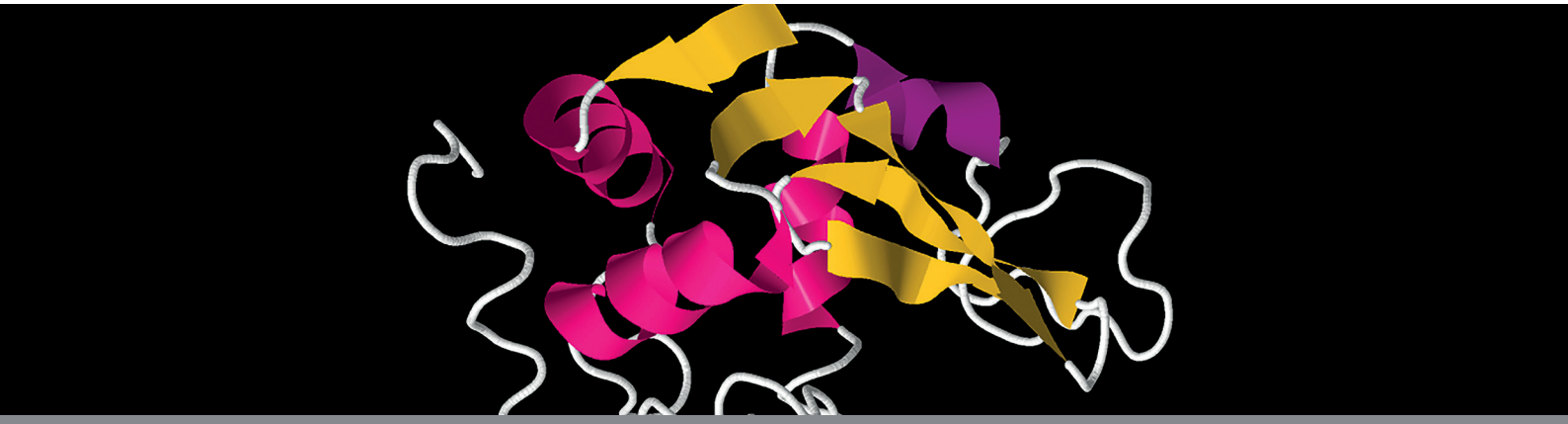

SAXS measurements of diluted solutions

SAXS investigations of Lysozyme solutions were performed enabling the calculation of the radius of gyration (R_g) and of the Pair-distance Distribution Function (PDDF).

Introduction

The Small Angle X-ray Scattering (SAXS) technique is now well known for studying biological systems and more specifically protein solutions. SAXS measurements allow the determination of the macromolecular shape conformation i.e. by an envelope reconstruction of the investigated protein. SAXS data were collected for the standard protein Lysozyme and enable the definition of both the R_g and the PDDF.

Measurements & results

Measurements were performed on 1.5, 3.0 and 5.0 mg/ml concentration solutions with the Xenocs Capillary Flow Cell, using the buffer *40 mM acetic acid 50 mM NaCl pH 4.0.*

c (mg/ml)	Exposure time	Guinier R_g (error) AutoRg[1]
1.5 mg/ml	10 min 30 min	1.36 nm (0.34) 1.41 nm (0.04)
3.0 mg/ml	10 min 30 min	1.43 nm (0.22) 1.40 nm (0.15)
5.0 mg/ml	10 min 30 min	1.38 nm (0.14) 1.39 nm (0.02)

Table 1 - Radius of gyration of Lysozyme depending on the concentration and exposure time.

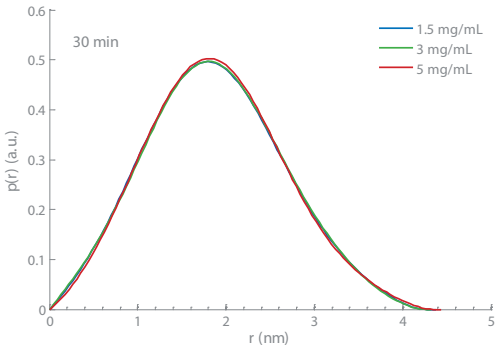

Fig. 1 - Pair-distance distribution function for c = 1.5, 3.0 and 5.0 mg/ml. Exposure time = 30 min.

The structural parameter R_g results were obtained from calculations with the PRIMUS[1] software. The values obtained for each concentration with different exposure times are reported in Table 1. The values are quite consistent with the synchrotron data[2] of R_g = 1.43 nm. Short exposure times of 10 min are sufficient to determine these essential structural parameters.

The PPDF p(r) is calculated using the GNOM[1] software. In Figure 1, it can be observed that the curves obtained from different concentrations overlap. This proves that consistent data can be collected from low concentration measurements.

A comparison of two exposure times for the concentration 5 mg/ml is shown in Figure 2. The curves almost overlap, which is an indication of how a short exposure time of 10 min is enough to provide relevant data.

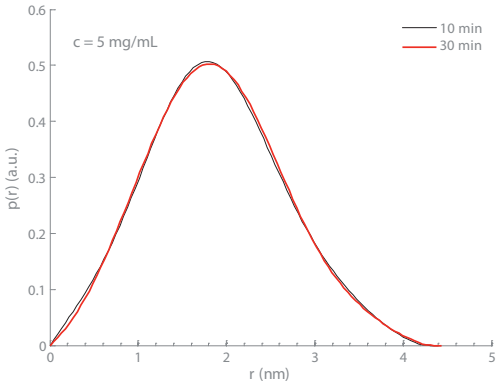

Fig. 2 - Pair-distance distribution function for c = 5 mg/ml. Exposure times = 10 and 30 min.

To go further

The Xenocs clean-beam technology[3] is fully integrated in the Nano-inXider, allowing accurate bio-macromolecular studies of highly diluted systems. Besides, the use of the Xenocs Low Noise Flow Cell reduces the container scattering to push further the limits of BioSAXS measurements in the lab.

[1]Petoukhov et al., J. Appl. Cryst., 2007,40, s223-s2282
[2]Svergun et al, J. Appl. Cryst., 2005, 38, 555–558
[3]TN-XE03 - Unique Signal-to-Noise ratio

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