SAXS studies of mutants of yeast Sis1: a flexible protein containing some structured domains

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Proteins with well structured domains linked by flexible regions are generally involved in important biological functions. These structural features are often observed in proteins that interact with DNA or RNA and in some chaperones. From the point of view of high resolution structure solving, these proteins pose a real challenge when NMR or crystallographic methods are employed. The existence of flexible regions in the molecules is sometimes a real obstacle. The small angle scattering of X-rays (SAXS) provides low resolution data that in many instances allows the investigation of important details in biological structure-function key problems. In addition to structural information, SAXS data can be used to obtain a low resolution model or molecular envelope for the proteins. In some cases, using existing high resolution structures of parts of the molecule solved by other techniques, it is possible to compose the complete structure. Even though the final result does not reach atomic resolution, the results are important in molecular biology since they may lead to identification of the interaction regions and binding sites. In this communication we will present a study about proteins that fit into the description given above.

In this work we describe results of experiments with mutants of the protein HSP40. In particular, domain deletion mutants of the yeast protein Sis1, whose functional activity is under, study by several biology groups. Several works about the HSP's (Heat Shock Proteins) can be found in the literature [1,2], but not much is known about their structure, principally about their mutants. In this work the following deletion mutants of the Sis1 protein were studied: Sis1_$\Delta_{124\text{-}174}$ (removing the residues 124-174) and Sis1_$\Delta_{121\text{-}257}$ (removing the residues 121-257). The dimensional parameters, such as radius of gyration (Rg) and maximum dimension (Dmax), were determined from SAXS data. The molecular weight was theoretically calculated (Protparam) and also estimated from SAXS data. The oligomeric state was found to be dimeric in each case. *Ab initio* methods using the well known "*dummy atoms*" and "*dummy residues*" routines, lead to the calculation of the average molecular envelopes for the two Sis1 mutants. Rigid body model refinement was performed, using the already solved crystallographic structures of some of the domains previously identified in the amino acid sequence of the proteins. Applying simulated annealing routines and taking into account the dimeric state of these proteins in solution, the low resolution structures of Sis1_$\Delta_{124\text{-}174}$ and Sis1_$\Delta_{121\text{-}257}$ were obtained.

References:

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[2].Sha, B., Lee, S. and Cyr, D.M. *Structure.* 2000, **Vol. 8**, pp. 799-807.

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