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New Physiological Function of Chaperones, Facilitating Reconstitution of Apoenzymes

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One of the important branches of modern biochemistry and molecular biology is the investigation of structure and function of molecular chaperones. The main function of the family of molecular chaperones named as small heat shock proteins (sHsps) is suppression of aggregation of non-native protein species formed under stress conditions or during folding of the newly synthesized polypeptide chains [1-3]. The low molecular mass of monomers (from 12 to 43 kDa) and tendency to form large oligomers with high molecular masses up to 1000 kDa are typical of this protein family [4-14]. The presence of a conservative α-crystallin domain in the structure of sHsp seems to be important for the formation of stable dimers, whereas variable N- and C-terminal ends seem to participate in the formation of large oligomers [2-4,7]. It is supposed that the polydispersity and quaternary structure dynamics play an important role in cellular sHsp chaperones function [2,12]. sHsps cannot provide folding of the polypeptide chain; however, they form complexes with non-native proteins and can transfer the latter either to ATP-dependent chaperones that provide assistance to protein folding or proteasomes, where proteolytic degradation of the unfolded proteins occurs [15-19].

The properties of sHSPs are being studied intensively over the past two decades. However the most of these investigations are carried on in the diluted solutions. In the living cell all the processes proceed in the medium with high concentrations of macromolecules (proteins, nucleic acids, polysaccharides) which occupy the significant part of the cellular volume (up to 40%). That is why the part of the cellular volume becomes inaccessible for proceeding biochemical processes. The term "molecular crowding" implies the effect of excluded volume.

One of the biochemical processes, which proceeds in the cell and may be sensitive to crowding environment, is the process of reconstruction of holoenzyme from apoenzyme and cofactor. Reconstruction of holoform of glycogen phosphorylase b (Phb; EC 2.4.1.1) is a convenient system for the study of the effect of crowding and chaperones on the reconstruction process. Phb in solution exists as a dimer which consists of two identical monomers with a molecular mass of 97.4 kDa. The catalytic site of the enzyme is located in a deep hydrophobic cavity in the center of the subunit; it contains one molecule of covalently bound cofactor pyridoxal 5'-phosphate (PLP), which is necessary for the catalytic activity. Removal of PLP results in the loss of the enzymatic activity of Phb and dissociation of dimer to monomers [20,21]. Monomeric form of apo Phb reveals a high propensity to self-association [20,22-24]. Reconstruction of Phb from apoenzyme and PLP is accompanied by the recovery of the catalytic activity and quaternary structure of the enzyme [20,25]. Therefore PLP can be considered as a catalytic and conformational cofactor of muscle Phb [26]. Since crowding affects protein conformation and processes of self-association of proteins, one would expect that the reconstruction of holo Phb is under control of crowding.

Chebotareva et al. [27] showed that crowding stimulated high-order association of apo Phb. These data agree with the predictions of the theory of molecular crowding and are supported by numerous experiments with various proteins [27-32]. Crowding-induced

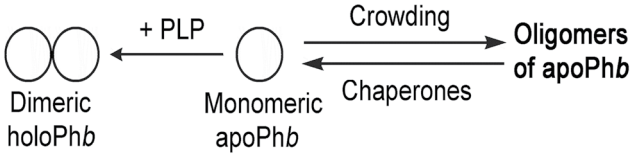

Figure 1: The scheme illustrating new physiological function of chaperones. Chaperones facilitate reconstitution of apoenzyme by preventing its self-association. ApoPhb and holoPhb are the apo- and holoforms of glycogen phosphorylase b, respectively.

association of apo Phb is accompanied by the diminishing of the rate of interaction with cofactor because of steric hindrances. The mobile equilibrium between oligomeric forms of apo Phb was found to be sensitive to chaperones. This was demonstrated for a-crystallin, a representative of the family of sHsps, and for chemical chaperone, proline. Chaperones favour the formation of small oligomeric forms of apo Phb resulting in the acceleration of reconstruction of Phb from apoenzyme and cofactor (Figure1). It is of interest that such a dissociating effect of chaperones was observed with phosphorylase kinase (PhK) from rabbit skeletal muscle. In the presence of Mg2+ and Ca2+ PhK reveals a high tendency to self-association. It has been shown that crowding agent trimethylamine N-oxide greatly favours self-association of PhK, a-crystallin and proline suppressing PhK self-association under crowding conditions [30-32]. Chebotareva et al. [33] showed that interaction of Hsp27 with native PhK under crowding conditions resulted in dissociation of large oligomers of PhK hexadecameric molecules.

