LOW RESOLUTION STRUCTURE AND STABILITY STUDIES OF THE PURINE PHOSPHORIBOSYLTRANSFERASE FROM PYROCOCCUS HORIKOSHII USING SAXS

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Purine phosphoribosyltransferases are key enzymes in the salvage of nitrogenated bases for cellular metabolism. Hypoxanthine-guanine-xanthine phosphoribosyltransferase is present as a hexamer in solution, as seen by dynamic light scattering and gel filtration. This represents a new aggregation state with respect to the other 6-oxo purine phosphoribosyltransferases, which form dimers or tetramers. Further studies with SAXS show that the protein forms a hexamer at low concentrations (below 5 mg/ml) and a dodecamer at higher concentrations, above that concentration. A low resolution model (10 Å) obtained from SAXS data show an elongated molecule that corresponds to a dimer of hexamers, as seen in the crystallographic structure deposited in the PDB. Treatment with the chemical agent guanidine hydrochloride shows that low concentrations of the agent (below 2 M) dissociates the dodecamer in hexamers and only at concentrations above 3.5 M the protein begins to unfold, and above 5 M the protein is completely unfolded. The existence of dodecamers does not seem to be of great importance, physiologically, due to the relatively high concentration for the dodecamer to form. Also, the low stability of this form to chemical treatment adds to this conclusion. In any case, it remains to be seen if this dodecamer might have any role in the thermal stability of this protein that comes from a hyperthermophilic organism, living at 98°C.