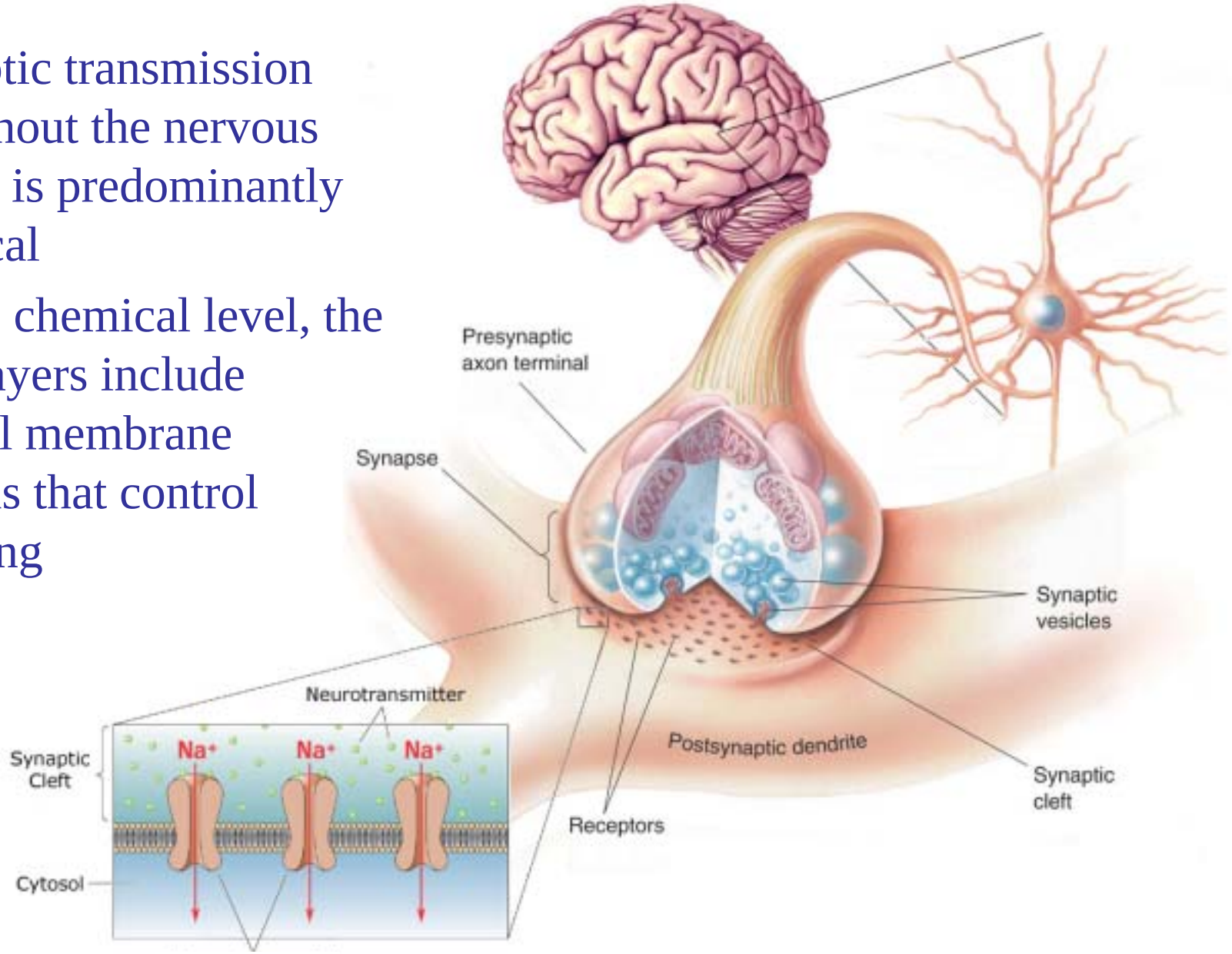
- Synaptic transmission throughout the nervous system is predominantly chemical
- At the chemical level, the key players include integral membrane proteins that control signaling

