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Translocation-Enhancing Peptides: Facilitate Transport of Molecules and Lipid Scrambling

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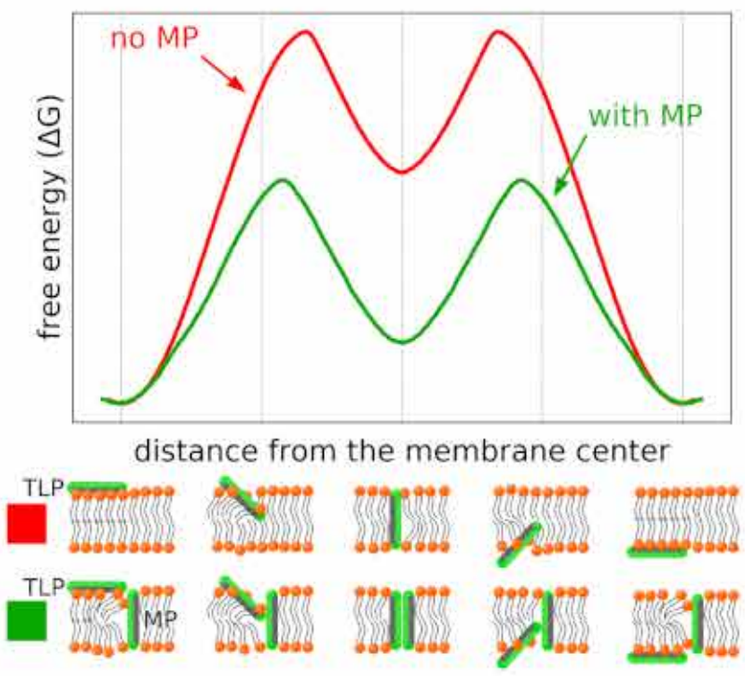
Motivation

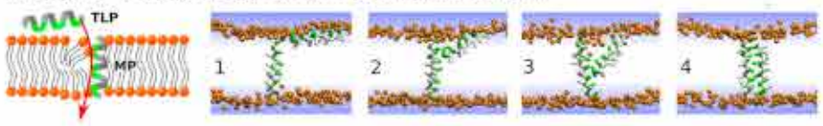
- Despite textbook knowledge that only small molecules can spontaneously cross cell membrane, amphiphilic molecules such as cell penetrating peptides were observed to do the same. Their translocation mechanism remains elusive[1-3]
- Scramblases are proteins facilitating bidirectional & energy-independent transport of phospholipids between membrane leaflets[4]
- Insertases facilitate membrane insertion of proteins by local membrane defect, which also facilitates lipids scrambling[5]



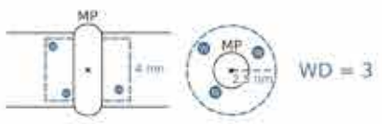
Methods

Molecular dynamics simulations:

- NPT molecular dynamics using Gromacs package versions 5.1.4 and 2021.4 [6]
- MARTINI 2.2 [7] with downscaled LJ interactions between protein beads[8]
- Lipid scrambling also all-atom with Slipids[9] and Amber 99SB-ILDN[10]
- Membrane composed of POPE:POPG 3:1
- Free energy calculations using umbrella sampling technique with collective variable including distance between peptide end and local membrane center of mass[11] for scrambling the lipid was pulled by phosphorus atom



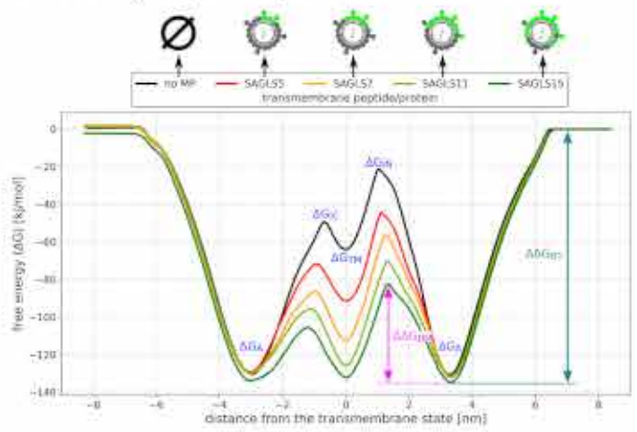
- water defect is average number of water beads in a cylinder around the transmembrane peptide