Thus, investigations of reconstitution of apo Phb allow putting forward an idea on a new physiological function of chaperones. This function consists in acceleration of reconstruction of holoenzyme from apoenzyme and cofactor by counteracting apoenzyme self-association that becomes especially significant under crowding conditions in the cell.

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References

1. Vos MJ, Hageman J, Carra S, Kampinga HH (2008) Structural and functional diversities between members of the human HSPB, HSPH, HSPA, and DNAJ chaperone families. Biochemistry 47: 7001-7011.
2. Mymrikov EV, Seit-Nebi AS, Gusev NB (2011) Large potentials of small heat shock proteins. Physiol Rev 91: 1123-1159.
3. Basha E, O'Neill H, Vierling E (2012) Small heat shock proteins and Îα-crystallins: dynamic proteins with flexible functions. Trends Biochem Sci 37: 106-117.
4. Van Montfort R, Slingsby C, Vierling E (2001) Structure and function of the small heat shock protein/alpha-crystallin family of molecular chaperones. Adv Protein Chem 59: 105-156.
5. Narberhaus F (2002) Alpha-crystallin-type heat shock proteins: socializing minichaperones in the context of a multichaperone network. Microbiol Mol Biol Rev 66: 64-93.
6. Haslbeck M, Buchner J (2002) Chaperone function of sHsps. Prog Mol Subcell Biol 28: 37-59.
7. Haslbeck M, Franzmann T, Weinfurtner D, Buchner J (2005) Some like it hot: the structure and function of small heat-shock proteins. Nat Struct Mol Biol 12: 842-846.
8. Sun Y, MacRae TH (2005) Small heat shock proteins: molecular structure and chaperone function. Cell Mol Life Sci 62: 2460-2476.
9. Nakamoto H, Vígh L (2007) The small heat shock proteins and their clients. Cell Mol Life Sci 64: 294-306.
10. Kappé G, Boelens WC, de Jong WW (2010) Why proteins without an alpha-crystallin domain should not be included in the human small heat shock protein family HSPB. Cell Stress Chaperones 15: 457-461.
11. Baldwin AJ, Lioe H, Hilton GR, Baker LA, Rubinstein JL, et al. (2011) The polydispersity of ÎαB-crystallin is rationalized by an interconverting polyhedral architecture. Structure 19: 1855-1863.
12. Baldwin AJ, Lioe H, Robinson CV, Kay LE, Benesch JL (2011) ÎαB-crystallin polydispersity is a consequence of unbiased quaternary dynamics. J Mol Biol 413: 297-309.
13. Jehle S, Vollmar BS, Bardiaux B, Dove KK, Rajagopal P, et al. (2011) N-terminal domain of alphaB-crystallin provides a conformational switch for multimerization and structural heterogeneity. Proc Natl Acad Sci U S A 108: 6409-6414.
14. Hilton GR, Lioe H, Stengel F, Baldwin AJ, Benesch JL (2013) Small heat-shock proteins: paramedics of the cell. Top Curr Chem 328: 69-98.
15. Lee GJ, Roseman AM, Saibil HR, Vierling E (1997) A small heat shock protein stably binds heat-denatured model substrates and can maintain a substrate in a folding-competent state. EMBO J 16: 659-671.
16. Ehrnsperger M, Gräber S, Gaestel M, Buchner J (1997) Binding of non-native protein to Hsp25 during heat shock creates a reservoir of folding intermediates for reactivation. EMBO J 16: 221-229.
17. Veinger L, Diamant S, Buchner J, Goloubinoff P (1998) The small heat-shock protein IbpB from Escherichia coli stabilizes stress-denatured proteins for subsequent refolding by a multichaperone network. J Biol Chem 273: 11032-11037.
