Designed Immobilization of Carbonic Anhydrase for Efficient Enzymatic CO₂ Sequestration

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Carbonic anhydrase (CA) is a prominent biocatalyst used for the enzymatic CO2 capture process. For industrial applications, the immobilization of enzyme on a matrix support is a useful technique for stabilizing the enzyme and improving its reusability. In this study, we investigated the effect of the immobilization position on the stability of CA using α-type Bovine CA (bCAII). Several CA mutants with a single mutation to Cys in a different position were designed for site-specific immobilization on support materials. Enzyme stability is typically related with the structural flexibility of the enzyme. The enzyme structure can be divided into several substructures according to the distinctive flexible features of each domain, and the flexibility of each part of the enzyme could contribute to the structural stability. We decopmosed the structure of bCAII into six substructures via a quasi-rigid domain analysis, and one candidate residue was selected considering the exposure ratio based on solvent accessibility. The selected residues were mutated to Cys for site-specific immobilization, and their effects on the thermostability and long-term stability of the CA were investigated.