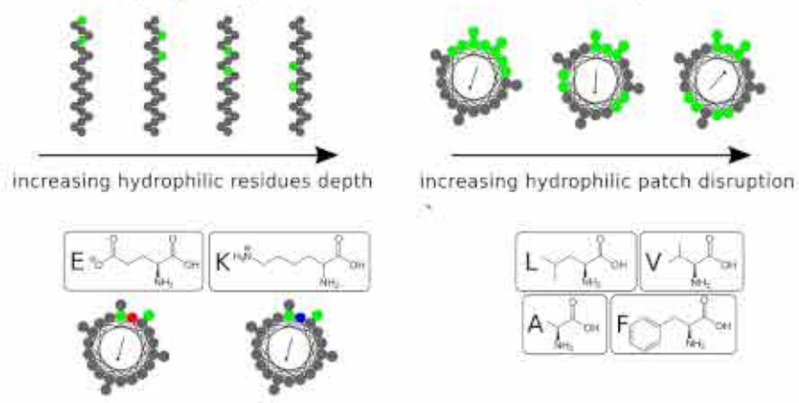
Results

Properties of transmembrane peptide affect the translocation barrier

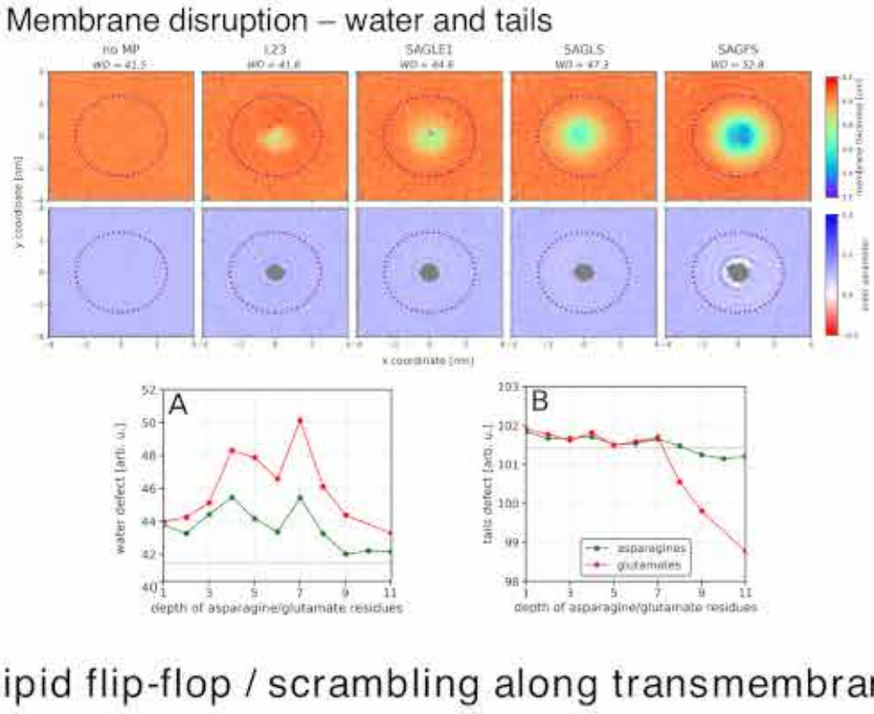
Size of hydrophilic patch[12]



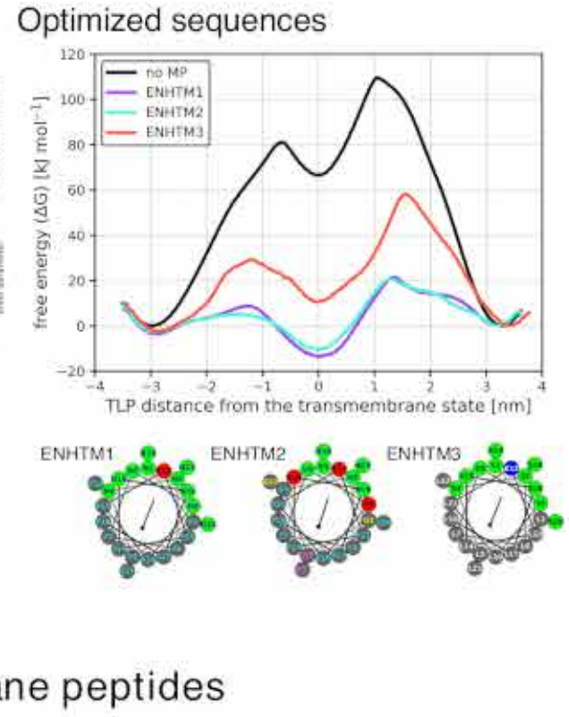
Patch depth, compactness and residue composition