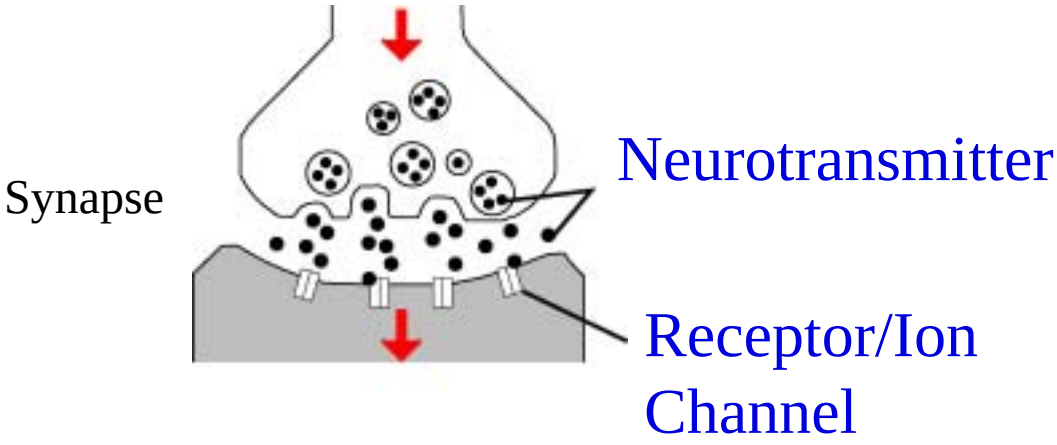


Ligand-Gated Ion Channels

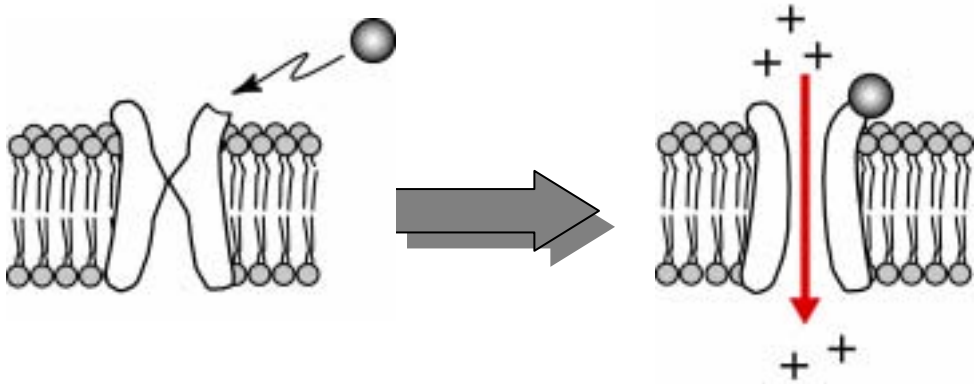
Neuroreceptors & Synaptic Transmission

A very large class of such proteins is the ligand-gated ion channels.

These complex proteins contain both a ligand binding site and an ion channel that is normally closed, but that opens in response to binding of the ligand (ie, the neurotransmitter).



