Neurotransmitters and physiology of synapses

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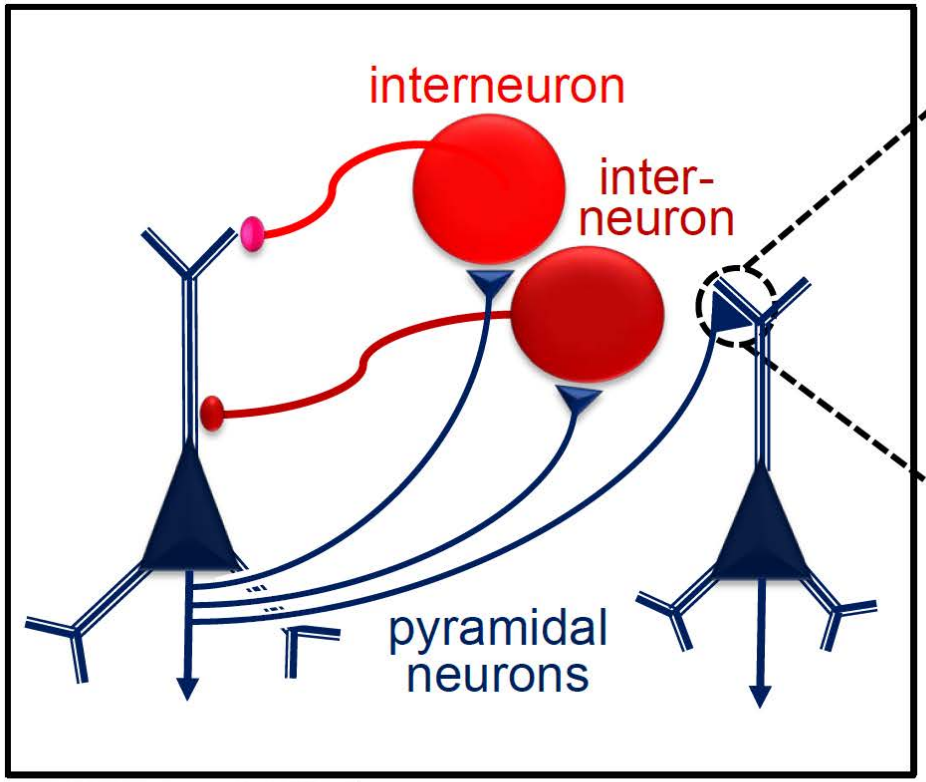
Neuronal communication

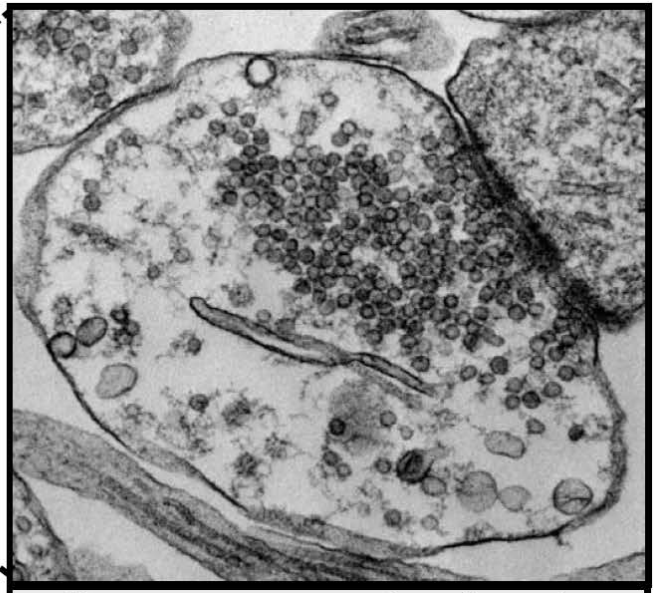
1) Electrical signals

2) Chemical signals

Neurotransmitter (mediator): a substance
that is released at a synapse by one neuron
and that affects a postsynaptic cell in a specific manner.

Thomas C. Südhof – Nobel Prize, 2013





Synapses: the basic computational units of the brain

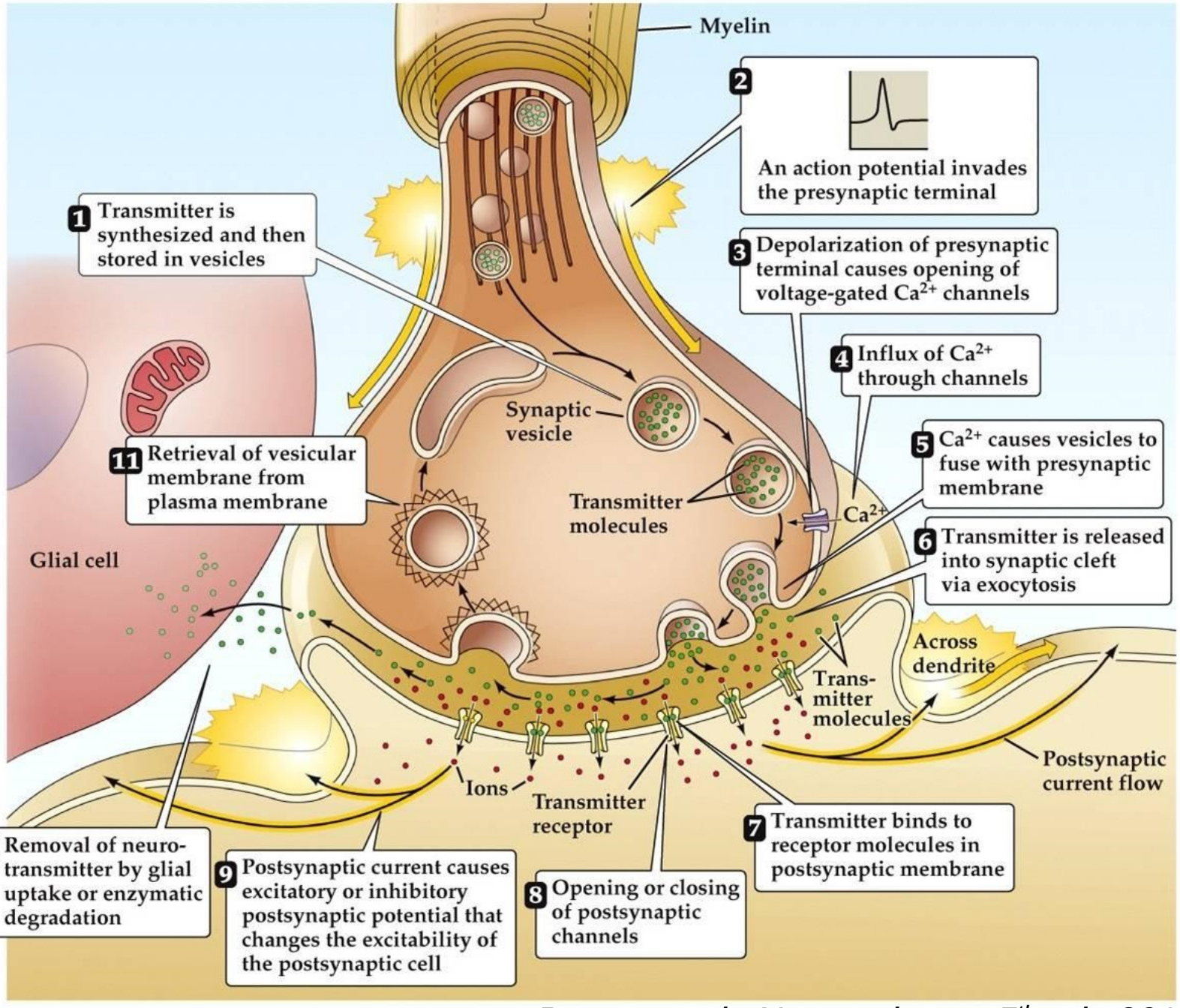
Although synapses differ in properties, all synapses operate by the same principle

Bernard Katz - Nobel Prize, 1970

Chemical signaling

- Cells release neurotransmitters, usually stored in synaptic vesicles in presynaptic endings.
- Fast secretion of neurotransmitters is triggered in Ca^{2+}-dependent way.
- Neurotransmitters diffuse from presynaptic to postsynaptic cells across the synaptic cleft.
- Neurotransmitters bind to postsynaptic cells at specific sites – postsynaptic receptors – activate them and generate postsynaptic response.

Synaptic transmission



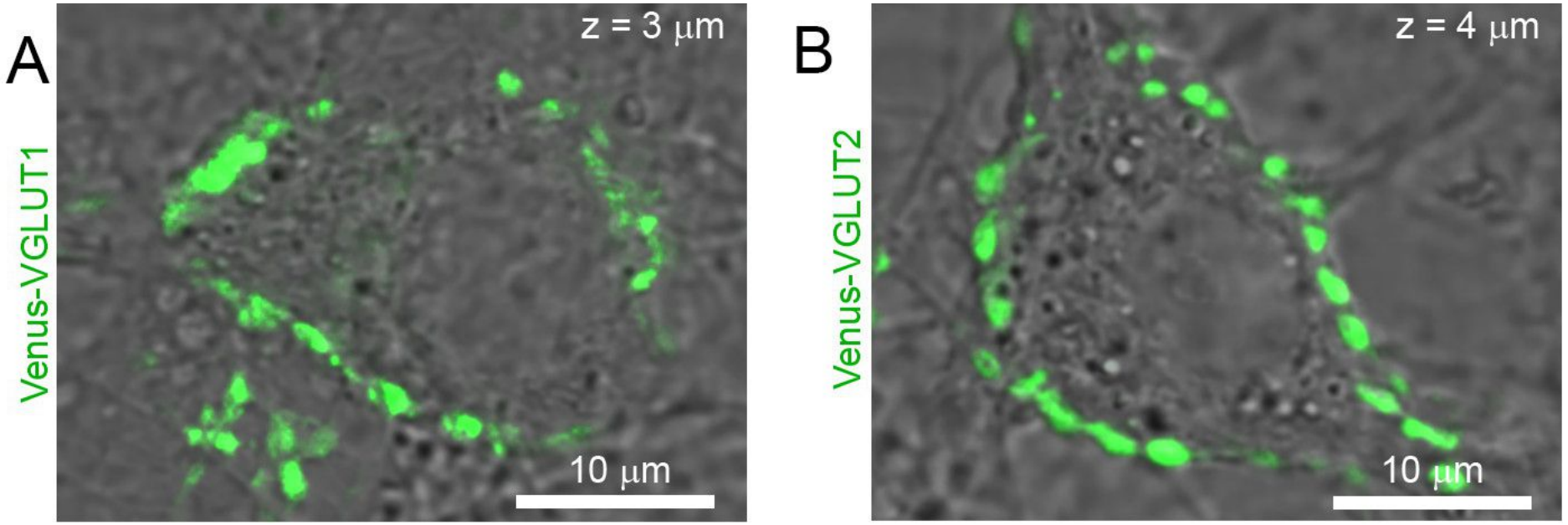
Purves et al., Neuroscience, 5th ed., 2012

Neurotransmitters (NT)

Criteria:

- it is localized in the presynaptic neuron
  (biochemical or immunocytochemical evidence of NT
  or synthesizing enzymes),
- it is released by presynaptic depolarization in
  Ca2+-dependent way (electrophysiological methods),
- postsynaptic cell contains specific receptors for candidate NT
  (electrophysiological, biochemical or
  immunocytochemical methods,
  application of exogenous agonists or antagonists)

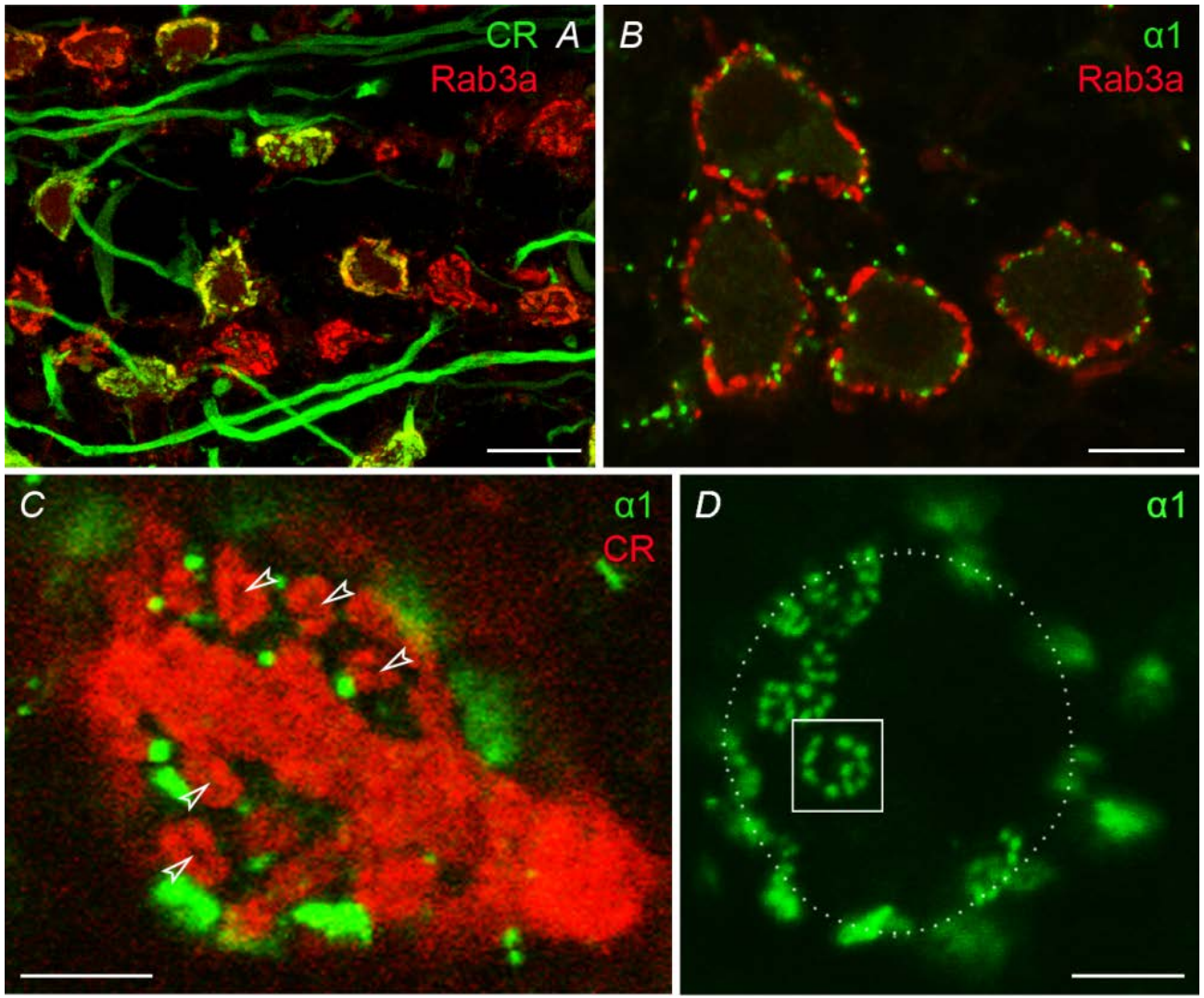
Vesicular glutamate transporter (vGluT)--positive terminals at central synapses



Guillaud et al., eLIFE 2017

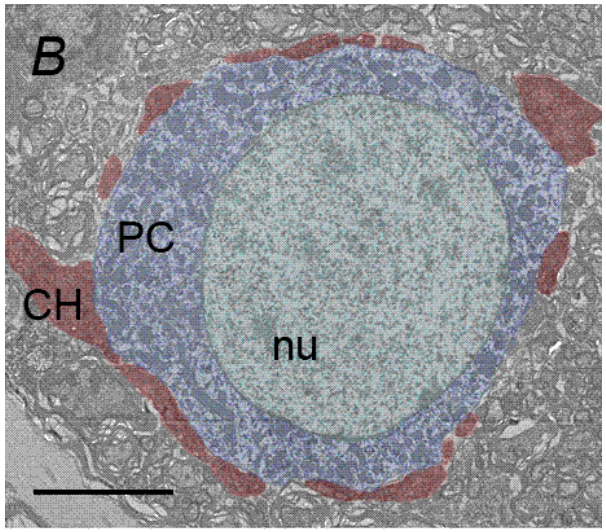
Immunohistochemical localization of glycine receptors in brain tissue

