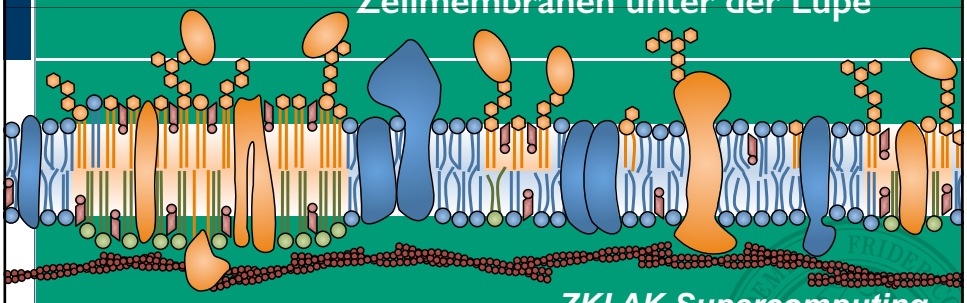
Biomolekulare Simulationen: Zellmembranen unter der Lupe

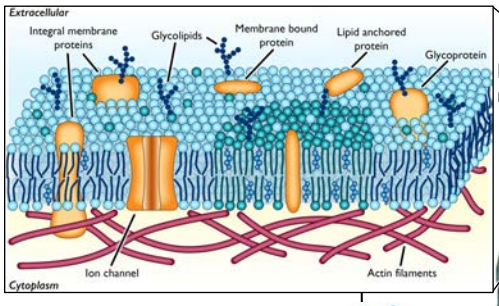


ZKI AK Supercomputing
September 10-11, 2020

Rainer Böckmann
Computational Biology

1

Biological cells and membranes



All living organisms consist of single or multiple units: *Cells*

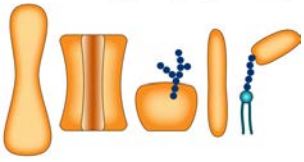
Plasma membrane

Membrane composition:

Lipids

hydrophilic
hydrophobic

) µm

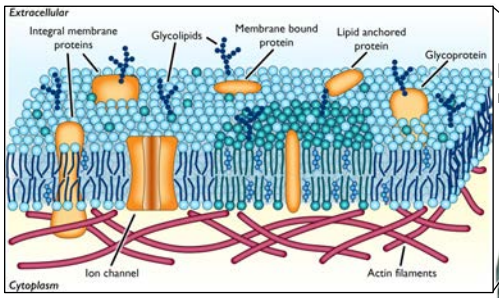


Proteins

by Russel Kightley

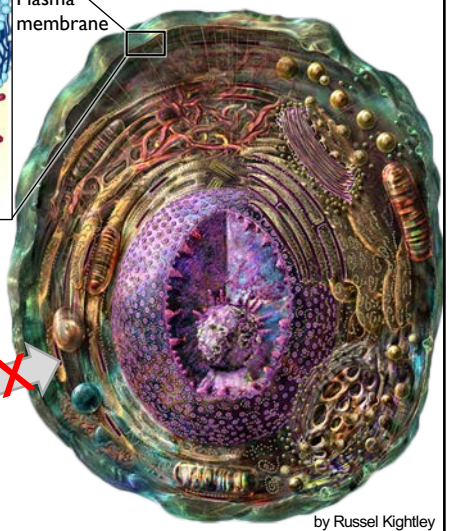
2

Biological cells and membranes



All living organisms consist of single or multiple units: ***Cells***

Plasma membrane



by Russel Kightley

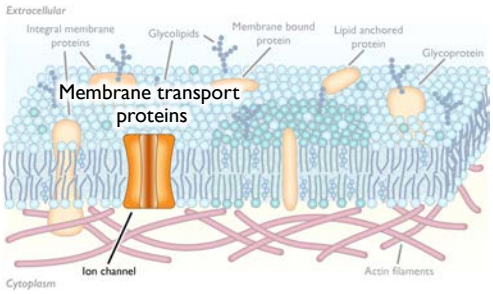
Membrane function:

- Protection of the cell interior from environment
- Intracellular compartmentalization

→ Barrier for macromolecules, charged compounds and mostly water

3

Membrane permeation



Cell survival requires exchange of materials

Signaling between and within cells involves passage of ions across membranes

Membrane function:

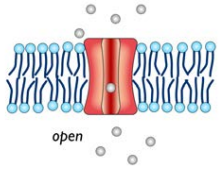
- Protection of the cell interior from environment
- Intracellular compartmentalization

→ Barrier for macromolecules, charged compounds and mostly water

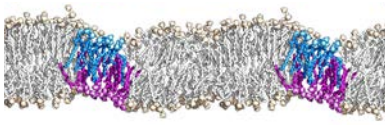
4

Outline

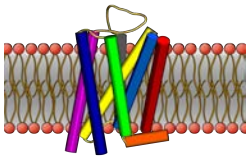
Transport *Across* Membranes: Two Pore Channels (TPC)



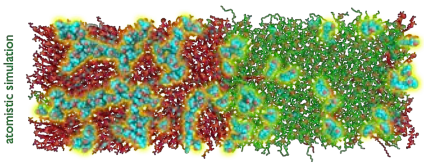
Domain Formation in Simple Phospholipid Membranes



Signaling *Through* Membranes: G Protein Coupled Receptors (GPCR)

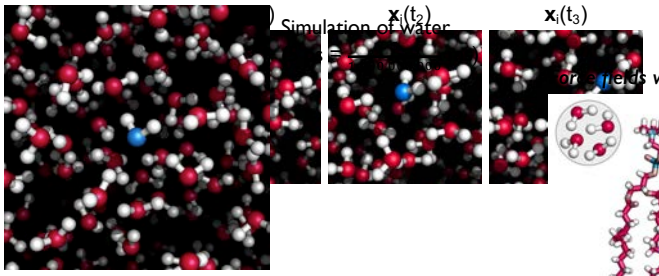


Domain Formation in Multi-Component Membranes



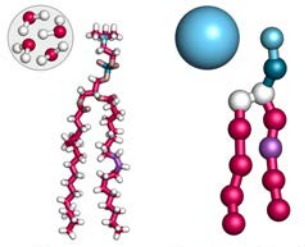
5

Molecular dynamics simulations



Simulation of water

Force fields with different resolutions:



Loss of detail
Gain of performance

Newton's law

$$\mathbf{a}_i(t) = \frac{\partial^2 \mathbf{x}_i(t)}{\partial t^2} = \frac{\mathbf{F}_i}{m_i} \qquad \mathbf{F}_i = -\frac{\partial V(\mathbf{x}_1, \mathbf{x}_2, \ldots, \mathbf{x}_N)}{\partial \mathbf{x}_i}$$

