a priori. A thoughtful spectroscopic characterization of CuNPs was performed by infrared, UV-Vis and X-ray photoelectron spectroscopies. These measurements demonstrated the formation of a PVP layer around a Cu(0) core. Size distributions and average shell thickness were obtained by statistical analysis on transmission electron microscopy micrographs. Cu@PVP particles were embedded in polymer matrices to prepare coatings for food packaging applications. Copper release kinetic was assessed by atomic absorption spectroscopy, and used to theorize the antimicrobial potential of the composites. The coatings showed a controlled release of Cu(II) ions, which maximum laid below the WHO regulatory limits (25 ppm) [4]. These coatings represent promising candidates for the preparation of bioactive food packaging, with negligible cytotoxicity associated to their use.

[1] M.C. Sportelli et al., TrAC, 84 Part A, 2016, 131. https://doi.org/10.1016/j.trac.2016.05.002
[2] S. Panigrahi et al., Polyhedron, 25, 2006, 1263. https://doi.org/10.1016/j.poly.2005.09.006
[3] M.C. Sportelli et al., ChemEuroJ, 29, 2023, e202203510. https://doi.org/10.1002/chem.202203510
[4] EFSA, "Revision of the currently authorized maximum copper content in complete feed", 2016 https://www.efsa.europa.eu/it/efsajournal/pub/4563.

PS1-09 Advanced method for simultaneous determination of Pb, Al, and Fe using HR-CS GF-AAS for the analysis of Antarctic moss and lichens

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High-resolution continuum source AAS offers some major advantages over traditional line-source AAS. As a narrow spectral interval around the monitored spectral line is observed, any adjacent spectral lines can also be monitored. While current instruments are not primarily designed for multielemental measurement, in recent years many methods for monitoring multiple analytes in one measurement have been published. As measuring using GF-AAS is relatively time-consuming, detecting more than one element in a single measurement is a highly appealing feature.

Here, we present an advanced method for the simultaneous measurement of lead, iron, and aluminum in a single shot. As the relative sensitivities of observed spectral lines vary substantially, the working range for the elements also varies accordingly. The method includes some advanced features, such as: a) slow ramp of atomization temperature to obtain the highest possible sensitivity of Pb and b) detecting Al using its two lines, which enables stretching the dynamic range over more than four orders of magnitude, which is not quite usual for AAS.

The method has been successfully used in the analysis of plant material, specifically Antarctic lichens and mosses from Nelson Island, Antarctica. The working range proved to be appropriate with respect to the content of the elements in these specimens.

PS1-10 Assessing Environmental Damage in Parchment by MALDI MS, ATR/FTIR & Raman Imaging

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Exposure to environmental pollutants puts parchment at risk of degradation and damage. UV light, humidity, and atmospheric pollutants constantly interfere with its surface, changing underlying molecular structures. This plays a crucial role especially for the preservation of cultural heritage objects. Parchment is made from animal skin and consists of many different compound classes, including proteins (especially collagens) and lipids. Different types of collagens are the main protein fraction. Sheep parchment in particular is also characterized by an inherently higher lipid content due to cutaneous lipids secreted. Oxidative stress plays an essential role in the structural and chemical modification of both, collagens and lipids. Here, we present the combination of ATR/FTIR & Raman imaging, with mass spectrometric imaging (MSI) to identify and study degradation of parchment after UV/VIS, humidity and SO2 exposure with a special focus on the distribution of protein and lipid modifications.

Hair/flesh sides of parchment were UV/Vis, humidity and SO2-aged under laboratory conditions for 4 weeks. Imaging experiments were performed on the sample surfaces, the hair and flesh side, and on cryo-sections after embedding. Spectroscopic analysis was performed on a LUMOS FTIR microscope (Bruker Optics) and a confocal Raman microscope (Horiba). MS imaging was performed by MALDI TOF

analysis. To assess lipid information the samples were coated with matrix (1,5 DAN). For in depth study of protein changes an "on-parchment" digestion with different enzymes was performed, before subsequent MALDI matrix application and MSI analysis (Bruker).

ATR/FTIR and Raman spectroscopy provided information on protein degradation of collagens. By utilizing the second derivatives of amide I, II, and III absorption bands, it was possible to trace the stepwise loss of the collagens' ordered structures. The light-induced degradation of lipids was studied via the CH-stretching bands.

This study assesses for the first-time also protein and lipid oxidation in parchment by MSI and correlates the results with spectral imaging data; horizontal and vertical changes in the parchment specimen were of interest. In-solution enzymatic digestion protocols for trypsin/LysC and collagenase III were optimized using collagen standards and optimum conditions were transferred to "on-parchment" digestion. The workflow allows the laterally resolved assessment of degradation-induced alteration by studying peptide modifications. Proteomic analysis of collagen was complemented by applying lipid investigation before and after light exposure. It enabled the identification of lipids present after the manufacturing process and the assessment of oxidation products.

PS1-11 Nanoparticles as a new tool to diagnose ischemic stroke

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Ischemic stroke (IS) was identified as the second most common cause of death worldwide in 2019 according to a survey [1]. A large percentage of patients remain with permanent consequences that can persist even fifteen years after the event [2]. This is also associated with economic impacts on healthcare and supportive services, which amounted to approximately 60 billion euros in the EU in 2017 [3]. The key to success is time. IS can already be precisely diagnosed nowadays. However, in each specific situation, there is a problem with detecting the onset of stroke, which is crucial for choosing the appropriate treatment and thus saving lives. Today's commonly used medical scanning devices can only visualize clots with high erythrocyte content. Other forms of thrombi become very difficult to recognize [4]. Polyiodinated biodegradable nanoparticles (IoNPs) have the potential to solve this problem. The current preclinical phase of our project is focused on researching the pharmacokinetics of these potential IoNPs theranostics in the body. A unique method LA-ICP-MS is used to determine the IoNPs distribution.

The study was supported by the Grant Agency of Masaryk University – Excellent diploma thesis, MUNI/C/0002/2023.
[1] Global Burdens of Disease – GBD Results. https://vizhub.healthdata.org/gbd-results/. (visited 9/1/2023)
[2] Gallacher, K. I. et al. Stroke, multimorbidity and polypharmacy in a nationally representative sample of 1,424,378 patients in Scotland: Implications for treatment burden. BMC Med 12, 1–9 (2014).
DOI: 10.1186/s12916-014-0151-0
[3] Luengo-Fernandez, R., Violato, M., Candio, P. & Leal, J. Economic burden of stroke across Europe: A population-based cost analysis. Eur Stroke J 5, 17–25 (2020).
DOI: 10.1177/2396987319883160
[4] Kim, J., Park, J. E., Nahrendorf, M. & Kim, D. E. Direct Thrombus Imaging in Stroke. J Stroke 18, 286–296 (2016).

DOI: 10.5853/jos.2016.00906

PS1-12 Fentanyl Specific Sensor using a Molecularly Imprinted Polymer

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This study presents, through a theoretical-experimental approach, the creation of a cheap and easy-to-handle material, Molecularly Imprinted Polymers (MIP), as a recognition phase in a sensor for selective and sensitive identification of the fentanyl1. It is known that the choice of reagents for the synthesis of MIP is far from being trivial and may require expenditure of reagents and resources. Therefore, Density Functional Theory (DFT) were used to screening and select the most suitable functional/structural monomer (FM) and solvent before the MIP synthesis2-5. After optimizations by DFT, the interaction energies for each complex were calculated through the equation ΔE = ($Efentanyl$+$Emonomer$)−($EComplex$), after the average between the set of fentanyl complexes was used with a given FM obtaining graphs of the interaction energy in order to understand whether or not the interaction is thermodynamically favorable. Futhermore, by calculating the energy difference between the highest occupied molecular orbital (HOMO) and the lowest unoccupied molecular orbital (LUMO), a graph representing the gap energy was generated and analyzed. Through these factors, it can be concluded which FM may be the most suitable for the synthesis of a MIP for fentanyl. FM such as acrylonitrile, aniline and acrylic acid are promising choices. To further improve the functionality of the MIPS for fentanyl, we propose to create magnetic MIPS by incorporating a magnetite (Fe3O4) core into the synthesis process. This will be achieved by silanizing the magnetite core with tetraethylorthosilicate and then using 3-methacryloxypropyltrimethyloxysilane to provide the activated C=C groups to further copolymerize the selective sites on top of the core using ethyleneglycoldimethacrylate and azoisobutyronitrile. The resulting magnetic MIPS will possess excellent selectivity and high binding affinity towards fentanyl, which can be experimentally validated through SEM, XRD, and FTIR analysis. To avoid using fentanyl in the MIPS synthesis process, we will use a synthesized equivalent called n-phenylpropionamide, which has a similar chemical structure to fentanyl with a phenyl group attached to a propionamide moiety. N-phenylpropionamide can mimic the interactions between fentanyl and the selected FM, making it a suitable alternative for use in MIP synthesis and characterization. Overall, the incorporation of a magnetite core in the MIPS synthesis process will enhance the selectivity and sensitivity of the resulting magnetic MIPS towards fentanyl, while the use of n-phenylpropionamide as an alternative to fentanyl will enable the characterization of the MIPS without the need for the controlled handling of a highly potent and addictive substance.

[1] Jones, A. A., Schneider, K. E., Mahlobo, C. T., Maggs, J. L., Dayton, L., Tobin, K. E., & Latkin, C. A. Psych. of Addic. Beha., 37(2), 191–198, 2023. DOI: 10.1037/adb0000834
[2] F. Neese, Wiley Inter. Reviews: Comp. Mol. Sci., vol. 8, 2018. DOI: 10.1002/wcms.1327
[3] Yu Takano, and Houk. K. N.; Jour. of Chem. Theo. and Comp. 1.1 70-77, 2005. DOI: 10.1021/ct049977a
[4] C. Lee, W. Yang, and R. G. Parr," Phys. Ver. B, vol. 37, no. 2, p. 785, 1988. DOI: 10.1103/PhysRevB.37.785
[5] A. Schäfer, C. Huber, and R. Ahlrichs. The Jour. of Chem. Phys, 100(8):5829–5835, 1994. DOI: 10.1063/1.467146

PS1-13 The comparison of MIP-based sensors developed for the detection of antiviral drugs with quantum chemical calculations

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Abstract

Antiviral drugs such as ribavirin (RIB) and molnupiravir (MOL) are frequently used to treat viral infections. RIB, known as virazol or 1-f8-d-ribofuranosyl-1,2,4-triazole-3-carboxamide, was first synthesized in 1972. It is a pyrimidine nucleoside designed as an in vitro broad-spectrum agent. In addition, RIB is a molecule with antiviral activity against different viruses and has been frequently used for the treatment of the hepatitis C virus in clinical practice [1]. MOL, known as N-hydroxy-5'-O-isobutyryl-3,4-dihydrocytidine[(2R,3S,4R,5R)-3,4-dihydroxy-5-[4(hydroxyamino)-2-oxopyrimidin-1-yl]oxolan-2-l]methyl-2-methylpropano ate,is an oral broad-spectrum antiviral agent originally designed for the treatment of Alphavirus infections. Recently, it has been utilized to treat COVID-19 disease, which is brought on by the nasopharyngeal SARS-CoV-2 infectious viruses [2,3]. In this study, different molecularly imprinted polymers (MIP)-based electropolymerization (EP) and photopolymerization (PP) techniques were investigated in detail for the selective and sensitive detection of RIB and MOL. 3-thiophene boronic acid (3-TBA) and para-aminophenyl boronic acid (p-APBA) as functional monomers for the EP techniques, methacryloyl guanidine (Gu-MA) and metacryloyl uracil (Ura-MA) as functional monomers for the PP techniques were designed and used for the selective and sensitive detection of RIB and MOL. The developed MIP sensors using EP were carried out by directly co-polymerization of 3-TBA and p-APBA monomer with pyrrole (Py) in the presence of target molecule RIB and MOL on a glassy carbon electrode (GCE) surface. The reusability and real sample applicability of the developed sensors depend on the correct determination of the optimization parameters. For this, the experimentally obtained data were supported by quantum chemical calculations. The optimized structure and electrostatic potential charge distribution were obtained by quantum chemical calculations considering the structures of the RIB, MOL, 3-TBA, p-APBA, Py, Gu-MA, and Ura-MA molecules. On the other hand, Py docking calculations of RIB-p-APBA and MOL-3-TBA pairs showed that up to 5 Py molecules could be attached to the structure, and it was found to be compatible with the experimental data obtained. In addition, the optimum template: monomer ratios for RIB-Ura-MA and MOL-Gu-MA were calculated as 1:1 for both sensors. As a result, the applicability of sensors developed using different MIP-based electrochemical methods was supported by quantum chemical calculations.

Acknowledgments

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[1] R.W. Sidwell, J.H. Huffman, G.P. Khare, L.B. Allen, J.T. Witkowski, R.K. Robins Broad-spectrum antiviral activity of Virazole: 1-beta-d-ribofuranosyl-1,2,4-triazole-3-carboxamide, Science, 177 (1972), pp. 705-706.

[2] F. Pourkarim, S. Pourtaghi-Anvarian, H. Rezaee. Molnupiravir: A new candidate for COVID-19 treatment, Pharmacol. Res. Perspect., 10 (2022), p. e00909.

[3] G.R. Painter, M.G. Natchus, O. Cohen, W. Holman, W.P. Painter. Developing a direct acting, orally available antiviral agent in a pandemic: the evolution of molnupiravir as a potential treatment for COVID-19, Curr. Opin. Virol., 50 (2021), pp. 17-22.

PS1-14 Au Metallized Polyethylene Terephthalate (PET) by Supercritical CO2-assisted Metallization toward Flexible Electrochemical Biosensors

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Flexible catalytic electrode for oxidation of biomolecules was realized by a supercritical carbon dioxide (scCO2)-assisted functionalization process [1]. The flexible catalytic electrode was an Au/Ni-P/polyethylene terephthalate (PET) composite. ScCO2 was used as the solvent in the catalyzation step of an electroless plating process. Palladium bis-hexafluoroacetylacetonate was used as the source of the Pt catalyst for the high solubility in scCO2. After the catalyzation step, Ni-P was firstly deposited on the catalyzed PET as the sacrificial layer for the later Au deposition. Electrical resistance of the Ni-P/PET composite was 0.27 Ω and maintained at 0.30 Ω after a tape adhesion test, which revealed the positive contribution of the scCO2 catalyzation on reliability of the metallized PET. After deposition of the Au layer, the flexible Au/Ni-P/PET composite was evaluated as the catalytic electrode in oxidation of urea, ascorbic acid and glucose to demonstrate the applicability in flexible biosensors. After introducing the biomolecule into the PBS solution (25 µM for urea and 2.5 mM for ascorbic acid and glucose), a sudden increase in the current density was observed. The increase of the current response represented the current generated from the oxidation reaction of these three biomolecules. The current density increased again after adjustment of the concentration to a higher value (50 and 75 µM for urea and 5.0 and 7.5 mM for ascorbic acid and glucose). In the oxidation of these three biomolecules, the current density increased immediately after introducing the biomolecules into the PBS solution, and then the current density gradually reduced to a steady-state level. The steady-state level was higher than the steady-state level at a lower concentration of the biomolecules. The gradual decrease in the current density is suggested to be caused by products produced from the oxidation reaction that adsorb on surface of the electrode and interfere with the main reaction. Nevertheless, the overall current density was higher when a higher concentration of the specific biomolecule was introduced into the PBS solution, which revealed the catalytic activity of the flexible Au/Ni-P/PET composite electrode in the oxidation reaction. Regarding the reliability of the Au/Ni-P/PET composite, after the tape adhesion test, the composite electrode still showed an obvious increase in the current density along with an increase in the glucose concentration. The result confirmed the reliability and intactness of the Au/Ni-P/PET composite toward flexible electrochemical biosensors.

[1] https://doi.org/10.1016/j.supflu.2021.105455

PS1-15 Sensitive detection of patulin in water and apple juice samples

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Research and development of new control technologies are essential given the importance of the cytotoxic effects associated with elevated levels of patulin. Therefore, an alternative viewpoint on patulin determination is absolutely essential. The electroanalysis of patulin was carried out employing a modified 2,3,7,8,12,13,17,18-octaethyl-21H,23H-porphine manganese (III) chloride (Mn(TPP)Cl) screen-printed carbon electrode through the application of a square-wave voltammetry technique. Several experimental parameters were investigated and optimized, including scan rate, buffer solution type, and pH. A linear concentration dependence was established in the range of 1.00×10-12 – 1.00×10-8 mol L-1. The quantification limit was determined to be 1.00×10-12 mol L-1, and the detection limit was found to be 3.33×10-13 mol L-1. The electroanalysis of patulin in water and apple juice samples resulted in a high level of reliability.

PS1-16 A novel time-dependent potentiometric glucose biosensor

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Glucose sensing has been on the frontline of biosensors for more than six decades, when the first glucose monitoring system was reported by Clark & Lyons in 1962. [1] Since then, thousands of such sensors have been reported, invasive or non-invasive, enzymatic or non-enzymatic, disposable or not, targeting blood or other bodily fluids, like sweat and urine. Considering the plethora of glucose studies reported in the literature, the remaining question is whether there is still room for improvement in the field. A considerable number of the reported sensors are enzymatic and based on electroanalytical techniques, like amperometry. Among them, the ones that have been commercialised use a mediator acting as an electron carrier (e.g. ferricyanide) between the electrode and enzyme. Also, they are typically screen-printed, which gives them the advantage of low cost to the detriment of accuracy. [2]
We report here a novel method for monitoring glucose based on a time-dependent response. The principle of measuring is based on a two-step process that involves the oxidation of a mediator and the subsequent monitoring of the open-circuit potential over time. This process enables one to record a time-dependent response that is a function of the level of glucose in contact with the sensor. As glucose is oxidised by the enzyme, the generated electrons gradually reduce the oxidised mediator manifesting a change in the open-circuit potential, the rate of which depends on the glucose levels. The readout is similar to that of chronopotentiometry where a transition time, rather than an electrochemical signal -like current- is monitored. The sensing membrane employed is based on ferrocene-modified poly(ethylenimine) used as mediator and glucose oxidase. In addition, a diffusion limiting membrane enables the steady-state diffusion of glucose towards the electrode and can be used to fine tune the selectivity, which is currently under examination.
This simple monitoring protocol requires low-power consumption and results in better reproducibility compared to the classic amperometric approach used for glucose sensing. The additional advantage of the specific approach lies in its universality, which makes possible the development of similar time-depended sensors for other analytes that we are currently investigating.

[1] L. C. Clark & C. Lyons, Ann. N. Y. Acad. Sci. 102 (1962) 29– 45. DOI: 10.1111/j.1749-6632.1962.tb13623.x
[2] E. Witkowska Nery, et al., Anal. Chem. 88 (2016) 11271–11282. DOI: 10.1021/acs.analchem.6b03151

[1] L. C. Clark & C. Lyons, Ann. N. Y. Acad. Sci. 102 (1962) 29– 45. DOI: 10.1111/j.1749-6632.1962.tb13623.x
[2] E. Witkowska Nery, et al., Anal. Chem. 88 (2016) 11271–11282. DOI: 10.1021/acs.analchem.6b03151

PS1-17 Nanostructured Zn doped TiO2 - carbon paste sensor for electrochemical determination of ofloxacin in water

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A simple, low-cost, fast response, and a highly sensitive potentiometric sensor for determining ofloxacin in wastewater. The Zn-doped TiO2 nanoparticles nanopowder has been synthesized via the sol-gel method and characterized by different analysis tools like FTIR, XRD, TEM, and SEM with EDX have been employed as a sensor. This work introduces an elegant modification of carbon paste electrode (CPE) by incorporation of zinc-doped TiO2 nanoparticles, All cyclic voltammetric and

potentiometric measurements was carried out by the modified CPE sensor as a working electrode vs. Ag/AgCl between 0 to 1200 mV in solutions of pH range (2.0 -10.0) containing a phosphate buffer solution (PBS). The highest potentiometric response was produced by a paste (20% and 80% graphite carbon, respectively which enhanced the electrode response with accurate results and better limit of detection (LOD) and the limit of quantification (LOQ). The results measurements by nano-zinc doped TiO2 modified CPE sensor gave a limit of detection of 0.00584 mM (5.8x10-6 M) and a limit of quantification of 0.0149 mM (1.49x10-5 M) and determination of different concentrations with simple and excellent accuracy exceeded 97.2% with instantaneous measurements within 2.0 seconds and long lifetime of stable response for about 10 weeks.). The developed sensor displayed a Nernstian response in the concentration range of 1.41 105 mol L-1 to 1 102 mol L-1, with a slope of 29.93 0.1 mV per. According to these positive results, the nano-zinc doped TiO2 modified CPE sensor was applied in potentiometric determinations of ofloxacin in some formulated drugs and also examination in different brands of the drug with high accuracy and 99.9% recovery. The good recoveries and low standard deviations obtained indicated the high accuracy and precision of the proposed method.

Zinc-doped TiO2 nanoparticles ,Cyclic voltammetric measurements, detection, modification, ofloxacin, potentiometeric sensor, quantification.

PS1-18 Separation-Free Enzyme-Immunosensor with Magnetic-Field-Driven Accumulation of Immunocomplexes and Pulstrode Delivery of Substrate

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Heterogeneous enzyme-immunoassays require labor intensive manipulation (washing steps) to remove unbound enzyme-labeled detection antibodies that would otherwise interfere with the output signal. If exposure of enzyme substrate to the unbound enzyme-labelled antibody in the bulk solution could be avoided by confining the substrate to the binding site, such assays could be greatly simplified. It has been shown before that an electrochemical excitation pulse can be used to selectively deliver the enzyme substrate through an ion-selective membrane (ISM), which also serves to monitor the enzyme kinetic cycle over time by zero-current chronopotentiometry after the release pulse [1]. However, direct biofunctionalization of polymeric ISMs with capture bioreceptors does not allow the reuse of those membranes for more than one assay, which is not ideal for practical operation. As an alternative, the capture antibodies can be immobilized on the surface of dispersible probes such as magnetic beads (MBs), which are attracted from the bulk solution to the membrane surface by a magnetic field to perform the measurement, and then released back to the solution [2].

Herein, we propose an integrated sensing system for enzyme-linked immunoassays, where the dispersible magnetic probe containing a sandwich-type enzyme-immunocomplex is spatially resolved from the excess detection conjugate in the bulk solution by applying a magnetic field. The streptavidin-modified magnetic beads bind to the biotinylated capture antibody, becoming a magnetic capture probe. After binding to the target analyte, a sandwich is allowed to form with the enzyme-labeled detection antibody. Once the beads are on the surface of the ion selective membrane, a well-established pulstrode protocol is used to deliver the enzyme substrate from the back side inner solution to the immunocomplexes side. Initially, the open circuit potential of the system is observed and confirmed to be stable with time. Subsequently, a controlled anodic current pulse of constant

amplitude and duration is applied, releasing a known quantity of enzyme substrate (choline). In the presence of the enzyme-linked immunocomplex on the surface of the ISM, the choline ions instrumentally delivered from the membrane are now partially consumed by the choline oxidase enzyme label. The potential response changes with time since the membrane is selective to choline activity and is proportional to the concentration of enzyme-immunocomplex, which allows the quantification of the target analyte. This integrated electrochemical immunosensor comprises the immunobinding, enzyme reaction, and electrochemical detection all occurring in the same system without any washing step, controlled by electrochemistry and magnetic forces.

[1] G. J. Mattos and E. Bakker, Biosens. Bioelectron. (2023) 100351.
DOI: 10.1016/j.biosx.2023.100351
[2] W. Qin, et al., Angew. Chem. 60 (2021) 2609.
DOI: 10.1002/anie.202011331

PS1-19 Visual detection of microRNAs from urine samples using a lateral flow strip

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MicroRNAs or miRNAs are considered as novel biomarkers that can be exploited in liquid biopsy applications. Liquid biopsy involves the collection of samples from various body fluids such as blood, saliva, urine, etc. As biomarkers for such applications, miRNAs, circulating tumor DNA and cells, as well as exosomes, are the preferred analytes. The detection of miRNA is particularly challenging due to its very small size (18-24 nt). Several methods have been already reported for miRNAs analysis. These methods include exponential amplification (PCR or isothermal), DNA sequencing and microarrays. A great number of reports, however, focus on biosensors that have been developed to overcome the cost, the long analysis time, the extensive sample preparation and the expensive instruments of the forementioned techniques. Nanotechnology has been greatly exploited in these biosensors to increase the produced signal. Many sophisticated signal amplification techniques have also been reported for miRNA analysis. In this project, we have developed a lateral flow strip assay for the detection of two miRNAs, miR-21 and miR-let-7a, in urine samples. The method is based on the amplification of miRNAs by reverse transcription – polymerase chain reaction (RT-PCR) using stem-loop specific primers. The amplified products were then detected by the lateral flow strip using gold nanoparticles as reporters. As low as 10^2-10^3 copies of miR-21 and 10^2-10^4copies of miR-let-7a spiked in urine were detectable by the proposed lateral flow strip. The proposed strip is very simple, provides fast analysis time, has good detectability and very good repeatability (%CVs < 4.5%), while with slight modifications of the oligonucleotides used, it can be applied for the detection of any other miRNA sequence.

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PS1-20 Fe2+/Fe3+ in internal solution of classical ISEs: prospects for the use in non-zero current modes

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Inspired by E. Bakker's voltammetry with solid-contact ISEs containing several ionophores, we tried to make voltammetric measurements possible for classically designed ISEs with internal solution and internal reference electrode. Classical ISEs are attractive due to their long lifetime and excellent piece-to-piece reproducibility of their potentials. We will report on the possibility to obtain voltammetric response which provide information on the activity of an analyte: in analogous to those described by E. Bakker's group [1-3]. The transition to classical ISEs with relatively thick membranes allowed to increase the lifetime of voltammetric ISEs: up to the moment of this abstract submission, they have demonstrated an operating period of three months. Their potentiometric, voltammetric behavior during this period will be presented.

To obtain a voltammetric response, we use Fe2+/Fe3+ as a part of the internal filling solution and platinum internal reference electrode. On the example of Ca-, Li- and K-ISEs we will show a voltammetric response of the ISEs constructed in such a manner, with a Nernstian potential shift of the oxidation and reduction peaks. We will also discuss how water sorption by ISE membranes, studied in detail recently by our group [4, 5], influences the voltametric curves. Although such voltammetric experiments have been successfully carried out with membranes containing only one ionophore, we believe that this is a useful intermediate result obtained for the first time for classical ISEs.

Moreover, it will be shown that such ISEs containing a redox system inside can appear surprisingly useful for chronoamperometric/coulometric measurements: it is possible to obtain the signal in a rather short time and use it for building a nice calibration without any capacitor connected in series with the ISE. According to our data, the latter is not possible with classical ISEs without such redox system inside and not connected to a capacitor. At the same time, measurements with ISEs without electronic capacitors allow to avoid measurement errors associated with capacitor leakage currents. On the other hand, the use of a capacitor in series to the classical ISEs can provide with their own benefits: the capacitor can be recharged independently of the ISE itself, and this allows to replicate peaks originated from a single concentration change. Such a signal replication can improve the accuracy and reliability of the analysis, and this possibility will be also presented on the example of ISEs containing Fe2+/Fe3+ in internal solution and platinum internal electrode.

1. G. Crespo, et al., Anal. Chem. 87 (2015) 7729. DOI: 10.1021/acs.analchem.5b01459
2. M. Cuartero, et al., Anal. Chem. 88 (2016) 1654. DOI: 10.1021/acs.analchem.5b03611
3. M. Cuartero, et al., Anal. Chem. 88 (2016) 5649. DOI: 10.1021/acs.analchem.6b01085
4. E. Solovyeva, et al., J. Membr. Science 619 (2020) 118798. DOI: 10.1016/j.memsci.2020.118798
5. V. Keresten, et al., Membranes 11 (2021) 1050344. DOI: 10.3390/membranes11050344

PS1-21 In-Situ Formation of a Solid-State Ag/AgCl Reference Membrane Using Intense Pulsed Light Photoreduction

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Miniaturized solid-state electrochemical sensor platforms enable gathering information unreachable to classical electroanalytical protocols based on bulky electrodes with liquid compartments. Consequently, they are promising for integration into wearable devices, agricultural and environmental uses, and industrial process monitoring. High demand for easily accessible chemical information requires a fast, low-cost, and ecologically friendly production technique. Based on digitally controlled selective ejection of a functional material, inkjet printing is becoming popular [1].
Accuracy, reliability, and precision of electrochemical sensor platforms are dictated by the stability of the solid-state reference electrode (SSRE). Miniaturized SSREs are mostly based on the Ag/AgCl pair, produced by chlorinating a silver layer deposited on a substrate material and protected from dissolution with a polymer-based membrane doped with chloride ions [2].

Presented here is the development of an inkjet printable solution for the in-situ formation of an Ag/AgCl SSRE. A previously reported poly(vinyl butyral) (PVB) based reference membrane prepared from an oversaturated AgCl methanol solution can be deposited on any electrically conducting material [3]. However, methanol's physical characteristics and nozzle blocking due to the AgCl precipitate formation make it unsuitable for inkjet printing.

We have prepared two separate inkjet printable solutions of $AgNO_3$ and NaCl in a solvent mixture of methanol, butanol, and ethylene glycol. Alternately drop-casting the two solutions, AgCl forms as a white precipitate on the glassy carbon electrode surface. To obtain Ag-AgCl clusters responsible for the potential stability of the SSRE, we replaced time-consuming light bulb photoreduction with a significantly faster high-intensity intense pulsed light (IPL) treatment. IPL can be in-line coupled with the inkjet printing process, making this a promising mass production protocol [4]. The Ag-AgCl modified glassy carbon electrode surface was covered with a PVB-protective membrane and the electrode potential stability was measured in chloride ion containing solutions. The IPL parameters, including voltage and energy of the incident light, were optimized. Finally, the same procedure was transferred to in-house inkjet printed Ag-electrodes on polyimide substrates. The electrodes were stable in the 10–6 to 10–3 M concentration range of KCl, with a drift of 1.04 mV/h. The prepared SSREs were further electrochemically characterized by the means of potentiometry, cyclic voltammetry and electrochemical impedance spectroscopy.

[1] A. Moya, et al., Curr. Opin. Electrochem. 3 (2017) 29. DOI: 10.1016/j.coelec.2017.05.003.
[2] E. Zdrachek, et al., Anal. Chem. 93 (2021) 72. DOI: 10.1021/acs.analchem.0c04249.
[3] T. Guinovart, et al., Anal. Chim. Acta 821 (2014) 72. DOI: 10.1016/j.aca.2014.02.028.
[4] H.S. Lim, et al., J. Mater. Chem. C. 5 (2017) 7142. DOI: 10.1039/C7TC01848G.

PS1-22 Planar reference electrodes based on ionic liquids

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Miniaturization is undeniably one of the most notable trends in modern chemical analysis. Particular interest is directed towards simple miniature sensors, enabling rapid analysis of samples by an inexperienced user. Potentiometric sensors have a very simple construction, are inexpensive and can be easily scaled up for mass production. In this type of sensor at least two miniature electrodes are needed: an indicator electrode and a reference electrode. While the miniaturization of indicator electrodes has been deeply investigated, little attention has been paid to reference electrodes, which

are essential for the sensor. To meet the need for planar reference electrodes, we decided to develop reference electrodes with a polymer membrane based on ionic liquids.

During study we were able to examine a number of ionic liquids that could find potential application in reference electrodes. In the first stage, we studied the change in EMF of classical electrodes containing ionic liquids in solutions with different concentrations of highly lipophilic perchlorate anions and tetramethylammonium cations. The electrodes exhibited very different behavior in these solutions, supporting the hypothesis that the structure of the ionic liquid strongly affects the stability of the reference electrode potential. The electrodes were also similarly tested for potential analytes (lactate and pH) and their long-term stability was examined. Electrodes containing different amounts of ionic liquid were also tested, to demonstrate that the ionic liquid content has no significant effect on the potential stability of the electrodes, only on the potential value.

Based on preliminary results, the most promising ionic liquid was selected and planar electrodes with a polymer membrane containing this substance were prepared and tested. Reference electrodes were also tested on a screen-printed multi-electrode transducer designed for future sensor. Tests of electrodes on multi-electrode transducers with membranes applied not manually, but automatically by the aerosol jet printing (AJP) method were also performed.

Based on the results, further studies are planned, including tests on an integrated sensor consisting of a reference electrode and several indicator electrodes (lactate and pH). Ultimately, a device will be developed for sweat analysis and detection of anaerobic threshold during exercise.

C. Bieg et al., Anal. Bioanal. Chem. 409 (2017) 45-61.
J. Kuczak et al., Sens. Actuators B Chem. 357 (2022) 131446.

PS1-23 Development of On-site Applicable Fluorescent Probe for Fire Blight

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Fire blight is an infectious plant disease with Erwinia amylovora as the pathogen. It hosts the Rosaceae family such as apples and pears and has symptoms of tissue darkening and drying out across all plant organs. Because there is no clear cure method for fire blight and the pathogen spreads rapidly, fire blight has globally brought a burden on the agricultural economy. Therefore, it is essential to prevent the spread of fire blight by diagnosing it in the early stage. However, current diagnostic methods rely on PCR and immune-strip, which require specialized equipment and experts and have limitations of low sensitivity to early diagnosis.

In this study, we disclosed a novel fire blight diagnosis platform based on fluorescence. We secured a lead compound, B-1, by performing confocal laser microscope imaging with dye libraries and a plant-based practical application. E. amylovora-specific fluorophore B-1 isn't fluorescent in an aqueous solution, but it emits red fluorescence (686 nm) with E. amylovora. B-1 is suitable for early diagnosis of fire blight by detecting E. amylovora within 10 seconds and at least 10^2 CFU/mL. It also shows that the diagnosis of fire blight is not limited to the experimental setting by efficiently detecting E. amylovora present in apple tree organs. We expect that this study will contribute to diagnosing various plant diseases beyond fire blight.

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PS1-24 A Novel Fluorescent Complex for Targeting Human Glioblastoma, Consisting of Dipolar Dye, Caveolin-Targeting Peptide, and Serum Albumin Proteins

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Glioblastoma (GBM) is a malignant disease with high patient mortality and recurrence rates. When performing surgical procedures, it is important to completely remove part of the tumor while preserving critical brain regions. To increase the accuracy of such surgeries, the use of fluorescence probes for resection is highly beneficial. However, there are difficulties in using fluorescence probes for fluorescence-guided surgery (FGS) in GBM due to the characteristics of the tumor. i) GBM is a conglomerate of tumor cells, so there is a possibility that the fluorescent agent may not uniformly penetrate the entire tumor. ii) the fluorescent agent must only emit light within the tumor to clearly distinguish the boundary between the tumor and surrounding tissue. iii), the technology should be highly selective for the tumor, as fluorescence reactions can occur in healthy tissue due to the nature of the fluorescent agent. To overcome these challenges, we have developed a GBM-homing hybrid complex (BSA-OXN-SIWV). This complex is composed of a fluorophore, a peptide, and a protein. Oxazepine (OXN) is a highly stable dipolar fluorophore that is responsible for visualizing brain tumors. The tetra-peptide SIWV (Ser-Ile-Trp-Val) and the protein albumin target caveolin-1 and gp60/SPARC, respectively, which are overexpressed in GBM, helping the complex to specifically target the brain tumor. The optical properties and molecular docking mode of BSA-OXN-SIWV were analyzed, and its stability and imaging of clinical GBM tissues were demonstrated. Our research results suggest the potential application of BSA-OXN-SIWV as a new FGS agent for surgery.

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PS1-25 Application of miniaturized solid-phase microextraction coupled with gas chromatography-mass spectrometry for determination food additives in beverages

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Food additives are synthetic chemical or natural substances that are used to impart certain qualities to wine products, such as taste, texture, color, smell, shelf life, appearance. Initially, natural ingredients made from natural raw materials were used as additives. With the development of the chemical industry, food additives began to be produced artificially. They began to produce such synthetic additives as dyes, preservatives, thickeners, stabilizers, antioxidants [1].

Propylene glycol, benzoic and sorbic acids have been identified as important food additives and contaminants to control in beverages, including juices, wines and spirits. Benzoic and sorbic acids are

used in wine products primarily to control microbial growth [2], while propylene glycol is used to mask artificially added flavors and dissolve colorants [3]. Benzoic acid is considered the most active against yeast and mold and the least active against bacteria [4]. Excessive amounts of these components pose a serious danger to consumers and must be strictly controlled.

High performance liquid chromatography is widely used for the analysis of sorbic and benzoic acid in foods and beverages [5]. In addition, since traditional gas chromatography methods require complex pre-treatment, i.e. multiple extraction, evaporation and derivatization steps can reduce the analytical accuracy of gas chromatography. Several new sample pretreatment methods, such as solid phase extraction, solid phase microextraction and stirred sorption extraction, have recently been introduced as a pretreatment procedure in analysis [6].

Propylene glycol, benzoic and sorbic acids were first identified in this work using miniaturized solid phase microextraction combined with gas chromatography-mass spectrometry. The components were extracted from the gas phase using a miniaturized solid phase microextraction process, and key parameters such as extraction temperature and time, preincubation time, solvent addition, and sample volume were optimized.

The proposed approach uses 2 ml vials, in contrast to the solid phase microextraction method, which requires only 100 µl of the target analyte. The time required for the liquid phase and fiber to reach equilibrium is drastically reduced when using miniature solid phase microextraction, which increases the overall accuracy of the study. . The proposed sample preparation method does not require the use of hazardous organic solvents, which makes it an environmentally friendly method of analysis. This study was carried out under the project "Effective development of highly sensitive methods of analysis of food based on miniaturized solid-phase microextraction" and funded by the Ministry of Science and Higher Education of the Kazakhstan Republic (Grant no. AP09058561).

[1] M. M. Silva, et al., Food Colour Additives. 2022 11(3) 379. https://doi.org/10.3390/foods11030379
[2] D. Mischek, et al., Food additives & contaminants. 2012 29(3) 371–382. https://doi.org/10.1080/19440049.2011.643415
[3] M. Momtaz, et al., Foods. 2023 12(1) 199 https://doi.org/10.3390/foods12010199
[4] R. Hazan, et al., Applied and environmental microbiology. 2004 70(8) 4449–4457 https://doi.org/10.1128/AEM.70.8.4449-4457.2004
[5] G. N. Sagandykova, et al., Food chemistry. 2017 220 162–167. https://doi.org/10.1016/j.foodchem.2016.09.164
[6] M. E. I. Badawy, et al., Analytical sciences : the international journal of the Japan Society for Analytical Chemistry. 2022 38(12) 1457–1487. https://doi.org/10.1007/s44211-022-00190-8

PS1-26 Mineral Content of Spanish Commercial Honey Samples

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One aspect of honey that relates its composition to its geographic and botanical origin is its mineral content. The mineral composition of honey can vary depending on the plant species from which the bees collected nectar, as well as the geological characteristics of the region where the plants grow. It also may affect some physicochemical properties of honey and influence its textural and organoleptically properties. Furthermore, the mineral content of honey can have potential health benefits.

An optimized green digestion procedure, in terms of small quantities of sample and reagents employed, was used for the analysis of 65 honey samples originated in Spain, with different geographic and botanical origin. Briefly, 0.5 g honey was mixed with 3 mL nitric acid and 1 mL hydrogen peroxide and digested via microwave digestion. The digestates were diluted to 50 mL, prior to filtration and injection in the ICP-OES. Major elements, Na, Ca, K, Mg, and minor elements Zn, Fe, and Mn were quantified, and results were evaluated via statistical analysis.

Among the honey analysed, carob honey, Ceratonia siliqua, originated in Mallorca, Spain, showed interesting particularities. The content of minerals (mean±SD) increased in the order Mn<Zn<Fe<Mg<Na<Ca<K (1.05±0.30mg/kg Mn, 2.96±1.48 mg/kg Zn, 4.47±3.46 mg/kg Fe, Mg with 108.52±5.35 mg/kg, Na with 199.80±65.97mg/kg, 549.35±74.303 for Ca and 2385.85±135.85 mg/kg for K). The mineral content was related with high electrical conductivity, 1.20±0.24 mS/cm. The amount of minerals in the other samples, of various botanical origins, was lower for all elements (Mg with 46.77±5.76 mg/kg, Na with 43.60±7.78 mg/kg, 92.68±8.69 for Ca and 871.83±90.83 mg/kg for K) Fe, Zn and Mn didn't differ significantly.

Based on these findings, mineral content could serve as a marker for the authenticity of carob honey originating in Mallorca, Spain. However, it should be noted that the electrical conductivity of this honey, which was higher than the limit set by the Codex Alimentarius could be an exception due to the unique characteristics of Mallorca carob honey.

B. Silva, et al., J. Food Compos. Anal 96 (2020) 1-9. DOI:10.1016/j.jfca.2020.103727
S. Valverde et al., J. Food Compos. Anal 119 (2023) DOI:10.1016/j.jfca.2023.105235
T. Liu, et al., Food Chem, 343 (2021) DOI: 10.1016/j.foodchem.2020.128455
P. Šedík et al. J. Food Nutr. Res.59 (3) (2020) 185–192 [Google Scholar]

PS1-27 Determination of Sugar Contents of Some Fruits According to the Degree of Ripening by HPLC-ELSD

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Context: Determination of sugars in food products is a major area of focus in the food science and food industry. The sugar content of a fruit is defined as the sum of monosaccharides and disaccharides. The most abundant mono and disaccharides in fruit products are fructose, glucose, and sucrose, all classified as major sugars.

To evaluate the variation of sugar content depending on the ripening degree of fruits using a simple, novel, rapid, robust and cost-effective HPLC-ELSD method.

All major sugar compounds and fruit samples were prepared in HPLC grade water. All solutions were prepared daily by diluting stock solutions with the mobile phase. Chromatographic separations were carried out on a Phenomenex Luna NH2 (5 µm particle size, 250 mm x 4.6 mm id, 100 Å) column with isocratic elution with acetonitrile:water mixture. ELSD drift tube temperature was set at 40 °C and N2 gas was adjusted to 350 kPa with Gain:7 and Filter:10 on the ELSD system.

Results: The regression equation revealed a good linear relationship within test ranges for each major sugars. The HPLC-ELSD method was found accurate and obtained RSD% values (lower than 1 %) showed that the precision of the method was good.

The present HPLC-ELSD method was successfully applied to determine the sugar content of fruits and also the sugar contents of fruits with various ripening degrees were compared statistically.

1. Koh, D. W., Park, J. W., Lim, J. H., Yea, M. J., & Bang, D. Y. (2018). A rapid method for simultaneous quantification of 13 sugars and sugar alcohols in food products by UPLC-ELSD. Food chemistry, 240, 694-700.
2. Alghamdi, B. A., Shasho, E., Saad, M., Rawas, G. M., Alharthi, N. T., Baeshen, M. N., ... & Suhail, M. (2020). Analysis of sugar composition and pesticides using HPLC and GC–MS techniques in honey samples collected from Saudi Arabian markets. Saudi J Biol Sci 27: 3720–3726.
3. Lu, Y., Guo, S., Zhang, F., Yan, H., Qian, D. W., Shang, E. X., ... & Duan, J. A. (2021). Nutritional components characterization of goji berries from different regions in China. Journal of Pharmaceutical and Biomedical Analysis, 195, 113859.
4. Montesano, D., Cossignani, L., Giua, L., Urbani, E., Simonetti, M. S., & Blasi, F. (2016). A simple HPLC-ELSD method for sugar analysis in goji berry. Journal of chemistry, 2016.
5. Wang, W., Wang, Y., Chen, F., & Zheng, F. (2021). Comparison of determination of sugar-PMP derivatives by two different stationary phases and two HPLC detectors: C18 vs. amide columns and DAD vs. ELSD. Journal of Food Composition and Analysis, 96, 103715.
6. Soyseven, M., Sezgin, B., & Arli, G. (2022). A novel, rapid and robust HPLC-ELSD method for simultaneous determination of fructose, glucose and sucrose in various food samples: Method development and validation. Journal of Food Composition and Analysis, 107, 104400.

PS1-28 Enhancing Bulgur Production through Artificial Intelligence for Sustainable Food Production

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This paper focuses on improving the bulgur production process through the utilization of an artificial intelligence-supported system, with the ultimate goal of ensuring sustainable food production. Key aspects such as time efficiency, food safety, quality enhancement, energy consumption reduction, and cost minimization are targeted for improvement.

The proposed AI-supported system will analyze data within the bulgur production process, enabling a more efficient production workflow, while simultaneously contributing to improved product quality and reduced environmental impacts. The implementation of low energy consumption techniques and cost-saving measures will significantly contribute to the realization of sustainable food production practices.

The study not only aims to enhance bulgur production through the application of artificial intelligence technologies but also strives to establish a framework that can be extended to other domains within the food production industry to ensure sustainability.

Furthermore, this paper aims to increase efficiency within the bulgur production process. The integration of artificial intelligence technology facilitates accurate and rapid decision-making, thereby enhancing overall production efficiency. Consequently, this will contribute to sustainable food production by increasing both the quantity and quality of the output. In addition to the aforementioned benefits, the study recognizes the importance of regional economic development.

Given that bulgur production holds a prominent position, particularly in Turkey, this study endeavors to bolster the competitiveness of producers, generate employment opportunities, and strengthen the regional economy.

In conclusion, this study represents a significant stride toward achieving a sustainable future from environmental and social perspectives. Through the adoption of an artificial intelligence-supported system, the research aims to optimize bulgur production, minimize environmental impacts, and foster economic growth in the region.

Yılmaz, V. A., & Koca, A. F. (2020). Quality, sensorial and textural properties of einkorn and durum bulgur produced with several methods. International Journal of Gastronomy and Food Science, 22, 100263.
ALIMENTARE, A. (2009). Application of food safety and quality management systems to bulgur processing. Volume XXI Number 4 2009, 21(385), 499.
Taner, T. (2015). Optimisation processes of energy efficiency for a drying plant: A case of study for Turkey. Applied Thermal Engineering, 80, 247-260.
Yağcı, S., Sutay Kocabaş, D., Çalışkan, R., & Özbek, H. N. (2022). Statistical investigation of the bioprocess conditions of alkali combined twin-screw extrusion pretreatment to enhance fractionation and enzymatic hydrolysis of bulgur bran. Journal of the Science of Food and Agriculture, 102(11), 4770-4779.
Bayram, M., Öner, M. D., & Eren, S. (2004). Effect of cooking time and temperature on the dimensions and crease of the wheat kernel during bulgur production. Journal of Food Engineering, 64(1), 43-51.
Bayram, M., Kaya, A., & Oner, M. D. (2003). Color-sorting systems for bulgur production. Cereal foods world, 48(4), 168.
YILMAZ, İ., & SÖYLEMEZ, M. (2020). A novel thermal analysis for cooking process in bulgur production: Design considerations, energy efficiency and wastewater diminution for industrial processes. Isı Bilimi ve Tekniği Dergisi, 40(1), 113-129.
Çetinkaya, Z., & Erdal, E. (2019, September). Daily food demand forecast with artificial neural networks: Kırıkkale University case. In 2019 4th international conference on computer science and engineering (UBMK) (pp. 1-6). IEEE.
Shafaei, S. M., Nourmohamadi-Moghadami, A., Rahmanian-Koushkaki, H., & Kamgar, S. (2019). Neural computing efforts for integrated simulation of ultrasound-assisted hydration kinetics of wheat. Information Processing in Agriculture, 6(3), 357-374.

PS1-29 Method development for the determination of water-soluble vitamins in enteral food with LC-MS

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Vitamins are essential nutrients that must be regularly supplied to the body in small amounts to perform various chemical and physiological functions in the human body [1]. The role of enteral nutrition is to ensure the optimal concentration of vitamins and prevent the risk of malnutrition [2]. The development of a single method for the simultaneous determination of water-soluble vitamins in fortified enteral nutrition is difficult due to the different structures and chemical properties of the vitamin compounds, the trace amounts of vitamins present, the complexity of the matrix, instability to light and heat and solubility problems [3].

The aim of this project is to develop a sensitive LC-MS method for the simultaneous determination of nine water-soluble vitamins, namely, ascorbic acid (C), thiamine (B1), riboflavin (B2), niacin (B3), pantothenic acid (B5), pyridoxine hydrochloride (B6), biotin (B7), folic acid (B9) and cyanocobalamin (B12), respectively. Various solvents and their mixtures have been tested for the extraction of vitamins from enteral food. The best results were obtained with a mixture of acetonitrile and ethanol, followed by protein precipitation. After precipitation, the supernatant was evaporated and reconstituted in 0.1 M TFA. After testing a range of chromatographic columns and gradients, the vitamins were well

separated on Waters Atlantis™ column dC18, 4.6 x 250 mm, 5 µm. The solvents used for the analysis were 0.1% TFA in water (solvent A) and acetonitrile (solvent B). The gradient was applied as follows: 0 min 100% A, 0-13.5 min 100% A-97% A, 13.5-15 min 97% A -85% A, 15-22 min 85% A-80% A, 22-25 min 80% A-0% A, 25-40 min 0% A, 40.1-45 min 100% A. The flow rate was 1.4 mL/min. Samples were analyzed on the Shimadzu LC-MS in single ion monitoring (SIM) mode.

[1] Y. Zhang et al., Molecules 23 (2018) 1484-1509.
[2] F. Martin et al., Journal of AOAC International 99 (2016) 19-25.
[3] E. Goh, Application note: Rapid Analysis of Water-Soluble Vitamins in Infant Formula by Standard-Addition, Waters, 2010

PS1-30 DNA-based detection of olive oil adulteration with other plant oils using a single rapid test

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Olive oil is one of the most prone to adulteration agricultural products due to its sensorial characteristics and high nutritional and health benefits. Olive oil adulteration involves, among others, the substitution of olive oil with other edible plant oils of lower nutritional value, and thus of lower price, for economical profit. Therefore, analytical methods for detection of olive oil adulteration are highly required to protect consumers and producers from fraud. DNA is the preferred analyte due to its high stability, especially in processed samples, and its high specificity for the discrimination of highly related species. We have herein developed a multiplex rapid test for the detection of olive oil adulteration. We used universal (common) forward and reverse primers for PCR amplification of a segment of the rbcl gene from 7 plant oils, i.e., olive, sunflower, soya, corn, sesame, hazelnut, and almond oils. The PCR product was subjected directly (without prior purification) to a genotyping reaction to identify species-specific polymorphisms in each sequence. Then, using a single strip, we were able to discriminate olive oil from 6 other plant oils, including sunflower, soya, corn, sesame, hazelnut, and almond. The detection is visual using gold nanoparticles as reporters to generate red spots on the membrane of the strip. Each red spot corresponds to a specific plant oil, allowing spatial discrimination and simultaneous detection of seven different plant species/oils. As low as 5% of adulteration of olive oil in binary mixtures with hazelnut, sesame, soya and corn were detected by the multiplex strip, while 10% of adulteration was detected for almond and sunflower, all with very good reproducibility. [The authors VM, AM and ES contributed equally to this project].

