

# Lipids as ligands: Elucidating the role lipids play in modulating neurotransmitter receptor response to agonist

**RUTGERS**

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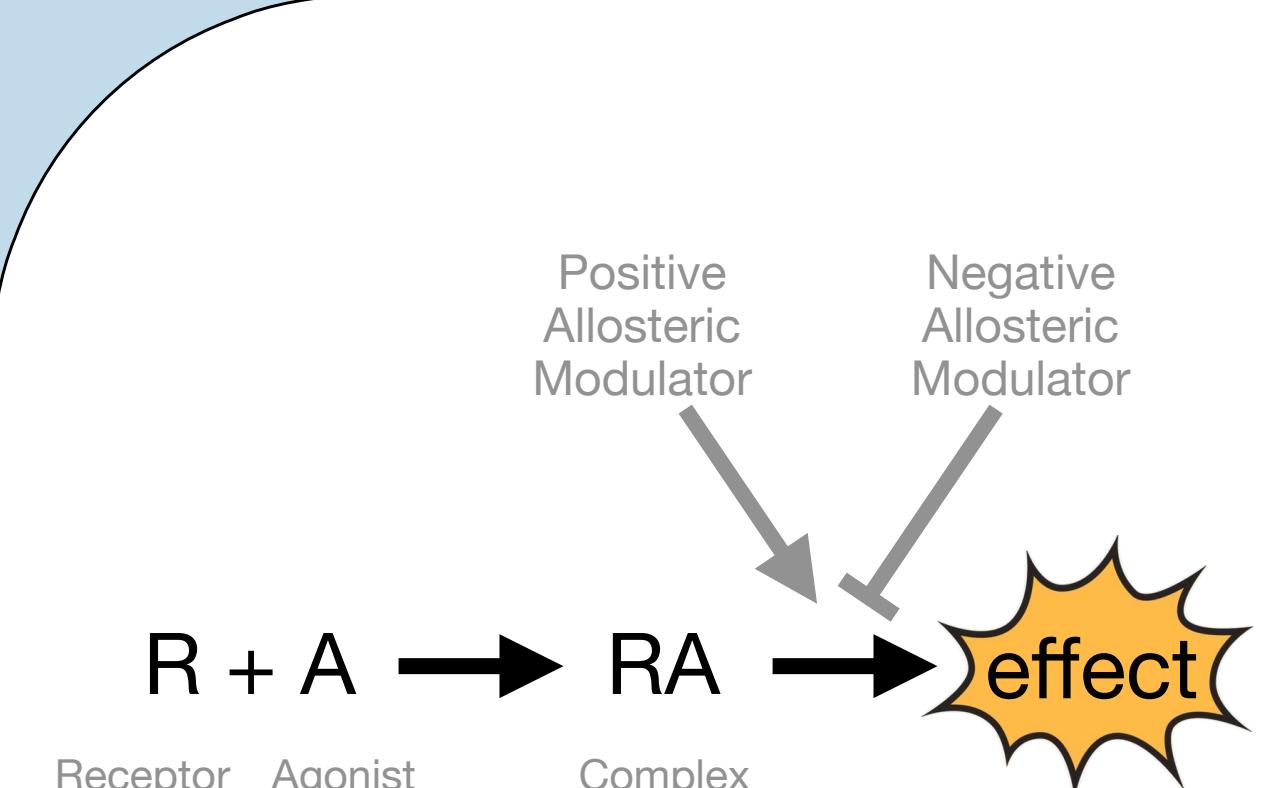