Force field

$$V(\mathbf{x}) = \left(\sum_{bonds} V_{bonds} + \sum_{angles} V_{angles} + \sum_{torsions} V_{torsions} + \sum_{improper} V_{improper} \right)_{bonded} + \left(\sum_{pairs\ i,j} V_{electrostatic} + \sum_{pairs\ i,j} V_{van\ der\ Waals} \right)_{non-bonded}$$

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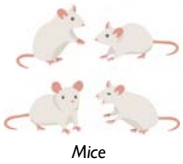
Transport *Across* Membranes: Two Pore Channels (TPC)

Humans

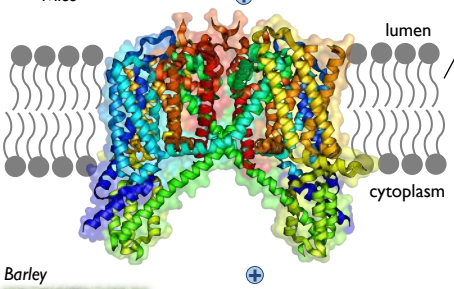
hTPC2

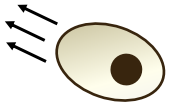


Mice

Zebra fish

Animals: endo-lysosomal membrane
Plants: vacuolar membrane





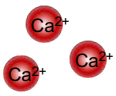
Cancer cell migration [1]

Ebola virus infection [2]

Arabidopsis thaliana

AtTPC1

Barley



Ca^{2+} signalling [3,4]

[1] O. Nguyen et al. Cancer Res. 77:6, 1427-1438 (2017)
[2] Y. Sakurai et al. Science 347:6225, 995-998 (2015)
[3] W. Choi et al. PNAS 111:17, 6497-6502 (2014)
[4] P. Calcraft et al. Nature 459:7246 596-600

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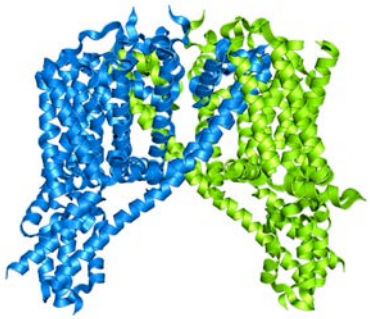
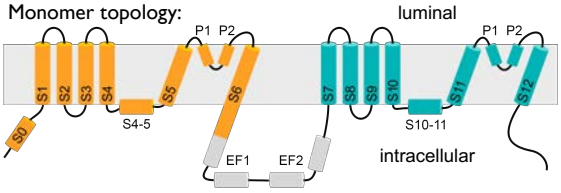
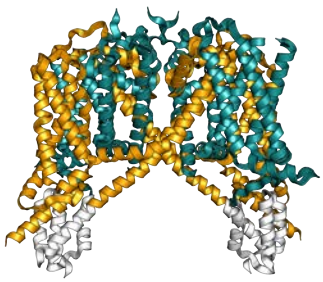
Structure of Two Pore Channels



TPCs are homo-dimers
(2 x S1-S12)

Monomer topology:

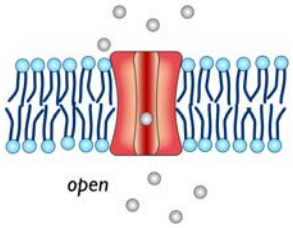




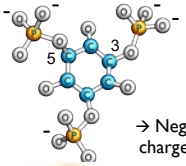
8

hTPC2 is activated by PI(3,5)P_2 – AtTPC1 is not

PI(3,5)P_2 activates hTPC2 [1,2]



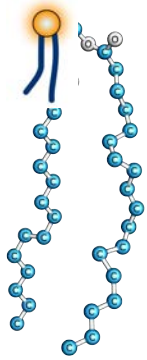
Signaling lipid:
Phosphatidylinositol-3,5-bisphosphate
(PI(3,5)P_2)



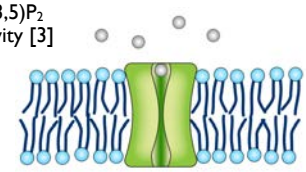
→ Negatively charged lipid

Why does PI(3,5)P_2 activate hTPC2, but not AtTPC1 ?

??



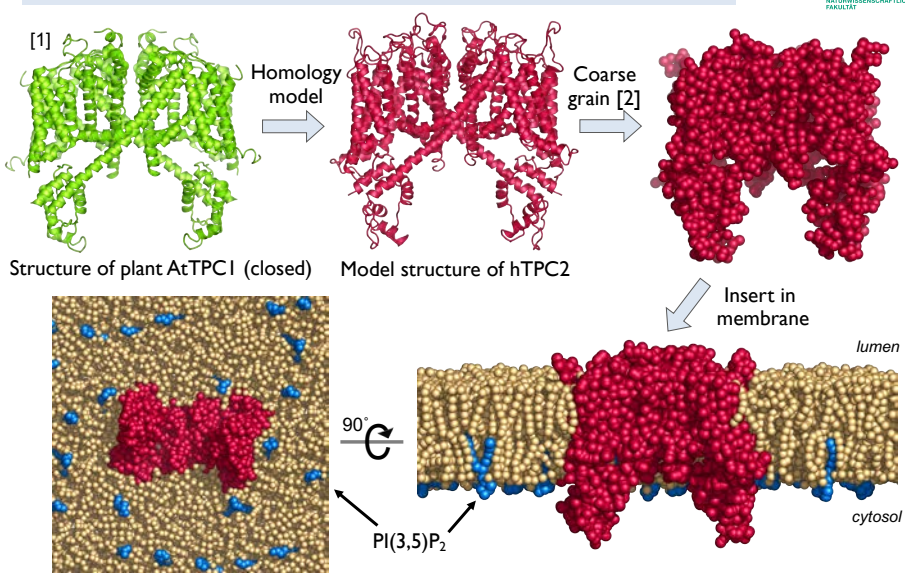
No effect of PI(3,5)P_2 on AtTPC1 activity [3]



[1] J. She, W. Zeng, J. Guo, Q. Chen, X. Bai, and Y. Jiang. eLife 8:e45222 (2019)
[2] X. Wang, X. Zhang, X.P. Dong, M. Samie, X. Li, X. Cheng, A. Goschka, D. Shen, Y. Zhou, et *al.* Cell 151 (2012)
[3] A. Boccaccio, J. Scholz-Starke, S. Hamamoto, N. Larisch et al., Cell. Mol. Life Sci. 71:4275-4283 (2014)