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PS1-32 Ultrasensitive assay of atrazine in food and water samples

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Atrazine is the pesticide that is found in surface water sources more frequently than any other pesticide, therefore, the paper describes ultrasensitive detection of atrazine from fruit and water samples, using an on-site screening platforms incorporating stochastic microsensors based on graphite powder modified with 4-tert-butylcalix [4] arene and calix[4]arene-25,26,27,28-tetrol. Limits of determination of 0.1 mol/L were recorded while the sensors could have been used on wide concentration ranges. Recoveries higher than 95,00% were recorded when used for the assay of atrazine in water, fruits and vegetables.

PS1-33 Lateral flow assay for DNA-based visual distinction between three important tuna species from tissue samples and heat-processed cooked mixtures

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Tuna in general, is an excellent food product, relatively low in calories, and is recommended for a balanced diet. The continuously increasing demand, especially for bluefin tuna species food preparations in fresh or processed forms and its relatively high market price, makes adulteration by intentionally mixing with other lower-priced tunas and bonitos, more likely. Accordingly, the development of methods to detect tuna adulteration is a challenge in food analytical science. This study aims to develop a simple, fast, and low-cost molecular method for visual detection of BFT (Bluefin tuna species) adulteration. The three species studied were Thunnus thynnus (BFT), Thunnus albacares and Katsuwonus pelamis. The method is based on the construction of a DNA biosensor comprising an immersion pad, a conjugate pad, a membrane, and an absorbent pad. The membrane consists of a control zone, containing biotinylated albumin, and a test zone with immobilized oligo-dTTP. DNA was isolated from fresh and cooked fish samples followed by PCR to amplify sequences of the D-loop region for Thunnus albacares, the Cytb gene for Katsuwonus pelamis and the NADH5 for BFT. The PCR products were hybridized (15 min) with specific, dATP-tailed probes. We prepared gold nanoparticle-antibiotin antibody conjugates for hybrid detection. The hybridized solution and the conjugates were applied to the conjugate pad. The signal was observed in 15 min. The effect of the amount (fmol) of PCR products on the intensity of the test zone was studied for the three species in the range of 0-100 fmol, in triplicate. The results were reproducible. The biosensor can detect 1.6, 6.3 and 12.5 fmol PCR product from albacares, pelamis and BFT, respectively. Cross-reactivity studies confirmed that the probe of one species does not react with DNA from the other two species. The method was evaluated using mixtures of fresh tissue PCR products and mixtures of DNA isolates from heat-treated tissues at adulteration percentages of 1-100 %. The three most ordinary case adulterated mixtures were tested, i.e., albacares with pelamis, BFT with pelamis and BFT with albacares. The results show that the method can identify 1% of adulteration by naked eye.

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PS1-34 CADMIUM ACCUMULATION IN ORGAN TISSUES AFTER INHALATION OF CADMIUM-BASED NANOPARTICLES

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Cadmium and its toxic effects are a very current issue at this time, as poisoning by this element can cause serious health problems or even death. Cadmium is transported through the bloodstream and is found in a wide range of tissues. Thus, long-termed exposure to cadmium can lead to cancer and adverse effects on organ systems (skeletal, reproductive, cardiovascular, etc.).[1]

The distribution of cadmium and other biogenic elements was studied in the organs of mice - lungs, livers and kidneys. For the inhalation experiment, mice were divided into 3 groups. The first group was continuously exposed to CdO NPs for 11 weeks. The second group was exposed to CdO NPs for 11 weeks and then inhaled clean air for 5 weeks to determine whether the CdO NPs were permanently deposited in the tissues. In the third group (the control group), mice were exposed to the same conditions at each time point but breathed only clean air. After each exposure period, the mice were sacrificed and the organs were dry frozen, placed in agar medium, and cut into 20 µm thin cryosections. Samples were analyzed by laser ablation combined with inductively coupled plasma mass spectrometry (LA-ICP-MS) using a Nd:YAG laser.

[1] Rafati Rahimzadeh M, Rafati Rahimzadeh M, Kazemi S, Moghadamnia AA. Cadmium toxicity and treatment: An update. Caspian J Intern Med. 2017 Summer;8(3):135-145.
doi: 10.22088/cjim.8.3.135. PMID: 28932363; PMCID: PMC5596182.

PS1-35 An On-line SPE-UHPLC-HRMS Method for the Determination of 11 Classes of Per- and Polyfluoroalkyl Substances (PFAS) in Water

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The analysis of PFAS in environmental samples typically requires highly sensitive analytical methods, which should also be able to perform under extremely complex environmental matrix that can interfere with the analysis. We developed a highly sensitive method for determining 41 PFAS in water using on-line solid phase extraction (on-line SPE) coupled with ultrahigh performance liquid chromatography - high resolution mass spectrometry (UPLC-HRMS). Low detection limits were achieved through a series of optimization experiments on the on-line SPE, UPLC and HRMS conditions. The targeted compounds belonged to 11 classes and comprised legacy and emerging short chain (C4)

and long chain (C18) PFAS. Background contamination was eliminated by using PFAS-certified polypropylene vials and caps, high-purity solvents and modifiers for mobile phases, replacing fluorinated solvent tubing with PEEK, removing PFTE filter frits, and installing a delay column between the pumps and the autosampler. A key drawback of conventional offline SPE in PFAS analysis is the possibility of sample contamination and loss of surface-active PFAS to container walls and other materials [1]. As a result, on-line SPE was used for simultaneous sample clean-up and enrichment, enabling quantitative transfer of analytes to the analysis system [2] and guaranteeing minimal contamination [3] by eliminating the use of perfluorinated materials such as SPE cartridges, tubing, and syringes. A polymeric weak anion exchange (Strata-X-AW) on-line SPE cartridge combined with a Luna Omega PS C18 column provided the best results allowing for three orders of linear dynamic range and LODs ranging from 0.3 to 23.4 ng/L. The method has been validated using isotope dilution with a mixture of 24 extracted internal standard (EIS) that are isotopically labeled analogs of the method analytes. Method analytes with no available isotopically labeled analogs were quantitated using the IS analogs of similar chemical properties. Recoveries in drinking and well water samples ranged from 87 to 118%, except for PFODA (61%), PFTrDA (77%), PFHpA (77%), and PFDoS (78%). Matrix effects were also evaluated and found to not impact method performance. The method was applied to real water samples collected from and around a suspected PFAS-contaminated site, and 13 PFAS, including PFOS, PFOA, PFHxS and PFBA, were found at concentrations ranging from 81 to 3460 ng/L. We acknowledge the funding received from the European Union's Horizon 2020 Research and Innovation program under grant agreement No. 101037509 (SCENARIOS).

[1] M. Mazzoni, et al., J. Anal. Methods Chem. 2015 (2015) 942016. DOI:10.1155/2015/942016.
[2] I. Kourtchev, et al., Sci. Total Environ. 835 (2022) 155496. DOI:10.1016/j.scitotenv.2022.155496.
[3] T. Sanan, et al., J. Chromatogr. A, 1626 (2020) 461324. DOI:10.1016/j.chroma.2020.461324.

PS1-36 Developing Mass Spectrometry Methods for the Characterisation of Viper Venoms

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Venoms are complex bioactive mixtures capable of inducing cytotoxic effects. For snake venoms, the main components are proteins and peptides, the composition of which varies widely between species. Viper venoms are composed mainly of three protein families: Phospholipase A_2 (PLA_2), snake venom metalloproteinases (SVMP) and snake venom serine proteinases (SVSP). Understanding what venoms are composed of is important for three reasons: this information is needed to understand the mechanisms of action, it is essential to develop new antivenoms to treat the 2.7 million cases of snake envenoming per year and to characterise these new proteins to allow for the evaluation of their possible therapeutic effects.

There are two main methods used for investigating venom composition. These are bottom-up proteomics and genomics. Although these methods are important for such investigations, they have limitations. The bottom-up proteomics approach tends to miss peptides due to the protein databases being incomplete. The genomic approaches are time-consuming, and the genes are not always expressed, therefore they do not accurately reflect the composition of the venom. Neither of these techniques is capable of providing information about the quaternary structures of a protein, which is essential to know its function. For these reasons, new approaches are required to investigate snake venoms.

To overcome these problems mentioned above, native mass spectrometry (MS) can be used to investigate snake venoms. It allows for the analysis of non-covalent complexes in the gas phase. This method can be used with MS/MS, top-down sequencing and cyclic ion-mobility spectrometry (cIMS) for detailed structural investigations. The combination of all these techniques allows researchers to determine the quaternary structure of a biomolecule, and to investigate complex mixtures, as analytes can be discriminated based on mass and conformation. Also, sequence data obtained from top-down sequencing can be used to identify the proteins present in the venom.

In this work, these MS techniques were applied to analyse the venom from the Mexican cantil viper snake, Agkistrodon bilineatus to test how effective these MS methods are for determining venom composition.

PS1-37 Point of care breath analysis in chronic liver disease with a focus on NAFLD – a SIFT-MS pilot study

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Analyses of trace amounts of volatile metabolites are a fast and non-invasive promising approach for diagnosing metabolic changes caused by various diseases. Increased concentrations of specific volatile organic compounds (VOCs) in patients' breath may be a potential biomarker in diagnosing liver disease, especially non-alcoholic fatty liver disease (NAFLD). Quantitative results showing a correlation between the concentration of these volatile compounds and the degree of hepatic impairment could be particularly valuable. Recently metabolic rate of exogenous substances such as limonene and trimethylamine seems the most promising method to assess liver function. Other endogenous metabolites, including acetone, isoprene, ethanol, acetaldehyde and pentane, are also important indicators of the metabolic or inflammatory processes.

Results will be presented on a pilot clinical study involving 56 subjects, including controls and patients with chronic liver diseases (NAFLD, cirrhosis, fibrosis). Baseline concentrations of the VOCs and their response after administering safe doses of d-limonene and trimethylamine will be discussed. Data were obtained in the clinical setting using selected ion flow tube mass spectrometry, SIFT-MS.
SIFT-MS is a non-separative method for direct quantitative analyses of volatile compounds, VOCs, in air and humid breath based on chemical ionization. Selected reagent ions, either positive or negative (unreactive with major air components), ionize analyte molecules during a defined time in a flow tube by gas phase ion-molecule reactions producing analyte ions characteristic of the neutral analyte VOCs. Concentrations can be calculated in real-time from the ion count rates.

The main outcome of this study will be developing a breath test to screen for chronic liver diseases.
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Quantification of volatile metabolites in exhaled breath by selected ion flow tube mass spectrometry, SIFT-MS. P. Španěl, D. Smith. Clinical Mass Spectrometry 16 (2020) 18–24. https://doi.org/10.1016/j.clinms.2020.02.001

PS1-38 Characterization of a prototype thermal desorption unit for high-throughput headspace analysis

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The analysis of complex gaseous samples with biological origin for metabolomic applications often requires a high separation capacity to differentiate and identify the different metabolites and high throughput to achieve statistical significance. It is proposed to use a Secondary ElectroSpray Ionization source coupled to a High-Resolution Mass Spectrometer (SESI-HRMS) and a custom-built thermal desorption (TD) unit in combination with a robotic arm. We hypothesize that this setup will produce high-quality data for thousands of samples at a rate of one sample per minute because SESI-HRMS spectra are easy to interpret even without chromatographic separation due to the extreme softness of the method. This work investigated and characterized the custom-built TD prototype (named SIROCCO) coupled with SESI-HRMS for high-throughput headspace analysis of volatile organic compounds (VOCs) using solid-phase microextraction (SPME) sample preparation. The analytes were chosen based on their involvement in human metabolism, m/z ratio, and chemical class, i.e., short-chain fatty acids, hydrocarbons, or gut microbiome-related molecules. Aqueous solutions of compounds of interest were prepared at known concentrations in 20 mL vials. They were left for 12 hours to reach an equilibrium between the liquid and the gas phase. Using an SPME fiber, the gas phase was then sampled directly from inside the vials. Preliminary testing was conducted to optimize the sampling and analysis conditions for individual compounds: the solution concentration, the adsorption time in the headspace, and the desorption temperatures and times. These parameters were investigated in dry and humid conditions. The temperature of the SIROCCO thermal desorption unit varied from 40 to 180 ℃, while the humidity was adjusted (SIROCCO incorporates a closed loop humidity controller system that sets the dew point with high precision and without bubbling). The system's repeatability was studied within one day and over a long period. Calibration curves were generated for several compounds. Subsequently, they were combined with the measurements of compound mixtures to elucidate the influence of the matrix. The limits of detection (LOD), limits of quantification (LOQ), rise and fall time, and the linear response for the concentration range investigated were recorded for several compounds.

PS1-39 GENDER CHANGE IN THE ASPECT OF ASSESSING THE ATHLETE'S BIOLOGICAL PASSPORT

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The Athlete Biological Passport (ABP) is one of the tools to identify the use of WADA Prohibited Substances that affect an athlete's steroid profile. The concept of the ABP is based on detecting doping-induced changes in selected biomarkers by their long-time monitoring and comparison with values obtained during previous analyzes.

Several factors, such as inter-individual variability of steroid synthesis and metabolism, pregnancy, contraceptive pills, a large intake of alcohol, the administration of ketoconazole, human chorionic gonadotrophin in males, inhibitors of 5α-reductase, the influence of microorganisms existing in urine samples, and the use of masking agents and diuretics might affect the steroid profile. The steroid profile values may also be influenced by mental stress or changes related to the age of athletes.

The aim of this study was to investigate shifts in the hormonal balance in terms of gender change. For this purpose, we collaborated with a gynecologist and a person undergoing a sex change. The collected results were analyzed for the steroid parameters which were observed when assessing the athlete's passport. The collected and tested samples, taken from the person undergoing the gender change, showed a significant effect of the treatment on the change of the passport. All analyzes were performed using GC/MS system. Different body responses were observed depending on the treatment cycle. The obtained results were compared with the passport of the athlete who had a therapeutic use exemption (TUE) granted for testosterone use.

This study was carried out in cooperation with an expert from the Warsaw APMU (Athlete's Passport Management Unit), Dr Arkadiusz Kapliński and with financial support from the Ministry of Sport and Tourism of the Republic of Poland under the project number 2020.0145/1575/UDOT/BM and 2021.0416/1575/Udot/DS./14/AM with the ethics committee's approval nr KEBN-20-55-DK.

D. Kwiatkowska et al., Recent Advances in Doping Analysis 30 (2022) 40-43

PS1-40 Determination of trace vancomycin in fishery products by liquid chromatography tandem mass spectrometry

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Vancomycin is a glycopeptide antibiotic against for gram-positive bacteria. It is known as the last line of defence against methciline-resistant bacteria. South Korea, the United States and other countries have implemented relevant laws and policies prohibiting the use of vancomycin in food. This study was conducted to develop an analytical method to determine vancomycin residues in aquatic products. The vancomycin was confirmed and quantified via liquid chromatography tandem mass spectrometry(LC-MS/MS) in the positive ion mode using multiple reaction monitoring (MRM). The sample was extracted with 50 mM ammonium acetate and 15% acetonitrile solution, and purified with the Mixed-Mode Cation (MCX) solid-phase cartridge. Chromatographic separation was performed on a C18 column (3 μm, 2.1 x 150 mm), and quantification was carried out by a AB sciex 4500 mass spectrometer. The method validation was performed based on the Codex Alimentarius Commission(CAC) guideline. The results showed that the method has good linearity (r>0.99) in the range of 0.5-20 ng/mL, and the average recoveries of vancomycin at 1, 2, 10 μgkg-1 spiking levels were between 60 to 120%, with the relative standard deviations below 30%. The limit of detection (LOD) and that of quantification (LOQ) were 0.5 and 1 μgkg-1, respectively. This method can be applied to various aquatic products for determination of traces of vancomycin.

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PS1-42 Development of prediction models for effective optimization of make-up solvent composition in SFC-MS with different ionization sources

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The coupling of the state-of-the-art supercritical fluid chromatography (SFC) with mass spectrometry (MS) detectors is usually enabled via the commercially available SFC-MS interface. These interfaces take advantage of a sheath pump delivering a make-up solvent with the aim to improve the MS ionization and sensitivity. The composition of both, the mobile phase and make-up solvent, has an important effect on MS response. The MS response, and thus sensitivity, can be improved by the careful optimization of make-up solvent composition without affecting the chromatographic separation. However, this optimization is usually time-consuming as it includes testing of numerous solvents to achieve the desired sensitivity.

The aim of this study was to propose a prediction model which will enable to estimate the MS response obtainable with different make-up solvent compositions based on the physicochemical properties of the analytes and used ionization sources. Overall, three ionization sources were tested in this study including electrospray ionization (ESI), ESI-based UniSpray, and atmospheric pressure chemical ionization (APCI). The suggested prediction models are based on experimental data obtained by analyzing > 60 compounds on the diol column using two organic modifiers of the CO2-based mobile phase and 24 make-up solvents. The tested make-up solvents included pure alcohols and methanol in combination with 6 commonly used additives with varying molarity. The MS detection was carried out in positive and negative modes using all 3 ionization sources resulting in over 14.000 data points. Subsequent calculations included a correction to QC samples to mitigate interday variations in MS responses and the adjustment of responses to 100 µL methanol entering the ionization source to eliminate differences caused by variations in retention times.

The same rigorous experiment protocol enabled us to describe in detail the correlations between make-up solvent composition, physicochemical properties of the analyte, and particular ionization source. To sufficiently describe the analytes, > 250 molecular descriptors were calculated for each compound and subsequently used for a statistical evaluation by multilinear regression analysis. Finally, the prediction equations were suggested and tested using a set of blind probes. This proved that the make-up solvent composition optimal for the target analyte can be effectively predicted using the developed models, simplifying the optimizing procedure, decreasing solvent consumption, and increasing the environmental friendliness of the SFC technique.

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PS1-43 Improved compound identification in GC analysis using an EI&CI-TOFMS

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A time of flight (TOF) mass spectrometer simultaneously operating an electron ionization (EI) and a chemical ionization (CI) source is presented for non-target analysis. By coupling both ionization sources directly to one single gas chromatograph (GC), structural as well as accurate mass molecular ion information is generated. Hence, target and suspect screening analysis as well as effective non-target analysis using GC-MS is improved considerably. In this presentation, various experiments will be discussed, illustrating the potential of the GC-ecTOF for non-targeted approaches, including applications in fields such as environmental contaminants, material emissions, food flavour analysis and metabolomic research.

An Agilent 6890A GC was coupled to an ecTOF (TOFWERK, Thun, Switzerland), using a 70 eV EI source and the newly developed Tofwerk HRP CI source [1,2]. Various GC methods and sampling procedures were employed depending on the analytical need of the study, including liquid injection of extracted samples, headspace sampling including SPME and thermal desorption using Tenax tubes. To generate the ideal molecular ion information different reactant ions (e.g., N2H+, H3O+ and NH4+) were used for the chemical ionization process.

Standard procedures employed by routine laboratories, e.g., target screening for material emissions or steroid screening, is shown to be feasible using the GC-ecTOF. Whilst standard methods mainly focus on target analysis, suspect screening is enabled and improved using the ecTOF. Especially when EI library hits are fair with low corresponding probability, chromatographic and CI information can be used to increase compound identification confidence. False positives from an EI only approach can easily be identified and often correctly annotated using the additional information from the ecTOF. Furthermore, compounds that are not listed in libraries have a high uncertainty for identification using an EI only approach. Using the accurate mass information generated by CI, molecular sum formula for these unknowns can be derived. Combining this with the structural information generated by EI, tentative structure elucidation becomes feasible in many cases. Hence, non-target approaches become viable using the GC-ecTOF.

[1] Bräkling, S. et al. Parallel Operation of Electron Ionization and Chemical Ionization for GC-MS Using a Single TOF Mass Analyzer. Anal Chem., 2022, 94 (15), 6057-6064.
[2] Bräkling, S. et al. Hydrogen Plasma-Based Medium Pressure Chemical Ionization Source for GC-TOFMS. J. Am. Soc. Mass Spectrom., 2022, 33 (3), 499-509.

PS1-44 Analytical mass spectrometry method for quantification of TriPPPro-prodrugs and their metabolites in cell extracts

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Nucleoside and nucleotide analogues have been applied in anticancer and antiviral chemotherapy and they comprise the frontline of drugs used to tackle infections caused by viruses. Generally, nucleoside analogue drugs have to be metabolized by host cell kinases to undergo stepwise phosphorylation to yield the active nucleoside triphosphate (NTP) analogue. This metabolic conversion often proceeds slow and insufficiently. To directly deliver the active metabolites, our group developed the TriPPPro-approach.[1] According to this technology the γ-phosphate of an NTP analogue is masked by two lipophilic units and is therefore able to penetrate through the cell membrane. After enzymatic cleavage of the lipophilic masking units, the ultimately bioactive NTP is formed. To investigate the successful uptake and intracellular delivery of the metabolites, uptake studies were performed. Liquid

chromatography coupled to tandem mass spectrometry (LC-MS/MS) is the gold standard for such investigation because of its high sensitivity and selectivity. For that, a Hydrophilic Interaction Liquid Chromatography (HILIC) method with Multiple Reaction Monitoring (MRM) detection was developed which enables the simultaneous analysis of lipophilic TriPPPro-prodrugs and the associated hydrophilic metabolites (such as NTP, NDP and NMP) within a single HPLC run. The set of analytes that were used to develop this method comprises two TriPPPro-prodrugs derived from the anticancer drug 5-FU and also the FdU-TP, -DP, -MP and Floxuridine including internal standards. The procedure involves the incubation of the TriPPPro-prodrugs in two different cancer cell lines (SW620 and HT29) and subsequent sample extraction using protein precipitation. The validation in terms of linearity, accuracy, precision, recovery, matrix effects and sensitivity to ensure reliable quantification results will be presented.

[1] T. Gollnest, et al., Angew. Chemie - Int. Ed. 55 (2016) 5255–5258.
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PS1-46 Towards Mass Spectrometry Analysis of Organoids and Gastruloids

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A nano desorption electrospray ionization mass spectrometry (nanoDESI-MS) platform will be developed in-house and optimized to studying organoids and gastruloids. Organoids are self-organized 3D organ-like structures, derived from e.g. stem cells. Organoids are however significantly less structured and mature than their human equivalents. Gastruloids constitute a distinct group of organoid models that recapitulate a gastrulation [1]. In this fundamental developmental process, the cells in the zygote reorganize themselves into a discernable organism profile [2].

A functioning in-house nanoDESI, platform was built, based on the established protocol with modifications to achieve high flexibility and spatial resolution. The nanoDESI platform is coupled to an Orbitrap Mass Spectrometer. Nanoemitters are produced in-house using the gravity-assisted self-termination etching process with hydrofluoric acid, and coated with SnO2 using atomic layer deposition to ensure conductivity [3,4]. A liquid bridge was obtained in positive ionization mode and desorption of the analyte from the slide was observed. Next steps involve improving desorption, optimizing flow, as well as producing images of samples. The data obtained with the nanoDESI-MS is to be correlated with Raman spectrometry to monitor spatial distribution of metabolites and develop a deeper understanding of organoid development and stem cell biology.

For comparison with commercially available mass spectrometric imaging, a Bruker timsTOF fleX instrument with MALDI-MSI was used. Matrix was applied using Tessem Lab protocol: 20mg/ml 2,5-dihydroxybenzoic acid in 70% methanol + 0.1% trifluoric acetic acid, 10 passes. Samples were embedded in 2% carboxymethylcellulose, dried in Leica cryostat chamber, cut into 10 µm slices and placed on dedicated Bruker's Intellislides.

With MALDI MSI, first MS images of 7 day-old gastruloids were obtained and correlated with DAPI (4',6-diamidino-2-phenylindole) staining and scanned images. For specific lipid identity, tandem MS and/or

ion mobility will be employed. Improved cutting and embedding material is needed to preserve cellular structure and improve spatial resolution. Collected images give however a good indication of potential finding using MSI. This pilot subproject gave us good basis for optimization of all steps in the process, from sample preparation, through matrix application to collecting images.

In the next stages, images of steatotic and normal liver organoids will be analyzed with DESI and MALDI MSI in the Waters laboratory with the goal of mapping metabolite distribution. The imaging will be also performed on organoids exposed to per- and polyfluoroalkyl substances (PFAS) compounds as a first step in mapping organic pollutions in various cells using organoids and gastruloids as models.

1. Simunovic, M. and A.H. Brivanlou Development, 2017. 144(6): p. 976-985.
DOI: 10.1242/dev.143529
2. Arias, A.M., Y. Marikawa, and N. Moris Developmental Biology, 2022. 488: p. 35-46.
DOI: 10.1016/j.ydbio.2022.05.002
3. Kogler, S. Master Thesis, UiO, 2019.
4. Midtøy, L. Project Thesis, UiO, 2021.

PS1-47 Response of Saccharomyces cerevisiae Lalvin EC1118™ to tetraconazole-based fungicides: a metabolomic approach

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Yeasts may modulate their genome through the overexpression or repression of several genes as an adaptative response to the presence of fungicides in the fermentative media [1]. In this work an untargeted GC-QqQ-MS metabolomics approach is proposed to evaluate the impact of tetraconazole and their adjuvant sodium docusate on the metabolome of S. cerevisiae Lalvin EC1118™. The individual and the joint effect of both substances was assessed at laboratory scale through microvinifications assays of synthetic must (n=3). Yeast cells were collected at different fermentation times (30, 54, 68, and 92 h), centrifuged and flash-frozen in liquid nitrogen to avoid degradation of labile metabolites.

Intracellular metabolites were extracted by incubation for 1 h at 4 °C with a methanol/water/chloroform (3/1/1, v/v/v) mixture. For LC-MS analysis, 400 µL of metabolite extract were reduced to dryness and redissolved in 200 µL of acetonitrile/water (1/9, v/v). For GC-MS analysis, 400 µL of the metabolite extract were reduced to dryness an submitted to a derivatization process in two-steps: first with 100 µL of MeOX (20 mg mL−1 in pyridine) for methoxymation, and then with 100 µL of MSTFA with 1 % of TMCS for silylation. The obtained results were processed with the MetaboAnalyst software program to identify those metabolites that were differently expressed (Fold change >2, at a significance level of $p<0.01$) in the yeasts grown in the media spiked with the fungicide treatment (tetraconazole, sodium docusate and tetraconazole+sodium docusate) with respect to the control yeasts (grown in a media without fungicides).

The most important changes were observed at time 92 h. At this point, 20 compounds registered differences in their abundance in the presence of tetraconazole, 10 in the presence of sodium docusate and 13 in the presence of both substances. The most affected pathways were those related with aminoacyl-tRNA biosynthesis, purine metabolism and the amino acids metabolism (especially glycine, serine and threonine).

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[1] T. Sieiro-Sampedro, et al., Food Microbiol. 92 (2020) 103554. DOI: 10.1016/j.fm.2020.103554

PS1-48 Grafting nanoMIPs onto core-shell gold silica nanoparticles Au@SiO2@nMIP

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Molecularly imprinted polymers (MIPs) are synthetic, highly cross-linked biomimetic materials that have cavities capable of selective molecular recognition. They are characterized by the presence of sterically oriented functional groups in such a way as to be complementary to a molecule used as a template in the polymerization process. Recently, molecular imprinting technology has undergone an evolution through the solid phase synthesis of nanoparticles (nanoMIPs).[1] The nanoMIPs have demonstrated to be efficient mimics of natural receptors and a promising alternative to antibodies for sensors and bioanalytical applications.[2]

This study proposed simple method for fabricating a new composite material that combines the molecular recognition properties of nanoMIP with the optical properties of a colorimetric tag. A hybrid material was synthesized by modifying the surface of the nanoMIP with gold nanoparticles coated with shells of functionalized silica (Au@SiO2). Different synthesis and preparation approach of Au@SiO2 were evaluated. All the tested methods have foreseen the use of TEOS (Tetraethyl orthosilicate) as a silica source for the synthesis of silica-based materials and APTMS (3-Aminopropyl trimethoxysilane) as a silanizing agent to provide a terminal amino function which subsequently allows their covalent attachment on the nano polymer's surface. [3], [4], [5]

The new synthetized materials were characterized in different stages of synthesis through transmission electron microscopy (TEM), UV-vis spectroscopy, Dinamic light scattering (DLS) and Scanning electron microscopy with energy dispersive X-ray (SEM/EDX). Then, the functionality and binding properties of the composite nanomaterial were investigated. The data obtained were treated with the mathematical model of the Langmuir binding isotherm to estimate the affinity constant towards the target molecule and the number of binding sites.

This work paves the way for the development of efficient and reproducible synthesis strategies in order to obtain a new labeled synthetic receptor which can be use in lateral flow assays as an alternative to antibodies to improve the fundamentals of rapid tests such as accessibility, robustness, and usability to end users.

[1] F. Canfarotta, et al., Nat. Protoc., 2016, doi:10.1038/nprot.2016.030.
[2] D. Refaat et al., Int. J. Mol. Sci., 2019, doi:10.3390/ijms20246304.
[3] H. B. Lee, et al., Scr. Mater., 2006, doi:10.1016/J.SCRIPTAMAT.2006.08.044.
[4] Y. Lu, et al., Nano Lett., 2002, doi:10.1021/nl025598i.
[5] S. Liu, et al., Adv. Funct. Mater., 2005, doi:10.1002/ADFM.200400427.

PS1-49 Solvatochromic ionophore-based optical creatinine sensors

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Along with glucose, creatinine is found on the top of the list of essential biomarkers and is used routinely for the monitoring of various chronic conditions. Variations of its blood and urine concentrations outside the established healthy ranges are indicative of the development of a number of renal and muscular illnesses, such as chronic kidney disease (CKD) [1] and Duchenne muscular dystrophy [2]. Despite its high importance, however, clinical creatinine analysis has not seen major change in the last 30 years. The majority of laboratory analyzes today are based on Jaffé's reaction – an interaction of creatinine with picric acid, that is accompanied by a color change. This method has been first reported over a century ago and is known to suffer from significant errors due to the need for extensive sample manipulation and high sensitivity to temperature, making the ongoing search for new robust approaches even more relevant [3].

One of the promising directions for creatinine analysis lies in the realm of ion-selective sensors. Ionophore-based electrochemical and optical sensors have become a reliable analytical tool over the last decades and found many applications including analysis of biological fluids. There have been numerous attempts to create suitable creatinine receptors. The most notable one, a calix[4]pyrrole-based molecule capable of serving as a host for creatinine or creatininum cation, has been successfully applied for potentiometric and optical sensing of creatinine in urine and serum, demonstrating good selectivity over sodium and potassium [4,5]. This comes at a cost: the necessity to fully protonate sample creatinine in order to have a classic ion-exchange response enforces the use of acidic conditions (pH 3.8) in the course of the analysis, significantly limiting the scope of usable chromoionophores and affecting the response characteristics.

This contribution builds on the strengths of the previously reported approach while exploring other readouts and sensing protocols. Solvatochromic dyes are shown to be a convenient alternative to chromoionophores, allowing both absorbance and fluorescence readout. Their signal is pH-independent, which enables one to fine tune the sensor response to the desired concentration range without any downgrades of the reporter's performance. In turn, controlled sample acidification is incorporated into the procedure in order to automatize the analysis and limit the required sample matrix alteration. With an optimized procedure it is possible to quantitatively protonate sample creatinine regardless of the pH of the sample, negating the influence of natural urine pH from patient to patient.

[1] S. Bakker, Nat Rev Nephrol 9 (2013) 710.
DOI: 10.1038/nrneph.2013.233
[2] A.M. Zygmunt et al., Muscle & Nerve 67 (2) (2023) 138.
DOI: 10.1002/mus.27760
[3] R. Cánovas et al., Biosensors & Bioelectronics 130 (2019) 110.
DOI: 10.1016/j.bios.2019.01.048
[4] T. Guinovart et al., Biosensors & Bioelectronics 87 (2017) 587.
DOI: 10.1016/j.bios.2016.08.025
[5] M.M. Erenas et al., ACS Sens. 4 (2019) 421.
DOI: 10.1021/acssensors.8b01378

PS1-50 A Tunable Colorimetric Carbon Dioxide Sensor Based on Ion-Exchanger- and Chromoionophore- Doped Hydrogel

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Carbon dioxide (CO_2) is an essential physiological component for humans to maintain normal respiration.[1] In the baking industry, CO_2 concentration is also an important parameter during yeast fermentation for the production of bread, beer, and other alcoholic beverages.[2] Current CO_2 detection are mainly based on electrochemical sensors and optical sensors. The most common electrochemical CO_2 sensor is known as the Severinghaus electrode, which has been employed in blood gas analysis. However, the relatively high detection limit (several hundred ppm) and low stability of the electrode cannot be ignored.[3-5] Among various optical sensing methodologies, the infrared absorption of CO_2 molecules has been developed into commercial sensors for the detection in the gas phase. However, they could suffer from high cost and interference from water vapor.[4,5]

To overcome the disadvantages and improve the performance of the CO_2 sensors, different strategies were reported such as optical CO_2 sensors similar to the Severinghaus electrode. For these optical sensors, a hydrophobic gas-permeable polymer membrane is often used in tandem with a pH sensing thin layer. The gas-permeable membrane serves to isolate contaminants such as large particulates and ions while allowing CO_2 to enter the pH sensing layer.[6] The pH indicator, embedded in the pH sensing thin layer, is a critical component of such CO_2 optical sensors since it directly influences the sensitivity, the response range, and the signal transduction mode (color change, fluorescence, etc.).[7] The user-friendliness, portability, and low-cost, made the optical CO_2 sensors promising in various CO_2 detection applications.

In this work, we report on a colorimetric CO_2 optode sensor with hydrophobic pH indicator incorporated in polyurethane hydrogel. The additional use of a cation exchanger, sodium tetrakis-[3,5-bis(trifluoromethyl)-phenyl] borate (NaTFPB), relative to the amount of pH indicator in the hydrogel was capable of adjusting the apparent pKa of the indicator and then tune the CO_2 response range. Moreover, a cationic primary amine was also embedded in the hydrogel and the characteristics was compared with the conventional sodium bicarbonate. Finally, the CO_2 release during flour fermentation was successfully monitored by the proposed sensor.

[1] S. Schutting, et al., Anal. Chem. 85 (2013) 3271. DOI: 10.1021/ac303595v
[2] A. Calvo-Lopez, et al., Anal. Chim. Acta 931 (2016) 64. DOI: 10.1016/j.aca.2016.05.010
[3] J. W. Severinghaus, et al., J. Appl. Physiol. 13 (1958) 515. DOI: 10.1152/jappl.1958.13.3.515
[4] J. Zosel, et al., Meas. Sci. Technol. 22 (2011) 072001. DOI: 10.1088/0957-0233/22/7/072001
[5] E. Dervieux, et al., Sensors 22 (2022) 1. DOI: 10.3390/s22010188
[6] T. Li, et al., Sens. Actuators, B 243 (2017) 566. DOI: 10.1016/j.snb.2016.12.026
[7] K. Ko, et al., Sci. Total Environ. 729 (2020) 138786. DOI: 10.1016/j.scitotenv.2020.138786

PS1-51 Development of Conjugated Polymers for High-Detectivity Organic Photodetectors

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Organic photodetectors (OPDs) have been extensively studied to utilize in flexible electronic devices and sensors. The development of conjugated polymers that absorbs light and converts it into electricity is the most important to increase responsivity (R), specific detectivity (D*) are dynamic properties of the OPDs. For efficient light detection, it is basically necessary to improve the photocurrent density (Jph), but more importantly, it is to keep the dark current density (Jd) low. From the control of the morphology, molecular orientation, and insulating properties of the conjugated polymers, we could find several synthetic strategies to obtain high D* over 1013 Jones by effective suppression of Jd in the OPDs. In addition, from the molecular engineering of the conjugated polymers by incorporating π-spacers, the faster dynamic propeties and the improved R while keeping low Jd could be achieved. Based on these promising OPD performance, the optical fingerprint sensors were demonstrated using those conjugated polymers.

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PS1-52 Selective solid phase extraction of U(VI) ions based on new ion-imprinted polymer and its application for determination of uranium in waters, wine and honey

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Uranium is an abundant chemical element and occurs as three natural isotopes: 238U, 235U and 234U. Uranium exposure can induce multifarious health problems due to its chemotoxicity and radiotoxicity. In this regard, the quality control of waters and food requires the development of analytical procedures that are suitable for the separation and determination of trace amounts of uranium in them.

The aim of the present study is synthesis and characterization of new U(VI) ion-imprinted polymer sorbent (U(VI)-IIP) and further investigations on the extraction efficiency toward U in surface and ground waters as well as in more complex matrices such as wine and honey. U(VI)-IIP is obtained by precipitation copolymerization of methacrylic acid and trimethylolpropane trimethacrylate in the presence of U(VI) complexes with 4-(2-pyridylazo)resorcinol. The particles prepared are characterized for their composition, structure and morphology by using elemental microanalysis, Fourier transform infrared spectroscopy, scanning electron microscopy, and nitrogen adsorption–desorption measurements. The adsorption properties of U(VI)-IIP toward U(VI) ions are studied by batch procedure. The optimal pH value for the quantitative U(VI) sorption is 7, and full desorption is achieved by 2 M HCl. The mechanism of the adsorption process toward U(VI) is best described by pseudo-first-order kinetic and Langmuir isotherm models. High extraction efficiency and capacity of synthesized sorbent toward U(VI) allows its application for SPE determination of U(VI) in wine and honey without preliminary sample digestion using ICP-OES as measurement method. Recoveries achieved varied: (i) between 88 to 95% for surface and ground waters, (ii) between 90-96% for 5% aqueous solution of honey, (iii) between 86-93% for different types of wine. Limit of detection/quantification (LOD/LOQ), for U, defined as three/ten times the standard deviation of the blank signal using ICP-OES as instrumental method are: 0.05/0.15 μg/L for surface/ground waters, 0.07/0.2 μg/L for wines and 1.0/3.0 μg/kg for honey. The validity and versatility of proposed analytical methods were confirmed

by parallel measurement of U in water samples using Alpha spectrometry and U analysis in wine and honey after sample digestion and ICP-MS measurement.

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V. Georgiev, et al., Molecules 27 (2022) 5516. DOI:10.3390/molecules27175516

PS1-53 Reducing flow-induced peak broadening in electric field gradient focusing by using AC electro-osmotic flow

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Preparative separation techniques such as capillary electrophoresis and liquid chromatography suffer from peak broadening over time due to molecular diffusion. This limits the maximally attainable separation resolution hence sample purity for downstream analysis. This time-dependent peak broadening can be prevented by focusing techniques, such as electric field gradient focusing (EFGF). Here, a gradient DC electric field along a pressure-driven flow channel focuses proteins in bands where the electrostatic force balances the hydrodynamic drag force, counteracting molecular diffusion. However, this focusing effect is still limited by the parabolic flow profile in a pressure-driven flow. The traditional approach of using DC electro-osmotic flow (DCEOF) to create a plug flow profile fails because of the non-uniformity of the DC electric field along the channel length.

We have recently proposed to use AC electro-osmotic flow (ACEOF) to generate a plug flow profile in EFGF to minimize the peak broadening [1]. ACEOF employs AC signals applied to pairs of electrodes placed along the separation channel, to form the electric double layer and create an electro-osmotic flow, resulting in a plug flow profile. This allows in principle to decouple the flow generation from the global DC electric field. However, the velocity still depends on the DC offset between the AC electrodes and the local electrolyte potential, and we showed by numerical simulations that a constant velocity can be generated along the EFGF separation channel by applying a DC offset to each electrode to match the local DC electrolyte potential.

Here, we will show the practical implementation of the concept. To generate the DC offsets, we use a driving circuit consisting of a resistance ladder, with coupling capacitors to apply the AC signal to sets of electrodes. This is combined with carefully designed metal-insulator-metal capacitors integrated in each electrode, to closely match the DC electrolyte potential. To suppress the unwanted DCEOF resulting from charges on the channel surface between the electrodes, a passivation layer was added with an isoelectric point around pH 6. First experimental verification results of flow profiles along the channel length were obtained using particle tracking velocimetry at different heights. We will also discuss how other non-idealities, such as the low ionic strength required to operate ACEOF, could be accounted for. With this proof-of-principle, we aim to demonstrate how to overcome the practical challenges to enable the promising concept of using ACEOF in EFGF.

[1] T. De Moor, et al., J. Chromatogr. A 1689 (2023) 463726. DOI: 10.1016/j.chroma.2022.463726

PS1-54 First method based on gas chromatography-mass spectrometry for the simultaneous quantification of ethinyl estradiol and drospirenone in contraceptive formulations

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17α-Ethinyl estradiol (EE) and drospirenone (DP, 6β,7β:15β,16β-dimethylene-3-oxo-17α-pregn-4-ene-21,17-carbolactone) are synthetic versions of an estrogen and a progestogen that are used together to inhibit the ovulation process [1]. Quantifying their accurate amounts in contraceptive tablets is, precisely, a key feature in the preparation of these formulations. Most of the analytical methods are based on high-performance liquid chromatography with spectrophotometric detection, either with a single UV detector, or with an UV detector for DP and a fluorescence detector in series for EE [2].

As an alternative, herein we report the first method based on gas chromatography-mass spectrometry (GC-MS) for the simultaneous determination of EE and DP in contraceptive formulations. Analytes are extracted from the solid tablet by ultrasound-assisted extraction (15 min); the suspension is diluted, centrifuged, and directly injected into the GC-MS system. To correct potential instrumental variations, calibration is performed by the internal standard method using cholesterol as internal standard. The method was validated in terms of linearity, trueness, precision, and limits of detection and quantification. A good linearity (R^2 >0.99) was achieved in the whole calibration range both for EE (3-12 µg/mL) and DP (300-1200 µg/mL). Trueness, assessed in terms of percentages of recovery, was also satisfactory: 106 ± 8 % for EE and 93 ± 9 % for DP. Intra-day and inter-day precision studies showed relative standard deviation values (expressed as percentages) below 6 % for both analytes. In terms of sensitivity, instrumental limits of detection were 0.25 µg/mL for EE and 6.6 µg/mL for DP, and instrumental limits of quantification 0.82 µg/mL for EE and 22 µg/mL for DP.

The method was successfully applied to the analysis of contraceptive tablets from three different pharmaceutical companies. No differences were observed between the measured and the claimed amount of active principle per tablet, proving the applicability of our procedure. In addition, a stability study performed in both standards and sample extracts showed that these can be stored at room temperature over a period of at least seven days.

[1] P. Rosenbaum, W. Schmidt, F.M. Helmerhorst, et al. Eur. J. Contracept. Reprod. Health Care 5(1) (2000) 16-24. https://doi.org/10.1080/13625180008500376.
[2] V.B. Silva, A.A.G. Galdos, C.M.A. Mothe, et al. Braz. J. Pharm. Sci. 49(3) (2013) 521-528. https://doi.org/10.1590/S1984-82502013000300013

PS1-55 Quantitative analysis of reference gas mixture using a gas chromatograph with a thermal conductivity detector under unstable retention time of peaks

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Gas chromatograph with thermal conductivity detector (GC-TCD) and simple structure, including manual mechanical mass flow controllers, is still used for routine gas analysis or at factories, because of its robustness and usability. Helium gas regulated by the mass flow controllers flow into the detector through analytical packed column and reference packed column.

An international comparison (APMP.QM-K111) was carried out for mutual comparison of reference gas mixtures (propane[C_3H_8] in nitrogen with concentration close to 1000 μmol/mol) of National Metrology Institutes in Asia-Pasific region. When the sample gas mixture of this comparison was measured using the GC-TCD in my laboratory, both peak areas and retention times of propane peaks were unfortunately unstable. The reason was unknown at that time. Later, it was found that the peak area was proportional to the retention time. Additionally, the peak areas were corrected using their linear retention time dependence. This correction enabled to determine the mole fraction of the sample gas mixture with a relative expanded uncertainty of 0.1 %, which was sufficient to analyze the sample of this international comparison with high accuracy. The analytical concentration was also well consistent with reference value of the international comparison. If the peak area was not corrected, the relative expanded uncertainty would be 1.1 %.

Several years after the measurements, it was discovered that malfunction of the mechanical mass flow controller might be the root cause of such unstable retention times. The carrier gas could not flow into the analytical packed column at all. After the controller was replaced to new one, the stability of retention time was greatly improved. The difference in average values between the fastest and slowest retention time was 0.0005 min, while the difference was 0.017 min at the international comparison analysis.

The details of the experimental conditions and analytical results have been described in this presentation, which will be hopefully a trouble-shooting for practitioners of reference gas mixture analysis using the GC.

<Reference>
1) Lin T.Y., et al. "APMP.QM-K111—propane in nitrogen", Metrologia, 2018, 55(1A), 08008.
2) Matsumoto, N., "Quantitative analysis of reference gas mixture using a gas chromatograph with a thermal conductivity detector under unstable retention time of peaks", J. Chromatogr. A [submitted].
10.1088/0026-1394/55/1A/08008

PS1-56 DEVELOPMENT OF MATERIALS AS ADSORBENT FOR RECOVERY OF HIGH-MOLECULAR WEIGHT POLYCYCLIC AROMATIC HYDROCARBONS FROM NON-AQUEOUS MEDIUM

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Polycyclic aromatic hydrocarbons (PAHs) constitute a group of persistent organic pollutants widely distributed in the environment and due to their lipophilic nature, they can easily

contaminate oils, mammals and marine organisms. Most of these compounds have carcinogenic properties and therefore must be controlled in the environment. Currently, the removal and quantification of polyaromatics in substances with predominant lipid composition is a challenge for

researchers and several hydrophobic adsorbents are widely used to decontaminate water sources with PAHs. However, such adsorbents, do not have satisfactory results for the purification of oils, fuels and other lipid solutions, since they do not have selectivity for PAHs and, therefore, are saturated by components of the hydrophobic matrix. Our idea is to use hydrophilic adsorbents with a higher affinity for PAHs removal from the hydrophobic matrix. Recently, it has been published in the literature that carbon dots (CNDs) can bind tightly to PAHs due to extra molecular pi-pi interactions, and our primary experiments have demonstrated that CNDs can be covalently immobilized on silica gel surfaces [1]. This effect may allow the preparation of a new type of selective PAHs adsorbent suitable for both analytical determination and decontamination of lipid solutions, such as natural oils. The nanocomposite with immobilized CNDs was obtained by covalent immobilization of CNDs fragments due to adsorption on aminosilica silica surface and nanoreactor approach. The adsorbent properties were performed in dynamic SPE modes for the model compounds: a organic mobile phases and PAHs mixture (16 pollutants) from organic medium. As a result,the nanomaterials show high affinity to PAHs with more that 5-cicles (as Dibenzo(a,h)antraceno, Benzo(ghi)perileno).

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References:
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PS1-57 Long-Term Retention Time Stability in SFC

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A broad spectrum of stationary phases (SPs) is employed in current supercritical fluid chromatography (SFC). SPs include various chemistries from polar silica and diol SP, across aliphatic and aromatic amines, to nonpolar carbonyl chain SP providing different interactions. Since most of the SPs commonly used in SFC are silica-based, the free acidic silanols also play an important role in SP selectivity and interactions. Indeed, free acidic silanols can react with an organic modifier, such as methanol, which is SFC mobile phase component, and form silyl-ethers. The simple reversible condensation is called silyl-ether formation (SEF) and can result in a decreased number of free silanols that cannot participate in the interactions of SP and analytes. Consequently, the selectivity change can be observed over time. Moreover, water formed as a side product of SEF can act as a polar mobile phase additive resulting in a decrease of retention time. The typical separation in SFC is carried out with mobile phase containing additives, such as ammonia, volatile acids, and ammonia salts of organic acids, that can act as a catalyst of the SEF. Additionally, the same additives can be irreversibly adsorbed on silica surface resulting in the selectivity change. Nevertheless, both processes can be reversed to some extent. In the case of SEF, the slight addition of water can shift the equilibrium towards free silanols and mitigate the SEF effect, as well as the flushing of the column with water during the column regeneration procedure. Additives can be effectively removed by long washing of columns using neat organic solvents, such as methanol. However, a deep investigation on long-term SP stability and selectivity is still missing as it is a phenomenon typical for SFC.

This study aimed to describe and evaluate the effect of column aging and additive adsorption in SFC. We tested silica SP and 8 different SP with hybrid silica support, including hybrid silica, C18, diol, 2-ethylpyridine, 2-picolylamine, 1-aminoanthracene, diethylamine, and fluorophenyl. 3 different mobile phase compositions, carbon dioxide with methanol, methanol + 10 mmol/L ammonia, and methanol with 2% water, were used in the same gradient elution to cover different mobile phase pH. The rigorous

protocol, where the retention behavior of 112 analytes was observed over one year and after the regeneration protocols, was carried out. The differences among SP were described using statistical evaluation.

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PS1-58 Simultaneous Analysis of Chromium Species Using µLC-ICP-MS

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Chromium (Cr) is a transition element that exists in oxidation states ranging from - 2 to +6. The common stable ones in the environment are trivalent Cr(III) and hexavalent Cr(VI) chromium. Cr(III) is an important micronutrient for the human body, while Cr(VI) is highly toxic and carcinogenic. The environmental concentrations of both oxidation states are low. Due to the differences in toxicity between Cr(VI) and Cr(III) compounds, speciation of Cr is very important. Therefore, an improved sensitive and robust method for the simultaneous determination of Cr(III) and Cr(VI) in water samples (saliva, plasma, gastric juice, sweat and urine) has been developed. The method uses a hyphenated micro liquid chromatography (µLC) system coupled to inductively coupled plasma mass spectrometry (ICP-MS). The optimised method incorporates a pH adjusted EDTA complexation step to stabilise Cr(VI) and Cr(III). The µLC system uses an anion exchange micro-sized column to separate the Cr species. Cr(III) and Cr(VI) were separated with different retention times at 170 and 230 sec, respectively. The method was optimized and validated by spiking Cr(III) and Cr(VI) in various water samples. Furthermore, the method was validated using a drinking water proficiency testing material sample. The developed method can be used for rapid routine determination of chromium species with high precision and reliability.

PS1-59 Evaluation of extraction potential of novel silica IL-based fibers using headspace solid-phase microextraction for the determination of organophosphorus insecticides in real samples

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The solid-phase microextraction technique (SPME) is one of the most popular sample preparation techniques widely used in analytical practice for sampling a wide range of analytes from media characterized by complex matrix composition (e.g., food products, biological substances, and environmental samples). However, one of the main drawbacks of using SPME is the limited choice of sorption materials of commercially available SPME fibers. Therefore, many scientific efforts are directed at searching for new sorption materials for coating SPME fibers as stationary phases. Recently, special attention has been paid to ionic liquids (ILs) due to their unique physicochemical properties. Our proposed solution is obtaining a porous solid silica structure on the surface of the SPME fiber and confinement of ionic liquid within its pores.

