Pure protein membranes made from fungal hydrophobins: Assembly and mechanical properties

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Compartmentalization of an aqueous solution is of utmost importance in biology. Typically, the matrix of the membranes forming the compartments is a phospholipid bilayer. Thereby, the amphiphilicity of the lipids is necessary for bilayer formation. Yet, for applications in, e.g., biomedicine or synthetic biology, phospholipids are limited in their variety in mechanical and biochemical properties and thus, alternative building blocks are needed. Proteins are promising candidates due to their biocompatibility and versatility via genetic engineering. A special family of strongly amphiphilic proteins, hydrophobins, appears to be particularly suited. In this study, we utilize fungal hydrophobins which self-assemble at water-interfaces into stable monolayer films. Contacting two interfacial films, stable bilayer membranes resembling lipid bilayers can be produced [1]. We study the assembly process of the monolayers and the mechanical properties of mono- and bilayers. At the interface, the proteins organize in clusters in which the proteins obtain a crystalline order [2]. In AFM membrane stretching experiments, we determine the elastic modulus of these monolayer films. We find a remarkably high value, very likely owing to the high cohesion in the 2D crystal structure. Yet, in the bilayer form, the modulus seems to be reduced, hinting at a protein reordering during bilayer formation.

[1] Hähl, H. *et al., Adv. Mater* 29, 1602888 (2017).

[2] Hähl, H. *et al.*, *Langmuir* 35, 9202 (2019).