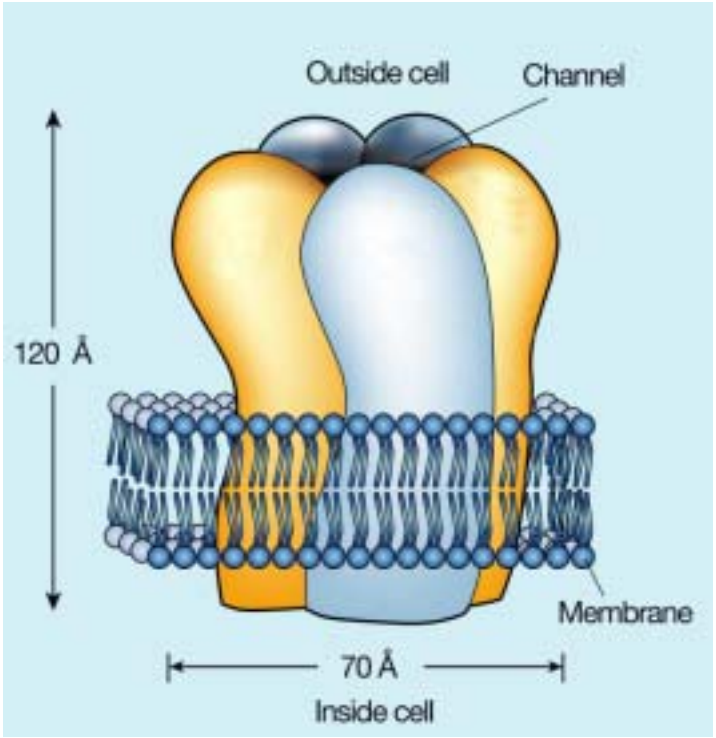
Ligand-Gated Ion Channel



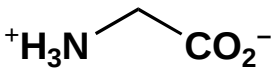
= Neurotransmitter

Ion Flow = Current

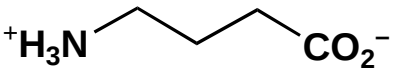
The "Cys Loop" Superfamily of Ligand-Gated Ion Channels



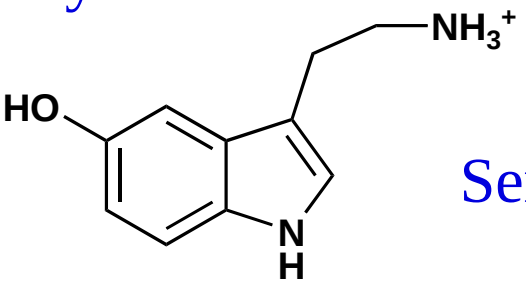
Receptors for:



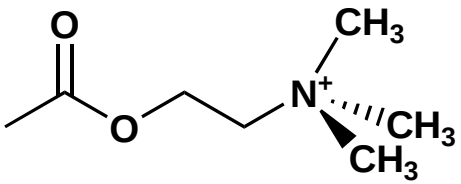
Glycine



GABA

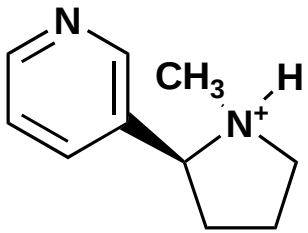


Serotonin (5-HT)



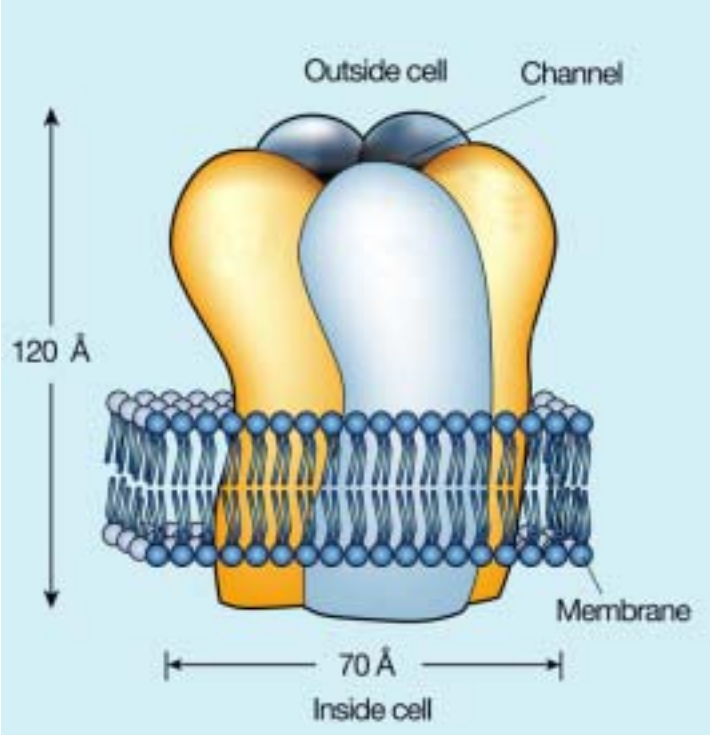
Acetylcholine (ACh)