Novel silica IL-based SPME fibers were prepared and applied to HS-SPME-GC-FID method of determining organophosphorus insecticides from water and food samples. The fibers were obtained according to a developed procedure involving immersing glass fibers in a mixture of potassium silicate (a precursor) and formamide (a pore-forming agent) in a specially designed container. After the final formation of silica coating, different ILs were physically immobilized in its pores. Four ILs based on the same bis(trifluoromethylsulfonyl)imide anion and different cations: 1-Butyl-1-methylpyrrolidinium, 1-Benzyl-3-methylimidazolium, 1-(2-Methoxyethyl)-3-methylimidazolium, and Butyltriethyl ammonium were investigated. These ILs were selected based on their beneficial parameter differences, such as moderate viscosity and high desorption temperatures.

The most important extraction parameters of HS-SPME-GC-FID method were optimized with a central composite design. The SPME fibers obtained higher selectivity for extracting the analyzed insecticides than commercially available fibers (polydimethylsiloxane, PDMS and polyacrylate, PA) and showed good precision and repeatability. All coatings achieved promising results for their application as extractants for SPME. However, the SPME coating produced with ionic liquid based on 1-Benzyl-3-methylimidazolium cation exhibited the best extraction efficiency results. Finally, the proposed ionogel fibers were employed to analyze the insecticides in fresh cucumber and grapefruit juices. It was revealed that the sample matrix could affect the extraction efficiency of SPME of insecticides.

A further selection of different ILs could be a simple and convenient method of tuning the required physicochemical properties of ILs depending on the class of the substances to be extracted.

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PS1-60 Electrostatic Repulsion Hydrophilic Interaction Liquid Chromatography: an underrated separation method for charged analytes

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Due to their high polarity and abundance of amines, spermine and spermidine pose multiple challenges for analysis via liquid chromatography. Reversed phases generally do not enable any retention for these analytes. Hydrophilic interaction liquid chromatography (HILIC) often leads to undesirable artefacts, such as carryover and tailing. Electrostatic repulsion hydrophilic interaction liquid chromatography (ERLIC) is a non-prominent separation technique, capable of separating analytes such as spermine and spermidine, while enabling good retention and peak symmetry.

The acronym ERLIC was introduced by A.J. Alpert (2008) and describes the use of an ion exchange column equally charged as the analyte in combination with a HILIC elution system (i.e., highly nonpolar with a small percentage of water). The mobile phase forms three distinct layers: an immobilized aqueous layer hydrating the stationary phase, a mobile diffuse aqueous layer on top, and a mobile organic layer in the middle of the mobile phase. Retention and elution depend on the two main principles electrostatic repulsion and hydrophilicity of the analyte. Electrostatic repulsion between the stationary phase and the analyte repels the analyte away from the stationary phase and towards the mobile layers in the middle of the mobile phase. The hydrophilicity of the analytes, however, keeps

them away from the organic layer in the middle of the mobile phase. Thus, an equilibrium between the electrostatic repulsion and the analyte's hydrophilicity is reached, which can be influenced by adjusting the water content of the mobile phase. A lower content of water in the mobile phase reduces the thickness of the diffuse aqueous layer and, hence, shifts the balance of the analyte towards the immobilized layer. This in turn leads to retention of the analyte. Up to now, this separation technique has mostly been applied in peptide analysis.
In the present study, a model ERLIC system has been optimized for the separation of the polyamines spermine and spermidine as well as the diamines cadaverine and putrescine. As far as the authors know, this is the first method using gradient elution with the ERLIC technique. The developed method can retard all analytes, while still eluting them in under six minutes. Furthermore, the polyamines spermine and spermidine are separated and differentiation of the diamines cadaverine and putrescine is enabled by tandem mass spectrometry detection.

A.J. Alpert, Anal. Chem. 80 (2008) 60.
DOI: https://doi.org/10.1021/ac070997p
A.J. Alpert, J. Chrom. A 1538 (2018) 45.

PS1-61 Phosphorus removal and recycled from tertiary effluent in sewage treatment plant using graphene modified with magnetic nanoparticles (M@GO)

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Phosphorus is employed in detergents, as fertilizers in agriculture, etc. As a nutrient for plants, too much phosphorus can cause increased growth of algae and large aquatic plants, which can result in decreased levels of dissolved oxygen– a process called eutrophication. On the other hand, P is a relatively limited resource, considered by the European Union as a strategic interest material. Thus, the removal and recycled of P from the sewage treatment plants is of great interest to the society.

In this work, a new patented magnetic graphene oxide (M@GO) for the removal of phosphorus from wastewater is studied. The main technical advantage of this solid adsorbent is its easy separation from the treated water by applying a magnetic field. The key factors affecting the sorption and elution efficiency are studied. The thermodynamic adsorption model that provides a best fit was the Langmuir isotherm. The mass transfer kinetic model indicates that the mass transfer of P between the bulk liquid and the solid surface is not the rate-limiting step of the adsorption process.

The P adsorption on M@GO was demonstrated by TEM, XPS, FTIR. After the adsorption, an ammonia aqueous solution has provided to be the best eluent to recover the phosphorus from the solid adsorbent, as ammonium phosphate, with recovery yields above 90%. The results of this work have driven to the design of a new magnetic reactor for the treatment of waste water.
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PS1-62 Why is paper recycling NOT a one-size-fits-all process: A case study on multi-residue analysis of semi-volatile pollutants in South African recycling paper grades using accelerated solvent extraction with gas chromatography-mass spectrometry

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Recycled paper is a valuable commodity which forms an intrinsic part of actualizing a circular economy. Research on pollutant prevalence in South African recycling paper grades is limited and may differ from studies emanating from the Global North, where wastepaper is usually segregated at source. The aim of this study was to develop a multi-residue analysis method for identification and quantification of semi-volatile organic pollutants present in the South African paper recycling chain. The analyzed samples were collected from various points of the recycled paper chain in Cape Town, South Africa and included magazines, newspapers, office paper, mixed waste and cardboard. Multi-residue analyses were performed using gas chromatography coupled with mass spectrometry after accelerated solvent extraction. The extraction conditions were optimized to an extraction temperature of 70 °C utilizing a solvent mixture of acetone: hexane (5:2). The method quantification limits ranged from 3.147 ηg/g for tris (2,4-di-tert-butylphenyl) phosphite to 14.962 ηg/g for benzophenone. Paper was spiked at different concentrations and yielded recoveries ranging from 73 to 104%, all with relative standard deviations below 10%. The pollutants found in the investigated samples were those previously reported as prevalent in European recycled paper, these included diethylhexyl phthalate, dibutyl phthalate, benzophenone and diisopropylnaphthalene. In addition, butylated hydroxytoluene, tris (2,4-di-tert-butylphenyl) phosphite and its degradation product tris (2,4-di-tert-butylphenyl) phosphate, which are associated with plastic and multi-material packaging, were also detected. These indicated the unique elements of the South African recycling paper chain.

PS1-63 Passive sampling of semi volatile organic compounds in urban atmospheres near petrochemical parks

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Air quality is an issue that affects more and more people as it is directly related to health problems such as respiratory infections, lung cancer and heart disease [1]. For this reason, the presence of contaminants in air has to be controlled. Air samples are mainly collected by active sampling using high volume samplers, but passive sampling is an interesting alternative because it is cheaper, is not noisy, takes up less space and does not need power supply [2]. However, to apply passive sampling as reliable sampling technique, the sampling time must be optimized and the diffusive uptake rates of each target compound for the selected sampling time are required.

In the present study, a multi-residue method based on passive sampling followed by the extraction of contaminants by pressurized liquid extraction and the determination by gas chromatography-mass spectrometry has been developed for monitoring semi-volatile organic compounds in urban air samples . More specifically, the compounds studied are polycyclic aromatic hydrocarbons and high

production volume chemicals (annual production > 1,000 tons per year), such as benzothiazoles, benzenesulfonamides, phthalate esters, organophosphate esters, ultraviolet stabilizers and phenolic antioxidants. For the passive sampling, aluminium collectors with polyurethane foam were used and the diffusive uptake rates of the target compounds were determined under real conditions, urban air near a petrochemical area, and from active sampling results as reference method [3]. The proposed method includes passive sampling for two months and the determination of diffusive uptake rates for 30 of the 64 compounds included in the study with values between 1,392 m3·day-1 (benzo(a)anthracene) and 25,641 m3·day-1 (2,4-di-tert-butylphenol). Validation results showed recoveries between 49 % for 2-amino-1-H-benzothiazole and 131 % for 2-(2H-benzotriazol-2-yl)-4-methyl-6-(2-propenyl)phenol and repeatability, as percent standard deviation (% RSD, n=3), lower than 21 % for all the target compounds. The method was applied for the determination of the contaminants at different sampling sites near the petrochemical parks of Tarragona and several contaminants were quantified.

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[1] S. Wang et al., Sci. Total Environ. 871 (2023) 162005, doi: 10.1016/j.scitotenv.2023.162005.
[2] B. Strandberg et al., Chemosphere 190 (2018) 35-42, doi: 10.1016/j.chemosphere.2017.09.106.
[3] T. Harner et al., Atmos. Environ. 75(2013) 123-128, doi: 10.1016/j.atmosenv.2013.04.012.

PS1-64 A low-cost portable system for on-site detection of soil pH and potassium levels using 3D printed sensors.

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Conventional analytical techniques for monitoring of nutrients and pH provide traceability, precision, and accuracy, but it requires expensive and complex instrumentation, while only providing low analysis frequency. This lack of frequent monitoring and real time information on nutrient availability in soils has led to excessive use of fertilisers resulting in pollution of groundwaters and waterways. Additionally, overuse of chemical fertilisers can lead to soil acidification, resulting in soil depletion, decreased fertility. and reduced crop yields.

Harvested agricultural land generally has potassium levels replenished annually by addition of synthetic based fertilisers. Its overuse could be avoided by more frequent on-site detection of potassium levels. Soil pH also plays a significant role in soil biogeochemical processes responsible for plant growth and biomass yield. It is a reliable predictor of soil properties as it's considered a major element that influences other soil properties.

This work has developed a miniaturised ISE platform consisting of 3D printed sensors for potassium and pH in soil pore water. The design of these arrays is novel in soil analysis as they allow the determination of the analytes in extremely small samples volumes without the requirement of sample preparation.

The prototype uses 3D printed electrodes that are spotted with appropriate Ion Selective Membranes (ISMs), attached to a PSoC4 microcontroller, and programmed using C language. A Universal Asynchronous Receiver/Transmitter (UART) transmits results over a serial link via USB to PC. The terminal emulator TeraTerm is then used to present results. For further development of this project

the number of sensors can be increased to 8 and a SigFox module is used for remote sensing. This advancement in electronic instrumentation and the fabrication of low-cost 3D printed sensors has made the possibility of more frequent portable in-situ measurements of potassium and pH in soil pore water much more realistic.

PS1-65 The Application of Electroanalysis for the Monitoring of the Photocatalytic Degradation of the Herbicide MCPA

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MCPA (2-methyl-4-chlorophenoxyacetic acid) is a herbicide used to control the growth of broad-leaf weeds in arable and horticultural crops. The herbicide is applied by broadcast spraying and despite regulatory buffer zones, MCPA can frequently enter watercourses. As MCPA is also highly soluble, it can leach from soil into waterways after rainfall. MCPA in these watercourses can contaminate natural ecosystems and disrupt biodiversity [1].

Water remediation processes such as adsorption and coagulation have limitations as they often only concentrate pollutants or simply transfer them to another phase or media, and in doing so, fail to completely remove the threat. Other processes such as filtration and chemical methods are associated with high operating costs as well as the possibility of generating toxic secondary pollutants [2].

Advanced oxidation processes (AOP), due to the generation of highly reactive and a non-selective reactive oxygen species, can degrade problematic organic pollutants. Among AOPs, photocatalytic processes have been considered as an effective method for the mineralisation of organic pollutants due to its reduced energy requirements, low cost and maintenance and minimal production of harmful by-products.

Current analytical methods for detecting and quantifying many organic pollutants include chromatographic and spectroscopic techniques. Although these detection methods are sensitive and reliable, they require high-cost instrumentation, involve time consuming sample preparation procedures, require skilled technical personnel, and use significant volumes of solvents [3].

Electrochemical sensor technology offers an attractive alternative method for the detection and quantification of electroactive organic pollutants. Electrochemical sensors can offer real time analysis with many advantages over conventional methods. These include the use of inexpensive, portable instrumentation, quick analysis time, minimal sample preparation, small sample volumes (in µL quantities) while also providing high sensitivity and selectivity to the analyte of interest [4]. The portability aspect of this detection technique offers in-situ monitoring yielding rapid onsite data.
To ensure the development of photocatalytic technology for water remediation applications continues, it's paramount that the degradation process can be accurately monitored. Monitoring the degradation of MCPA and its main degradation product, CMP, is usually performed by gas or liquid chromatography. These methods can be costly, time-consuming and/or requiring complicated sample preparation. A potential solution to this problem is the utilisation of an electrochemical sensor for in-situ detection providing fast, reliable, and cost-effective monitoring.

Therefore, presented here is a study, which has shown the simultaneous detection and monitoring of MCPA and its degradation product CMP by electroanalysis during a photocatalytic process.

[1] P. A. Morton, et al., Sci. Tot. Env. 755 (2021) 142827 DOI:10.1016/j.scitotenv.2020.142827
[2] G. Crini, et al., Env. Chem. Lett. 17 (2019) 145.
DOI: 10.1007/s10311-018-0785-9
[3] S. Tajik, et al., Int. & Eng. Chem. Res. 60 (2021) 1112
DOI: 10.1021/acs.iecr.0c04952
[4] R. Umapathi, et al., Co. Chem. Reviews (2022) 214305
DOI: 10.1016/j.ccr.2021.214305

PS1-66 Multi-process control of trace contaminants fate in surface waters probed at nano-scale by using asymmetrical flow field flow fractionation linked to ICP-MS

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Flow field-flow fractionation (FlFFF) is well-suited state-of-the-art technique finding growing applications in the separation and size characterization of natural and engineered nanoparticles. The hyphenation of the FlFFF with a very sensitive elemental analyzer, such as inductively coupled plasma-mass spectrometry (ICP-MS) and single particle ICP-MS, opens novel avenues to explore the interactions of metal-containing forms, e.g. traces metals and engineered nanoparticles, with different abiotic and biotic components in the aquatic systems. Determining the speciation of dissolved trace metals in the complex environmental and biological systems is paramount for the assessment of their reactivity. Some of the recent advances with respect to the understanding of the trace metals and metallic nanoparticles behavior in the aquatic systems by using the asymmetrical FlFFF coupled or not to ICP-MS will be exposed. With examples of our own research we will illustrate the capabilities of the AFlFFF-multidetection system (i) to explore metal association, size or molar mass distribution of metal complexes with dissolved and colloidal organic matter; (ii) to characterize engineered nanoparticles and stability under natural conditions (iii) to explore the interaction of manufactured nanomaterials with (metallo)proteins, (iv) to distinguish between the contaminant forms e.g. dissolved and nanoparticulate.

More than a sizing technique, and thanks to detectors available online, AFlFFF give rise to multiplex information and can be used to probe-out at the nanoscale the behavior and bioreactivity of the various metal containing forms in complex environment.

PS1-67 Development of Urban Particulate Matter Reference Material for the Analysis of Hazardous Chemicals and Source Identification

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Airborne particulate matter (PM) is a growing concern due to its negative impact on both the environment and human health. Exposure to PMs has been linked to respiratory and cardiovascular illnesses, and there are even claims of a connection to neurodegenerative diseases. The chemicals contained within PMs, as well as their size, are significant sources of toxicity. The composition of chemicals found in PMs can provide valuable information about their origins and growth mechanisms. However, accurately measuring these chemicals is difficult due to the complex sample matrix. To address this challenge, certified reference materials (CRMs) are needed for method validation and quality control purposes. KRISS has developed an urban PM CRM for various chemical analysis using raw materials collected from intake air filters in the Greater Seoul area of South Korea. The analytical methods used for the characterization of various elements, polycyclic aromatic hydrocarbons (PAHs), water-soluble ions, total carbon, and isotope ratios of carbon/nitrogen/sulfur/lead in the CRM will be discussed, as well as homogeneity and stability assessments. Additionally, progress on the development of two additional PM CRMs (subway station and on-road PM CRMs) and other potential measurands will be presented. The PM CRMs developed by KRISS will improve the quality of chemical analysis in PM samples, enabling reliable assessments of PM-mediated toxicity and source identification.

PS1-68 Assessment of microwave assisted extraction efficiency for the determination of herbicides in soil and maize cob: cumulative and health risks assessment

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The effectiveness of microwave-assisted extraction (MAE) was used for the determination of herbicides (atrazine, 2,4-D, mesotrione and glyphosate) in soil and maize crop followed by gas chromatography with flame ionization detector. The parameters optimised were extraction time (2, 8 and 15 minutes) and extraction solvent volume (5, 12, and 25 mL) and extraction solvents (hexane: acetone (1:2 v/v), acetonitrile: acetone (1:2 v/v) and the mixture of methanol: ethyl acetate (1:1 v/v). The recoveries of herbicides in maize and soil were 80-98% and 85-101%. The analysis repeatability, represented as relative standard deviations were less than 20% for all herbicides. All the herbicides calibration curves showed a good correlation coefficient (R^2) ≥0.996, indicating good linearity. The limits of detection and quantification ranged between 0.1-0.29 µg/L and 1.0-2.9 µg/L. These findings showed that MAE method is more accurate and sensitive, thus can be accurately applied for the determination of the assessed herbicides in soil and maize cop. Herbicides concentrations obtained ranged from 2.7 – 20.4 µg/L in maize and 1.2 - 30.5 µg/L in soil samples. The concentrations obtained in maize were higher than the maximum residue limits suggesting that health effect may occur upon continuous consumption. The herbicides toxicity index further confirmed the possible high toxicity effect of the studied maize crop as it exceeded the threshold value of 1. However, the health risk index was lower than 100% limit and did not exceed the acceptable daily intake of the maize crop in both adult and children indicating no possible health effects.

1. S. Singh, V. Kumar, J. P. K Gill, S. Datta, S. Singh, V. Dhaka, D. Kapoor. Int. J. Environ. Res. Public Health, 2020, 17, 7519.
2. S. Moret, C. Conchione, A. Srbinovska, P. Lucci, Foods, 2019, 8, 503.
3. M. Czaplicka, H. Barchanska, K. Jaworek, B. Kaczmarczyk, J. Soils Sediments, 2018, 18, 827 – 83

PS1-69 Enrichment and clean-up of steroid hormones from water samples

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Synthetic hormones such as 17α ethinyl estradiol (EE2), known as an active ingredient of the birth control pill, but also the natural or nature-identical hormone estradiol (E2) are specifically used to regulate hormone balance in humans. Both substances, as well as their common degradation product estrone (E1), are excreted by the human body and are therefore found in the environment. They are known to have a lasting effect on fish reproduction even in the very low ng/l range [1]. By the publication of the COMMISSION IMPLEMENTING DECISION (EU) 2015/495 of 20 March 2015 a first watch list of substances for Union-wide monitoring in the field of water policy pursuant to Directive 2008/105/EC of the European Parliament and of the Council was established [2]. In the first EU Watch List describes very challenging detection requirements for these endocrine disruptors. The LLOQ for estrone and 17-beta-estradiol were set at 0.4 ng/l and for 17-alpha-ethinyl estradiol at 0.035 ng/l. The EU Commission´s predicted no-effect concentration (PNEC) values are 3.6 ng/l for estrone, 0.4 ng/l for 17-beta-estradiol, and 0.035 ng/l for 17-alpha-ethinylestradiol.

This work presents the reliable and successful determination of 17α-Ethinylestradiol, 17β-Estradiol, and Estrone from drinking water. By using a spherical, hydrophobic polystyrene-divinylbenzene resin, CHROMABOND® HR-X, it was possible to extract efficiently the steroid hormones before cleaning the extract with silica gel, CHROMABOND® SiOH. In combination with large volume injection, high recovery rates about 80% with good reproducibility could be achieved. The elution step from the PS/DVB copolymer was optimized with a hydrophobic solvent mixture (hexane-ethyl acetate (90:10; v/v)) so that the cleaning procedure could follow directly without timewasting eluent exchange.

The identification and the quantification of the focused analytes was finally carried out by ESI mass spectrometry on a NUCLEOSHELL® column. The polar monomeric octadecyl modification successfully helps to separate analytes from matrix for a wide calibration range from 10 pg/mL up to 1000 pg/L.

[1] Steroid hormones in the aquatic environment, J.O. Ojoghoro, M.D. Scrimshaw, J.P. Sumpter, Science of The Total Environment, Volume 792, 20 October 2021, 148306
[2] COMMISSION IMPLEMENTING DECISION (EU) 2015/495 of 20 March 2015 establishing a watch list of substances for Union-wide monitoring in the field of water policy pursuant to Directive 2008/105/EC of the European Parliament and of the Council.

PS1-70 Preparation of volatile gas-based probe and its application in identification of drug resistant bacteria

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Bacterial infections are the main cause of many human diseases and a significant entail factor of mortality. The ability to rapidly identify bacteria has major implications in terms of the treatment of infections in a clinical setting. Typical bacterial identification methods (culture-based, serological and genetic methods using DNA replication) are highly time-consuming (several hours, sometimes even days) and expensive. Too often the diagnostic data arrive late, with attendant increases in costs, morbidity and mortality. In recent years, colorimetric and fluorescent probe method have become

useful tools for monitoring enzyme activity because of their advantages of convenient use, low cost and fast response. However, most of the studies are based on the in vitro level, and less research on the in vivo level. Therefore, in order to achieve rapid identification and drug treatment after biological infection, it is very necessary to develop rapid, in situ, non-invasive and sensitive detection methods for drug-resistant bacteria.

Based on this, a novel design and development strategy for gas-based probes (CS) is proposed. The probe was prepared by grafting volatile gas (2-methyl-3-mercaptofuran, MF) onto cephalosporin intermediate through nucleophilic substitution reaction. NMR and HRMS spectra analysis confirmed the successful preparation of the probe. The probe can react with Class A TEM-1 Bla to release MF molecules as a marker of corresponding drug-resistant bacteria, and the released gas was analyzed by headspace solid phase microextraction-gas chromatograph mass spectrometry (HS-SPME-GC-MS/MS). The results show that this method can be used for the qualitative and quantitative detection of drug-resistant strains in vivo, with a limit of detection 0.2 nM, which provides a new idea for the detecting enzyme activity and screening of drug-resistant strains in vivo, and is expected to realize the rapid diagnosis of drug-resistant bacteria and corresponding diseases.

PS1-71 Screen-printed DNA-based sensors for detection of the prostate cancer biomarker miR-21 – a feasibility study

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The miR-21 was reported as one of the potential microRNA (miRNA) biomarkers to be used as diagnostics tool for the detection of prostate cancer. Statistically relevant over-expression in comparison with healthy controls has been reported for this marker for urine samples of prostate cancer patients. [1]

In the present feasibility study, we are investigating the use of electrochemical detection based on screen printed electrodes and DNA-based hybridization recognition of the miRNA strand of interest. The results of the present work will be the basis for an evaluation of the present technology for the development of a non-invasive, simple to use, cost-effective and rapid point of need diagnostic system. This preliminary study is based on a DNA probe, designed analogously as similar literature reported probes used for the detection of nucleic acids of similar length (e.g. [2]). miR-21 was spiked in buffer solutions of different concentrations of NaCl, mimicking the range found in urine.

The selective binding of miR-21 to the DNA probe induces its conformational change, which displaces the electrochemical marker methylene blue. The signal is detected by square wave voltammetry. At a frequency of 15 Hz the sensors displays signal-on behaviour with a maximum signal gain of 96.0% ± 5.7% and an estimated dissociation constant (KD) of 137.7 ± 4.4 nM (n=4). The useful dynamic range (defined as the range from 10 to 90% of the maximum signal change) is from 55 nM to 343 nM with a limit of detection (LOD) of 31 nM (n=4).

This preliminary study will be followed by an extensive study aimed at the test and optimization of further parameters related to the target application in view, such as pH sensitivity, shelf life, stability of the biomarker in solution, etc. The analytical optimization will be followed by evaluating performance in real matrix, i.e. urine.

1. N. Ghorbanmehr, S. Gharbi, E. Korsching, M. Tavallaei, B. Einollahi, S. J. Mowla, miR-21-5p, miR-141-3p, and miR-205-5p levels in urine—promising biomarkers for the identification of prostate and bladder cancer, The Prostate. 2019, 79, 88–95
2. S. Cinti, G. Cinotti, C. Parolo, E.P. Nguyen, V. Caratelli, D. Moscone, F. Arduini, A. Merkoçi, Experimental Comparison in Sensing Breast Cancer Mutations by Signal ON and Signal OFF Paper-Based Electroanalytical Strips, Analytical Chemistry. 2020, 92, 1674-1679.

PS1-73 The impact of column hardware on efficiency in liquid chromatography

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Sample band broadening in the immediate vicinity of the column ends can be characterized by column reversal experiments [1]. In the case of packed columns, measurements have shown that the columns are heterogeneous and some differences can be observed between the two ends of the column.

Column reversal has peak compression effect, the peaks obtained with reversed flow are always narrower and more symmetric than those without flow reversal, and therefore column reversal is suitable for determining the local plate height values of the columns and for detecting the difference between the two column ends.

We can conclude that shorter columns are more axially homogeneous than longer columns, so that column length is an influential factor in the column packing procedure.

Column reversal was also attempted with macromolecules to eliminate the effect of pores. Similar conclusions could be drawn, however, due to the complexity of the measurements, we will continue to perform the column reversal with small molecules in the following.

In addition to this, electron microscopy measurements were also carried out, where the purpose of the measurements was to determine whether any damage was visible on the frit or on the particles as a result of the column loading procedure. The inlet and outlet frit of five types of columns were examined. None of the cases showed visible damage to either the frit or the particles. However, the silica gel particles were visibly entrapped in the pores of the frit due to their size and the heterogeneous structure of the pressed frit. The particles embedded in the frit may also be responsible for the band broadening effect near the frit.

PS1-74 Constructing Colorimetric Vernier Caliper for Distance-Based Self-Powered Signal Transduction Using an Array Optical Sensor

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Potentiometric ion-selective electrodes (ISEs) are well-established analytical tools for measuring a range of target ions because of their simple operation, low cost, fast response, and ability to be integrated with other technologies. Prussian Blue (PB) film electropolymerized onto a transparent

electrode is analytically useful and gives reversible colorimetric signals that track the potential of the coupled potentiometric probe.1,2 However, the absorbance-based readout using image capturing devices as an indispensable tool to analyse the colour signal, does not work sufficiently quantitatively with naked human eyes. More conducive to naked eyes detection, distance-based chemical sensors have been developed as a popular instrument free sensing technique in recent years. Here we introduce a new approach that realizes a distance-based chemical sensor by a caliper-type slider to connect different PB electrodes. When the connection was slid, the average potential would increase by use of a voltage divider. The colour of PB only starts to change when the voltage reaches a predetermined threshold. After this point, the PB colour saturates with a further increasing cell voltage. Sliding the connector changes the PB colour one by one, minimizing the transient charge flow across the ion-selective sensor.

[1] Jansod, S., Cherubini, T., Soda, Y., Bakker, E. Optical sensing with a potentiometric sensing array by Prussian blue film integrated closed bipolar electrodes. Anal. Chem., 2020, 92(13), 9138-9145.
[2] Jansod, S., Bakker, E. Self-powered electrochromic readout of potentiometric pH electrodes. Anal. Chem., 2021, 93(9), 4263-4269

PS2-01 SPME analysis of organic compound of Lactobacillus plantarum 17M and its antagonistic activity against Erwinia amylovora

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This research presents the results of the antagonistic action of Lactobacillus plantarum 17M, which was isolated from the phyllosphere of a garden, and collection strains of lactic acid bacteria from SPC of Microbiology and Virology against the pathogen of bacterial fire blight, Erwinia amylovora. The L. plantarum 17M strain was superior in antagonistic activity to the collection strains. Solid phase microextraction (SPME) was used to obtain organic compound profiles of the culture broth of L. plantarum 17M. Organic compounds in the headspace were adsorbed onto SPME fibers coated with 85 mm carboxen/polydimethylsiloxane (CAR/PDMS) (Supelco, Bellefonte, PA, USA). Sample preparation carried out in the 20mL vials, which was maintained at 30 °C for 30 min until it reached equilibrium. The Lactobacillus plantarum 17M strain can produce large amounts of certain major flavor compounds. In particular, fermentation by Lactobacillus strains generated characteristic aromatic compounds such as acetaldehyde, acetone, butan-2-one, butane-2,3-dione (diacetyl), and 3-hydroxybutan-2-one (acetoin) as fermented products [1]. In this study, acetic acid, lactic acid and 2,3-butanedione from L. plantarum 17M were established as active components against fire blight, and their contents were high in the culture broth, with relative abundances of 53.2±4.3%, 16.3±2.3% and 14,84 ±4.1%, respectively. In the L. plantarum 17M culture broth sample, in addition to acetic acid and lactic acid, other acids with low contents were identified, including propanoic acid, butanoic acid, pentanoic acid, hexanoic acid, heptanoic acid and octanoic acid. Analysis of the component composition of the main organic compounds of L. plantarum 17M revealed that the contents of acetic acid, with a value of 53.2±4.3%, lactic acid, with a value of 16.3±2.3%, and 2,3-butanedione, with a value of 14.84 ±4.1%, were high, and the presence of other organic compounds was detected. The inhibitory activity of various concentrations of acetic acid, lactic acid, and 2,3-butanedione against E. amylovora was studied. It was found that at high concentrations, these compounds completely suppressed the growth of the causative agent of fire blight. The concentrations of acetic and lactic

acids that had inhibitory activity against E. amylovora was 5%, which is nontoxic for apple plants, and 10% for 2,3-butanedione. This study confirms the potential use of L. plantarum 17M as active agents of microbial biopesticides to combat fire blight of fruit crops in Kazakhstan. The use of this biological product will optimize phytosanitary conditions for gardeners, reduce the pesticide load and obtain environmentally friendly products.

Kaseleht K. et.al., Int J Food Sci Technol. (2011) 1940–1946.

PS2-02 Evaluation of Phytocannabinoid Bioavailability Rates using the Caco-2 Cell Model

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Phytocannabinoids are biologically active secondary metabolites occurring in Cannabis sativa L. plants. Due to their therapeutic potential, attention has focused mainly on delta9-tetrahydrocannabinol (Δ9-THC) and its non-psychotropic isomer cannabidiol (CBD)[1]. In the recent decade, research studies have reported a number of other cannabinoids that also have interesting biological activities, for example antibacterial, analgesic, anticonvulsant, or neuroprotective; the examples are cannabigerol (CBG), cannabinol (CBN), cannabichromene (CBC) or cannabidivarin (CBDV)[2-4]. Cannabinoid acids (precursors of neutral phytocannabinoids) and other biologically active substances found in cannabis may also have these properties, but sufficient scientific information is not available. In addition to the effectiveness of individual substances, a synergistic effect was also observed[5].

In the current project, we have been investigating the anti-inflammatory effect of phytocannabinoids in the context of the potential to treat/prevent atherosclerosis. For this purpose, the in vitro bioavailability including possible biotransformation of phytocannabinoids (CBD, CBC, CBG, CBN, CBDV, and relevant acids) using a model system with Caco-2 cells (human colon adenocarcinoma cells) was investigated.

Cells were exposed either to phytocannabinoids or their mixture with some other non-cannabinoid components of cannabis extract obtained by preparative high-performance chromatography. To study the transfer of analytes, a Boyden chamber containing tissue culture medium Ham's Nutrient Mixture F12 and a monolayer of Caco-2 cells on a semipermeable membrane was used. After 24 hours of incubation, individual layers (apical medium, basolateral medium, and cells) were subjected to Analysis by ultra-performance liquid chromatography coupled with tandem mass spectrometry was used for both parent phytocannabinoids and potential metabolites.

Relatively significant differences between simulated bioavailability neutral phytocannabinoids and relevant phytocannabinoid acids were observed indicating that the polarity of respective compounds plays an important role. After 24 hours of incubation, neutral substances were retained to a greater extent in Caco-2 cells, and part of them was transferred to the basolateral medium. In contrast, phytocannabinoid acids were less retained by cells and reached the basolateral part. This trend was observed regardless of whether cannabinoids were applied alone or in combination with cannabis extract. Any metabolites of phytocannabinoids were not detected.

1. *Levinsohn, E.A. and K.P. Hill, Clinical uses of cannabis and cannabinoids in the United States. Journal of the Neurological Sciences, 2020. 411: p. 116717.*

2. Appendino, G., et al., Antibacterial cannabinoids from Cannabis sativa: a structure-activity study. J Nat Prod, 2008. 71(8): p. 1427-30.
3. Maione, S., et al., Non-psychoactive cannabinoids modulate the descending pathway of antinociception in anaesthetized rats through several mechanisms of action. Br J Pharmacol, 2011. 162(3): p. 584-96.
4. Hill, A.J., et al., Cannabidivarin is anticonvulsant in mouse and rat. Br J Pharmacol, 2012. 167(8): p. 1629-42.
5. Chacon, F.T., et al., Secondary Terpenes in Cannabis sativa L.: Synthesis and Synergy. 2022. 10(12): p. 3142.

PS2-03 Simultaneous analysis of 5 biomarkers of oxidative and nitrative stress in urine by SPE+HILIC-MS/MS

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The determination of oxidative and nitrative stress biomarkers plays a crucial role in comprehending the impact of diverse factors on biological systems. Biomarkers serve as reliable indicators of biological processes or conditions, and their accurate measurement is essential for assessing the impact of different stimuli. Among the commonly studied biomarkers, five key ones are malondialdehyde (MDA), 8-hydroxydeoxyguanosine (8-OHdG), 8-hydroxyguanosine (8-OHG), 3-nitrotyrosine (3-NT) and its metabolite 3-nitro-4-hidrosyphenylacetic acid (NHPA). MDA is a widely used biomarker for lipid peroxidation, an oxidative degradation process of polyunsaturated fatty acids. Elevated levels of MDA indicate cellular oxidative stress and damage. Likewise, 8-OHdG and 8-OHG are biomarkers associated with oxidative DNA or RNA damage. These biomarkers are employed to assess the extent of damage caused by reactive oxygen species (ROS). Finally, 3-NT and its metabolite NHPA are biomarkers associated with nitrative stress of proteins.

In this work a new confirmatory method has been developed based on HILIC-MS/MS for the simultaneous quantification of these five urinary biomarkers in urine samples. For those compounds present in urine at very low levels (3-NT and NHPA) a preconcentration step employing Isolute ENV+ (a hyper crosslinked hydroxylated polystyrene-divinylbenzene copolymer) as SPE sorbent has been developed. The developed method has been applied to the analysis of several urine samples, reaching limits of detection between 1.02 and 1.49 mg L -1 for 8-OHdG and MDA respectively (no SPE required) and 0.11 and 0.18 mg L -1 for NHPA and 3-NT (including SPE).

HILIC-MS/MS offered notable advantages in the analysis of these biomarkers in urine. Its capacity to retain and separate highly polar compounds, such as the five biomarkers here studied, makes it well-suited for their analysis in hydrophilic matrices like urine. Furthermore, the high sensitivity and selectivity of tandem mass spectrometry enables precise and reliable quantification of these biomarkers, by means of the standards addition approach, even at trace levels.

The application of HILIC-MS/MS in biomarker analysis contributes to our understanding of oxidative and nitrative stress, DNA, RNA, lipids and proteins damage, and related biological processes, bolstering research in various fields including biomarker research. environmental toxicology, clinical diagnostics, and personalized medicine.

PS2-04 Vibrio-Sequins - dPCR-traceable DNA standards for quantitative metagenomics of Vibrio spp.

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Vibrio spp. are a diverse group of ecologically important marine bacteria that have caused several foodborne outbreaks of gastroenteritis around the world. Their detection and characterization is moving away from conventional culture-based methods towards next generation sequencing (NGS)-derived metagenomics. However, metagenomic methods are relative in nature and are facing technical biases arising from library preparation and sequencing. Here, we developed a quantitative metagenomic approach enabling the quantification of Vibrio spp. at the limit of quantification (LOQ) through artificial DNA standards and their absolute quantification via digital PCR (dPCR). We present six newly developed DNA standards, called Vibrio-Sequins together with optimized TaqMan assays for their quantification via dPCR in individually sequenced DNA libraries. Therefore, we validated three duplex dPCR methods to quantify six targets. LOQs were ranging from 20 to 120 cp/µl for the six standards, whereas the limit of detection (LOD) was approximately at 10 cp/µl for all six assays. Subsequently, a quantitative metagenomics approach was applied to quantify Vibrio-DNA in a pooled mix of several Vibrio species. The method adds metrological traceability through the coupling of NGS and dPCR to existing quantitative metagenomic protocols. Hence, we significantly advance existing quantitative metagenomic methods by ensuring metrological traceability of NGS-based DNA quantification in environmental samples. Our method represents a useful tool for future metagenomic studies with the aim of quantifying microbial DNA in an absolute manner. The inclusion of dPCR into metagenomic methods now allows the development of statistical approaches for the estimation of measurement uncertainties (MU) for NGS, which is still in its infancy.

PS2-05 Kdo substitution and endotoxin quantification using the novel chemical Kdo-DMB-LC endotoxin content assay

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Endotoxins (ETs) are critical contaminations of e.g., injectables. If they enter the human blood stream, ETs initiate strong immune responses such as fever, or organ failure with potentially fatal outcome. All ETs contain the rare, almost ET specific sugar acid 3-deoxy-d-manno-oct-2-ulsonic acid (Kdo). It is used as biomarker for ET quantification in our chemical Kdo-DMB-LC ET content assay [1].

Kdo is released during mild acidic hydrolysis from the ET core without breaking other e.g., neutral sugar bonds. It is derivatized with the fluorophore 1,2-diamino-4,5-methylenedioxybenzene-2 HCl (DMB) to enable its chromatographic separation from matrix compounds such as sialic acids by RP-HPLC and sensitive fluorescent detection. ET quantification is based on an external Kdo and / or ET standard [2]. Like sialic acids, Kdo can be non-stoichiometrically modified with functional groups such as phosphoethanolamines (PEtN), phosphates, different hexoses, or amino acids. Those Kdo derivatives are separated by the Kdo-DMB-LC assay. In this work, the impact of Kdo derivatization on ET quantification was investigated for different ET standards. The large S-type ETs standard of Pseudomonas aeruginosa 10 only has underivatized Kdos whereas Escherichia coli O55:B5 bears additionally Kdo modified with PEtN (15%). Kdo-PEtN was also found in the small R-type ET standards of E. coli EH100 (31%) and E. coli F583 (13%). They have in addition hexose modifications such as galactose (E. coli EH100, 33%) and D-glycero-D-manno-heptose (E. coli F583, 45%).

For all, Kdo and Kdo-derivates, the release in dependence on the hydrolysis time is described by a saturated curve. Hydrolysis kinetics for the same Kdo-derivates found in different ET standards are the same. Different Kdo-derivates showed differences in their release kinetics. Unmodified Kdo was released the fasted followed by Kdo-Gal, Kdo-PEtN, and Kdo-Hep. Since all Kdo-derivates and Kdo show a constant maximum of signal versus release time, total ET content determination can be performed at the 100% release time of the Kdo-derivate with the slowest release kinetics, e.g., Kdo-Hep. Assuming the same fluorescence intensity for all Kdo-derivates a 100% recovery of the Kdo content was achieved for the small ETs from E. coli F583 and E. coli EH100.

In conclusion: Quantification of the total ET content can be performed in case of an available ET standard for the ET in question based on the unmodified Kdo alone. In case of unknown ET composition and no standard, all Kdo-derivates must be used for the calculation of the total ET content.

[1] A. Hoffmann et al., A Novel Chemical Assay that Brings Endotoxin Analysis into the 21st Century. CHIMIA, 2022, 76(11), 966; DOI: 10.2533/chimia.2022.966
[2] B. Bucsella et al., Novel RP-HPLC based assay for selective and sensitive endotoxin quantification. Anal. Methods, 2020, 12, 4621-4634; DOI: 10.1039/d0ay00872a

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PS2-06 ERROR PROPAGATION STUDIES IN microRNA QUANTIFICATION

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MicroRNAs (miR) constitute an extremely important class of new biomarkers at the forefront of bioanalysis as they regulate fundamental biological functions. Hence, more and more assays are being reported for miR quantification in various clinical samples. By reviewing the extensive recent literature, we observed a lack of data regarding the ability of the methods to distinguish small changes in miR concentration (within the range of linear response) although this is particularly important for monitoring disease progression and patient's response to treatment. In the present work, calibration data were collected from 90 papers published in high impact factor journals, including Anal. Chem., Biosens. Bioelectron., Sens. Act. B, etc. In the reported calibration graphs, the signal (S) is either a linear function of the logarithm of miR concentration (S vs. logC) or a linear function of concentration (S vs. C). The majority (81%) of the methods that are based on S vs. logC graphs present a wide linear response, covering several orders of magnitude. However, the corresponding change of the signal is relatively small. Consequently, a large change in miR concentration is required to cause a statistically significant change in the signal. The calibration data were used to calculate, in each case, the minimum distinguishable change (MDC), expressed as fold (F), in miR concentration. We found that F>3 in 45% of the papers based on S vs. logC graphs and extends up to F=17 (i.e., MDC in concentration >200% and >1600% respectively). In methods based on S vs. C graphs, F ranged from 1.1 to 1.6. Furthermore, we estimated the LODs from S vs. C calibration equation data and compared with the reported LODs. For most of the methods the reported LODs, were ≥10 times lower than the estimated ones. Also, we show the effect of (a) the signal uncertainty and (b) the slope of the S vs. logC calibration graph on the MDC in miR concentration. Our results were confirmed by Monte Carlo simulations. It is anticipated that our study will provide a guide, for the selection, from a plethora of reported miR assays, those that are more suitable for clinical laboratory practice.

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PS2-07 Method development for detection and determination of carotenoids in the cap of the middle spotted woodpecker

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The middle spotted woodpecker (Dendrocoptes medius L.) is a species from the woodpecker family, not very numerous in Poland, which is distinguished by a red cap on the head of representatives of both sexes. The color of these feathers is due to carotenoids, water-insoluble compounds with a chain structure consisting of isoprene units, that gives a series of conjugated C=C bonds in their structure. At the ends of this chain there may be two carboxyl groups, ester groups, cyclohexyl rings, as well as its carbonyl and/or hydroxyl derivatives. However, carotenoids are not synthesized by the middle spotted woodpecker but are supplied to it with food (woodpecker feeds mainly on insects and their larvae dug out of rotten wood or simply collected from the surface of bark and leaves, as well as on seeds and fruits). It has been noted that the staining degree of the red cap feathers of the middle spotted woodpecker varies with the availability of food. Thus, the research is based on the hypothesis that the concentration of carotenoids in the feathers of the middle spotted woodpecker reflects the condition of the individual, while the number of carotenoid derivatives may depend on the degree of diversity of its food.

The aim of the research was to develop a method to track changes in the content of carotenoids extracted from the feathers of the middle spotted woodpecker, so that in the longer term, the work can be extended to research on (i) the impact of the condition of individuals on the content of carotenoids in feathers, and (ii) transformations of carotenoids in the body of the woodpecker or (iii) taxonomic relationships of different species. Due to the very small amounts of samples (the weight of one feather is between 0.5 to 1.0 mg), as well as the complex composition of carotenoids present in the feathers, it was necessary to use appropriate separation techniques combined with sensitive and selective detection techniques. Therefore, the research was carried out using reversed-phase high-performance liquid chromatography combined with spectrophotometric detection and tandem mass spectrometry detection with electrospray ionization (HPLC-UV-Vis-ESI-MS/MS). Research involved optimizing the separation, ionization and fragmentation of eight carotenoids, which, due to their highly non-polar nature of this group of compounds, was a key step in the development of the analytical method.

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PS2-09 New challenges in early diagnosis of cancer

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Early diagnosis of cancer is the only hope for fast and full recovery from this disease. While conventional methods of analysis such as ELISA, chemiluminescence are not able to determine more

than one biomarker in a run, and also their working concentration range is narrow, there is a high need to develop new screening methods able to determine more than one biomarker in any stage of the development of cancer.

Therefore, we proposed 3D stochastic microsensors and miniplatforms which had the advantage of performing reliably the qualitative analysis of biomarkers (in the molecular recognition step) as well as the quantitative analysis of the biomarkers in the quantitative step.

In this regard, there will be discuss the role of enantioanalysis of amino acids as key factor in the early diagnosis of cancer. Wide working concentration ranges and low limits of determination were obtained for the amino acids assay in whole blood and tumoral tissues.

PS2-10 Hyphenated MS-Methods as a Tool for Orthogonal Metabolite Annotation in On-Line Breath Analysis with SESI-HRMS

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On-line breath analysis with secondary electrospray ionization (SESI) coupled to high-resolution mass spectrometry (HRMS) is a powerful method for rapid and non-invasive examination of the human metabolome. Various clinical research projects have shown the effectiveness of this technique. [1] However, the identification of metabolites and biomarkers using SESI-HRMS is still limited due to the lack of a hyphenated separation method before the MS analysis. Comparing annotated metabolites found in exhaled breath condensate (EBC) with detected features in on-line data can merge this gap and serve as a step toward building a database of metabolites for the human exhalome.

For the comparison, the breath of 16 healthy adults was measured on-line for 10 days with a SESI source (Fossil Ion Tech, Spain) coupled to a Q-Exactive Plus Orbitrap mass spectrometer (Thermo Fischer Scientific, Germany) while also simultaneously condensing part of the exhaled breath. EBC was analyzed using an Acquity UPLC system (Waters Corporation, USA) coupled to the same mass spectrometer, employing a reverse phase and a hydrophilic interaction column. In addition, a data-independent MS2-acquisition method derived from PAcIFIC[2] was utilized. GC-MS analysis was carried out using dynamic headspace vacuum transfer in-trap extraction coupled to GC-MS (DHS-VTT GC-MS) [3]. On-line and EBC MS2 data were processed by custom workflows and EBC MS1 data by standard workflows.

An immense number of features were obtained in the LC-MS analysis, especially with the MS2 method. Employing CANOPUS, the features were assigned chemical classes, revealing that primarily amino acids, amines, and carboxylic acids in the negative mode were detected. The GC-MS analysis mainly revealed compounds of external origin, such as additives from oral hygiene products like menthol or other fragrances and flavoring agents. Before sampling, food, and beverage consumption restrictions will be needed to mitigate this issue. Comparison of the detected off-line features of both methods with the detected on-line features showed partial overlap; however, the m/z range from 150-200 needs to be better covered by the off-line techniques. While these results demonstrate the potential of combined LC-MS and GC-MS analysis of breath condensate as an orthogonal annotation tool for SESI-HRMS, additional on-line fragmentation is needed for annotation with higher confidence.

[1] Bruderer, T. et al., On-Line Analysis of Exhaled Breath. Chemical Reviews 2019, 119 (19), 10803-10828
[2] Panchaud, A. et al., Precursor Acquisition Independent From Ion Count: How to Dive Deeper into the Proteomics Ocean. Analytical Chemistry 2009, 81 (15), 6481-6488
[3] Fuchsmann, P. et al., Development and performance evaluation of a novel dynamic headspace vacuum transfer "In Trap" extraction method for volatile compounds and comparison with headspace solid-phase microextraction and headspace in-tube extraction. Journal of Chromatography A 2019, 1601, 60-70

PS2-11 New nanopores sensors for the detection of DNA

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Aerolysin-like proteins are a family of β-pore-forming toxins (PFTs) which are widely present in all kingdoms of life. Recently, this family of proteins is gaining attention because of their biotechnological application as nanopore sensors for the sensing and sequencing of biomolecules. To identify novel and more sensitive PFTs for DNA and peptide sensing, we explored the possibilities of using the knowledge of the sequence and structure of proteins to screen and explore new potential nanopore candidates. In spite of the conserved structural fold within the aerolysin family, the sequence identity is very low and complicates their sequence alignment, hindering into the understanding of their pore structure and properties, limiting further biotechnological applications. We analyzed the pore structure of three family members, Clostridium perfringens epsilon toxin (ETX), Laetiporus sulphureus lectin (LSL) and Bacillus thuringiensis parasporin-2 and compared it to aerolysin. Their structure and sensing capabilities for ssDNA were first assessed by in silico methods and further explored experimentally for ETX. We found that 3 types of ETX pores with different conductance can be formed and that only one of them is able to translocate DNA. Moreover, the depth of the current blockage is higher compared to aerolysin, which indicates a higher sensitivity for DNA sensing. Our findings open a new venue for improving and diversifying nanopore capabilities for diverse molecular sensing.

PS2-12 A novel Cu(II)-Schiff base complex catalyzed synthesis of Synthesis of Benzamide Derivatives via C-H Bond Functionalization of Arenes

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Abstract

A novel Cu(II)-Schiff base nano-particles has been prepared using the bidentate Schiff base ligand namely, N-(3,4-dimethylphenyl)-(pyridine-2-ylmethylene)amine and CuCl2 was prepared and characterized using FT-IR, UV–vis, 1H NMR spectroscopy, elemental analysis and X-ray crystallography. The catalytic potential of the complex was evaluated in preparation of benzamide derivatives via C-H bond functionalization of arenes. All products were successfully formed in high yields.

Keywords:
Cu(II)-Schiff base complex; Pyrimidines; Nano-Particles; Pyran; X-ray crystallography

1. Diederich, F.; Stang, P. J. Metal-Catalyzed Cross-coupling Reactions; Wiley-VCH: Weinheim, 1998.
2. De Meijere, A.; Diederich, F. Metal-Catalyzed Cross-Coupling Reactions; Wiley-VCH: Weinheim, 2004.

3. Brennfuuhrer, A.; Neumann, H.; Beller, M. Angew. Chem. Int. Ed. 2009, 48, 4114.
4. Sambiagio, C.; Marsden, S. P.; Blackera, A. J.; McGowan, P. C. Chem. Soc. Rev. 2014, 43, 3525.

PS2-13 Granulometric characterization and quantification of TiO2 nanoparticles in pharmaceutical products by single particle ICP-MS

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Titanium dioxide (TiO2) particles are widely used in drug products mainly to obtain a white and glossy coating onto tablets. However, their potential toxicity raises safety concerns that have not been yet considered by pharmacopeias. As some medicines are administered daily, they may pose the same risk as that associated with the presence of the additive E171 in food. In this case, the European Food Safety Authority (EFSA) recently concluded that the nanoparticle fraction contained in this additive can cause potential immunotoxicity, inflammation, and neurotoxicity and therefore cannot be considered as safe. Since these effects depend mainly on the TiO2 particle size and concentration, the determination of these two parameters is crucial to evaluate the exposure level and then establish adapted risk management procedures. In this study, a single-particle inductively coupled plasma mass spectrometry (spICP-MS) method was then developed for granulometric characterization and quantification of TiO2 particles in pharmaceuticals. First, to obtain a quantitative extraction, several extracting reagents were tested such as water, surfactants (HMPNa and SDS), and alkaline solutions (TMAH) using cardiovascular pills containing titanium dioxide in the list of ingredients. All these conditions gave quantitative extraction yields showing that the particles are present on the surface of this drug and are easily extracted. Because of its ease of use, water was then retained and extraction was optimized considering different influencing parameters such as the solid/liquid ratio, the sonication procedure (ultrasonic bath or ultrasonic probe), and the sonication duration. In the optimized conditions corresponding to a solid/liquid ratio of 1/20 (1 tablet per 20 mL of water) with agitation in an ultrasonic bath for thirty minutes, an extraction recovery of 90 ± 1 % was obtained showing that the developed procedure allowed a complete extraction of the particles. The TiO2 particle concentration in the studied drug was 1.52 ± 0.09 g/kg with a median diameter of 181 ± 3 nm and the nanoparticle fraction (< 100 nm) was 19 ± 2 %. After validation, the method developed thus made it possible to show the significant presence of nanoparticles in the drug studied. It was applied to other drugs administered for chronic diseases and similar results were obtained. This study shows the importance of not neglecting this contribution of nanoparticles in the daily intake and must be taken into account in risk studies.

