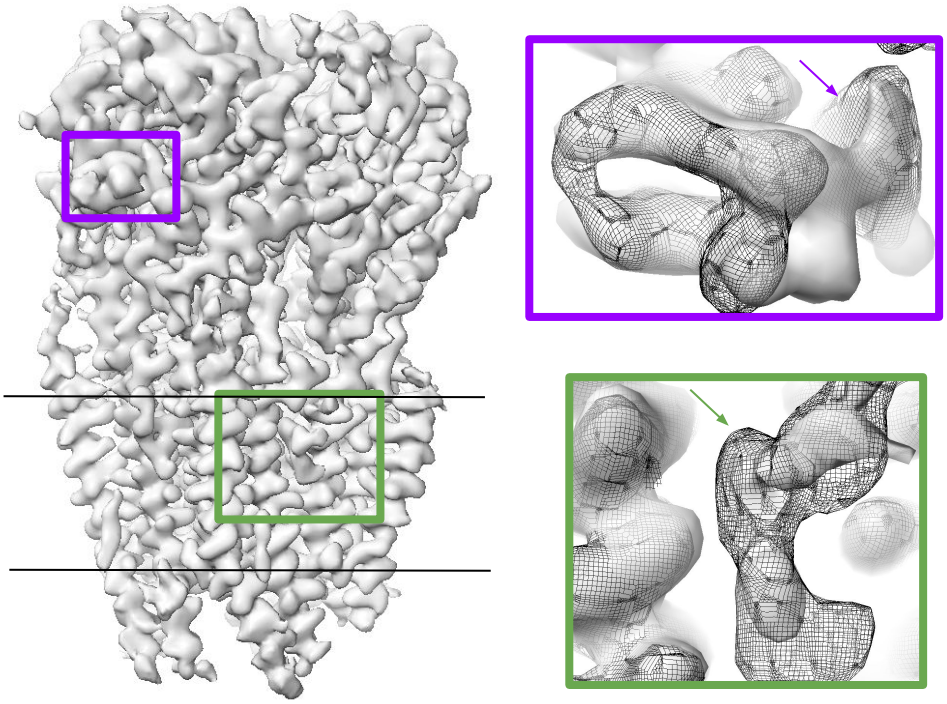
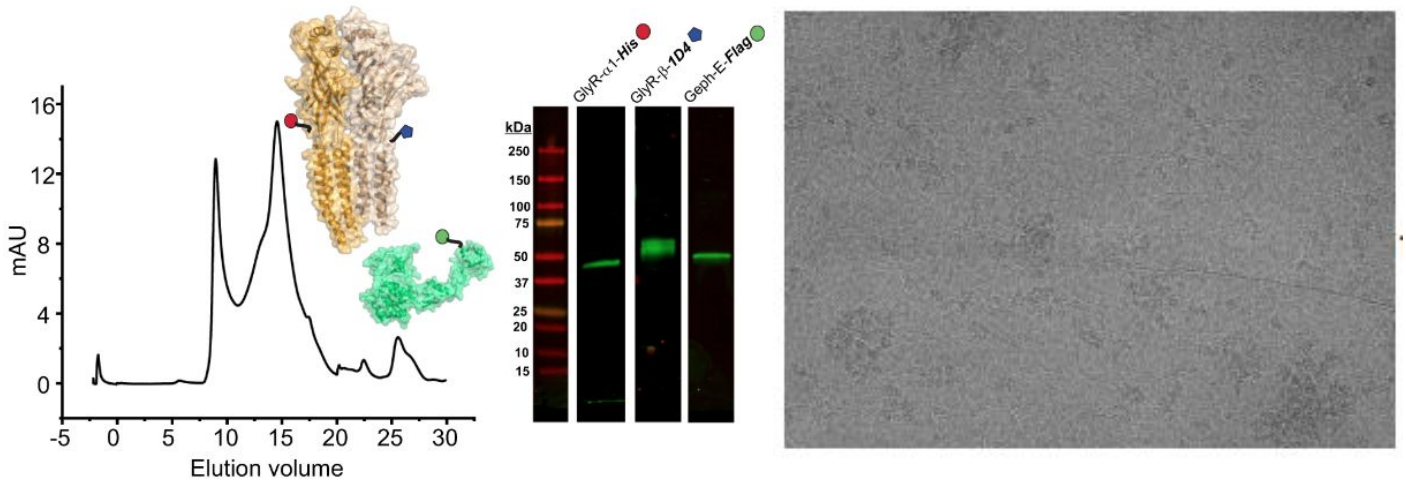
Figure 1



Preliminary Structure of GlyR in the Presence of Saturating Strychnine and Ivermectin (3.5 angstrom). Binding sites for strychnine (purple arrow) and ivermectin (green arrow) are highlighted. An approximate membrane boundary is shown as a black line. To show differences in ligand binding between interfaces, the density was copied and rotated one subunit relative to the original. The rotated density is shown as a black mesh while the other with transparent gray. There are significant differences in the positioning of the ligand and surrounding protein. These regions are highly correlated with channel activity in past GlyR studies. The pore is also desensitized in this structure suggesting the effects of ivermectin trump those of strychnine.

Figure 2



Purification of GlyR and Cryo-EM Imaging: Top Left: Heteromeric GlyR was purified using a double affinity purification scheme using 1D4 and 8x His tags for β and α subunits respectively. This was followed by size-exclusion chromatography with fractions from the 15 mL peak taken for cryo-EM samples. Top Right: A typical micrograph of heteromeric GlyR in the presence of 100 μM strychnine on graphene-oxide grids. Bottom Right: Final 2D classes from 84,437 particles from 14,270 micrographs similar to the one shown. Figures are from Gibbs et al. 2023.