9

Simulation setup to investigate PI(3,5)P_2 binding



[1] J. Guo, W. Zeng, Q. Chen, C. Lee, L. Chen, Y. Yang, C. Cang, D. Ren, and Y. Jiang. Nature 531 (2016)
[2] S.J. Marrink, A.H. Vries, and A.E. Mark. J. Phys. Chem. B 108 (2004)

10

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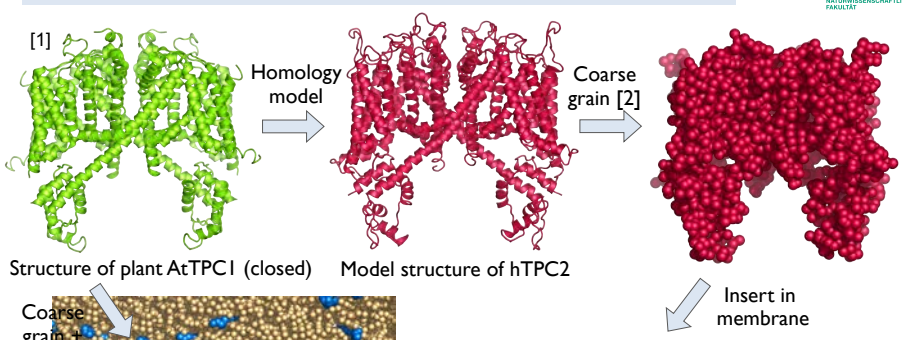

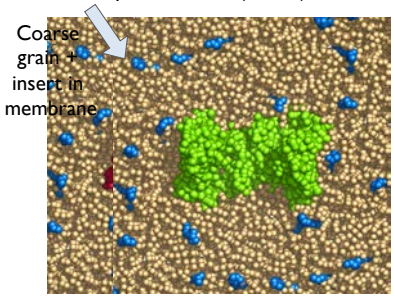

[1] J. Guo, W. Zeng, Q. Chen, C. Lee, L. Chen, Y. Yang, C. Cang, D. Ren, and Y. Jiang. Nature 531 (2016)
[2] S.J. Marrink, A.H. Vries, and A.E. Mark. J. Phys. Chem. B 108 (2004)

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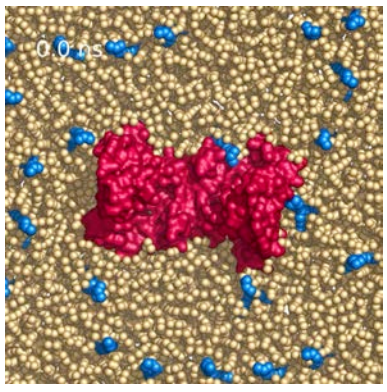
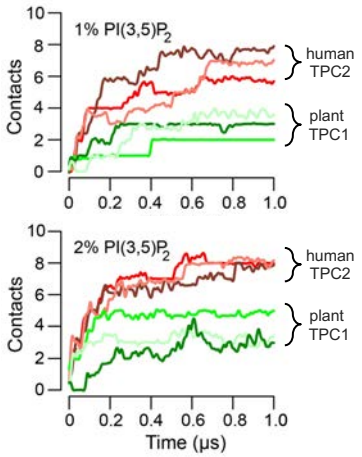
hTPC2 – PI(3,5)P_2 domain formation

1 µs simulation of hTPC2 with PI(3,5)P_2



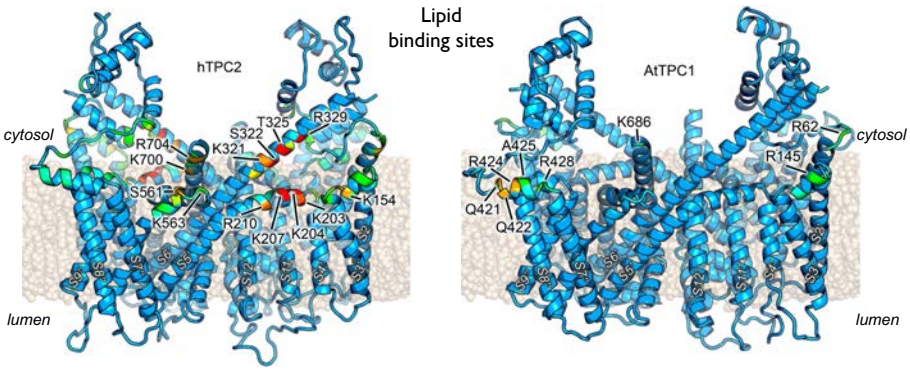


→PI(3,5)P_2 binding to hTPC2 is enhanced compared to AtTPC1

S.A. Kirsch*, A. Kugemann*, A. Carpaneto, R.A. Böckmann and P. Dietrich. Cell. Mol. Life Sci. 75:3803-3815 (2018)

12

$PI(3,5)P_2$ binding sites differ significantly

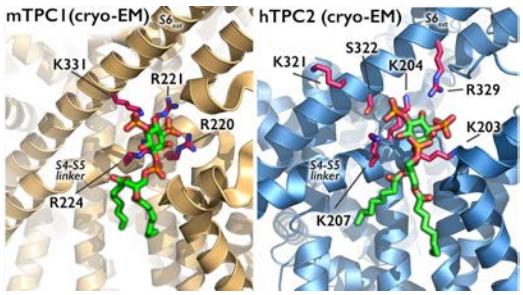


S.A. Kirsch*, A. Kugemann*, A. Carpaneto, R.A. Böckmann and P. Dietrich. Cell. Mol. Life Sci. 75:3803-3815 (2018)

13

Predicted binding site from MD in line with cryo-EM structures of hTPC2 and mTPC1

Cryo-EM structures of PI(3,5)P2-sensitive mTPC1 [2] and hTPC2 [3]



→ Binding site predicted by simulations coincides with binding site identified by cryo-EM!