PS2-14 Analytical chemical characterization of engineered nanomaterials in complex sample matrices

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A variety of analytical chemistry techniques and methodological approaches are used for isolation/purification and determination of the composition of pristine engineered nanomaterials (ENMs) and for the detection, identification, and quantification in nano-enabled consumer products. The ENMs assessment frequently requires interdisciplinary approaches and multi-modal analysis methods. Metrology of nanomaterials, including available reference materials and the development and validation of standardized methods become available to address characterization and analysis. Analytical methods applied to analysis in complex matrices of environmental samples, food, cosmetics, and samples of biological origin as well as those used to monitor the fate of ENMs in the environment and biological systems are reported. There is a rapid development on the field mostly in the stage of accumulation of factual material.

It can be concluded that: (i) there is a fundamental difference between an analyte being bulk material and nanomaterials, (ii) together with the determination of chemical composition it is necessary to evaluate the physical parameters including size, shape, and structure important for specific application area, (iii) validated and standardized methods and reference materials are required to support quality control of existing products and risk assessment for regulation, (iv) many methods capable of analyzing chemical composition with the nanoscale resolution are not routinely available or applied (analysis of particles with a complex core/shell structure and surface coatings).

Among challenges are: (i) techniques that introduce minimal artifacts of sampling to the analysis are needed, (ii) the development of non-denaturing sample preparation and/or de-agglomeration methods and, on-line fractionation / pre-concentration techniques, (iii) analytical techniques that differentiate between manufactured ENMs and their possible transformations and fate.

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References:
J. Labuda, J. Barek, Z. Gajdosechova, H. Goenaga-Infante, L. J. Johnston, Z. Mester, S. Shtykov, Analytical chemistry of engineered nanomaterials: Part 1. Scope, regulation, legislation, and metrology (IUPAC Technical Report), Pure Appl. Chem. 2023, 95(2), 133-163. https://doi.org/10.1515/pac-2021-1001
https://doi.org/10.1515/pac-2021-1001

PS2-15 Development of a nanoparticle-based lateral flow assay for malaria prognostic

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Introduction. Malaria is the leading cause of fever in international travelers and in the 2-23% of the cases produces severe symptoms (including death)(1,2). Although appropriate treatment can almost entirely reduce mortality, current methods for prognosing malaria, which assess the risk of developing severe symptoms, are not reliable(3). Therefore, current clinical guidelines recommend hospitalization for all malaria-positive patients, leading to the unnecessary use of healthcare resources for over 75% of patients. Therefore, a tool able to precisely prognose malaria during the initial visit would allow for a better patient management. In order to provide a solution for this need, we developed a lateral flow assay (LFA) for the detection of two prognostic biomarkers: angiopoietin-1 (ANG1) and angiopoietin-2 (ANG2)(4–6).

Methods. First, in order to identify the cut-off levels of three host biomarkers (ANG1, ANG2 and sTREM) reportedly associated with malaria severity(4–8), we analyzed a retrospective cohort of 132 patient samples using commercially available ELISA kits. Then, following a recently published protocol(9), we developed a LFA for the detection of ANG2 and ANG1, whose ratio provided the best prognostic performance. Specifically, we designed and developed both competitive and non-competitive assays using gold nanoparticles as colorimetric labels(10). To optimize the assay, we tested 12 different antibodies, 3 nitrocellulose membranes, and 3 running buffers. Finally, we used the plot profile function of the ImageJ software to quantify the test results.

Results. From the analysis of the biomarker levels, the ANG2/ANG1 ratio provides the best performance with an area under the ROC of 0.82, which is similar to the values obtained by the current gold-standard, laboratory-based microscopic method. The measurements of the developed LFA was able to detect concentrations of ANG1 between 50 ng/ml to 1000 ng/mL and of ANG2 between 40 ng/ml and 1000 ng/ml(11), concentrations respectively within the clinical relevant range of ANG1 (5-200 ng/mL) and just above the one of ANG2 (2-15 ng/mL). To test the specificity of the antibodies used in the LFA, we employed ANG1 to test the anti-ANG2 antibodies and ANG2 to test the anti ANG1-antibodies. The results confirmed that the developed LFA is specific for both targets. The use of the ImageJ software was feasible for the quantification of the biomarker concentrations.

Conclusions. We believe that developing and implementing more point-of-care prognostic devices is essential to shift from the common 'one-size-fits-all' approach to precision medicine, which allows for personalized patient management and optimizes available healthcare resources.

1. World Health Organization. World malaria report 2020 - 20 years of global progress & challenges. 2020.

2. Bruneel F, Tubach F, Mira JP, Houze S, Gibot S, Huisse MG, et al. Correction to: Imported falciparum malaria in adults: host- and parasite-related factors associated with severity. The French prospective multicenter PALUREA cohort study. Intensive Care Med [Internet]. 2018 Jan 1 [cited 2021 Nov 15];44(1):141–3. Available from: https://pubmed.ncbi.nlm.nih.gov/29138897/

3. Krampa FD, Aniweh Y, Kanyong P, Awandare GA. Recent Advances in the Development of Biosensors for Malaria Diagnosis. Sensors (Basel) [Internet]. 2020 Feb 1 [cited 2021 Nov 8];20(3). Available from: /pmc/articles/PMC7038750/

4. Balerdi-Sarasola L, Parolo C, Cruz A, Subirà C, Rodriguez-Valero N, Almuedo-Riera A. Parasite and host biomarkers for the diagnosis of severe Plasmodium falciparum malaria in non-endemic. 32nd ECCMID. the European Congress of Clinical Microbiology and Infectious Diseases. Lisbon, Portugal; 2022.

5. Lovegrove FE, Tangpukdee N, Opoka RO, Lafferty EI, Rajwans N, Hawkes M, et al. Serum angiopoietin-1 and -2 levels discriminate cerebral malaria from uncomplicated malaria and predict clinical outcome in African children. PLoS One [Internet]. 2009 Mar 20 [cited 2022 Jan 20];4(3). Available from: https://pubmed.ncbi.nlm.nih.gov/19300530/

6. Gomes LT, Alves- ER, Rodrigues-Jesus C, Nery AF, Gasquez-Martin TO, Fontes CJ. Angiopoietin-2 and angiopoietin-2/angiopoietin-1 ratio as indicators of potential severity of plasmodium vivax malaria in patients with thrombocytopenia. PLoS One. 2014 Oct 2;9(10).

7. Conroy AL, Lafferty EI, Lovegrove FE, Krudsood S, Tangpukdee N, Liles WC, et al. Whole blood angiopoietin-1 and-2 levels discriminate cerebral and severe (non-cerebral) malaria from uncomplicated malaria. Malar J [Internet]. 2009 Dec 15 [cited 2022 Jun 17];8(1):1–7. Available from: https://malariajournal.biomedcentral.com/articles/10.1186/1475-2875-8-295

8. Balanza N, Erice C, Ngai M, Varo R, Kain KC, Bassat Q. Host-Based Prognostic Biomarkers to Improve Risk Stratification and Outcome of Febrile Children in Low- and Middle-Income Countries. Front Pediatr. 2020 Sep 18;8:595.

9. Parolo C, Sena-Torralba A, Bergua JF, Calucho E, Fuentes-Chust C, Hu L, et al. Tutorial: design and fabrication of nanoparticle-based lateral-flow immunoassays. Vol. 15, Nature Protocols. Nature Research; 2020. p. 3788–816.

10. Sena-Torralba A, Álvarez-Diduk R, Parolo C, Piper A, Merkoçi A. Toward Next Generation Lateral Flow Assays: Integration of Nanomaterials. Chem Rev [Internet]. 2022 Sep 28 [cited 2023 Apr 27];122(18):14881–910. Available from: https://pubs.acs.org/doi/full/10.1021/acs.chemrev.1c01012

11. Sena-Torralba A, Torné-Morató H, Parolo C, Ranjbar S, Farahmand Nejad MA, Álvarez-Diduk R, et al. A Novel Ratiometric Fluorescent Approach for the Modulation of the Dynamic Range of Lateral Flow Immunoassays. Adv Mater Technol [Internet]. 2022 Aug 1 [cited 2023 Apr 27];7(8):2101450. Available from: https://onlinelibrary.wiley.com/doi/full/10.1002/admt.202101450

PS2-16 Studying the entropic pulling of Hsp70/DnaK at the single-molecule level using a biological nanopore

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The Hsp70 family of ATP-dependent chaperons is essential in cellular networks to ensure protein quality. Despite their crucial roles in areas such as prevention of protein aggregation and transmembrane transport, the underlying mechanism of their functions is so far only theoretically described in the entropic pulling model[1]. Due to a lack of suitable methods, no experimental data has proven or quantified this pulling mechanism. Recently, nanopores have emerged as a novel approach to analyze the function of motor enzymes[2], as well as protein-protein interactions[3]. Here, we designed a system that uses a biological nanopore to investigate the physical mechanism of the molecular motor DnaK (Hsp70 isoform in E. Coli) on the single-molecule level. In nanopore measurements, we observed DnaK pulling against defined voltage-induced forces on a substrate that is trapped inside the pore. Comparison between the behavior of the substrate in the presence and absence of DnaK support the entropic pulling as physical mechanism of the motor. Furthermore, the escape experiments were used to quantify the pulling force of DnaK. These results provide a direct observation of the entropic pulling mechanism and emphasize the utility of nanopores for studying protein function at the single-molecule level.

[1] Sousa, R. & Lafer, E. M. The Physics of Entropic Pulling: A Novel Model for the Hsp70 Motor Mechanism. Int J Mol Sci 20, E2334 (2019).
[2] Laszlo, A. H., Derrington, I. M. & Gundlach, J. H. MspA nanopore as a single-molecule tool: From sequencing to SPRNT. Methods 105, 75–89 (2016).
[3] Thakur, A. K. & Movileanu, L. Real-time measurement of protein-protein interactions at single-molecule resolution using a biological nanopore. Nat Biotechnol 37, 96-101 (2019)

PS2-17 DFT and dynamics simulation studies to understand probing of folic acid using β-cyclodextrin functionalized copper nanoclusters and vitamin B6 cofactor pyridoxal by displacement approach

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The β-cyclodextrin (β-CD) functionalized nanomaterials are widely employed for the development of fluorescent nanochemosensors for the detection of bioactive analytes.1,2 The hydrophobic cavity of β-CD formed inclusion complexation with the target analyte that perturbed the optical properties of nanomaterials through aggregation of nanoparticles and electron or energy transfer phenomenon.3 Herein, green emissive β-CD capped copper nanoclusters (β-CD-CuNCs) was synthesized and inclusion complexation was studied with the vitamin B6 (VB6) cofactor pyridoxal (PL). The addition of PL formed inclusion complex with the functionalized β-CD and induced a significant fluorescence enhancement at 435 nm. The PL decorated β-CD-CuNCs was employed for the detection of folic acid (FA). The

addition of FA displaced the PL from the β-CD because of the high inclusion complexing ability of FA than PL. DFT and molecular dynamics simulations were performed to complement the experimental evidence. Using the PL decorated β-CD-CuNCs, the concentration of FA can be detected down to 0.47 µM and the practical utility was validated by quantifying FA in blood serum.

Keywords: Host-Guest chemistry; Inclusion complexation; β-cyclodextrin; DFT and dynamics simulation, Fluorescent probe.

References

1. *S. K. Sahoo, Sensing and biosensing with optically active nanomaterials, Elsevier 2022, ISBN: 9780323902441.*
2. *S. K. Sahoo, M. Reza Hormozi-Nezhad, Gold and silver nanoparticles: synthesis and applications, Elsevier 2023, ISBN: 9780323994545.*
3. *Sahoo et al., Decorating vitamin B6 cofactor over beta-cyclodextrin stabilized silver nanoparticles through inclusion complexation for fluorescent turn-on detection of hydrazine, ACS Applied Bio Materials, 2020, 3, 10, 7021–7028; ACS applied Nano Materials, 2021, 4, 6231-6238.*

PS2-18 Direct and Selective Quantification of Cr(VI) in Waste Waters using Raffinose Capped Silver Nanoparticles as Sensitive Optical Sensor

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Chromium is a relatively abundant element in the earth's crust, existing predominantly as Cr(III) compounds. The Cr(VI) compounds are also found in nature at much lower quantities as a result of natural oxidation of Cr(III) in the presence of manganese minerals. The biological activity of Cr(III) and Cr(VI) species, their chemical behavior and toxic effects are quite different: Cr(III) is nontoxic and involved in several biochemical processes; Cr(VI) is toxic to most living organisms. The industrial application of Cr(VI) compounds is still very high, leading to release of toxic Cr(VI) species and calls for strict analytical control of Cr(III) and Cr(VI) content in wastes, waste waters and other environmental matrices. That is why the environmental quality standards and individual permissible limits for waste waters, which have been introduced through European and national legislations, require reliable speciation analysis of Cr or at least selective quantification of the highly toxic Cr(VI) species.

In the present study, a rapid, easy, and time-efficient sensing strategy for direct and selective determination of Cr(VI) in water samples at pH 4 is presented using raffinose capped silver nanoparticles (Ag/Raff NPs) as a simple optical LSPR based sensor. The nanoparticles used are prepared by a green synthesis procedure utilizing raffinose as both reducing and capping agent. The analytical method is based on the variation of LSPR absorption band intensity as a result of electrostatic interaction between the negatively charged Ag/Raff NPs and positive Cr(III) ions, in-situ produced by chemical reduction of Cr(VI) with ascorbic acid, combined with the fast kinetics of Cr(III) coordination to the –OH groups of raffinose on the nanoparticle surface, causing further the nanoparticle aggregation. Careful optimization of sensor response time (only 5 min) permits accurate and reliable determination of Cr(VI) in the presence of excess concentrations of Cr(III) without any preliminary separation of both species. The interference studies, performed in the presence of various metal ions, show very good selectivity of Ag/Raff NPs toward Cr(VI) species. The calibration curve for Cr(VI) is linear in the range 2.5 - 7.5 µmol/L, limit of quantification achieved is 1.9 µmol/L, and values of relative standard deviation vary from 3 to 5 % for concentration level 1.9 - 7.5 µmol/L. The added-found method is used to confirm accuracy and precision of developed analytical procedure. The analytical

approach developed is suitable for routine application and might be used also as a fast screening method on site.

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PS2-19 Quality by Design Approach for a Multicomponent Quantification Using HPLC-PDA and HPLC-MS: Application to Dosage Form and Biological Body Fluids

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A multicomponent pharmaceutical that contains loratadine, paracetamol, and pseudoephedrine was quantified using HPLC-PDA. The three analytes were well-separated and quantified in the dosage form on a C-18 column using a gradient mobile phase. A quality by design strategy was followed to achieve the challenging separation. Screening and optimization steps were carried out to investigate the effect of many factors on the studied responses with a minimum number of runs. The ANOVA of the factorial model showed that % acetonitrile (factor A), flow rate (factor B), and pH (factor C) were significant. The detection of the analytes' peaks was carried out using a PDA detector at 248nm for loratadine and paracetamol, and 214 nm for pseudoephedrine. The second method was SPE-HPLC-MS, where the three analytes and desloratadine, the active metabolite of loratadine, were quantified in spiked plasma and urine, using betamethasone valerate as an internal standard. The recovery of the analytes from body fluids was above 96%, and the LOQ was below 0.5 ng/mL. The validation of the developed HPLC-PDA method was achieved as per ICH guidelines, whereas the HPLC-MS method was validated according to FDA guidelines for bioanalytical method validation. The results were compared with the reported method, and no significant differences were found.

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PS2-20 Expanding the exposomics toolbox towards metals

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The emerging field of exposome research aims to comprehensively assess and understand the totality of environmental exposures and their effects on human health. [1] By considering the complex

interplay of various environmental factors, including chemical pollutants, lifestyle choices, and social determinants, the goal is to provide a holistic perspective on the factors influencing human health and disease. However, while many recent exposome studies focus on organic chemical pollutants, heavy metals and (potentially) toxic trace elements have received rather little attention, despite their ubiquitous occurrence and wide array of known detrimental effects on human health.
Therefore, we aim to establish analytical workflows based on inductively coupled plasma mass spectrometry (ICP-MS) for the determination of a broad range of metals, metalloids and non-metals in biological samples. The complexity of sample preparation will be reduced as much as possible, to enable a smooth integration into existing exposomics workflows. Further, focus will be given to high throughput and at the same time minimized sample consumption, considering the requirement to work with small scale samples, such as human tissue biopsies or body fluids.

On the presented poster, key aspects of these goals will be demonstrated on the example of human placenta samples. Although humans are exposed to xenobiotics during their whole lifetime, the time span of fetal development and early childhood is a specifically critical phase, because the immune system and the metabolism are still developing and are therefore more sensitive to exposure. [2] For a first proof-of-principle study, placenta samples of 12 healthy participants were taken directly after birth. [3] Small aliquots were digested with nitric acid in a microwave-assisted pressurized system and analyzed for around 50 elements with ICP-MS/MS. In parallel, extracts were prepared for the nontargeted analysis of organic pollutants. Aliquots of these extracts were further diluted and then subjected to analysis via ICP-MS/MS as well. The results were compared, and the workflows evaluated for their suitability for routine, high-throughput nontargeted metal(loid) analysis within an established exposomics workframe.

[1] C.P. Wild, Int. J. Epidemiol. 41 (2012) 24–32. https://doi.org/10.1093/ije/dyr236
[2] M. Krausová et al., Annu. Rev. Pharmacol. Toxicol. 63 (2023) 517-540. https://doi.org/10.1146/annurev-pharmtox-051922-113350
[3] B. Warth et al., Environ. Health Perspect. 127 (2019) CID: 107004. https://doi.org/10.1289/EHP4860

PS2-21 3D stochastic microsensor based on graphene for the simultaneous determination of p53, HER-3, and HER-4

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The 3D stochastic microsensor was proposed as a new tool for the fast screening of biological samples. The design of the stochastic sensor was based on NB dop-10 graphene. NB dop-10 graphene solvothermal synthesis was done followed by its morphological characterization. Biological fluids (whole blood, gastric tumor tissue, saliva, and urine) were screened using the proposed microsensors for selected biomarkers: p53, HER-3, and HER-4. The biomarkers were determined simultaneously in the biological samples, based on their specific signatures. High sensitivities and wide linear concentration ranges, as well as low limits of quantification, were achieved. The student t-test showed that the suggested 3D stochastic sensor may be employed reliably for the simultaneous determination of p53, HER-3, and HER-4 in whole blood, gastric tumor tissue, saliva, and urine. The proposed sensor was thus validated using real biological samples.

PS2-22 Stochastic sensors as new tools for the assay of CA72-4, CA19-9, CA12-5 and CEA in biological samples

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Three diagnostic biomarkers, namely carcinoembryonic antigen (CEA), carbohydrate antigen 19-9 (CA19-9), and carbohydrate antigen 72-4 (CA72-4), are utilized for the diagnosis of gastric cancer (GC) [1]. The correlation between the elevation and the incidence, reappearance, and spread of GC has been extensively studied. Recent research has demonstrated that serum tumor markers, particularly CA12-5, possess significant clinical value in the identification of GC. According to a recent study, the combination of CEA, CA19-9, CA72-4, and other biomarkers may offer enhanced precision in the diagnosis of GC patients [2]. Stochastic sensors have been employed in the field of biomedical research due to their ability to accurately and quantitatively analysis a variety of biological samples [3]. Therefore, this paper proposes the utilization of two stochastic sensors based on graphenes decorated with heteroatoms used for the simultaneous determination of CA72-4, CA19-9, CA12-5 and CEA in various biological samples. The conductivity of the matrix material, specifically graphene, was enhanced through the selection of N- and S-doped graphene. The utilization of graphene material was found to be more convenient due to its high signal stability and greater reproducibility of measurements when compared to graphite pastes. Oleamides (used as modifiers for the design of the stochastic sensors) are a novel class of materials that exhibit a three-dimensional "V" conformation, resembling the required pores essential for the stochastic sensor design. The proposed sensors exhibited high sensitivities, low limits of quantification, and wide working concentration ranges, enabling the simultaneous assay of CA72-4, CA19-9, CA12-5 and CEA despite the stage of gastric cancer.

[1] T.H. Zheng, et al., Crit. Rev. Eukaryot. Gene Expr. 25 (2015) 299.
[2] L. Guo, et al., J. Gastrointest. Surg. 25 (2021) 2470.
[3] R.I. Stefan-van Staden, et al., Symmetry 15 (2023) 958.

PS2-23 Comparison of different sample preparation techniques for degradation products of nerve agents in biological fluids

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Nerve agents such as sarin and VX are highly toxic organophosphorus compounds that inhibit the activity of cholinesterase. Their production and storage have been controlled by international law(1). Moreover these compounds have been used in various crimes(2). Therefore reliable analysis of these compounds is important for appropriate documentation, remediation and case control. Although various on site detection equipment have been developed to confirm the presence of these compounds. Most nerve agents are easily metabolized in human body and it is difficult to detect agents in long period. Thus, there is a great importance to detect metabolized products to confirm nerve agents and also detection of nerve agent metabolites in body fluids is important to treat rapidly victims.

In literature various analytical methods have been developed to analyze these metabolites in body fluids. Most of these methods are based on chromatographic based methods. Especially technical developments in mass spectroscopy have been offer great opportunity for high resolution and sensitivity to detect low abundance metabolites in body fluids. Liquid chromatography coupled mass spectroscopy (LC/MS) based methods have been used simultaneous analysis of nerve agents metabolites(3, 4). However there are not standard sample preparation techniques before LC/MS analysis.

In present work, we extracted EMPA (VX degradation products), PMPA (product of soman degradation), CHMPA (a product of cyclohexyl Sarin degradation) and BuMPA (a product of RVX degradation) with four different sample preparation techniques wand analyzed with LC/MSMS. We compared efficiency of four different sample preparation methods in urine and plasma. 1: 1 Dilution and shot, methanol extraction, C18 SPE and Easy SPE methods have been applied biological samples. Metabolites separated C18 stationary phase and water (A), acetonitrile (B) mobile phase. In negative mode MRM parameters were optimized for all metabolites. Validation parameters in terms of LOD, LOQ, linearity, accuracy, precision and selectivity were evaluated for all sample preparation techniques.

1) J. Kassa, J Toxicol Clin Toxicol. 40 (2002) 803.
DOI: 10.1081/clt-120015840]
2) D. Noort, et al., Arch Toxicol. 72 (1998) 671.
DOI: 10.1007/s002040050559
3) J.R. Smith, et al.,J Appl Toxicol. 21 (2001) 27.
DOI: 10.1002/jat.802]
4) M. Otsuka, et al., Forensic Toxicol 41 (2023) 71.
DOI: 10.1007/s11419-022-00633-x

PS2-24 Enzyme-based platform immunoassay for the simultaneous quantification of drug and anti-drug antibodies.

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TNF-α inhibitors (TNFi) are therapeutic monoclonal antibodies used extensively in immune-mediated diseases. One such condition is Inflammatory bowel disease (IBD), a chronic and relapsing condition that affects 6.8 million cases globally. TNFi are the mainstay of the patients' treatment course and Infliximab (IFX) is one such drug example. Despite their proven clinical efficacy, the administration of TNFi elicits an immune response, resulting in the development of anti-drug antibodies (ADA), reported in more than 40% of TNFi-treated patients. ADA promotes drug blocking and/or clearance, consequently reducing its efficacy. In the therapeutic drug monitoring (TDM) framework, ADA and drug levels are measured and their levels guide therapeutic decisions, namely dose increase or therapy change. Currently, drug and ADA levels are evaluated by methods that do not accurately quantify the neutralization effect in patients, cannot detect drug and ADA simultaneously, and mostly require lab testing. Here, we present the development of a novel paper-based assay (PBA) for evaluating both drug levels and drug neutralization by ADA at the point of care (POC) within the same device. We developed a new architecture of classic PBA, utilizing both competitive and direct binding that enables the quantification of both analytes (drug & ADA) in a single-step test. This assay aims to facilitate physicians' decision-making, by enabling the performing of the test with high sensitivity. In this

research, Infliximab was used as the model for the therapeutic mAb, however, the assay can be adapted to detect other therapeutics and ADA specific to them.

1. https://doi.org/10.3389/fimmu.2015.00109
2. https://doi.org/10.3389/fimmu.2020.01365
3. doi:10.1136/gut.2009.201533
4. doi: 10.1056/NEJMoa030815

PS2-26 Fast screening of biological and food samples using miniplatforms based on 3D stochastic microsensors

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Enantioanalysis of aspartic acid is of high importance for metabolomics in colon cancer, as well as for the quality of food. Therefore, we proposed two miniplatforms based on incorporation of 3D stochastic microsensors for fast screening tests of biological and food samples. The 3D stochastic microsensors were designed using nanographene paste decorated with spheroidal Cu, and CuO, respectively, and modified with a solution of beta-cyclodextrin. Different values for the signatures of L- and D-aspartic acid were recorded making possible its enantioanalysis in biological samples and in food samples. The miniplatforms, and fast screening tests (6minutes/screening test) were validated using real samples.

PS2-27 Harnessing programmable zwitterionic coacervates as versatile sensing platforms

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We have recently developed associative coacervates based on zwitterionic heterotypic polymers generated by RAFT polymerization, which exhibit a series of attractive features for bioseparation and biosensing [1]. These coacervates possess responsiveness to external stimuli and preferentially exclude most molecules, making them an ideal baseline material for designing specific uptake of target components through polymer engineering [1]. One exciting application involves functionalizing the zwitterionic coacervates with affinity tags to locally concentrate and separate target molecules from complex biofluids [1].

In this study, we leverage this principle to develop an abiotic affinity-based coacervates technology for the analysis of extracellular vesicles (EVs) in urine, which holds promise as a noninvasive diagnostic bioassay for liquid biopsy. EVs have emerged as important mediators of cell-cell communication, and their potential as diagnostics and therapeutic tools has generated significant interest. Here, we functionalized the polymeric chains with a recently developed membrane-sensing peptide [2] and demonstrate the ability of the coacervates to isolate both model liposomes and EVs from complex biofluids.

[1] U. Capasso Palmiero, et al., Adv. Mater. 34 (2022) 2104837.

DOI: 10.1002/adma.202104837
[2] A. Gori, et al., J Extracell Vescicles 9 (2020) 1751428.
DOI: 10.1080/20013078.2020.1751428

PS2-28 The development of a MIP-based electrochemical sensors for antiviral drug detection using different electroanalytical techniques

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Abstract

Umifenovir (UMI), (Arbidol, ethyl 6-bromo-4-[(dimethylamino)methyl]-5-hydroxy-1-methyl-2-[(phenylsulfanyl)methyl]-1H-indole-3-carboxylate), is an antiviral drug commonly used to treat and prevent COVID-19 and some other viral infections. Umifenovir, which exhibits a broad spectrum of anti-influenza activity, was first licensed for the treatment of influenza in Russia in 1993 and in China in 2006. It has also been shown to be effective against the human herpes virus, hepatitis B and C, and Ebola virus, inhibit enterovirus C and have antioxidant activity [1]. In this study, methacryloyl butyl (Bu-MA) functional monomer was designed, synthesized, and used for the selective and sensitive detection of UMI using the photopolymerization (PP) technique on a glassy carbon electrode (GCE) surface. In the Bu-MA/RIB@MIP sensors, Bu-MA as a functional monomer was designed, synthesized, and obtained in the presence of basic monomer (2-hydroxyethyl methacrylate), crosslinker (ethylene glycol dimethacrylate), pore former (polyvinyl pyrrolidine), and initiator (2-hydroxy-2-methyl propiophenone) by keeping it under a UV lamp at 365 nm. The surface and electrochemical characterizations of MIP-based sensors were performed using Fourier transforms infrared (FT-IR) spectroscopy, atomic force spectroscopy (AFM), scanning electron microscope (SEM), cyclic voltammetry (CV), contact angle, and impedance spectroscopy (EIS). The electrochemical analyses of the modified sensors were investigated using differential pulse voltammetry (DPV) and EIS. Under optimum experimental conditions, the dynamic linear range of the developed sensors was found as 0.50-7.5 pM and 0.25-5.0 pM for DPV and EIS, respectively. Moreover, the developed MIP-based electrochemical sensors were evaluated by examining the interference effects of structurally similar antivirals on UMI analysis. According to the results obtained, these sensors, which were developed for the first time upon the detection of UMI were proved to be UMI-specific.

Acknowledgments
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[1] E.V. Proskurnina, D.Y. Izmailov, M.M. Sozarukova, T.A. Zhuravleva, I.A. Leneva, A.A., Poromov. Antioxidant potential of antiviral drug umifenovir. Molecules 25, (2020), 1577. https://doi.org/10.3390/molecules25071577.

PS2-29 Online biomass monitoring of Chlorella vulgaris cultures by dielectric spectroscopy

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Microalgae are seen as one of the potential strongholds in the fight against global warming and a rich, sustainable source of valuable ingredients. Prominent application areas include carbon fixation, nutrition (microalgae are rich in proteins and unsaturated fatty acids) and biofuels production. A large variety of novel compounds originating from algal biomass has been chemically determined and is being investigated for commercial exploitation. They include carotenoids, antioxidants, fatty acids, enzymes, polymers, peptides, toxins, and sterols.

Although the biotechnological potential of algal cultures has been widely recognized, the toolkit for process control and analytical monitoring technologies for microalgae is still lagging behind the standard of other fields in bioprocessing. In particular, the online determination of biomass poses challenges owing to the largely variable configuration of microalgae cells. Dielectric spectroscopy is handled as a promising, yet still under-explored technology in this regard. In the present study, we critically compare different off-line methodologies of biomass estimation with dielectric spectroscopy and present conclusions on the applicability and limits of the dielectric spectroscopy as a tool to estimate the biomass of microalgae cell suspensions.

Chlorella vulgaris was used as a model microorganism, cell suspensions at different concentrations were analysed. The measurements were carried out with a commercially available sensor (Incyte, Hamilton), which measures the dielectric spectrum of cell suspensions in frequencies between 300 kHz and 10 MHz. The dielectric spectrum was analysed by a software developed in-house. This software fits the dielectric spectrum of the cell suspension to a complex nonlinear model based on the Cole-Cole equation. The resulting parameters provided by the software were compared with standard biomass estimation technologies, such as cell dry weight (CDW) and optical density (OD) measurements. As additional, non-standard data, particle counts from flow cytometry measurements were also included. Very high correlations were found between the different standard techniques and dielectric spectroscopy, validating the use of this methodology as an online biomass monitoring and control tool for microalgae cultivations.

Critically, parameters influencing the dielectric spectroscopy were varied experimentally providing understanding on their interference in the resulting dielectric spectrum. Changes in temperature, conductivity and cell morphology were. Of relevant interest, it was found that changes on temperature have limited effect on the measurement, although changes on conductivity during the cultivation were shown to have a negative impact on the measurement and will require further analysis.

PS2-30 Electrochemical classification of benzodiazepines: a comprehensive approach combining insights from voltammetry and liquid chromatography – mass spectrometry

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The class of benzodiazepines were once the most prescribed medications globally due to their sedative, anxiolytic, anticonvulsant, muscle-relaxing and numerous other properties. Although initially branded as a safer alternative to barbiturates, their potential for abuse and dependence became clear and the prescribing of benzodiazepines became increasingly regulated. However, the non-medical use of this class has since only increased and is seen as a significant threat to public health. Particularly problematic is their involvement in drug-facilitated sexual assault (also called 'date rape') and in polydrug abuse combined with opioids.

The increasing abuse of benzodiazepines, combined with the emergence of new 'designer benzos' and influx of counterfeit pills on the market, has urged the development of highly accurate and sensitive analytical methods for benzodiazepine containing samples. Confirmatory methods such as high-performance liquid chromatography (HPLC) and gas chromatography coupled to mass spectrometry (GC-MS) are considered the gold standard for drug analysis in a laboratory environment. Meanwhile, fast and portable methods for the screening of samples in the field are needed to allow law enforcement and health-related services to make timely and informed decisions on the spot.

Electrochemical sensors are increasingly seen as a promising analytical tool in forensics thanks to their high potential for miniaturization, low cost, rapid measurements and strong analytical performance. Herein, we report the development of a comprehensive electrochemical approach for the classification of a variety of benzodiazepine samples using screen-printed electrodes. Through a combination of voltammetry and liquid chromatography – mass spectrometry, the characteristic redox signals of three representative benzodiazepines (diazepam, clonazepam and alprazolam) are linked to structural elements and based on this, three classes are defined. To confirm the potential use of the approach in a general pill testing context, 22 confiscated pills (containing 14 different benzodiazepines) are analysed and assigned to a class while discarding a set of commonly encountered non-benzodiazepine substances in pills. Finally, the three representative benzodiazepines were spiked in five different alcoholic beverages and analysed in the context of combatting drug-facilitated sexual assault.

The use of screen-printed electrodes and portable potentiostats makes this approach highly suitable for on-site deployment. It could provide law enforcement, first aid responders and drug-checking services with important information on the content of unknown samples on the spot.

J.Y. Wick, Consult Pharm 28 (2013) 9
DOI: 10.4140/TCP.n.2013.538
Z. Qriouet, et al., J Anal Methods Chem (2019)
DOI: 10.1155/2019/2035492
L. Shaw, et al., Curr Opin Electrochem 3 (2017) 1
DOI: 10.1016/j.coelec.2017.05.001

PS2-31 A label-free insight into the molecular aspects of electrochemical DNA sensors for mercury ion detection

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An area of research of continuing interest in modern bioanalysis is the search for methods to determine small-molecule analytes. Electrochemical DNA biosensors, due to their low manufacturing cost and simplicity of signal readout, are valuable tools of increasing importance in environmental analytics. At the same time, they are an alternative to current tools for determining metal cations, which need improvement and are more affordable for daily use.

One example of the risks from toxic heavy metal ions in liquid samples (e.g. drinking water) is mercury ions (Hg2+). Hg2+-sensitive DNA biosensors rely on the phenomenon of thymine bridge formation involving the detected ion. Various mechanisms of ion-aptamer complex formation have been described in the literature [1]. Depending on the aptamer sequence used, one can distinguish: (i) the primary mechanism of T-Hg-T bridge formation - a sequence built from thymines alone, which under the influence of mercury ions adopts the structure of a single-stranded 'hairpin', (ii) the assisted mechanism, where, in addition to thymines, other nucleotides are present in the non-binding regions to facilitate the formation of the target conformation (usually a 'hairpin') in the presence of Hg2+, (iii) the mechanism based on competition of interactions, where DNA with the initial 'hairpin' structure undergoes a conformational change under the influence of mercury ions through its unbinding. Therefore, the design of an aptasensor with good analytical performance requires the selection of a mechanism that would allow the most significant measurable changes in the electrochemical signal (typically associated with a significant change in DNA conformation) as well as the most stable Hg2+-aptamer interaction possible.

The presented research is a comparison of different DNA aptamer sequence variants for the detection of mercury ions. They include a thorough investigation of the mechanism of T-Hg-T bridge formation (both within a single and two adjacent strands), a comparison of the binding strength of such structures and classical dsDNA duplexes, and the selection of analyte-binding fragment lengths. The study results will provide new knowledge and facilitate the design of proprietary sequences with the desired receptor properties. Various techniques have been used to investigate surface interactions (indirect or direct monitoring of interaction kinetics using QCM/SPR) and voltammetric methods. Using a rationally designed analyte recognition sequence could result in an extremely useful and innovative biosensor with attractive analytical performance for environmental analysis. It could become a low-cost and easily miniaturised alternative to routine spectroscopic methods.

[1] Y. Miyake, et al., J. Am. Chem. Soc. 128 (2006) 2172–2173, DOI: 10.1021/ja056354d]

PS2-32 Paper-based Device for Point-of-care Nucleic Acid Quantification Combining CRISPR/Cas System and Personal Glucose Meter

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Nucleic acid testing plays an important role in various fields from basic biology to clinical diagnosis. Recently, clustered regularly interspaced short palindromic repeat (CRISPR)-based assays have been reported as attractive tools for the nucleic acid detection due to their high specificity and sensitivity [1]. Taking advantage of the low cost, portability and wide availability of personal glucose meters (PGMs) [2], methods combining the CRISPR/Cas system and PGMs for nucleic acid quantification have been established for point-of-care testing (POCT). They rely on the conversion of the target nucleic acid concentration into a glucose signal through an enzymatic reaction. However, most reported assays combining CRISPR/Cas with PGMs require multi-step operations involving pipetting and separation,

which can only be performed by trained personnel, which is against the concept of POCT [3]. On the other hand, µPADs (microfluidic paper-based analytical devices) are drawing attention as tools for POCT due to their low cost, ease of fabrication, user-friendliness and disposability [4]. By integrating the CRISPR/Cas system and PGM-based assays into a µPAD approach, it is expected to achieve specific, sensitive, low cost, portable, and quantitative POCT. In this work, we are developing a sensitive and highly specific paper-based biosensor for quantification of nucleic acids by combining the CRISPR/Cas system and PGMs. Pre-deposition of all required reagents on a multi-layer paper device enables assays to be performed by end-users without multiple operation steps and reagent handling. The device consists of three layers of hydrophobic wax-patterned paper. A target DNA-specific CRISPR/Cas12a-RNA (crRNA) complex and single-stranded DNA-conjugated invertase immobilized on magnetic beads (MB-ssDNA-invertase) are deposited on the first layer, while sucrose is dried on the third layer. Application of a sample containing target DNA (tgDNA) onto the first layer of the paper device activates the CRISPR/Cas12a-crRNA complex, resulting in release of invertase through nonspecific cleavage of ssDNA at the surface of MB-ssDNA-invertase. After the cleavage reaction, removing a hydrophobic film separating the first and second paper layers allows the released invertase to flow through the second layer, while magnetic beads are retained. When reaching the third device layer, the released invertase converts the pre-deposited sucrose to glucose, which is subsequently detected by the PGM. A tgDNA concentration-dependent PGM signal is obtained from a single sample application (10 µL) without any further user intervention except for the hydrophobic film removal. For proof-of-concept, the detection of human papilomavirus-DNA (43 base pairs) has been achieved.

[1] Jeanne E. van Dongen, et al., Biosens. Bioelectron. 166 (2020) 112445. DOI: 10.1016/j.bios.2020.112445
[2] Y. Xiang, et al., Nat. Chem. 3 (2011) 697. DOI: 10.1038/nchem.1092
[3] B. Fang, et al., Talanta, 249 (2022) 123657. DOI: 10.1016/j.talanta.2022.123657
[4] K. Yamada, et al., Lab Chip 17 (2017) 1206. DOI: 10.1039/C6LC01577H

PS2-33 Platform for verification of electrochemical sensors for biomedical applications

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Electrochemical sensors and biosensors have great potential for biomedical applications [Zhang 2008], a prospect that motivates many researchers in the field. However, from first prototype to actual deployment in biomedical practice is a long and challenging journey with many barriers to overcome [Hoekstra 2018], and stringent requirements to be addressed: verification, analytical validation, clinical validation, safety, data privacy, usability, and economic feasibility [Coravos 2020].

Step one -verification- entails the systematic in-vitro evaluation of the sensor performance compared to ground truth, against pre-specified criteria [Goldsack 2020]. It includes metrics like sensitivity, selectivity, response time, working range, limit of detection, repeatability, reproducibility, precision, accuracy, drift and ruggedness [Eurachem 2014]. While academic researchers typically focus on novel sensor concept development, from an industrial de-risking perspective, completed verification is considered as an essential starting condition to decide on industrial uptake.

To bridge this gap, at imec we are developing a platform for verification of electrochemical sensors, composed of the following elements:

• A generic sensor substrate that allows for die-level electrode functionalization of up to 50 spatially multiplexed electrochemical sub-sensors integrated into a compact footprint (<1 cm2).
• Optionally, leveraging the compatibility with wafer-scale processing techniques, a single wafer-level electrode functionalization results in 100+ (diced) sensor substrates (with each up to 50 parallel sub-sensor units).
• A computer-controlled experimental setup, holding the sensor substrate embedded into a microfluidic flowcell, connected to a fluidic system for automated analyte mixing and delivery. Additionally, a multi-channel potentiostat allows for parallel read-out of all electrochemical sub-sensors.
• A scalable data processing framework based on the SensorThings API [Liang 2021] that allows for fine grained analysis along the various dimensions and parameters included in the verification dataset.

The design of this platform is geared towards automated high-throughput verification experiments, to facilitate both the required large number of variations in experimental conditions, as well as the large number of (sub-)sensor reproductions and statistics, combined in an efficient workflow.
We are open for collaboration opportunities with industry and academia, to deploy the imec platform for verification of electrochemical sensor materials and designs from partners.

• *Coravos et al., Modernizing and designing evaluation frameworks for connected sensor technologies in medicine, Digital Medicine 3, p.37 (2020), https://doi.org/10.1038/s41746-020-0237-3*
• *Eurachem, The Fitness for Purpose of Analytical Methods, second edition (2014), ISBN 978-91-87461-59-0*
• *Goldsack et al., Verification analytical validation and clinical validation (V3) biometric monitoring, Digital Medicine 3, p.55 (2020), https://doi.org/10.1038/s41746-020-0260-4*
• *Hoekstra et al., Distributed electrochemical sensors recent advances and barriers to market adoption, Analytical and Bioanalytical Chemistry 410, p.4077 (2018), https://doi.org/10.1007/s00216-018-1104-9*
• *Liang et al, OGC SensorThings API Part 1: Sensing Version 1.1, Open Geospatial Consortium (2021), https://docs.ogc.org/is/18-088/18-088.html*
• *Zhang et al, Electrochemical Sensors, Biosensors and Their Biomedical Applications, Academic Press (2008), ISBN 978-0-12-373738-0*

PS2-34 Self-powered optical potentiometric sensors array based on electronic paper

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We report here a novel optical readout method for ion-selective potentiometric probe arrays. The electronic paper (e-paper) was chosen here as the signal transducer owing to its fast response on the order of seconds, its wide dynamic range of about 1.5 V and the capacitive character of the display, which allows one to reach the desired zero current condition in the potentiometric measurement.[1] While an ion-selective electrode (ISE) responds to the sample in a Nernstian manner at zero current, a transient current of limited amplitude provides the charge needed for the e-paper to exhibit a visual color change.

The cathodes of three different pixels of the e-paper were connected to three different ISEs responsive to Na+, K+, and Ca2+, while their common anode was connected to the shared reference electrode (RE). In such a manner, the voltage applied to each pixel is mapped to the behavior of one sensor, making it possible to detect multiple ions with a e-pixel array. Here, the sensor configuration gave quantitative information on Na+, K+, and Ca2+ from 10-5 M to 10-1 M by analyzing the RGB information

of the three pixels. The e-paper exhibited a stable response within half a minute, much faster than previously established electrochromic material transducers such as Prussian Blue. With this sensor, we successfully carried out multi-ion concentration sensing without external power, which is potentially attractive in environmental monitoring and clinical assays.

1. Wu, Y. and E. Bakker, Self-Powered Signal Transduction of Ion-Selective Electrodes to an Electronic Paper Display. ACS Sens., 2022. 7(10): p. 3201-3207.

PS2-35 Hierarchical architectures of graphene as sensitive membranes for electrochemical sensors

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Carbon-based low-dimensional nanomaterials have gained enormous research interest in the last decade due to their excellent electrochemical and photochemical properties. Among them are 2D nanoparticles, such as graphene (G) and graphene oxide (GO); 1D nanostructures, such as carbon nanotubes; and 0D nanoparticles, such as nanodiamonds and carbon nanodots (CNDs). Recently we studied a new type of carbon-based 2D and 0D nanomaterials such as mesoporous graphene (PG) and tremella-like graphene (TG), which, contrary to G and GO, have porous morphology. In the search for materials that can increase the sensitivity of electrochemical sensors, PG and TG were immobilized on the surface of a glassy carbon electrode, and analytical properties of the sensors were tested in the determination of endocrine disruptors, clinically essential chemicals such as glucose, and neurotransmitters. The results were compared with sensors having electrocatalytically active membranes with GO, graphene quantum dots. Due to the porous and hierarchical electroactive structure, the nanomaterials have increased sensitivity and improved sensor selectivity.

PS2-36 In-depth Study of Tyrosine Oxidation Using Electrochemistry, Capillary Electrophoresis, and Mass Spectrometry

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It is of utmost importance for many bio(organic) molecules to closely study the processes happening on the electrode, considering the electrode materials and oxidation or reduction conditions.

Tyrosine (Tyr) is one of the crucial targets of oxidation in living systems. A phenolic amino acid with relevance, not only as a constituent of proteins but also as a precursor of the neurotransmitters, norepinephrine, and epinephrine. In this context, several methods are developed for understanding and studying the oxidation behavior of Tyr, enzymatic oxidation by tyrosinase, and non-enzymatic oxidation by hydroxyl free radicals, for instance [1]. Since Tyr is an electrochemically active compound, electrochemical oxidation can be used as an alternative oxidation method. Electrochemical oxidation drives benefit from well-controllable parameters such as applied oxidation potential, reaction time, and the wide variety of electrode materials. While classical electrochemical techniques represent several advantages, the limitation of being unable to identify the electrogenerated species for

understanding the mechanistic details, cannot be neglected. Therefore, more elaborate analytical techniques are hyphenated with electrochemistry (EC), involving mass spectrometry (MS), and capillary electrophoresis (CE), to investigate the redox behavior. CE is implemented as a strong separation technique between EC and MS. MS as a versatile method provides structural information alongside the exact molecular masses [2].

In this study, a real-time mass voltammogram was recorded by online EC-MS using an electrochemical flow cell with an integrated disposable electrode DRP-110, providing the potential dependent product profile. On the other hand, EC-CE-MS electropherograms resulting from measurements with and without electrochemical pre-treatment were investigated to study the separation behavior and the structural properties of the individual species.

[1] https://doi.org/10.1016/j.ab.2022.114856
[2] https://doi.org/10.1002/celc.202000442

PS2-37 Modification-free boron-doped diamond as a sensing material for direct and reliable detection of the anti-HIV drug nevirapine

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Human immunodeficiency virus (HIV) is a retrovirus harming the cells of the immune system and weakening the body's ability to fight regular infections and diseases [1]. One of the most extensively prescribed anti-HIV drugs worldwide is nevirapine (NVP) [1]. It is important to monitor the NVP concentration in the patient's blood serum to establish the optimum therapeutic dosage to avoid negative effects and to maintain the suitable management of HIV infection treatment. Moreover, NVP is an emerging environmental contaminant, which is highly resistant against conventional water treatment procedures [2]. Evidently, it is crucial to establish suitable procedures for monitoring and detection of NVP in pharmaceutical, clinical and environmental samples.

So far, none of the electrochemical sensing approaches targeting NVP has utilized conductive boron-doped diamond (BDD), despite the fact that BDD possesses a range of unique properties such as broad potential window, low and stable background currents, weak molecular adsorption making BDD surface resistant to (bio)fouling and deactivation, which in consequence enables long-term reliability and stability of sensors, and biocompatibility [3].

Hence, in this work, non-modified BDD was employed first time ever as the sensing material for the in-depth voltammetric study of the antiretroviral drug NVP [4]. Two types of electrode surface pre-treatments, anodic oxidation and alumina-polishing, yielded BDD of different surface chemistry, denoted as O-BDD and p-BDD, respectively. Induced alterations in BDD surface composition reflected in distinct voltammetric responses of NVP, also depending on the pH of the medium. The electrochemical oxidation of NVP on both electrodes, whose mechanism has also been proposed, has an irreversible character and is controlled by diffusion. Overall, NVP provided signals of excellent intra- and inter-day repeatability (RSD ≤ 5.0%) which remained unaffected even in the presence of common interfering compounds (e.g., glucose, ascorbic acid, uric acid, and dopamine). Even though the O-BDD electrode outperformed the p-BDD electrode in terms of sensitivity and the lowest detection limit achieved (0.04 µM), both O-BDD and p-BDD provided highly favourable analytical parameters fulfilling the requirements for clinical application for NVP monitoring in biofluids. This was also proved by

electroanalysis of NVP in synthetic serum samples where recovery values of 96.3−103.0% were achieved. Finally, unique properties of BDD allowed to develop a direct, modification-free, and reliable protocol for NVP detection, which paves the way for the full sensor development.

Acknowledgements
Financial support from the Dutch Research Council (NWO) through the Open Technology Programme (project no. 16361) is gratefully acknowledged.
[1] A.O. Adeola, et al., Environ. Toxicol. Chem., 41 (2022) 247. DOI: 10.1002/etc.5127.
[2] V. Simon, et al., Lancet, 368 (2006) 489. DOI: 10.1016/S0140-6736(06)69157-5.
[3] S. Baluchová, et al., Anal. Chim. Acta, 1077 (2019) 30. DOI: 10.1016/j.aca.2019.05.041.
[4] S. Baluchová, et al., Electrochim. Acta, 450 (2023) 142238. DOI: 10.1016/j.electacta.2023.142238.

PS2-38 Simultaneous voltammetric determination of prothioconazole and bixafen on a boron-doped diamond electrode

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Fungicides are chemical compounds that penetrate plant tissues and spread throughout the plant, preventing the development of fungal pathogens [1]. The representative fungicides are bixafen, fluopyram, and prothioconazole which are the components of the ASCRA Xpro formulation. Bixafen and fluopyram are succinate dehydrogenase inhibitors, while prothioconazole is a sterol biosynthesis (demethylation) inhibitor [2]. AscraXpro delivers broad-spectrum disease control, including more curative activity against Septoria than any other fungicide formulations and the highest yields.

The main objective of this work was to evaluate the possibility of the simultaneous voltammetric determination of three fungicides, i.e., bixafen, prothioconazole, and fluopyram on a boron-doped diamond electrode (BDDE). Detailed studies of the electrochemical activity of each fungicide were carried out using cyclic voltammetry in a supporting electrolyte (Britton-Robinson buffer (BRB)) within a wide pH range of 2.0–12.0. It was found that prothioconazole and bixafen are being oxidized on the BDDE in a wide pH range from 2.0 to 12.0, while fluopyram has no electrochemical activity on the BDDE. In the next step, the effect of the pH of the BRB on the separation of the square-wave voltammetric oxidation signals of prothioconazole and bixafen was investigated. As the optimal medium for the simultaneous determination of both pesticides, the BRB of pH 3.0 was selected. It was found that the simultaneous voltammetric determination of prothioconazole and bixafen on the BDDE was possible in the range of 5.0–80.0 µmol L–1 (LOD = 1.53 µmol L–1 for prothioconazole and LOD = 1.54 µmol L–1 for bixafen).

