SAXS analysis of intrinsically disordered protein with a tendency to aggregate

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Small angel X-ray scattering (SAXS) is commonly used for the low-resolution structure analysis of the macromolecules in solution. SAXS is especially useful for intrinsically disordered proteins (IDPs) elongated and flexible chains studies, where other methods fail. We performed SAXS analysis to determine the character of *D. melanogaster* Germ-cell expressed protein C-terminal fragment (GCEC). The GCE, as the juvenile hormone (JH) receptor, mediates JH function in preventing the precocious development of *D. melanogaster* during metamorphosis. What important, GCEC is probably responsible for specific modulation of protein action. During irradiation GCEC experienced severe radiation damage what resulted in aggregation. Since the SAXS scattering signal is a function of molecular weight, this technique is sensitive to the presence of a very small amounts of aggregates, higher oligomers or larger impurities. These significantly affect the measurements results and make it not interpretable. Finally, a qualitative diffraction patterns were obtained for several samples measured directly after protein purification. Initial scans, collected for each of the samples, were combined and used for further analysis. The obtained Kratky plot for GCEC does not present a maximum and reaches plateau at higher values of scattering vector. Such a shape is characteristic for IDPs. The calculated radius of gyration (R_g=56 Å) is significantly higher as 26.5 Å calculated for GCEC with the assumption of globular structure. The maximal intramolecular distance within the molecule (D_{max}) is 174.5 Å. All parameters indicate highly asymmetric and expanded GCEC conformation. Finally, we performed EOM analysis to generate exemplary conformers adopted by GCEC. All determined structures present characteristic bend in the middle of the sequence and significant tangling at both N- and C-termini.

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