[1] S.A. Kirsch*, A. Kugemann*, A. Carpaneto, R.A. Böckmann and P. Dietrich. Cell. Mol. Life Sci. 75:3803-3815 (2018)
[2] J. She, J. Guo, Q. Chen, W. Zeng, Y. Jiang and X.C. Bai. Nature. 556:130-134 (2018)
[3] J. She, W. Zeng, J. Guo, Q. Chen, X.C. Bai and Y. Jiang. eLife. 8:e45222 (2019)

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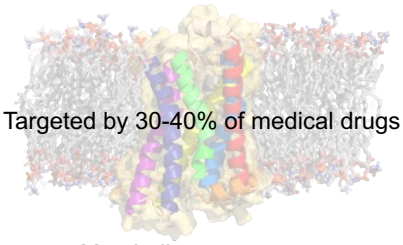
Signaling *Through* Membranes: G Protein Coupled Receptors (GPCR)

Dopamine and Opioid receptors

Development



Targeted by 30-40% of medical drugs

Cardiovascular System

Adrenergic receptors

Olfactory System

Immune System

Chemokine receptors

Metabolism

Taste

Vision

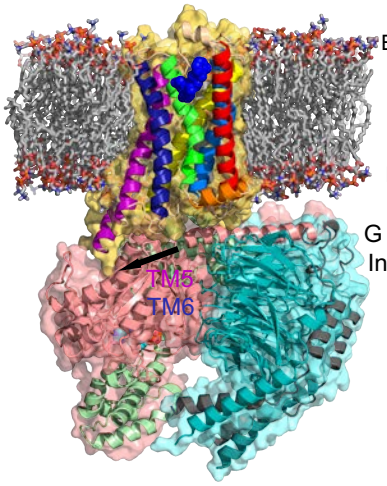
Rhodopsin

15

15

Signaling *Through* Membranes: G Protein Coupled Receptors (GPCR)

Ligand



Rasmussen S.G.F, et al., *Nature*, 2011
Kobilka B.K., Lefkowitz R. J., *Nobel Prize in Chemistry* 2012

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Signaling *Through* Membranes: G Protein Coupled Receptors (GPCR)

- **GPCRs can form dimers or higher order oligomers**
  - Affects ligand binding

Monomer Dimer Monomer

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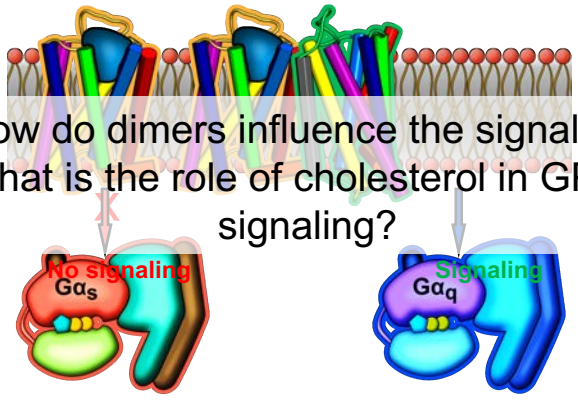
Signaling *Through* Membranes: G Protein Coupled Receptors (GPCR)

- **GPCRs can form dimers or higher order oligomers**
  - Affects ligand binding → **Different signaling than monomers**
  - Modulates intracellular signaling



How do dimers influence the signaling?
What is the role of cholesterol in GPCR signaling?

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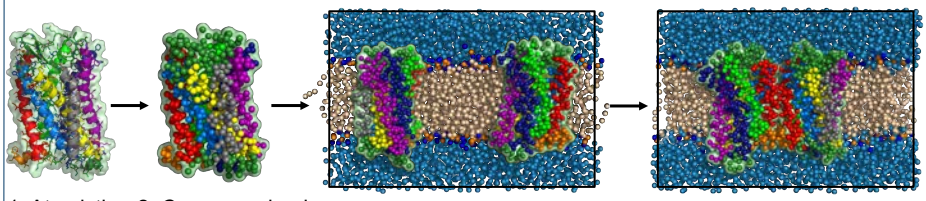
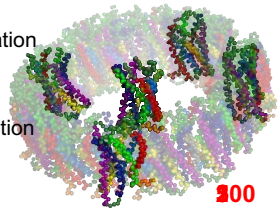
Signaling *Through* Membranes: G Protein Coupled Receptors (GPCR)



1. Atomistic model
2. Coarse-grained model
3. System setup
4. Simulation (several µs)

Simulation ensemble

- 500 independent simulation
- 2 receptors at same starting distance, but different starting orientation
- Unbiased dimerization processes



→ Statistical analysis of preferred dimer interfaces

[1] Wassenaar T.A., et al., *J. Chem. Theory Comput.* 117, 2015 [2] Wassenaar T.A., et al., *J. Chem. Theory Comput.* 11, 2015

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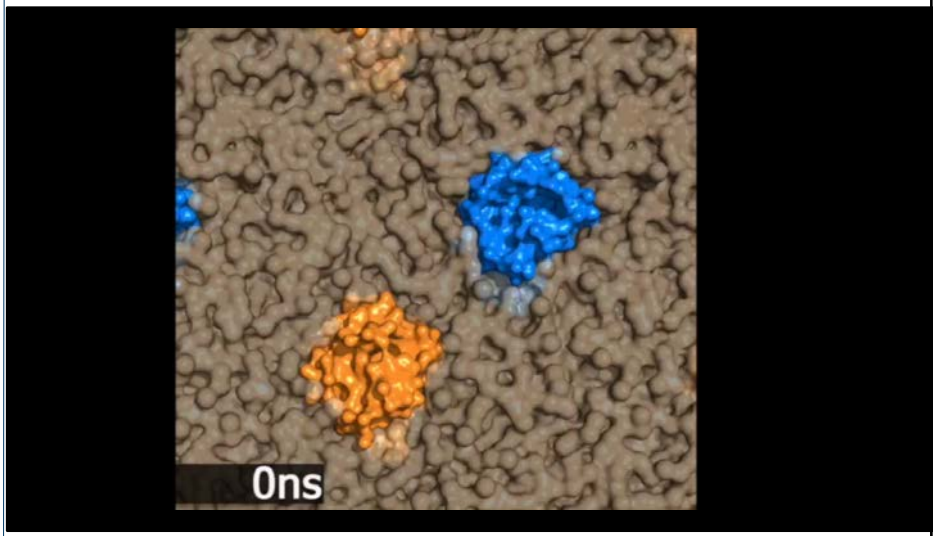
Sample GPCR Dimerization Simulation



Pluhackova K.*, Gahbauer S.*, Kranz F., Wassenaar T.A., Böckmann R.A., *PLoS Comput. Biol.* 12, 2016