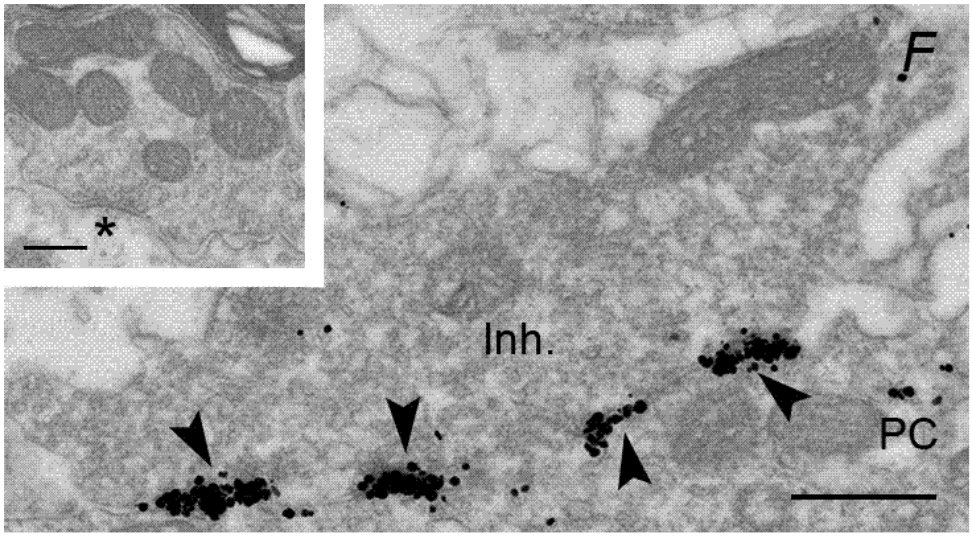
Confocal microscopy

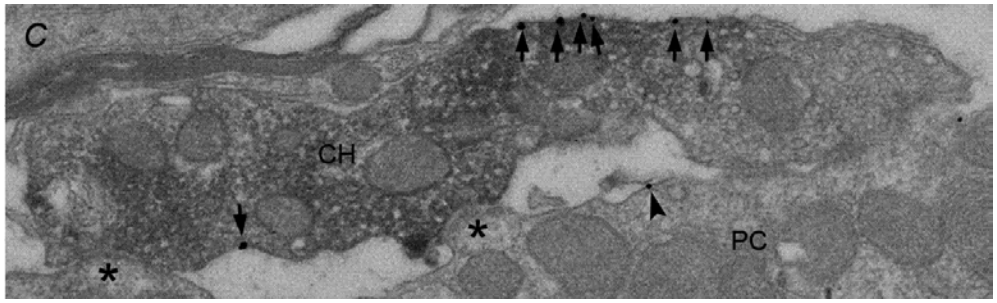


Hruskova et al., J. Neurosci. 2012

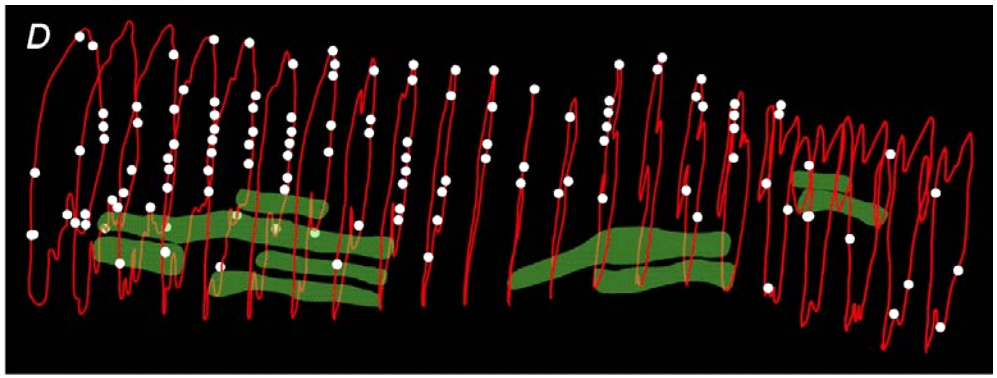
Immunoelectron microscopy







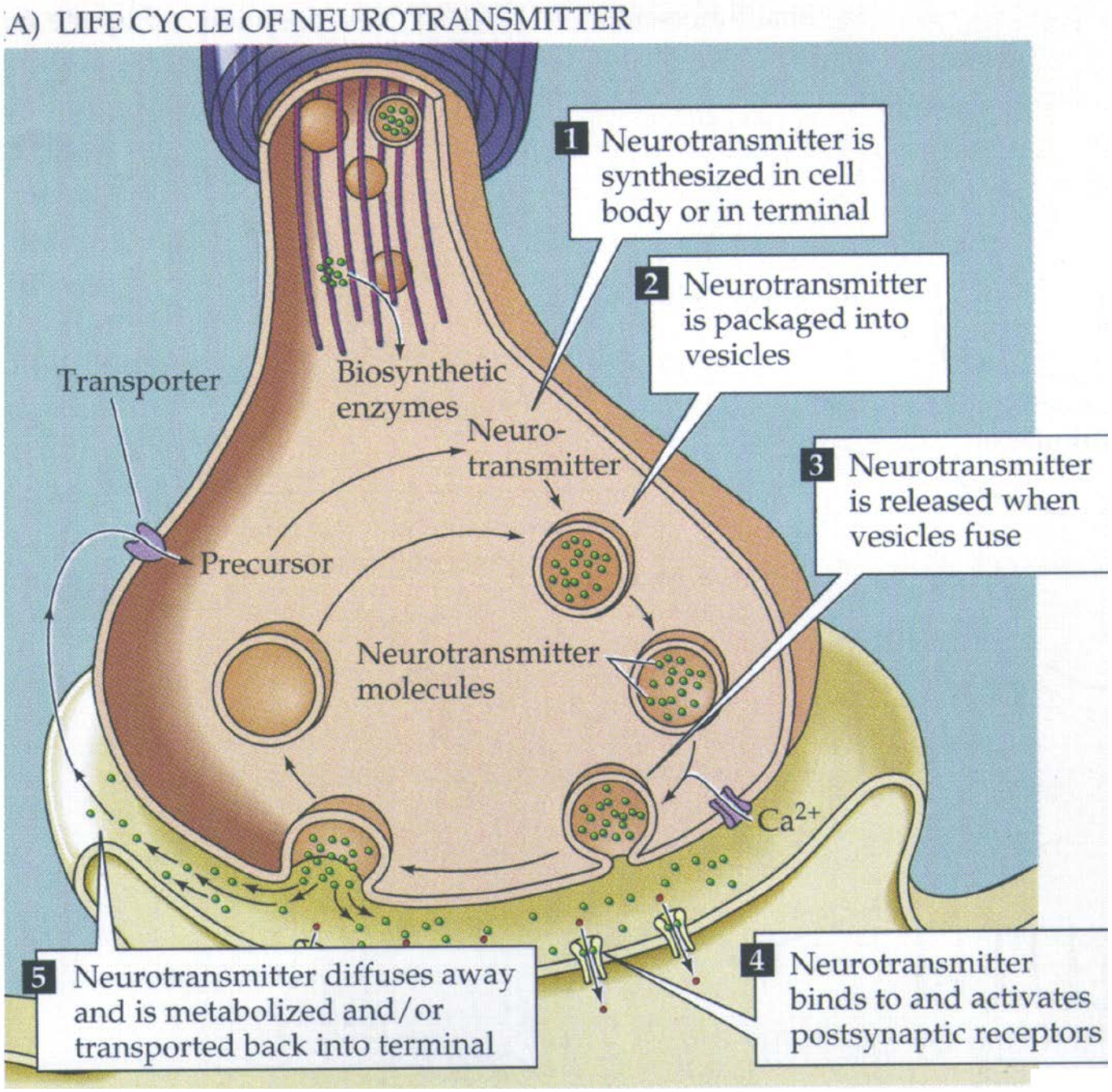
vGluT - peroxidase



GlyRs – immunogold particles

Hruskova et al., J. Neurosci. 2012

Synthesis and storage of NT



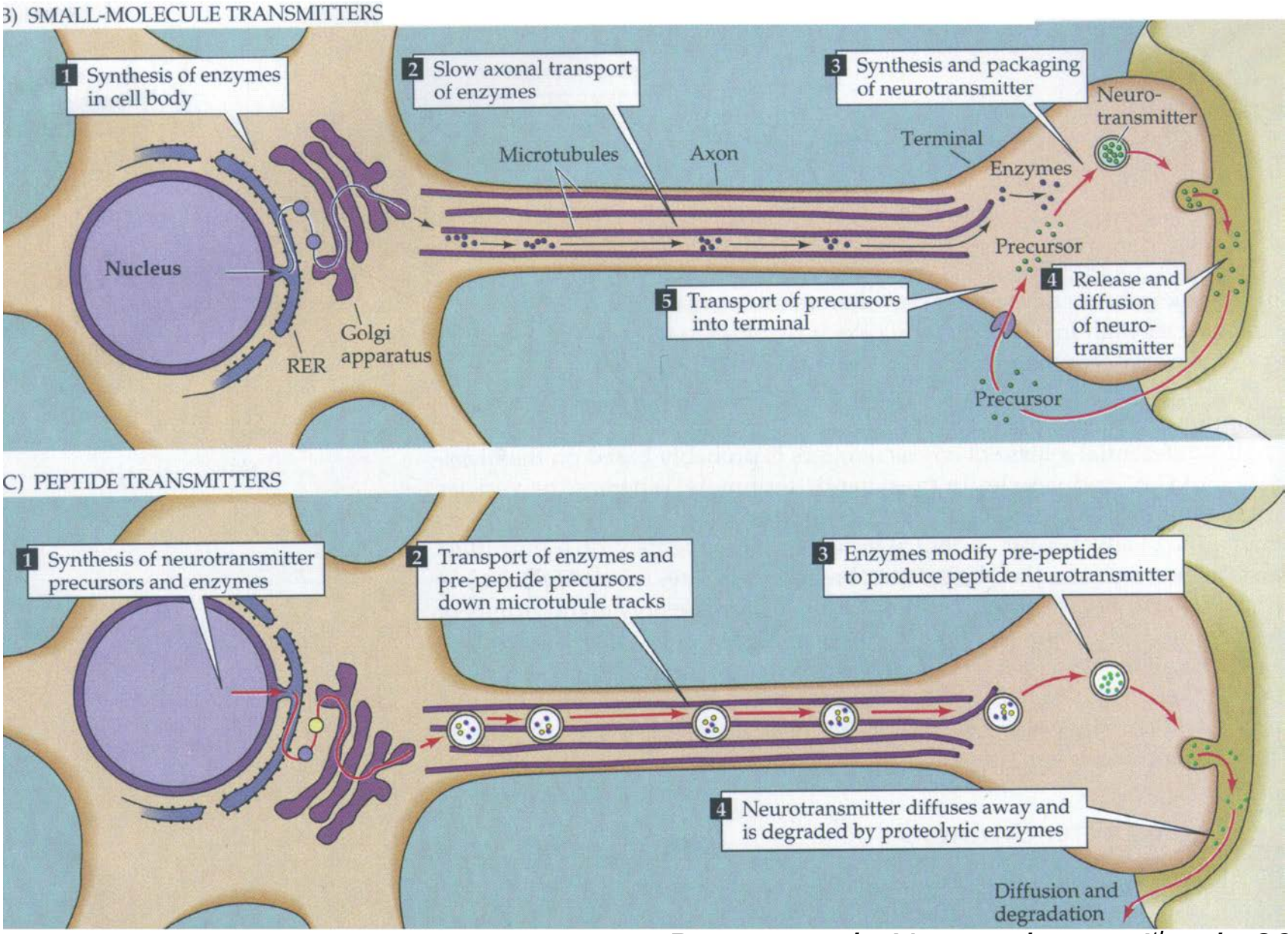
Purves et al., Neuroscience, 4th ed., 2008

Two Major Categories of NT

1. Small-molecule mediators (low molecular weight, e.g. ACh, GABA, glutamate, serotonin, histamine)
2. Neuropeptides (high MW, contain several to tens of AA, e.g. substance P, neuropeptide Y, enkephalin)

Co-transmitters – 1. together with 2. in the same nerve terminal.

Differential synthesis of NTs

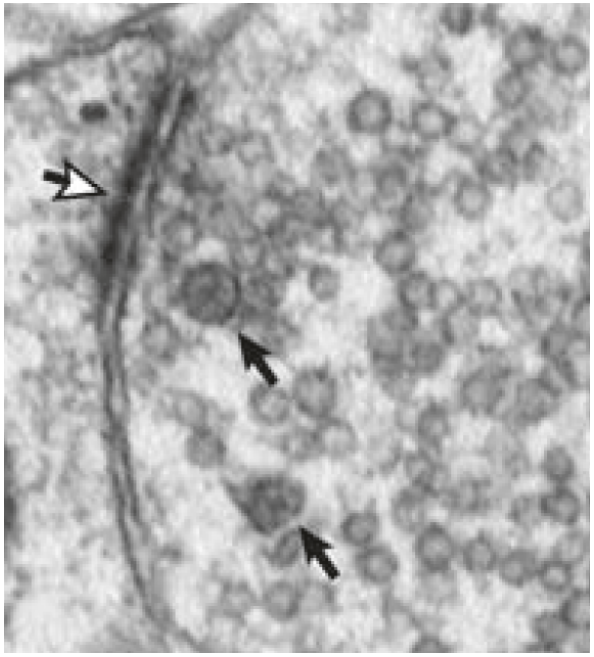


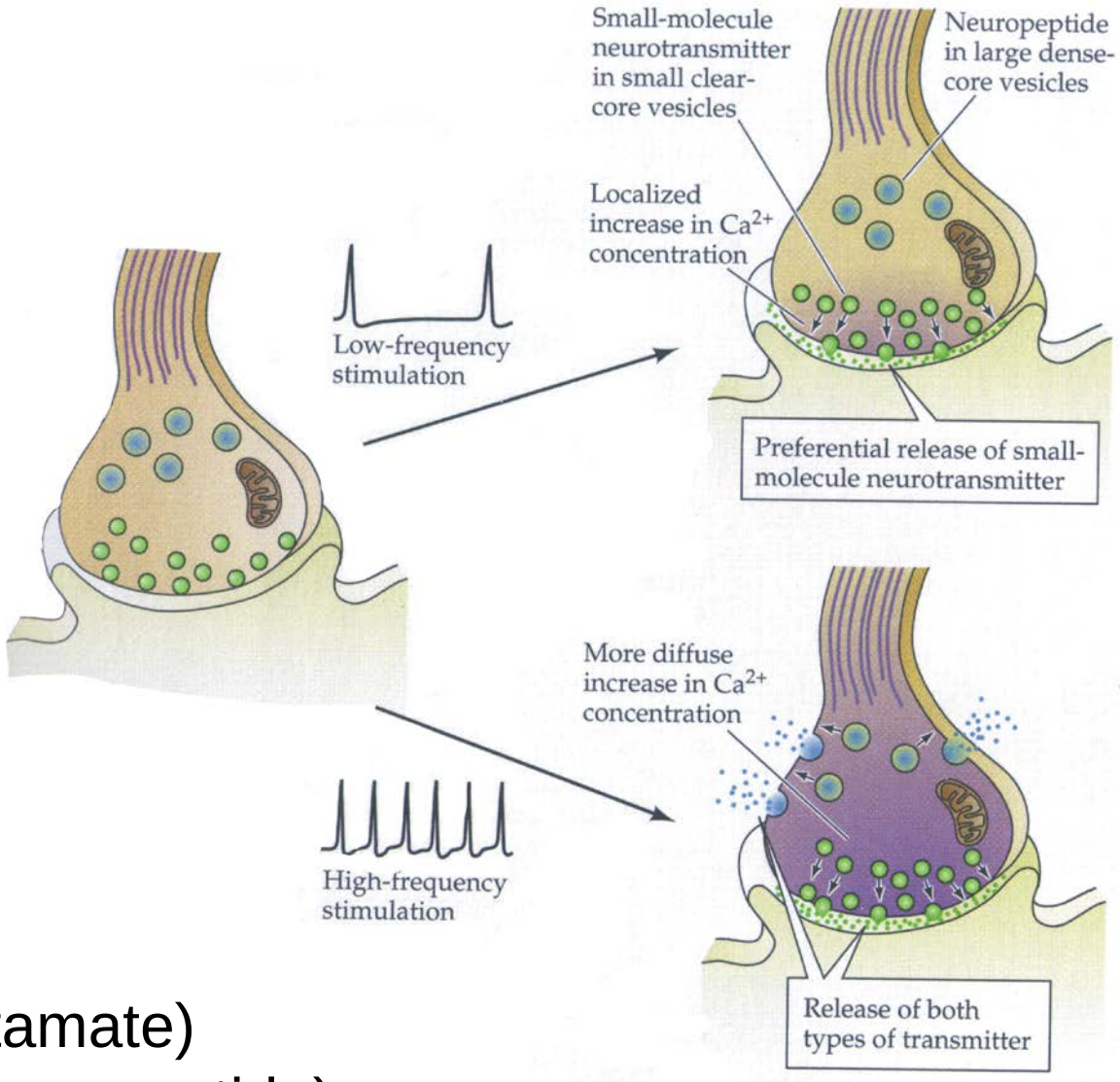
Purves et al., Neuroscience, 4th ed., 2008

Co-transmitters

examples:

GABA/somatostatin
ACh/substance P

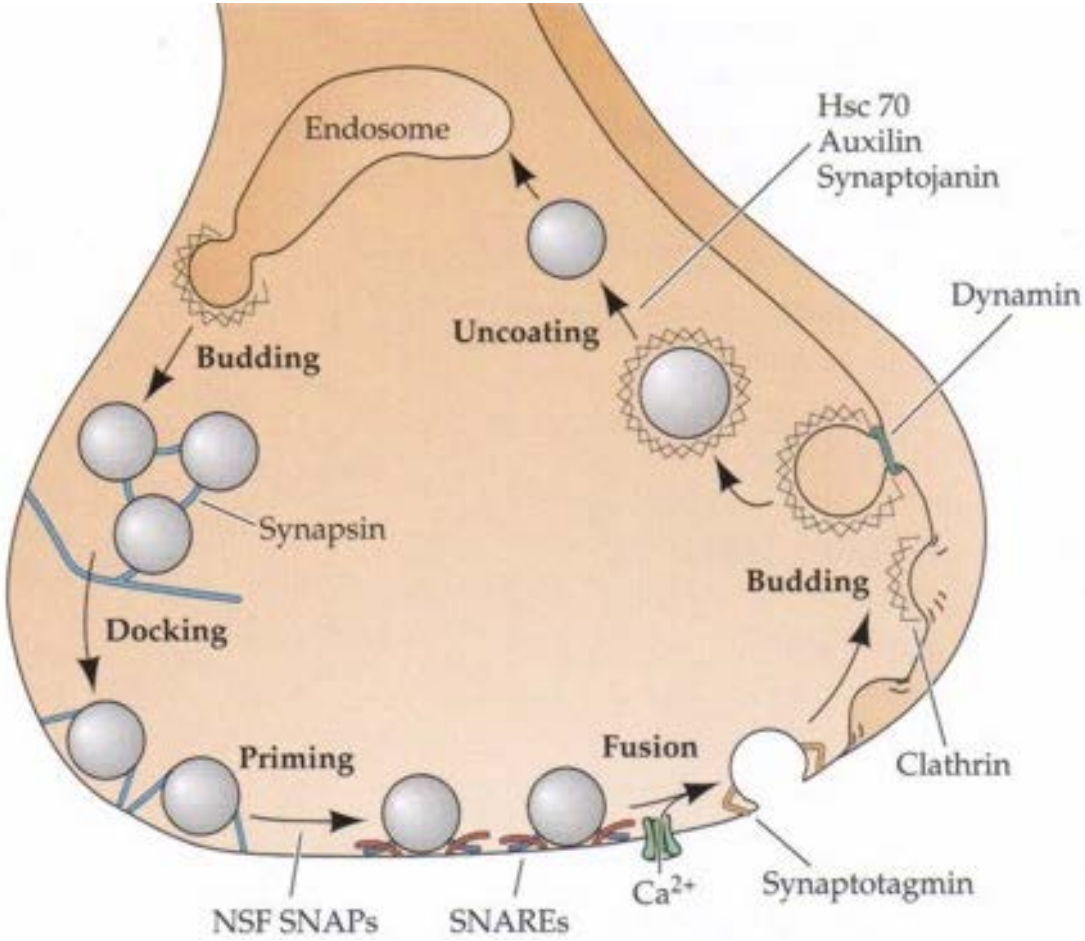


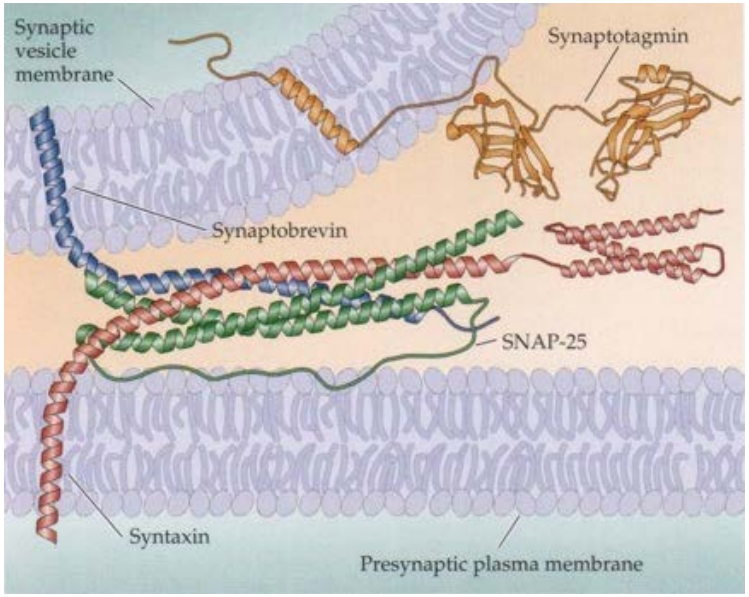


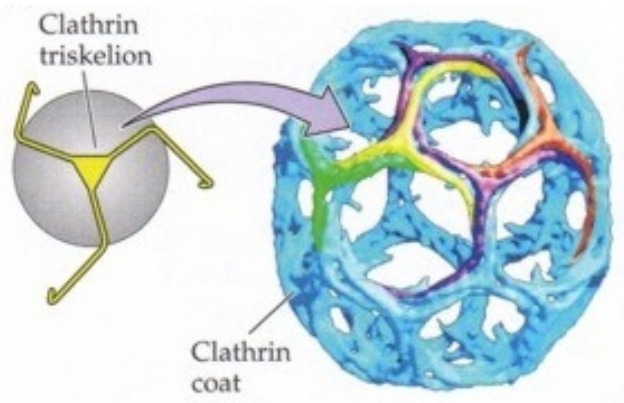
small and clear SV (glutamate)
large dense-core SV (neuropeptide)

Purves et al., Neuroscience, 4th ed., 2008

Synaptic vesicle cycle

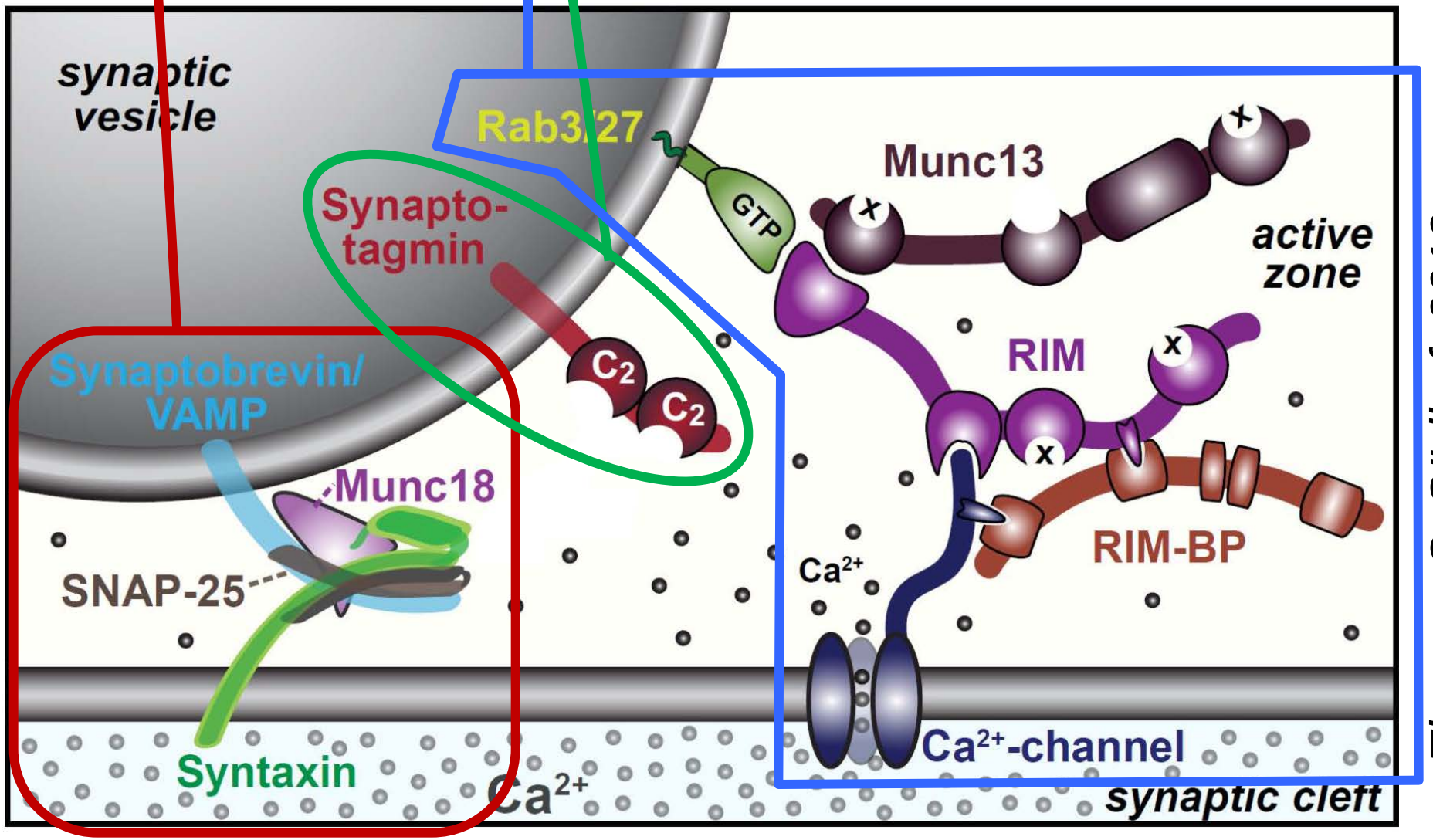






Purves et al., Neuroscience, 4th ed., 2008

Presynaptic release apparatus mediates fusion, Ca^{2+}-triggering and Ca^{2+}-channel tethering

