Nutraceutical phycocyanobilin binding to catalase protects the pigment from oxidation without affecting catalytic activity

Abstract

Phycocyanobilin is a dark blue linear tetrapyrrole chromophore covalently attached to protein subunits of phycobiliproteins present in the light-harvesting complexes of the cyanobacteria Arthrospira platensis (Spirulina "superfood"). It shows exceptional health-promoting properties and emerging use in various fields of bioscience and industry. This study aims to examine the mutual impact of phycocyanobilin interactions with catalase, a life-essential antioxidant enzyme. Fluorescence quenching experiments demonstrated moderate binding (K_a of 3.9×10^4 M^{-1} at 25 °C; n = 0.89) (static type), while van't Hoff plot points to an enthalpically driven ligand binding (ΔG = -28.2 kJ mol^{-1}; ΔH = -41.9 kJ mol^{-1}). No significant changes in protein secondary structures (α-helix content ~22%) and thermal protein stability in terms of enzyme tetramer subunits (T_m ~ 64 °C) were detected upon ligand binding. Alterations in the tertiary catalase structure were found without adverse effects on enzyme activity (~2×10^6 IU/mL). The docking study results indicated that the ligand most likely binds to amino acid residues (Asn141, Arg 362, Tyr369 and Asn384) near the cavity between the enzyme homotetramer subunits not related to the active site. Finally, complex formation protects the pigment from free-radical induced oxidation (bleaching), suggesting possible prolongation of its half-life and bioactivity in vivo if bound to catalase.