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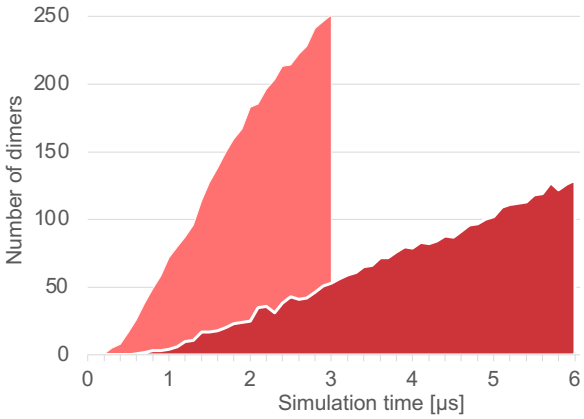
Homodimerization of CXCR4



POPC:
~250 dimers after 3μs in 500 simulation

POPC/30% cholesterol:
~120 dimers after 6μs in 500 simulations

Pluhackova K.*, Gahbauer S.*, Kranz F., Wassenaar T.A., Böckmann R.A., *PLoS Comput. Biol.* 12, 2016

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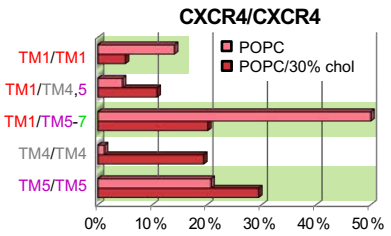
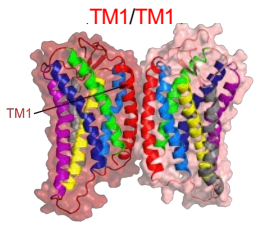
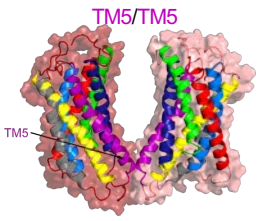
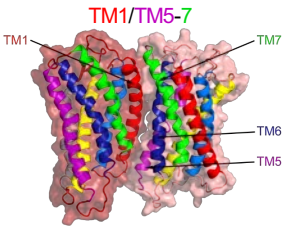
Homodimerization of CXCR4









Pluhackova K.*, Gahbauer S.*, Kranz F., Wassenaar T.A., Böckmann R.A., *PLoS Comput. Biol.* 12, 2016

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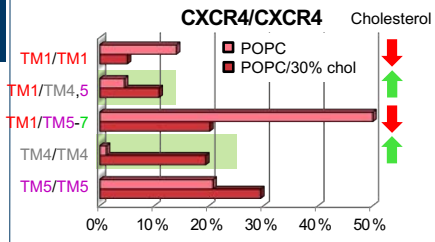
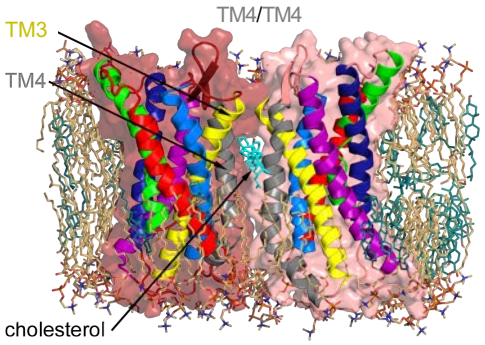
Homodimerization of CXCR4



- Cholesterol is required for CXCR4 function [1]
- TM4 was suggested in experiments to be important for function, dimerization, and HIV infection [2]



Pluhackova K.*, Gahbauer S.*, Kranz F., Wassenaar T.A., Böckmann R.A., *PLoS Comput. Biol.* 12, 2016

[1] Wang J., et al., *Mol. Cancer Ther.* 5, 2006
[2] Percherancier Y., et al., *J. Biol. Chem.* 280, 2005

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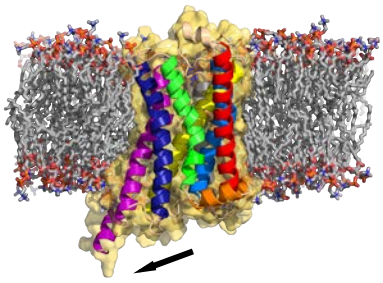
Homodimerization of CXCR4

- **TM1/TM5-7** (POPC)



- **TM5** and **TM6** trapped at interface
- Likely ***inactive***

- **TM4/TM4** (POPC/30% cholesterol)

- **TM5** and **TM6** freely accessible
- Potentially ***active***

Pluhackova K.*, Gahbauer S.*, Kranz F., Wassenaar T.A., Böckmann R.A., *PLoS Comput. Biol.* 12, 2016

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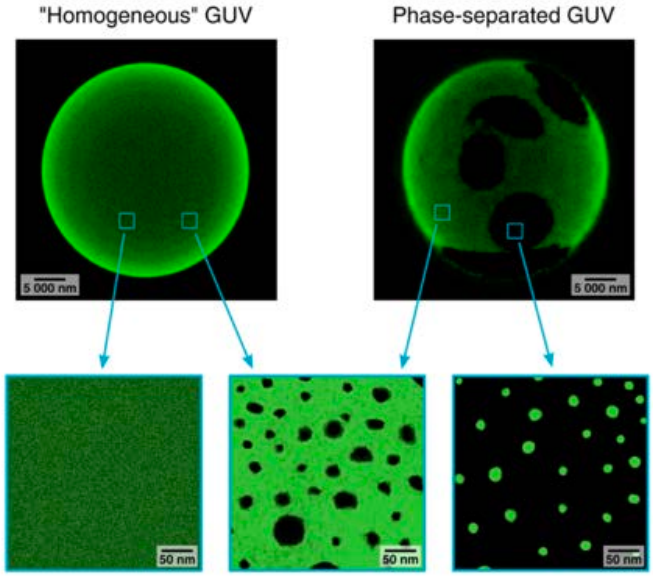
Domain Formation in Phospholipid Membranes



19.09.19 25

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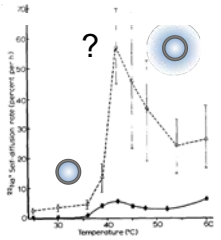
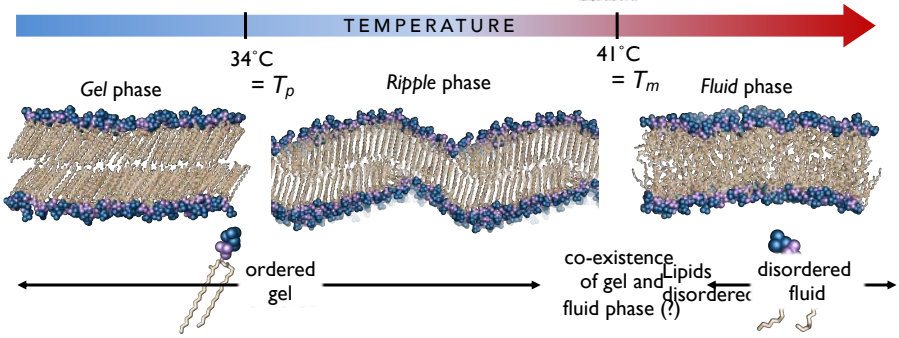
Domain Formation in Phospholipid Membranes

How does the bilayer structure look like at and slightly above T_m and how can it explain the increased permeability?



