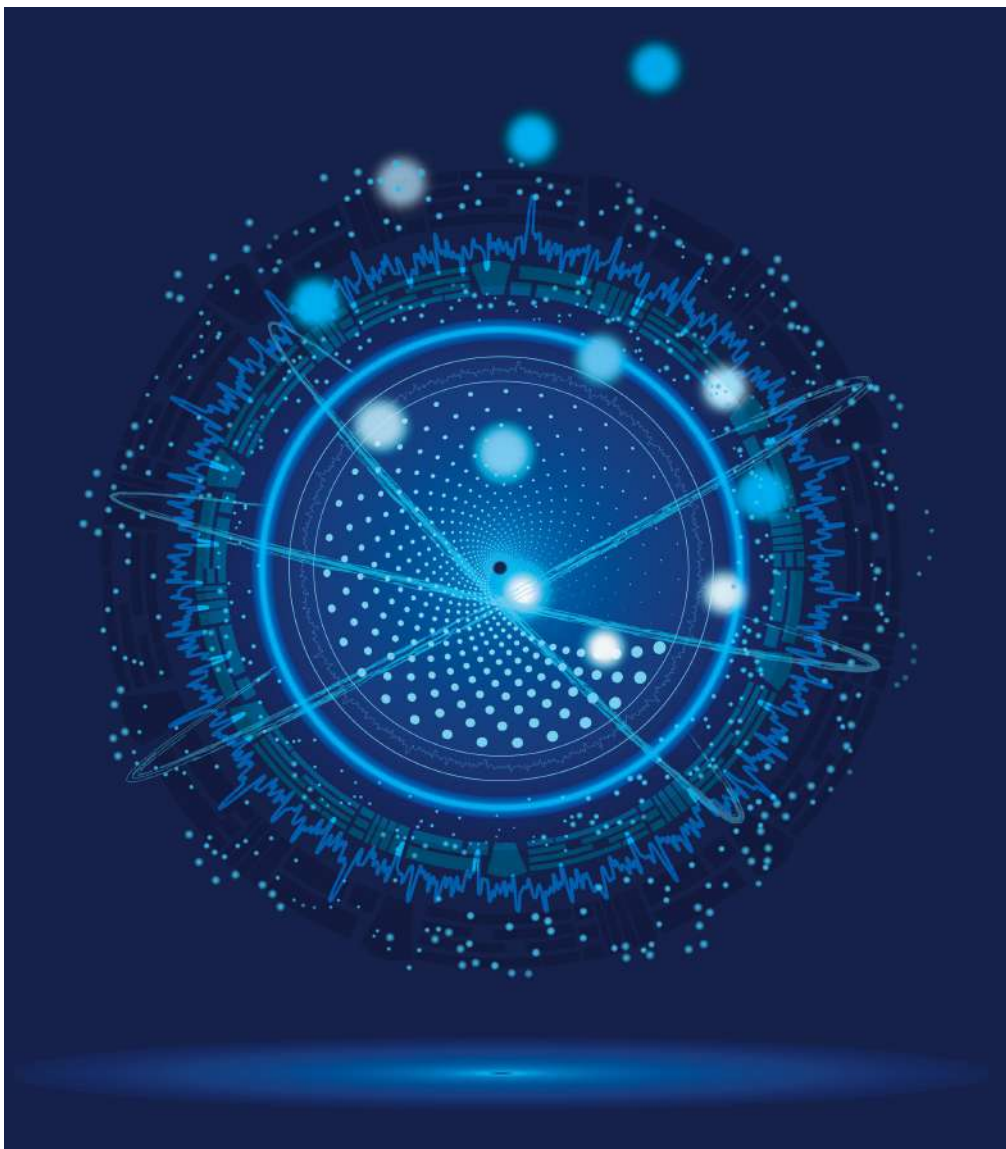


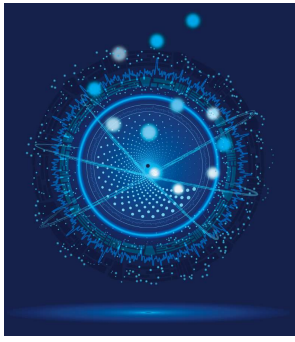
Small New World 2.0

4-5 September 2023

Abstract Book



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Small New World 2.0

4-5 September 2023., Graz, Austria

Joint Meeting of



Austrian Society for Extracellular Vesicles - ASEV
Hungarian Section for Extracellular Vesicles - HSEV
Slovenian Network for Extracellular Vesicles - SiN-EV
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Influence of SEC to resistance of EVs to purity control treatments and stability in different handling/storage conditions

Sofija Glamočlija, Marina Bekić; Alisa Gruden-Movsesijan; Sergej Tomić; Maja Kosanović

Institute for the application of nuclear energy, INEP, Serbia

Introduction: To utilize diagnostic/therapeutic potential of extracellular vesicles (EVs), their integrity should be kept throughout the separation, purification and storage process. However, some techniques, such as size exclusion chromatography (SEC), can affect the corona of EVs and influence both diagnostic/therapeutic molecules and stability of EVs. Therefore, the aim of this work was to investigate whether SEC affects EVs' resistance to purity control treatments and stability in different handling/storage conditions.

Materials & Methods: We separated EVs by ultracentrifugation (UC) on 17000xg (T17) and 100.000xg (T100) and subjected half of obtained EVs to SEC (Sephacryl S400, 10 x 1.2 cm column). In each fraction, the number of particles was determined by NTA, protein concentration by BCA and EVs' marker profile by dot-blot. Fractions containing peak of EVs were pooled. EVs before and after SEC were incubated with different detergents, buffers and were subjected to different temperatures after which the median size and concentration were determined by NTA. Trypsin was used for direct removal of corona.

Results: Most detergents had high number of detected particles and could not be used in NTA/scatter mode. EVs separated by UC only and EVs further purified by SEC were differentially sensitive to temperature and differentially stable in different buffers. Notably, trypsin treatment caused increase in the total number of particles but reduction in median size, possibly indicating disruption of EVs or .

Discussion: Obtained results point out to the importance of quality controls following each step of separation and purification processes and handling/storage.

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