[C5. S. Longeville] Influence of macromolecular crowding on protein folding and stability: a model for unfolded chain

The cytoplasm, the interior of cells, is filled with a very high quantity of objects with respect to shape and size. In most of the cases, each species is present at rather low concentration but the overall occupied volume fraction can reach Φ~0.3-0.4. The term "crowding" is generally used to describe this environment rather than high concentration which appears less appropriate in this context.

Under crowding environment protein-protein interactions play a fundamental role because the distances between molecules are of the order of few tens of Å. The crowding environment can affect some physical, chemical or biological properties of biological macromolecules [1][2]. The structure and the reactivity can be very strongly modified as a function of inert crowding agents. A particularly interesting aspect concerns the effect of crowding on protein folding and stability. Usually protein folding is studied *in-vitro* at very low concentration. Under such conditions small globular single chain proteins can unfold and refold quite rapidly depending mainly on the nature of the solvent.

The aim of our project is to search for the possible differences between the process of protein folding/unfolding studied *in-vitro* where the protein are very diluted and surrounded by solvent only and the mechanism *in-vivo* where proteins are in very crowded environment.

Theoretically the problem was studied by the introduction of the concept of excluded volume [3]. In a recent paper [4], Minton uses a statistical thermodynamic model to address the question. He predicted that inert cosolutes stabilize the native state of proteins against unfolding mainly by destabilizing the unfolded state and that the dimension of the unfolded state decreases with increasing the concentration of solute in a measurable way.

In a first series of experiment we have measured by SANS the effect of a classically used inert co-solute F70 on the conformation of a deuterated polymer (PEG). We choose a solvent mixture of D_20 and H_20 at the matching point of the F70 in order to observe only the polymer in good solvent, assumed to be a model for an unfolded chain [5].

On the contrary to what is generally assumed chemical interactions can not be neglected leading to partial segregation of the two components but we show that the polymer density is increase when adding F70.

[Collaboration: S. Longeville, LLB, B. Demé ILL]

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[C6. D. Lairez] Phase transition of metastasic extracellular matrix: theory and experiment

The extracellular matrix is a gel made of various macromolecules that isolates organs. In tumour dissemination, invasive cells liquefy the extracellular matrix gel by producing proteolytic enzymes. We study the physical aspects of their actual role in cell invasion: proteinases by hydrolyzing peptide bonds between gel crosslinks, catalyze a phase transition from a gel and solid state to a liquid [1]. A key feature has to be considered: *in vivo*, the enzyme concentration range is so small that enzymes must diffuse within the gel to significantly damage it. Enzyme diffusion introduces space correlations and then controls the gel degradation mechanism [2] and its universality class [3].

Recently [4], a model system consisting in gelatin (denatured collagen) and thermolysin as proteinase was studied at different gel volume fraction, ϕ_{gel}, and enzyme concentration, *[E]*, and varying the solvent viscosity η_r. The degradation time t_c varies as:

$$t_c \mu \eta_r \times \phi_{gel}^{2.50\pm0.05} \times [E]^{-1.46\pm0.07}$$

This result provides clear evidence, which was missing until now, that the gel degradation kinetics is diffusion-limited. We propose a scaling argument and reduced variables for anomalous enzyme diffusion that fully account for experiments. Plotting $[E]\xi_0^3$ as a function of t_c/τ_0 with ξ_0 the mesh size of the gel network and τ_0 the diffusion time of enzyme over this length, allows us to obtain a single master curve independent of ϕ_{gel}. This scaling argument is consistent with self-attracting memory effect on enzyme random walk, i.e. enzyme has some facilities for going back in previously visited area (already damaged gel) rather than for exploring new area (intact gel).

This self-attracting random walk leads to a "Swiss Cheese" model for gel degradation that belongs to the continuum percolation class.

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