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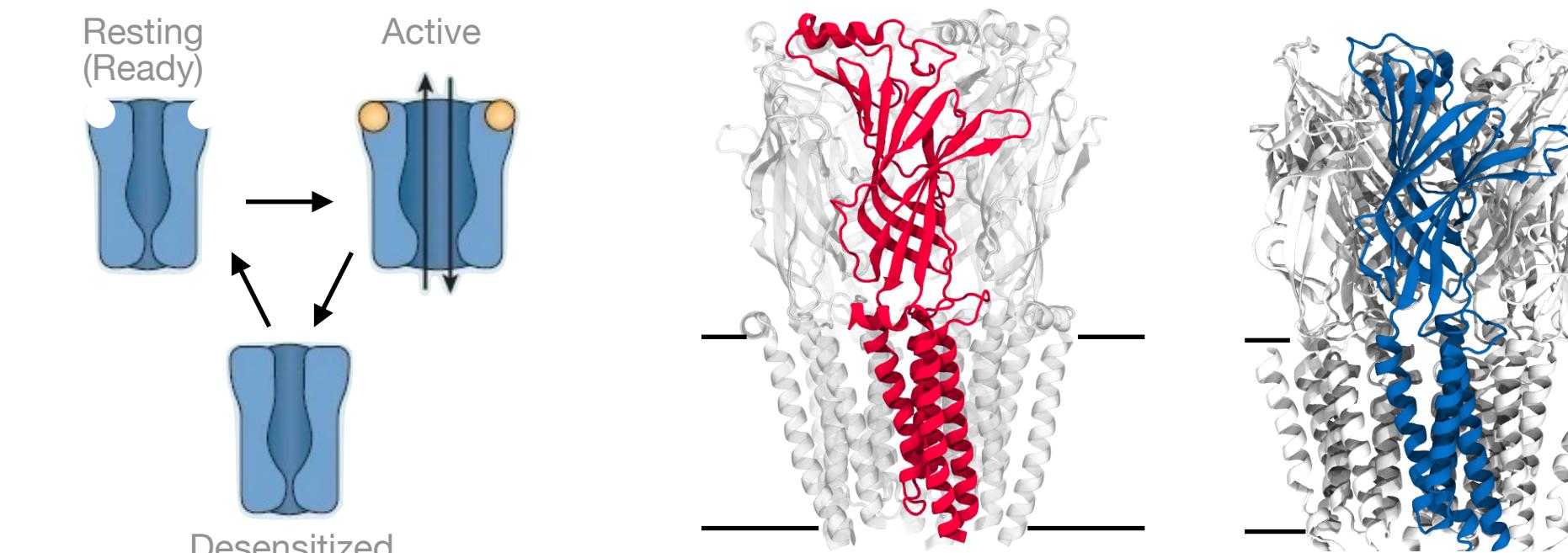
## Abstract

Neurotransmitter receptors, several of which belong to the pentameric ligand-gated ion channel (pLGIC) family, are known to be sensitive to their lipid environment. Gain or loss of function is regularly observed in membranes of varying composition, suggesting lipids can modulate protein function. While it has been known for some time that lipids can directly bind to and allosterically modulate membrane proteins, measuring a lipid's binding affinity has remained a challenge. Our lab has introduced the Density-Threshold Affinity (DTA), a technique for measuring a lipid's binding affinity from unbiased coarse-grained molecular dynamics simulations. In the present work, we use the DTA toolkit to investigate two lipid interactions with pLGICs: arachidonic acid (AA) inhibiting the nicotinic acetylcholine receptor (nAChR), and cardiolipin (CDL) rescuing the resting state of a prokaryotic ion channel (ELIC5). We demonstrate that AA binds to an intrasubunit site on the nAChR and effectively saturates the subunits at conditions matching those reported by experimentalist colleagues. We also show that cardiolipin biases the ensemble of states towards the resting state by binding to an intrasubunit site on ELIC5.