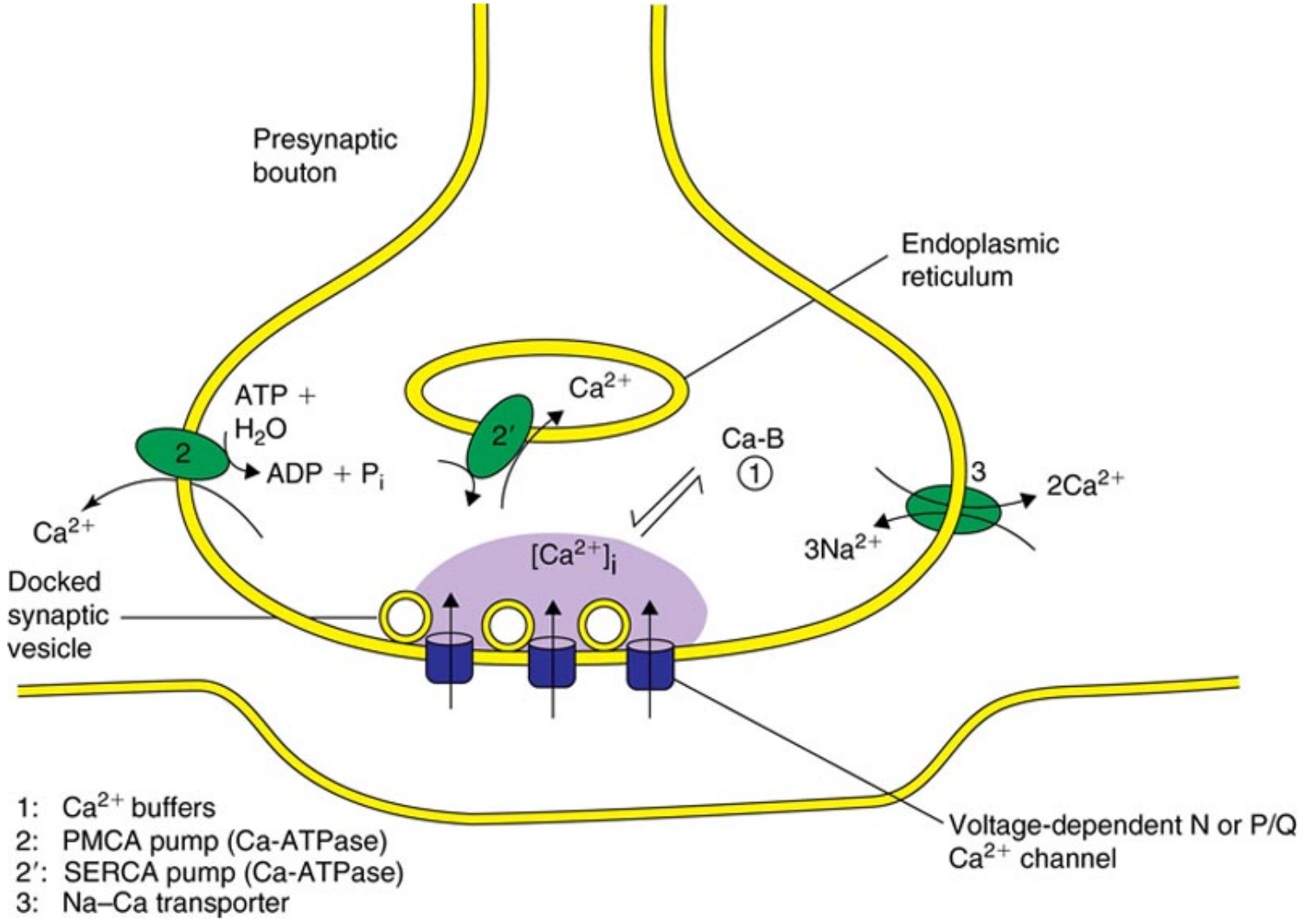


Thomas C. Südhof, 2013

Disturbances in SV exocytosis → neurological disorders

- myasthenic syndromes, defects in synaptic transmission at neuromuscular junctions (-> muscle weakness). Lambert-Eaton myasthenic syndrome (assoc. with lung cancer) – autoimmune reaction to presynaptic Ca2+ channels, reduced ACh release, imunosuppressive drugs
- Congenital myasthenic syndrome – failures in SV cycle - reduced ACh release, inhibitors of AChE

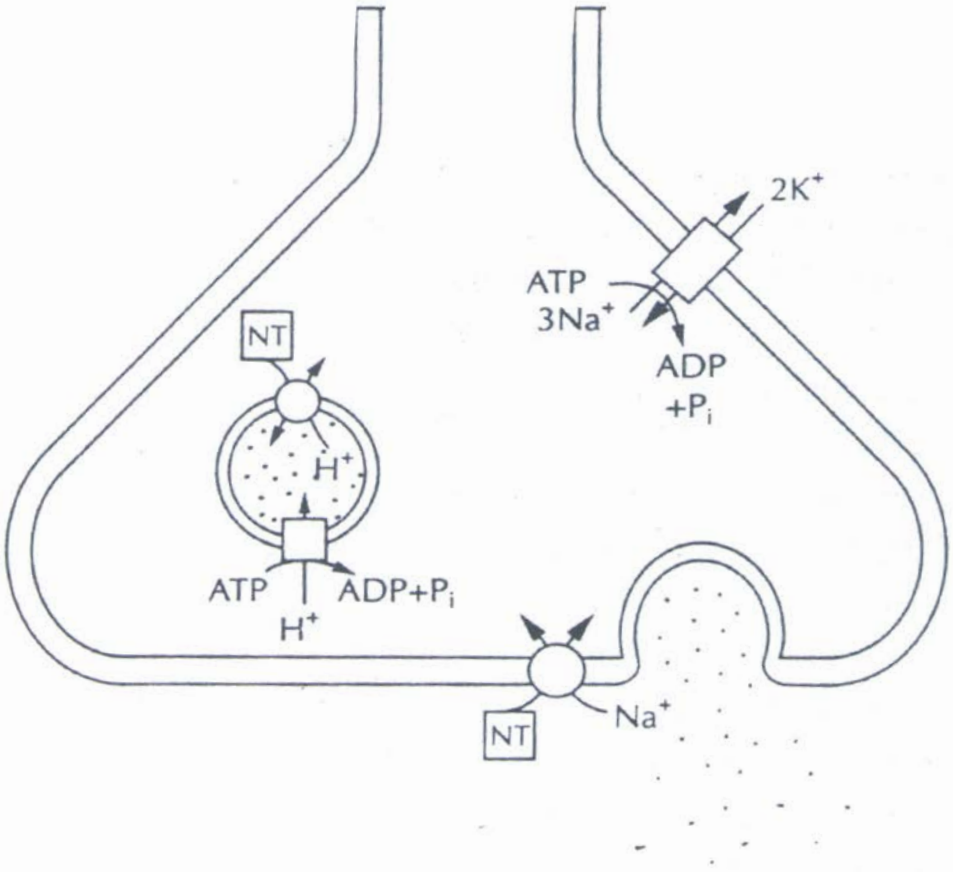
Mechanisms of presynaptic Ca^{2+} clearance



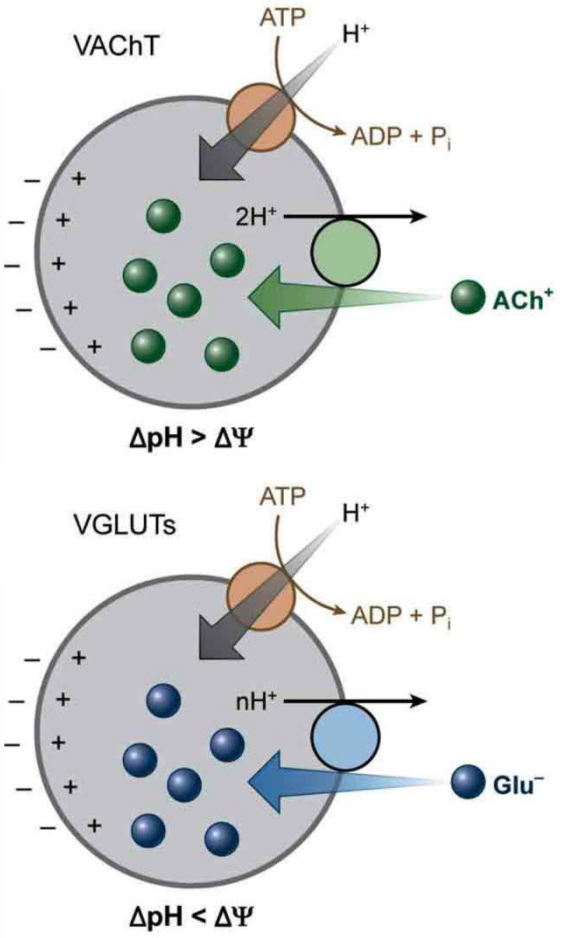
Hammond, Cell. Mol. Neurophys, 4th ed., 2015

Mechanisms of NT re-uptake

Specific transport proteins use presynaptic Na+ gradient

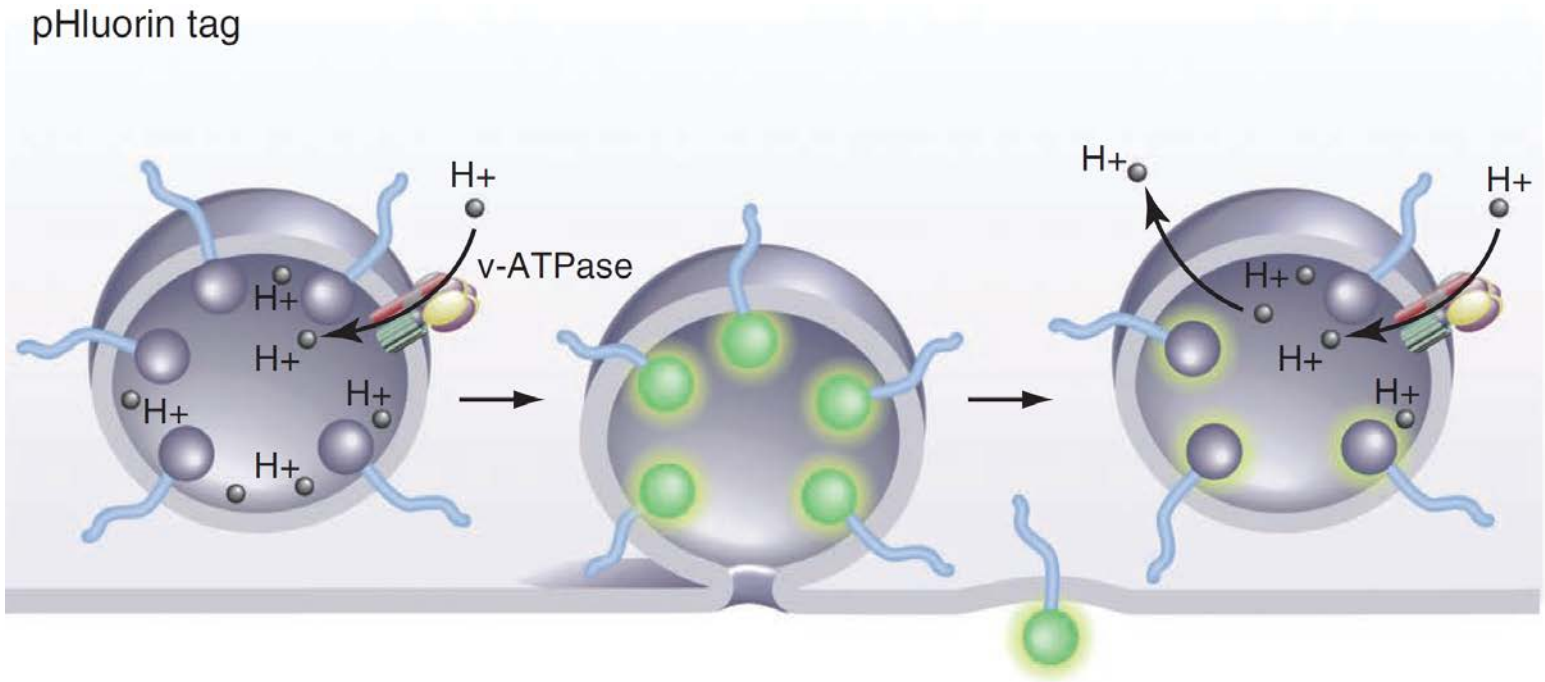


Vesicular transporters use electrochemical gradients: pH (ΔpH) and electrical (ΔΨ)



VanLiefferinge, Front. Cell. Neur. 2013

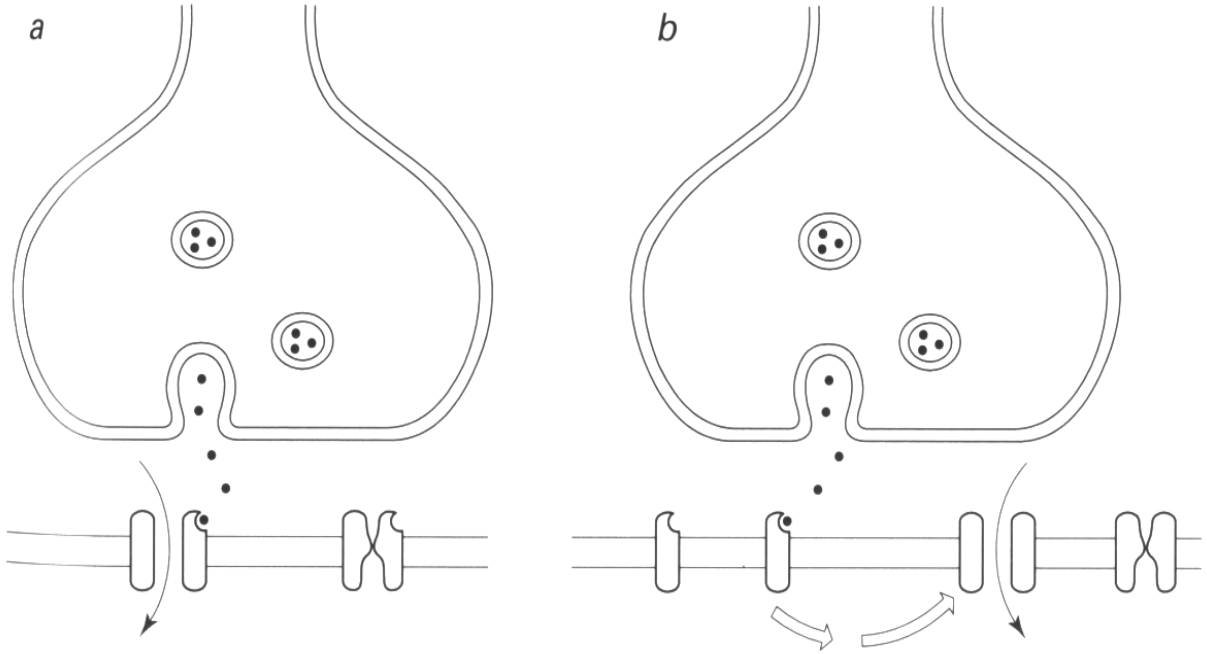
Optogenetics: monitoring of SV and NT cycles



Kavalali, Nat. Neurosci., 2013

Postsynaptic receptors

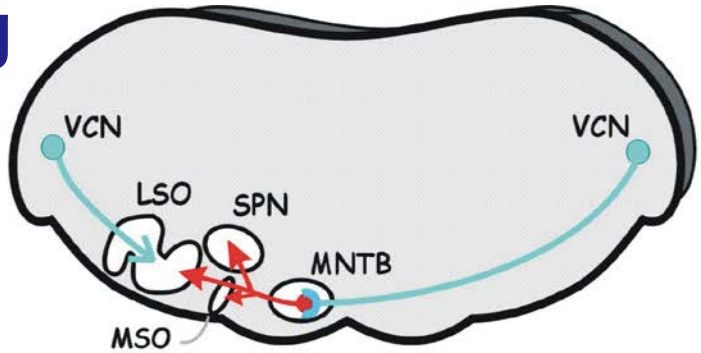
Synaptic transmission: - fast: ligand-gated ion channels
- slow: metabotropic receptors

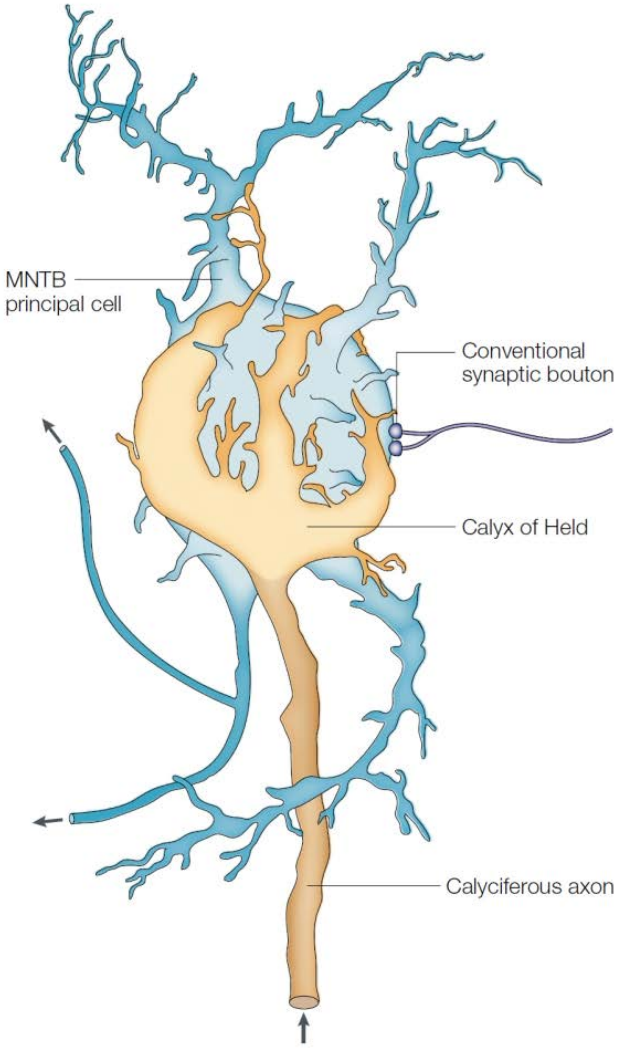


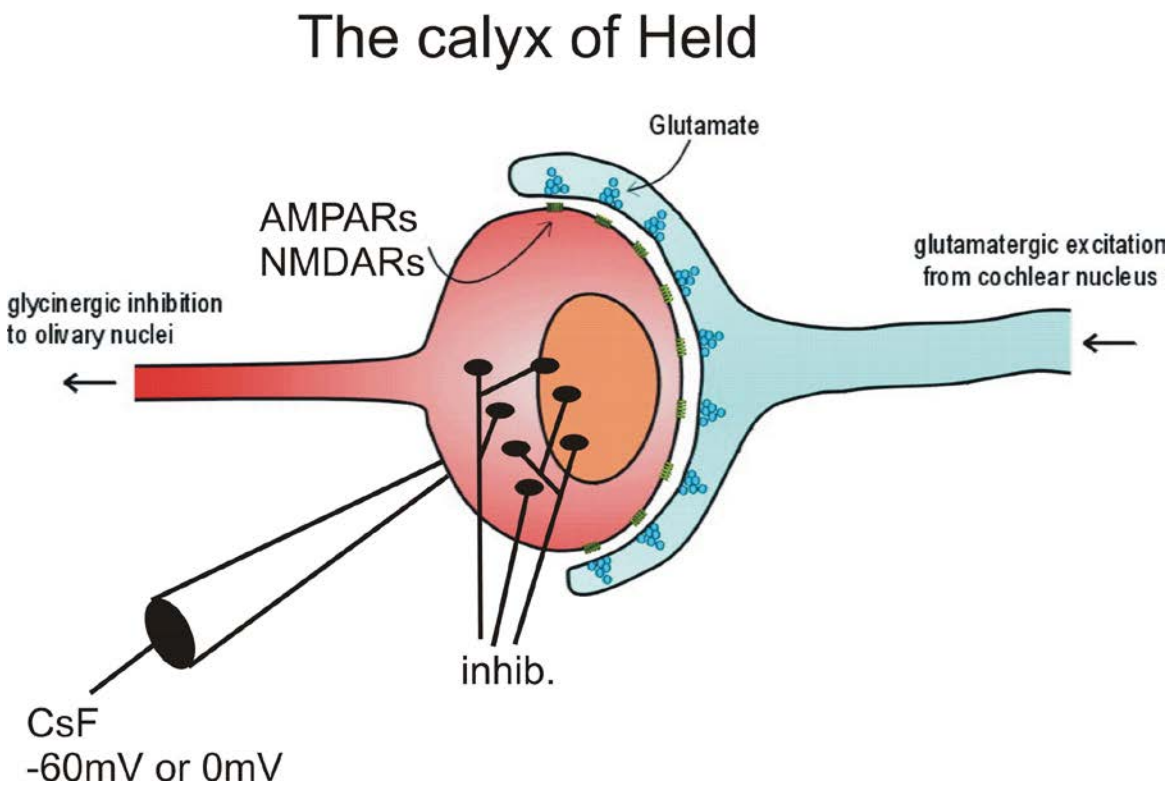
Ligand-gated ion channels

3 families:
-cys-loop (GABA-A, GlyR, 5-HT, nAChR)
-Glutamate (AMPA, KR, NMDA)
-ATP (P2X)

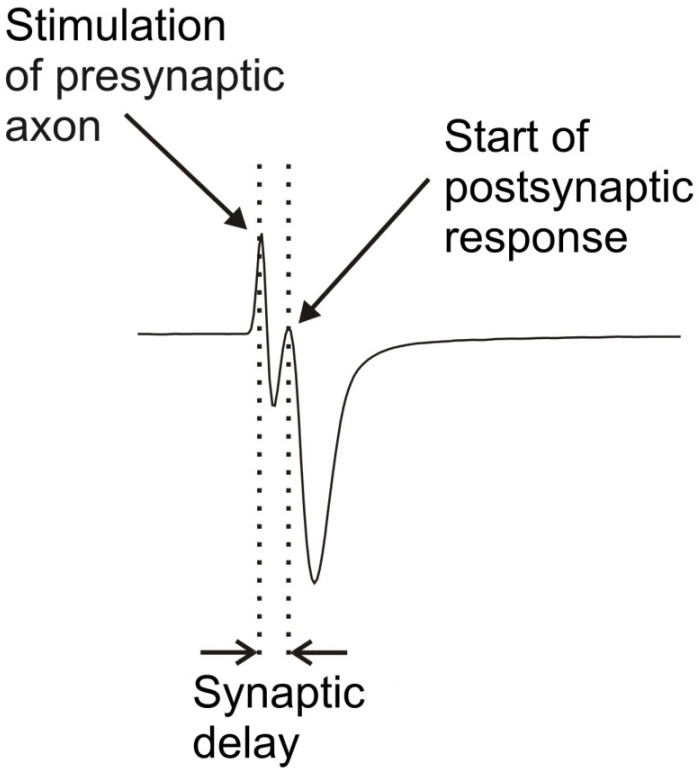
Fast synaptic transmission is typically studied using live brain slices

