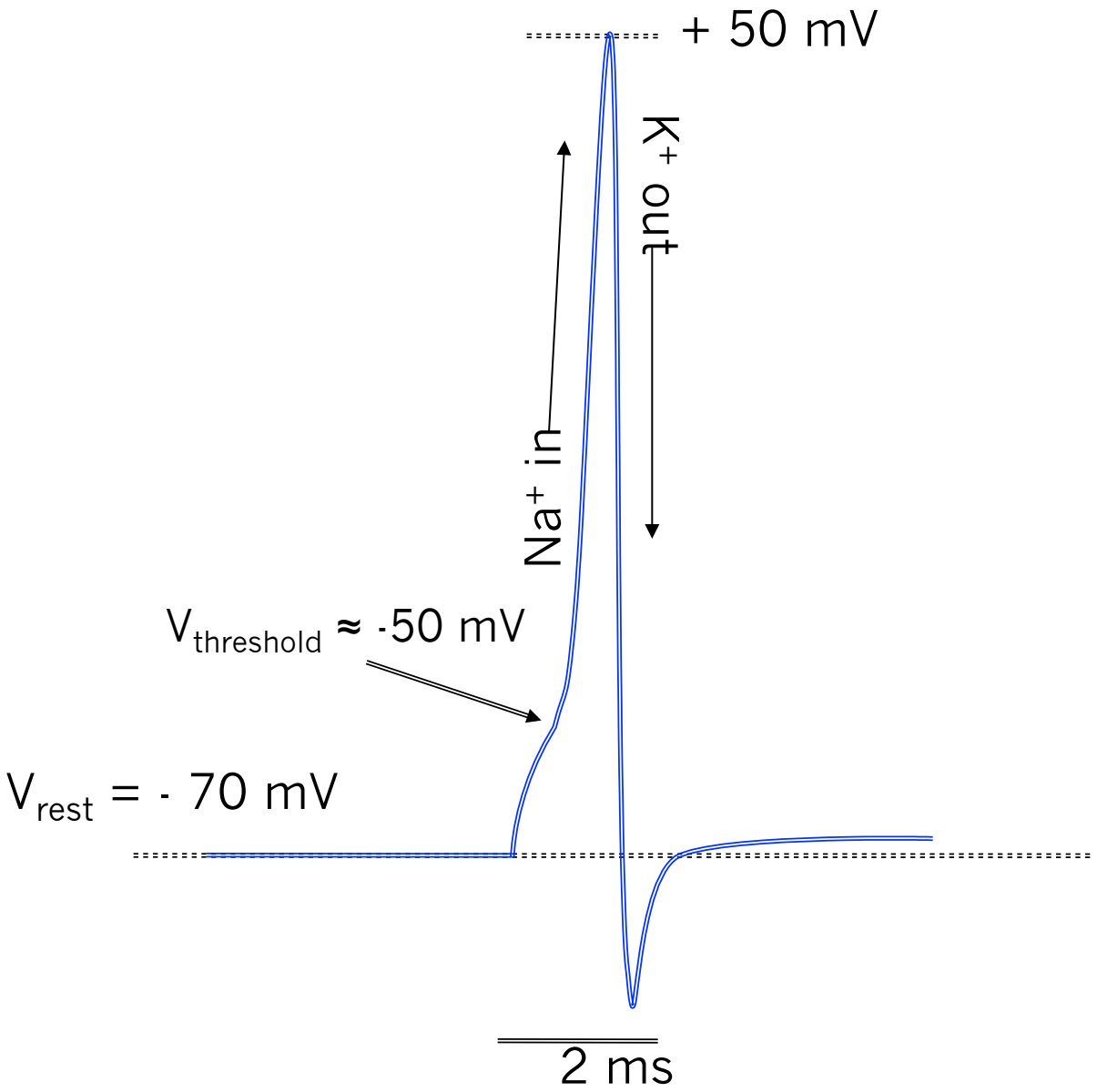
Class 3: Chemical transmisstion 2/23/2013