The ACh receptor also responds to nicotine, and so is called the "nicotinic" acetylcholine receptor - nAChR

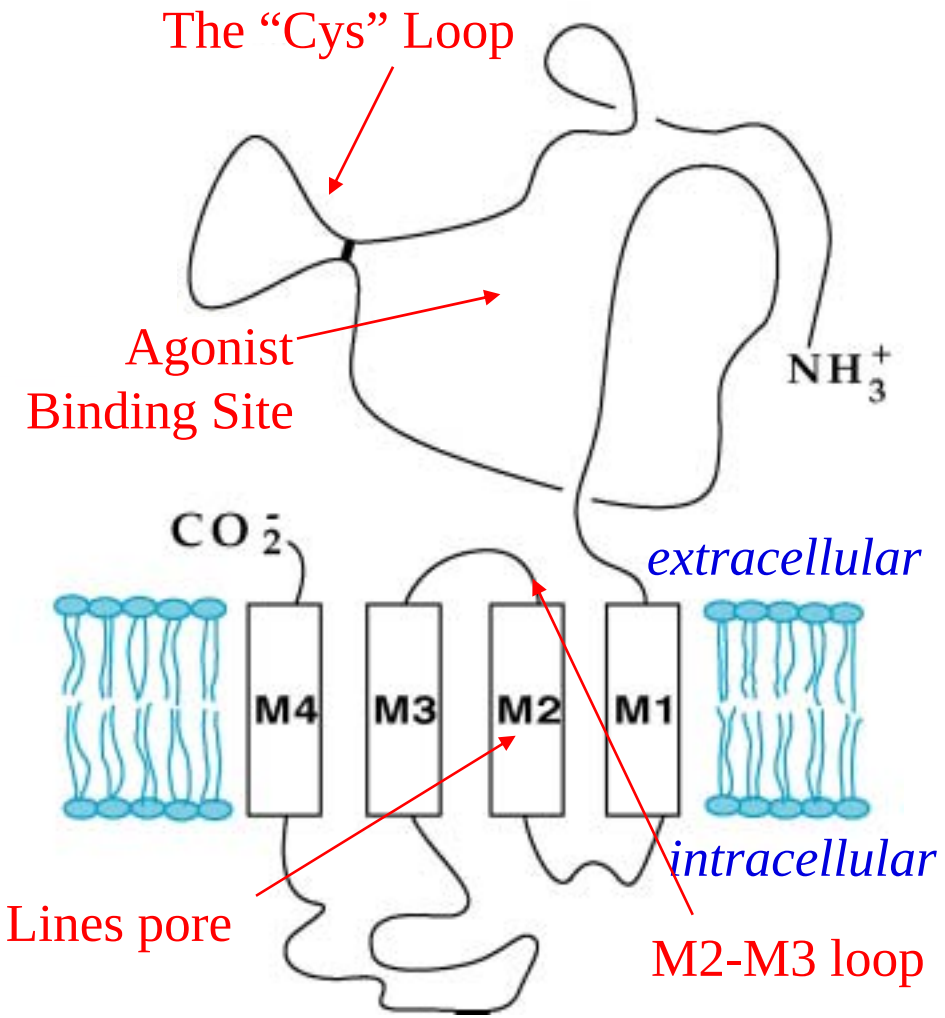


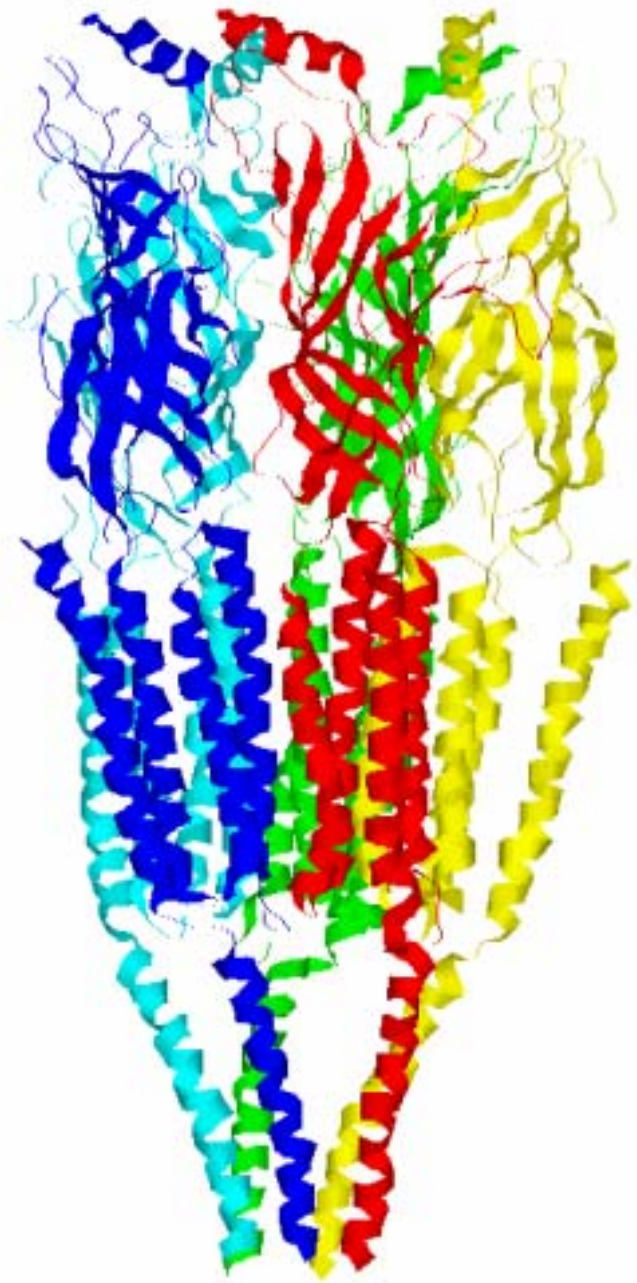
Nicotine

The “Cys Loop” Superfamily of Ligand-Gated Ion Channels



Five homologous/identical subunits





nAChR

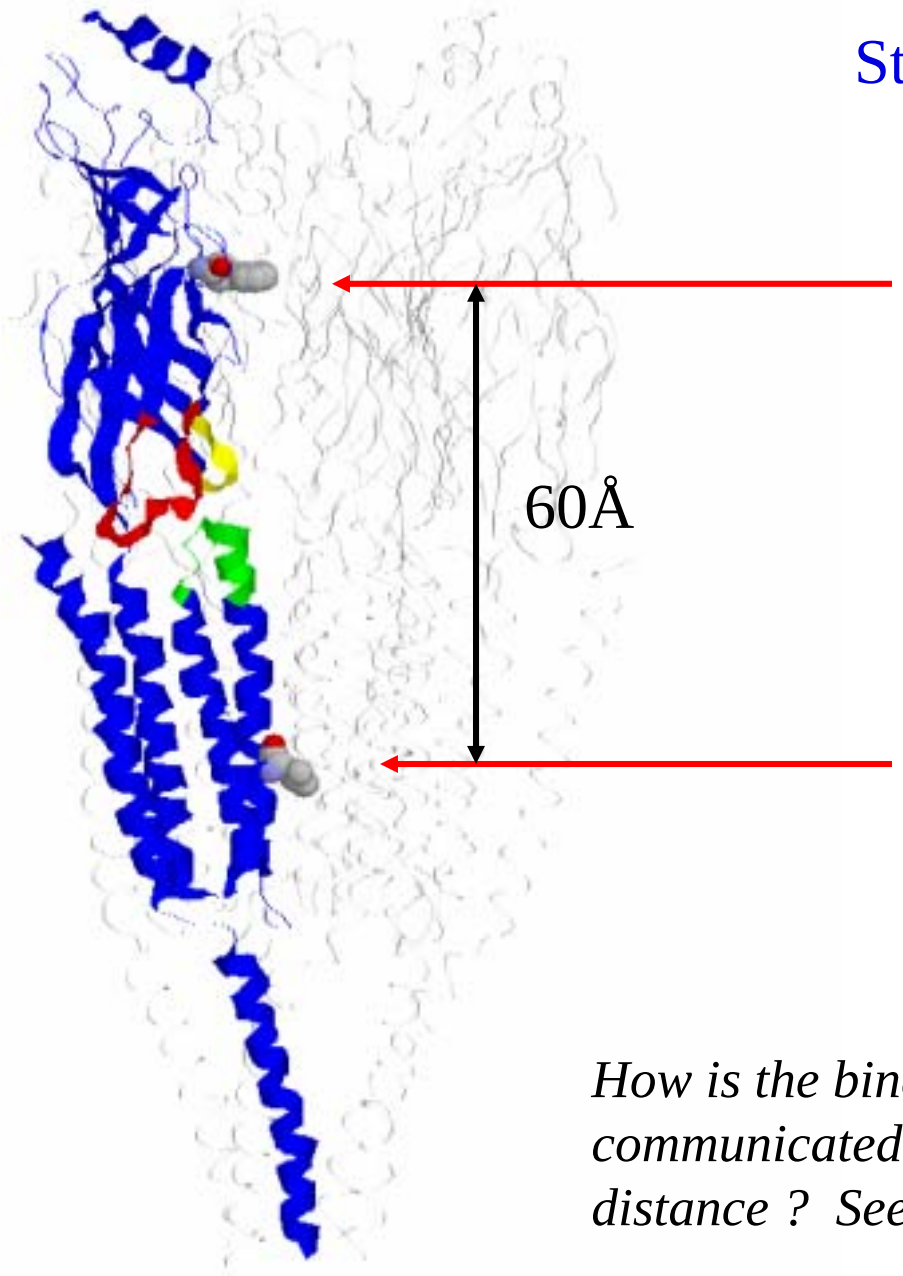
A moderate resolution model of the receptor has been built using homology modeling and Cryo-EM image.