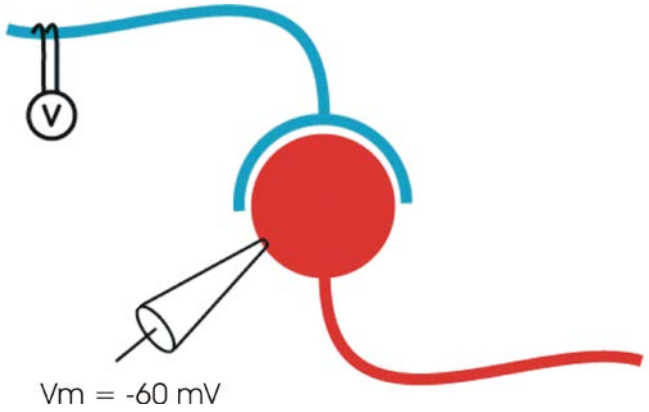


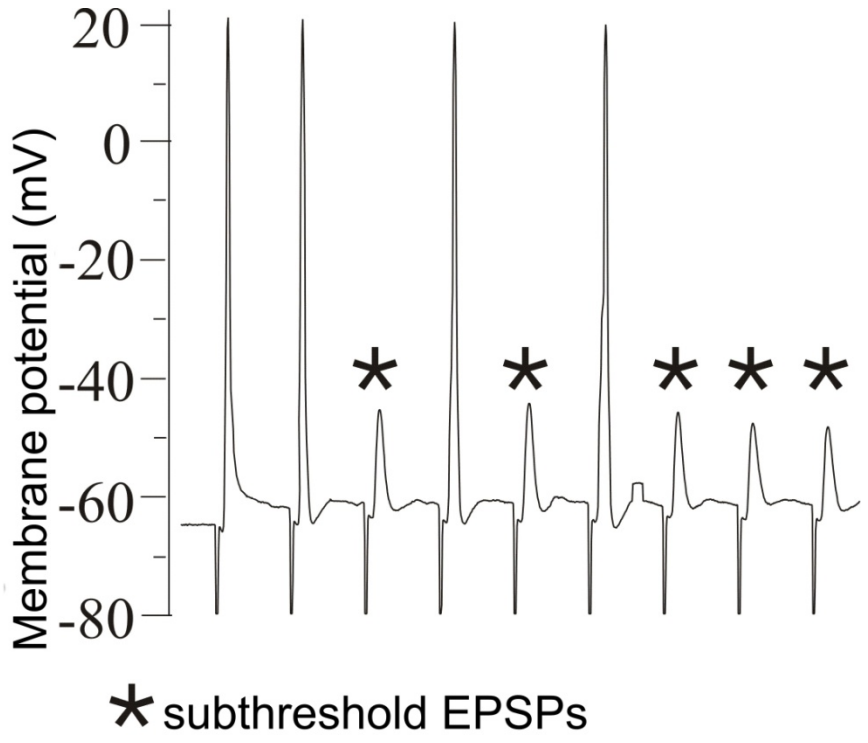




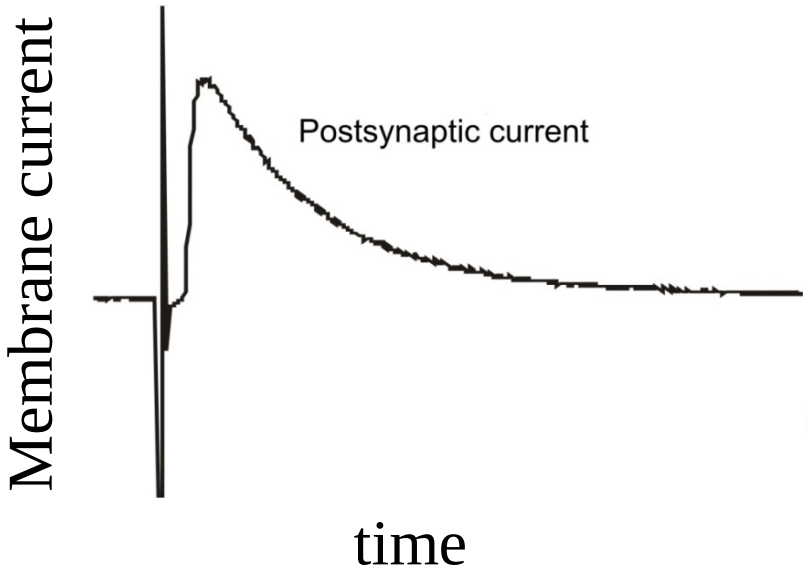
Excitatory postsynaptic current (EPSC) and potential (EPSP)

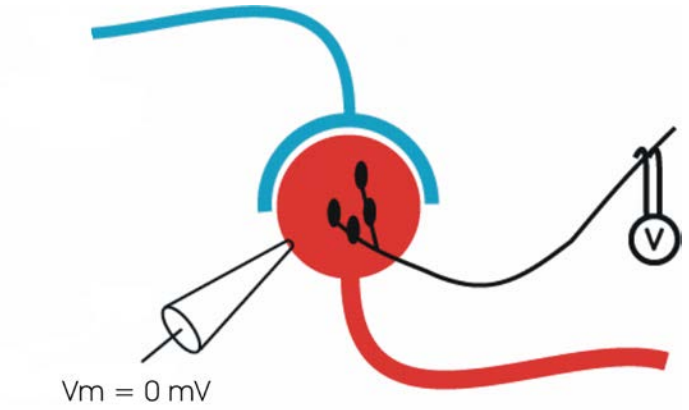


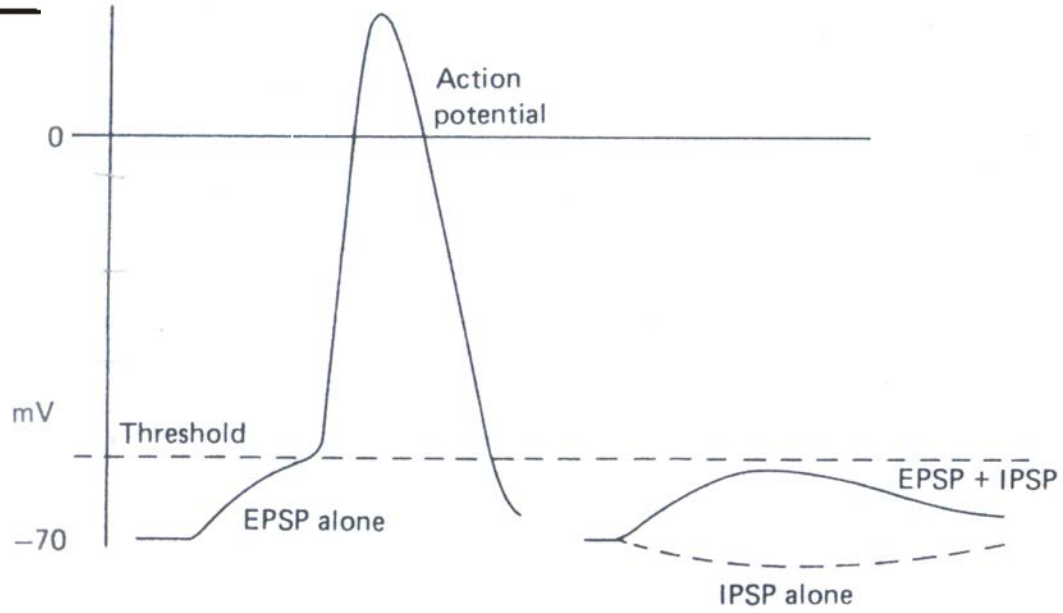




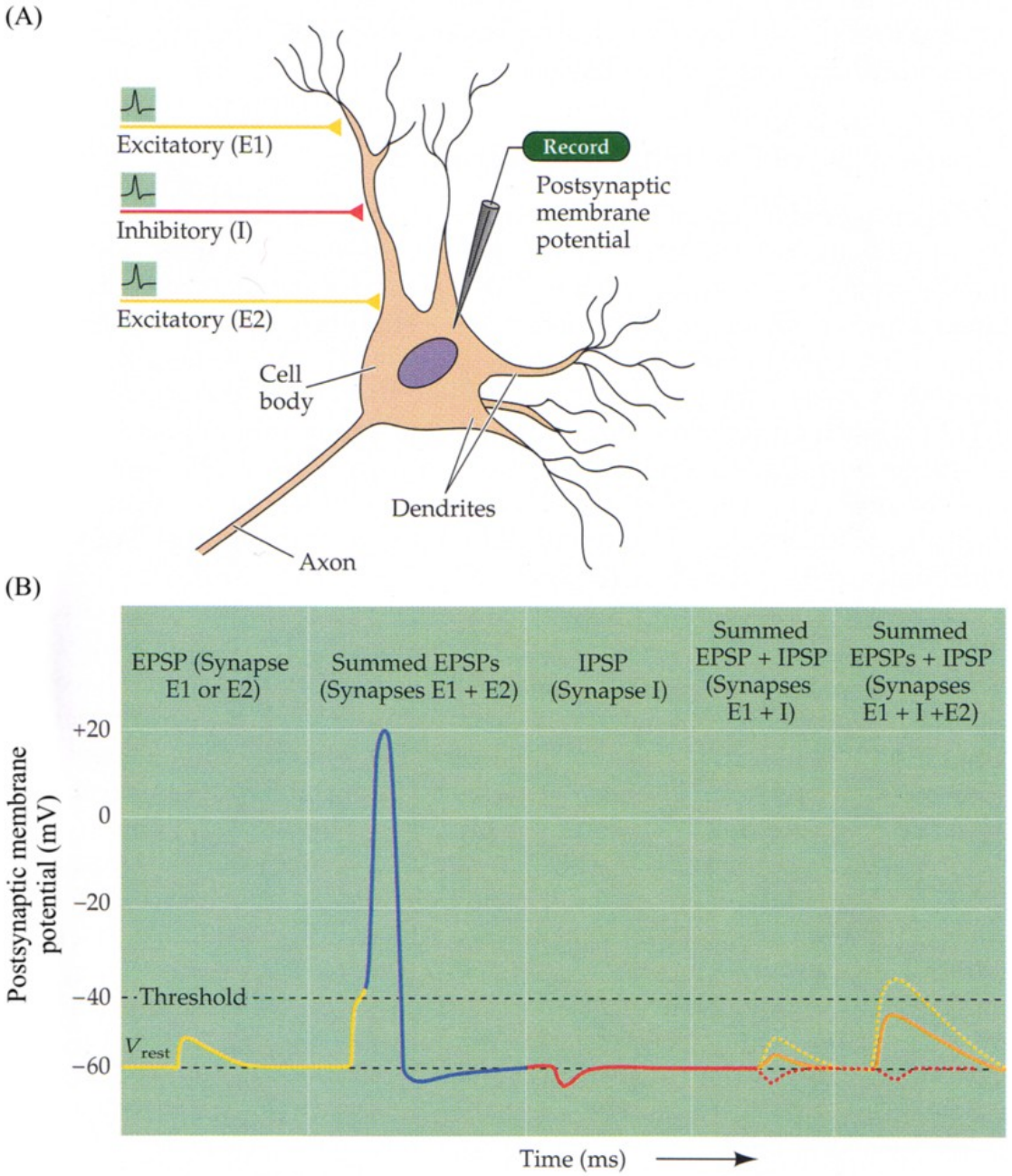
Inhibitory postsynaptic current (IPSC) and potential (IPSP)







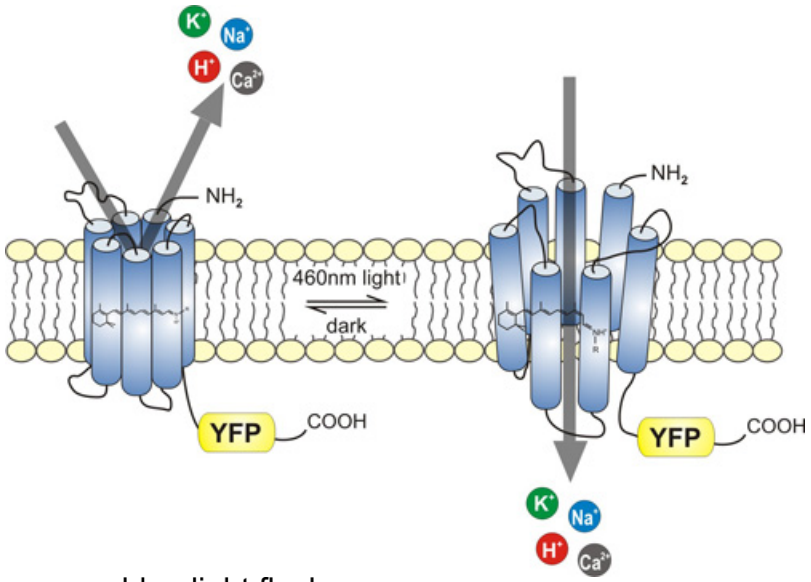
Summation of postsynaptic potentials

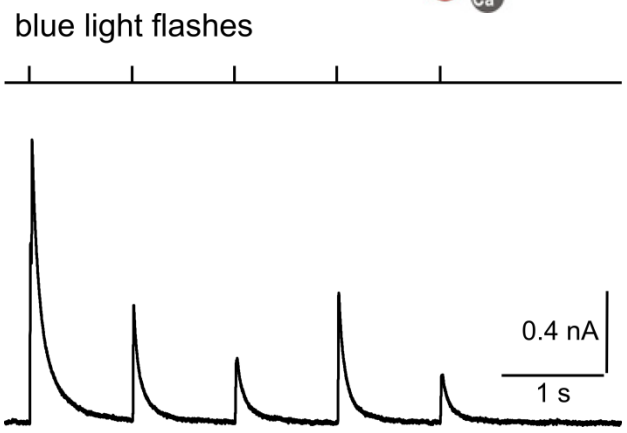


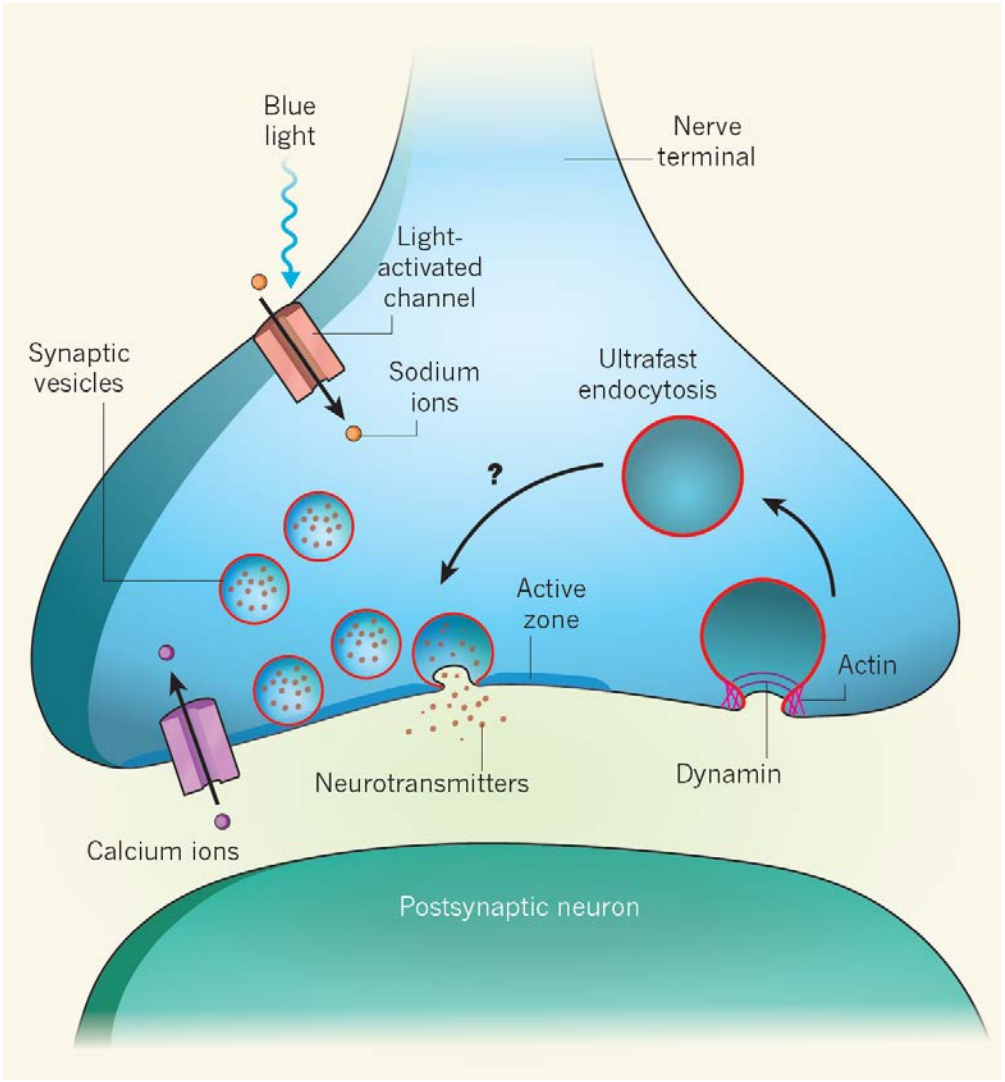
Purves et al., Neuroscience, 4th ed., 2008

Optogenetic techniques used to stimulate synaptic transmission

Channelrhodopsin 2







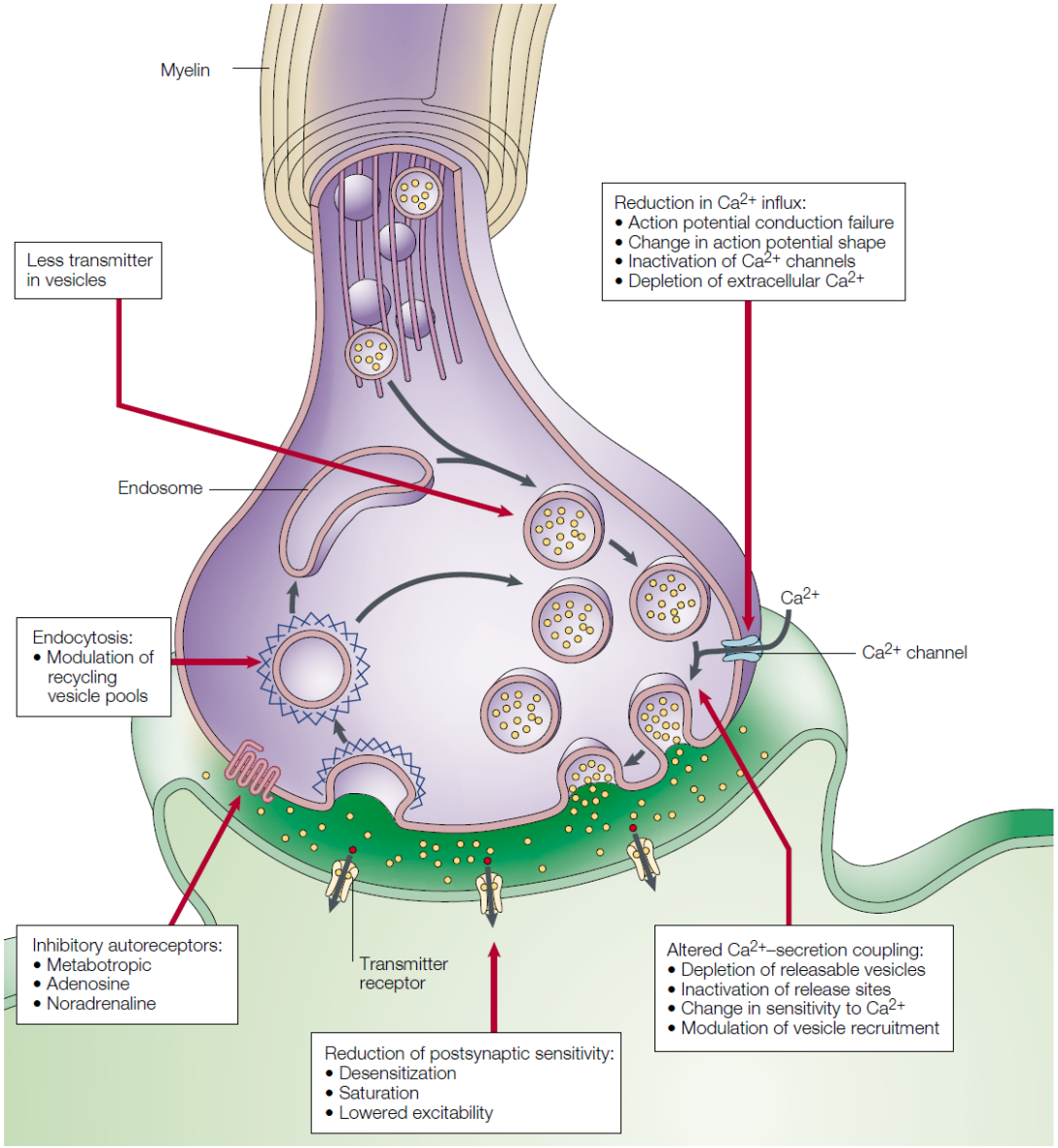
Nagel et al., PNAS, 2003

Plasticity of synaptic transmission

SV cycle vs. NT cycle

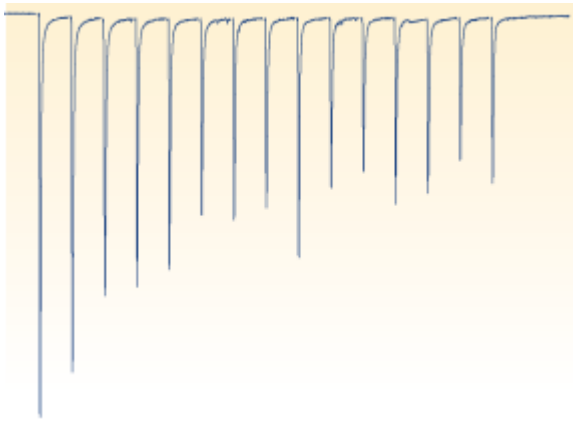
- short-term plasticity
  vs.
  long-term plasticity

