

Structural study of hNck2 SH3 domain protein by X-ray solution scattering I. Concentration dependence at pH 6.

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Introduction

hNck2 SH3 domain protein takes β -structured conformation at neutral pH. However, Liu and Song found it takes α -helix-rich conformation at acidic pH by means of NMR spectroscopy and CD [1].

We have started the series of conformational study of this protein by means of X-ray solution scattering, and report in four reports; the first report (the present one) shows protein concentration dependent monomer-dimer transition at neutral pH, the second one reports pH-dependent monomer-dimer transition, the third one reports conformational study at pH 2, and the fourth one reports gross structures at pH 2 and pH 8.

Experimental

X-ray scattering experiments were done at the beamline of Bio-SAXS in Stanford Synchrotron Radiation Light Source (SSRL). Experiments were done at pH 6 at 12°C for protein concentration dependent study.

Results and Discussion

Fig. 1 shows dependence of R_g of hNck2 SH3 domain on protein concentration. R_g value was 15.0 Å when the protein was 0.139 mg/mL, whereas R_g was nearly constant (20.8 Å) above 0.35 mg/mL. The former R_g value is in good agreement with that of the native state of src SH3 domain (14.6 Å) [2], whereas the latter R_g values are significantly bigger.

$I(0)$ values were also obtained from Guinier analysis of the same experiments, and normalized $I(0)$ values, $I(0)/c$, were plotted against the protein concentration (Fig. 2). As seen in the figure, $I(0)/c$ value above 1 mg/mL is found to be c.a. twice as big as that at 0.139 mg/mL, indicating that hNck2 SH3 domain structure changed from monomer to dimer.

Fig. 3 shows normalized Kratky plots of hNck2 SH3 domain at various protein concentration. It is clear that all Kratky plots show obvious one peak, indicating that hNck2 SH3 domain was compact globule state at any concentrations both at monomeric and dimeric states.

Acknowledgement

We are grateful to SSRL for giving the beam time in order to do X-ray solution scattering experiments.

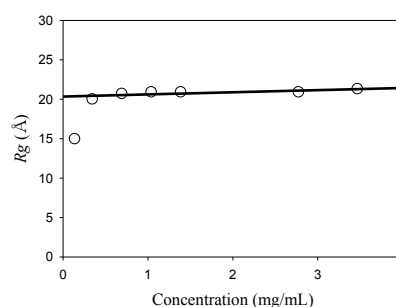


Fig. 1. R_g dependence of hNck2 SH3 domain concentration.

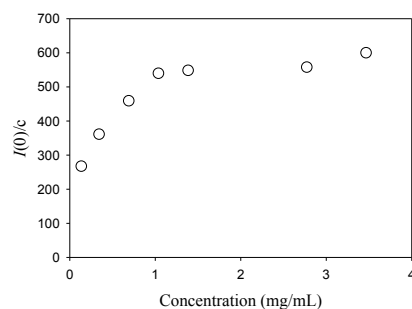


Fig. 2. $I(0)/c$ plots against hNck2 SH3 domain concentration.

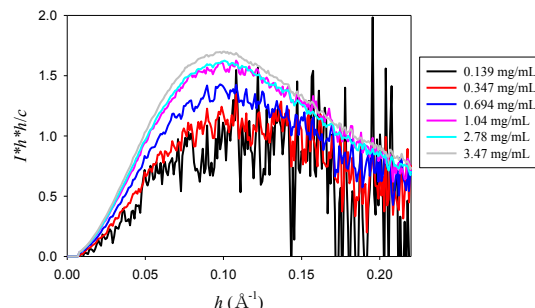


Fig. 3. Normalized Kratky plots of hNck2 SH3 domain.

References

[1] Liu & Song (2008) Biophys. J. 95, 4803-4812.

[2] Li *et al.* (2007) Biochemistry. 46, 5072-5082.

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