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Living in a crowded environment: A thermodynamic approach

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During recent decades it has gradually become recognized that crowding can considerably alter the reactivity of individual macromolecules like proteins, both qualitatively and quantitatively. Crowding can be mimicked experimentally by adding high concentrations of inert synthetic or natural macromolecules, termed crowding agents or crowders, to the system *in vitro*. The importance of crowding in protein folding is of particular interest in biophysics. Here, the crowding effect can accelerate the folding process, since a compact folded protein will occupy less volume than an unfolded protein chain. The process of protein refolding *in vitro* has been studied extensively as a mean of understanding how proteins fold inside cells. These experiments are, mainly for practical reasons, commonly carried out in simple buffer system of 20-50 mM with low concentrations of protein (~ 1-2 mg/ml) in order to avoid aggregation during the refolding reactions. A major difference between these idealized conditions and those encountered within cells is that the intracellular environment is highly crowded due to the presence of high concentrations of soluble and insoluble macromolecules in the cytoplasm. This has major thermodynamic and kinetic consequences on the properties of macromolecules present in the cell. These effects can be orders of magnitude different from those in the typical dilute solution used to study proteins in vitro. Biochemical equilibrium in a living cell may be quite different from those under idealized conditions. It is therefore surprising that the effects of macromolecular crowding on protein refolding have been mostly neglected with a few exceptions.

In this study we have created artificially crowded environment through the use of dextran, an artificial molecular crowding agent. Various biophysical techniques including UV Vis spectroscopy, fluorescence, CD, light scattering are used to study the effect of macromolecular crowding on proteins at various pH values. We found that secondary structure of the protein increases at high concentration of dextran. High concentration of dextran also increases the stability of the protein. We could observe increase in T_m, the mid point of denaturation, of the protein in the presence of high concentration of dextran, which is more prominent at low pH.

Biography

Asimul Islam has completed his Ph.D on the topic "Studies of Conformation and Conformational stability of Nef Protein of HIV-1" from Jamia Millia Islamia, a Central University, New Delhi under coloration with International Centre for Genetic Engineering and Biotechnology. He is an Assistant Professor at Centre for Interdisciplinary Research in Basic Sciences, JMI, New Delhi. He has published 10 papers in reputed journals. He has been awarded the Young Scientist Award under Fast Track Scheme by the Department of Science and Technology, India.

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