- presynaptic mechanisms
  vs.
  postsynaptic
  mechanisms

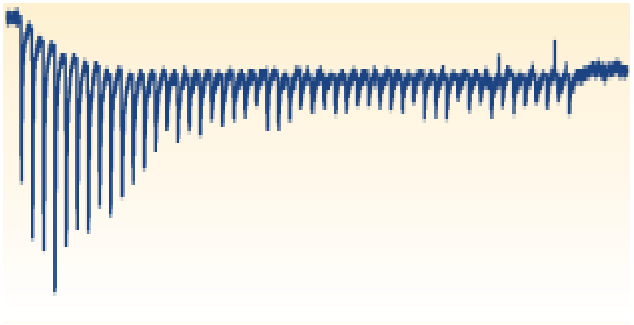


von Gersdorff and Borst, Nat. Rev. Neur. 2001

High frequency synaptic transmission



Short-term depression – repeated stimulation of presynaptic neuron leads to transient depletion of synaptic vesicles. Gradual decrease of EPSC amplitude.
Postsynaptic depression – desensitization of postsynaptic receptors

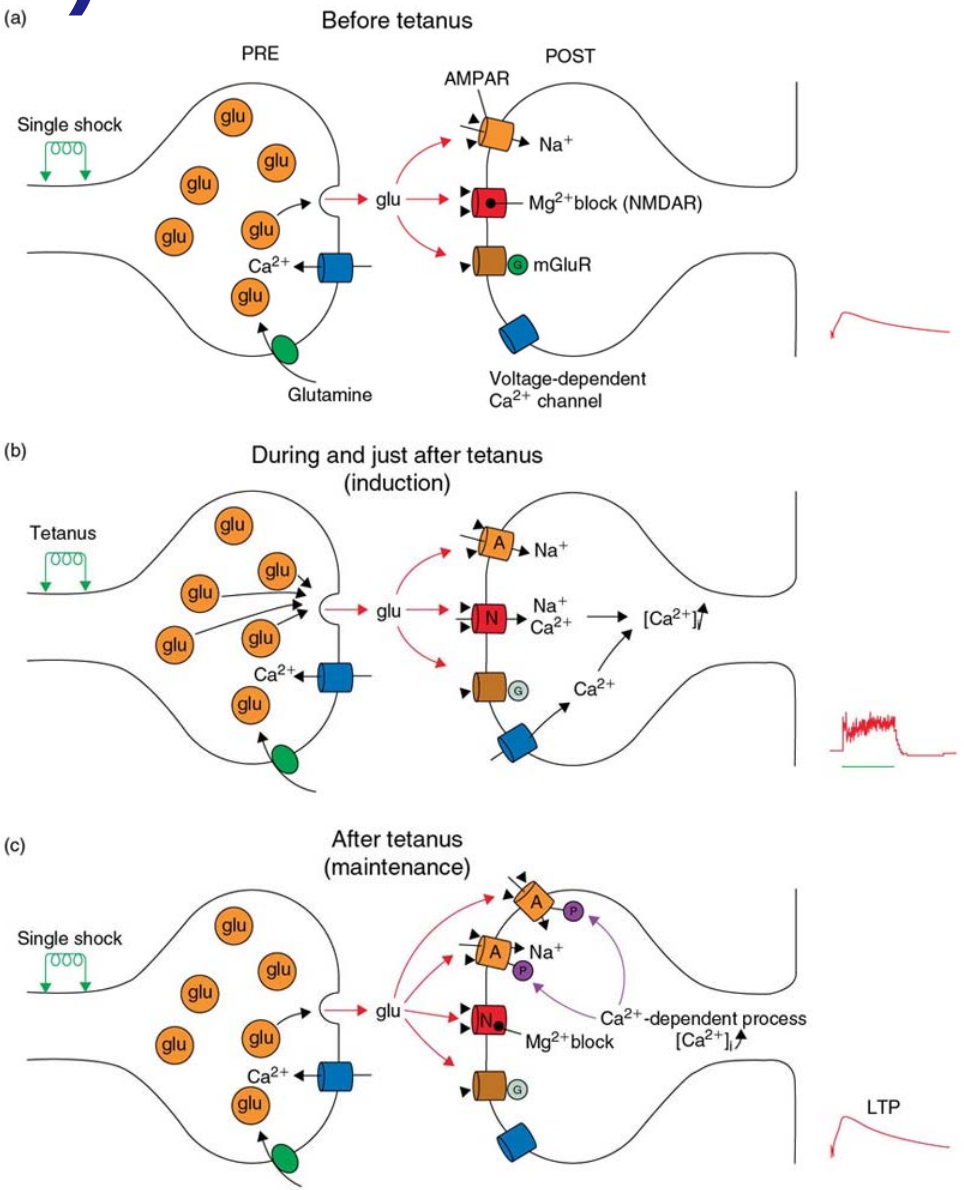


Facilitation –high-frequency stimulation causes transient accumulation of Ca in presynaptic neuron and increase of presynaptic release probability.

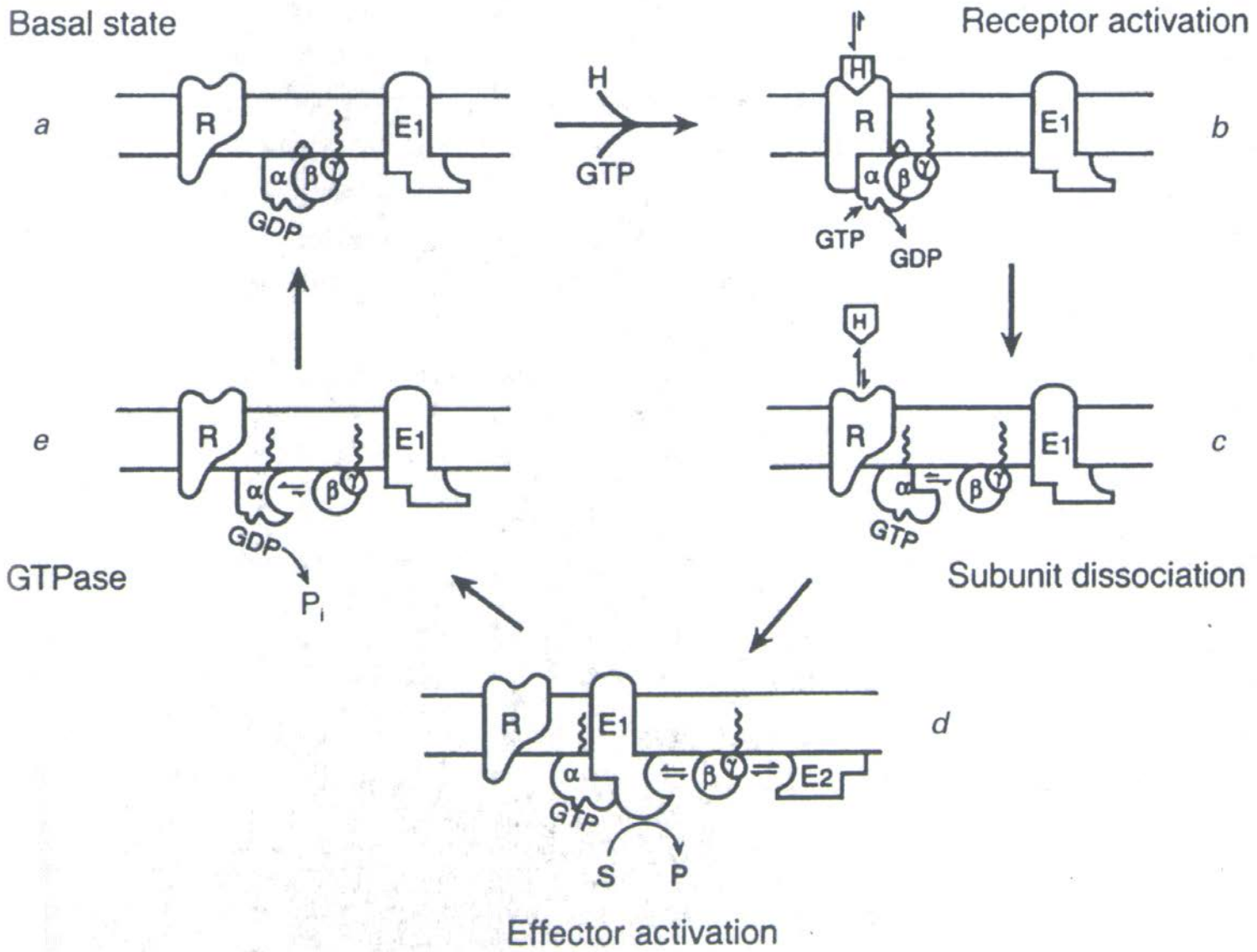
Long term potentiation (LTP) and depression (LTD)

Silent synapses (NMDA-dependent LTP)

High-frequency stimulation – LTP.
Low-frequency stimulation - LTD

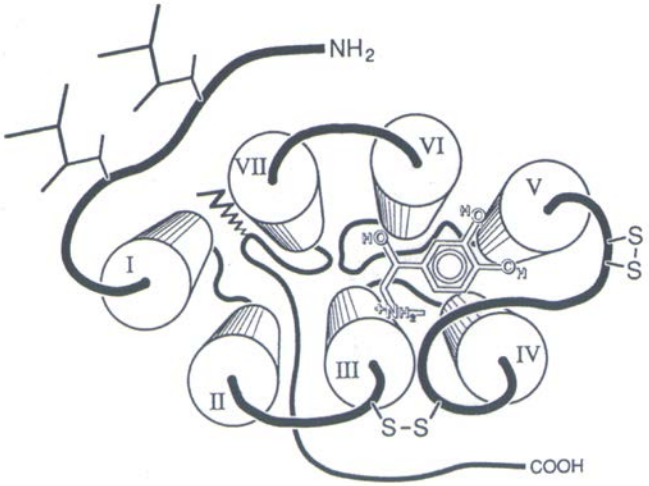


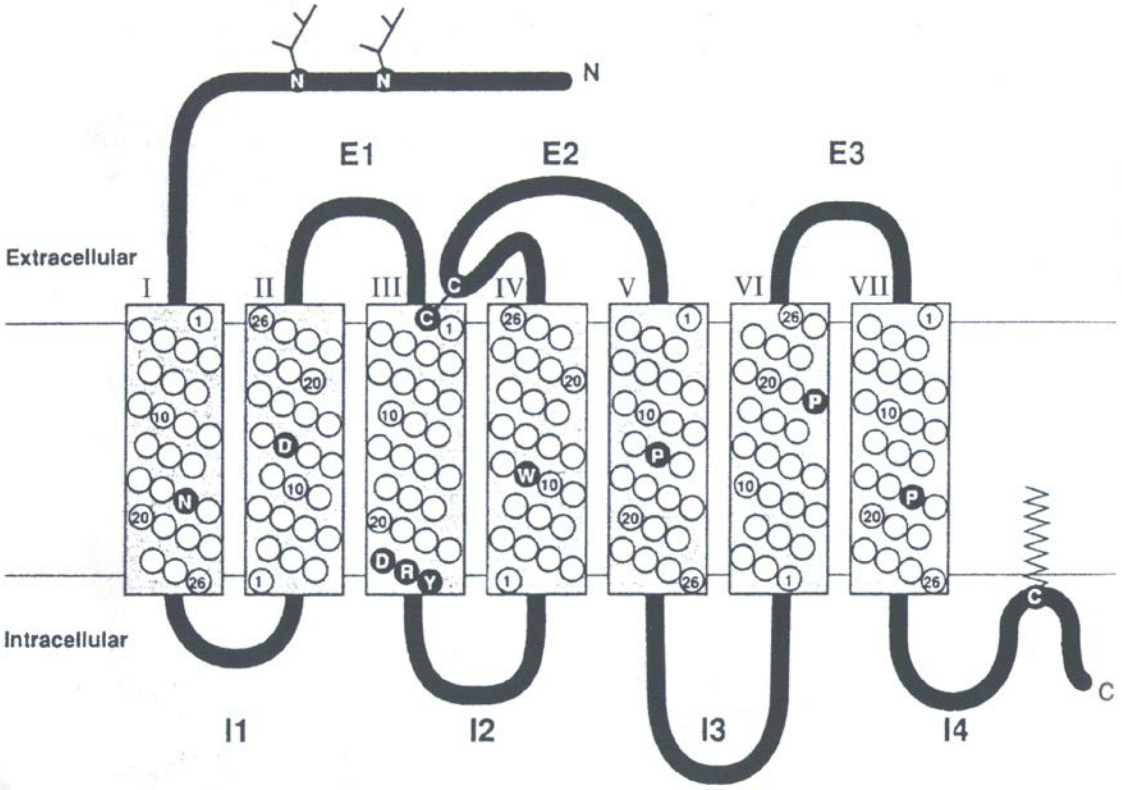
Slow synaptic transmission and G-protein coupled receptors (GPCRs)



Topology of GPCRs (heptahelical Rs)

7 TM domains, 3 extracellular (agonist binding site, low M) and 3-4 intracellular loops (G-protein interactions)





GPCR classes

- A (rhodopsin-like) (acetylcholine, amines, peptides)
- B (secretin receptor family) (peptides, hormones)
- C (mGluR/pheromone) (glutamate, GABA)
- D (pheromone)
- E (cAMP receptors)
- Frizzled

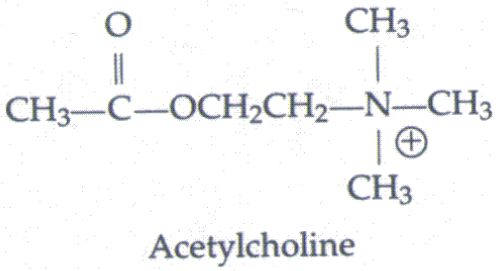
Neurotransmitters

1) Acetylcholine
2) Biogennic amines
3) Glutamate
4) GABA and glycin
5) Purines
6) Neuropeptides
7) Endocannabinoides
8) Gas mediators

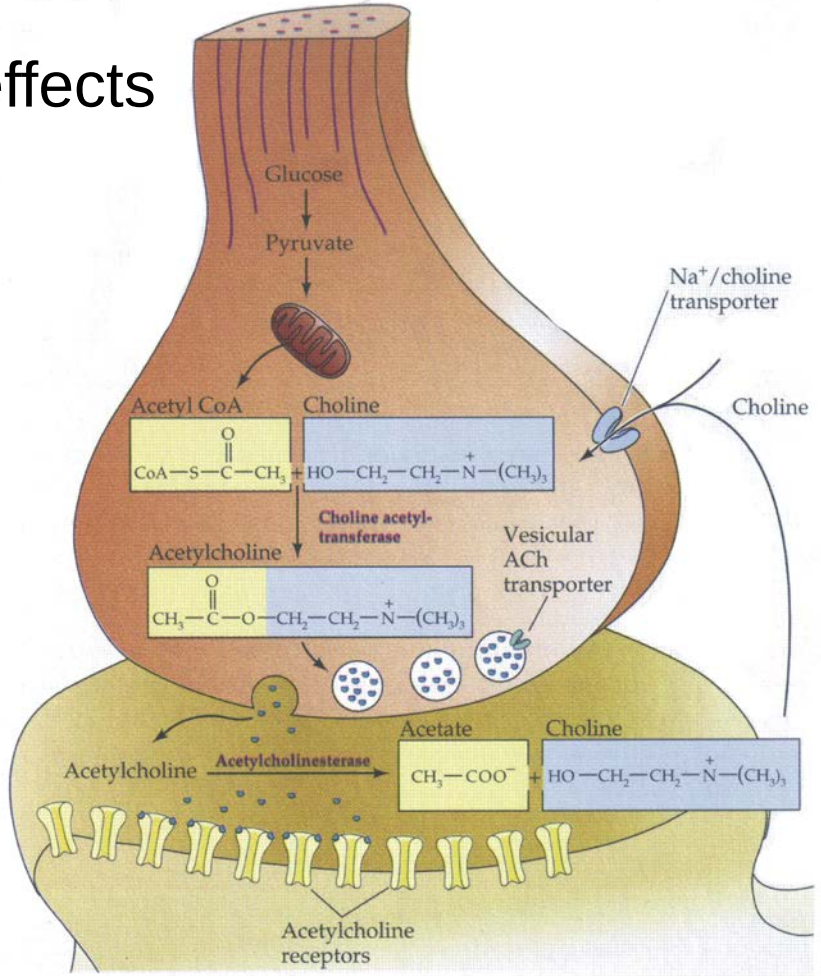
Acetylcholine

-Neuromuscular junctions, ganglia of visceral motor system, CNS
-Excitatory (mainly) and inhibitory effects

-Precursors: Cholin + acetyl CoA
-Inactivation: ACh esterase



Acetylcholine



Purves et al., Neuroscience, 4th ed., 2008

Acetylcholine receptors

1) Nicotinic, nAChR (ionotropic)
   agonist: nicotine (leaves of Nicotinia tabacum)
   antagonist: tubocurarine (bark of Chondrodendron tomentosum)

2) Muscarinic, mAChR(metabotropic) GPCR
   agonist: muscarine (Amanita muscaria)
   antagonist: atropine (Atropa belladona)

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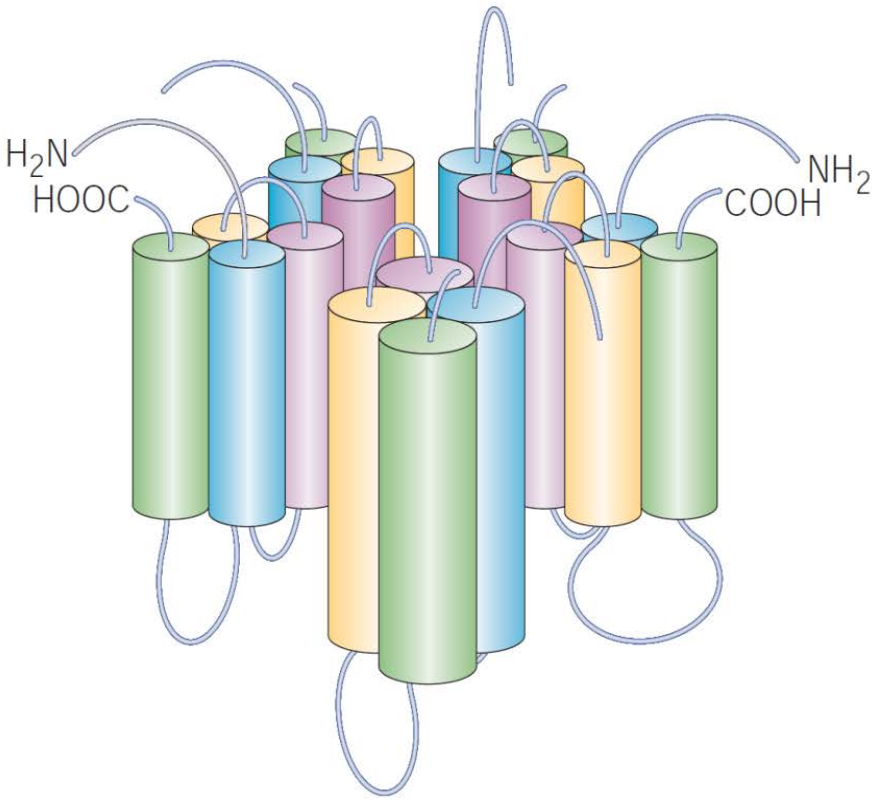
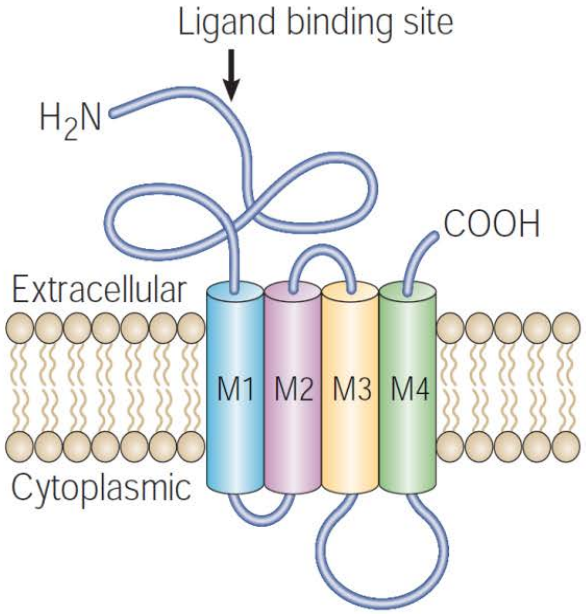
   agonist: muscarine (Amanita muscaria)
   antagonist: atropine (Atropa belladona)

nAChR:

Subunits:
-α1-10 (agonist binding)
-β1-4
-γ
-δ
-ε





Laviolette and van der Kooy, Nat. Rev. Neurosci 2004

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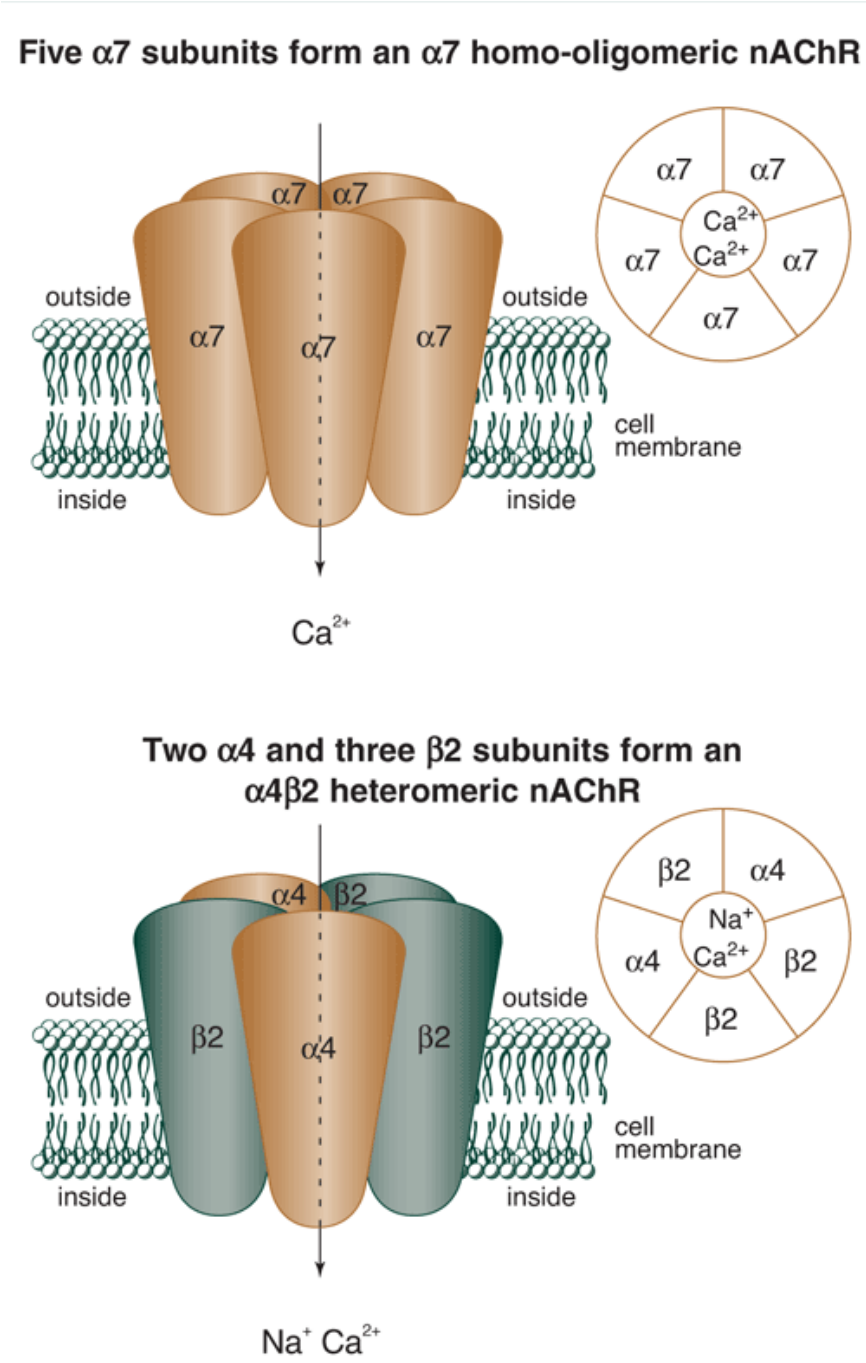
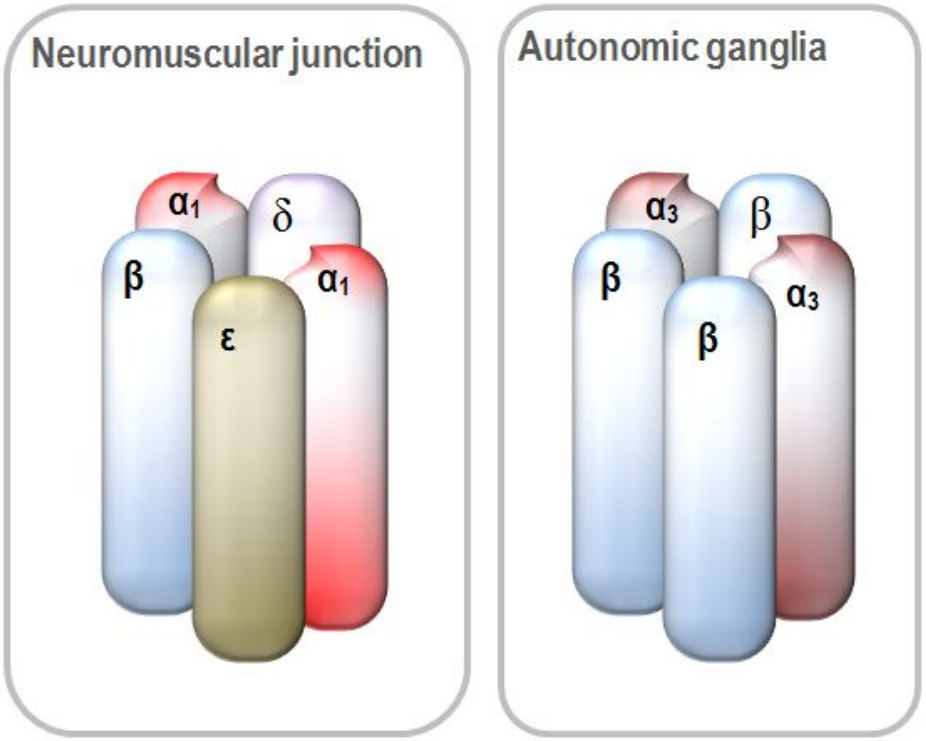


Davis and de Fiebre, Alcohol Res. Health 2006

nAChR:



Variable composition of native receptors:

- Homopentamers of α7, α8, α9 – in CNS
- Heteropentamers, e.g. α1β1ε(γ)δ – neuromuscular junction
  Embryonic (γ) vs adult (ε)
- α2-10, β2-4 neuronal subtypes

Deranged Physiology 2015

mAChR:

1. **inhibit adenylyl cyclase**

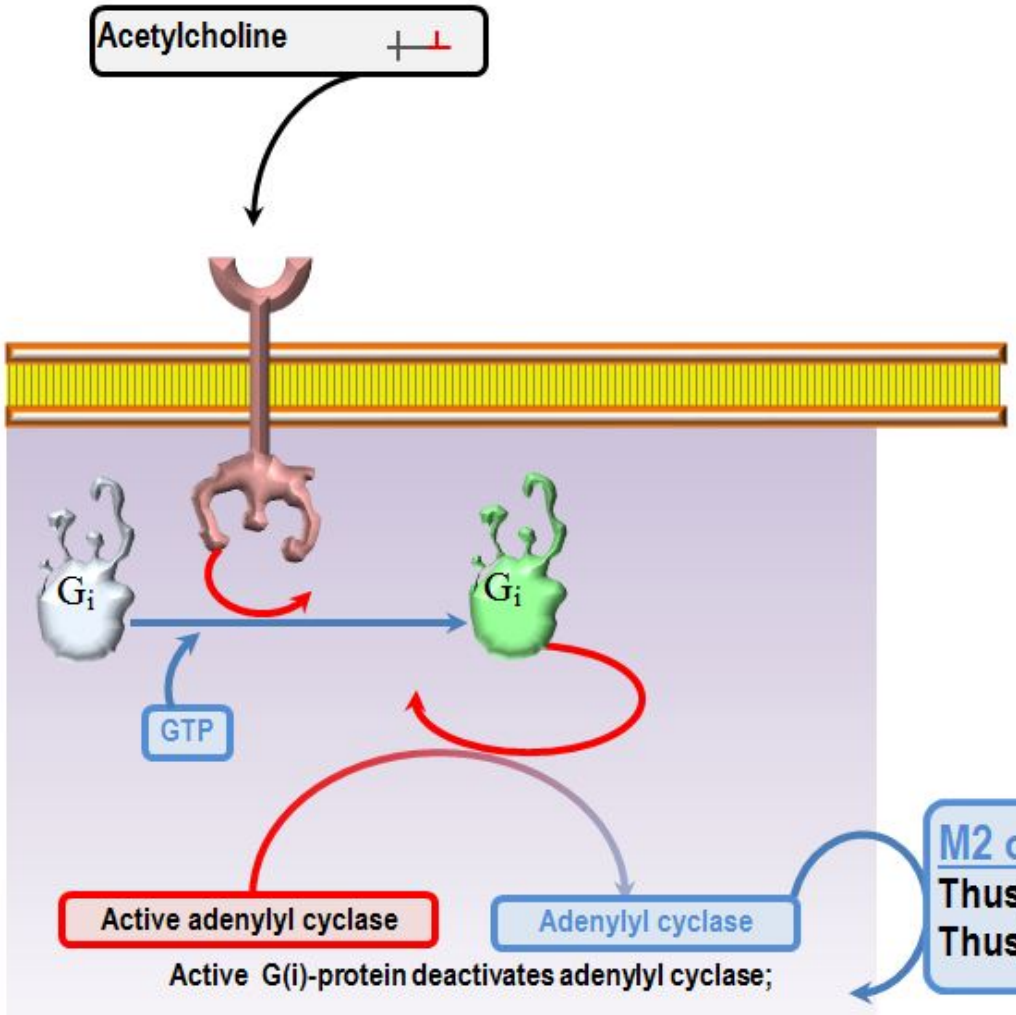
   (α: M2, M4)

2. stimulate phospholipase C

   (M1, M3, M5)

3. regulate ion channels

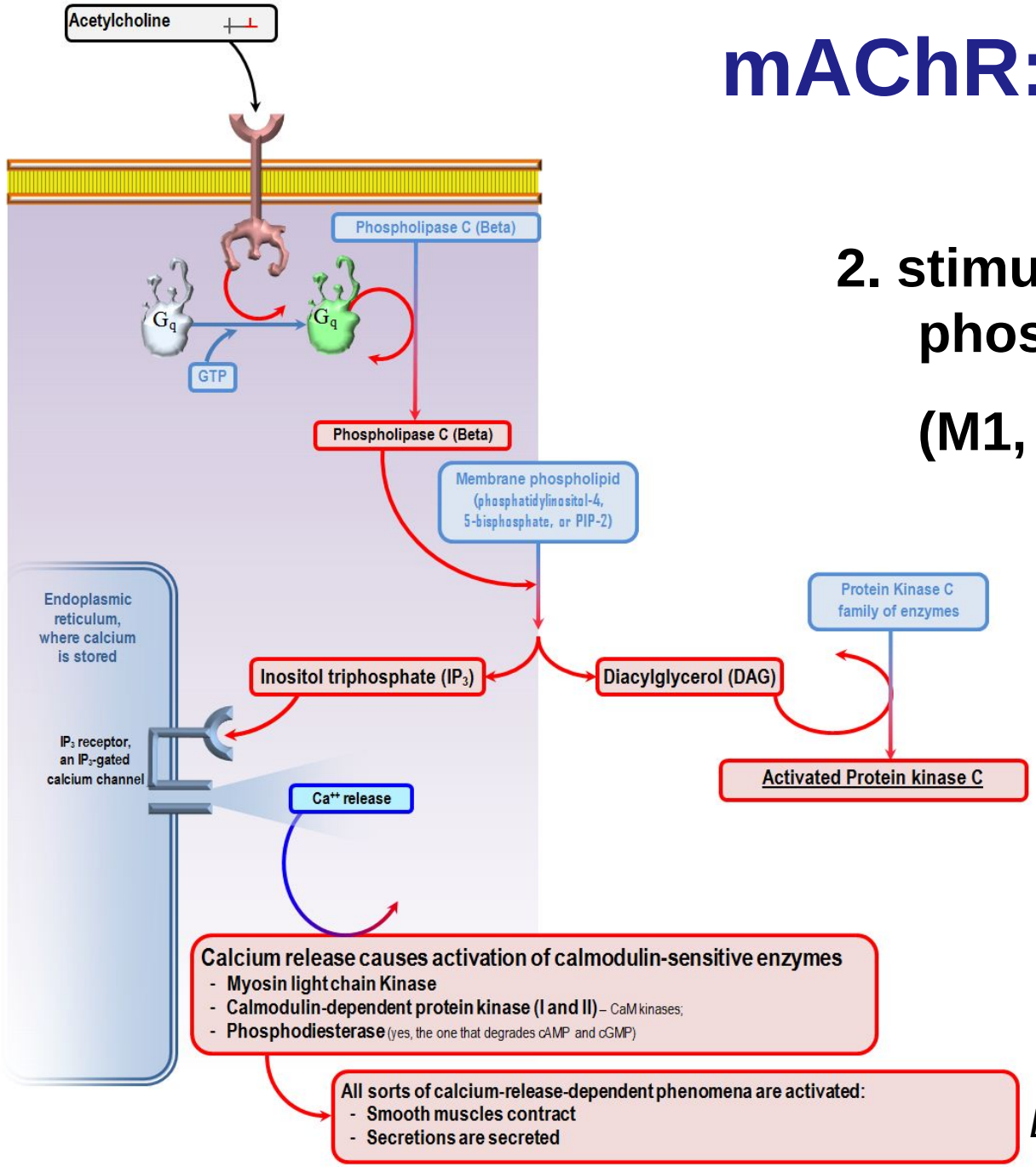
   (βγ: M2, M4)



M2 or M4 activation = A decrease in cAMP
Thus, no Protein Kinase A
Thus, no phosphorylated Phosphorylase Kinase.

Deranged Physiology 2015

mAChR:

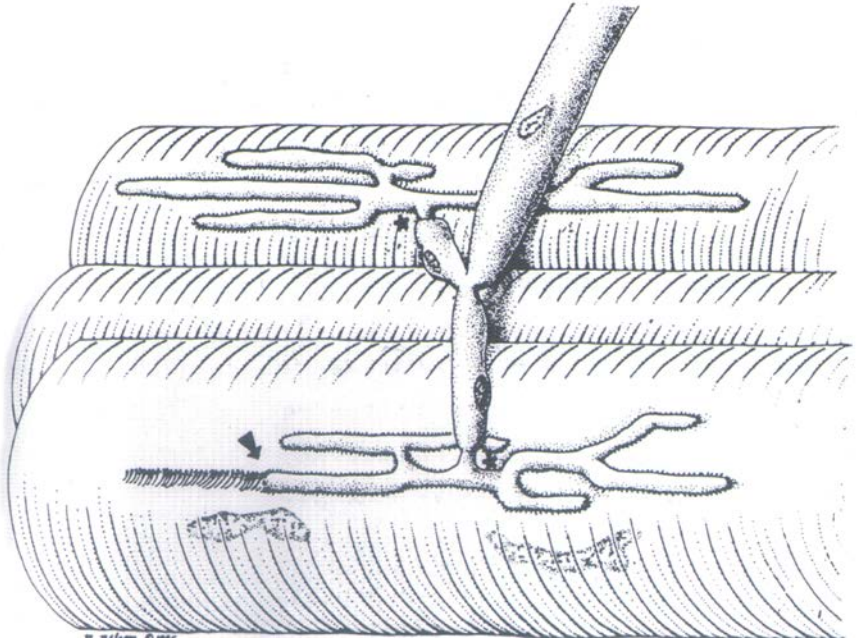


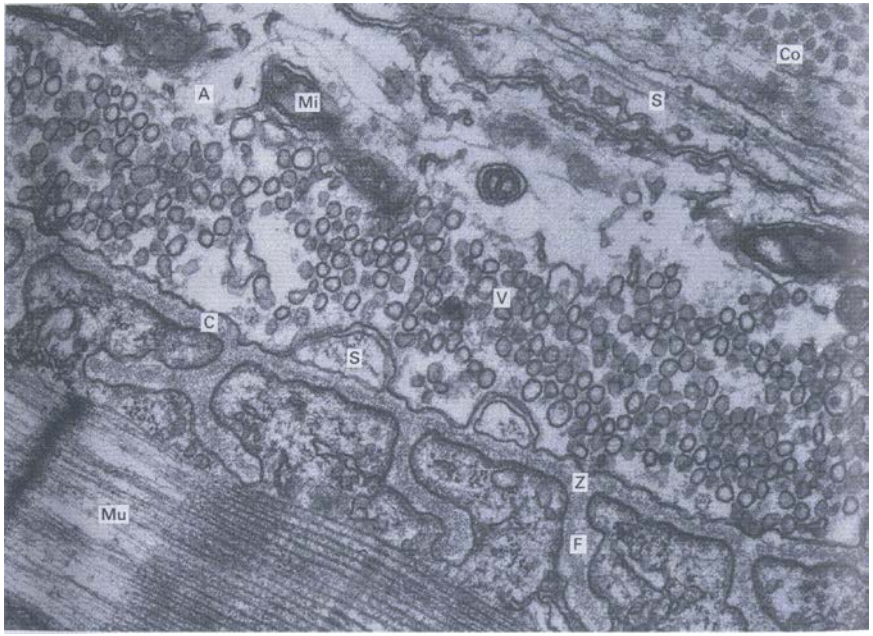
2. stimulate phospholipase C

(M1, M3, M5)

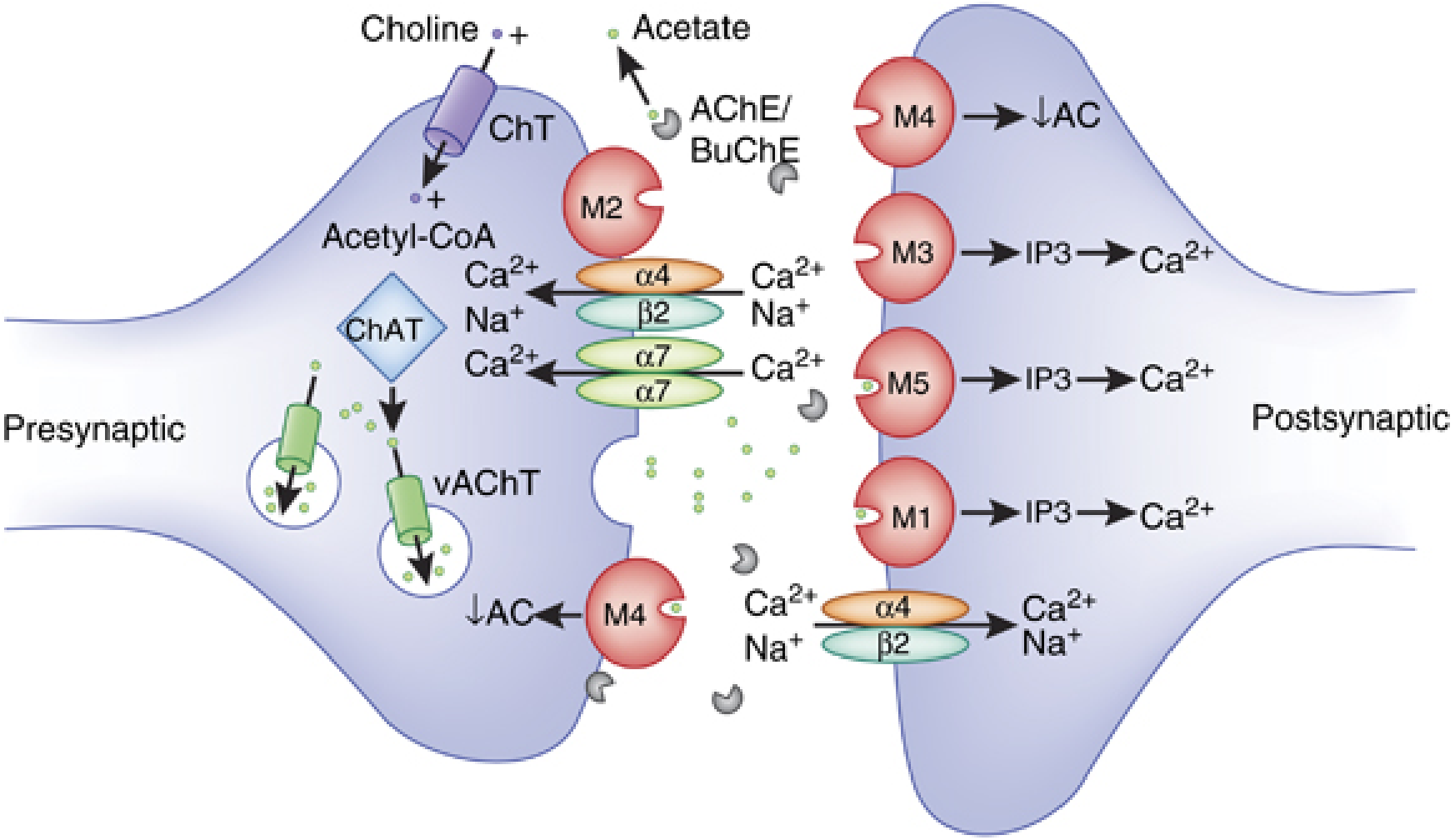
Deranged Physiology 2015

Neuromuscular junction





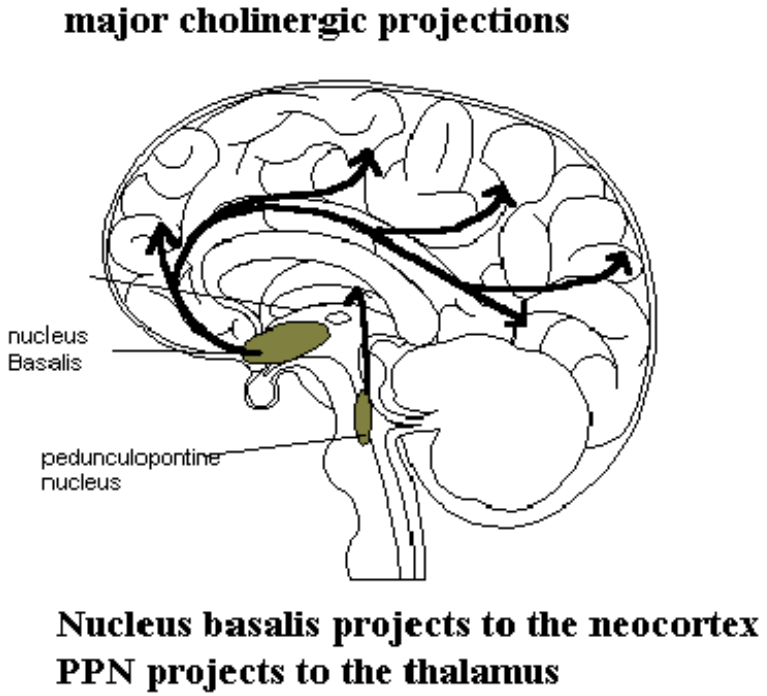
Cholinergic synapse



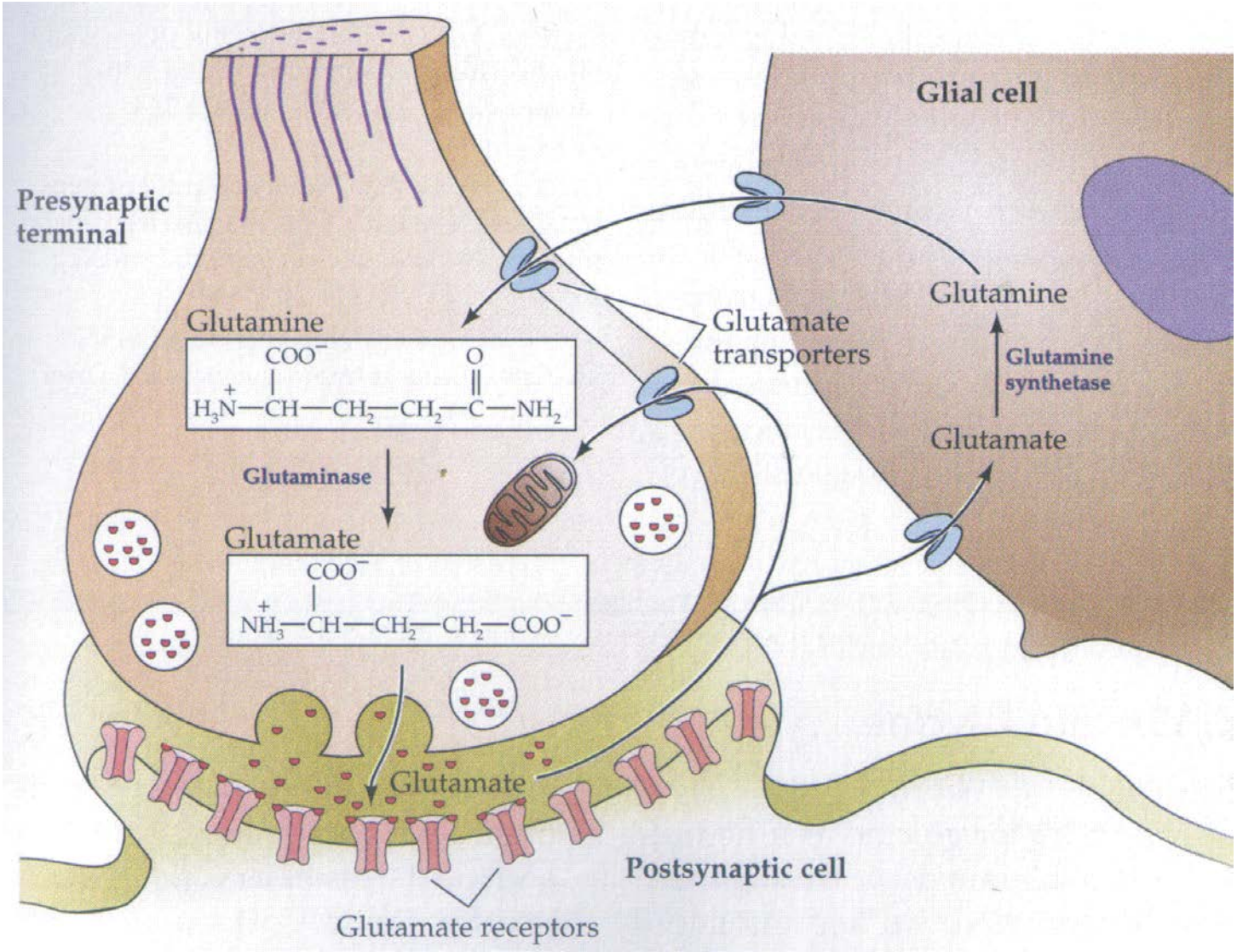
Jones et al., Neuropsychopharmacology 2012