Unwin, *JMB*, **346**, 967 (2005)

A great starting point, but:

Receptors exist in multiple states, and it is the collection of interconversions among them that defines their function

Structural Landmarks



Agonist Binding Site.
A tryptophan in the extracellular domain
See Case Study 1.

"Gate" of the Channel.
A hydrophobic occlusion formed by leucine residues

How is the binding event rapidly communicated over such a long distance ? See Case Study 2

Structure-Function Studies

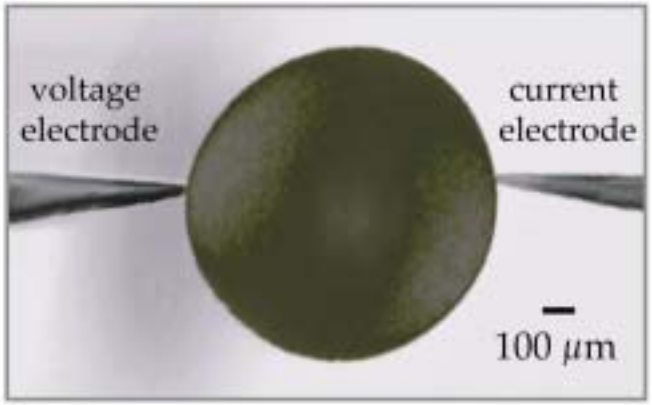
- High-resolution structural information is difficult to obtain for such large, integral membrane proteins
- Best tool is to vary structure and evaluate how that impacts function
- A convenient expression tool is the oocyte from a frog named *Xenopus laevis*
- We vary structure using unnatural amino acid mutagenesis - *see separate link*
- We probe function using *electrophysiology*

