Effect of sterols on the interaction between crystalline proteins and membranes

Ming-Tao Lee (李明道) 1,2 , Jui-Kai Chen (陳睿凱) 1* , and Jheng-Hao Lin (林政豪) 1
1National Synchrotron Radiation Research Center, 101 Hsin-Ann Road, Hsinchu Science Park, Hsinchu, Taiwan
2Department of Physics, National Central University, Jhongli, Taiwan
mtlee@nsrrc.org.tw

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The cataract, induced by light scattering from eye lens clouding, is the major cause of the blindness in the group of aged people. The most effective treatment of cataract is replacing the clouding eye lens with an artificial one. It not only has a surgical risk but also accompanies with the high medical expense. Thus, to develop a non-surgical and preventive treatment on cataract is important and urgent. Unfortunately, the lack of understanding on mechanism of cataract makes it difficult to be carried out.

In the vertebrate eye lens, alpha-crystallin(α-crystallin) is the major structural protein and consists of two subunits, αA and αB, which are used to maintain lens transparency throughout life. As a member of the small heat shock protein family (sHsp), α-crystallin exhibits chaperone-like activity to prevent misfolding as well as aggregation of key proteins in the lens associated with cataract diseases. It was believed that lens membrane is a key factor to reduce its function and eventually result in cataract[1]. The previous studies reported that binding capacity of α-crystallin to lens lipids increases with age[2], and high molecular complex, comprising α-crystallin and misfolding protein, showed higher association with membrane[1]. Furthermore, some sterols compound can improve lens transparency[3].

To clarify the interaction between membrane and α-crystallin, high purity αA and αB crystallin proteins were expressed from E. coli and purified by affinity and size exclusion chromatography. Size exclusion chromatography experiments showed that both αA and αB crystallin proteins exhibited oligomeric complexes in solution. The influence of membrane on chaperone-like activity of αA and αB were checked by the assays of insulin, lysozyme and alcohol dehydrogenase (ADH). Circular dichroism (CD) was used to monitor the secondary structure changes of crystallin proteins induced by binding to membranes. Finally, lamellar X-ray diffraction (LXD) was used to probe crystallin-induced structural change of membranes. Finally, the effect of cholesterol and ergosterol on the interaction between crystalline proteins and membranes will be discussed.

Keywords –Alpha-crystallin, membrane interaction, lamellar X-ray diffraction (LXD), Circular dichroism (CD).

References

[1] Cobb BA, Petrash JM. alpha-Crystallin chaperone-like activity and membrane binding in agerelated cataracts. Biochemistry. 2002;41(2):483–490.
[2] Vahid Grami, Yernan Marrero, Li Huang, Daxin Tang, Marta C. Yappert, Douglas Borchman. α-Crystallin binding in vitro to lipids from clear human lenses, Experimental Eye Research, 2005;81(2):138-146.
[3] Leah N. Makley, Kathryn A. McMenimen, Brian T. DeVree, Joshua W. Goldman, Brittney N. McGlasson, Ponni Rajagopal, Bryan M. Dunyak, Thomas J. McQuade, Andrea D. Thompson, Roger Sunahara, Rachel E. Klevit, Usha P. Andley, and Jason E. Gestwicki, Pharmacological Chaperone for Alpha-Crystallin Partially Restores Transparency in Cataract Models, Science, 2015;350(6261):674–677.