[1] R. Özcan, Ç. Büyükpınar, S. Bakırdere, Journal of Environmental Science and health. Part. B, Pesticides, Food Contaminants, and Agricultural Wastes 55 (2015) 1041.
[2] X. Jing, X. Huang, Y. Zhang, M. Wang, H. Xue, X. Wang, L. Jia, Food Chemistry 367 (2022) 130664.

PS2-39 The development of molecularly imprinted polymer-based electrochemical sensor for the selective and sensitive determination of tolvaptan

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Tolvaptan (TOL) is a vasopressin-2-receptor (V2R) antagonist. Also, this drug is a p-glycoprotein substrate that interacts drug-drug with digoxin in vivo. TOL treats heart failure, liver cirrhosis, and antidiuretic hormone secretion syndrome [1]. In this study, a highly sensitive electrochemical sensor was designed for the first time detection of TOL using the molecular imprinted polymer (MIP) method. A MIP-based electrochemical sensor is developed on the glassy carbon electrode (GCE) surface using thermal polymerization method. TOL showed very high sensitivity and selectivity towards the template molecule in the designed sensor. The surface and morphological characterizations of the MIP-based electrochemical sensor were performed using cyclic voltammetry (CV), electrochemical impedance spectroscopy (EIS), scanning electron microscopy (SEM), and energy distribution X-ray spectrometry (EDX). For the quantitative determination of TOL, 5.0 mM [Fe(CN)6]3−/4– measurement with a redox probe was performed in solution using a differential pulse voltammetry (DPV) technique. The analysis of TOL in standard solution, commercial serum sample, and tablet dosage forms were successfully applied. After the optimization experiments, the calibration range was found between 25 pM and 250 pM. The limit of detection (LOD) and the limit of quantitation (LOQ) were calculated as 0,188 pM and 0,627 pM, respectively. The accuracy of the sensor was proved by the recovery study and the recovery values were calculated as 101.0% and 104.0% in the tablet dosage form and commercial serum samples, respectively. Moreover, the selectivity of the sensor was proven using common interference agents such as KNO3, MgCl2, Na2SO4, uric acid, ascorbic acid, dopamine, and paracetamol. Imprinting factor (IF) were calculated using substances with similar molecular structures, such as sorafenib, regorafenib, imatinib, dasatinib, and nilotinib. The proposed method was proven to be highly sensitive and selective compared to other reported analytical methods.

PS2-40 Molecularly imprinted sensor based on CNFs for voltammetric detection of dasatinib

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Dasatinib (DAS), a multi-targeted tyrosine kinase inhibitor, is used to treat chronic myeloid leukemia and acute lymphoblastic leukemia [1,2]. DAS treats chronic, accelerated, or blast-phase myeloid leukemia that has developed resistance or intolerance to previous treatments. In addition, DAS has successfully passed clinical trials for treating non-Hodgkin lymphoma, metastatic breast cancer, and prostate cancer [2]. In this work, we fabricated a molecularly imprinted polymer (MIP)-based electrochemical sensor by using a carbon nanofiber (CNF) for ultrasensitive detection of DAS via photopolymerization technique. The MIP-based electrochemical sensor was constructed on a glassy carbon electrode (GCE) in the presence of DAS as a template molecule, using N-methacryloyl-L-phenylalanine, 2-hydroxyethyl methacrylate ethylene glycol dimethacrylate by keeping it under a UV lamp at 365 nm. Cyclic voltammetry (CV), differential pulse voltammetry (DPV), electrochemical impedance spectroscopy (EIS), Fourier transform infrared spectroscopy (FT-IR), scanning electron microscopy (SEM), and contact angle were used for surface and morphological characterizations of the modified electrode. Under optimum conditions, the current response for DAS was found in the linear

range of 10.0 fM to 100.0 fM and a detection limit of 1.76 fM. In addition, the sensor exhibited high reproducibility, anti-interference ability, and good stability for 1 week toward DAS detection. Finally, the developed sensor demonstrated excellent detection performance in complex tablet dosage forms and commercial serum samples with satisfactory recoveries. This sensor could be a promising tool due to its fast detection capabilities, cost-effectiveness, low sample consumption, high sensitivity, and strong stability.

[1] D.E. Bayraktepe. A voltammetric study on drug-DNA interactions: Kinetic and thermodynamic aspects of the relations between the anticancer agent dasatinib and ds-DNA using a pencil lead graphite electrode. Microchemical Journal, 157, (2020), 104946. https://doi.org/10.1016/j.microc.2020.104946.
[2] C.S.H. Jesus, V.C. Diculescu. Redox mechanism, spectrophotometrical characterisation and voltammetric determination in serum samples of kinases inhibitor and anticancer drug dasatinib, Journal of Electroanalytical Chemistry, 752, (2015), pp. 47-53. https://doi.org/10.1016/j.jelechem.2015.06.006.

PS2-41 The application of the modified carbon paste electrode in voltammetric sensing of ibuprofen

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Ibuprofen (2-(4-isobutylphenyl)propanoic acid, IBU) is a typical representative of non-steroidal anti-inflammatory drugs with anti-inflammatory, analgesic, and antipyretic activities [1]. The mechanisms of action are based on the non-selective reversible inhibition of the cyclo-oxygenase enzymes that are required for the synthesis of prostaglandins, the important mediators of pain, inflammation, and fever, and inhibitory effects on leucocytes [2]. Some studies revealed that regular intake of IBU can be useful in the prevention of some diseases including certain types of cancer, and neurodegenerative diseases [3, 4]. Although IBU is one of the most widely used drugs, monitoring the content of this drug in different samples represents a difficult challenge for analysts.

The aim of this study was to develop an electroanalytical methodology for the sensitive and selective determination of a particular drug in various real samples. To achieve this, a modified carbon paste electrode was utilized. As a modifier of carbon paste electrode functionalized multi-walled carbon nanotubes supported with Fe electrocatalyst (Fe/fMWCNTs) have been synthesized and used to enhance the performance of CPE. Fe/fMWCNTs through modification improved the electrochemical properties of the bare CPE by enlarging its surface and improving its conductivity. By using square-wave voltammetry as an electroanalytical technique, linearity was obtained in two concentration ranges of 10-100 µmol/L and 100-500 µmol/L, with an accomplished LOD of 4.6·10-8 mol/L. Finally, the developed electroanalytical methodology enabled the quantification of the drug in formulation, water, and urine samples.

1. Higton F (2015) The Pharmaceutics of Ibuprofen. In: Rainsford KD (ed) Ibuprofen Discovery, Development and Therapeutics. John Wiley & Sons, Ltd, Oxford, pp 51–80
2. Rainsford KD (2015) Pharmacology and Toxicology of Ibuprofen. In: Rainsford KD (ed) Ibuprofen Discovery, Development and Therapeutics . John Wiley & Sons, Ltd., Chichester, pp 132–237
3. Rainsford KD (2015) Ibuprofen in Prevention of Neurodegenerative Diseases. In: Rainsford KD (ed) Ibuprofen in Prevention of Neurodegenerative Diseases. John Wiley & Sons, Ltd, Oxford, pp 547–570
4. Bittoni MA, Carbone DP, Harris RE (2017) Ibuprofen and fatal lung cancer: A brief report of the prospective results from the Third National Health and Nutrition Examination Survey (NHANES III). Mol Clin Oncol 6:917–920. https://doi.org/10.3892/mco.2017.1239

PS2-42 All-solid-state potentiometric sensors based on graphene oxide as novel ion-to-electron transducer for nitrate and nitrite detection in environmental waters

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Ion-selective electrodes (ISEs) are popular for the analysis of ions in complex biological and environmental samples due to their high sensitivity, cost-effectiveness, and miniaturization ability [1]. However, the formation of a thin aqueous layer between the ion-selective membrane (ISM) and the transducer in conventionally all-solid-state ISEs results in the potential drift that restricts their long-term applications [2].

In this work, graphene oxide (GO) was proposed as a novel ion-to-electron transducer to develop improved and durable all-solid-state anion-selective potentiometric electrodes. Commercially carbon screen-printed electrodes modified with GO were used for the detection of nitrate and nitrite ions in environmental water samples. A surface characterization based on Raman spectroscopy confirmed its identity and the modification with the corresponding ISM ensured the sensor preparation. The nitrate- and nitrite-ISM were based on tetradodecylammonium nitrate and nitrite ionophore VI, respectively, entrapped in plasticized polymeric matrices.

The potentialities of GO were demonstrated by the absence of an intermediate aqueous layer [3] and the decrease in charge-transfer resistance of the proposed all-solid-state electrodes. Nitrate-ISE showed a sensitivity of 52.0±1.2 mV/decade within the linear range from 3.0 µM to 0.01 M and a limit of detection of 2.2 µM at 0.1 M phosphate buffer background (pH 5.0). A fast response time (<20s), good selectivity against common ions found in waters, great reproducibility (RSD<1.4%), long-term potential stability of 0.3 mV/h, and durability of four weeks are some of the remarkable properties achieved. Likewise, the nitrite-ISE provided a sensitivity of 45.4±0.4 mV/decade over the linear range from 3.0 µM to 0.01 M and a limit of detection of 2.8 µM within the same conditions. A response time <50s, good selectivity, excellent reproducibility (RSD<0.4%), and long-term potential stability of 2.0 mV/h but a shorter lifespan of only a week were observed. The concentration of nitrate was determined in river and well water samples. Appropriate recovery percentages (88–108%) and an excellent agreement with a commercial probe (difference<8.5%) confirmed the reliability of the proposed all-solid-state electrodes.

The next steps envisage the study of GO to different anions detection as well as the sensor's incorporation into a microfluidic system to build up a portable platform for in situ analysis.

1. *G. Crespo, Electrochim. Acta 245 (2017) 1023. DOI: 10.1016/j.electacta.2017.05.159*
2. *R. De Marco, et al., Phys. Chem. Chem. Phys. 10 (2008) 73. DOI: 10.1039/b714248j*
3. *M. Fibbioli, et al., Electroanal. 12 (2000) 1286. DOI: 10.1002/1521-4109(200011)12:16<1286::AID-ELAN1286>3.0.CO;2-Q*

PS2-43 Spectroelectrochemical approaches for the qualitative and quantitative analysis of acetaldehyde in wine, fentanyl in drug of abuse and pesticide detection.

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Spectroelectrochemistry is a hybrid technique that joins the advantages of electrochemistry and spectroscopy. In a single experiment, spectroelectrochemistry provides two signals of different nature, being a very powerful feature to obtain valuable information about the system under study. However, this technique has been traditionally limited to the development of new devices to facilitate the performance of the experimental measurements. Nowadays, commercial cells, set-ups and instruments [1] offer an alternative to improve the spectroelectrochemical applications.

In order to demonstrate the utility of this multirresponse technique, several examples are shown in this work. UV-Vis spectroelectrochemistry has been used for the detection of acetaldehyde in wines [2]. The obtained results in the analysis of different wines agree with the results previously reported in literature with complex and tedious methods, demonstrating that spectroelectrochemistry saves cost and time respect to traditional detection methods.

In addition, Raman spectroelectrochemistry based on electrochemical surface-enhanced Raman scattering (EC-SERS) effect displays an excellent alternative for the detection of a huge variety of analytes. For instance, the enhancement of Raman intensity thanks to EC-SERS effect offers a rapid, efficient, and accurate approach for the detection of fentanyl and its analogs in drugs of abuse [3]. In that way, the analysis of the characteristic Raman band of fentanyl at 1000 cm-1 allows the rapid detection of 0.33 ug/mL of this drug.

Detection of dithiocarbamate, chloronicotinyl and organophosphate pesticides is also achieved thanks to the combination of EC-SERS effect and screen-printed electrodes. Particularly interesting are the results obtained in the detailed analysis of Raman bands at 1380 cm-1 of thiram and 1107 cm-1 of imidacloprid. These characteristic bands allow the detection of 2.4 ug/L thiram and 25 ug/L imidacloprid. In addition, tab water samples were analyzed, obtaining suitable results to demonstrate the capabilities of the spectroelectrochemical method [4].

[1] http://www.dropsens.com/en/spectroelectrochemical_instruments.html
[2] D. Ibáñez, M. B. González-García, D. Hernández-Santos, P. Fanjul-Bolado, Biosensors 12 (2022) 1032. https://doi.org/10.3390/bios12111032.
[3] C.E. Ott, A. Burns, E. Sisco, L.E. Arroyo, Forensic Chemistry 34 (2023) 100492. https://doi.org/10.1016/j.forc.2023.100492.
[4] D. Ibáñez, M. B. González-García, D. Hernández-Santos, P. Fanjul-Bolado, Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy, 248 (2021) 119174. https://doi.org/10.1016/j.saa.2020.119174.

PS2-44 On-site simultaneous determination of calcipotriol and betamethasone in topical pharmaceutical formulations and surface water samples using an intelligent mini platform based on carbon nanotubes-gold nanoparticles screen-printed electrode modified with calix[6]arene

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Calcipotriol and betamethasone are drugs used in combination to treat psoriasis. As psoriasis is a chronic disease requiring prolonged treatment, there are some concerns about the long-term use of betamethasone because of the adverse effects of corticosteroids [1]. In addition, once disposed of in water, corticosteroids may have eco-toxicological impacts due to their various effects on aquatic organisms [2,3]. For this reason, there is a need for rapid and reliable methods for the determination of these compounds in various sample matrices. This work proposes the use of a stochastic platform using a calix[6]arene-modified carbon nanotubes and gold nanoparticles screen-printed electrode for the simultaneous determination of the two analytes from surface water and pharmaceutical formulation samples. This platform is advantageous over other determination methods in that it is portable, cost-effective, and uses disposable sensors, thus avoiding cross-contamination of samples.

[1] K. Kragballe et al., Br. J. Dermatol. 154 (2006) 1155. DOI: 10.1111/j.1365-2133.2006.07236.x
[2] J. Gong et al., Environ. Pollut. 251 (2019) 102. DOI: 10.1016/j.envpol.2019.04.110
[3] C.M. Hamilton et al., Environ Int. 162 (2022) 107163. DOI: 10.1016/j.envint.2022.107163

PS2-45 An electrochemical sensor for trace analysis of morphine in human serum and saliva

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Morphine is a narcotic drug that can relieve severe and chronic pain, particularly chronic cancer pain [1]. Nevertheless, this drug can cause many potential side effects such as slow heart rate, slow breathing, weakness, CNS disturbance, itching, choking and low blood pressure. Even excessive consumption of this drug leads to death due to respiratory depression. [2]. In addition, the user of morphine becomes addicted to this drug after three days. Considering that morphine is considered as an illegal drug, therefore, it needs careful monitoring in many biological samples, which is very important in the clinical and forensic fields [3].

In this study, an Au nanodendrites/broken hollow carbon spheres (Au NDs/BHCS) nanocomposite was constructed to manufacture an effective electrochemical sensor for the detection of ultra-trace morphine (MPH) and exhibited advanced performance. The morphology, structure, and electrochemical behavior of the synthesized BHCS showed that the fractured spheres have a large specific surface area, abundant pores, and good conductivity, which can facilitate the transfer of ions and electrons at the sensor interface and afford an effective platform for electrochemical deposition of gold nanodendrites. Additionally, the catalytic property of Au nanodendrites is integrated with the electrochemical assay to reach high sensitivity for MPH detection. Benefiting from the synergistic effect of Au NDs and BHCS, the Au NDs/BHCS sensor demonstrated an outstanding electrochemical sensing performance toward MPH with a wider linear range of 0.01-300 µM and an ultralow detection limit of 0.0083 µM. Furthermore, the fabricated sensor was employed for determination of MPH in spiked serum and saliva samples and indicated satisfactory recovery in the range of 97–100%. This work supplies a new road for the expansion of validated platforms for on-site screening of illegal drugs, which is essential in clinical settings and forensics.

[1] E. Verrinder, N. Wester, E. Leppanen, T. Lilius, E. Kalso, B.r. Mikladal, I. Varjos, J. Koskinen, T. Laurila, ACS omega, 6 (2021) 11563-11569.
[2] B. Yu, C. Cao, P. Li, M. Mao, Q. Xie, L. Yang, Talanta, 186 (2018) 427-432.

[5] S.S. Kish, H. Ehzari, Z. Shekarbeygi, M. Shahlaei, S. Moradi, A.R. Jalalvand, Sensing and Bio-Sensing Research, 40 (2023) 100555.

PS2-46 Uncovering the multiple adsorption mechanisms of heavy metals by eggshells

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Recently, there has been a growing interest in cheap and effective materials for heavy metals and organic compounds adsorption], focusing on waste materials from industrial, agricultural and domestic sources. Such materials have great potential as sustainable solutions for wastewater treatment, while simultaneously contributing to waste minimization. Examples of cheap adsorbents include rice husks, corn cobs, bird feathers, cotton, and sawdust. Ideally, the adsorbent material only requires minimal pre-processing and adjustments prior to use.

Eggshells are a common waste material, which is difficult to dispose of. Consequently, there has been much interest in the use of eggshells as adsorbents. Many studies report the efficiency and optimal conditions for organic and inorganic pollutant adsorption using eggshells and eggshell membranes. Examples of pollutants documented in the literature include fluoride, actinides, dyes and various toxic metals (lead, chromium, cadmium, copper, arsenic, manganese, nickel and mercury)
Our research focuses on uncovering the various adsorption mechanisms of the eggshells. A better understanding of the adsorption mechanism will lead to a more efficient use of eggshells for adsorption applications. We will present preliminary results for the eggshell adsorption capacity of seven toxic metals and suggestions for the various adsorption mechanisms of the metals to the eggshells.

Reference
[1] – Alternative Low-cost Adsorbent for Water and Wastewater Decontamination Derived from Eggshell Waste, An Overrview. F.Castro et al. Waste and biomass valorizatio 2 157-167 (2011)
[2] - Characteristics and adsorption capacities of low-cost sorbents for wastewater treatment; A review. M.Notarnicola et al. Sustainable Materials and Technologies 9 10-40 (2016)
[3] – Applications of eggshell and egg shell membrane as adsorbents: A review
A. Mittal, M. Teotia, R.K. Soni, J. Mittal. Journal of molecular liquids 222 276-288 (2016

PS2-47 Assessment of metal content in agricultural soils and vegetables and their risk to human health in rural Roma communities in Transylvania, Romania

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The aim of this work was to evaluate the content of nine metals (As, Bi, Cd, Zn, Cr, Fe, Cu, Pb and Ni) in agriculture soils and vegetables (potatoes, onion, carrots, parsley, kale, and parsnips) and their possible risk to human health in some rural Roma communities in Transylvania, Romania.

Metals content was determined by Optical Emission Spectrometry in Inductively Coupled Plasma (ICP-OES), with the sequential determination of the elements in aqueous solution. For ICP-OES analysis, soil samples were subjected to mineral digestion with HNO3:HCl (1:3 v/v), while the vegetables were digested with microwaves (1800 W) with HNO3 (65%) and H2O2 (30%) mixture. A total of 23 soil samples and 23 vegetables were collected from 14 rural Roma communities.

In soil samples, the concentration found were: Bi (1 sample, 3 mg/kg), Fe (23 samples, 10.332 to 33.784 mg/kg), Cu (16 samples, 72 to 277 mg/kg), Pb (9 samples, 4 to 35 mg/kg), Ni (7 samples, 11 to 25 mg/kg) and Cr (23 samples, 9 to 73 mg/kg). For vegetables, the concentration found were Bi (20 sample, 7.02 to 12.5 mg/kg), Fe (10 samples, 0.87 to 16.71 mg/kg), and Cu (23 samples, 18.00 to 34.51 mg/kg).
The rate of metal accumulation in vegetables were: potatos (30.88 to 56.19 mg/kg), carrot (32.72 to 51.99 mg/kg), parsley (37.62 to 47.54 mg/kg) and onion (34.82 to 36.70 mg/kg).

The daily intake concentration (DITM) of Cu was from 0.116 to 0.168 (mg/kg/day) for adults and from 0.34 to 0.494 (mg/kg/day) for children. For Bi the DITM was from 0.034 to 0.0.061 (mg/kg/day) for adults and from 0.10 to 0.176 (mg/kg/day) for children.

The hazard quotient (HQM) of Cu was calculated to assess risks to human health from consumption of locally grown produce. The obtained values were from 2.90 to 4.21 for adults and from 8.51 to 12.35 for children. This suggests that consumption of these vegetables regularly and in large amounts may exposure population to risk.

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PS2-48 Conception of a test gas system for simulating complex air mixtures of biogenic volatile organic compounds in the ppt range

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The qualitative and quantitative analysis of biogenic volatile organic compounds (VOC) is essential for the protection of our environment, as they enable health screening of plants and the identification of infestation by pests. However, due to the complexity, heterogeneity, and the occurrence of many different substances in low concentrations, it poses particular difficulties. A challenging step in the instrumental analysis of VOC is sampling since enrichment is often unavoidable. Standards with a known concentration that are also representative of the real sample are required to quantify an analyte. In case of biogenic VOC, which are dominated by reactive hydrocarbons such as terpenes, gas standards are mostly unavailable. The designed test gas system is intended to close this gap by generating representative gas standards in the ppb-ppt range.

The construction of the test gas system is integrated in a temperature-controlled oven. Analytes of interest are introduced into a purified zero-air gas stream by one of three common sample application systems: test gas, dynamic injection and diffusion or permeation, respectively. They can be operated simultaneously and independently of each other. A second dilution step enables further dilution down to the ppt range and humidification of the gas stream. Physical characteristics of the test gas such as volume flow, temperature, pressure, and relative humidity are adjusted and measured at every key

point within the gas system. With the help of a self-designed valve block, the test gas can be enriched on up to five adsorbents independently, which can then be analyzed by thermal desorption gas chromatography mass spectrometry (TD-GC-MS). All components are controlled and synchronized by a self-written program based on LabVIEW, including the calculation of the theoretical concentration of the gas stream and the theoretical amount of substances on the adsorbents. The actual test gas concentration is monitored with a proton transfer reaction time of flight mass spectrometer (PTR-TOF). In conclusion, the system simulates the emission of VOC by plants and, therefore, allows for reliable quantification using common analytical devices.

PS2-49 Development of a multiresidue method including organotins, based on liquid chromatography coupled to tandem mass spectrometry, for the quantification of emerging micropollutants in Gammarus fossarum

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Emerging micropollutants in the environment due to our way of life (agricultural activities, industry, household use) which are generally not monitored by Water Agency, are however of great interest for a better understanding of environmental pollution.

Chemicals monitored in this study include different families such as pharmaceuticals and personal care products (PPCPs), some pesticides, flame-retardants, perfluorinated compounds and even organotin. These persistent molecules are not removed totally by sewage treatment plants, and are finally found in rivers (1).

Since the last decade, the European Directive includes arguments in favor of biomonitoring chemicals by the use of sentinel species (2). Among them, one distinguished from the others: Gammarus fossarum. This little-30mg-shrimps lives in rivers and mostly bioaccumulates apolar compounds with a logKow > 3. Gammarids can be encaged for example upstream and downstream sites presenting a risk of pollution such as industry or wastewater treatment sites.

This study presents a method for the quantification of emerging micropollutants including organotins in Gammarus fossarum using liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS). One originality of the development is to include organotins which are mainly analyzed by GC-MS/MS. That is why various parameters, such as chromatographic conditions and sample preparation, were optimized with the aim of achieving a reliable and suitable method for the quantification of organotin in Gammarus fossarum samples. The optimized multiresidue method was validated according to ICH guidelines, using fresh gammarids with accuracy rates ranging from 80% to 120%, and limits of quantification between 0.04 ng/g and 313.5 ng/g. The application of the method was further demonstrated in 15 batches of gammarids exposed under environmental conditions. The results prove that the developed LC-MS/MS method is reliable and suitable for the quantification of emerging micropollutants in real samples. To the best of our knowledge, this is the first time a method allows the quantification of emerging micropollutants including organotins in a single method of analysis by liquid chromatography coupled to tandem mass spectrometry.

(1) H.B. Hawash et al., Journal of Water Process Engineering 52 (2023) 103490
DOI: 10.1016/j.jwpe.2023.103490
(2) Directive 2013/39/UE

PS2-50 Strategies for on-site determination of trace elements in officinal plants by stripping voltammetry

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In the present work, Cd and Pb were determined simultaneously in officinal plants by differential pulse anodic stripping voltammetry (DP-ASV) using a glassy carbon electrode modified with ex-situ/in-situ mercury film (MF-GCE) and ex-situ/in-situ bismuth film (BF-GCE). A portable voltammetric analyzer was used, suitable for both laboratory and field analyses. Firstly, the repeatability, accuracy and linearity of the techniques were tested in standard solutions. Then, certified reference materials and three samples, namely passionflower (Passiflora incarnata L.), thyme (Thymus vulgaris) and rosemary (Rosmarinus Officinalis L.) were analysed. Two approaches were adopted for mineralization: microwave oven digestion (only suitable for laboratory use) and extraction in mini dry bath (suitable for field analysis); the subsequent voltammetric analyses provided comparable results for the passiflower sample, while in the case of the others a difference between Pb and Cd content determined after using microwave digester and the mini bath dry was observed: this behaviour is probably due to the fact that elements were partially bound with the undissolved residues left after the extraction carried out in the mini bath dry; for this reason their concentrations were underestimated.

A comparison between the analyses performed in laboratory and those carried out in field, showed that the values obtained for Pb were comparable, while for Cd improvements on the extracting procedure are still needed. In any case, the voltametric analysis in field seems to be reliable.

The main advantage of on-site analyses is the availability of data in real time, which offers the possibility of choosing further sampling points based on the results just obtained and reducing the number of samples requiring additional investigations in the laboratory.

PS2-51 Determination of Benzo(a)pyrene adsorbed onto plant pollen samples by microwave extraction and HPLC-FLD

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Benzo(a)pyrene is a known toxic atmospheric pollutant regulated in Europe by Directive 2004/107/EC. Once emitted, BaP can be found both in the gas and the aerosol phases. Gas-particle partitioning of BaP is favoured to the particulate phase due to its low volatility. Larger aerobiological particles such as pollen grains can adsorb gaseous BaP or smaller-sized atmospheric particulate matter (including PM-adsorbed BaP).

The objective of the study was to develop and test an analytical methodology based on UNE-EN 15549 for determining the concentration of BaP adsorbed onto pollen samples collected in urban areas as an indicator of BaP-pollen interaction in ambient air.

Pollen samples were obtained manually by shaking the branches of Cupressus sempervires specimens inside polyethylene plastic bags. Twenty-one sampling points were selected in the city of Badajoz (SW, Spain). The samples were sieved through a 30 µm mesh. Fifteen milligrams of pollen were deposited in Teflon vessels, and 20 ml of acetone/n-hexane (1:1 v/v) was added to extract the analyte by microwave-assisted extraction at 55 °C for 60 min at 1200 W. The extract was filtered, evaporated to dryness and redissolved in 1 ml acetonitrile for analysis.

The analysis of BaP was performed by high-performance liquid chromatography with fluorescence detection (HPLC-FLD) using an Ace 5 C18 column thermostated at 30ºC and water/Acetonitrile mobile phase (50:50). Quantification of the analyte was performed by external calibration (0.1 to 200 µg L-1). The limit of detection (LOD) was estimated to be 1.33 ng by analyzing blank quartz fiber filters for PM10 sampling because blank pollen samples were unavailable.. The average recovery from fortified samples was 84% (63 – 93%).

The results showed a BaP concentration range in pollen between 0.070 and 0.18 mg kg-1, with an average of 0.12 mg kg-1. This range is significantly lower than obtained for PM10 samples collected in the same period in the suburban areas of Badajoz, whose concentrations ranged between 2.4 and 11.3 mg kg-1.

The proposed methodology has proven useful for determining BaP in pollen collected at urban areas, providing an environmentally useful information which can be relevant to environmental managers and the general public, especially allergic people.

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PS2-52 Fingerprinting of Chlorinated Paraffins and Olefins in Sewage Sludge of a Swiss Wastewater Treatment Plant

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Chlorinated paraffines (CPs) are polychlorinated (nCl ≈ 3–12) n-alkanes (nC ≈ 10–30) with the molecular formula CnH(2n+2-x)Clx. CPs are high production volume chemicals (>1Mt/y) widely used in metalworking fluids or as flame retard-ants and plasticizers in the plastic industry. Therefore, CPs are contained in materials of our daily life, such as electronics, plastic mats, foils, and toys [1]. In other words, we are in contact with CPs on a frequent basis. Short-chain CPs (SCCPs: C10–C13) have been restricted by the Stockholm Convention of persistent organic pollutants (POPs) since 2017.
Technical CP-mixtures contain hundreds of C- and Cl-homologues. C-homologues differ in carbon (nC) and Cl-homologues in chlorine (nCl) numbers, respectively. Together with the complex isotopic clusters

of polychlorinated compounds, such mass spectra contain ten thousand of ions. Therefore, the mass spectrometric analysis of the universe of different CP-homologues and transformation products is a challenge.

We hypothesize that CPs and transformation products such as unsaturated chlorinated olefins (COs), diolefins (CdiOs), and even triolefins (CtriOs) are released from the industry and households. The effluents end up in municipal wastewater treatment plants (WWTPs) and with it, in sewage sludge.
We present a comprehensive analysis of CPs and olefinic side-products in a sewage sludge collected from a Swiss WWTP in 1993, which includes effluents from industry (metal working and plastic industry) as well as communal wastewaters of about 100000 residents (40000 households). This archived sludge can be compared with more recent ones to provide information on temporal trends.
The CP analysis was performed by an LC-APCI-Orbitrap-MS. This relies on the non-destructive, soft ionization of the APCI source method forming unfragmented singly charged chloride adduct ions [M+Cl]-. Respective high-resolution mass spectra were examined for the presence of about 1320 homologues of CPs and olefinic materials. In other words, about 23000 ions had to be extracted from these mass spectra and evaluated with an R-based automatic spectra evalua-tion routine (RASER), which allowed a time efficient data processing. The method has been introduced lately and ap-plied for the analysis of plastic consumer products collected from the Swiss market [1, 2, 3]. Herein, we present charac-teristic fingerprints of Swiss sewage sludge samples. In fact, we prove that various CPs and COs (nC = 10–33, nCl = 3–12) are released into WWTPs and consequently into sewage sludge.

[1] O. Mendo Diaz, et al., submitted (2023).
[2] M. Knobloch, et al., Anal. Chem., 94 (2022) 13777-13784.
https://doi.org/10.121/acs.analchem.2c02240
[3] M. Knobloch, et al., Chimia., 77 (2023) 68.
https://doi:10.2533/chimia.2023.68

PS2-53 Tyre wear ingredients: Markers and environmental behaviour in soil

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Tyre wear is considered one of the main contributors to microplastics in the environment [1]. Car tyres contain various materials, including rubber, carbon black and polyester, as well as several additives such as vulcanization accelerators, corrosion inhibitors or UV light stabilizers. Since tyres are complex mixtures, whose composition varies greatly depending on the manufacturer, marker substances for tyre abrasion are sought for estimating the total load into the environment. In this study, 2-(4-morpholinyl)-benzothiazole (MoBT), N-cyclohexyl-1,3-benzothiazol-2-amine (NCBA) and 1,2-dihydro-2,2,4-trimethylquinoline (TMQ) were investigated as marker substances for tyre particles specifically in soil.

Nine different soil samples and four tyre samples from different manufacturers were extracted using accelerated solvent extraction (ASE) followed by gas chromatography coupled to time-of-flight mass spectrometry (GC-QTOF). As data on environmental behaviour of these additives is scarce, measurements of the octanol-water partition coefficient (Kow) were done according to OECD 117 and degradation experiments were performed with TMQ spiked in activated sludge from a sewage treatment plant.

The extraction method was optimized, instrumental detection limits were evaluated on the GC-QTOF instrument, and recoveries for total sample work-up were determined using isotopically labelled internal standards. The mass fractions of these additives were determined in the soil samples and were in the range of a few µg·kg-^{1}. NCBA was found in all soil samples except one soil sample collected remotely off roads. Analysis of the tyre samples revealed that MoBT was found in two of four different tyres, while NCBA and TMQ were found in all tyre samples in the range of 5 - 85 mg·kg-^{1}. The gas chromatographic separation applied together with the high-mass resolution allowed the identification of markers in complex mixture of extracted soil with or without spiked tyre wear particles.

Measured partition coefficients of four measured additives had all a $\log_{10}$ Kow > 2. The sludge degradation experiment done with TMQ revealed that around 90% of this marker contained at the start time has been degraded in active sewage sludge after 72 h.

[1] B. Baensch-Baltruschat, B. Kocher, C. Kochleus, F. Stock, G. Reifferscheid, Sci. Total Environ. 752 (2021) 141939. doi.org/10.1016/j.scitotenv.2020.141939.

PS2-54 Analysis of Per- and Polyfluoroalkyl Substances in Aqueous Samples by SPE and LC-MS/MS according to EPA Draft Method 1633

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In September 2021, the United States Environmental Protection Agency (US EPA) has published a draft method for the analysis of per- and polyfluoroalkyl substances (PFAS) [1]. The draft method is a single laboratory validated method to test for 40 PFAS compounds in a diverse range of environmental matrices including wastewater, surface water, groundwater, soil, biosolids, sediment, landfill leachate, and fish tissue. The guideline can be used in various applications, exemplarily for use in the Clean Water Act (CWA) or the National Pollutant Discharge Elimination System (NPDES) [2]. These bio-accumulative pollutants are characterized by a linear aliphatic backbone, a high degree of fluorination, and often feature a carboxylic- or sulfonic- acid functionality. People can be exposed to PFAS in a variety of ways, including drinking water. But the exposure to PFAS can lead to adverse health effects. Many studies have examined possible connections between the level of per- and polyfluoroalkyl substances in the blood and adverse health effects in humans [3]. The research suggests that high levels of certain PFAS can lead to an increased cholesterol level, a reduced vaccine response in children, changes in liver enzymes, an increased risk of high blood pressure or pre-eclampsia in pregnant women and so on.
In this work, a SPE method according to EPA Draft Method 1633 using CHROMBOND® WAX is presented. High recovery rates with very good reproducibility are achieved for drinking water matrices. Finally, the extracts are analyzed using HPLC-MS/MS on a NUCLEODUR® PFAS column.

[1] Draft Method 1633, Analysis of Per- and Polyfluoroalkyl Substances (PFAS) in Aqueous, Solid, Biosolids, and Tissue Samples by LC-MS/MS; August 2021.
[2] National Pollutant Discharge Elimination System (NPDES), 832-F-12-033.NTP
[3] MONOGRAPH ON IMMUNOTOXICITY ASSOCIATED WITH EXPOSURE TO PERFLUOROOCTANOIC ACID (PFOA) OR PERFLUOROOCTANE SULFONATE (PFOS), September, 2016, Office of Health Assessment and Translation Division of the National Toxicology Program National Institute of Environmental Health Sciences National Institutes of Health.

PS2-56 Determination of 2-chloroethanol as a marker of fumigant ethylene oxide in sesame seeds by HS-SPME-GC-MS

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Ethylene oxide (EO) gas is used as a fumigant for the control of insects and microorganisms in food commodities [1]. But it reacts as well with natural chlorides present in the food matrix to form 2-chloroethanol, a known carcinogen, that may persist in the food product for long periods of time, even throughout food processing [2,3]. Due to the demonstrated harmful health effects of these compounds, the employment of EO as a fumigant for food commodities is being progressively regulated or banned in several countries [4]. Currently, Europe controls the use of this fumigant in food by regulation (EU) 2015/868, which defines its concentration as a sum of EO and 2-chloroethanol, with a permissible concentration of 0.05 mg/kg in nuts, oil fruits, and oil seeds [5]. The aim of this study was to develop a fast screening method by HS-SPME-GC-MS to detect and quantify 2-chloroethanol as a marker for EO fumigation in sesame seeds.

Four commercially available SPME fibers were used to determine the best selectivity for headspace extraction of 2-chloroethanol from sesame seeds followed by GC-MS analysis. Beside of fiber selectivity, extraction time and temperature were studied and optimized. The method was validated in terms of linearity, limits of detection (LOD), limits of quantification (LOQ), accuracy, and repeatability. The final method was applied on different sesame seed samples.

An HS-SPME-GC-MS method using a Carboxen-PDMS fiber was developed as a fast screening approach for 2-chloroethanol as a marker of ethylene oxide in sesame seeds. This SPME method yielded good sensitivity, accuracy, and reproducibility when applied to the analysis of sesame seed samples.

[1] S. Conviser, AICCM Bulletin, 28 (2003) 71-75. https://doi.org/10.1179/bac.2003.28.1.015
[2] D.Z. Hun, et al., J Clin Toxicol. 6 (2016) 1-3. DOI: 10.4172/2161-0495.1000314
[3] B.L. Oser, et al., Food Chem Toxicol, 13 (1975) 313-315. DOI: 10.1016/s0015-6264(75)80291-4
[4] J. Fowles, et al., Food Chem Toxicol. 39 (2001) 1055-62. DOI: 10.1016/s0278-6915(01)00052-7
[5] Commission Regulation (EU) 2015/868 of 26 May 2015 as regards maximum residue levels for 2,4,5-T, …, ethylene oxide, … in or on certain products. OJ L 145, 10.6.2015, p. 1–71. http://data.europa.eu/eli/reg/2015/868/2015-06-10

PS2-57 Development and analysis of flavonoids and phenolic acids from mandarin fruits by LC-DAD/MS

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Flavonoids and phenolic acids are the major compounds in citrus fruit. Flavonoids are found in various amounts in different parts of the fruit. Generally, citrus peels had more abundant flavonoids and higher contents than pulps and juices. In this study, we have developed and validated the method for the determination of 20 compounds of flavonoids and phenolic acids in mandarine juice, pulp, and peel. A 0.1 g of sample of dried mandarin peel or pulp was extracted by 1 ml of a mixture of methanol–dimethylsulfoxide (v/v; 50:50). A mandarine juice (0.6 mL) was extracted with the same volume of the mixture of methanol– dimethylsulfoxide. Two types of columns were tested: a diisobutyl n-octadecylsilane (C18) stationary phase (Poroshell 120 SB-C18 4.6x150mm, 4-µm) and a phenyl-hexyl

stationary phase (Phenyl-Hexyl 100 A CL 4.6x250mm, 5-µm). A better resolution was achieved for the majority of the compounds with a diisobutyl n-octadecylsilane (C18) stationary phase. The recoveries range from 95 % to 100.6 % for both columns used. At phenyl-hexyl stationary phase recoveries were only better for polymethoxyflavanones; sinesetin, nobiletin, and tangeretin. There were no significant differences between the quantification of phenolic acids and flavonoids in both columns. For further analysis of samples, Poroshell 120 SB-C18 4.6x150mm 4-µm was used at 40 ^{o}C with a gradient method using 2 % of formic acid as mobile phase A and methanol as mobile phase B. Following gradient was used (t/min, %B): (0, 10), (10, 20), (20, 30), (30, 40), (35, 40), (42, 50), (52, 90), (53,10) at flow rate 0.8 mL/min. A diode array detector was used for quantification at 280, 320, 330, and 360 nm. Mass spectrometry was used for identification. The quantity of free and bound phenolic acids in mandarin peels that had previously been hydrolyzed by NaOH was also determined.

PS2-58 Evaluating the potential of Irish Faba Beans as a protein alternative using multiple analytical techniques (FAAS, GFAAS & Kjeldahl Method)

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Faba beans have commonly been used as animal feed due to their high protein and energy concentration but are increasing in popularity as an animal protein replacement in the human diet. They are well suited to the Irish temperate climate with a high yield potential and can be harvested in both winter and spring. The bean is easy to grow, has plant hardiness and can withstand harsh and cold climates. The beans produce fixed nitrogen improving soil quality for itself and succeeding crops. They have a low nitrogen requirement which results in lower greenhouse gas emissions. Nutrient levels in faba beans were determined using Flame Atomic Absorption Spectroscopy: 620mg/kg calcium, 1199mg/kg magnesium, 36mg/kg iron, 38mg/kg zinc, 9mg/kg manganese and 9321mg/kg potassium. Graphite furnace atomic absorption spectroscopy was used to determine the levels of undesirable toxic metals cadmium and arsenic in the beans. This study found that Irish faba beans contained no arsenic and mean cadmium concentration of 25 µg/kg, comparable with what is found in vegetables such as cabbage and lettuce. Crude protein concentration of faba beans was determined using the Kjeldahl method with a mean value of 28%. Faba beans were found to be rich in many minerals important in the diets of both humans and animals. Crude protein of 28% confirms faba bean's suitability for use as a protein rich animal feed alternative. The world is facing a huge shortage of protein sources for livestock feed. With their suitability for growth in a range of climates and soils, large scale cultivation of faba bean could enhance Europe's capability of becoming self-sufficient in their animal feed requirements. This in turn would greatly reduce need for importation which has both environmental and financial impacts.

PS2-59 A novel HPLC-DAD method for determination of hydrogen peroxide in milk

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Hydrogen peroxide is one of the major adulterants in milk. Milk adulteration with hydrogen peroxide has been widely reported in developing countries. Food and Drug Administration (FDA) of the US allows the presence of hydrogen peroxide only in milk used to make cheese. Although, as a result of chemical processes in raw milk it may contain small amounts of hydrogen peroxide (1–2 mg/L), the concentration must be 10 times higher in order to destroy pathogens. At the same time, high concentrations of H2O2 in milk can lead to changes in its chemical composition, which, in turn, can lead to negative effect for the consumers.

In this work, a diode-array detection HPLC method based on oxidation of leuco-methylene blue to methylene blue was used for the determination of hydrogen peroxide in milk. Leuco-methylene blue was synthesized in situ using sodium borohydride and methylene blue in a reactor flask with an argon medium. Then the alkaline products were neutralized using a citric acid solution and a milk sample were added. The process of reduction of methylene blue to leuco-methylene blue takes about an hour. Even though milk consists of a lot of substances, it does not contain oxidizing agents that can react with leuco-methylene blue. The leuco-methylene blue formation was proved using an NMR-spectroscopy.

We have validated the determination of hydrogen peroxide in milk using suggested technique. We have established time of reducing and oxidizing reactions and developed HPLC conditions. We have used C18 column with mobile phase consisted of acetonitrile and 0.3 H3PO4 in gradient elution. LOQ was about 10 mg/L, the standard deviation was about 7 %. Recovery of hydrogen peroxide was from 97 % to 102 %. It was also found that the percentage of fat content of milk has no effect on the determination of hydrogen peroxide. Our method is applicable for raw milk, but it must be boiled beforehand, because raw milk can reduce some amount of methylene blue.

The proposed method can be considered suitable for use in most laboratories (routine technique) and is sensitive enough for the determination of hydrogen peroxide at the level of authorized standards in different countries.

PS2-60 Continuous monitoring of Lactoferrin for real-time process control

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Lactoferrin is a multifunctional iron-binding glycoprotein that is present in secretory fluids and plays a crucial role in supporting the immune system. Due to its beneficial properties (antiviral, antibacterial, anti-inflammatory, anti-tumor, antioxidant), lactoferrin is extracted from bovine milk for use as a supplement in infant nutrition and skin care products. However, milk exhibits fluctuating levels of lactoferrin, which complicates the extraction process and results in suboptimal yields. This calls for the

development of real-time measurement-and-control strategies that can optimize the extraction efficiency and minimize resource usage. Here, we describe the development of a biosensor technology for real-time lactoferrin measurements. The sensor is based on Biosensing by Particle Motion (BPM), a continuous sensing principle with single-molecule resolution [1]. In BPM, particles functionalized with antibodies hover over a biofunctionalized substrate. In the presence of target protein, a sandwich bond is formed between the particle and the substrate, which is optically detected by the restricted motion of the particle. To achieve lactoferrin quantification within short timespans, it is crucial to quantify and understand the mechanisms that limit the speed of the assay. We will present results on the development of the lactoferrin BPM assay and elucidate the parameters influencing its time resolution [2]. Finally, we discuss how the sensing methodology can enable real-time process control for sustainable food, biotechnology, and life science processes.

1. A. D. Buskermolen, et al., Continuous biomarker monitoring with single molecule resolution by measuring free particle motion. Nature Communications, 13.1 (2022), 6052.
DOI:10.1038/s41467-022-33487-3
2. C. Michielsen, et al., Sandwich immunosensor with particle labels: how do reactant concentrations and diffusion determine the speed of the sensor response?, in preparation

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PS2-61 Classification of Soybean Paste Products Using Laser-Induced Breakdown Spectroscopy, Inductively-Coupled Plasma Optical Emission Spectroscopy, and Inductively-Coupled Plasma Mass Spectrometry

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Soybean paste is one of the most popular fermented foods consumed in East Asia. South Korea exports a large amount of soybean paste to China, and Korean soybean paste is also produced in China. In this work, the elemental analysis techniques of laser-induced breakdown spectroscopy (LIBS), inductively-coupled plasma optical emission spectroscopy (ICP-OES), and inductively-coupled plasma mass spectrometry (ICPMS) were employed to distinguish Korean soybean pastes produced in South Korea and China from each other. 101 products from South Korea and 66 ones from China were collected to model their provenances. In the LIBS spectra, atomic or ionic emissions of Na, Cl, Mg, Ca, K, P, C, and H were identified. Among them, the emissions of Mg and C were found to possess most of the power discriminating soybean paste products from South Korea and China. From the ICP-OES analysis, the concentrations of Al, Ba, Sr, Cu, Fe, Mn, Zn, Ca, and Mg could be determined. The discrimination power obtained from these elements was found to be dominated by Mg and Sr. The provenances, South Korea and China were modeled by partial least squares discriminant analysis (PLS-DA). The PLS-DA models based on LIBS (C and Mg) and ICP-OES (Mg and Sr) showed the discrimination accuracy of 90.3% and 91.2%, respectively. For the samples that could not be correctly distinguished, Ni, Mn, and Co, analyzed by ICP-MS, were found to provide additional independent discrimination power. The accuracy from the two-step PLS-DA model based on ICP-OES and ICP-MS reached up to 98.6%. Our results suggest that the elemental analysis techniques can provide highly reliable methodologies for screening the soybean paste products with fake origin labels.

PS2-62 Developing analytical method for the determination of Inpyrfluxam and its metabolites residues in agricultural products

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Inpyrfluxam is a novel, broad-spectrum fungicide that prevents energy production of tricarboxylic acid cycle in pathogenic fungi. It shows high efficacy against major plant disease, such as brown rust on wheat and rice sheath blight, by suppression of Complex II succinate dehydrogenase. There is no suitable experimental method for the determination of Inpyrfluxam and its three metabolites(3'-OH-S-2840, 1'-CH2OH-S-2840A, 1'-CH2OH-S-2840B) residue in agricultural products. In this study, we established an analytical method to determine the residue of Inpyfluxam and its three metabolites simultaneously in foods. For a test to be reliable, five agricultural products(mandarin, potato, soybean, hulled rice, and chili pepper) showing group representatives were chosen. The sample preparation process was conducted in two steps with QuEChERs(quick, easy, cheap, effective, rugged, and safe) method for making the extraction and purification process efficient. First, in order to extract the samples, acetonitrile was added and shaken for 10 minutes. Thereafter, magnesium sulfate anhydrous, sodium chloride, disodium hydrogencitrate sesquihydrate and trisodium citrate dihydrate were added, followed by centrifugation at 4,000 G for 10 minutes. Second, to remove the interferences in samples, purification was performed using dispersive solid-phase extraction(d-SPE) with anhydrous magnesium sulfate and primary and secondary amine(PSA). After centrifugation at 4,000 G for 10 minutes, the supernatant was filtered by 0.2 μm of PTFE before LC-MS/MS analysis using C18 column. The standard calibration curves were linear within the ranges of 0.001-0.075 mg/L with determination coefficient, R2 > 0.99. Mean recovery rates of Inpyrfluxam and its metabolites were shown 79.2~113.3%. Also, the relative standard deviation(RSD) were shown to be less than 6.0% for all five samples, which were suitable for CODEX guideline on the pesticide residue analysis(CAC/GL 40). In summary, the developed method for determination of Inpyrfluxam and its metabolites residues in foods will be applied on the safety management of pesticides by including in the Korean Food Code.

PS2-63 Stability of water-soluble vitamins in enteral food

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Consumption of enteral formulas for hospitalized, critically ill and home enteral patients has dramatically increased over the past few decades [1]. Enteral foods are used to provide all nutrients for individuals with limited, impaired, or disturbed capacity of taking, digestion, absorption, metabolism and excretion of common food. [2] The stability of vitamins in such formulations can be influenced by various factors, such as the type of packaging and condition of storage (e.g., oxygen exposure, light, humidity and temperatures) [3].

We have developed a sensitive liquid chromatography-mass spectrometry (LC-MS) method in single ion monitoring (SIM) mode for simultaneous stability testing of nine water-soluble vitamins, ascorbic acid (C), thiamine (B1), riboflavin (B2), niacin (B3), pantothenic acid (B5), pyridoxine hydrochloride (B6), biotin (B7), folic acid (B9) and cyanocobalamin (B12), respectively, in different type of formulations. Depending on rheological characteristic of particular formulation, extraction protocol for water-soluble vitamins was optimized. Vitamin

content was determined in enteral food formulations and stability tests were performed. The samples were analyzed at the beginning of the storage period and three months after storage of the packaging

in temperature chambers at 37 °C and also after storage for six months at 23°C.The results obtained on 7 different formulations were correlated with the results obtained by other method.

[1] R. Baéz, G. Rojas, J. Sandoval-Guillén and M.A. Ángeles Valdivia-López, Adv. J. Food Sci. Technol., 4 (2012) 235-242.
[2] T. Delompre, E. Guichard, L. Briand and C. Salles, Nutrients, 11 (2019) 2050–2066.
[3] M. Edelmann, B. Chamlagain, M. Santin, S. Kariluoto and V. Piironen, Food Chem., 204 (2016) 21–28.

PS2-64 GC/MS/MS AS A THE BEST TECHNIQUE FOR DETECTION AND IDENTIFICATION OF LONG-TERM STEROID MARKERS IN DOPING CONTROL

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The nonmedical use of anabolic androgenic steroids (AASs), along with the improved oral bioavailability and beneficial effects on muscle, started shortly after the first identification of testosterone structure in 1935. In the following years, the synthesised designer steroids appeared on the black market, tempting both athletes and amateurs of sports with outstanding performance results. In response to this phenomenon, the use of the AAS, in- and out-of-competition by athletes, has been prohibited since 1974. Nevertheless, the performance-enhancing drugs containing the AASs are the most commonly abused products, whereas the AASs itself constitute above 60% of the adverse analytical findings (AAFs) worldwide.

The sophisticated violators of anti-doping rules know that the timing of AAS consumption is crucial for the so-called “safe” use in the “training“ period. When their use is discontinued in an adequate time preceding the scheduled competition, the anti-doping tests are believed to ensure negative test results. Therefore, all anti-doping laboratories are obligated to constantly update and improve their analytical methods to detect and identify the parent compound as well as the short- and long-term metabolites of prohibited substances.