Structure-Function Study

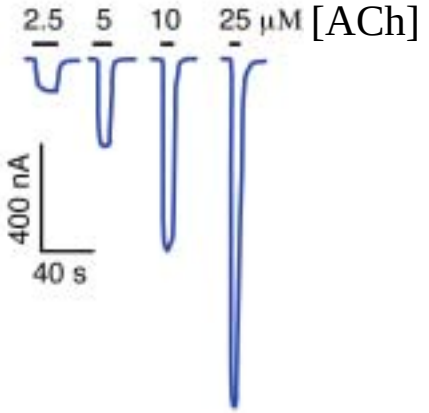
Flow of ions through a channel is equivalent to electrical current

Functional Probe: Electrophysiology

Two Electrode Voltage Clamp Recording



Xenopus oocyte



Simply inject DNA or mRNA into oocyte, return 24 hours later.

Ligand-gated ion channel is expressed and it responds to neurotransmitter (eg., ACh) by generating a current

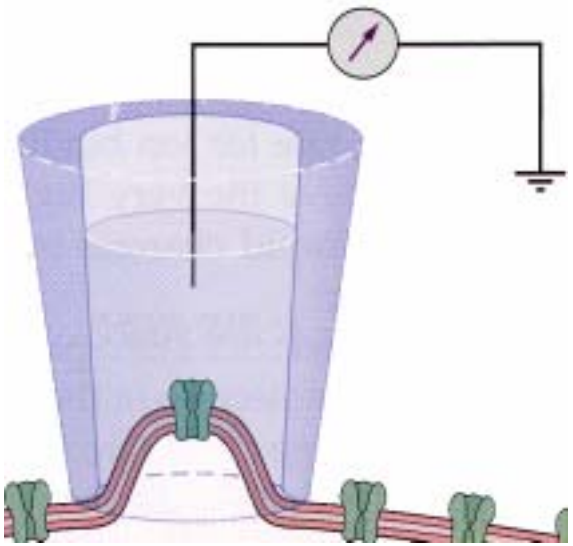
Physiology and pharmacology same as in natural environment.

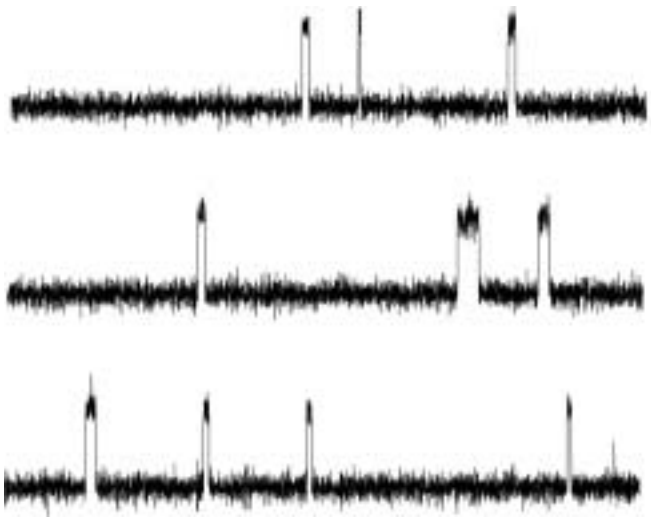
Structure-Function Study

Single molecule recording via the patch clamp

Functional Probe: Electrophysiology

The Patch Clamp





τ_{open} = 2.0 msec

Single molecule kinetics in real time