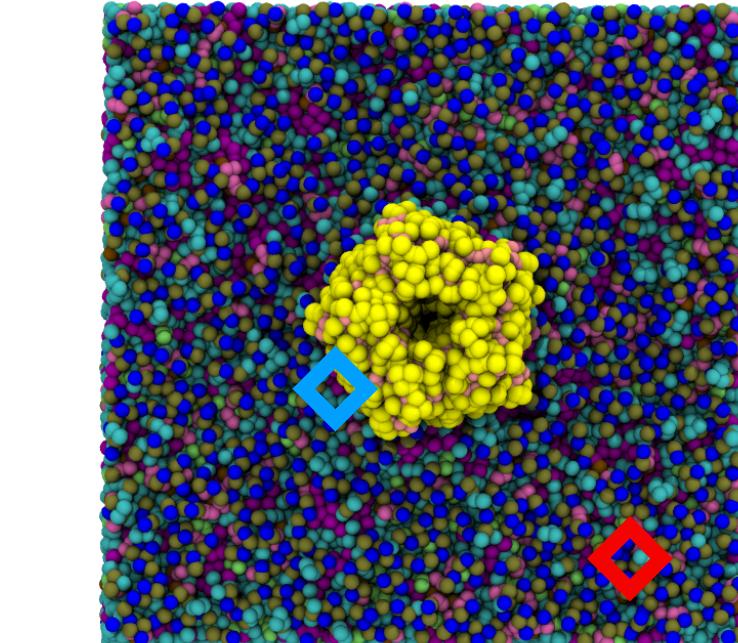


Molecules that bind to the receptor outside of the active site and change its response to agonist are called allosteric modulators. Increasingly, lipids are being recognized as potential allosteric modulators.

## Background



Three stage model of ion channel activation. Lipids can bias the ensemble of states by lowering the free energy of certain conformations relative to the others.



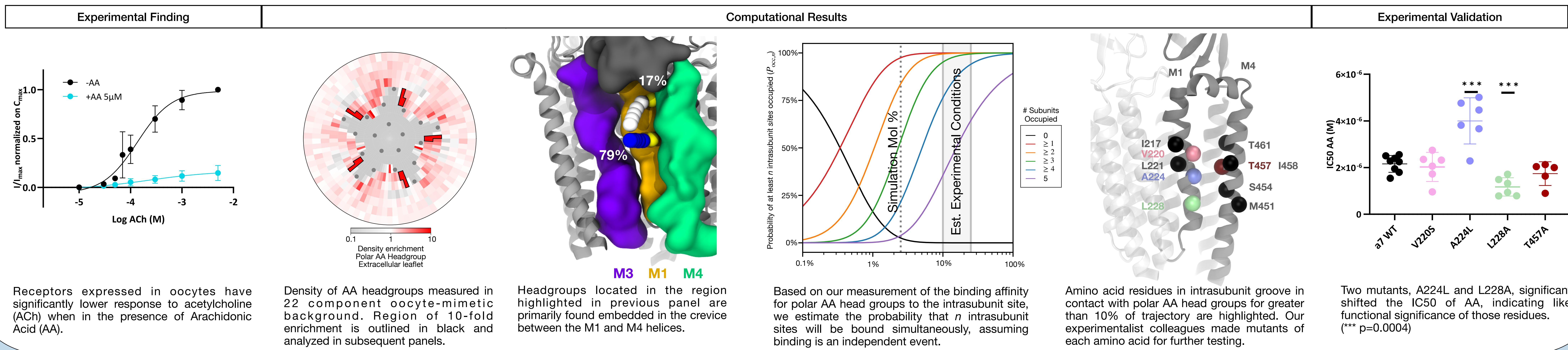
Two members of the pentameric ligand-gated ion channel (pLGIC) family: the nicotinic acetylcholine receptor (nAChR, red) and a prokaryotic relative (ELIC5, blue). Both bind agonist to open the ion channel pore in the center. Both are sensitive to their lipid environment.