Dipalmitoyl - phosphatidylcholine vesicles (DPPC, di-C16:0)

[1]



[1] D. Papahadjopoulos, K. Jacobson, S. Nir and T. Isac. Biochim. Biophys. Acta. 311:330-348 (1973)

INTRO – DOMAIN AND ELECTROPORE FORMATION 19.09.19 26

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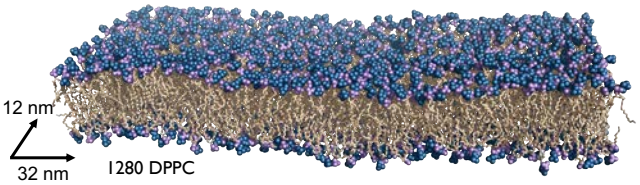
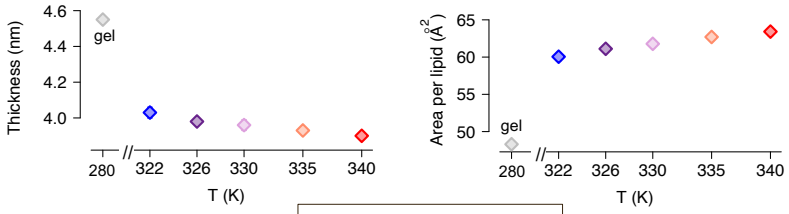
Structural features of fluid bilayers at different temperatures above T_m

Fluid phase bilayer to investigate effect of phase transition *ab initio:*

T_m = 321 K (48°C)



Average thickness and area per lipid show expected behavior:



Local bilayer properties ?

S.A. Kirsch and R.A. Böckmann. Biophys. J. 116:2131-2148 (2019)

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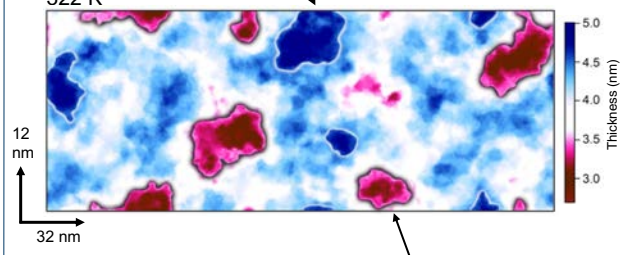
Local membrane thickness maps are very different

T_m = 321 K

Thick domains (**≥ 4.5 nm**)

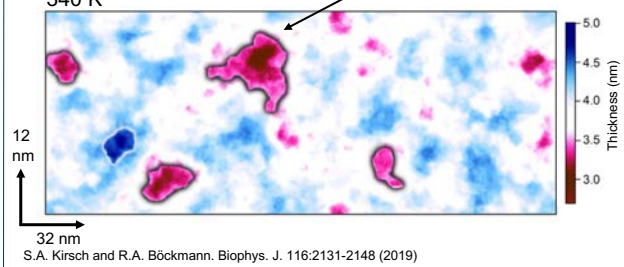
~12 nm²

322 K

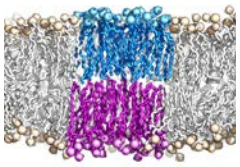


Thin domains (**≤** 3.5 nm)

340 K

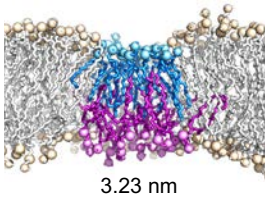


Thick domains: ordered lipids



4.71 nm

Thin domains: interdigitating lipids



3.23 nm

Lipid tail interdigitation more pronounced at T_m

S.A. Kirsch and R.A. Böckmann. Biophys. J. 116:2131-2148 (2019)

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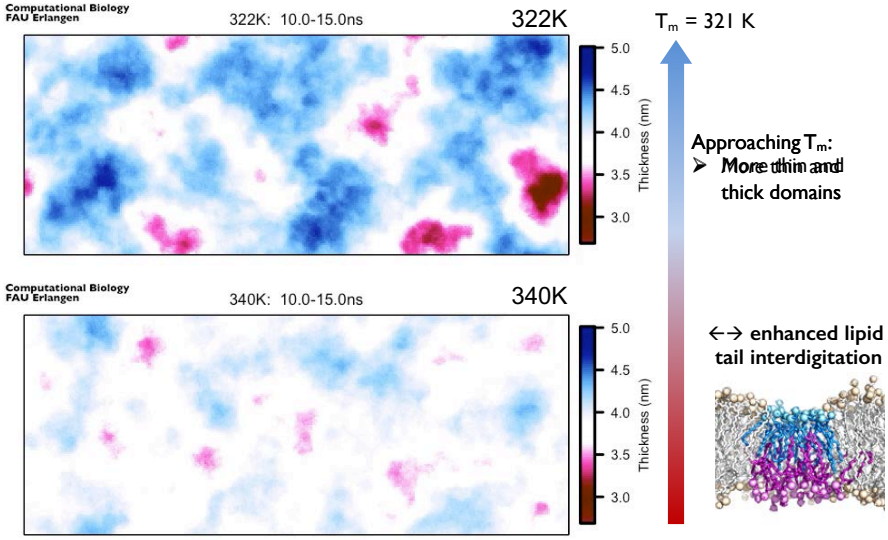
Increased lifetime of nanodomains close to T_m



T_m = 321 K

Approaching T_m:
- More thin and thick domains

←→ enhanced lipid tail interdigitation

S.A. Kirsch and R.A. Böckmann. Biophys. J. 116:2131-2148 (2019)

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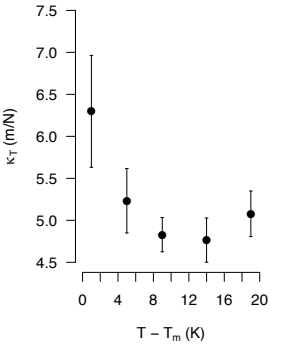
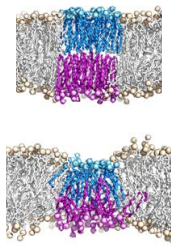
Membrane elastic constants are indicator for phase transition