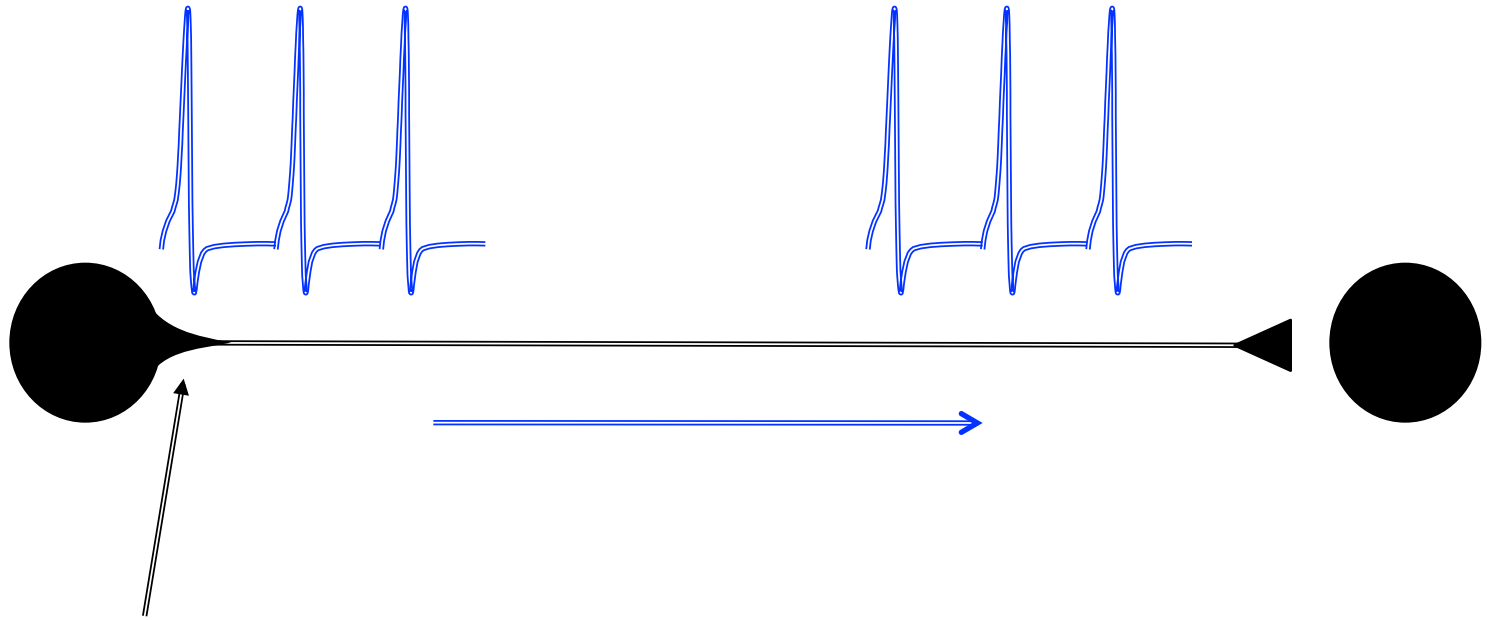
- Review of action potentials
- The Synapse: molecules and mechanisms
- Post-synaptic potentials
- Neurotransmitters

The Action Potential



Action potentials:

- "all-or-none" (always the same size)
- rapid (spike and return to baseline in < 2ms)
- start in initial segment
- propagate to axon terminals
- myelin speeds propagation (reduces leak of ions)



initial segment
(axon hillock)

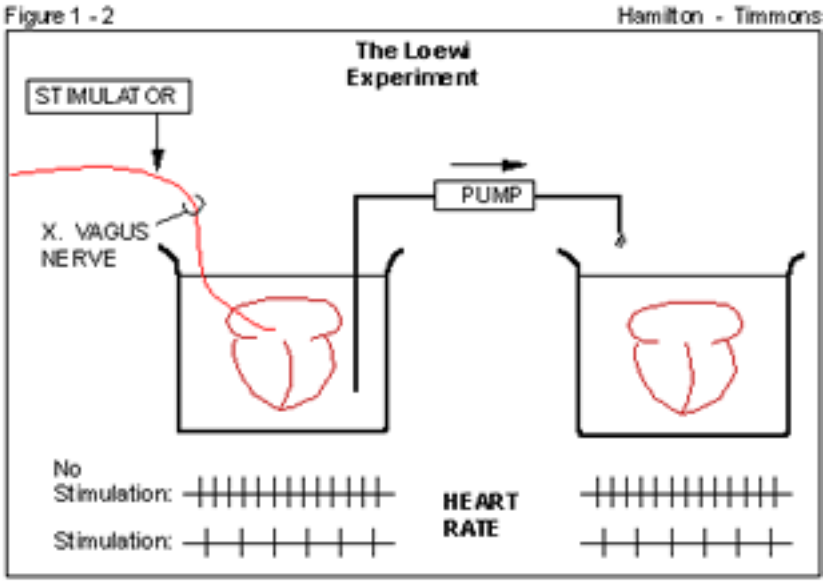
Chemical neurotransmission

Chemical or electrical?