## Research Questions

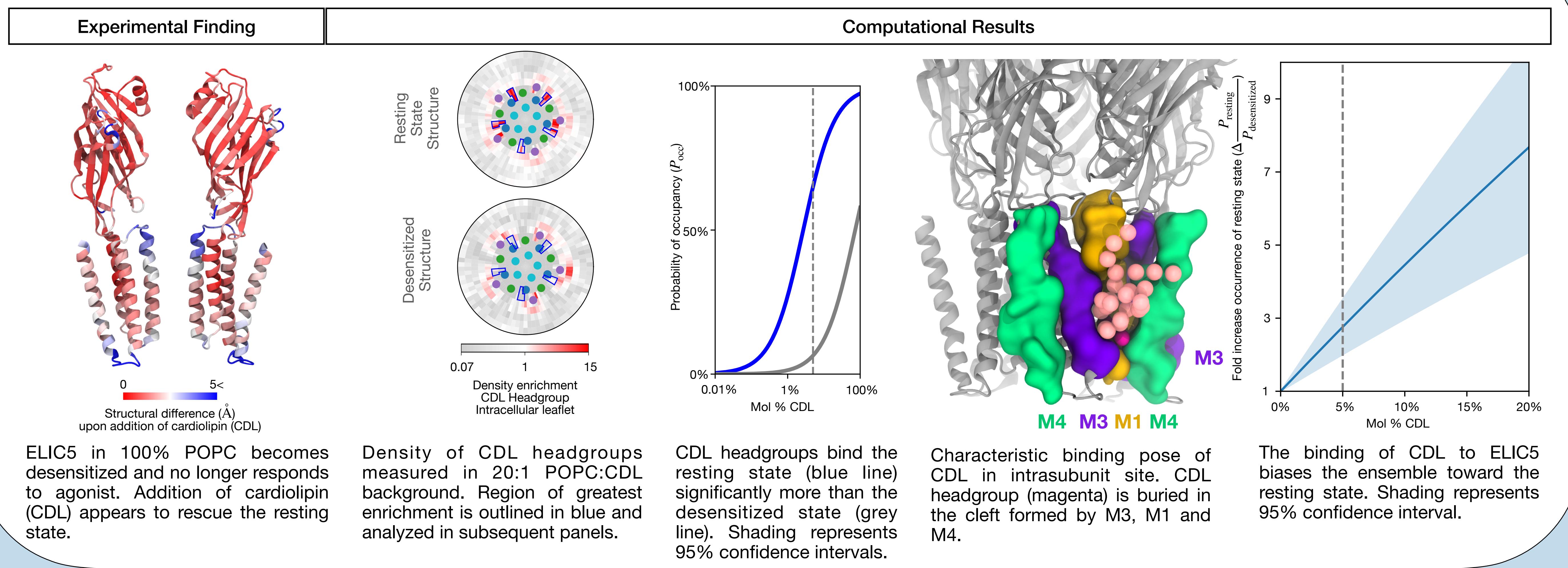
What is the molecular mechanism of Arachidonic Acid (AA) inhibition of the  $\alpha 7$  nicotinic acetylcholine receptor (nAChR)?

Does cardiolipin (CDL) stabilize the resting state of a prokaryotic pentameric ion channel (ELIC5)?

## Nicotinic acetylcholine receptor (nAChR) inhibited by Arachidonic Acid (AA) binding to intrasubunit site



## Cardiolipin (CDL) stabilizes resting state of prokaryotic pLGIC (ELIC5)



## Summary

- Arachidonic acid is expected to occupy 3-5 intrasubunit sites simultaneously, and may allosterically inhibit channel activation via ALA224 or LEU228.
- Cardiolipin stabilizes the resting state of ELIC5 by binding to an intrasubunit site in the intracellular leaflet, rescuing channel activity.

## Methods

$P_{occ} = \frac{x_B}{e^{\Delta G/RT} + x_B}$

$$P_{occ,n} = \sum_{i=n}^5 \binom{5}{i} (P_{occ})^i (1 - P_{occ})^{5-i}$$

$$P_{occ,0} = 1 - P_{occ,1}$$

$$\frac{P_A}{P_B} = \frac{x_{PC} + x_{CDL} e^{-\Delta G_B/RT}}{x_{PC} + x_{CDL} e^{-\Delta G_B/RT}}$$

Read our protocol paper!

All simulations had 4 replicas, each 10 us long. GROMACS 2024. Martini 2.2 Forcefield.

## Acknowledgments

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