AASs such as oxandrolone (OXN), metandienone (MTD), desoxymethyltestosterone (DMT) and dehydrohloromethyltestosterone (DHCMT) show how the identification of long-term metabolites can improve the detection of AASs in urine samples collected from athletes in the time period up to even several months after the drug application. Moreover, the development of analytical instrumentation has also contributed significantly to the fight against doping.

The number of AAFs in the Polish Anti-Doping Laboratory has escalated in the last 11 years due to the increased number of analysed samples, instrumentation improvement and methods development. The significate influence on the method development had the paper of the DMT and DHCMT long-term metabolites identification published by Tim Sobolewski1 in 2012.

The anti-doping urine samples collected in the Polish Anti-Doping Laboratory, in the period of January 2010 – December 2022, were tested for the presence of OXN, MTD, DMT, DHCMT and their metabolites. Qualitative analyses were conducted by gas chromatography-mass spectrometry (GC/MS) up to May 2012 and gas chromatography-tandem mass spectrometry (GC/MS/MS) since June 2012. The obtained AAF results have shown 1 – 6 cases per year with the presence of OXN, 1 – 13 cases per year with the presence of MTD, 1 case per year with the presence of DMT and 1 – 19 cases per year with the presence of DHCMT.

T. Sobolevski, et al., Drug Test. Analysis, 4 (2012) 682-691.
DOI: 10.1002/dta.1407

PS2-66 Investigation of bioactive metabolites in 36 Iris species and cultivars grown under different cultivation conditions

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The Iris genus comprises hundreds of species, but only some have been tested for antioxidant, antibacterial, anti-cancer, and other favorable bioactivities. Nevertheless, identification of the respective bioactive metabolites remains a challenge, mainly due to lack of analytical standards. The objective of our study was to (i) characterize the metabolic profile of a unique set of 160 extracts of leaves and rhizomes with roots of 36 Iris species and cultivars, (ii) measure their bioactivity (antioxidant, antimicrobial, anti-inflammatory, cytotoxic) and identify potential bioactive secondary metabolites, and (iii) evaluate the effect of cultivation conditions (aeropony/hydropony) and plant treatment (animal hydrolysate/vesiculo-arbuscular mycorrhiza) thereon. We have developed a high-throughput UHPLC-ESI-HRMS/MS target screening method using an in-house created spectral database containing elemental formulas of 293 metabolites reported to occur in IrisThe optimized chromatographic separation used BEH C18 column and gradient elution with (A) H2O and (B) methanol, both containing ammonium formate buffer. Metabolites were detected using the Q-TOF mass analyzer in positive and negative ionization. In total, 893 compounds complying with the m/z value of target metabolites were detected in MS1, some of which were further identified using analytical standards, online and in silico MS/MS fragmentation spectra. Among the detected compounds were (iso)flavonoids, phenols, terpenoids, phenolic acids, xanthones, steroids, and quinones. In terms of signal intensity, xanthones dominated in the leaves, of which the bioactive mangiferin was in general the most intense. On the other hand, in the rhizomes with roots the signals of terpenoids prevailed. The analysis revealed distinct chemical profiles of different Iris species, whereas the profiles of different cultivars were fairly similar. Most compounds were detected in the rhizomes with roots of Iris squalens and least in the leaves of Iris lactea, both cultivated hydroponically. The effect of cultivation mode (aeroponic/hydroponic) on the metabolic profile proved to be statistically significant (t-test, PCA, PLSDA) with different effects, depending on the specific metabolite. Statistically significant effect of plant treatment with animal hydrolysate or vesiculo-arbuscular mycorrhiza on the profile of bioactive compounds was not confirmed. Additionally, correlational analysis revealed a strong relationship between the content of four secondary metabolites (4’-hydroxy-3’-methoxyacetophenone/4'-hydroxy-2'-methoxyacetophenone/3’-hydroxy-5’-methoxyacetophenone, selenone, neomangiferin and eupatorin, the latter two confirmed by MS/MS) and antibacterial and anti-inflammatory activity. In our view, this study opens doors to further research of these metabolites, their identification and assessment of bioactive potential.

PS2-67 Adaptative response to tetraconazole-based fungicides shapes the proteome of Saccharomyces cerevisiae Lalvin EC1118™

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Yeasts may change their protein profile due to exposure to certain stress situations during their growth and development stages. Thus, the presence of fungicide residues can lead to changes in the gene expression pattern of yeasts that subsequently can be translated into changes in the protein profile (like post-translational modifications, phosphorylation, and increments or decrements in the protein abundance) altering the metabolic pathways of fermentation. This work aimed to analyse the impact of tetraconazole and the adjuvant sodium docusate on the proteome of S. cerevisiae Lalvin EC1118™. The individual and the joint effect of both substances was evaluated at laboratory scale through microvinifications assays of synthetic must (n=3). Yeast cells were collected at different fermentation times for transcriptomic (20, 36, and 54 h) and proteomic (30, 54, and 68 h) analyses. Yeasts were centrifuged, snap-frozen in liquid nitrogen, and stored at -80 °C until use. After isolation, RNA was sequenced on a NextSeq Sequencing System from Illumina [1] and soluble protein extracts were digested with trypsin prior to LC-MS/MS analysis on a hybrid high-resolution LTQ-Orbitrap Elite Mass Spectrometer [2].

From a transcriptomic point of view, major changes were observed after 36 h of fermentation for tetraconazole (80 genes differently expressed) and at time 20 h for the mixture tetraconazole + sodium docusate (69 genes differently expressed). No important changes were observed after the individual addition of sodium docusate, independently of the time point considered. Concerning the proteome, the addition of tetraconazole or the mixture tetraconazole + sodium docusate promoted a decrement of the abundance of an elevated number of proteins (186 and 172 proteins, respectively) with respect to the control yeasts (grown in a synthetic medium without fungicides) after 54 h of fermentation. However, for sodium docusate alterations were predominant observed after 30 h of fermentation (210 proteins with a decreased abundance). The principal biological process affected by the presence of these compounds are related with the metabolism of amino acids, lipids, and carbohydrates, stress response, transport of electrons and carbohydrates, and phosphorylation.

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[1] J. Alonso-del-Real, et al., Environ. Microbiol. 21 (2019) 1627. DOI: 10.1111/1462-2920.14536
[2] T. Sieiro-Sampedro, et al., Food Microbiol. 92 (2020) 103554. DOI: 10.1016/j.fm.2020.103554

PS2-68 Development of a HILIC-MS/MS method for covering short, medium and long chain acyl-CoA in one analytical run

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Acyl-CoAs are a group of thioesters compound and have a pivotal role in various metabolic processes such as fatty acid beta oxidation, biosynthesis of lipids, signalling, xenobiotics metabolism etc.[1,2] The most important biological function of acyl-CoA is in the metabolism of fatty acids via beta oxidation. Since acyl-CoA are involved in numerous physiological and pathophysiological pathways, it is important to develop analytical methods for their identification and quantification. The physicochemical properties of acyl-CoA vary greatly depending on carbon chain length, degree of saturation, and functional groups. Various efforts have been made to cover the full range of acyl-CoA in a single analytical run, although with limitations for application in routine clinical analysis. We have developed a bioanalytical method using hydrophilic interaction chromatography (HILIC) coupled with mass spectrometry as detector. With of help of our HILIC-MS/MS method, we were able to cover entire range of acyl-CoA compounds in one analytical run. The neutral loss of 507 is the diagnostic fragment that has been used for the identification of acyl-CoA. Various other chromatographic parameters such effect of buffer, injection solvents, pH etc. were determined.

1. Carvalho, et al., Molecules 2018, 23, 2583.
Doi:10.3390/molecules23102583.
2. Brass, et al., Chemico-Biological Interactions 1994, 90, 203–214.
Doi:10.1016/0009-2797(94)90010-8.

PS2-69 SCORE-metabolite-ID – Identification of metabolites from complex mixtures by correlation of 1D-1H or 2D-HSQC NMR, MS and LC data

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The identification of metabolites is an extremely relevant factor in metabolomics studies. Due to the large number of metabolites, which also occur in strong concentration differences, resulting in strong signal overlaps, the identification of individual metabolites from either NMR or MS spectra of natural products is hardly possible.

Since NMR and MS provide complementary information, the combination of both analytical methods is a promising approach. In our SCORE-metabolite-ID approach, this combination is achieved by correlation of NMR and MS data of the time course of a chromatographic fractionation. After liquid chromatography, NMR and MS spectra of each fraction are acquired individually. The resulting EDCs (extracted delta chromatograms) and EMCs (extracted mass chromatograms) can be correlated by Pearson correlation using the MATLAB app “SCORE-metabolite-ID”. This correlation strategy leads to the detection of related signals in the NMR spectra themselves as well as associated mass-to-charge ratios from the ESI mass spectra. This correlation of NMR and MS data can be performed semi-automatically. As a result, the MATLAB based app enables simple, efficient and reliable identification of metabolites without the need for individual isolation. The tool can be used either with ^{1}H NMR spectra or 2D HSQC spectra.

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DOI: 10.26434/chemrxiv-2022-mv6cl-v2

PS2-70 Application of microdialysis combined with UHPLC-QTOF/MS to screen for endogenous metabolites in aquatic organisms as biomarkers of exposure to an emerging contaminant, triclosan

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Triclosan (TCS), as an emerging contaminant, is a commonly used antibacterial agent widely present in the environment. Microdialysis (MD), as a sampling technique, can overcome some of the deficiencies of traditional approaches to sampling. In this study, we coupled MD with analysis using UHPLC-QTOF/MS to identify the endogenous metabolites in the liver as biomarkers of the exposure of crucian carp to TCS. The identified biomarkers were then quantified using UHPLC-MS/MS to continuously monitor the effect of TCS on endogenous metabolites in the liver of living crucian carp, which contributes to a better understanding of the toxicological effect of TCS. In the OPLS-DA model, the differences among groups are smaller but get noticeably further away from the control group with time. Metabolites resulting from the exposure to TCS at different exposure time, were studied in a combined V+S-plot. The production and levels of metabolites observed were attributed to the behavior of the fishes when exposed to TCS. The measured concentration of the 6 different metabolites from TCS exposure varied with time. The results demonstrated that TCS exposure interfered with the metabolic pathways of amino acids (L-isoleucine and L-histidine), purines (xanthine and hypoxanthine), and small nerve molecules (acetylcholine and choline).

PS2-71 Untargeted urinary metabolomics for identification of bladder cancer biomarkers using HPLC-MS

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Bladder cancer (BCa) constitutes tenth amongst the most common cancers and is considered one of the most widespread diseases worldwide with about 400,000 new cases each year. BCa has a high mortality and recurrence rate and high treatment costs as well.

Traditional methods of dealing with BCa such as invasive cystoscopy (the gold standard) and invasive cytologic screening have certain disadvantages both for the patient and for the reliability of cancer diagnosis. The former is painful and can cause infectious complications, the latter is characterized by low sensitivity and often gives false-negative results.

Detection of a noninvasive, highly sensitive and specific disease-related marker is urgently needed to improve the control and quality of life of BCa patients and to supplement current clinical methods.
In this regard, urinary metabolomics is a powerful approach as urine has direct contact with malignancies and thus is enriched with final products of the cellular processes or cancer-specific metabolites.

In this study we suggest an approach based on high-performance liquid chromatography coupled to mass detection (HPLC-MS) as one of the most powerful analytical techniques in terms of sensitivity and information value for profiling all the metabolites present in a given specimen, ensuring better analytical coverage of a metabolomics profile in an untargeted metabolome analysis.

We propose an algorithm for obtaining study results that includes integration of chromatographic peaks and retention time alignment, signal drift correction and elimination of undesirable variance in the data followed by statistical analysis to obtain an optimal panel of biomarkers for reliable classification of samples by experimental groups (PLS-DA, PCA, dendrograms were applied for data interpretation and urinary metabolite profiling).

The HPLC separation was conducted on a C18 column in a gradient elution mode with MS detection in a positive mode. “Dilute and shoot” technique was used for sample preparation. MetFrag software and databases (KEGG, HMDB, and Metabolomics Workbench) were used to identify the obtained biomarkers and corresponding regulation pathways.

The developed approach enables to fully separate the samples into initial groups using multivariate statistical analysis. Using machine learning techniques, 4 potential biomarkers were identified and chromatographic and mass spectral characteristics were established. Preliminary identification was performed and possible metabolic regulation pathways of these metabolites in humans were proposed. Current approach compared to traditional medical diagnostic methods benefits from high degree of automation, cost-effectiveness, noninvasiveness and ease of data interpretation. Obtained results can be applied in the diagnosis of BCa.

PS2-72 Electrochemical Approach on Interaction of Nerve Agent Metabolite and Albumin

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Albumin is one of the most abundant proteins, and it also plays an important role in the transport in blood [1]. The interactions between albumin and drugs/biomolecules/metabolites have great interest in the study of pharmacokinetics and pharmacodynamics of molecules (drug, biomarker, biomolecules, etc.). Biomolecule/drug-protein interaction needs to be carefully investigated and understood since it has a great influence on molecule metabolism, circulation and free concentration. Thus, investigation of drug molecules or metabolites with respect to their binding with human serum albumin (HSA) is important. Ethyl methylphosphonic acid (EMPA) is hydrolysis products of the nerve agents VX (S-2-diisopropylaminoethyl O-ethyl methylphosphonothiolate) [2]. The electrochemical techniques are effective tools to investigate the interaction between serum albumin and drug/biomolecule [3]. The interaction between EMPA and HSA was investigated via electrochemical techniques, including cyclic voltammetry (CV) and differential pulse voltammetry (DPV). The CV results demonstrated that the interaction of EMPA with albumin on a glassy carbon electrode (GCE) surface was controlled by an adsorption process in 5 mM K3Fe(CN)6/K4Fe(CN)6 with a scan rate of 100 mV/s in the potential range of -0.2-0.8 V. The addition of HSA was found to decrease the peak potential of redox probe without altering the electrochemical parameters, which is likely due to the formation of an electro-inactive complex between the drug and protein, as supported by CV and DPV measurements (Pulse potential: 0.2 V; Pulse time: 0.02 s; Scan rate: 0.1 V/s; Step potential: 8 mV). The binding constant and stoichiometry of the complex EMPA-HAS were investigated both CV and DPV techniques.

[1] J.J. Vallner, Binding of drugs by albumin and plasma protein, J. Pharm. Sci. 66 (1977) 447–465.

https://doi.org/10.1002/jps.2600660402
[2] M Katagi 1, M Nishikawa, M Tatsuno, H Tsuchihashi, Determination of the main hydrolysis product of O-ethyl S-2-diisopropylaminoethyl methylphosphonothiolate, ethyl methylphosphonic acid, in human serum J Chromatogr B Biomed Sci Appl. 1997 Feb 21;689(2):327-33. https://doi.org/10.1016/s0378-4347(96)00356-8
[3] J. Wang, C. Xiang, F.F. Tian, Z.Q. Xu, F.L. Jiang, Y. Liu, Investigating the interactions of a novel anticancer delocalized lipophilic cation and its precursor compound with human serum albumin, RSC Adv. 4 (2014) 18205– 18216. https://doi.org/10.1039/C3RA46997B

PS3-01 The study of topotecan sorption/desorption kinetics for poly(2-hydroxyethyl methacrylate) gels by UHPLC-MS/MS

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Topotecan (TPT) is a chemotherapy drug used to treat retinoblastoma. TPT undergoes hydrolysis of the lactone ring and changes to the carboxylate form (1). However, only the lactone form exhibits antitumor activity (2). The ratio of both forms at equilibrium and the rate of lactone conversion to the carboxylate form are influenced by the pH value. At physiological pH, the lactone and carboxylate form are in balance. In acidic pH, the lactone form predominates and in alkaline, TPT exists mainly in the carboxylate form (3).

Retinoblastoma is the most common primary malignant intraocular tumor in children and systemic chemotherapy is currently the most common treatment. However, a significant disadvantage of this drug administration is that only a very small part of the distributed drug reaches the tumor. The treatment is often associated with serious side effects. Therefore, it would be advantageous to use a system for the local distribution of chemotherapy drugs to achieve a high concentration of the drug in the target organ and reduce the drug toxicity to other tissues. Polymer carriers, for example in the form of nanoparticles or hydrogels, may be useful as local drug delivery systems (4).
In this work, a two-layer lens-shaped hydrogel implant was developed for the distribution of TPT via transscleral diffusion. The implant consists of an inner hydrophilic poly(2-hydroxyethyl methacrylate) (pHEMA) layer adjacent to the sclera (TPT reservoir) and an outer covering hydrophobic poly(2-ethoxyethyl methacrylate) (pEOEMA) layer impermeable to TPT. An ultra-high-performance liquid chromatography with tandem-mass spectrometric detection (UHPLC–MS/MS) method was developed to determine the TPT of the lactone and carboxylate form in water and aqueous buffer solutions. This method was used to study the sorption/desorption kinetics of TPT in a hydrophilic gel simulating in vivo conditions.

(1) Fassberg J, Stella VJ. A kinetic and mechanistic study of the hydrolysis of camptothecin and some analogues. J Pharm Sci. 1992;81(7):676-84.
(2) Kollmansberger C, Mross K, Jakob A, Kranz L, Bokemeyer C. Topotecan – A novel topoisomerase I inhibitor: Pharmacology and clinical experience. Oncology. 1999(56):1-12.
(3) Underberg WJ, Goossen RM, Smith BR, Beijnen JH. Equilibrium kinetics of the new experimental anti-tumour compound SK&F 104864-A in aqueous solution. J Pharm Biomed Anal. 1990;8(8-12):681-3.
(4) Cocarta AI, Hobzova R, Sirc J, Cerna T, Hrabeta J, Svojgr K, et al. Hydrogel implants for transscleral drug delivery for retinoblastoma treatment. Mater Sci Eng C Mater Biol Appl. 2019;103:109799.

PS3-02 Synthesis, Characterization, and Anticancer Evaluation of Phenanthroline-Based Macrocyclic Ligand and Nickel Complex: DNA Binding and Thermal Stability Studies

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By condensation of neocuproine with a 2,9-dial derivative of phenanthroline and complexation processes, a novel 1-10 phenanthroline-based macrocyclic ligand (compound A) and its nickel complex (compound B) were produced. Fourier Transformed Infrared, Proton (1H), and Carbon 13 (13C) Nuclear Magnetic Resonance (NMR), and Thermal analyses were used to structurally analyze the produced compounds. The compounds' anticancer efficacy was assessed using Deoxyribonucleic Acid (DNA) and Guanine-Cytosine (GC) -rich Amplicon binding assays, as well as antioxidative assays. UV/Vis spectrophotometer, Polymerase Chain Reaction (PCR), and Gel Electrophoresis were used to assess their binding capacities. The compounds demonstrated high DNA-Ligand/complex binding activities, implying superior DNA intercalation and external binding capable of suppressing DNA PCR amplification. The interactions of compounds A and B with the GC-rich Amplicon resulted in binding constants of 1.00x106M-1 and 1.07 x108M-1, respectively, and 6.00 x 104 M-1 and 6.02 x106 M-1 with cell-free (cf)-DNA. They demonstrated a higher binding constant with cf-DNA than with GC-rich Amplicon. The hypsochromic shift seen in Amplicon-compound B interactions suggested a probable reduction in compound B's aromatic conjugation. At 500µg/ml, the compounds had approximately 26% RSA and individual IC50s of 1.95mg/ml for compound A and 1.11mg/ml for compound B. ThermoGravimetric Analysis (TGA) and differential thermal analysis revealed that they all had superior thermal stability (breakdown at an average of about 950K) and decompose via first-order kinetics. These compounds can aid in cell function regulation by altering transcription and/or interfering with cell replication and DNA repair processes.

Coury, J., McFail-Isom, L., Williams, L., Bottomley, L. (1996). A Novel Assay For Drug-dna Binding Mode, Affinity, and Exclusion Number: Scanning Force Microscopy.. Proc. Natl. Acad. Sci. U.S.A., 22(93), 12283-12286. https://doi.org/10.1073/pnas.93.22.12283
Cao, Q., Li, Y., Freisinger, E., Qin, P. Z., Sigel, R. K. O., Mao, Z. (2017). G-quadruplex Dna Targeted Metal Complexes Acting As Potential Anticancer Drugs. Inorg. Chem. Front., 1(4), 10-32. https://doi.org/10.1039/c6qi00300a
Dröge, T., Glorius, F. (2010). The Measure Of All Rings-n-heterocyclic Carbenes. Angewandte Chemie International Edition, 39(49), 6940-6952. https://doi.org/10.1002/anie.201001865
Giri, N., Passantino, L., Sun, H., Zoroddu, M., Costa, M., Maroney, M. (2013). Structural Investigations Of the Nickel-induced Inhibition Of Truncated Constructs Of The Jmjd2 Family Of Histone Demethylases Using X-ray Absorption Spectroscopy. Biochemistry, 24(52), 4168-4183. https://doi.org/10.1021/bi400274v

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PS3-03 A technique to analyze and measure the amount of tar generated from the pyrolysis of waste tyres

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To conduct practical research, a technology was chosen that involves quickly heating finely-ground oil shale. This is done by using a solid heat carrier, like hot ash, in a rotary drum reactor. A rotary kiln reactor located in Daugavpils, Latvia was used for the pyrolysis experiments. During the investigation of tar sampling from non-condensable gas produced during tyre pyrolysis, researchers discovered that

the most effective sampling device contained 500 mg of an amino-phase sorbent and 100 mg of activated coconut charcoal. This device was successful in capturing tar and the volatile organic compounds associated with it. The first column used was a 3 mL ISOLUTE® single fritted reservoir (Biotage), which contained 500 mg of aminopropyl-bonded silica adsorbent (ISOLUTE® NH2) with an exchange capacity of 0.6 meq g-1, a particle size of 50 µm, and an average pore size of 60 Å. The second column was a 1 mL ISOLUTE® single fritted reservoir (Biotage) containing 100 mg of activated coconut charcoal with a surface area of 1070 m2 g-1 and a particle size of 20/40 mesh, 420-840 µm.
The method suggested in this research for measuring the concentration of tar compounds requires less sampling time compared to existing methods. This enables faster monitoring of pyrolysis conditions based on tar analysis results in pyrolytic gas. Moreover, unlike other techniques, the proposed method can measure both heavy tar compounds and light tar compounds like benzene and toluene. There was no significant difference observed in the total amount of tar and its component compounds with the increase in the volume of pyrolytic gas. However, an increase was noted in the quantity of lighter compounds like benzene and toluene that passed through the amino-phase adsorbent and were collected on the activated coconut charcoal as the gas volume increased. As the volume of pyrolytic gas increased, more compounds were detected and identified on the amino-phase adsorbent. It is crucial to consider the concentration of tar in the pyrolytic gas while determining the sampled gas volume and identifying individual tar compounds with low concentrations.

PS3-04 Development of a high-throughput screening assay to identify glutathione S-transferase (GST P1) inhibitors for potential use in cancer treatment

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Glutathione transferases (GSTs) are a superfamily of phase II metabolizing enzymes that catalyze the conjugation of reduced glutathione (GSH) to endogenous and exogenous electrophiles, including carcinogens and a variety of chemotherapeutic agents. GSTP1 is the predominantly expressed GST subclass and its overexpression can be directly correlated with resistance against chemotherapeutic agents [1]. A wide range of marketed chemotherapeutic drugs, including alkylating agents and anthracyclines, are substrates of GSTP1 [2]. Thus, the overexpression of GSTP1 results in accelerated drug metabolism, leading to significantly decreased therapeutic efficiency.

To date, the most widely used GST isoform assay measures turnover using the model substrate 1-chloro-2,4-dinitrobenzene (CDNB) coupled with UV–vis detection. However, it has only been practiced in cuvette and 96-well formats and suffers from small signal windows (absorbance detection) and large protein consumption [3].

Hence, a high-throughput screening assay (HTS) was developed to assess 5830 compounds in order to identify putative drug compounds that target GST P1. Twenty-four hit compounds identified from the HTS were validated via dose-response inhibition assays and further evaluated for cytotoxicity in MCF-7 (estrogen and progesterone receptor + breast cancer cells) and MDA-MB-231 (triple-negative breast cancer cells).

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References
[1] Bräutigam M., et al. ChemMedChem 10(4) (2015) 629. DOI: 10.1002/cmdc.201402553.
[2] Singh, S. Cancer Chemother Pharmacol 75 (2015) 1. DOI: 10.1007/s00280-014-2566-x
[3] Sigma-Aldrich. Glutathione S-Transferase (GST) Assay Kit (Sigma Technical Bulletin for Catalog Number CS0410). St. Louis, MO, Author, 2009.

PS3-06 Polyampholite hydrogels organized by dynamic bonds

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Hydrogels have a wide range of uses due to their structural arrangement, water content, and ability to respond to external factors. However, the poor mechanical performance of conventional hydrogels narrows this spectrum [1,2]. In order to overcome this poor mechanical performance caused by chemical and rigid cross-links, researchers try to give hydrogels a dynamic structure where physical and chemical bonds coexist [1-4]. In this study, we investigated the swelling, mechanical and flow properties of polyampholite hydrogels that combine electrostatic and hydrophobic interactions. As a result of the modification of materials with various hydrophobic side chain polymers (tetra- C14A, hexa- C16A and octa-decyl acrylate C18A), the effect of the interaction in the dynamic structure on the material properties and possible applications were investigated. The self-healing and shape memory properties of the selected gels were tested.

1. https://doi.org/10.1016/j.eurpolymj.2017.01.029
2. https://doi.org/10.1007/s00396-020-04605-8
3. Toleutay, G., Su, E., & Kudaibergenov, S.E. (2019). Swelling and mechanical properties of quenched polyampholyte hydrogels based on 2-acrylamido-2-methyl-1-propanesulfonic acid sodium salt (AMPS) and (3-acrylamidopropyl) trimethylammonium chloride (APTAC).
4. https://doi.org/10.1016/j.eurpolymj.2022.111098

PS3-07 Pregnancy as a factor influencing the change of the steroid profile in terms of assessment of athlete's biological passport

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The concept of an Athlete Biological Passport (ABP) in a doping control is based on detecting doping-induced changes in selected biomarkers by monitoring them in the long term and comparing them with the values already obtained in the previous anti-doping analysis. It may indirectly allow the detection of the effects of doping, which may be an alternative to the detection of the substance itself or the use of a prohibited method to prove an athlete's anti-doping rule violation. Unfortunately, there are several factors that might influence steroid profile such as e.g. inter-individual variability of steroid

synthesis and metabolism (UGT2B17 polymorphism), pregnancy, contraceptive pills, alcohol, the administration of ketoconazole, human chorionic gonadotrophin in males, inhibitors of 5α-reductase and the influence of microorganisms existing in urine samples.

In the present work the changes in the hormonal balance in terms of factors changing the steroid profile of the athlete during pregnancy have been investigated [2]. For this purpose, a collaboration with a gynaecological clinic and pregnant women was established. The urine samples collected from seven healthy pregnant volunteers in the range of at least from seventy-third to one hundred and sixth day of pregnancy, were prepared according to the routine procedure in the Polish Anti-Doping Laboratory for steroid profile determination and analyzed using GC/MS system. Six steroid parameters such as androsterone, etiocholanolone, 5α-Androstane-3α,17β-diol, 5β-Androstane-3α,17β-diol, testosterone and epitestosterone, and five their derivatives such as T/E ratio, A/Etio ratio, A/T, 5α-Adiol/5β-Adiol ratio and 5α-adiol/E were analyzed. Additionally, the concentrations of hCG were estimated using the immunochemical analyzer.

The observations confirmed that pregnancy, even the early one, which the athlete may not know about, can affect the concentrations of the tested hormones as well as their ratios/indicators. The obtained results might to help interpret the steroid profile results in terms of assessment the ABP and in terms of commissioning additional tests, including e.g. hCG testing in women to confirm pregnancy associated with changes in the steroid profile.

[1] Mareck U, Geyer H, Opfermann G, Thevis M, Schänzer W: Factors influencing the steroid profile in doping control analysis. J Mass Spectrom 2008; 43:877–891.
[2] Kwiatkowska D, Kaplinski A, Urbaniak-Zbyszynska A, Jarek A, Tarka M, Chajewska K, Siek P, Pasik M, Szczepanska Z, Wojtowicz-Zawadka M: Pregnancy as a factor influencing the change of the steroid profile in terms of assessment of the athlete's biological passport. Thevis M., Geyer H., Mareck U. (Eds.). Recent Advances in Doping Analysis (30). Sportverlag Strauss, Köln 2022, 35–39.

PS3-08 Development of a multi-targeted UHPLC-MS/MS method for steroid profiling in biological samples

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Steroids are a crucial chemical group inside the human metabolome. When dysregulated, steroidogenesis and steroid metabolism can be responsible for a wide variety of pathologies [1,2]. An extended steroid profiling is thus required to diagnose many biomedical issues such as diabetes, prostate cancer or male infertility [2]. The concurrent monitoring of the highest possible number of steroids is of great interest for the evaluation of distinct pathways of steroid metabolism [3].
A current trend in biomedical analytics is the increasing use of untargeted analytical workflows for global metabolic profiling. The lack of absolute quantitative information, the difficulty of unequivocal ion annotation, the combination of multiple sample preparation procedures with multiple LC stationary phases or ionization methods and the necessity of HRMS instruments, not available in all laboratories, represent some issues of these untargeted approaches [2,4]. With the growing availability of analytical standards, the extension of multi-targeted analysis methods involving low-resolution mass spectrometers, more readily available in clinical environment, is a promising alternative for steroid profiling at very low concentration levels. Furthermore, this kind of method leaves room for fine tuning of chromatographic separation of critical isomers pairs to enable accurate absolute quantification of

target steroids. In this view, the present work has the objective of carefully optimizing various analytical parameters for a sensitive and selective single-run determination of hundreds of steroids. UHPLC separation of steroids was optimized with a Biphenyl stationary phase. Several columns with different dimensions (e.g., 100 mm vs. 150 mm length, 1.7 vs 2.7 µm particles) were compared to obtain the best possible compromise between method throughput, robustness, and resolution of steroids, with a particular focus on possibly co-eluting isomers.

MS detection of the analytes was optimized by using ammonium fluoride (NH4F) as post-column reagent, leading to an enhancement of electrospray ionization for numerous analytes, as previously reported [3]. MS/MS transitions were carefully selected for each steroid, and other parameters such as dwell times were also optimized to obtain good peak shapes and intense signals, and to circumvent the high dynamic range of steroid concentrations in biological samples, knowing that some are commonly detected at the low pg/mL range, while others can be abundant at hundreds of ng/mL. The developed UHPLC-MS/MS method has shown promise despite the challenges due to the number and diversity of target compounds which included phase I and phase II metabolites and was applied for steroid profiling of plasma and seminal fluid samples.

[1] E.Olesti, et al., J. Chromatogr. B 1136 (2020) 121929.
[2] F.Jeanneret, et al., J. Chromatogr. A 1430 (2016) 97–112.
[3] L.Schiffer, et al., J. Chromatogr. B 1209 (2022) 123413.
[4] D.J.Floros, et al., J. Chromatogr. A 1697 (2023) 463985.

PS3-09 Construction of a generalized interaction model for molecular pattern-recognition of pectic heteropolysaccharides by TLR4

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The intricate composition of pectin derived from terrestrial plant cell walls has captivated the attention of scientific inquiry as a promising reservoir of novel innate immune modulators. The ongoing discovery of numerous bioactive polysaccharides intricately linked with pectin, though an annual occurrence, is hampered in its full comprehension by the intricate heterogeneity and complexity inherent to pectin. This research focuses on a systematic exploration of the interplays governing pattern recognition, focusing on the common glycostructures encompassed within the intricate pectic heteropolysaccharides (HPSs) as they interface with Toll-like receptors (TLRs). Through a meticulous analysis of the glycosyl residues originating from pectic HPS, their compositional parallels are substantiated via rigorous systematic evaluations, thereby affording the basis for the subsequent construction of intricate molecular models of prototypical pectic segments. The outcome of the structural scrutiny herein conducted unveils a noteworthy forecast – the inner concavity of TLR4's leucine-rich repeats emerges as a plausible binding motif, mediating carbohydrate recognition. Sequential simulations prognosticate the precise binding modalities and conformations. Moreover, the orchestrated clustering of pectic HPS with TLR4 during endocytosis is demonstrated, inciting downstream signaling cascades that underlie the phenotypic activation of macrophages. Collectively, this study not only augments our comprehension of the pattern recognition nuances intrinsic to pectic HPS but also propounds a cogent paradigm for unraveling the intricate interplay between intricate carbohydrates and protein moieties.

Gyu Hwan Hyun, et al., Carbohydr. Polym. 314 (2023).
DOI: 10.1016/j.carbpol.2023.120921

PS3-10 Trace Determination of Silicones in Pharmaceutical Devices Using Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES)

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Silicones (poly(organo)siloxanes) are used in pharmaceutical applications as part of formulations, as well as during manufacturing and in packaging. The most commonly used substances are trimethylsilyloxy-terminated polydimethylsiloxanes (PDMS) which are used as active ingredients and antifoaming agents as well as excipients in numerous formulations and cosmetics. Furthermore, silicone oils are used as lubricants, reducing the break loose and gliding forces within medical devices like syringes and metered-dose inhalers. Due to increasingly stringent quality requirements, effective monitoring of the applied silicone quantities is critical for medical device development and quality control. A challenge for the development of suitable analytical methods is presented by the hydrophobicity of silicone oil and the often very small quantities applied as well as Si background levels from inorganic Si.

A suitable approach for the determination of silicone impurities is the analysis of total elemental silicon by inductively coupled plasma-optical emission spectrometry (ICP-OES). Without prior species separating steps, the method does neither differentiate between organic (e.g. silicones) and inorganic (e.g. SiO2) Si species nor between different organic Si species. However, extraction and determination of total organic silicon emitted by a medical device during the administration of a drug can be used as a worst-case scenario for possible PDMS contaminations.

In this study, a trace determination of organic silicone in medical devices by ICP-OES was developed. Due to the hydrophobic nature of silicone oils, quantitative extraction of the analytes is most feasible by using organic solvents like white spirit or methyl isobutyl ketone (MIBK). To allow for determination of trace amounts of silicone oil in medical devices, a low limit of quantification (LOQ) was a main target. To avoid additional dilution steps of the organic extracts, an organic solvent ICP-OES method was developed using oxygenated argon as auxiliary gas. Initially, the analytical parameters independent from the extraction technique like LOQ, linearity and stability of solutions were assessed.

Additional steps are the development and feasibility testing of the combined extraction and measurement method by performing spike recovery testing by applying known amounts of silicone oil on prefillable glass syringes since they are one of the most widespread siliconized devices for pharmaceutical applications.

B. Zeiss, TechnoPharm 6 (2016), 264.
F. Felsovalyi, et al., J. Pharm. Sci. 101 (2012), 4569.
https://doi.org/10.1002/jps.23328

PS3-11 Bio-based antimicrobial peptides for smart response self-disinfected surfaces

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We are focused on the development of innovative self-disinfectant nano-coatings designed with unique antimicrobial nanoparticles, incorporating bioactive elements such as Antimicrobial Peptides (AMPs) sourced from chicken feathers. At the current preliminary stage, we are actively engaged in the extraction, characterization, and optimization process of obtaining these AMPs. The extraction process of keratin employs enzymatic and alkaline hydrolysis methods, while the resulting peptides are being characterized using a spectrum of analytical techniques which will be further presented and discussed.

PS3-12 Residual enzyme limit test by UHPLC-MS

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Currently, Aspen Oss B.V. produces Testosterone using three chemical process steps. However, to create a more sustainable, efficient, and cheaper process, Aspen Oss B.V. developed a biocatalytic route towards Testosterone. The precursor 4-androstene-3,17-dione is converted into Testosterone by a single process that utilizes an enzyme. To guarantee an enzyme free Active Pharmaceutical Ingredient (API), the analytical department developed a method that can detect the residual enzyme up to a limit of 1 ppm in the final product.

The principle of this limit test is based on tryptic digested Testosterone samples that contain the enzyme and the use of isotopically labeled peptides as an internal standard. Trypsin always cleaves a protein at the carboxy or C-terminal side of the amino acids Lysine and Arginine, except when it is flanked by a Proline on the C-terminal side. Therefore, protein fragments resulting from a tryptic digestion can be predicted. Some characteristic peptides were selected to serve as a marker for the presence of the residual enzyme. The quantification of the selected peptides was performed by spiking the sample with the isotopically labeled equivalent of these peptides at 1 ppm level equivalent to the intact enzyme. By comparison of the peak areas of the natural and labeled peptides, the residual enzyme can successfully be quantified using UHPLC with a Q-Exactive Orbitrap High resolution mass spectrometer.

DOI: 10.33140/JPR

PS3-13 Systematic assessment of feature selection methods with PLS-DA model for photonic in vitro detection of lung cancer

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Lung tumors remain a primary cause of cancer-related mortality globally, emphasizing the critical need for early detection to improve survival rates [1]. This study is based on in vitro analysis of human blood plasma samples by Raman spectroscopy and provides a thorough, rigorous examination of feature selection methods for optimizing Partial Least Squares-Discriminant Analysis (PLS-DA) model for lung

cancer detection, resulting in improved classification performance and insights into the most significant features.

In our methodological framework, five distinct feature selection methods were applied to the spectral dataset obtained through Raman spectroscopy: Fisher score, minimum Redundancy maximum Relevance (mRMR), SHapley Additive exPlanations (SHAP), a synergistic combination of SHAP and mRMR (SHAP+mRMR), and statistical analysis utilizing Independent Samples T-Test and Mann-Whitney U test [2]. The performance of the PLS-DA model was examined for each feature selection method considering from 2 to 15 features and two latent variables. Model evaluation was carried out using KFold cross-validation with three splits, and performance metrics encompassed accuracy, F1 score, precision, and recall. All data analysis was performed using Spyder IDE, a scientific Python development environment (version 3.9.13).

Our comprehensive research demonstrates that the novel combination of SHAP+mRMR outperformes all other feature selection methods, producing improved classification metrics with lower standard deviation and requiring fewer features for optimal performance. This finding highlights the potential of combining complementary feature selection approaches to enhance the effectiveness of machine learning models, here for lung cancer detection.

Furthermore, we delved into the wavenumbers that played a pivotal role in differentiating the two groups, healthy and diseased. By conducting a meticulous statistical analysis on the selected wavenumbers and employing box whisker plots for visualization, we corroborated the statistical significance of the identified features, which can be assigned to specific chemical bonds, and hence, biomarkers.

In conclusion, this work provides a reliable, systematic approach to lung cancer detection using an improved PLS-DA model supported by a variety of feature selection methods. Our novel approach of combining SHAP with mRMR emerged as the preeminent method for feature selection, delivering superior classification performance and invaluable insights into the most influential wavenumbers. Moreover, our holistic approach considers the entirety of physiological parameters accessible by Raman spectroscopy. These highly promising findings can enable new precise and more efficient diagnostic tools for lung cancer, ultimately contributing to enhanced patient outcomes.

[1] Dela Cruz, C. S., et al., Clinics in Chest Medicine 32 (4), (2011) 605-44. DOI: 10.1016/j.ccm.2011.09.001
[2] Effrosynidis, D., et al., Ecological Informatics 61 (2021) 101224. DOI: 10.1016/j.ecoinf.2021.101224

PS3-15 Unified Approach to Univariate Analytical Calbration

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The presentation concerns a recently published my book on univariate calibration in analytical chemistry, which is titled “Calibration in analytical science. Methods and procedures” (Wiley VCH, Weinheim 2023). This is the first monograph to address these fundamental analytical issues in a comprehensive and systematic manner.

The theoretical part of the book is based on the definition of analytical calibration [1], which has been expanded and generalized with introduced concepts of three calibration functions: true, true and

model functions. The main focus is on the properties of various calibration methods that are or can be used in qualitative and quantitative analysis. Their specific features and analytical performance have been verified by mathematical calculations. Great is paid to the sources and role of various analytical effects and the possibilities for their elimination and compensation in the calibration process. It is shown how some calibration approaches can be used to detect, examine and compensate for uncontrolled analytical effects.

Analytical calibration is presented in the book in a unified form. This is evidenced by, among other things:

- every analytical method requires calibration (including gravimetric and volumetric methods), so terms such as ''analysis without calibration,'' ''standardless calibration'' or ''absolute analysis'' are not justified,
- analytical calibration requires the use of standards that are not only chemical, but also mathematical, biological or physical in nature,
- calibration methods used in qualitative analysis can be defined and classified in a similar way as in quantitative analysis.

All the theoretical considerations and the resulting conclusions are documented with numerous experimental examples, mainly from the field of forensic chemistry and flow analysis.

1. K. Danzer, et al., Pure Appl. Chem., 70 (1998) 993. DOI: org/10.1351/pac199870040993

PS3-16 A model for the identification of wood-derived mordant dyes in cultural heritage objects using mass spectrometry and chemometric tools

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Procedures based on modern analytical techniques are increasingly used to study works of art, allowing historians and art conservators to explore the essence of historic objects more deeply. The results provide an insight into the nature of the examined objects, and also enable the selection of appropriate methods of their conservation and restoration. An important area of interest is the identification of mordant dyes, which were the most numerous and the most commonly used group of natural dyes for coloring textiles over the centuries. Each of the dye contains from a few to a dozen or so coloring compounds, many of which have not yet been identified. Some of these preparations, such as wood-derived mordant dyes, although they come from different regions of the world, show a great similarity in chemical composition, which makes their identification much more difficult. In addition, given the historical value of the objects and small the amount of sample available for research, it is important to develop methods that allow obtaining a large amount of information with a minimum amount of material. The study of color matter in antique textiles is therefore a complex issue that requires appropriate separation technique combined with sensitive and selective detection techniques.

The aim of the study was to develop a model for distinguishing and identifying four wood-derived mordant dyes (peachwood, pernambuco, sappanwood, and logwood). The study was conducted using high-performance liquid chromatography combined with UV-Vis spectrophotometric detection and tandem mass spectrometry detection with electrospray ionization (HPLC-UV-Vis ESI-MS/MS) and analysis of the resulting data with chemometric tools.

Potential markers of peachwood, pernambuco, sappanwood, and logwood determined using MS/MS spectra were included in the ESI-MS/MS detection method for profiling of extracts from natural fibers dyed with these four mordant dyes. The registered chromatographic profiles revealed a significant difference between the extracts of logwood-dyed fibers and the others, which were highly similar to each other. Therefore, chemometric similarity analysis was necessary to distinguish between these three red dyes. The 2D-PCA graphs and heat maps generated for the selected conditions and method of normalization, enabled the difference of sappanwood (Asian tree) from pernambuco and peachwood (American trees). Finally, the developed model was used to identify wood dyes in samples from historical fabrics, which confirmed its usefulness.

PS3-17 Leveraging physics-informed machine learning to expand use of electronic tongues for environmental applications

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Ionophore-based sensors have a number of desirable characteristics for in-situ environmental measurements, such as the relatively low cost, low power demand, and small size compared to other types of electrochemical sensors. Yet the lack of perfect specificity presents an increasingly difficult challenge in environmental applications compared to, for instance, medical sensing needs, as the background concentrations of interfering ions are usually changing in complex and heterogeneous ways. Thus application of sensor arrays (electronic tongues) in place of single-transducer sensors is critical, however for environmental applications this is typical still insufficient to sense ions of interest, e.g., nutrients, which are often minority constituents of the total charge balance of the sample. To overcome these challenges, our team uses an approach wherein the selection of membrane composition, total number and variety of sensor elements, and data fusion algorithm are co-designed to achieve targeted detection accuracy and precision with, ideally, minimum sensor array size. This process is being validated on the design of printed arrays of sensors for pH and, eventually, nitrate sensing in estuarine and coastal waters and has the co-benefit of simultaneously quantifying many other ions contributing to charge balance of the sample. Specific aspects that will be discussed in this talk include: (1) variability in characteristics of individually printed sensors and effect of this measure on the design of the sensor array makeup (replicate copies of identical membrane elements), (2) correlation/difference in interferences experienced by different membrane compositions using different ionophores and effect of this measure on range of membrane types needed in the sensor array, and (3) data fusion approaches that leverage both knowledge of sensor physics and knowledge of physics of the target environment to optimize estimation accuracy for target parameter concentration.

PS3-18 Sensitive detection and electrochemical evaluation of the anticancer drug tofacitinib in pharmaceutical and biological samples using two different electrodes

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Tofacitinib citrate (TOF), 3-((3R,4R)-4-methyl-3-(methyl(7H-pyrrolo[2,3-d]pyrimidin-4-yl)amino)piperidine-1-yl)-3-oxopropanenitrile citrate, is a new oral Janus Kinase (JAK) selective inhibitor developed for the treatment of rheumatoid arthritis (RA). RA is a chronic autoimmune disease characterized by systemic, destructive, and progressive inflammatory polyarthritis in the immune system [1]. The sensitive, time-saving, and environmentally friendly voltammetric methods were developed, for the first time, for the quantification of TOF using glassy carbon (GCE) and boron-doped diamond electrodes (BDDE). The effects of pH, supporting electrolyte, and scanning rate on TOF's peak current and potentials were investigated in detail using both electrodes. Irreversible anodic peaks were obtained at pH 4.7 acetate buffer and pH 8.0 phosphate buffer media using cyclic voltammetry at 0.65 V and 0.55 V for GCE and 0.74 V and 0.66 V for BDDE, respectively. Furthermore, the electrochemical oxidation process of TOF was determined by cyclic voltammetry (CV). The results were found to be diffusion-controlled and irreversible at both electrodes. In addition, the mechanism of the oxidation process was investigated using model compounds. Under optimized conditions, low detection limits of 2.75×10-7 and 5.22×10-8 M were obtained for the electrochemical signal of the electrodes, GCE and BDDE, respectively. Also, the linear concentration ranges were found between 2.0×10-6 M and 1.0×10-4 M for GCE; and 1.0×10-6 M and 1.0×10-4 M for BDDE. These proposed methods were applied to determine the amount of TOF in pharmaceutical dosage forms and serum samples with excellent recovery results.

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PS3-20 ELECTRODE DESIGN AND ANALYSIS OF Cr DOPING INTO NASICON-STRUCTURED Na3V2(PO4)3 CATHODE WITH SELF-CARBON-COATING

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NASICON-structured Na3V2(PO4)3 (NVP) is one of the most promising cathode material for rechargeable sodium-ion batteries. NVP is characterized by a robust 3D structural framework and a high operating potential; these properties have enabled it to be widely studied as a stable and high-energy density cathode material for sodium-ion batteries (SIBs).[1-2] In the present study, we designed a Na3V1.6Cr0.4(PO4)3/C (NVCrP@C) cathode by implanting Cr into the crystal structure of NVP and simultaneously coating the surface of NVP with carbon for realizing high power density SIBs. NVP is fabricated using a low-cost pyro synthesis technique with the advantage of self-carbon-coating and nano sized particles are gained through this facile technique. To know the exact composition of vanadium and chromium in NVCrP@C, the ICP-AES analysis was performed and the results indicate that the stoichiometric composition of the prepared sample corresponds to Na3V1.6Cr0.4(PO4)3. The substitution of Cr with vanadium in the NVP structure significantly enhanced the structural stability of the electrode while the uniform and thin carbon layer improved the electrical conductivity. Interestingly, the NVCrP@C cathode showed high electrochemical activities with multiple V3+/4+/5+ redox reactions triggered by Cr3+ substitution in a wide voltage range (2.5–4.1 V). Consequently, the NVCrP@C cathode delivered excellent cycling stability over 500 cycles even at 15 C-rate and power capability up to 70 C-rate.

[1] S. Fang, et al., Adv. Energy Mater., 2019, 10, 1902485
DOI:10.1002/aenm.20190248
[2] D. H. Liu, et al, Adv. Mater., 2018, 30, 1706317
DOI:10.1002/adma.201706317

PS3-21 Combining Electroanalysis with Photocatalysis: Moving Beyond Remediation

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As climate change continues to be a global issue, many countries are transitioning towards a green and sustainable net-zero society. Consequently, there is an urgent need to develop new technologies to help in this advancement. To this end, photocatalysis continues to gain considerable attention. This is highly evident in the increasing number of publications in the literature, which span a range of applications [1].

A key consideration when deploying these applications is the analytical technique used to monitor the process. Whilst the literature is vastly dominated by spectroscopic and chromatographic approaches, there is a limited, but increasing number of reports on electroanalytical methods for monitoring photocatalytic processes. These reports mainly focus on monitoring the photocatalytic degradation of a specific environmental pollutant [2].

It has recently been shown that electroanalytical monitoring can progress beyond this scope. Herein, it is reported that electroanalysis for monitoring photocatalytic reactions is a rapid and accurate alternative, that can circumvent many problematic failings associated with the more commonly used monitoring methods.

A recent study by this group reported the in-situ electrochemical monitoring of the oxidation of 5-hydroxymethylfurfural towards 2,5-diformylfuran, which highlighted that a more accurate reaction profile was obtained relative to a standard HPLC method. This investigation provided a more detailed understanding of the 2,5-diformylfuran production process which could support the development of photocatalytic technology for biomass valorisation [3]

In another study, this group showed how electroanalysis can provide a rapid, sensitive, and accurate in-situ method for the monitoring of OH radical formation using coumarin as a probe. This investigation highlighted that issues associated with standard conventional spectroscopic analysis are circumvented by avoiding inner filter affects associated with probing the production of hydroxyl radicals via fluorescent spectroscopy.

These advances coupled with the well understood redox profiles of many desirable products and reactive oxygen species currently been produced via photocatalytic technologies, support the integration of electroanalysis into photocatalytic processes.

[1] F. Zhang, et al., Appl. Sci. 9 (2019) 24890
DOI:10.3390/app9122489
[2] A. Saravanan, et al., Environ. Chem. Lett. 19 (2021) 441
DOI:10.1007/s10311-020-01077-8
[3] P. McDonagh, et al., Electrochem.com. 142 (2022) 107365
DOI:10.1016/j.elecom.2022.107365

PS3-22 Biosensor development: Employing Self-Assembled Monolayers and Electrochemical Transducers

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Point-of-care (POC) analysis has attracted a lot of attention as they can be applied easily on-site, such as the iconic at home pregnancy tests commercially available at most pharmacies. The SARS-CoV-2 pandemic has highlighted how critical it is for rapid and accessible testing to help mitigate the spread of a contagious and potentially deadly virus. On the other hand, POC analysis is also needed for environmental applications, such as monitoring harmful contaminants in water sources. Electrochemical sensors have gained lots of interest partly due to the success of glucose sensors. However, adapting electrochemical sensors to biological detections has many challenges, which will require us to develop new sensing designs.