Membrane disruption – water and tails

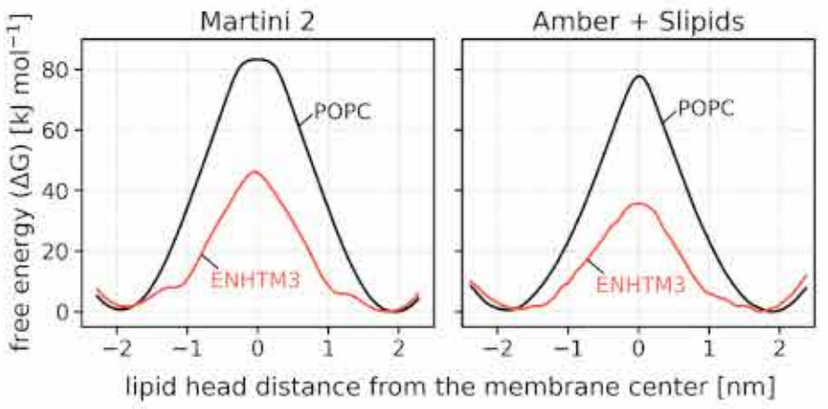


Optimized sequences



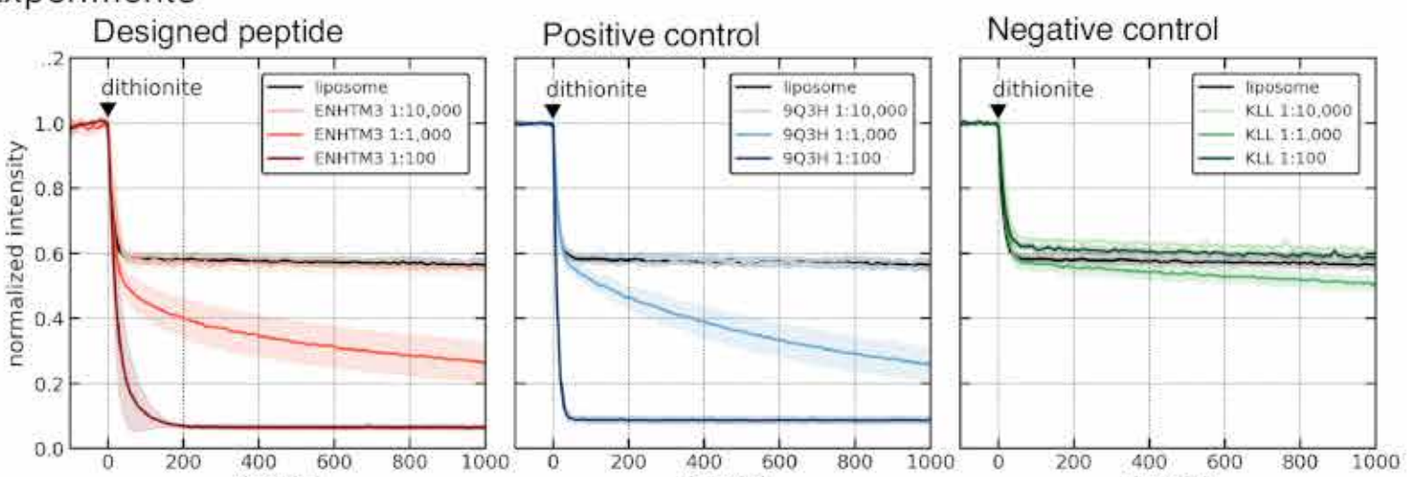
Lipid flip-flop / scrambling along transmembrane peptides

Simulations



Coarse-grained
All-atom

Experiments



Conclusions

- transmembrane proteins/peptides with a hydrophilic patch enhance the translocation of amphiphilic molecules (e.g. peptides and lipids) by stabilizing them in the membrane[13]
- local membrane disruption is mainly responsible for the translocation enhancement
- transport of amphiphilic molecules in cells could be enhanced by native scramblases and insertases or artificial translocation-enhancing peptide

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