# Facial recognition of biological lipid membranes

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The present field of targeted drug design is predominantly protein-centric not just because of historical reasons but especially because of the persisting lack of an efficient design platform for drugs that enable selective targeting of biological membranes beyond the level of molecular structure. Design of (peptide) drugs able to selectively target distinct, collective structural features in biological lipid membranes (e.g., leaflet curvature and lipid composition) is severely challenged by the dynamic, disordered fluid nature of lipid membranes, i.e. its fluid interface, rendering existing molecular structure-based peptide drug design strategies ineffective. Our evolutionary molecular dynamics (evoMD) approach [1] overcomes these current limitations by 'learning' peptides how to optimally recognize distinct fluid membrane interfaces via evolutionary optimization (artificial intelligence) directed by highly efficient free energy calculation approaches. What makes this inverse design approach particularly valuable is that the obtained insights can simultaneously gain an unique mechanistic understanding on how native proteins recognize membrane interfacial features such as membrane curvature or lipid composition and especially how protein-membrane interactions have determined the evolution of these proteins. To demonstrate the utility of the evoMD method, we will first resolve the trans-membrane domain sequence which maximally attracts/clusters cholesterol [1]. Surprisingly, the global solution features an unusual short hydrophobic block, consisting of typically only eight short chain hydrophobic amino acids, surrounded by three successive lysines. We will discuss the underlying molecular mechanism of cholesterol attraction and compare our findings with recent experimental results [2] as well as the proposed cholesterol recognition amino acid consensus (CRAC) motif [3]. Last but not least, we will demonstrate the utility of evoMD in the targeted design of broad-spectrum antiviral peptide drugs.

[1] J. Methorst, N. van Hilten, Herre Jelger Risselada, Inverse design of cholesterol attracting transmembrane helices reveals a paradoxical role of hydrophobic length, bioRxiv 2021.07.01.450699; doi: https://doi.org/10.1101/2021.07.01.450699

[2] Lorent, J.H., Diaz-Rohrer, B., Lin, X. *et al.* Structural determinants and functional consequences of protein affinity for membrane rafts. *Nat Commun* **8,** 1219 (2017). https://doi.org/10.1038/s41467-017-01328-3

[3] Fantini J, Barrantes FJ. How cholesterol interacts with membrane proteins: an exploration of cholesterol-binding sites including CRAC, CARC, and tilted domains. *Front Physiol*. 2013;4:31.