Importance of cholinergic systems

- Motor activity
- Learning and memory: nucleus basalis
- Alzheimer disease



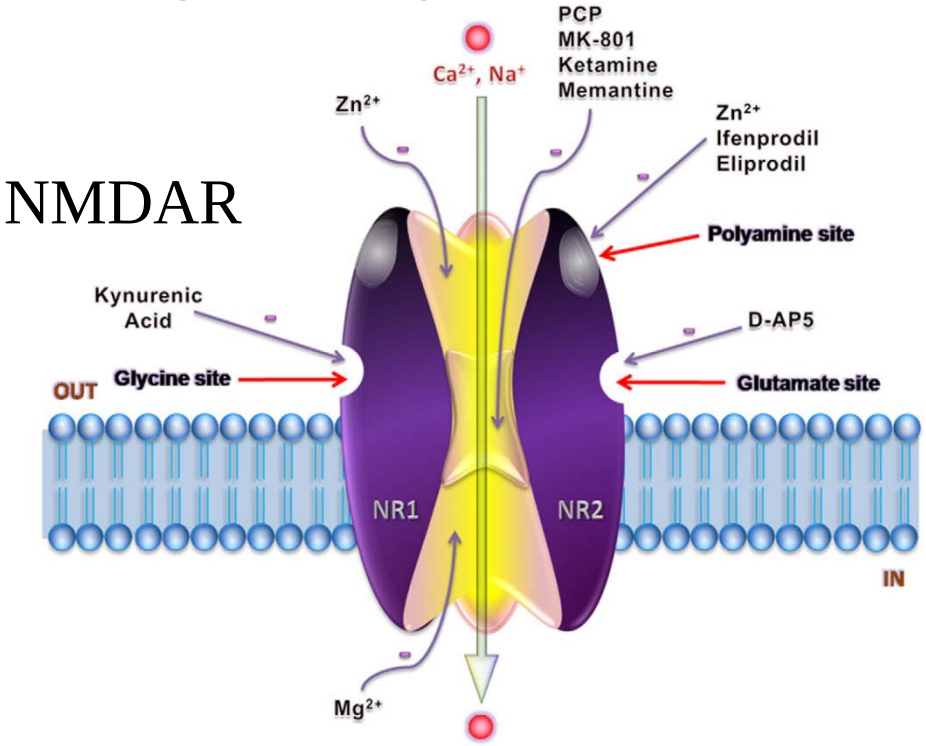
Glutamate – the main excitatory NT in CNS



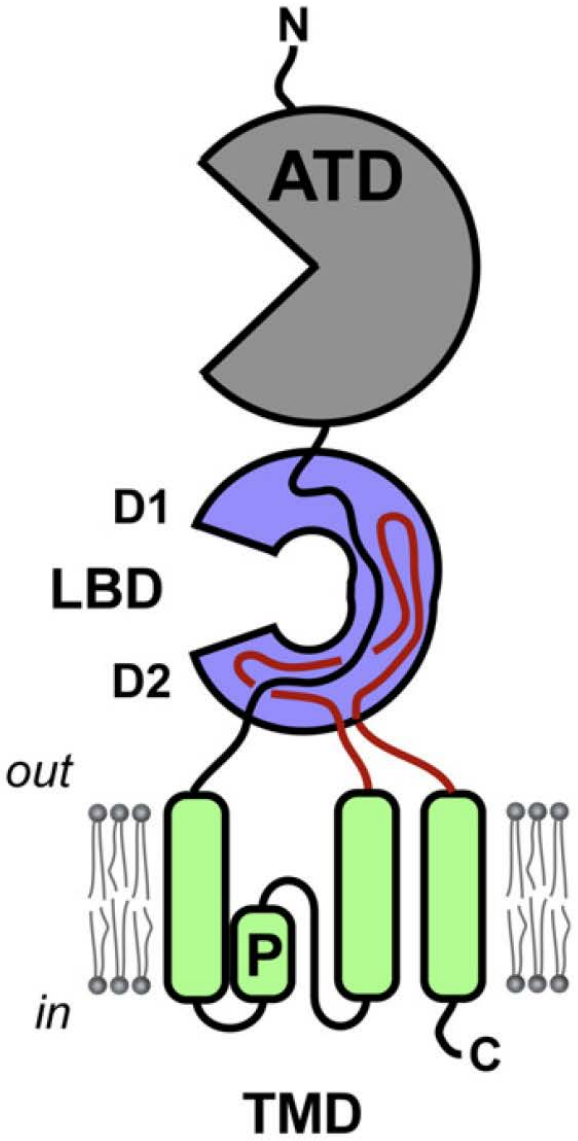
Purves et al., Neuroscience, 4th ed., 2008

Ionotropic GluRs: cation permeable channels

Subtypes:
-NMDA (GluN1, GluN2A-D, GluN3A,B)
-AMPA (GluA1-4)
-Kainate (GluK1-5)

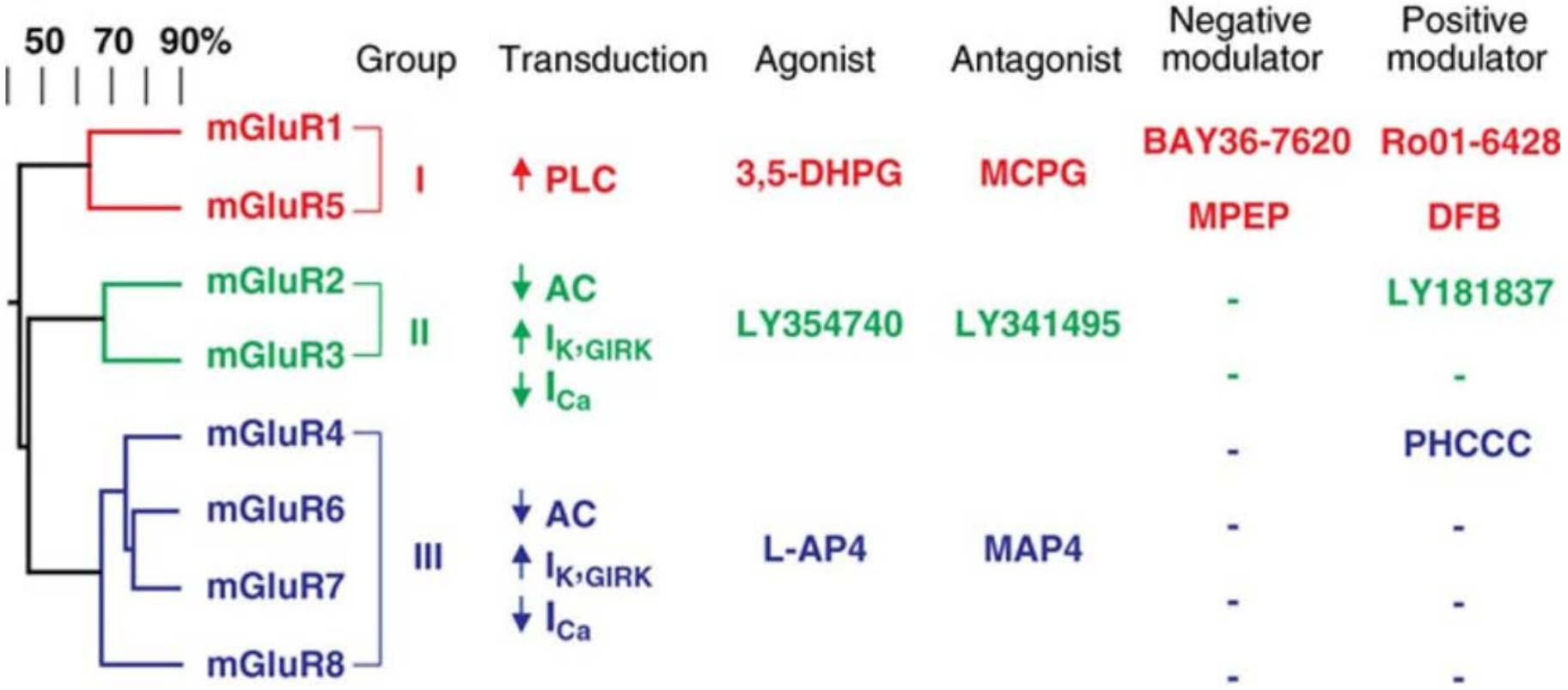


Ghasemi and Schachter, Epilepsy&Behavior, 2008

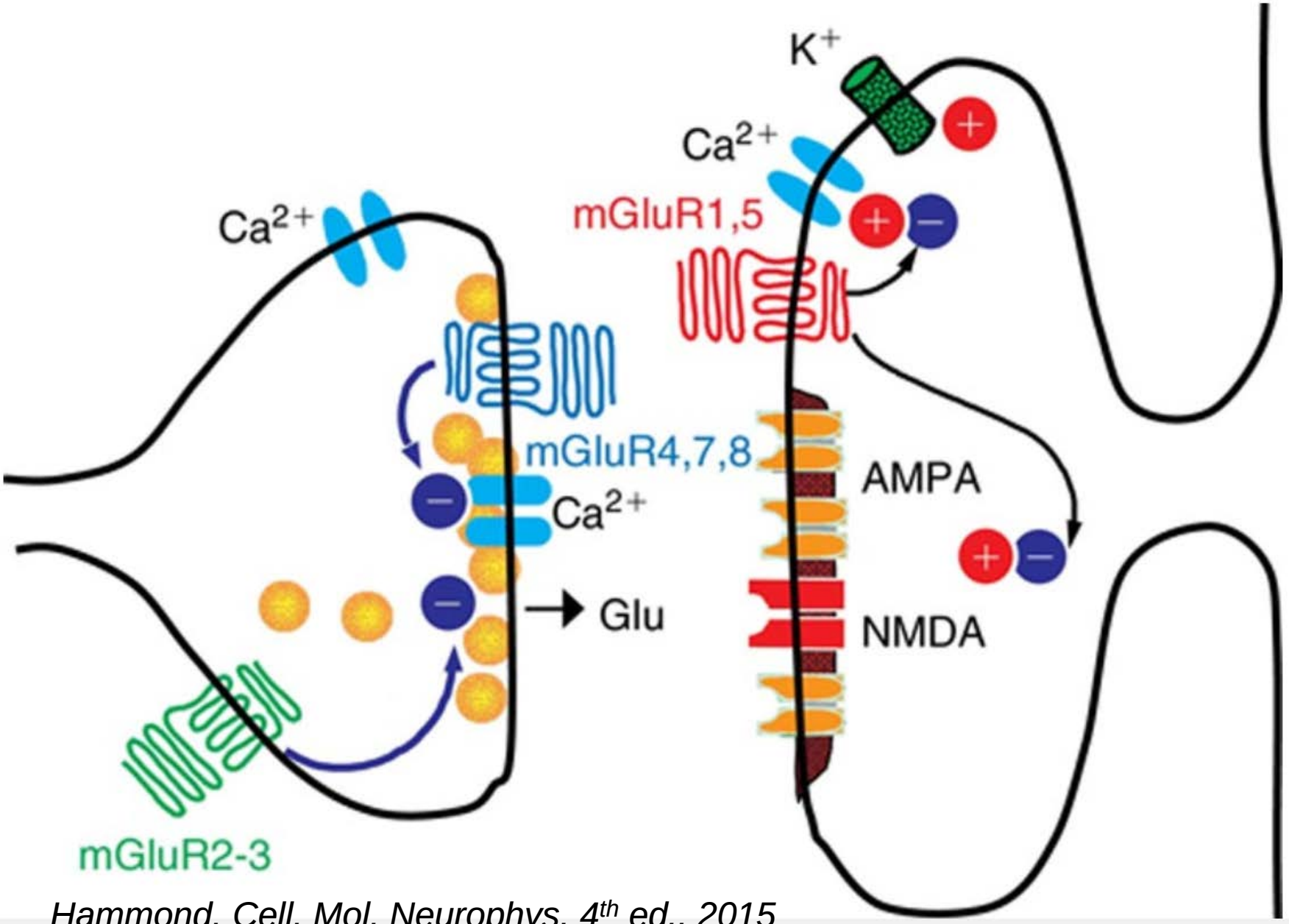


Stawski et al., Bioorg. Med. Chem., 2010

Metabotropic GluRs: GPCRs



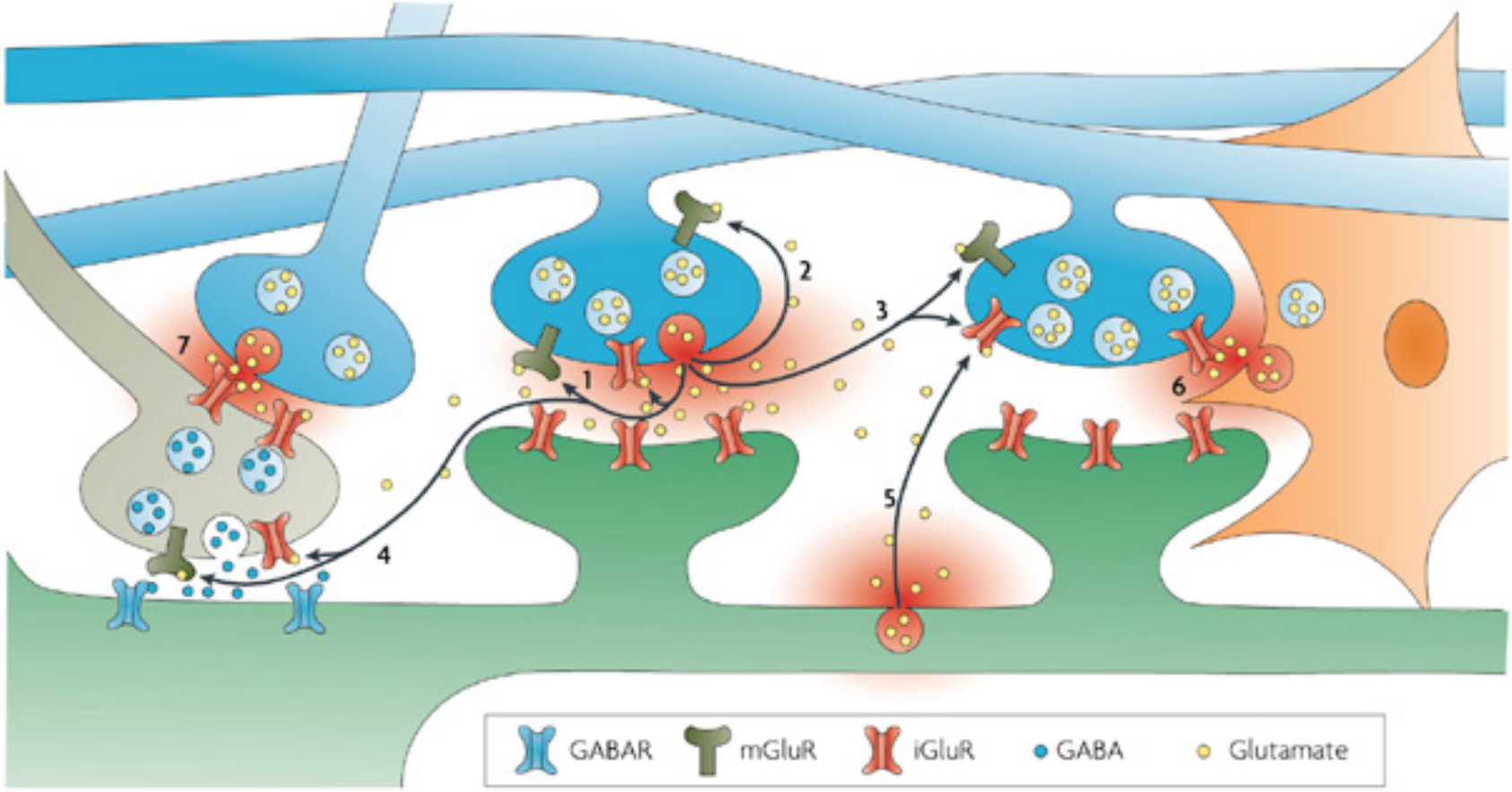
Glutamatergic synapse



Hammond, Cell. Mol. Neurophys, 4th ed., 2015

Presynaptic receptors

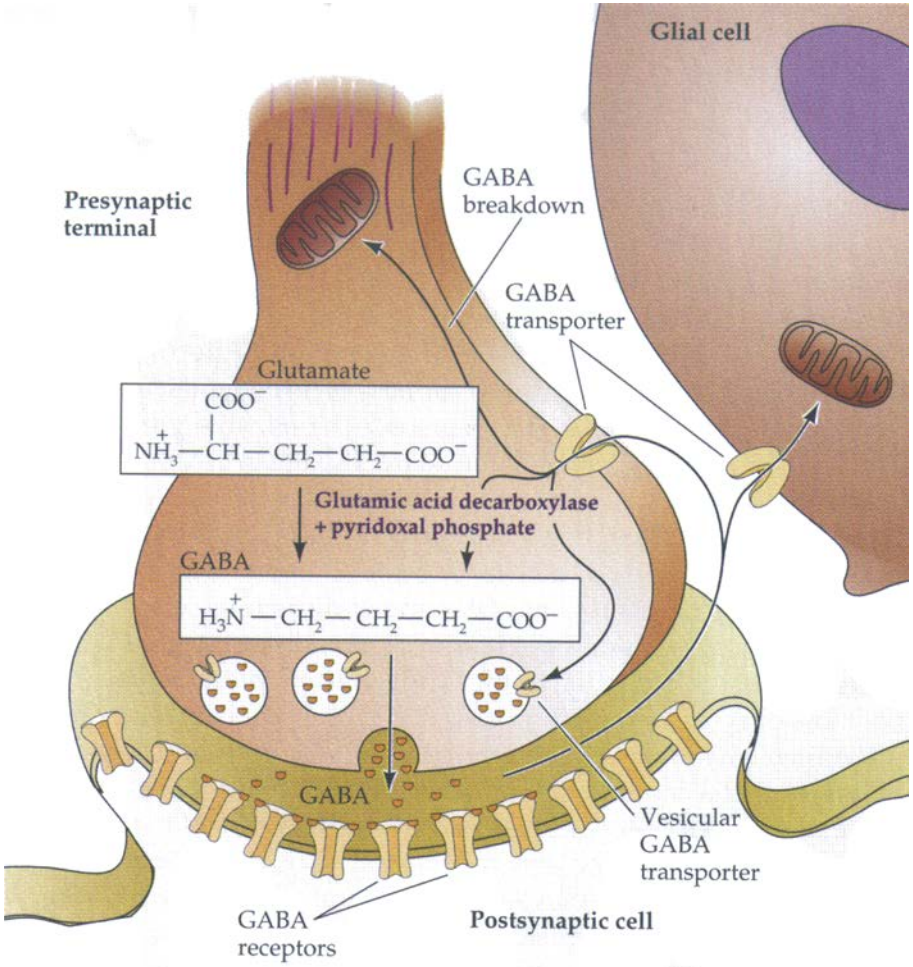
Presynaptic glutamate receptors – spillover, autoreceptors vs. heteroreceptors, homosynaptic vs. heterosynaptic modulation