A biosensor contains three key parts, a biorecognition element (BRE), a sensor chip, and a linker molecule which connects the first two parts. While sensitivity of the detection is important, it is also essential to accommodate additional requirements such as selectivity and robustness of designs.
The work presented will focus on the design of a biosensors which utilize different types of gold sensor chips, from commercially available to home built using rapid 3D printing prototyping. Different linking molecules bound to toll-like receptor BREs are employed for broad spectrum whole cell pathogen detection. Additionally, sensor storage strategies are employed extending the sensor shelf life. The result demonstrates versatility in chip and linker molecule design for the successful detection of whole cell bacteria, with affordable prototyping methods and effective storage strategies.

J. McLeod, et al., Analyst, 2020, 145, 6024 - 6031
DOI: 10.1039/d0an01050b
I. Singh, et al., Chem. Commun., 2021, 57, 8421 - 8424
DOI:10.1039/d1cc03030b

PS3-23 Fabrication of cobalt oxide-supported carbon paste electrode for sensitive and selective Levofloxacin sensing

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Antibiotics are widely used in human medicine worldwide because of their effectiveness in treating bacterial infections. Fluoroquinolones are among the most important classes of synthetic antibacterial agents used in human and veterinary medicine since the 1980s. [1] Levofloxacin (LEV) is one of the most promising fluoroquinolones, and its antibacterial activity is much higher than that of other drugs in the fluoroquinolone family. Since LEV and similar antibiotics are widely used in agriculture and

aquaculture [2], a sensitive and discriminatory system for the discovery of LEV is essential for both human health and the environment.

In this study, a modified cobalt oxide (Co3O4) carbon paste electrode was prepared for the detection of LEV. Co3O4 nanoparticles were synthesized by the chemical coprecipitation method. The electrochemical properties of LEV at this electrode were investigated by cyclic voltammetry (CV), square wave voltammetry (SWV), and differential pulse voltammetry (DPV). In addition, electrochemical impedance spectroscopy (EIS), inductively coupled plasma–optical emission spectrometry (ICP-OES), transmission and scanning electron microscopy (TEM and SEM) and X-ray diffraction (XRD) were used to characterize the synthesized materials. The prepared electrode showed better electrocatalytic response than the bare carbon paste electrode. After square wave voltammetry (SWV) optimization, the electrode showed a wide linear working range from 1 to 100 µM at pH 5 of Britton–Robinson buffer solution (BRBS) as the supporting electrolyte. The excellent selectivity of the proposed method, with good repeatability and reproducibility, strongly suggests a potential application of the method for the determination of LEV in pharmaceuticals. The practicality with good recoveries indicates that the morphology of the materials is closely related to other parameters, which in turn suggests that the developed approach can provide a cost effective, rapid, selective, and sensitive method for LEV monitoring.

Rkik M, Brahim M Ben, Samet Y. Electrochemical determination of levofloxacin antibiotic in biological samples using boron doped diamond electrode. Journal of Electroanalytical Chemistry. 2017;794:175-181. doi:10.1016/j.jelechem.2017.04.015
2. Siporin 1949- C, Heifetz 1935- CL, Domagala 1951- JM, eds. The New Generation of Quinolones / Edited by Clifford Siporin, Carl L. Heifetz, John M. Domagala. M. Dekker; 1990.
ISBN:0824782240

PS3-24 Ultrasensitive fluoride detection in aquatic environments.

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Fluoride concentration in unpolluted waters typically ranges from 0.01 to 0.3 and 1.2 to 1.5 mg L-1 in fresh and sea waters respectively.[1] A concentration exceeding 0.5 mg L-1 confuses migratory fish trying to reach their reproductive site.[2] Thus, it is crucial to develop an ultrasensitive in situ fluoride detection method. Potentiometry with fluoride selective electrode (F- ISE) is a method of choice but suffers from limited sensitivity. Bobacka and co-workers proposed a novel readout method which (1) introduces a capacitive element at the working electrode (WE) and (2) maintains a constant potential between the WE and the reference electrode (RE).[3] However, flowing currents across an ion selective membrane induces polarization in the organic phase resulting in a potential drift. This can be overcome by switching the WE and the RE positions, treating the ISE as RE.[4]

In this work, a LaF3 single crystal-based F- ISE was used as RE and an Ag/AgCl reference element as WE for constant potential coulometry. A home-made electronic circuit was designed to allow a complete electronic control. This system was implemented in a flow cell to assess 0.01 (2 % concentration change) and 0.001 (0.2 % concentration change) logarithmic change in fluoride activity with outstanding reproducibility (concentration uncertainty of 0.08 % and 0.03% respectively). The same concentrations changes were assessed in two river samples and single point measurement with an in-lab calibration curve. The results were compared with ion chromatography analysis. The principle could be implemented in a field deployable probe for fluoride concentration profile.

(1) Camargo, J. A. (2003). Fluoride Toxicity to Aquatic Organisms: A Review. Chemosphere, 50 (3), 251–264. https://linkinghub.elsevier.com/retrieve/pii/S0045653502004988
(2) Damkaer, D. M.; Dey, D. B. (1989). Evidence for Fluoride Effects on Salmon Passage at John Day Dam, Columbia River, 1982–1986. North Am. J. Fish. Manag, 9 (2), 154–162. http://doi.wiley.com/10.1577/1548-8675(1989)009<0154:EFFEOS>2.3.CO;2
(3) Vanamo, U.; Hupa, E.; Yrjänä, V.; Bobacka, (2016). J. New Signal Readout Principle for Solid-Contact Ion-Selective Electrodes. Anal. Chem, 88 (8), 4369–4374. https://pubs.acs.org/doi/10.1021/acs.analchem.5b04800
(4) Kraikaew, P.; Soda, Y.; Nussbaum, R.; Jeanneret, S.; Bakker, E. (2023). Portable Instrument and Current Polarization Limitations of High Sensitivity Constant-Potential Capacitive Readout with Polymeric Ion-Selective Membranes. Sens. Actuators B Chem., 379, 133220. https://linkinghub.elsevier.com/retrieve/pii/S0925400522018639

PS3-25 MERCURY: FROM ATMOSPHERIC POLLUTION INTO BLOOD. ULTRASONIC MICROEXTRACTION AND DISPOSABLE SCREEN-PRINTED GOLD ELECTRODES FOR VOLTAMMETRIC MONITORING OF Hg IN BLOOD SAMPLES

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Mercury has been recognized as a neurotoxicant as well as immunotoxic and designated by the World Health Organization as one of the ten most dangerous chemicals to public health [1]. Directive 2004/107/EC establishes that the effects of mercury in particular through the food chain, and the environment as a whole, occur through ambient air concentrations and deposition [2]. Atmospheric pollutants such as Hg, can be accumulated in human tissues and organs, causing irreversible damages and the relationship between atmospheric exposure and blood Hg levels have also been reported [3]. This research aims to demonstrate the applicability of ultrasonic (US) microextraction and the use of gold screen printed electrodes for the voltammetric determination of mercury in blood samples, as a low-cost alternative method to other existing methodologies that require more complex digestion procedures followed by determination through expensive techniques such as ICP-MS. Therefore, a new method for US probe extraction of Hg in blood samples has been developed using a Box Behnken experimental design. In addition, a new method for the determination of Hg by anodic stripping voltammetry on a gold screen-printed electrode has also been developed. A suitable detection limit of 2 µg/L has been obtained for the determination of Hg in blood samples. The applicability of the combination of these two methodologies for the determination of Hg in blood samples has been demonstrated by analysing several samples of certified reference material, obtaining recoveries between 81-105 % and has also been employed to analyse real samples. As we have already demonstrated within our research group, the application of this voltammetric determination on gold printed electrode is applicable to determine atmospheric gaseous mercury [5], so our future purpose would be to find the relationship between these atmospheric Hg levels and those obtained in blood samples in field monitoring campaigns.

[1] K. Grace Pavithra et al., Chemosphere 312 (2023). DOI: 10.1016/j.chemosphere.2022.137314
[2] Directive 2004/107/EC of the European Parliament and of the Council of 15 December 2004 relating to arsenic, cadmium, mercury, nickel and polycyclic aromatic hydrocarbons in ambient air.
[3] Geir Bjørklunda et al., Environmental Research, 159 (2017). https://doi.org/10.1016/j.envres.2017.08.051
[4] Samuel Frutos-Puerto et al., Microchemical Journal 180 (2022). https://doi.org/10.1016/j.microc.2022.107642

PS3-26 Development of a Novel Molecularly Imprinted Polymer-Based Electrochemical Sensor for the Selective Determination of Ethyl Methylphosphonic Acid

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Organophosphorus-based nerve agents are highly toxic chemicals that are used as chemical weapons. Because of their use in wars and various attacks, a vast number of people are affected by nerve agents, accidentally or deliberately [1,2]. Nerve agents show their effect on humans very quickly, and they are excreted in the urine as various metabolites in 24-72 hours. Ethyl methylphosphonic acid (EMPA) is one of the metabolites of nerve agent VX. In 7 days, EMPA is decomposed to methyl phosphonic acid (MPA) like other nerve agent metabolites isopropyl methylphosphonic acid (IMPA) and pinacolyl methylphosphonic acid (PMPA) [1,3]. Analysis of biological samples of victims affected by nerve agents to determine the concentration levels of nerve agent metabolites has critical importance in terms of confirming the alleged use of nerve agents. The Organisation for the Prohibition of Chemical Weapons (OPCW) manages these analyses globally [4]. Currently, available methods for the sample preparation and determination of nerve agent metabolites are based on chromatographic methods coupled with mass spectrometry. However, these procedures are complex, expensive, and time-consuming. In search of a more advantageous analysis option, electrochemical sensors can be considered as easy-to-use, low-cost, sensitive, and portable analysis options. Despite these advantages, the greatest drawback of electrochemical sensors is the lack of selectivity. Integrating molecularly imprinted polymers (MIPs), specifically formed polymers to recognize the target analyte with specific cavities, with electrochemical sensors enables remarkable selectivity [5,6].

In this study, a MIP-based electrochemical sensor was developed using the functional monomer 4-aminobenzoic acid (4-ABA). The thermal polymerization (TP) technique was used in the presence of sodium dodecyl sulfate (SDS) as the pore-maker, NH3, and tetraethyl orthosilicate (TEOS) for the hydrolysis/condensation. Significant parameters related to the MIP process (monomer:template ratio, TP temperature, TP time, dropping volume, removal solution, removal time, and rebinding time) were optimized. Under the optimized conditions, the developed sensor was used to determine EMPA in standard solution and human urine samples. The developed sensor's full validation was performed in terms of linearity, the limit of detection (LOD), the limit of quantification (LOQ), accuracy, precision, sensitivity, selectivity, and stability. According to the obtained results, this novel sensor can be a great option for the routine analysis of nerve agents' metabolites.

[1] V. Van Son et al, Anal. Chim. Acta. 1069 (2019) 82–88.
DOI:10.1016/j.aca.2019.04.012.
[2] K. Bouchouareb et al,Talanta. 249 (2022) 1–10.
DOI: 10.1016/j.talanta.2022.123704.
[3] T. Baygildiev et al, J. Chromatogr. B Anal. Technol. Biomed. Life Sci. 1058 (2017) 32–39.
DOI:10.1016/j.jchromb.2017.05.005.
[4] T. Baygildiev et al, J. Chromatogr. B Anal. Technol. Biomed. Life Sci. 1132 (2019) 121815.
DOI: 10.1016/j.jchromb.2019.121815.
[5] S. Irem Kaya, et al, Electrochemical sensing of aclidinium bromide with metal ion-assisted molecularly imprinted films, Microchem. J. 184 (2022) 108128.
DOI: 10.1016/j.microc.2022.108128.
[6] F. Budak et al, J. Pharm. Biomed. Anal. Open. 1 (2023) 100005.
DOI: 10.1016/j.jpbao.2023.100005.

PS3-27 Self-referencing Pulstrode: Further Optimization and New Electrode Designs

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Point-of-Care Testings (POCTs) and wearable sensors have attracted a tremendous interest in the past decades. As opposed to traditional analysis, which are costly and time-consuming, POCTs and wearable sensors present, among others, the following advantages: they are cost-effective and allow rapid or continuous measurements, which lead to better reaction time and thus fewer costly complications [1]. Electrochemical sensors in that regards represent a good example of POCTs.

The reference electrode is an essential component of an electrochemical system, resulting in a high research activity in that domain [2]. The gold standard remains the Ag/AgCl double junction reference electrode. However, owing to its electrolyte-filled inner compartment its design is cumbersome and impractical for wearable sensors applications, which require miniaturization. From that point of view, all-solid state reference electrodes provide a promising alternative. Gao et al. proposed a solid-state reference electrode which relies on an Ag/AgI element and acts as a pulstrode to self-generate a reference potential [3]. The pulstrode protocol consists of four distinct steps: 1) potentiometric measurement of the initial state of the system (OCP), 2) a cathodic current pulse, leading to the reduction of Ag+ into Ag and the local release of a controlled amount of iodide, 3) measurement of the EMF (reference pulse) 4) application of the original OCP to regenerate the system into its initial state. The protocol has proven its reliability in terms of precision and stability over cycles on a macro-electrode.

In the interest of finding a reference electrode suitable for miniaturized systems, this work investigates the application of the pulstrode protocol on inkjet-printed electrodes provided by the Swiss Center for Electronics and Microtechnology. Additionally, in the context of manufacturing efficiency, the use of a fluid dispenser to form the AgI layer by chemical oxidation instead of electro-chemical oxidation is explored. Moreover, using the fluid dispenser, an agarose gel layer is deposited to cover the electrode surface area to improve the robustness of the described system against sample convection and sample density fluctuations.

[1] A. Lewenstam, Electroanalysis, 26 (6) (2014), 1171–81.
DOI: 10.1007/s00216-016-9945-6
[2] H. Jinbo, A. Stein and P. Bühlmann, TrAC Trends in Analytical Chemistry, 76 (2016), 102–14.
DOI: 10.1016/j.trac.2015.11.004
[3] W.Gao, E. Zdrachek, X. Xie, and E. Bakker, Angewandte Chemie International Edition 59, 59 (6) (2020), 2294–98.
DOI: 10.1002/anie.201912651

PS3-28 Sensitive simultaneous electrochemical determination of reduced and oxidized glutathione in urine sample using modified carbon paste electrode

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In this work, an electroanalytical sensor using an antimony trioxide modified-carbon paste electrode (Sb_2O_3–CPE) was proposed for the sensitive simultaneous determination of glutathione (GSH) and glutathione disulfide (GSSG), which are biomarkers commonly associated with oxidative stress. Based on the obtained results by scanning electron microscopy, CV and electrochemical impedance spectroscopy, it was realized that the modification of CPE with Sb2O3 enhanced the charge transfer at the electrode surface. Hence, compared to the bare CPE, the modified electrode showed a significant enhancement in the peak current response of GSH and GSSG with good separation of the peaks. On Sb_2O_3–CPE, GSH and GSSG exhibited well-defined oxidation peaks at +1.08 V and +1.36 V (vs. Ag/AgCl, 3 mol/L), respectively. Further, optimization of experimental and instrumental parameters such as the amount of modifier, type and pH value of supporting electrolyte, amplitude, and frequency for SWV determination was done. Under optimized conditions, the voltammetric responses were linear in the concentration range of 2–200 µmol/L for GSH and GSSG, with limits of detection of 0.34 µmol/L and 0.10 µmol/L for GSH and GSSG, respectively. The proposed sensor also showed good reproducibility (RSD=4.8%), stability (RSD=3.8%), and selectivity. Finally, using a modified carbon electrode, it was possible to simultaneously electrochemically distinguish between oxidized and reduced glutathione. Results revealed that this method provides a way to analyze the levels of both forms of glutathione, which are important markers of oxidative stress, and can help in understanding the underlying mechanisms of oxidative stress in biological systems.

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PS3-29 Electrochemical determination of phenolic antioxidant BHT in cosmetic and food samples

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Butylated hydroxytoluene (BHT) is a commonly employed antioxidant agent to inhibit the process of oxidative rancidity in various commercial products, including food and pharmaceutical items. Several research studies have indicated that synthetic constituents have the potential to infiltrate the human body via the consumption of food items, pharmaceutical products, and other means. The potential adverse impact of these additives on human health requests consideration. Artificial phenolic antioxidants have been discovered to potentially induce nutrient loss and toxic effects. Electrochemical techniques, particularly voltametric methods, present a viable substitute for chromatographic approaches owing to their cost-effectiveness (facile sample preparation, brief analysis duration), akin selectivity, sensitivity, and detection thresholds. Two sensors utilizing graphene powder modified with iron (III) oxide and europium (II) oxide were proposed for the purpose of detecting butylated hydroxytoluene (BHT) in food and cosmetic samples. The new sensors were evaluated and characterized using differential pulse voltammetry. Subsequently, they were subjected to testing and validation procedures to determine their efficacy in detecting butylated hydroxytoluene (BHT) in samples of food and cosmetics. The two sensors exhibited considerable stability, selectivity, sensitivity, and reproducibility in their respective measurements. The scientific community has shown interest in

rare earth elements as potential components for nanocomposites utilized in electrode modification. These nanocomposites are expected to perform similarly to those composed of noble metals. The element Europium has been considered significant within the group of lanthanides due to its capacity for redox reactions, which is attributed to its ability to exist in multiple oxidation states [1]. Various techniques have been employed to synthesize transition metal oxide nanoparticles, including iron (III) oxide, with diverse shapes and structures. The favorable electrical and photocatalytic characteristics of these metal oxide nanoparticles can be attributed to their dimensions, morphology, robustness, and increased interfacial area.

[1] P. Mohammadzadeh Jahani, et al., Measurement 173 (2021) 108616.

PS3-30 Antimony remediation using a new magnetic system in potable aqueous samples.

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Heavy metals are serious pollutants because of their toxicity, persistence, and non-degradability in the environment. Sb is easily accumulated in organism and will cause deleterious effects on human beings when the content goes beyond the allowable limit. As a cumulative toxic element, antimony has chemical and toxicological properties.

Currently, Drinking Water Treatment Plants (DWTP) are incapable to eliminate totally the Sb concentration in natural waters, and due to its toxicity is needed. In this work, the adsorption process of a recently patented magnetic material (M@GOPS) towards Sb has been studied.

Magnetic solid phase extraction (MSPE) with magnetic particles (MPs) as the adsorbents has aroused great interest in analytical community in recent years. Superparamagnetic iron oxide particles as sorbents in SPE have received increasing attention because they are attracted to a magnetic field but do not retain any magnetism after the field is removed. Thus, a chelating sorbent which employs magnetic nanoparticles (MNPs) and graphene oxide functionalized with [1,5-bis (2-pyridyl) 3-sulfophenylmethylene] thiocarbonohydrazide M@GO-PS was used to adsorb trace amounts of metal ions of Sb (III) in natural waters sources of drinking water for citizens. Also, the material presents magnetic properties, biocompatibility and low toxicity. The Sb determination in the treated water was aborded by Graphite Furnace Atomic Absorption Spectrometry (GFAAS).

The kinetics of the process have been studied, showing a good approximation to the Langmuir's theoretical model. The magnetic adsorption procedure has shown a performance of 50% in 60 min for the elimination of Sb, with a dosage of 1 g/L of M@GO-PSTH in a potable water with an initial concentration of 0.001 g/mL of Sb.

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PS3-31 In situ seasonal monitoring of the potentially bioavailable Nickel dissolved fraction in Lake Geneva

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Assessing the impact of trace metals (TM) on aquatic ecosystems and ultimately human health is challenging. Trace metals are distributed in a variety of redox states and chemical species (speciation) which proportion may vary continuously in space and time [1,2]. Only some TM species are bioavailable. The development of robust and adaptive submersible sensitive TM bioavailability-assessment tools is therefore required to support the establishment of environmental quality standards (EQS) and guidelines (EQG) based on realistic metal risk assessment to protect aquatic life and biodiversity, and ultimately human health. Innovative antifouling gel-integrated microelectrode arrays (GIMEs) and submersible sensing probes (VIP, TracMetal) were developed towards this aim. They enable in situ, high-resolution, autonomous measurements of the dynamic (potentially bioavailable) fraction of a range of trace metals at sub-nanomolar to low picomolar levels [2–4].

We report here on the development of an analytical protocol enabling to apply these devices to the direct quantification of Ni(II) in aquatic systems [4]. A GIME-VIP probe using the developed protocol was successfully applied to monitor the hourly variation of the potentially bioavailable Ni(II) fraction in Lake Geneva over 3 seasons. In parallel, master variables were monitored in situ, and water samples were collected for complementary analyses of total dissolved metals concentrations in the operationally defined dissolved <0.2 µm and <0.02 µm fractions, water composition and proxies for primary production. The combination of the provided data allowed for the very first time to assess (i) the temporal concentration of the potentially available Ni(II) fraction and (ii) the biotic and/or abiotic processes that controlled the seasonal Ni speciation.

1. Tercier-Waeber, M.-L.; Stoll, S.; Slaveykova, V. Trace metal behavior in surface waters: emphasis on dynamic speciation, sorption processes and bioavailability. Archives des Sciences 2012, 65, 119–142.
2. Tercier-Waeber, M.-L.; Confalonieri, F.; Abdou, M.; Dutruch, L.; Bossy, C.; Fighera, M.; Bakker, E.; Graziottin, F.; van der Wal, P.; Schäfer, J. Advanced Multichannel Submersible Probe for Autonomous High-Resolution in Situ Monitoring of the Cycling of the Potentially Bioavailable Fraction of a Range of Trace Metals. Chemosphere 2021, 282, 131014, doi:10.1016/j.chemosphere.2021.131014.
3. Creffield, S.; Tercier-Waeber, M.-L.; Gressard, T.; Bakker, E.; Layglon, N. On-Chip Antifouling Gel-Integrated Microelectrode Arrays for In Situ High-Resolution Quantification of the Nickel Fraction Available for Bio-Uptake in Natural Waters. Molecules 2023, 28, 1346, doi:10.3390/molecules28031346.
4. Layglon, N.; Creffield, S.; Bakker, E.; Tercier-Waeber, M.-L. On-Field High-Resolution Quantification of the Cobalt Fraction Available for Bio-Uptake in Natural Waters Using Antifouling Gel-Integrated Microelectrode Arrays. Marine Pollution Bulletin 2023, 189, 114807, doi:10.1016/j.marpolbul.2023.114807.

PS3-32 Photodegradation of Textile Pollutants in Wastewater by Nanocomposite Membranes

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Textile effluents have a significant environmental threat due to toxic nature and visibility of the dyes even at very less concentration. Membrane technology is a highly desirable filtration process but most

of polymeric membranes exhibit surface fouling due to their hydrophobic nature, so these membranes cannot be used in long term applications. In this research, the PVDF (polyvinylidene fluoride) – PANI (polyaniline) –titanium nanotube (TNT) based nanocomposite membranes were synthesised through phase inversion method. The composition and structural properties of nanocomposite membranes were characterized by X-ray photoelectron spectroscopy (XPS), Fourier-transform infrared spectroscopy (FTIR), X-ray diffraction (XRD), atomic force microscopy (AFM) and scanning electron microscope (SEM). The significant properties of synthesized membranes such as distribution of pore size, thermal properties, mechanical properties, and photocatalytic behaviour of membranes were also studied. The hydrophilic properties of the composite membranes increased with filler content (PANI-TNT) and results in improved pure water flux (484.8 ± 2.9 L/m2h−1) compared to that (312.0 ± 1.91 L/m2h−1) of the pure PVDF membrane. The pure PVDF and nanocomposite membrane were further analysed in terms of their filtration properties such as adsorption of dyes (methyl orange, Allura red) and UV self-cleaning properties. The newly developed nanocomposite membranes showed excellent pollutant removal efficiency (~90%). The synthesized nanocomposite membranes also showed photocatalytic activities due to the presence of TNTs, and adsorption of methyl orange (MO) reduces significantly with the UV light irradiations. The UV self-cleaning property of the composite membrane was further confirmed due to their high flux recovery ratio of about 94%. The results show that embedded PANI-TNT within nanocomposite was photo-catalytically active and degraded the dye molecules from the surface of the nanocomposite membrane.

Nawaz, Hifza, et al. "Photodegradation of textile pollutants by nanocomposite membranes of polyvinylidene fluoride integrated with polyaniline–titanium dioxide nanotubes." Chemical Engineering Journal 419 (2021): 129542. https://doi.org/10.1016/j.cej.2021.129542

PS3-33 Pollution assessment and source apportionment of Persistent Organic Pollutants in soil of Rural Roma Communities in Transylvania

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This work aims to assess the pollution and source apportionment of three classes of persistent organic pollutants (POPs), namely organochlorine pesticides (OCPs), polychlorinated biphenyls (PCBs), and polycyclic aromatic hydrocarbons (PAHs) in 22 soil samples collected from 13 rural Roma communities in Transylvania, Romania. A total of 20 OCPs, 12 PCBs and 16 PAHs were analysed using gas chromatography coupled with an electron capture detector for halogenated compounds and mass spectrometry for PAHs. POPs were isolated from soil matrix by ultrasound-assisted extraction followed by purification of the extract by open column chromatography.

Ratio of low and high molecular weight PAHs, (∑LMW/∑HMW), Anthracene/(Anthracene + Phenanthrene), ANT/(ANT + PHE), Fluoranthene/(Fluoranthene + Pyrene), FLT/(FLT + PYR), and ΣDDTs/ΣHCHs, α-HCH/γ-HCH and, (p,p`-DDE + p,p`-DDD)/ΣDDTs were used as a tool for identification of the pollution sources. Soil quality standard guides (ERL and ERM) and toxic equivalent factors of PAHs were used to assess the ecotoxicological risk of the soil contamination.

The values of ∑LMW/∑HMW under 0.1, ANT/(ANT + PHE) over 0.1 and FLT/(FLT) + PYR) over 0.5 suggest a pyrogenic nature of PAHs such as biomass, coals, and petroleum combustion. The ΣDDT/ΣHCHs ratio

show that the DDT was especially used in the north-west of Transylvania (value over 0.5) while HCH isomers prevails in the central and eastern part (values under 0.5). The ratio of α-HCH/γ-HCH is over 1 in all analysed samples which suggest that technical HCHs was used instead of pure lindane. The ration of (DDE + DDD)/ΣDDT have a median value of 0.50 which suggest that the sources of DDT are mainly from historical use. The PCBs of 3 to 4 and 6 to 8 chlorine atoms prevail in the analysed soil samples suggesting their use in transformers, capacitors, lubricants, paints, and plasticizers.

The PAHs concentrations (individual and total) were lower than ERL, indicating that the ecological risk of PAHs is considerably low. For ΣPCBs, in 12 out of 22 samples, the concentrations were between ERL and ERM, suggesting a potential ecological risk. Highest potential ecological risk was observed for p,p'-DDD in 4 out of 22 samples and for p,p'-DDT 19 out of 22 samples. For ΣDDT, in 10 out of 22 samples, the concentrations were between ERL and ERM which indicates that adverse effects would occasionally be observed.

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PS3-34 Direct measurement of organic micropollutants in natural water and wastewater using fluorescence spectroscopy

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Special concerns have been raised about the presence of organic micropollutants (OMPs) in aquatic environments due to their known or potential adverse ecological and/or human health effects[1-2]. In this context, many environmental agencies including the Swedish Medical Products Agency, recommend monitoring the occurrence of many OMPs in a range of environmental compartments, including water, sludge, and biota. OMPs of emerging concern include antibiotics, non-steroidal anti-inflammatories drugs and sedative hypnotics[3] and usually enter the environment via wastewater treatment plants (WWTPs). Generally, to quantify OMPs in aquatic samples and assess their removal efficiency in wastewater treatment plants requires expensive and time-consuming analytical methodologies[4]. Therefore, this study evaluated the potential of using inexpensive fluorescence spectroscopy combined with a priori models developed with parallel factor analysis (PARAFAC) to directly quantify fluorescent OMPs following limited sample pre-treatment (filtration and pH adjustment). Three OMPs (ciprofloxacin, naproxen and zolpidem) were studied in samples from a wide range of environments including natural waterbodies (river, lake, pond, stream), drinking water and wastewater. These OMPs were recovered from natural water and wastewater samples at 80-120% of their spiked concentration, with limits of detection vs quantification 1.0-3.3 µg/L vs 2.9-9.3 µg/L for natural water samples, and 0.6-8.9 µg/L vs 1.5-22.9 µg/L for wastewater, respectively. This study demonstrated a simple approach to quantify OMPs directly via fluorescence despite interfering natural organic matter fluorescence of varying concentration and complexity. Prospective applications of this methodology include its use in lab-scale studies of OMPs fate and removal.

[1] *https://doi.org/10.1016/j.jece.2023.109613*
[2] *https://doi.org/10.1016/j.chemosphere.2022.134631*
[3] *https://doi.org/10.1016/j.envsci.2020.06.011*
[4] *https://doi.org/10.1016/j.watres.2020.116749*

PS3-35 Analysing sorption Kd of fluoroquinolone antibiotics in soils and soil components

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The presence of fluoroquinolone (FQ) antibiotics in the environment is of concern due to their disrupting effects on bacterial communities. The evaluation of FQ sorption in soils and pure soil components, by obtaining solid-liquid distribution coefficients (Kd), is valuable information to address their (bio)availability and mobility in the environment. However, sorption processes are complex due to the ability of FQ to bind to different soil phases, which depend on multiple factors such as pH, FQ concentration and speciation, and soil solution composition, contributing to a large variability of the Kd value. Therefore, quantifying the Kd values of different FQs in multiple soils and pure soil components and varying experimental conditions is of interest to elucidate the impact of each soil and FQ factor on the overall Kd and derive Kd values more representative of specific scenarios and with lower variability.

In this study, we compiled sorption Kd (FQ) data for humic substances, metal oxides, phyllosilicates and soils, as well as ancillary information for the corresponding systems. Compilations included literature data and own experimental data. Batch sorption experiments were carried out following OECD guidelines, with specific changes in the experimental setup such as pH and Ca concentration in solution, to identify the main factors affecting sorption. FQ concentration in the respective solutions were quantified by HPLC with fluorescence detection.

Sorption data revealed a bell-shaped sorption trend with varying pH in all phases analysed, being the factor responsible for the highest Kd (FQ) variability, confirming the key role of FQ speciation in sorption. Analysis of Kd (FQ) values through cumulative distribution functions (CDF) showed an increasing sorption affinity for FQs in the sequence: humic substances > phyllosilicate minerals > metal oxides. Besides the respective absolute sorption affinity, the abundance of each phase in soil also determines its final sorption contribution. Thus, higher sorption was expected for organic soils. This was confirmed by the analysis of Kd (FQ) data in bulk soils, where pH was identified as the main parameter affecting FQ sorption, followed by organic carbon and clay minerals. Prediction of Kd (FQ) values was assessed with partial least square (PLS) regression and CDF based on relevant soil physicochemical properties. Kd (FQ) values in soils were above 1,000 L/kg under most environmental conditions, suggesting strong sorption onto soil particles and low predicted mobility. However, higher environmental mobility might be expected in scenarios with alkaline pH, low OC and high sand contents.

PS3-36 DIRECT MERCURY SPECIATION IN SOLID SAMPLES USING THERMAL RELEASE COUPLED TO ELECTROTHERMAL ATOMIC ABSORPTION SPECTROMETRY

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Mercury is an element involved in global processes occurring in Biosphere. The distribution of mercury in various natural environments depends on its chemical form. For this reason, the development of methodology and new methods for mercury speciation in natural and man-made media is of particular value. Wherein, the solid samples are the most problematic in terms of determining the chemical forms. Traditionally used conventional analytical procedures usually include extraction of the species from the matrix followed by their subsequent separation and element-selective detection, at that the most critical stage - extraction of the analytes - may be accompanied by their transformation. Moreover, the use of extraction does not allow the isolation of water-insoluble species, for example, mercury sulfide. Therefore, the most promising methodology seems to be the direct analysis with application of the thermal release as separation stage in combination with electrothermal atomic absorption detection (TR-ETA-AAS). Nevertheless, a number of issues remain that hinder its practical application as a reliable analytical technique.

The thermal behavior of the most common mercury specie in order to eliminate the gaps of this approach were studied in present work on the example of pure substances as a starting point for a subsequent transition to the real objects . Studies were carried out using a commercial installation RA-915+ (Lumex, Russia) has been modified by adding of a module providing programmable heating of the sample which allows a continuous increase in temperature with a subsequent stop until the thermal peak is formed. It was shown on the example of the mixture of methylmercury and mercury chlorides, mercury sulfide and mercury sulfate that the transformation of the studied compounds happens under thermal exposure, which leads to a change in their physicochemical characteristics. But, nevertheless, it does not prevent their baseline separation [1]. This methodology has been applied in analysis of real natural and technogenic samples as well as standard reference materials which were diluted by inert material, Al2O3, to eliminate matrix effects. As a result the absolute detection limits at the level of 2-5 ng (as mercury) were achieved.

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PS3-38 EXTREMADURAN CHARCOAL: QUALITY AND POSSIBILITIES AS A BIOCHAR

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1. Introduction:
Biochar is a product usually generated for the pyrolysis of organic matter rich in carbon. Biochar can be used in agriculture as a way to improve soil health and sequester carbon. Currently, other applications range from animal feed to building. However, it is necessary an exhaustive control of quality in order to avoid possible environmental affections. On the other hand, charcoal is a carbon obtained from wood or other organic matter by heating in absence of air. The production of charcoal in Extremadura (Spain) is based in traditional processes by mainly using wood from holm oak. The result is a high-quality fuel for heating or barbecues. This work studied the possibilities of this product as a possible source of biochar by an exhaustive quality analysis in line with compliance with international standards.

2. Experimental:
For this purpose, six samples collected in different areas of Extremadura were analysed according to the main quality parameters set by some of the most recognized standards to assess the quality of biochar: the European Biochar Certificate (EBC) and the International Biochar Initiative (IBI). A wide range of tests: elemental analysis, physical parameters, nutrients, heavy metals and organic pollutants (PAHs, PCBs) were carried out and the results compared to the guidelines of ECB and IBI.

3. Results:
In general terms, the results showed a high quality for the agricultural uses (EBC-Agro and EBC-AgroOrganic), urban environment (EBC-urban) and materials (EBC-Consumer Materials and EBC-Basic Materials). With reference to IBI standards, the average parameters fulfilled requirements for all categories (A, B and C). Regarding animal feed (EBC-FeedPlus and EBC-Feed), the analysis of PCBs were carried out taking into account the expected use for agronomic practices so it cannot therefore be concluded the suitability for this purpose.

4. Conclusions:
The charcoal from Extremadura has excellent quality criteria in terms of EBC and IBI parameters and could be used as a type of biochar: addition to the soil, composting, building, but a possible use for animal feed must be checked in more detail. Besides, other parameters such as economic ones limit the options and it would be necessary to study these new uses compared to the current ones (fuel).

PS3-39 Ion-Selective Electrodes as Companion Diagnostics for Personalized Treatment of Mental Health Disorders

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Mental health diseases account for 32% of years lived with disability and pose an estimated 2.5 trillion economic burden to the US. About 5% of US population will be impacted by the bipolar disorder. Bipolar disorder is associated with major mood swings with deep depressed states and emotional highs. Lithium therapy (Li+) is currently the primary and the most reliable treatment for bipolar disease and mood disorders. Lithium therapy has been reliable and effective, but it comes with a big challenge of a small therapeutic window of lithium, and acute toxicity of lithium. The effective and safe serum Li+ levels are 0.4 to 1.0 mM. Higher dosages (≥1.2 mM) pose a risk of acute toxicity. There is a high rate (15%–35%) of acute kidney damage in bipolar patients that receive lithium therapy. Current paradigm of lithium testing requires the patient to visit the hospital, submit blood samples, and wait for sample analysis in the pathology lab. A companion diagnostic device capable of real-time monitoring of lithium levels in biofluids of the patient will revolutionize lithium therapy and enable a personalized dosage of this life-saving drug. This device enables frequent monitoring of Li+ in biofluids to detect toxic dosages, and substantially lower the acute toxicity and organ damage caused by Li+. This work shows a low-cost and compact potentiometric sensor bundle that can selectively measure Li+ in different biofluids (saliva, serum, sweat, urine) at the convenience of home, and inform the patient on a toxic or effective lithium dosage. Technical challenges for addressing selectivity issues, and sample volume will be discussed.

PS3-40 Point-of-care testing of LDL cholesterol using molecularly imprinted polymers

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Cholesterol is one of the main constituents of cell membranes and is the starting molecule for the biosynthesis of hormones and vitamin D. In the body, cholesterol is transported within micellar assemblies made up of proteins, phospholipids and triglycerides: the lipoproteins. Among these, Low Density Lipoproteins (LDL) are colloquially called “bad cholesterol” because they distribute cholesterol from the liver to the body and tend to accumulate on blood vessels’ interior walls causing arteriosclerosis.

The determination of LDL cholesterol (LDL-C) levels in blood is therefore a key parameter to prevent the risk of developing cardiovascular diseases. Nowadays, LDL-C is quantified indirectly using the Friedewald equation by assessing, using enzymatic/colorimetric tests, the quantity of total cholesterol and subtracting the amount of non-LDL lipoproteins. The analytical performance of this method is limited as the uncertainties of the individual measurements accumulate. The direct and specific determination of LDL-C using a dedicated assay would be a more accurate strategy. To develop such a specific assay, Molecularly Imprinted Polymers (MIPs) are an interesting class of polymer-based molecular recognition reagents engineered to bind to one single target compound. Selectivity is introduced during MIP synthesis thanks to a template molecule that guides the formation of specific cavities or imprints that are sterically and chemically complementary to the target analyte.

We will present the results of a project aiming at the development of a novel point-of-care test (POCT) of LDL-C using MIP. POCT are medical diagnostic tests that can be quickly performed at the patient’s bedside. We will describe the challenges and pitfalls encountered when using MIP with lipoproteins and the analytical performance that can be achieved.

PS3-41 Smart Portable Device Based on the Utilization of a 2D Disposable Paper Stochastic Sensor for Fast Ultrasensitive Screening of Food Samples

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Since the determination of the high toxicity of bisphenol A, alternative structures for bisphenols were synthesized, resulting in bisphenols C, E, F, S, Z. These bisphenols replaced bisphenol A in plastic bottles, toys, cans – used for preserving food. Later, the toxicity and negative effects of all these bisphenols on people’s health were proven. Therefore, there is a need for a fast ultrasensitive screening method able to detect the presence of these bisphenols in any condition directly from food samples. This paper presented a disposable device based on the utilization of a 2D disposable paper stochastic sensor for fast ultrasensitive screening of food samples for bisphenols A, C, E, F, S, and Z. The 2D disposable sensor was obtained by the deposition of graphene and silver nanolayers on paper using cold plasma. Furthermore, the active side of the sensor was modified using 2,3,7,8,12,13,17,18-octaethyl-21H,28H Mn porphyrin. The limits of quantification of these bisphenols were of 1 fmol/L for

the bisphenols C and E, 10 fmol/L for the bisphenols A and F, 10 pmol/L for bisphenol S, and 1 pmol/L for bisphenol Z. The recoveries of these bisphenols in milk, canned fruits, vegetables and fish were higher than 99.00% with RSD (%) values lower than 1.50%.

PS3-42 IDENTIFICATION OF SELECTED NONSTEROIDAL ANTI-INFLAMMATORY DRUGS IN HORSES' URINE AND BLOOD – GC/MS/MS METHODS VALIDATION.

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Horses occupy a special place in the discussion on the administration of drugs due to the fact that they may be treated as companion animals as well as animals whose tissues or products may be intended for human consumption, and also as sport horses that in addition comply with the anti-doping regulations of the International Federation of Equestrian (FEI – Fédération Équestre Internationale). FEI presents a list of prohibited substances that include androgenic-anabolic steroids, stimulants, beta-agonist, etc. and a wide range of nonsteroidal anti-inflammatory drugs (NSAIDs).

In human medicine, there are hundred preparations available, mainly for analgesic and antirheumatic purposes. Athletes often seek artificial ways to gain an advantage over other competitors as well as to prolong their participation in competition, and unlike horse races events that kind of practice is allowed in athletic events.

FEI regulation states that the use of NSAIDs in horses has to be strictly controlled and any amount of nonsteroidal anti-inflammatory drugs present in blood or urine samples obtained from horses during anti-doping controls shall have positive results for anti-doping tests. In order to meet the requirements of the FEI, a method for the detection and identification of NSAIDs, in both blood and urine samples, has been developed. For this purpose, a gas chromatograph system, coupled with a triple mass spectrometer (GC/MS/MS), has been applied. The aim of this study was to validate the proposed methods and applied them to routine analysis. The result for 40 analyzed substances showed very good limit of detection (lower than 1 ng/mL) compared to the already existing method conducted on a gas chromatograph system coupled with a single mass spectrometer (25 ng/mL). Also, the number of positive cases increased significantly after applying the new methods to the routine analysis.

PS3-43 A chemometric approach to discrimination of isobaric β- and γ-isoforms of tocopherol and tocotrienol using RPLC-ESI-MS

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Tocopherols and tocotrienols, collectively called "tocols" or "vitamin E", are essential oil-soluble secondary plant metabolites. Their molecules consist of polar chromanol and non-polar phytyl moieties. The two groups differ in the unsaturation degree: tocotrienols (T3) possess 3 unsaturated bonds in their phytyl tail, while tocopherols (T) have all C-C bonds saturated in this part. Both

tocopherols and tocotrienols exist in 4 isoforms differing in substituting the aromatic ring in the chromanol part. Tocols play several biological roles, the most prominent being free radical scavenging. As components of edible oils, they provide strong antioxidant activity, crucial for maintaining the oil quality during storage. Even more importantly, vitamin E delivered with an oil-based diet or dietary supplements is necessary to protect human cells from oxidative stress. The concentration and antioxidant activity of individual tocols varies depending on the plant and environmental conditions, with α-tocopherol and tocotrienols being usually the major and the most active component [1]. For this reason, the knowledge of the concentration of each isoform of all tocols is crucial in monitoring the quality control of lipid-based food products.

In the present contribution, we describe a new approach to determine all tocols (α-, β-, γ- and δ-tocopherols and tocotrienols) content using a relatively straightforward method based on reversed-phase liquid chromatography coupled with electrospray mass spectrometry (RPLC-ESI-MS). The well-known problem of the inability to resolve the β- and γ- isomers using RPLC was circumvented by applying a chemometric procedure based on a transmission signal ratio, which correlated with their amounts in the whole quantitation range (0.4 – 120 ng/mL). Such a low LOQ was achieved by improving the recovery of tocols from a C4 stationary phase and their MS ionization using a pH 7.5 mobile phase. The new method was validated using dietary supplements and successfully applied to follow the aging of cold-pressed false flax oil and to determine tocols in several vitamin E-abundant plant oils.

H. Y., Peh, at all., Pharmacology & Therapeutics 162 (2016) 152, DOI:10.1016/J.PHARMTHERA.2015.12.003

PS3-44 New method to increase the efficiency of DNA extraction using dielectrophoresis

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Nucleic acid extraction is crucial for applications spanning from liquid biopsies to climate-resistant crop development. Dielectrophoresis has been described as possible alternative to the more laborious and toxic phenol-chloroform protocol currently employed, but so far, has been limited to analytical applications [1], not being used as preparative technique due to poor yield, particularly for small fragments. The dielectrophoretic attraction is counteracted by the electrostatic and osmotic repulsion due to negative charges in the nucleic acids' backbones. This prevents the high localized concentration as well as the aggregation required to capture smaller fragments.

We tried to circumvent this challenge using principles from solid-phase reversible immobilization, namely: a) inducing DNA condensation using crowding agents, what will result in a configuration with less negative charges exposed and b) shielding the remaining charges using mono or di-valent cations [2].

We used a set of interdigitated gold electrodes, fabricate using conventional CMOS technology. Each electrode was 2 µm wide with a spacing of 2 µm. These dimensions were optimized to maximum capture of DNA.

We started by investigating the role of monovalent cations by assessing the capture of lambda-DNA in buffer with increased concentration of Na+. The increased concentration of sodium resulted in higher conductivity. However, at the frequency used (10 MHz), this has no impact in the real part of Clausius-

Mossotti factor, hence on the dielectrophoretic force. The capture of lambda-DNA and retention after the electric field was turned off (measured by fluorescence) increased proportionally with the concentration of sodium for an interval between 1.4 mM and 1400 mM.

Next, we investigated the addition of IgG protein (100 mg/L) to the buffer with highest sodium content. At physiological pH, IgG is positively charged and has a long molecular chain, able to induce crowding effect [3]. It presents only minimal negative dielectrophoretic response at the employed frequency. We observed that, 15 s after the field is applied, the capture of lambda-DNA reached a maximum in the control solution (not containing IgG), decreasing after that. In solutions containing the protein, the retention remained constant at the maximum value until the field was turned off. Additionally, DNA concentration after the field was turned off remained 2x higher in presence of IgG than in the control solution.

Those initial experiments show a clear possibility of improving the performance of dielectrophoretic capture of DNA through manipulation of the sample composition.

(1) Sonnenberg, A.; Marciniak, J. Y.; Skowronski, E. A.; Manouchehri, S.; Rassenti, L.; Ghia, E. M.; Widhopf II, G. F.; Kipps, T. J.; Heller, M. J. Dielectrophoretic Isolation and Detection of Cancer-Related Circulating Cell-Free DNA Biomarkers from Blood and Plasma. ELECTROPHORESIS 2014, 35 (12–13), 1828–1836. https://doi.org/10.1002/elps.201400016.
(2) Campos, C. D. M.; Gamage, S. S. T.; Jackson, J. M.; Witek, M. A.; Park, D. S.; Murphy, M. C.; Godwin, A. K.; Soper, S. A. Microfluidic-Based Solid Phase Extraction of Cell Free DNA. Lab Chip 2018, 18 (22), 3459–3470. https://doi.org/10.1039/C8LC00716K.
(3) de Vries, R. DNA Condensation in Bacteria: Interplay between Macromolecular Crowding and Nucleoid Proteins. Biochimie 2010, 92 (12), 1715–1721. https://doi.org/10.1016/j.biochi.2010.06.024.

PS3-45 A lateral flow smartphone-based biosensor for rapid on-site assay of carcinoembryonic antigen

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Carcinoembryonic antigen (CEA) is a protein not detectable in the blood of healthy adults but may be increased in certain types of cancer [1,2]. CEA tests are usually conducted to detect colorectal or bowel cancer, but also to ascertain if treatment is successful during cancer treatment or if cancer has reappeared after treatment [3,4]. Considering the importance of a CEA detection, it is of immediate priority to develop rapid, cost-efficient and sensitive methods for CEA monitoring. The detection of CEA in blood samples is mostly based on commercial immunoassays such as enzyme-linked immunosorbent assays (ELISA) [5], immunonephelometry [6], chemiluminometric immunoassays (CLIA) [7], etc. However, these assays require trained personnel, specialized equipment and are time-consuming. On the contrary, lateral-flow assays (LFAs) are more convenient for on-site measurements, are simpler and provide satisfactory sensitivity, therefore are ideal for rapid point-of-care detection of CEA [8]. In this work, we describe a simple, portable and lateral flow biosensor for rapid CEA determination in serum samples. The sandwich-type assay involves a CEA-specific capture antibody (Ab1) immobilized on the surface of the test line of the strip and a second CEA-specific Ab2 conjugated with gold nanoparticles (AuNPs) as the biorecognition element. In the presence of CEA, the Ab2-AuNPs conjugates are bound with the analyte and the CEA-Ab2-AuNPs conjugates are captured by the immobilized Ab1. Qualitative detection of CEA is performed by visual inspection of the intensity of the test line while quantification is performed by reflectance colorimetry using a smartphone and image analysis. Under optimal conditions, the response is linear over the range 2.5-500 ng mL-1 CEA while the visual limit of detection of the strip for qualitative detection is 2.5 ng mL-1. The assay takes 20 minutes to complete, while the strips are stable over a long time period, indicating that the proposed biosensor could be a potential useful tool for rapid on-site detection of CEA in a wide concentration range.

[1] S. Hammarström, Semin Cancer Biol. 9 (1999) 67-81. DOI:10.1006/scbi.1998.0119
[2] R. Lázaro-Gorines, et al., Sci Rep. 9 (2019) 11680. DOI: 10.1038/s41598-019-48285-z
[3] D. Shinmi, et al., Cancer. 6 (2017) 798. DOI: 10.1002/cam4.1003
[4] T. Ozawa, et al., J Surg Oncol. 124 (2021) 97. DOI: 10.1002/jso.26497
[5] T.L. Wu, et al., J. Clin. Lab. Anal. 17 (2003) 241. DOI: 10.1002/jcla.10104
[6] X.J. Wu, et al., J. Int. Med. Res. 39 (2011) 838. DOI: 10.1177/147323001103900317
[7] S. Qu, et al., Anal Chim Acta. 766 (2013) 94. DOI: 10.1016/j.aca.2012.12.043
[8] T. Mahmoudi, et al., Biosensors (Basel). 11 (2021) 392. DOI: 10.3390/bios11100392

PS3-46 Multiplexed LC - MS permeation analysis in artificial cell systems

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The membrane permeation profile characterizes the permeable compounds within the mixture of chemical species across a biological membrane. In any membrane separated system, the membrane permeation profile thus emerges as a function of protein and lipid composition as well as the surrounding chemical environment. The gap between biological and artificial membranes remains a contentious factor due to limitations in the reconstitution of compositional and functional membrane

complexities. This gap has driven an emergent hypothesis regarding the relevance of simple diffusion in biological systems.1

Artificial cell materials are assembled from minimal biological components as an engineering platform for biomimetic in vitro systems. In looking up to cells, artificial ones begin with the compartmentalization of an aqueous volume and typically an imitation of a biological process. Information extraction in artificial cell systems can be limited by dependencies on fluorescence-based readouts. The Bioanalytics group joins microfluidic platforms with downstream analytical techniques such as LC-MS to assay chemically and biologically diverse analytes. 2,3

Droplet interface bilayers (DIBs) are a leading artificial cell architecture for mimicking chemical gradients across biological membranes, with studies spanning across fundamental and translational science. To design droplet microfluidic based artificial cell systems with chemical libraries, the permeability rate constant and oil partition dynamics were assessed across an FDA-approved library. Analysis was performed within a temperature controllable, automated open microfluidic system with access to droplet sequestration. Mixtures of small molecules were assembled based on compound retention time, where LC-MS characterisation of droplets ascertained whether a compound was permeable or not, defining the membrane permeation profile. The large number of analysed compounds allowed for an assessment of basic physicochemical descriptors as predictors of the permeability. This strategy of membrane permeation profile characterisation may be used to rationalise the biophysical differences between artificial and biological membranes.