18. Lee GJ, Vierling E (2000) A small heat shock protein cooperates with heat shock protein 70 systems to reactivate a heat-denatured protein. Plant Physiol 122: 189-198.
19. Wang K, Spector A (2000) α-Crystallin prevents irreversible protein denaturation and acts cooperatively with other heat-shock proteins to renature the stabilized partially denatured protein in an ATP-dependent manner. Eur J Biochem 267: 4705-4712.
20. Hedrick JL, Shaltliel S, Fischer EH (1966) On the role of pyridoxal 5'-phosphate in phosphorylase. 3. Physicochemical properties and reconstitution of apophosphorylase b. Biochemistry 5: 2117-2125.
21. Chebotareva NA, Sugrobova NP, Bulanova LN, Poznanskaia AA, Kurganov BI et al. (1995) Reconstruction of muscle glycogen phosphorylase b from an apoenzyme and pyridoxal-5'-phosphate and its analogs. Interaction of apophosphorylase and the reconstructed enzyme with specific ligands. Biochemistry 60: 2030-2039.
22. Steiner RF, Greer L, Gunther C (1977) Structural changes accompanying the removal of pyridoxal 5'-phosphate from phosphorylase b. Biochim Biophys Acta 494: 233-244.
23. Gunar VI, Sugrobova NP, Chebotareva NA, Stepanova SV, Poznanskaya AA et al. (1991) Synthesis of pyridoxal-5'-phosphate analogs and their interaction with apoenzyme of glycogen phosphorylase b. In: Fukui T, Kagamiyama H, Soda K, Wada H (Eds) Enzymes Dependent on Pyridoxal Phosphate and Other Carbonyl Components as Cofactors. Pergamon Press, Oxford, 417-419.
24. Livanova NB, Chebotareva NA, Eronina TB, Kurganov BI (2002) Pyridoxal 5'-phosphate as a catalytic and conformational cofactor of muscle glycogen phosphorylase B. Biochemistry (Mosc) 67: 1089-1098.
25. Klungsoyr L (1974) Interaction of 8-anilino-1-naphthalenesulfonic acid with holo- and apophosphorylase b. Ligand effects, resolution, and reconstitution with pyridoxal 5'-phosphate. Biochemistry 13: 1751-1757.
26. Chebotareva NA, Eronina TB, Roman SG, Poliansky NB, Muranov KO, et al. (2013) Effect of crowding and chaperones on self-association, aggregation and reconstitution of apophosphorylase b. Int J Biol Macromol 60: 69-76.
27. Hall D, Minton AP (2003) Macromolecular crowding: qualitative and semiquantitative successes, quantitative challenges. Biochim Biophys Acta 1649: 127-139.
28. Chebotareva NA, Kurganov BI, Livanova NB (2004) Biochemical effects of molecular crowding. Biochemistry (Mosc) 69: 1239-1251.
29. Zhou HX, Rivas G, Minton AP (2008) Macromolecular crowding and confinement: biochemical, biophysical, and potential physiological consequences. Annu Rev Biophys 37: 375-397.
30. Chebotareva NA, Andreeva IE, Makeeva VF, Kurganov BI, Livanova NB at al. (2002) Self-association of phosphorylase kinase from rabbit skeletal muscle in the presence of natural osmolyte, trimethylamine N-oxide. Prog Colloid Polym Sci 119: 70-76.
31. Chebotareva NA, Meremyanin AV, Makeeva VF, Kurganov BI (2006) Self-association of phosphorylase kinase under molecular crowding conditions. Prog Colloid Polym Sci 131: 83-92.
32. Chebotareva NA, Meremyanin AV, Makeeva VF, Livanova NB, Kurganov BI (2008) Cooperative self-association of phosphorylase kinase from rabbit skeletal muscle. Biophys Chem 133: 45-53.
33. Chebotareva NA, Makeeva VF, Bazhina SG, Eronina TB, Gusev NB, et al. (2010) Interaction of Hsp27 with native phosphorylase kinase under crowding conditions. Macromol Biosci 10: 783-789.