STRUCTURAL DIVERSITY OF PROLINE CATABOLIC ENZYMES REVEALED BY SMALL ANGLE X-RAY SCATTERING, X-RAY CRYSTALLOGRAPHY AND LIGHT SCATTERING

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ABSTRACT

The proline catabolic enzymes catalyze the 4-electron oxidation of proline to glutamate. The reaction involves two enzymes, proline dehydrogenase (PRODH) and Δ^1-pyrroline -5-carboxylate dehydrogenase (P5CDH). Some bacterial organisms have both of these enzymes fused together, and the fused bifunctional enzymes are called Proline utilization A (PutA). In addition to these bifunctional enzymes, some PutAs are trifunctional, because they moonlight as transcription repressors of their own gene. Our lab recently reported that the quaternary structure of the bifunctional PutA from B. *japonicam* (BjPutA) is a ring-shaped tetramer. However, the structural organization of PutAs from other organisms is still unknown. In particular, there are no structures available for moonlighting trifunctional PutAs. We therefore utilized small angle X-ray scattering (SAXS) to obtain the overall shape of a trifunctional PutA from *Escherichia coli* (EcPutA). In addition, rigid body modeling of full-length PutA has been done with the help of SAXS data and crystal structures of DNA-binding and PRODH domains of EcPutA, and BjPutA crystal structure. Unique structural features of PutA have also been explored through multiple sequence alignments and homology modeling using the webservers like ClustalW, Espript, Phyre, and Swiss Model. Finally, the structural basis of HPII disease that is related to disorder in human P5CDH was determined through X-ray crystallographic studies.