Abstract of PhD Thesis

Catalytic Action of Microbial Enzyme DapE and Rational Design of its Potential Inhibitors

by
Debodyuti Dutta
Department of Chemistry, IIT Kharagur, India

The fight against bacterial infections and antimicrobial drug resistance requires unrelenting research efforts in finding new classes of antibiotic targets and their small molecule inhibitors. To this end, the lysine biosynthetic pathway of bacteria offers many potentially safe antibiotic targets due to the fact that this pathway is crucial for bacterial survival while absent in human beings **(Chapter 1)**. In this thesis, the action of a bacterial enzyme (DapE), critical in the lysine biosynthetic pathway, has been investigated using a wide range of computational techniques that include, force field based molecular dynamics simulations, quantum chemical calculations, and their hybrid combination (QM/MM) **(Chapter 2)**. The substrate binding has been shown to significantly influence the global dynamics in the dimeric enzyme DapE. In the DapE-SDAP complex, the two catalytic domains of the dimeric enzyme tend to fold onto the corresponding dimeric domains, leading to a tight binding of the substrate SDAP **(Chapter 3)**. The functional roles of the two Zn metal centres in the enzyme active site have been explained, wherein it is shown that Zn1 ion facilitates the catalytic reaction, while Zn2 ion is necessary for substrate binding **(Chapter 4)**. The presence of the substrate at the active site of the enzyme has been shown to be necessary for the deprotonation of the catalytic water molecule, thereby initiating the hydrolysis reaction. The multistep catalytic hydrolysis reaction follows a general acid- base mechanism, where two steps exhibit proton transfer processes, while the rate determining step involves a nucleophilic attack **(Chapter 5)**. The QM/MM-MD simulations performed under NPT conditions reproduce the experimental energy barriers of the catalytic reaction more accurately than the stationary state QM/MM methods. In the catalytic reaction, the Glu134 residue is found to play a crucial role in facilitating proton transfer and guiding the nucleophile to the site of nucleophilic attack. The His67 and His349 residues coordinated to the Zn ions are shown to be essential for maintaining the structural integrity of the active site, and hence are essential for enzyme catalysis **(Chapter 6)**. On the basis of a combined approach of drug repurposing and structural similarity, the present work proposes a series of captopril based inhibitors that can be potential small molecule inhibitors of the bacterial enzyme DapE **(Chapter 7)**.

Keywords: Antimicrobial Resistance; DapE Enzyme; Hybrid QM/MM; Enzyme Catalysis; Small Molecule Inhibitors