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Optimization of synthesis methodology for α-chymotrypsin enzyme nanoparticles

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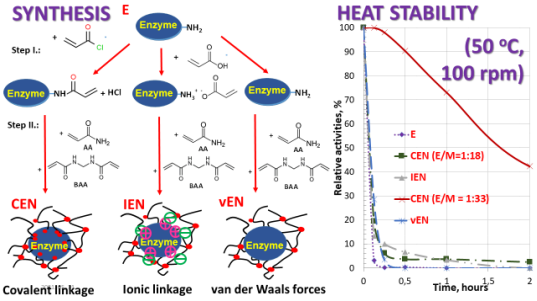
Statement of the Problem: Enzyme nanoparticles represent a new class of nanomaterials with acceptable bio-catalytic activity and very long life-time. Polymerization initiated from the modified surface of the enzyme (see Image, Step I and II) leads to enzyme molecules covered with a polymer layer. These may have at least one order of magnitude longer lifetime than that of free enzymes. However, their bio-catalytic activity is at least about 50% less than that of free enzymes. It is not clear how the size of the polymer layer and the strength of enzyme-polymer bond influence the stability of enzyme nanoparticles.

Methodology & Theoretical Orientation: α-Chymotrypsin enzyme (bovine) was used for the synthesis. Enzyme nanoparticles containing covalent bond (CEN), ionic binding (IEN) and connected with van der Waals forces (vEN) between enzyme molecule and polymer layer are synthesized (see image: SYNTHESIS) and their bio-catalytic activity was investigated at 50 °C (optimal temperature 37 °C). All products were stirred with 100 rpm and samples were withdrawn from time to time for standard activity measurements (ref. 1.)

Findings: The results show that there is a significant difference between the stability of enzyme nanoparticles synthesized by different methods. CEN has the longest lifetime (about 1.7 hour even at 50 °C), when the enzyme:monomer ratio (E/M) during the synthesis is 1:33, but it is the shortest when E/M = 1:18. There is no significant difference between half-lives of E, IEN, vEN and CEN when E/M=1:18 (value is about 0.1 h in all of these cases) (See image: HEAT STABILITY).

Conclusion & Significance: It seems that the minimal amount of E/M for synthesis of efficient product is about 1:30. This ratio results significant enhance of its half-lifetime (ca. 17 times). Enzymes stabilized by polymer layer with ionic and van der Waals interactions could not result in significant stabilization.



Biography

Imre Hegedus has started his research activity at Department of Chemistry and Chemical Informatics, University of Szeged in 2003 and he studied quantum chemistry for modelling the optimal spatial structure of selenocysteine amino-acid. From 2005 he had worked at Research Institute of Biomolecular and Process Engineering at University of Pannonia (Veszprem, Hungary) where he has earned his PhD degree. His main research areas are synthesis and study of enzyme nanoparticles and he applied them as industrial biocatalysts as well as protein drug carriers. He has synthesized dendrimers for application them as drug carriers. Other drug carriers as protein nanoparticles, and nano-emulsions have also been synthesized. From 2020 he is working in Department of Biophysics and Radiation Biology in Semmelweis University, Budapest in the Nanobiotechnology and In Vivo Imaging Centre. His research interests lie in nanoparticle synthesis for clinical therapeutic and diagnostic PET/SPECT applications, and nanomaterial characterization, using atomic force microscopy.