

STABILITY OF TRIMERS OF DIMERS OF *Np*SR_{II}/*Np*Htr_{II} COMPLEX AT LOW SALT CONCENTRATION

D. D. Kuklina¹, Yu.L. Ryzhykau^{1,2}, V. I. Gordeliy^{1,3,4,5} and A.I. Kuklin^{1,2}

¹Research Center for Mechanisms of Aging and Age-Related Diseases, Moscow Institute of Physics and Technology, 141700 Dolgoprudny, Russian Federation

²Frank Laboratory of Neutron Physics, Joint Institute for Nuclear Research, 141980 Dubna, Russian Federation

³Institute of Biological Information Processing (IBI-7: Structural Biochemistry), Forschungszentrum Jülich, 52425 Jülich, Germany

⁴JuStruct: Jülich Center for Structural Biology, Forschungszentrum Jülich, 52428 Jülich, Germany

⁵Institut de Biologie Structurale Jean-Pierre Ebel, Université Grenoble Alpes–Commissariat à l’Energie Atomique et aux Energies Alternatives–CNRS, F-38027 Grenoble, France

E-mail: kuklina.dd@phystech.edu

The subject of conducted research is a protein complex of sensory rhodopsin II (SR_{II}) and its cognate transducer (Htr_{II}) from archaea *Natronomonas pharaonis*. The complex represents a family of TCS (Two-Component Systems), which consist of a transmembrane receptor and adaptor proteins attached at their cytoplasmic tips. TCS are the most common signaling systems in all domains of life, however are absent in mammalian cells. This fact forms the interest in clarification of signal pathways through the bacterial membrane.

*Np*SR_{II}/*Np*Htr_{II} mediates negative phototaxis in halobacterial *N. pharaonis* and, similarly to chemoreceptors, the *Np*SR_{II}/*Np*Htr_{II} complex forms trimers of dimers in the *N. pharaonis* membrane [1]. *N. pharaonis* grows optimally at 3.5 M NaCl. It has been shown that structure and oligomerization state of the *Np*SR_{II}/*Np*Htr_{II} strongly depend on salt concentration [2]. In a previous research [3,4] small-angle neutron scattering (SANS) measurements were performed on the YuMO spectrometer (IBR-2, Dubna, Russia) with two-detector system [5,6]. It has shown that only at high salt concentrations the *Np*SR_{II}/*Np*Htr_{II} complex is able to form trimers of dimers, while at low salt concentrations the scattering curve is perfectly fit by dimers.

In this research, we present the result of chromatographic analysis, showing the stability of trimers of dimers. The fraction of trimers of dimers was transferred into low-salt buffer by gel-filtration. Further analysis by size exclusion chromatography shows that the protein is eluting at the same volume position as the trimers of dimer. It has proved the stability of trimers of dimers of the *Np*SR_{II}/*Np*Htr_{II} at low salt concentration even on time scales of several days.

The discovered stability of oligomeric state of trimers of dimers at low salt conditions can be used for the following studies in procedures that work better with low-salt buffers, such as reconstitution to nanodiscs or lipid vesicles, preparation of grids for cryo-EM studies, etc.

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