SAXS investigation of the Heat shock protein (Hsp70) structure and its interaction with ATP and substrate peptides

I. Eliseev[1], A. Yudenko[1], V. Ukleev[2], A. Ischenko[3], S. Grigoriev[2] and M. Dubina[1]

[1]Saint-Petersburg Academic University, Khlopina str. 8/3, 194021 Saint-Petersburg, Russia

[2]Petersburg Nuclear Physics Institute, Orlova Roscha, 188300 Gatchina, Russia

[3]State Research Institute of Highly Pure Biopreparations, Pudozhskaya str. 7, 197110 Saint-Petersburg, Russia

Heat shock protein (Hsp70) plays major role in chaperoning misfolded proteins and demonstrates immunomodulatory activity through its ability to transport tumor-specific peptides and present them on the cell surface. Structurally Hsp70 molecule consists of N-terminal domain that has ATPase activity and C-terminal domain interacting with other proteins. Although individual structures of separate domains have been solved by X-ray crystallography, their interactions as a whole molecule while operation is still remains not fully clear.

In this experiment we applied small-angle X-ray scattering to investigate Hsp70 conformation in solution as well as its transformations upon the interaction with ATP and substrate peptides.

We found two stages of Hsp70 conformational transitions (see **Fig. 1**): 1) addition of ATP caused compaction of the whole Hsp70 molecule (radius of gyration R_g decreased from 43.3 Å to 41.5 Å); and 2) after the addition of peptides to the Hsp70+ATP complex R_g decreased further to 39.0 Å.

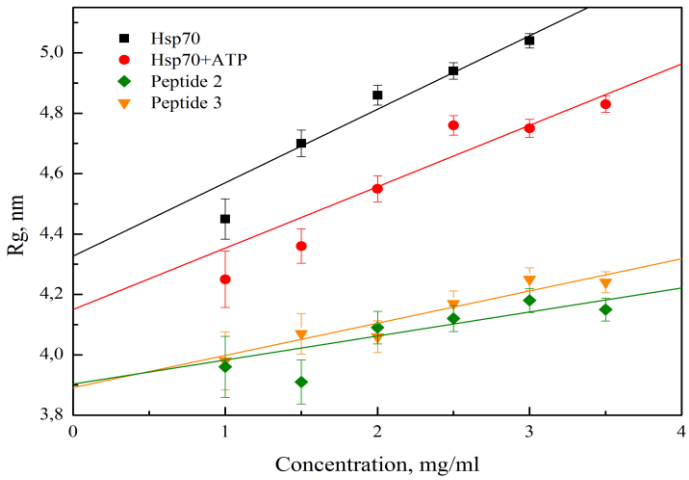

Figure 1 Radius of gyration for Hsp70 and its complexes with ATP and peptides.

The same effect was observed for the diameter of the molecule (D_{max}). The analysis of the pair distribution function (see **Fig. 2**) shows that the domains forms a nearly globular structure after binding with peptide. These results complement the previous SAXS data on Hsp70 in solution [1] and a recent NMR study of the substrate-bound Hsp70 [2].

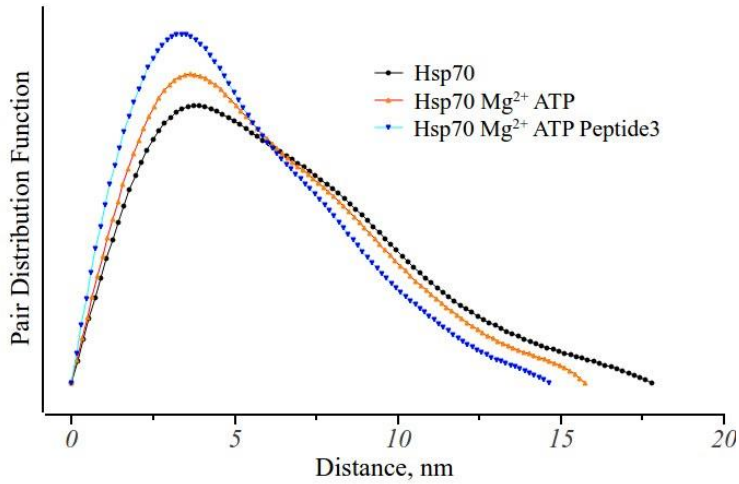

Figure 2 Pair distribution function for Hsp70 and its complexes with ATP and substrate.

References

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[2] E.B. Bertelsen, L. Chang, J.E. Getswicki, E.R.P. Zuiderweg, "Solution conformation of wild-type E. coli Hsp70 (DnaK) chaperone complexed with ADP and substrate", *PNAS*, **106**, 8471-8476 (2009)