Lipid-free, pure-protein bilayers and vesicles from native fungal hydrophobins

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The compartmentalization of an aqueous solution by semi-permeable membranes is of utmost importance in biology. Typically, the matrix of these membranes is composed by bilayers made from phospholipids. Thereby, the amphiphilic character of the lipids is necessary for bilayer formation. Since phospholipids are limited in their variety in mechanical and biochemical properties, alternative building blocks are sought for specialized applications. Proteins seem to be ideal candidates promising biocompatibility and versatility via genetic engineering. A special type of amphiphilic proteins, hydrophobins, appears to be particularly suited. These proteins occur naturally in filamentous fungi being involved in, e.g., sporulation or adhesion. In this study, bilayers made purely from hydrophobins were created using a microfluidic platform. The ability of these bilayer to be formed between any type of fluid compartments, be it gas, water, or oil is demonstrated, which renders hydrophobins much more versatile than lipids. Via microfluidic jetting, vesicles were formed from these different types of bilayers. In the case of vesicles in aqueous surrounding, gramicidin-A ion channels could be inserted into the bilayer allowing the transport of monovalent ions [1]. Thus, these vesicles are the first example of vesicles with lipid-free, artificial bilayers containing inserted functional proteins.

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