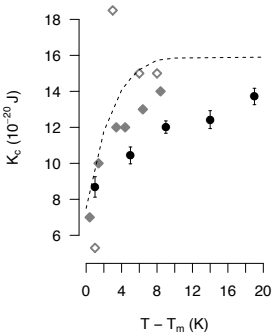
Area compressibility displays a **maximum** at T_m





Simulation results

Bending rigidity displays a **minimum** at T_m



→ Increased membrane heterogeneities explain maximum/minimum of membrane compressibility/bending rigidity when approaching T_m

S.A. Kirsch and R.A. Böckmann. Biophys. J. 116:2131-2148 (2019)

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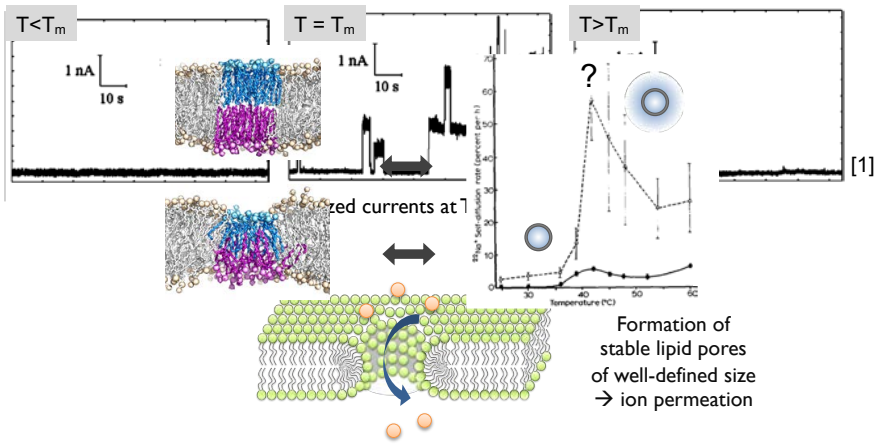
Quantized current events were observed at T_m

Relation of bilayer structure and dynamics to increased permeability?



[1] V.F. Antonov, A.A. Anosov, V.P. Norik and E.Y. Smirnova. Eur. Biophys. J. 34:155-162 (2005)

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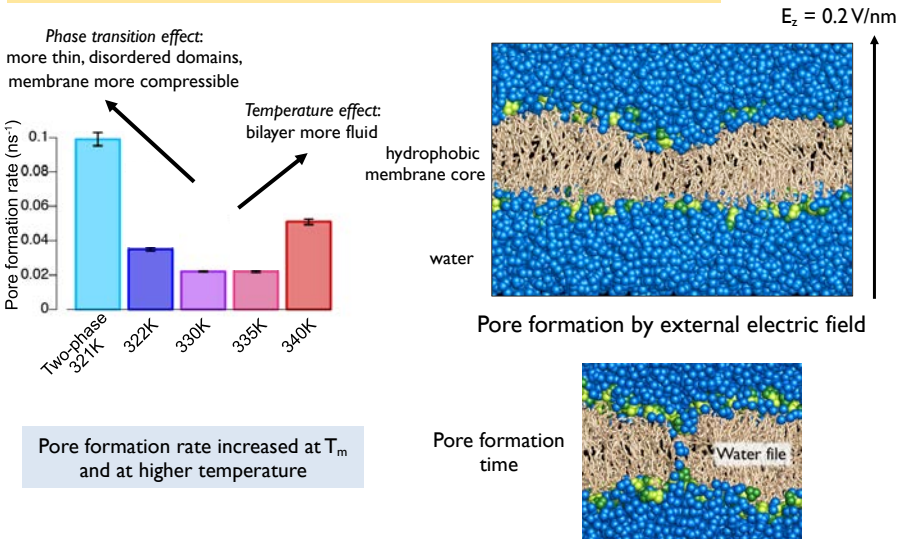
Temperature-dependent bilayer stability against electric field



Pore formation rate increased at T_m and at higher temperature

S.A. Kirsch and R.A. Böckmann. Biophys. J. 116:2131-2148 (2019)

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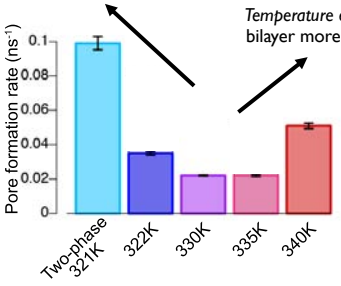
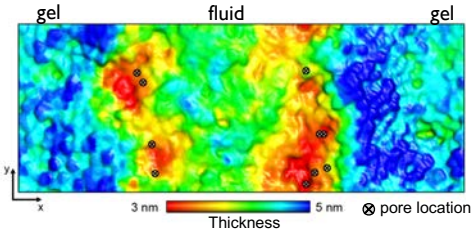
Temperature-dependent bilayer stability against electric field

Phase transition effect:
more thin, disordered domains,
membrane more compressible

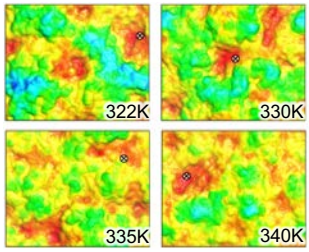
Temperature effect:
bilayer more fluid



Pore formation rate increased at T_m and at higher temperature



Pore formation in pre-existing thin membrane domains



S.A. Kirsch and R.A. Böckmann. Biophys. J. 116:2131-2148 (2019)

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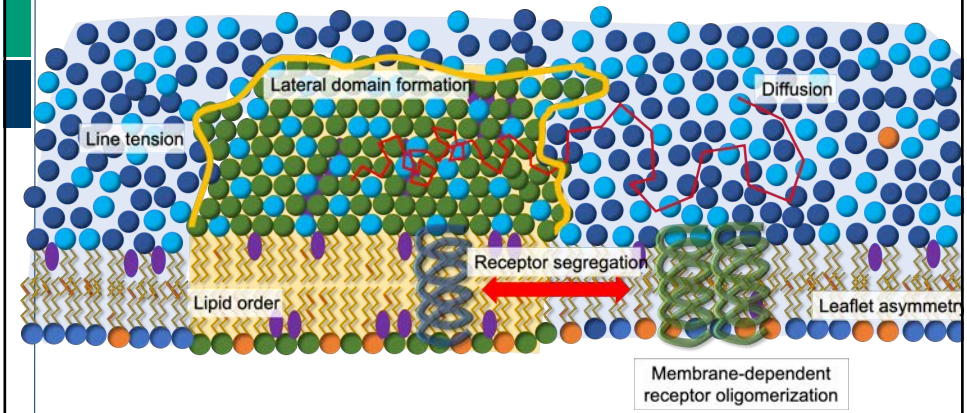
Domain Formation in Multi-Component Membranes



How are the different domains in multi-component membranes composed?

