

Bicelles as versatile platforms for rhodopsin in biophysical applications

Patryk Bielski^{1*}, Justyna Bożek², Valerio Marino³, Daniele Dell'Orco³, Carsten Dosche², Izabella Brand^{2,4}, Karl-Wilhelm Koch^{1,4}

¹Carl von Ossietzky Universität Oldenburg, Department of Neuroscience, 26129 Oldenburg, Germany,

²Carl von Ossietzky Universität Oldenburg, Institute of Chemistry, 26111 Oldenburg, Germany,

³Department of Neurosciences, Biomedicine and Movement Sciences, Section of Biological Chemistry, University of Verona, 37134 Verona, Italy,

⁴Neuroscience Research Center for Neurosensory Sciences, Carl von Ossietzky Universität Oldenburg, 26111 Oldenburg, Germany,

corresponding author email: patryk.bielski@uni-oldenburg.de

*Presenting author

1. Introduction

Rhodopsin is localised within the lipid bilayers of the photoreceptor outer segments discs and is one of the best-characterized membrane proteins that plays a crucial role in processing the light signal into visual information^[1]. The lipid composition of the disc membrane in rod outer segments influences rhodopsin activity by modulating its stability, kinetic properties, and functional performance^[2]. However, the correlation between the lipid composition and rhodopsin functionality is not well understood. Bicelles, disc-shaped lipid aggregates capable of forming a single lipid bilayer, can provide a compelling medium for investigating this relationship^[3].

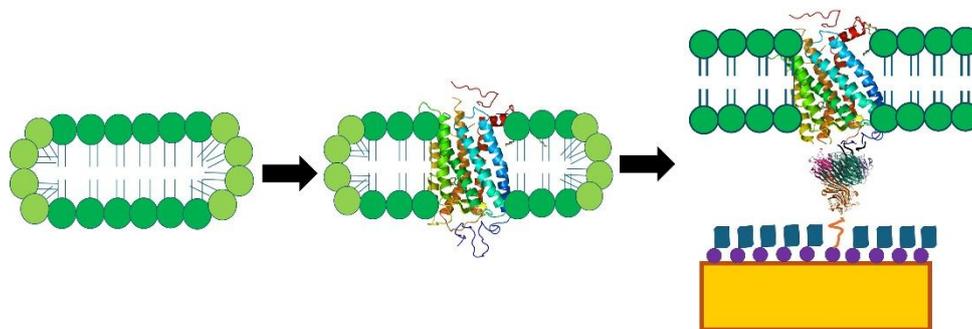


Fig. 1. Schematic model of DMPC/DHPC bicelles with inserted rhodopsin and spreading of bicelles containing rhodopsin on a gold surface, protein structures from PDB.

2. Methods and results

In our research, we characterized the structural properties of rhodopsin reconstituted in bicelles. The size difference of bicelles with and without rhodopsin was confirmed by Dynamic Light Scattering (DLS) measurements, and they were captured by transmission electron microscopy (TEM). Using infrared (IRS) and circular dichroism spectroscopy (CDS), we found that rhodopsin forms a more packed structure compared to the detergent-solubilized condition. Additionally, the bicelle-reconstituted rhodopsin forms exhibited increased thermal stability. The capability to form bilayers starting from bicelles incorporating rhodopsin was investigated by Polarization-Modulation Infrared Reflection-Absorption Spectroscopy PM IRRAS analysis, which revealed that the application of transmembrane potential induces pronounced conformational and orientational changes within the helical bundle of rhodopsin. We also immobilized rhodopsin in bicelles on a sensor chip surface via concanavalin A anchoring and checked the efficiency of binding to the G protein transducin in comparison to the detergent environment by Surface Plasmon Resonance (SPR).

3. Acknowledgement

We acknowledge financial support from the Deutsche Forschungsgemeinschaft (DFG) to IB (BR 3961/13-1; project number: 510809665) and KWK (KO 948/18-1; project number: 510809665), DFG grant (INST 184/ 106-1 FUGG) (HR TEM). Project „MIX IT" Erasmus+ Traineeships for students/graduates; projekt-code: 2024-1-DE01-KA131-HED-000220537.

4. References

- [1] Hargrave, P. A., McDowell, J. H. "Rhodopsin and phototransduction: a model system for G protein-linked receptors". *FASEB journal* 6(6), 2323–2331. (1992).
- [2] Senapati S, Poma AB, Cieplak M, Filipek S, Park PSH. „Differentiating between Inactive and Active States of Rhodopsin by Atomic Force Microscopy in Native Membranes". *Analytical Chemistry*. 91(11):7226-7235. (2019).
- [3] Bożek, J., Dziubak, D., Grempeka, A., Sek, S., and Brand, I. (2025). Monitoring of Bicelles Spreading into Floating Lipid Bilayers *Langmuir* 41, 20942-20953 10.1021/acs.langmuir.5c02620