1 E. Girardi, et al., Nat Chem Biol 16 (2020) 469–478. https://doi.org/10.1038/s41589-020-0483-3
2 S. Bachler, et al., Anal Chem 93 (2021) 5137–5144. https://doi.org/10.1021/acs.analchem.0c04939
3 S. Bachler, et al., Commun Biol. 3 (2020) 769.
https://doi.org/10.1038/s42003-020-01489-w

PS3-47 Electromembrane extraction of peptides based on hydrogen bond interactions

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Electromembrane extraction (EME) of peptides have been reported in several research papers recently. Conceptually, said research have involved transfer of net positively charged peptides from an aqueous sample, through a liquid membrane, and into an aqueous acceptor solution, under the influence of an electrical field. The liquid membrane has been an organic solvent held by capillary forces in a porous solid membrane (support membrane), and ionic carriers have been added to the liquid membrane for efficient mass transfer of peptides. The purpose of the ionic carriers has been to facilitate peptide solvation in the organic solvent based on ionic interactions. Unfortunately, ionic carriers increase the conductivity of the liquid membrane; the current in the system increases, the electrolysis in sample and acceptor is accelerated, and the extraction system tend to be unstable and suffers from drifting pH. In the present work, a broad selection of organic solvents were tested as pure liquid membrane for EME of peptides, without ionic carrier. Several alkylated phosphates provided

high mass transfer, and tri(pentyl) phosphate was selected since this solvent also provided high operational stability. Among 16 different peptides used as model analytes, tri(pentyl) phosphate extracted those with net charge +1 and with no more than two polar side chains. Tri(pentyl) phosphate served as a very strong hydrogen bond acceptor, while the protonated peptides were hydrogen bond donors. By such, hydrogen bonding served as the primary interactions responsible for mass transfer. Tri(pentyl) phosphate as liquid membrane, could exhaustively extract leu-enkephalin, met-enkephalin, and endomorphin from human blood plasma and detected by LC-MS/MS. Calibration curves were linear (r2 >0.99) within a concentration range from 1 to 500 ng/mL, and a relative

standard deviation within 12% was observed for precision studies. The current experiments are important because they indicate that small peptides of low polarity may be extracted selectively in EME based on hydrogen bond interactions, in systems not suffering from electrolysis.

References

1. S. Pedersen-Bjergaard and K.E. Rasmussen, Electrokinetic migration across artificial liquid membranes: New concept for rapid sample preparation of biological fluids. J. Chromatogr. A. 1109 (2) (2006) 183-190.

2. M. Drobnjak, F. Hansen, E.L. Øiestad, T. Løvli, R. Trones, Ø.G. Martinsen, and S. Pedersen-Bjergaard, Electromembrane Extraction with Vials of Conducting Polymer. LCGC North America. 38 (8) (2020) 435-439.

3. T.G. Skaalvik, E.L. Øiestad, R. Trones, S. Pedersen-Bjergaard, and S. Hegstad, Determination of psychoactive drugs in serum using conductive vial electromembrane extraction combined with UHPLC-MS/MS. J. Chromatogr. B. 1183 (2021) 122926.

4. E. Santigosa-Murillo, S. Maspoch, M. Muñoz, and M. Ramos-Payán, An efficient microfluidic device based on electromembrane extraction for the simultaneous extraction of acidic and basic drugs. Anal. Chim. Acta. 1160 (2021) 338448.

5. N.C. Dominguez, A. Gjelstad, A.M. Nadal, H. Jensen, N.J. Petersen, S.H. Hansen, K.E. Rasmussen, and S. Pedersen-Bjergaard, Selective electromembrane extraction at low voltages based on analyte polarity and charge. Journal of chromatography. A. 1248 (2012) 48-54.

6. C.X. Huang, A. Gjelstad, and S. Pedersen-Bjergaard, Organic solvents in electromembrane extraction: recent insights. Rev. Anal. Chem. 35 (4) (2016) 169-183.

7. M.S. Restan, H. Jensen, X.T. Shen, C.X. Huang, O.G. Martinsen, P. Kuban, A. Gjelstad, and S. Pedersen-Bjergaard, Comprehensive study of buffer systems and local pH effects in electromembrane extraction. Anal. Chim. Acta. 984 (2017) 116-123.

8. M. Ramos Payán, E. Santigosa, R. Fernández Torres, and M.Á. Bello López, A New Microchip Design. A Versatile Combination of Electromembrane Extraction and Liquid-Phase Microextraction in a Single Chip Device. Anal. Chem. 90 (17) (2018) 10417-10424.

9. S. Seidi, M.H. Ranjbar, M. Baharfar, M. Shanehsaz, and M. Tajik, A promising design of microfluidic electromembrane extraction coupled with sensitive colorimetric detection for colorless compounds based on quantum dots fluorescence. Talanta. 194 (2019) 298-307.

10. F. Zarghampour, Y. Yamini, M. Baharfar, G. Javadian, and M. Faraji, On-chip electromembrane extraction followed by sensitive digital image-based colorimetry for determination of trace amounts of Cr(vi). Anal. Methods. 12 (4) (2020) 483-490.

11. F.A. Hansen, N.J. Petersen, J.P. Kutter, and S. Pedersen-Bjergaard, Electromembrane extraction in microfluidic formats. J. Sep. Sci. 45 (1) (2022) 246-257.

12. Y. Ma, Y. Luo, X. Feng, C. Huang, and X. Shen, Smartphone-controlled biosensor for viral respiratory infectious diseases: Screening and response. Talanta. 254 (2023) 124167.

13. F. Zarghampour, Y. Yamini, E. Alipanahpour Dil, A. Shokrollahi, and G. Javadian, A new microfluidic-chip device followed by sensitive image analysis of smart phone for simultaneous determination of dyes with different acidic–basic properties. Talanta. 254 (2023).

14. Q. Shang, H. Mei, C. Huang, and X. Shen, Fundamentals, operations and applications of electromembrane extraction: An overview of reviews. Microchem J. 181 (2022) 107751.

15. L.V. Eie, S. Pedersen-Bjergaard, and F.A. Hansen, Electromembrane extraction of polar substances – status and perspectives. J. Pharm. Biomed. Anal. 207 (2022) 114407.

16. N. Drouin, P. Kubáň, S. Rudaz, S. Pedersen-Bjergaard, and J. Schappler, Electromembrane extraction: Overview of the last decade. Trends Anal. Chem. 113 (2019) 357-363.

17. A. Šlampová and P. Kubáň, Micro-electromembrane extraction through volatile free liquid membrane for the determination of β-lactam antibiotics in biological and environmental samples. Talanta. 252 (2023).

18. H. Rahbarian, S. Nojavan, M. Maghsoudi, and H. Tabani, In-tube gel electromembrane extraction: A green strategy for the extraction of narcotic drugs from biological samples. J. Chromatogr. A. 1688 (2023).
19. R. Hu, Y. Yan, L. Jiang, C. Huang, and X. Shen, Determination of total cathinones with a single molecularly imprinted fluorescent sensor assisted by electromembrane microextraction. Microchimica Acta. 189 (9) (2022) 324.
20. G.L. de Araújo, G.F. dos Santos, R.O. Martins, G. da Silva Lima, I. Medeiros, R.M. de Carvalho, R.C. Simas, L.F. Sgobbi, A.R. Chaves, and B.G. Vaz, Electromembrane Extraction of Naphthenic Acids in Produced Water Followed by Ultra-High-Resolution Mass Spectrometry Analysis. J. Am. Soc. Mass. Spectrom. 33 (8) (2022) 1510-1517.
21. S. Yousefi, S. Makarem, W. Alahmad, F.D. Zare, and H. Tabani, Evaluation of complexing agents in the gel electro-membrane extraction: An efficient approach for the quantification of zinc (II) ions in water samples. Talanta. 238 (2022) 123031.
22. A. Sahragard, W. Alahmad, and P. Varanusupakul, Application of electrocolorimetric extraction for the determination of Ni(II) ions in chocolate samples: A green methodology for food analysis. Food Chem. 382 (2022) 132344.
23. L. Wan, H. Gao, H. Gao, R. Du, F. Wang, Y. Wang, and M. Chen, Selective extraction and determination of steroidal glycoalkaloids in potato tissues by electromembrane extraction combined with LC-MS/MS. Food Chem. 367 (2022).
24. C. Román-Hidalgo, G. López-Pérez, M. Villar-Navarro, and M.J. Martín-Valero, Green electromembrane extraction procedure based on biodegradable chitosan films for determination of polyphenolic compounds in food samples: Greenness assessment of the sample preparation approach. Talanta. 253 (2023).
25. F.A. Hansen and S. Pedersen-Bjergaard, Electromembrane extraction – looking closer into the liquid membrane. Advances in Sample Preparation, (2022) 100020.
26. M. Balchen, L. Reubsaet, and S. Pedersen-Bjergaard, Electromembrane extraction of peptides. J. Chromatogr. A. 1194 (2) (2008) 143-149.
27. M. Balchen, T.G. Halvorsen, L. Reubsaet, and S. Pedersen-Bjergaard, Rapid isolation of angiotensin peptides from plasma by electromembrane extraction. J. Chromatogr. A. 1216 (41) (2009) 6900-6905.
28. M. Balchen, H. Jensen, L. Reubsaet, and S. Pedersen-Bjergaard, Potential-driven peptide extractions across supported liquid membranes: Investigation of principal operational parameters. J. Sep. Sci. 33 (11) (2010) 1665-1672.
29. M. Balchen, A.G. Hatterud, L. Reubsaet, and S. Pedersen-Bjergaard, Fundamental studies on the electrokinetic transfer of net cationic peptides across supported liquid membranes. J. Sep. Sci. 34 (2) (2011) 186-195.
30. M. Balchen, H. Lund, L. Reubsaet, and S. Pedersen-Bjergaard, Fast, selective, and sensitive analysis of low-abundance peptides in human plasma by electromembrane extraction. Anal. Chim. Acta. 716 (2012) 16-23.
31. N. Drouin, S. Rudaz, and J. Schappler, Dynamic-Electromembrane Extraction: A Technical Development for the Extraction of Neuropeptides. Anal. Chem. 88 (10) (2016) 5308-15.
32. C.X. Huang, A. Gjelstad, and S. Pedersen-Bjergaard, Exhaustive extraction of peptides by electromembrane extraction. Anal. Chim. Acta. 853 (2015) 328-334.
33. C.X. Huang, A. Gjelstad, and S. Pedersen-Bjergaard, Selective electromembrane extraction based on isoelectric point: Fundamental studies with angiotensin II antipeptide as model analyte. J. Membr. Sci. 481 (2015) 115-123.
34. A. Pourahadi, S. Nojavan, and S.S. Hosseiny Davarani, Gel-electromembrane extraction of peptides: Determination of five hypothalamic agents in human plasma samples. Talanta. 217 (2020) 121025.
35. T.K. Rye, G. Martinovic, L.V. Eie, F.A. Hansen, T.G. Halvorsen, and S. Pedersen-Bjergaard, Electromembrane Extraction of Peptides using Deep Eutectic Solvents as Liquid Membrane. Anal. Chim. Acta. 1175 (2021) 338717.
36. K.F. Seip, J. Stigsson, A. Gjelstad, M. Balchen, and S. Pedersen-Bjergaard, Electromembrane extraction of peptides – Fundamental studies on the supported liquid membrane. J. Sep. Sci. 34 (23) (2011) 3410-3417.
37. L.V. Eie, T.K. Rye, F. Hansen, T.G. Halvorsen, and S. Pedersen-Bjergaard, Electromembrane extraction of peptides and amino acids – status and perspectives. Bioanalysis. 13 (4) (2021) 277-289.
38. F.A. Hansen, E. Santigosa-Murillo, M. Ramos-Payán, M. Muñoz, E. Leere Øiestad, and S. Pedersen-Bjergaard, Electromembrane extraction using deep eutectic solvents as the liquid membrane. Anal. Chim. Acta. 1143 (2021) 109-116.
39. A. Gjelstad, K.E. Rasmussen, and S. Pedersen-Bjergaard, Simulation of flux during electro-membrane extraction based on the Nernst–Planck equation. J. Chromatogr. A. 1174 (2007) 104-111.

PS3-48 VIRTUAL INSTRUMENTS FOR FILLING A GAP IN PEAK EVALUATION SOFTWARE FOR FLOW-BASED METHODS

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A virtual instrument (VI) is a code that can operate modular hardware, acquire signals, read files, modify, evaluate, present, and store data. A VI can comprise all the functionalities or some of them, depending on the objectives. NI LabView is a graphical programming environment dedicated to the development of the VIs. Graphical programming is easier to learn than text-based programming. Icons are selected from the pallets by a drag-and-drop approach. It enables researchers and engineers who are not qualified programmers to develop instruments tailored to their professional needs. The process has two aspects. For operating the VI through a computer screen, a front panel consisting of controls and indicators is designed first. It becomes the user interface. The block diagram as an executable code is derived from it. A block diagram consists of nodes comprising functions, sub-Vis, and structures. They are connected with wires which correspond to variables in a text-based programming language. Liquid chromatography and other flow-based analytical methods require software for peak evaluations. General-purpose commercial packages lack some functionalities. User-developed VIs can fill the gap. The modularly structured multifunctional VIs for zone penetration studies are going to be presented and examples of applications provided.

PS3-51 Multiple Critical Quality Attributes Assessment of mAbs for Process Control - Agilent InfinityLab Online LC Solution for automated heart-cutting 2D-LC experiments

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Monoclonal antibodies (mAbs) belong to one of the most important biopharmaceutical drug classes. A monoclonal antibody is uniform on a molecular level, but during production, purification, and formulation, changes might happen, which can compromise their efficacy and safety. These changes may include the formation of higher molecular weight aggregates like dimers or trimers or modifications resulting in charge variants. These modifications are two of the most important critical quality attributes (CQAs). Therefore, it is important to control the CQAs of a mAb during production (upstream) as well as during and after purification and formulation (downstream).

Analysis of the CQAs during the production or the purification process for the automated control of the product quality of mAbs can be performed using the Agilent InfinityLab Online LC Solution.[1] For the analysis of mAb CQAs, a two-dimensional heart-cutting LC analysis can be applied with online analytics. The combination of Protein A affinity chromatography in the first dimension and a method for the determination of a quality attribute in the second dimension enhances efficiency in contrast to separated workflows. The combined automated workflow reduces analysis time and cost especially if used in process analytical technology (PAT) for up- and/or downstream product quality assessment.[2]
A protein A affinity chromatography in the first dimension is applied for titer determination and/or purification for the subsequent analyses. The effluent from the Protein A column is collected in a loop and transferred to the second dimension column to separate the mAb on, for example, a size exclusion or ion-exchange column. This setup allows the determination of titer and aggregates or titer and charge variants in one method at a time.

The use of the Agilent Online LC Monitoring Software for online process control by an Agilent Online LC applied for a two-dimensional heart cutting experiment for the determination of two CQAs of a mAb is demonstrated. This setup enables complete automation of up- and downstream online process

monitoring of mAb production for their CQAs in an economic and time saving fashion and enables automated intact multi attribute method (MAM) analysis during the bioprocess.

1. Bonnington, L.; Naegele, E.; Kutscher, D. Online LC Monitoring of Downstream Processing in the Production of Therapeutic mAbs. Agilent Technologies application note, publication number 5994-5186EN, 2022.
2. Chemmalil, L.; Naegele, E.; Kutscher, D. An automated Sampling System for Online Monitoring as a Process Analytical Technology (PAT) Tool, Agilent Technologies application note, publication number 5994-3716EN, 2021

PS3-52 Calix[6]arene and TiO2 modified reduced graphene oxide electrode-based portable stochastic platform for the determination of nonivamide from topical pharmaceutical dosage forms and water samples

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A new portable stochastic detection platform based on an integrated sensor constructed by modifying a reduced graphene oxide and TiO_2 paste with calix[6]arene was proposed for both quantitative and qualitative assay of nonivamide in topical pharmaceutical dosage forms and water samples. By employing the stochastic detection platform, a wide linear analytical range from 1.00 × 10−18 mol L−1 to 1.00 × 10−1 mol L−1 was achieved, with a very low limit of quantification of 1.00 × 10−18 mol L−1. The proposed platform was applied for the determination of nonivamide in real samples, obtaining good recovery values in the performed tests. Moreover, the pharmaceutical samples were analyzed without any pretreatment, while the surface water samples required minimal preliminary processing, demonstrating an approach that is not only simple but also quick and reliable. In addition, the designed detection platform is easily transported, making it suitable for the on-site and continuous assay of nonivamide for the assessment of surface water quality and the quality control tests performed in the pharmaceutical industry.

PS3-53 Efficient sol-gel immobilization of microporous polymer on silica-based adsorbent for the enrichment of non-steroidal anti-inflammatory drugs

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Para-cyano conjugated microporous polymer (PCCMP, surface area of 784 m^2 g^-1 and pore size of 1.29 nm) was immobilized on the outer surface of the silica microsphere by means of intercalation within sol-gel based outer shell. This simple, efficient, and cost-effective method of immobilization allowed for a stable combination of multi-functional organic polymer with the silica microsphere without any pre-or post-synthesis chemical treatments. The prepared core-shell adsorbent was packed in an SPE cartridge and used for the enrichment of non-steroidal anti-inflammatory drugs (NSAIDs) in water and acetate buffer samples prior to the analysis by HPLC-UV. The prepared adsorbent was characterized by Fourier-transform infrared spectroscopy (FTIR), X-ray photoelectron spectroscopy (XPS), elemental analysis, field emission scanning electron microscope (FESEM), transmission electron microscopes (TEM). The optimization of the sampling-, enrichment-, and elution-conditions were

thoroughly investigated. Detailed investigations of the analytical parameters influencing the extraction efficiency were validated. At optimum pre-determined conditions, the prepared adsorbent allowed for good linearity (r2> 0.9982), run-to-run reproducibility (RSD < 7 %) with good limits of detection, and limits of quantitation of (0.33 – 1.46 ng L^-1) and (1.1 – 4.8 ng L^-1), respectively. Compared to the commercially recommended SPE adsorbent for NSAIDs, the PCCMP-SPE adsorbent showed enhanced affinity and enrichment capabilities.

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PS3-54 Application of microextraction in a packed syringe approach for determination of phosphate in natural water samples

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Microextraction in a packed syringe (MEPS) is a new miniaturized version of solid phase extraction. The determination is based on the modification of known procedures [1,2] of the formation of phosphoantimonylmolybdenum blue complex (PMB) and its sorption [3,4]. The formation of PMB is followed by the formation of an ion associate with cetyltrimethylammonium bromide (CTAB) and its sorption on the C-18 sorbent. After the sorption, the analyte is eluted and quantified spectrophotometrically. Sorption was performed using an eVol® XR semi-automatic device equipped with a 500 µL MEPS syringe and a C-18 sorbent cartridge (BIN). The absorbance was measured using a Lightwave II UV-Vis spectrophotometer (Biochrom, UK) with a quartz microcuvette (Starna Scientific Ltd., England) with an optical path length of 1 cm and a volume of 5 µL.

A calibration plot constructed under optimized experimental conditions was linear in the range 1.55–24.8 µg/L. The limit of detection (LOD) and limit of quantification (LOQ), calculated from blank test was 0.39 and 1.3 µg/L, respectively. The preconcentration factor calculated from the slope of the calibration plots with and without preconcentration step was around 46. The precision and accuracy of the developed method were checked by performing 3 extractions of spiked water samples at two concentration levels (2.48 and 22.32 µg/L) over two days. The relative standard deviation percentage (RSD, 4.6–13.1%) was used to calculate the method's precision, while the accuracy of the method was characterized by the recovery percentage (R, 95.3–101.7%). The procedure was applied to the determination of phosphate in real water samples (river water, spring water).

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[1] L. Drummond, W. Maher, Anal. Chim. Acta 302 (1995) 69. DOI: 10.1016/0003-2670(94)00429-P
[2] J. Murphy, J.P. Riley, Anal. Chim. Acta 27(C) (1962) 31. DOI: 10.1016/S0003-2670(00)88444-5
[3] Y. Liang, et al., Mar. Chem. 103 (2007) 122. DOI: 10.1016/j.marchem.2006.06.013
[4] V. Phouthavong, et al., Sci. Rep. 11 (2021) 23073. DOI: 10.1038/s41598-021-02516-4

PS3-55 Assessment of Hollow Fiber and Dispersive Solid Phase Microextraction combined with Total Reflection X-ray Spectrometry (TXRF) for Inorganic Arsenic Speciation Analysis in Water

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Arsenic contamination in drinking water is a global health concern due to its toxicity, and accurate speciation analysis is crucial for assessing the associated health risks. In this study, we evaluate two microextraction techniques, namely hollow fiber liquid phase microextraction (HF-LPME) and dispersive solid phase microextraction (DSPME), combined with total reflection X-ray spectrometry (TXRF), for the sensitive and selective determination of inorganic arsenic species in water.

Inorganic arsenic species, such as arsenite (As(III)) and arsenate (As(V)), are the toxic forms of arsenic found in water that can lead to adverse health effects. While conventional methods for speciation analysis, such as liquid chromatography inductively coupled plasma mass spectrometry (LC-ICP/MS), are effective, they can be expensive and suffer from matrix effects. In this study, we aim to explore the use of cost-effective and efficient microextraction techniques, namely HF-LPME and DSPME, combined with TXRF, for the determination of trace amounts of inorganic arsenic species in water.

In the present study and taking into account the microanalytical capability of total reflection X-ray spectrometry (TXRF), we investigated a hollow fiber three phase liquid-phase microextraction system and a dispersive solid phase extraction method using multiwalled carbon nanotubes impregnated with the ionic exchanger Aliquat 336, both combined with TXRF for the determination of trace amounts of inorganic As species in waters. We evaluated the performance of HF-LPME and DSPME for the extraction of As(III) and As(V) from water samples under various conditions, including extractant type, organic solvent, pH, and extraction time. Our results show that As(III) extraction is optimal at pH 13, while the optimum pH for As(V) extraction is 8.5. Both methods, when optimized, demonstrated excellent detection limits that meet current regulatory requirements for the determination of inorganic arsenic species in water samples. Overall, our study demonstrates the potential of HF-LPME and DSPME combined with TXRF as cost-effective and reliable methods for the determination of inorganic arsenic species in water.

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PS3-57 Polymer nanofibrous disks for preconcentration of environmental contaminants prior to HPLC determination

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Low concentration levels of environmental pollutants in water generally require extraction and preconcentration step prior to HPLC analysis. Solid phase extraction (SPE) enables high enrichment of target analytes. Another advantage of SPE is the variety and availability of a wide range of sorbent selectivity and, more recently, the intensive research on new and advanced nanosized materials for extraction. Polymer nanofibers represent sorbents for solid phase extraction attempting

miniaturization and diversity and feature a large surface area to volume ratio leading to good adsorption capacity and fast kinetics of adsorption and desorption. Nanofibrous polymeric sorbents can be customized using a great variety of organic monomers, fabrication procedures, and surface modifications. Furthermore, fibrous sorbents can be adjusted in various formats of SPE. In water contaminants analysis, when preconcentration of trace-level contaminants is enabled from quite an unlimited sample volume, filtration through an SPE disk is preferable due to the high flow rate and easy automation. Similarly, high-volume samples can be treated using sorbent stirring in full volume, facilitating analytes diffusion and adsorption, such as stir bar sorptive extraction, followed by elution to a minimum volume of solvent. Novel methods applying original nanofibrous disks will be presented for pollutants preconcentration using centrifugal filters or simple stirring in water samples. Different polymers were tested regarding extraction efficiency, such as polycaprolactone, polyhydroxybutyrate, polyurethane, and hybrid nanofibrous material composed of organic polymer highly doped with graphene. At least a twofold increase in preconcentration of analytes was achieved with the addition of graphene into the polymer in comparison with the plain polymer. Finally, the optimized methods were validated with reasonable recoveries and precision of extraction. The use of fibrous disks brings several advantages to analytical applications, such as simple preparation and handling, miniaturization, avoiding plastic waste from disposable material, and high preconcentration factors.

PS3-58 New zwitterionic materials for the selective extraction of analytes from environmental samples

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Sample preparation is a key step in analytical procedure and sorptive-based extraction techniques are the most preferred for liquid samples. In the recent years, mixed mode ion-exchange sorbents have been developed to increase the retention of ionic or ionizable compounds which show low retention in the commonly used sorbents. Mixed mode ion-exchange sorbents retain compounds through reversed-phase and ion-exchange interactions and selectivity can be achieved when an effective clean step is included in the extraction protocol [1]. Mixed mode ion-exchange sorbents that are commercially available are only based in one type of ion-exchange interaction, either anionic or cationic interactions, and this may be a significant limitation for the simultaneous extraction of acidic and basic analytes.

To overcome this limitation, different strategies have been developed to combine cationic and anionic sorbents [2]. A new strategy is to prepare zwitterionic sorbents that combine cationic and anionic moieties in the same sorbent [3]. In this sense, two strategies to create homemade silica-based zwitterionic sorbents have been explored. The first one was the functionalization of silica through sol-gel approach with C18 chains, sulfonic groups and quaternary amines, so the sorbent can perform reversed-phase, strong cation-exchange and strong anion-exchange interactions [4]. The second strategy evaluated was the functionalization of the silica with zwitterionic groups that combine sulfonic and quaternary amine moieties. These sorbents were evaluated for the solid-phase extraction (SPE) of a group of acidic and basic pharmaceuticals.

To promote the ion-exchange interactions, the loading pH was carefully optimized and to increase the selectivity, a clean-up step with methanol was included, which enables the compounds only retained by reversed-phase interaction to be washed up. To elute the compounds, the pH of the elution solvent

is also critical because the ion-exchange interactions must be disrupted. Sample volume was optimized to increase the sensitivity for each group of analytes.

The best performing sorbents were applied and the methods including SPE and liquid chromatography-mass spectrometry-based detectors were validated for the determination of analytes in river water and wastewater. Good quality parameters were obtained which demonstrate the suitability of these new zwitterionic sorbents and encourage their use for the selective extraction of other polar acidic and basic analytes from other complex samples.

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1.N. Fontanals et al., Anal. Chim. Acta 117 (2020) 89-107. DOI: 10.1016/j.aca.2020.03.053
2.D. Salas et al., Anal. Bional. Chem. 410 (2018) 459-469. DOI: 10.1007/s00216-017-0736-5
3. J.C. Nadal et al., J. Chromatogr. A 1661 (2022) 462715. DOI: 10.1016/j.chroma.2021.462715
4. A. Moral et al., J. Chromatrogr. A 1676 (2022) 463237.DOI: 10.1016/j.chroma.2022.463237

PS3-59 Vacuum Assisted Sorbent Extraction : VASE, a qualitative and green approach for VOCs to SVOCs analysis using GCMS

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A novel method based on Vacuum-Assisted sorbent extraction (VASE™) used with gas chromatography-mass spectrometry (GCMS) for determination of volatile compounds is described here. The method is based on extraction of analytes into sorbent traps (Sorbent Pen™) filled with Tenax in a vacuum system: vials with traps from which air is evacuated. The method can be applied for extraction of volatile organic compounds (VOCs) from aqueous or solid matrix and flavor matrix such as perfumes or food samples were used as examples. Terpenes, aldehydes/ketones, esters, alcools, acids, phenols, and other micropollutants in method development optimal extraction parameters were elaborated. For the analysis of those VOCs the method is really characterized with satisfactory repetability and sensibility.

Limits of detection (LODs) for some analyzed compounds are ranged from 0.05 to 0.5 µg/L and repeatability for majority of compounds is under 5% for a single trap extraction. The idea of using vacuum to be utilized in sorbent extraction was also developed and commercialized based on specific sorbent traps (Sorbent Pens™), which are used with specially designed vial caps allowing evacuation of air from the vial by a membrane pump and special injection port, in which desorption of analytes into chromatographic column is performed. The system is manufactured by Entech Instruments and distributed by Quad Service.

Keywords: Vacuum-assisted sorbent extraction (VASE™) ; VOCs; flavor compounds; Gas Chromatography-Mass spectrometry (GCMS).

PS3-60 A new environmentally friendly procedures for preconcentration and online monitoring of selected analytes

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We present some new, fast and, above all, environmentally friendly procedures for the preconcentration and online monitoring of selected analytes, environmental contaminants, such as Hg, Cd, and Fe. For each of process, the reaction conditions were studied.

The first procedure is based on the formation of a specific ion association complex between Hg(II) and the polymethine dye Astra Phloxine in the presence of iodide ions. The analytical wavelength was chosen in the region of an intense narrow band with maximum at 600 nm that bathochromically shifted related to the band of the aggregated dye. An automated flow-based system was used to reducing of reagents consumption. The limit of detection was 10 µg/L Hg.

The second procedure is cloud point extraction (CPE) for the preconcentration of traces (Hg and Cd) before its spectrophotometric determination. The process is based on extraction of Hg(II) and Cd(II) using surfactant Triton X-114 after complexation reaction with the hydrophobic azo dye 6-hexyl-4-(2-thiazolylazo)resorcinol at pH 11 and pH 9.5, respectively. Experimental conditions for CPE were found: incubation time 15 min at 60°C, mass fraction of the surfactant Triton X-114 (1%). A micro-volume setup for UV-Vis detection was applied. The limit of detection for cadmium was 3.5 µg/L.

The third procedure is an automated liquid phase microextraction of iron. The procedure uses flow system for miniaturization of liquid phase extraction of Fe(III) after reaction with Astra Phloxine dye in the presence of chloride ions. Proposed procedure has some advantages, such as closed system for the handling of organic solvent, low sample and reagents consumption, and full automation. Using spectral pipette for detection lead to reduce the organic solvent consumption and thus also to increase the sensitivity of the determination. The limit of detection was 5 µg/L Fe.

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PS3-61 In Situ Rapid Electrochemical Fabrication of Porphyrin-based Covalent Organic Frameworks Fibers for Electro-enhanced Solid-phase Microextraction

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Solid-phase microextraction (SPME), a promising sample pretreatment technique, has been widely applied in the field of environmental, biological, food analysis since it was introduced in the 1990s. In the process of SPME, diffusion equilibrium of analytes between sample matrix and extraction phase is via passive diffusion. It has relatively low extraction efficiency and takes a long time for polar or ionic com-pounds. In order to improve the extraction efficiency and selectivity of targets, auxiliary electric

field technology have been introduced to accelerate the diffusion equilibrium of analytes, which is defined as electro-enhanced solid-phase microextraction (EE-SPME). The extraction principle depends on applying different electrical potential to change the relative distribution of polar or ionic compounds between the two phases for improvement of extraction efficiency and selectivity, where fibers (extraction phase) with high extraction capacity and strong conductivity is the key point of EE-SPME. Over the past decade, a limited variety of fibers, including commercial polydimethylsiloxane/divinylbenzene fiber (PDMS/DVB), metal oxides, carbon materials, and conductive polymers, have been developed and applied in this field. However, few of them have good extraction performance, attributing to low specific surface area or poor conductivity of coating materials on the surface of fibers.

Herein, we propose and demonstrate a facile and novel preparation strategy of high conductive fibers based on one-step electropolymerization, where porphyrin was used as a conductive active monomer for the construction of covalent organic framework coatings (POR-COF) on the surface of fiber in situ. The electric field-based preparation method can more easily and orderly regulate the morphology and thickness of the coatings by adjusting the applied positive potential and preparation time. These factors directly affect the electrical properties of the fiber, which can effectively solve the problems of poor reproducibility for existing conductive fibers. At the same time, electric field endows the extraction technology with a controllable adsorption process and efficiently achieves selective extraction of targets. To evaluate the performance of POR-COF in EE-SPME, several phthalates esters (PAEs) were selected as the model analytes, which have been most commonly existing in industrial wastewater, lake water, beverages and oyster samples from local areas. The results show satisfactory sensitivity and excellent recovery through gas chromatography-tandem mass spectrometry (GC-MS/MS) analysis. In summary, the construction of novel conductive COF materials and their fibers provides the possibility for vigorous development of EE-SPME technology.

PS3-62 Nanoparticle-directed metal organic framework and ionic liquids@metal organic framework nanocomposites hybrid monolith for efficient capillary microextraction

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The integration of micro- or nanocrystalline materials in porous organic polymer monoliths is a promising strategy for the development of high-performance adsorbents-based capillary microextraction (CME). Metal–organic frameworks (MOFs), an emerging class of porous crystalline materials, have attracted intensive interest in analytical sample preparation, owing to their designable chemical composition, adjustable pore size, large specific surface area and abundant coordinated unsaturated centers. The polymeric MOFs-derived adsorbent (MOFs-polymer) did facilitate adsorption and ensure reproducibility since it inherits favorable properties of both MOFs and polymers. Unfortunately, the preparation of MOFs-polymer composites still faces with difficulty that limits their real application, because of the stark differences between the nature of crystalline MOFs and flexible organic polymers. The currently adopted strategies (e.g., physical blending, copolymerization, mixed matrix membrane and electrospinning) suffer from the inevitable aggregation, pore blockage and limited accessibility of MOFs, resulting in a negative effect on the adsorption performance of the composites. The development of MOFs-polymer composites sorbents with high efficiency and facile preparation routes is of great importance.

Herein, we reported a novel nanoparticle-directed strategy, instead of the conventionally used metal salts-based method, to synthesis MOFs and ionic liquids/metal–organic frameworks (ILs@MOF) nanocomposites hybrid monolith for efficient capillary microextraction (CME). The ZnO nanoparticles (ZnO-NPs) were initially introduced into a precursor polymer monolith, and acted as the metal sources and anchoring seeds to construct zeolitic imidazolate framework (ZIF-8) or ILs@ZIF-8 nanocomposites via a nanoparticle-directed in situ growth route in DMF/H2O or confined imidazolium ionic liquids. The in-situ growth of ZIF-8 or ILs@ZIF-8 coating significantly increased the surface area of the parent monolith and exhibited multiple interactions with target analytes, which contributed to the excellent extraction performance of the resultant hybrid monolith. During the ionothermal synthesis for the construction of ILs@ZIF-8 nanocomposites, the confined ILs not only served as solvent for in situ growth but also acted as structure directing agent, which tuned the structure and performance of ILs@ZIF-8 nanocomposites. By coupling the hybrid monolith-based CME with LC-MS, a sensitive and environment-friendly analysis of ultra-trace perfluoroalkyl phosphonic acids (PFPAs) and microcytins (MCs) in complex matrix was achieved with satisfactory recoveries.

PS3-63 A new concept for the control of functional food creation methods by speciation analysis of various elements present in microalgae

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Consumers are searching for healthy foodstuffs containing various nutrients and extraordinary health-beneficial features. Nowadays, the consumer demands more natural foods with health benefits, and as a result, microalgae are expected to perform an important role in the novel food industry in the future. Microalgae have been used for centuries as a human food source or nutritional supplement. Microalgae can accumulate valuable substances with potential applications in the food, feed, pharmaceutical, and cosmetics industries.

The main goal of the investigation was to develop an analytical method to control the content of microelements and trace elements in microalgae, including selenium, arsenic, copper, and zinc. During the project implementation, the content of selected elements will be determined, and their presents in different species of Spirulina will be compared. The task will be carried out using the inductively coupled plasma mass spectrometry technique (ICP-MS/MS). Additionally, speciation analysis of selected elements present in microalgae was performed using liquid chromatography techniques (SEC/HILIC) coupled with ICP-MS/MS and ESI-MS/MS.

The investigation's second goal was to develop a process of accumulating selected trace elements, making it possible to obtain a product suitable for human and/or animal consumption. The task will be accomplished by optimizing the content of selected elements in microalgae grown under controlled conditions with a specific content of ions of selected elements. The speciation analysis of selected elements in cultured microalgae will be made using liquid chromatography coupled with ICP-MS/MS. The research will identify the form of selected elements in the cultivated microalgae.

The obtained research results will become the starting point for designing new, more effective methods of creating functional food, which will directly impact the development of the environment and the economy.

PS3-64 Honey characterization and classification based on chromatographic profiles and antioxidant capacity

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Honey is a very appreciated product for its nutritional characteristics and its benefits for human health, comprising antioxidant, anti-inflammatory, antifungal, and antibacterial activities. These attributes depend on the specific composition of each honey variety, with the botanical origin as one of the distinctive features. Firstly, honey can be classified as honeydew and blossom honeys, depending on the raw material bees use to produce it. For honeydew honeys bees use plant secretions or sugar-rich materials that plant-sucking insects excrete. Contrary, the nectar of flowers is used to produce blossom honeys. Honeydew and blossom honeys show different physicochemical properties, being the antioxidant capacity, mainly relying on the phenolic compound content, one of the most important. In addition, within these two groups, honey from each specific botanical origin may have particular attributes relying on the honey composition.

In this work, honey samples were first characterized based on their bioactive compound contents. Different spectroscopic methods were used for the determination of total phenolic content (TPC), total flavonoid content (TFC), and antioxidant capacity. The Folin-Ciocalteu assay was used for the TPC determination. Regarding flavonoid content, two different methods, both based on the formation of aluminum chelates, were evaluated, observing that the response of compounds belonging to different flavonoid subfamilies depends on the experimental conditions. Lastly, the ferric reducing antioxidant power (FRAP) method was selected for determining the antioxidant capacity. Data obtained with these spectroscopic assays were treated by means of chemometric tools. As a result, a satisfactory discrimination (error 5%) between honeydew and blossom honeys were accomplished with the built partial least squares-discriminant analysis (PLS-DA) model. However, a complete classification of honeys according to their botanical variety was not fulfilled. Hence, for further characterization of the studied samples, a non-targeted C18 reversed-phase HPLC-UV-MS methodology was assessed. The obtained LC fingerprints were subjected to PLS-DA to evaluate their viability as sample chemical descriptors for classification purposes, obtaining good discrimination results between blossom- and honeydew-honey samples. In addition, the characterization and classification of honey samples according to their specific botanical origin was also achieved. Finally, several characteristic polyphenols of each botanical variety were tentatively identified by LC-MS/MS to propose possible honey markers for future experiments.

PS3-65 Dietary fatty acids as a new binding partner of C - phycocyanin: a fluorimetric study

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C-Phycocyanin (C-PC) is a phycobiliprotein from cyanobacteria, where it harvests light energy that is then transferred to chlorophylls during photosynthesis. It has an intense blue color due to a covalently bonded tetrapyrrole chromophore, and owing to this property is used in the food industry as a good natural alternative for food coloring. In addition to its coloring properties, C-PC has anti-inflammatory, antioxidant, anti-cancer, and immune-enhancing effects that qualify it as a dietary supplement already included in various formulations, mainly Spirulina extract powders. Since it is used as a food colorant and as a dietary supplement, it may interact with food ingredients, affecting its stability, digestibility, or antioxidant properties. Palmitic acid and linoleic acid (which can be metabolized to linolenic acid) are abundant in meat, milk, and edible oils, so that they could interact with C-PC. C-Phycocyanin isolated from the cyanobacterium Arthrospira platensis (Spirulina) was incubated with increasing concentrations of these three fatty acids, and its fluorescence intensity was monitored. Incubation resulted in a fluorescence quenching effect, indicating that binding had occurred. The binding equations indicated that the association constants were of the same order of magnitude and that the number of approximate binding sites was more than one ($Ka = 4.64 \times 10^4\ M^{-1}$, $n = 1.5$ for linoleic acid; $Ka = 2.88 \times 10^4\ M^{-1}$, $n = 1.9$ for linolenic acid; $Ka = 0.44 \times 10^4\ M^{-1}$, $n = 0.8$ for palmitic acid). This moderate interaction between C-PC and fatty acids could influence its behavior as a nutraceutical and food colorant.

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PS3-66 Determination of Veterinary Drug Residues in Foods of Animal Origin Using QuEChERS methology by LC–MS/MS

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The control of veterinary drug residues in food is of paramount importance for ensuring the quality and safety of food products in the European marketplace. Therefore, the European Parliament and of the Council the European Union have placed with the Directive 2019/06 a regulatory framework. This regulation lays down rules for the placing on the market, manufacturing, import, export, supply, distribution, pharmacovigilance, control and use of veterinary medicinal products [1]. The availability of safe and effective veterinary medicines is essential - to protect animals themselves, but also to protect humans from the transmission of diseases by animals, the so-called zoonoses [2, 3]. For industry and national regulatory laboratories, the challenges of controlling veterinary drug residues in food include the high number of drugs (antibiotics, antiparasitics, anti-inflammatory agents, etc.) and the diversity of foods of animal origin. It is critical to use an efficient sample pretreatment method for analyte extraction, concentration and matrix clean-up.

In this work, a sensitive QuEChERS method with an efficient cleanup for animal origin sample matrices like milk, eggs and beef was developed. The sample raw extract was purified with a cleanup-mix with customized composition. Sodium sulfate was used instead of traditionally used magnesium sulfate to allow establishing multi-residue methods because of certain veterinary drug groups tend to chelate with magnesium ions. High recovery rates of veterinary drugs like benzimidazoles, glucocorticoids and

sulfonamides and the matrix-reduction for different sample materials are presented and discussed. The identification and the quantification of the focused analyts was finally carried out by ESI mass spectrometry on NUCLEOSHELL® RP18 column.

[1] REGULATION (EU) 2019/6 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 11 December 2018 on veterinary medicinal products and repealing Directive 2001/82/EC.
[2] Lekshmi, M., Ammini, P., Kumar, S., & Varela, M.F. (2017) Microorganisms 5, 11–36. doi:10.3390/microorganisms5010011.
[3] WHO (2011) Critically Important Antimicrobials for Human Medicine, 3rd ed.

PS3-67 Simultaneous determination of vitamins B5, B7 and B9 using stochastic sensors as tools

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Vitamins play a vital role in supporting daily physiological functions and are integral to metabolic processes, including the prevention of vascular events and the postponement of diabetic nephropathy progression. Accurate evaluation of food quality necessitates the ultrasensitive assessment of vitamins B5, B7, and B9. Hence, a prompt screening assay for multivitamin tablets, pharmaceutical tablets, water, and biological fluids such as urine is essential to ensure their accurate detection and quantification. The present research proposes a miniplatform that employs a 2D sensor based on Cobalt-Phthalocyanine/Carbon for detecting vitamins B5, B7, and B9 in various samples, such as multivitamin tablets, pharmaceutical tablets, water, and biological samples like urine. The utilization of stochastic sensors and the stochastic mode was chosen as the screening method for the diverse samples, given the complex structure of the matrix. Additionally, the stochastic sensors are capable of performing reliable qualitative and quantitative analyses. The identification and quantification of vitamins B5, B7, and B9 were achieved by determining their specific signatures and utilizing them to identify their signals in the diagrams obtained during the rapid screening of the samples. The sensor proposed in this research provided high sensitivities and low determination limits. The validation process involved the utilization of pharmaceutical tablets, supplement tablets, water samples, and biological samples, specifically urine. The sensor under consideration exhibits cost-effectiveness and can be employed for rapid screening tests, with a duration of 6 minutes, across a variety of samples, with over 150 measurements possible, and a lifespan of up to one month.

PS3-69 Extra-virgin olive oil phenolic compounds as modulators of the gut microbiota in diabetics: unravelling their colonic metabolism

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Recent studies have shown that individuals with Type 2 Diabetes Mellitus (T2DM) often exhibit alterations in the composition and diversity of their gut microbiota (dysbiosis) compared to individuals without diabetes. Microbiota metabolites have a strong influence in T2DM, through their diverse relationships with the host metabolism and implications in several pathways. In this sense, a bidirectional relationship between microbiota and diabetes coexists (i.e., T2DM changes the composition of the microbiota, and the changed microbiota influences the pathophysiology of the disease).

Intervention studies in humans have revealed the extent to which the microbiota can be modulated by dietary changes. Mediterranean Diet (MedD) is a major factor that promotes gut microbiota composition. Extra virgin olive oil (EVOO), apart from being the main source of dietary fat at the core of MedD, is a recognised functional food with a wide variety of healthy components, mainly phenolic compounds, with multiple beneficial effects at least in part exerted along with modulation of the gut microbiota.

In this study a high-phenolic EVOO obtained by co-crushing 'Brava Gallega' and 'Mansa de Figueiredo', two ancient autochthonous varieties from north-western Spain, was selected to perform a simulation of gastrointestinal digestion according to the INFOGEST standardised protocol. The extract containing the non-absorbable phenolic compounds was immediately subjected to faecal fermentation (from 0 h to 48 h) under simulated colonic conditions; fresh faecal samples were collected from two diabetic volunteers, one of whom was also obese. This in vitro model, which mimics the human gastrointestinal tract, made it possible to evaluate the effect of the oily substrate on:
i) Diabetic gut microbiota composition using 16S rDNA sequencing, differentiating between obese and non-obese profiles: only in the obese subject the decrease in Shannon and Simpson alpha diversity was significant at 48 h compared to baseline (0 h). Differences of beta diversity at genus level were observed for both volunteers between 0 and 48 h (wUnifrac, Bray, Jaccard, ANOVA, p=0.001).
ii) Colonic microbiota metabolism, assessing the production of short-chain and medium fatty acids (SCFAs and MCFAs) by GC-MS and deciphering the main pathways of EVOO phenolic microbial metabolites generated over time by HPLC coupled to several detectors: diode array (DAD), fluorescence (FLD) and tandem-mass spectrometry (MS/MS).

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PS3-70 Development of ultrafast PCR assays to detect Artemisia annua and Ambrosia artemisiifolia

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Artemisia annua is well-known herbal medicine which has anti-cancer and anti-oxidant effects. Ambrosia artemisiifolia has very similar morphological characteristics with Artemisia annua and can cause stomach pain when ingested. To discriminate between the two species, ultrafast PCR assays were developed based on a microfluidic chip. Species-specific primer sets for the identification of the two plants were selected targeting the ribosomal protein S12 (rps12) gene, and estimated for their specificity against the two species. The sensitivity of each primer set was independently validated in triplicated and was as little as 0.01 ng of DNA extracted from the two plants. A total of 15 commercial

processed products were tested to evaluate their reliability. The ultrafast PCR assays developed in this study take approximately 20 min, and additionally confirm target species through post-PCR melt curve analysis. Thus, the two species could be rapidly and accurately identified in commercial food products using specific and sensitive ultrafast PCR methods.

Kim, G.S. & Jang, C.S., The Korean Society of Crop Science. 2019. Available from: https://www.cropscience.or.kr.

PS3-71 Simplified LC-MS/MS method for glyphosate and related compounds in oat cereals using a new Carbon HPLC column

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Glyphosate is the most often used herbicide globally. [1] In the US tolerance levels in the ppb to ppm range are set up for glyphosate in various food products. [2] Detection of glyphosate and other polar pesticides is possible without derivatization by employing mass spectrometry. In the past, a variety of different HPLC column chemistries were used in the separation during analysis including ion-exchange and HILIC stationary phases. In many cases, the detection of glyphosate was done under acidic mobile phase conditions utilizing positive ion mode.

In this work, the use of a new carbon-based HPLC column for analysis of glyphosate by MS/MS without derivatization and the validation of this analytical method including the extraction using QuPPe (Quick Polar Pesticides) methodology is presented. The carbon U/HPLC column and ammonium bicarbonate pH 9 mobile phase were used to retain and separate the analytes and allowed a sensitive detection using basic mobile conditions for negative ESI.

The extraction and clean-up of samples using HLB SPE resulted in good recovery of analytes. While using stable-isotope labelled internal standards (SIL IS) for all compounds, the spiked amounts of analytes at both 80 ppb and 800 ppb levels were recovered with accuracy in the range of 80-130% with the reproducibility below 15% RSD against a prepared calibration curve in solvent. The lower quantitation limits for the three analytes (glyphosate, (aminomethyl)phosphonic acid (AMPA) and glufosinate) were determined at 6-11 ppb utilizing a triple-quadrupole instrument. Multiple oat cereal samples were analyzed for the presence of glyphosate, AMPA and glufosinate. Only one of the "organically" labelled cereals contained glyphosate at less than 25 ppb with no detection of other compounds. The cereals not labelled "organic" contained 179-259 ppb of glyphosate and 12-41 ppb of AMPA, but no glufosinate.

[1] Baylis, A. D. 2000. Why glyphosate is a global herbicide: strengths, weaknesses and prospects. Pesticide Management Science, 56: 299-308.
[2] 40 CFR 180.364 - Glyphosate; tolerances for residues.

PS3-72 Characterization of the diurnal pattern of exhaled fatty acids and enteric methane emissions in dairy cows

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Exhaled breath contains hundreds of volatile organic compounds (VOCs) that can reflect on animal physiological processes. Currently, the commonly used methods to assess and evaluate rumen functions are invasive. We thus aim to explore the breath metabolome of dairy cows as a non-invasive technique and characterize their diurnal patterns of rumen fermentation and enteric methane emissions. Enteric methane emissions of 7 lactating cows were measured 8 times over 2 consecutive days using a head chamber system to represent every 3 hours of a day. Simultaneously, exhaled breath samples were collected in Tedlar gas sampling bags. Breath samples were analyzed using a secondary electrospray ionization (SESI) source coupled to a high-resolution mass spectrometry (HRMS) system. SESI-HRMS is a powerful, well-established, and robust analytical technique ideal for in-depth volatile metabolomics characterization offering high sensitivity, fast and accurate analysis. The mass spectra obtained were then processed in Matlab, and volatile fatty acids (VFA) were annotated using their extract m/z ratio. The intensity of short-chain VFA (i.e., acetate, propionate, butyrate) increased immediately after feeding and followed a similar pattern also observed for methane emissions. The initial results from this study revealed a great potential to assess rumen fermentation and health in a non-invasive approach to improve animal welfare.

PS3-73 Methodological and kinetic aspects of Oxygen Radical Absorbance Capacity assay for evaluation of radical scavenging capacity

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The Oxygen Radical Absorbance Capacity (ORAC) assay involves the measurement of the protection afforded by an antioxidant compound to a target molecule (fluorescein (FL), for example) against oxidation by biologically relevant oxygen radical species (R. Apak, 2022). This is one of the most widely used methods for estimating total antioxidant capacity, based on the generation of peroxyl radicals that are relevant in food and biological systems. However, it may be influenced by the existence of secondary reactions, while methodological aspects concerning assay conditions are also variable among studies, often hampering data comparison.

In this context, this work aimed at the critical evaluation of ORAC data and interpretation of different parameters when FL is used as a target molecule through a microchemical, sustainable approach, and at disclosing their meaning as antioxidant capacity indicators (J.R.B. Carvalho, 2023). For this purpose, several antioxidant compounds (caffeic acid, gallic acid, reduced glutathione and quercetin) with different scavenging kinetics towards 2,2'-azobis(2-amidinopropane) (AAPH)-derived radicals were analysed, with monitoring of the intrinsic fluorescence of the target molecule FL over time. The information retrieved by the ORAC curves was disentangled into three main parameters: area under FL decay curve (AUC), lag time and FL decay rate. The influence of ethanol (as a model for organic solvent effect) in these parameters was also evaluated, and the presence of ethanol caused the delay of the reaction for both FL and antioxidant compounds. For complex food samples, namely red wines and orange juices, the Trolox equivalent values, commonly used to express ORAC, were increased

when calculated from AUC than from lag time. These findings stressed the importance of choosing calibrator compounds presenting ORAC kinetics similar to that presented by samples (L.M. Magalhães, 2012) to prevent biased estimation of the antioxidant capacity.

Acknowledgments

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R. Apak, et al., Pure Appl. Chem. 94 (2022) 87. DOI: 10.1515/pac-2020-0902

J. R. B. Carvalho, et al., Antioxidants 94 (2022) 87. DOI: 10.3390/antiox12020505

L. M. Magalhães, et al., Talanta 97 (2012) 473. DOI: 10.1016/j.talanta.2012.05.002

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