How can we control membrane domain formation?

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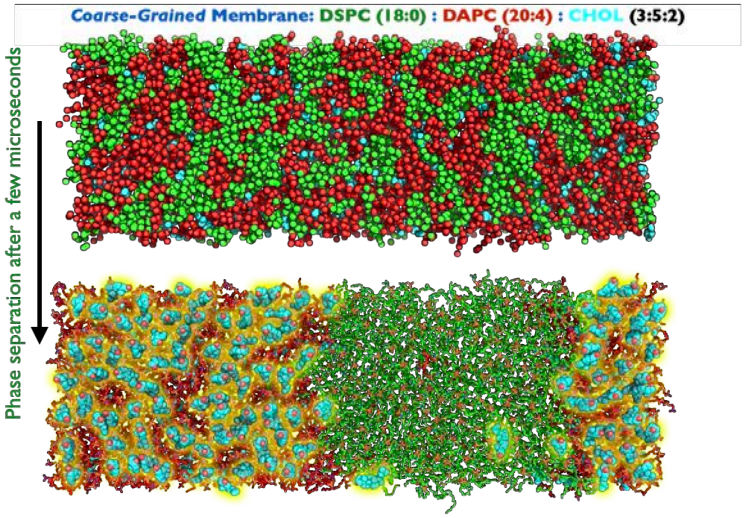
Domain Formation in Multi-Component Membranes

A coarse-grained view…



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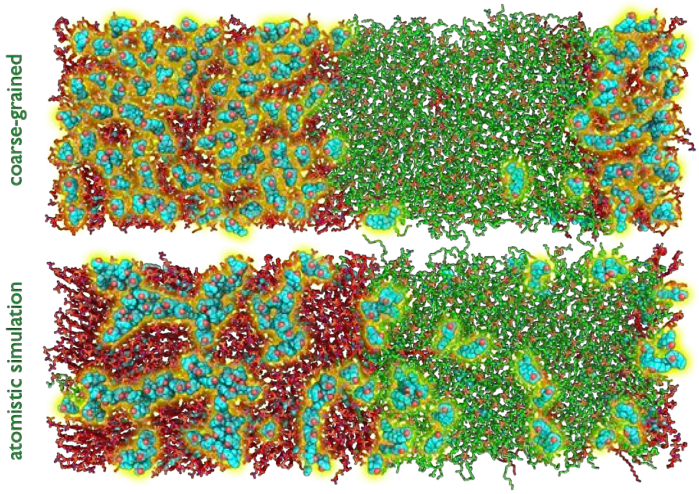
Domain Formation in Multi-Component Membranes

Switching to atomistic resolution…



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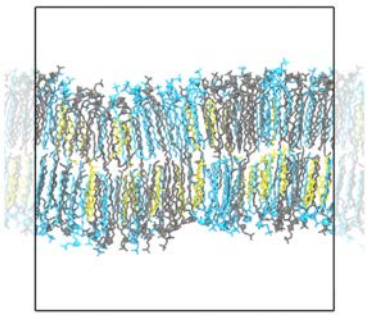
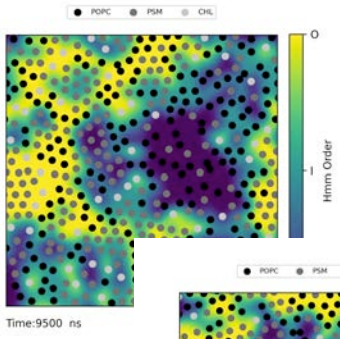
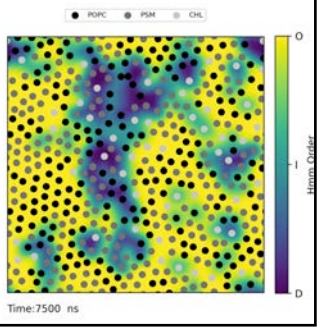
Domain Formation in Multi-Component Membranes

Ternary membrane with POPC/ PSM/Cholesterol:





303K
ordered
disordered



294K
ordered
disordered

Temperature-dependent formation of membrane domains

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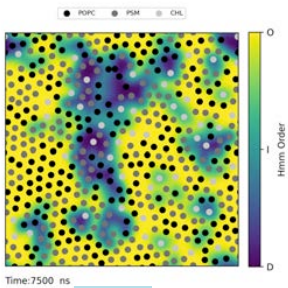
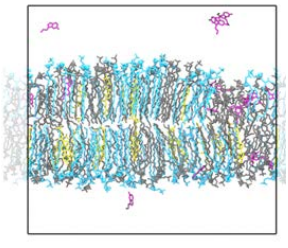
Domain Formation in Multi-Component Membranes

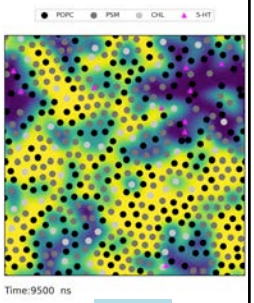
Ternary membrane with POPC/ PSM/Cholesterol**: Influence of Neurotransmitters**



294K

+ Serotonin





294K

Neurotransmitter modulate the membrane domain formation!

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THANK YOU!

Computational Biology
Anna Bochicchio
Astrid Brandner
Matthias Pöhnl
Sonja Kirsch
Stefan Gahbauer
Marius Trollmann
Franziska Spiegel
Linkai Zhang
Theo Konopatzki

Collaborators
Petra Dietrich (Molecular Plant Physiology, FAU)
Armando Carpaneto (University of Genoa)
Alexandra Schambony (Developmental Biology)
Daniel Huster (NMR, Leipzig)
Tobias Unruh (Crystallography and Structural Physics)
Thomas Gutsmann (Borstel)
Ivan Bogeski (Göttingen)
Monika Pischetsrieder (Food Chemistry, FAU)
Lorenz Meinel (Pharmaceutics, Würzburg)
Falk Nimmerjahn (Genetics, FAU)
Diana Dudziak (Dendritic Cell Biology, FAU)

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