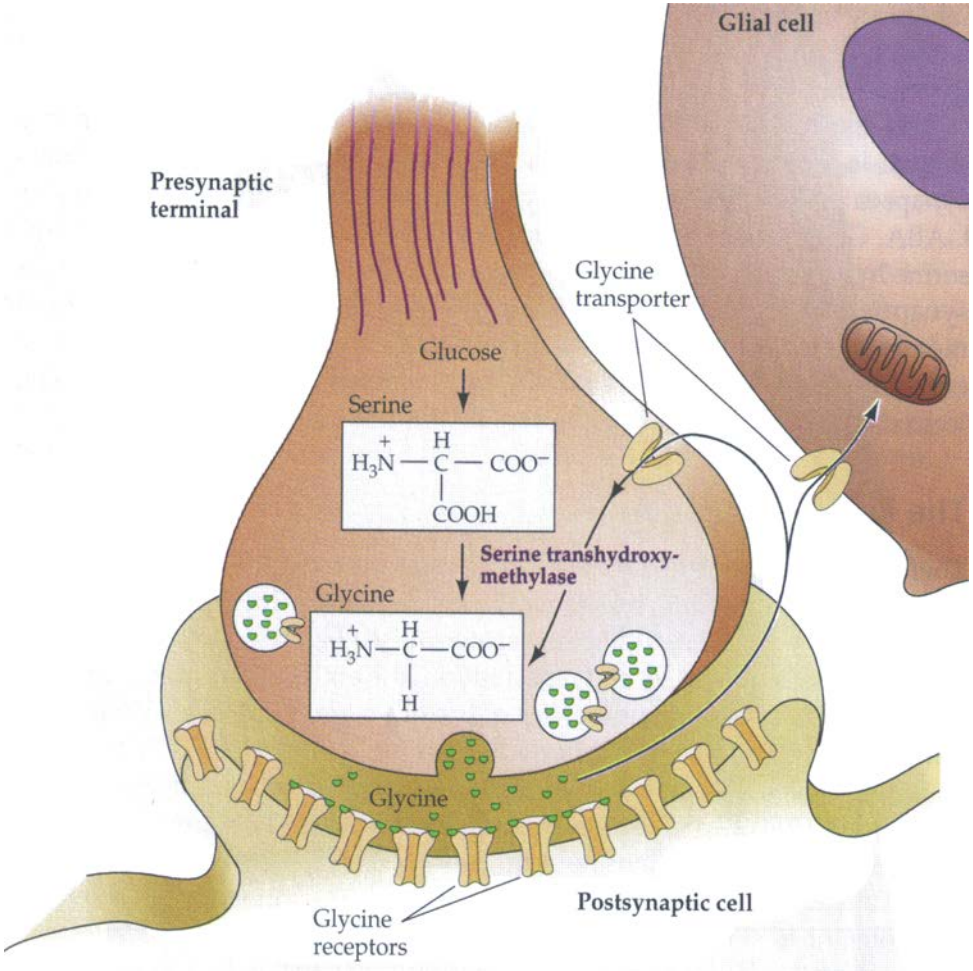


Nature Reviews | Neuroscience

GABA and glycine

the main inhibitory NTs in the CNS





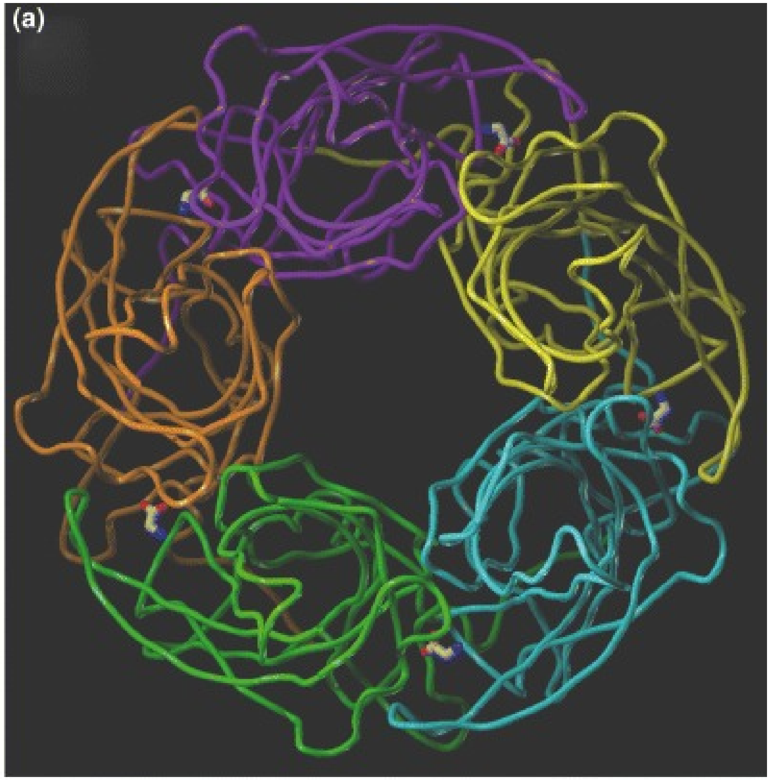
Purves et al., Neuroscience, 4th ed., 2008

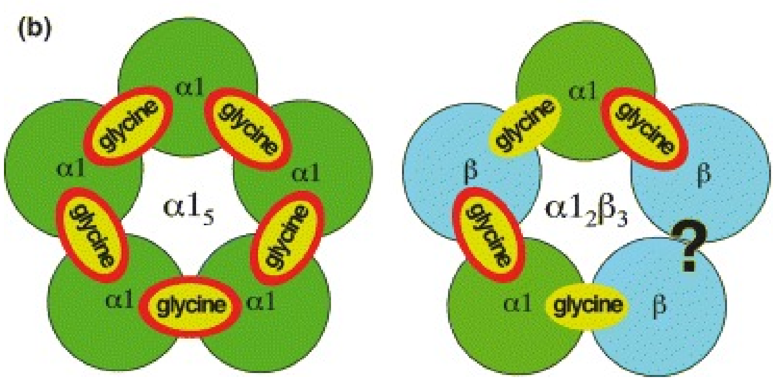
Receptors for inhibitory aminoacids

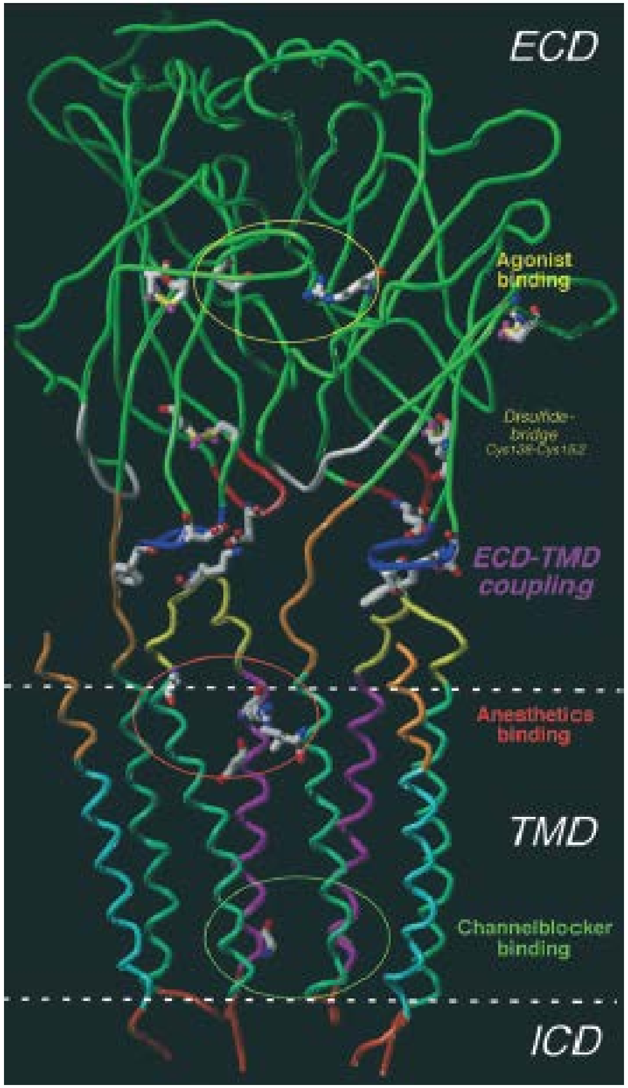
1. Ionotropic ($GABA_{A,C}$, glycine)
- anionic channels (Cl^-, $HC0_3^-$)
- low $[Cl^-]i$
- hyperpolarization, shunting of postsynaptic excitability

2. Metabotropic ($GABA_B$)
- GPCR
- inhibition of AC or VDCC (presynaptic), activation of K+ channels (postsynaptic)

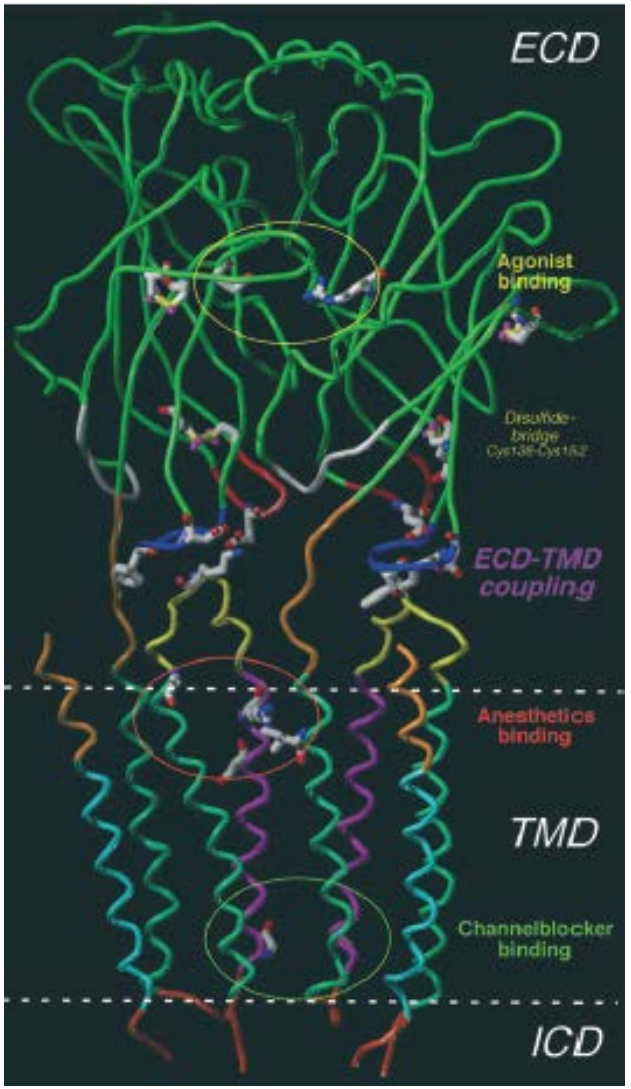
Glycine receptor





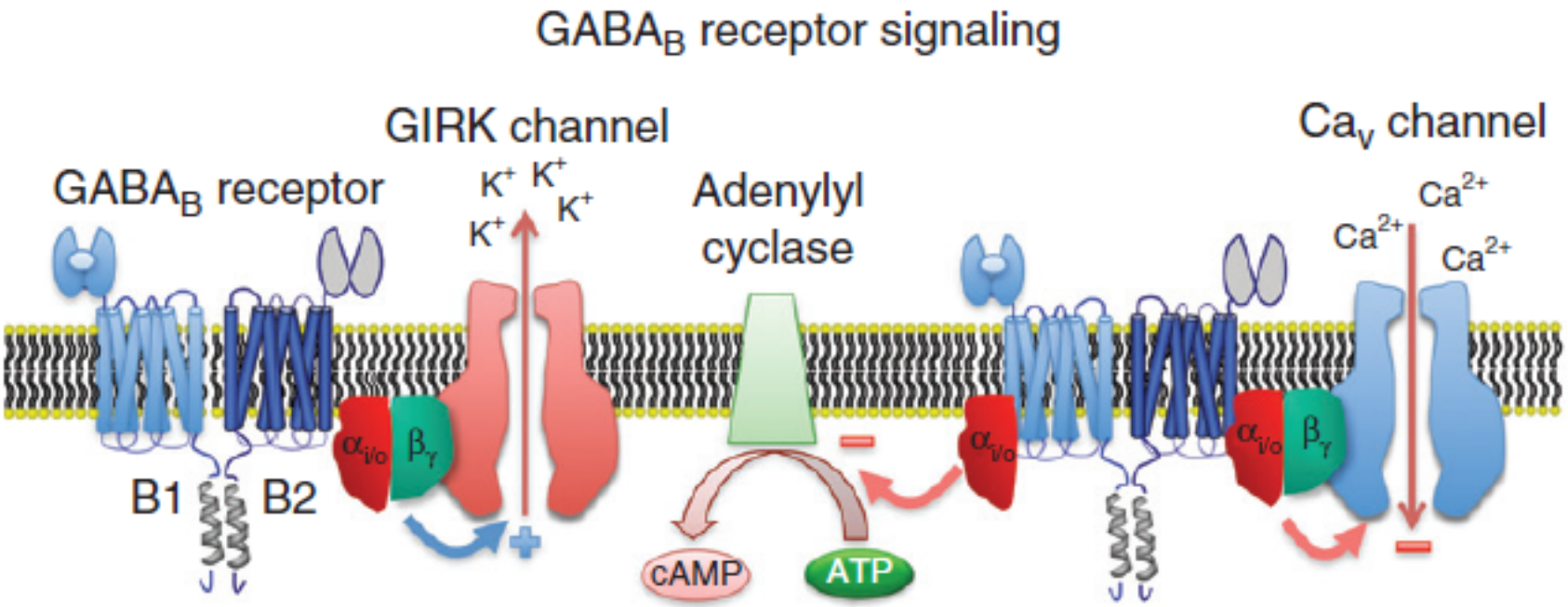


Glycine receptor



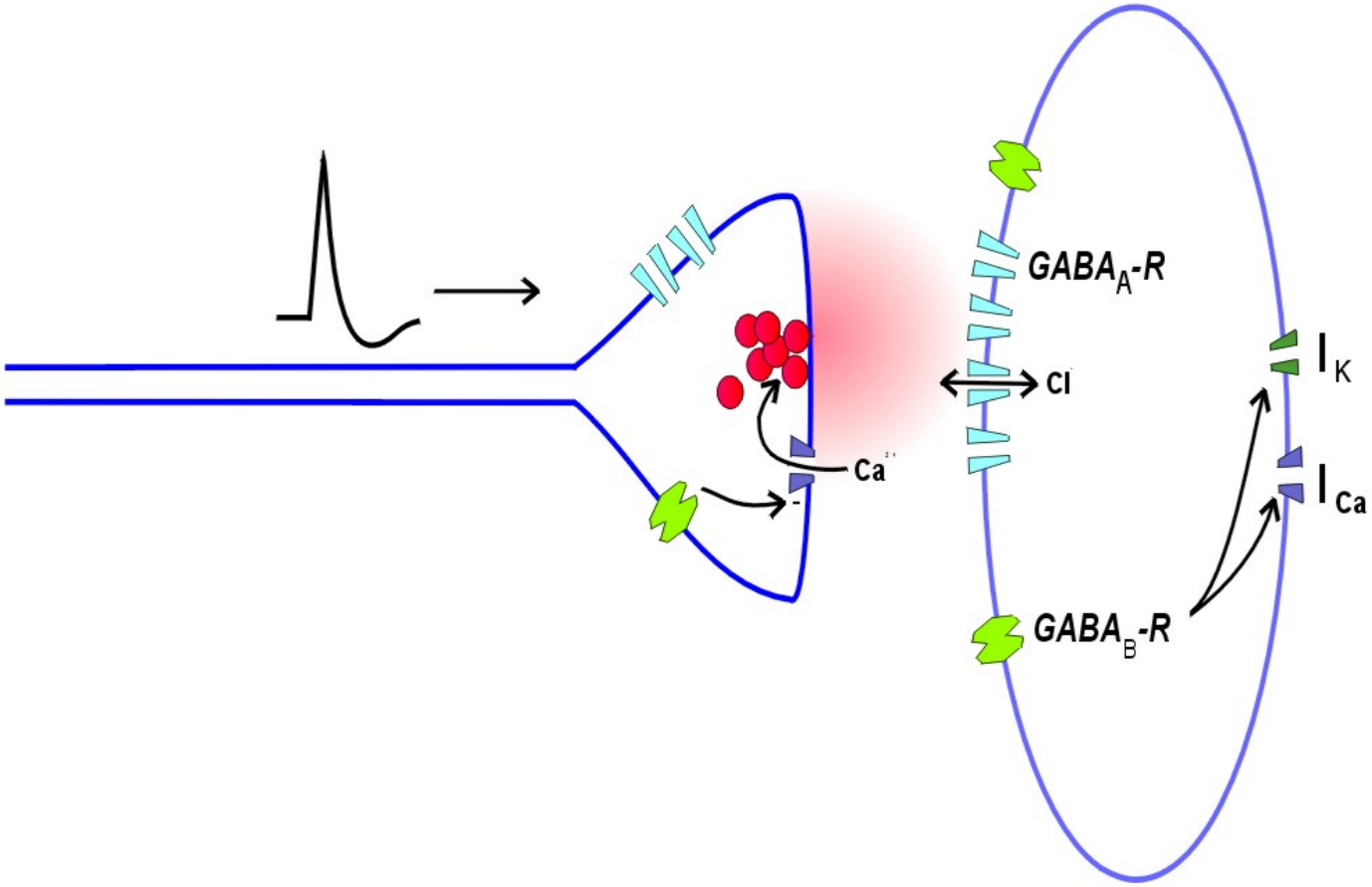
antagonist – strychnine (seeds of Strychnos nux-vomica)

GABA-B receptor - GPCR



Padgett and Slesinger, Adv. Pharmacol, 2010

GABAergic synapse

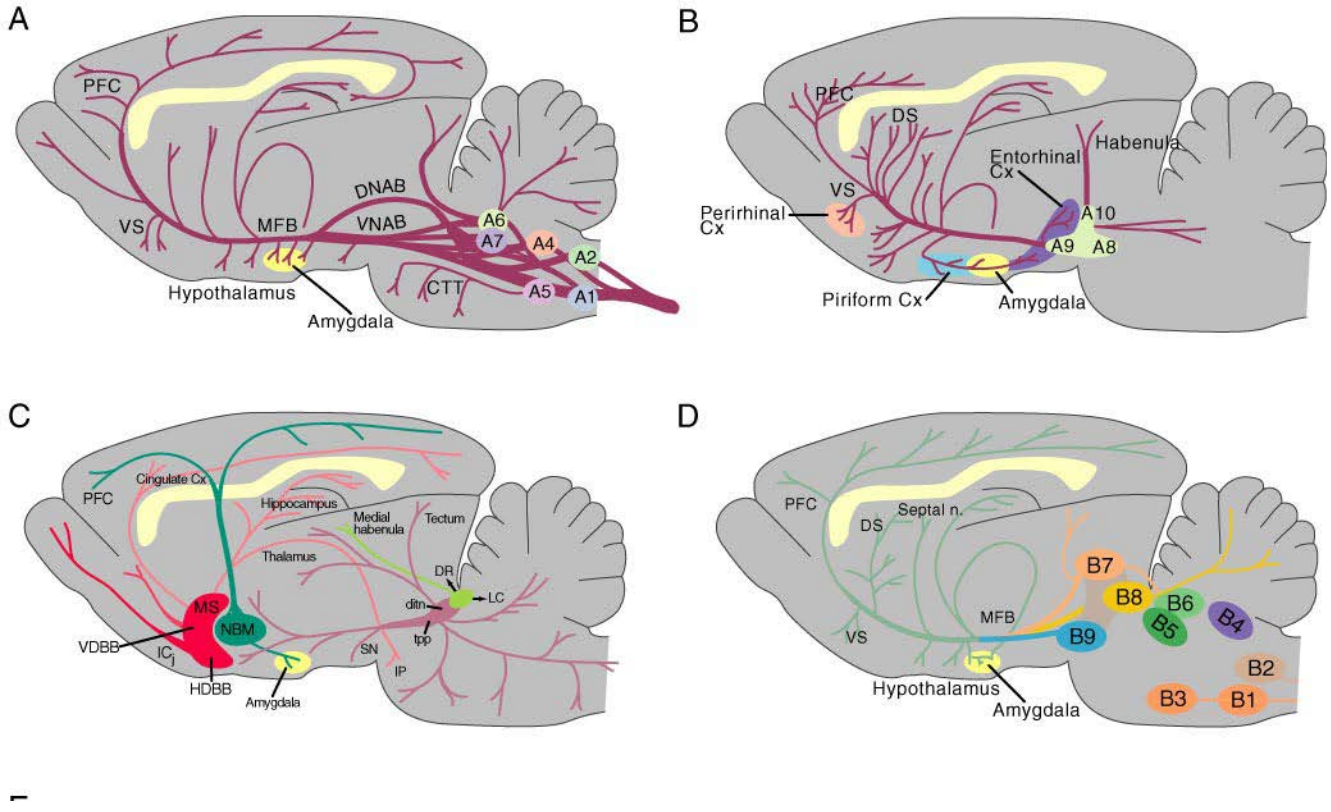


Biogennic amines

1. Catecholamines: dopamine, epinephrine (= adrenaline)
norepinefrine (= noradrenaline)
2. Histamine
3. Serotonin

Biogenic neurotransmitters

- Produced by nuclei concentrated in brainstem but terminals exhibit widespread distribution in the CNS
- Mediate slow neurotransmission
- Main function in CNS is neuromodulation



Ascending monoamine neurotransmitter systems. Figure shows schematic sagittal (A–D) sections through the lateral hypothalamus of a rat brain.

(A) Origin and distribution of central noradrenergic pathways. Note noradrenergic cell groups A1–A7, including the locus ceruleus (A6).

(B) Origin and distribution of central dopamine pathways. Note dopaminergic cell groups A8–A10. Substantia nigra, VTA

(C) Origin and distribution of central cholinergic pathways.

(D) Origin and distribution of central serotoninergic pathways. Note cell groups in the raphe nucleus, B4–B9. MFB, medial forebrain bundle; PFC, prefrontal cortex; VS, ventral striatum; DS, dorsal striatum.

Receptors for biogennic amines

Catecholamines:

- Dopamine receptors - GPCRs, $D1_{A,B}$, D2, D3, D4, activation or inhibition of AC
- NE and epinephrine: α and β -adrenergic receptors, α_1 - inhibition of K^+ channels, α_2 – activation of K^+ channels or inhibition of AC, $\beta_{1\text{-}3}$ – stimulation of AC

Histamine:

H_1 - H_3, GPCRs, stimulation of PLC or AC

Serotonin:

$5HT_{1\text{-}7}$, GPCRs except $5HT_3$ – ion channel permeable for cations, GPCRs – modulation of K^+ channels, AC or PLC