Experimental report

01/09/2022

Proposal: 9-13-954 **Council:** 10/2020

Title: Structure and dynamics of elastin-like peptides upon hydrophobic collapse

Research area: Soft condensed matter

This proposal is a new proposal

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Samples: elastin-like peptides in phosphate buffer

Instrument	Requested days	Allocated days	From	To
IN5	3	2	15/06/2021	18/06/2021
D22	1	0		

Abstract:

Elastin-like peptides (ELPs) are artificial polypeptides mimicking the hydrophobic repeat units of the protein elastin which provides elasticity to several biological tissues. One of the main features of ELPs is a hydrophobic collapse upon crossing a lower critical solution temperature (LCST), which induces stimulus-response useful for advanced biomaterials, protein purification and drug delivery. In addition, this hydrophobic collapse is key for the elastic properties of elastin. However, a comprehensive structural and dynamic characterization of the collapse process and the final, collapsed state is still missing, and debated in literature. Using fully hydrogenated and partially deuterated ELP molecules, we aim for a comprehensive understanding of the hydrophobic collapse in ELPs by combining structural, dynamical and thermodynamic signatures using SANS (D22), QENS (IN5), complementary techniques and simulations.
Crucially, the partial deuteration will allow for elucidation of the dynamic contributions of the peptide backbone and side chains. Our study will provide valuable insights into the molecular nature of the hydrophobic collapse of ELPs and its role in bioapplications.

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Scientific background

Elastin is a key protein of the extracellular matrix, and provides elastic properties to biological tissues – such as lung, arteries, skin, ligaments and cartilage – with extraordinary long-term stability and resilience. Besides this fundamental role for life, elastin and the related elastin-like peptides (ELPs) have been intensively and successfully used for biomaterials and biomedical applications such as drug delivery and protein purification in the last decade. ELPs are artificially designed biomolecules mimicking the hydrophobic repeat units in elastin. These repeat units undergo a hydrophobic collapse upon crossing a lower critical solution temperature (LCST), which causes both compaction of individual chains, and association of chains into coacervates (Fig.1). Although key to the elasticity of elastin and the stimulus response of ELPs, a comprehensive mechanistic characterization of the static and dynamicaspects of the collapse is missing. This is due to the difficulties in disentangling dynamical and structural aspects of assemblies, individual molecules and chain motions. The hydrophobic collapse and the related phase transition has been studied in detail for a range of ELP sequences. However, the dynamical state in the collapsed hydrophobic domains is still highly debated, the alternatives being a fluid-like structure 6 versus a specific stacking of beta-turn motifs. In addition to their imminent physiological and biomedical relevance, ELPs also represent models for intrinsically disordered proteins (IDPs) whose coacervation has a profound impact on cellular processes and organization. A mechanistic description of the driving forces behind coacervation in these systems open new opportunities for fundamental understanding and applications.

Experimental results

We measured the identical samples of ELP GVG(VPGVG)$_{42}$ as in a D22 experiment right before this beamtime 9-13-916 for a range of temperatures from 290 to 333K. Data were sliced into an equidistant q array from 0.4 to 1.4 A^{-1}, normalized to the vanadium scattering and reduced for scattering of the sample cylinder and buffer background using standard scripts available at IN5.
As an initial data analysis, we used a single Lorentzian fit to the profile which already results in a reasonable representation of the data. This analysis will be adapted and optimized to include effects of an EISF and a second time scale, thereby carefully evaluating the statistics and potentially emplozing q-global fits.
Analyzing the relaxation rates, we obtain a change in characteristics from a diffusive signature (~q^2) at lower temperature to a jump-diffusive signature at larger temperatures.

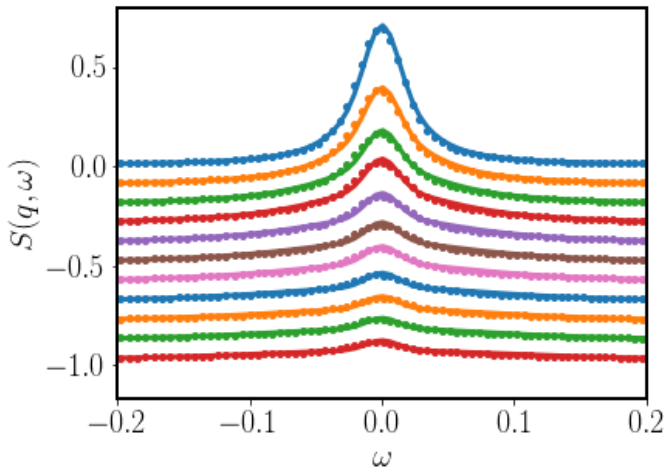

Fig.1: Dynamic structure factor of the ELPs at a temperature of 318 K (symbols) after normalization and background reduction. All data were fitted with a single Lorentzian shape as a first explorative analysis (line).

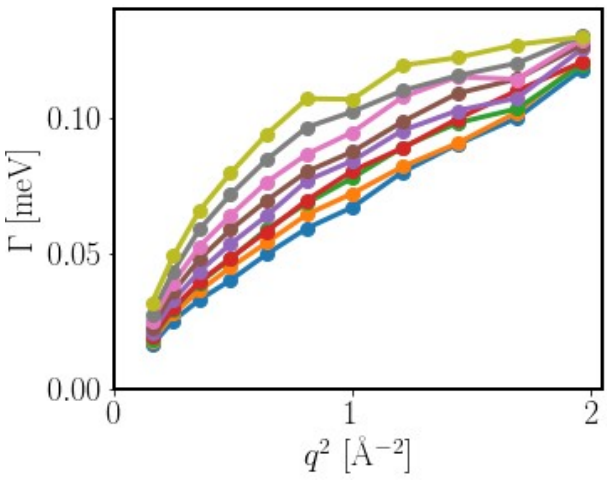

Fig.2: The relaxation rates from the Lorentzian HWHM indicate a change of motion on the local scale from a more diffusive signature at low T (blue) to a more jump-like motion at higher T (yellow).

Conclusions

The preliminary analysis clearly outlines a change of dynamical behavior upon heating beyond the chain collapse transition. However, no abrupt change is observed at the transition. A detailed analysis will provide interesting insights into potential effects of dynamical confinement in this thermoresponsive material.