Loewi's exp

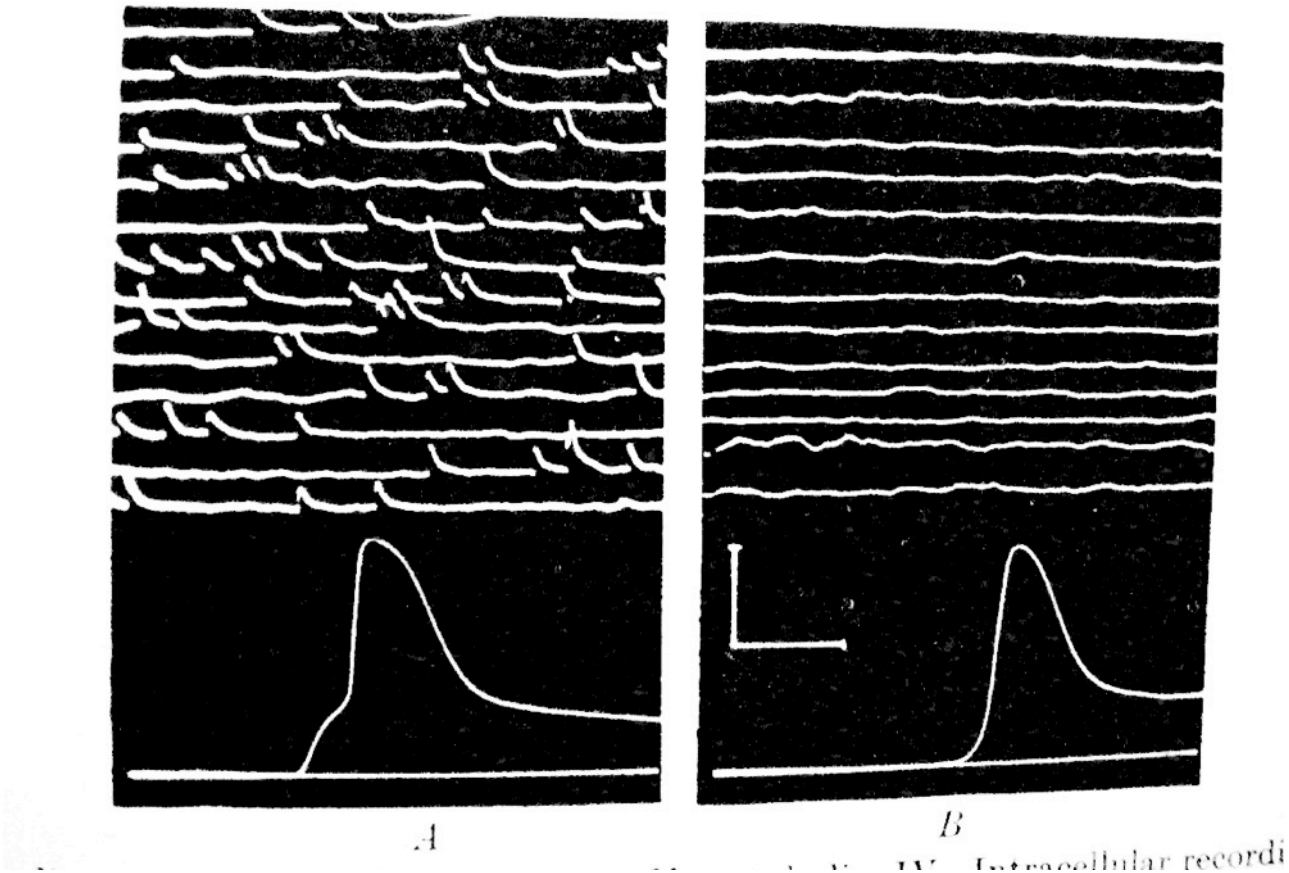
Loewi’s experiment 1921





Fatt and Katz 1952

A more decisive point was that the discharge could never



mini End Plate Potential (mEPP)

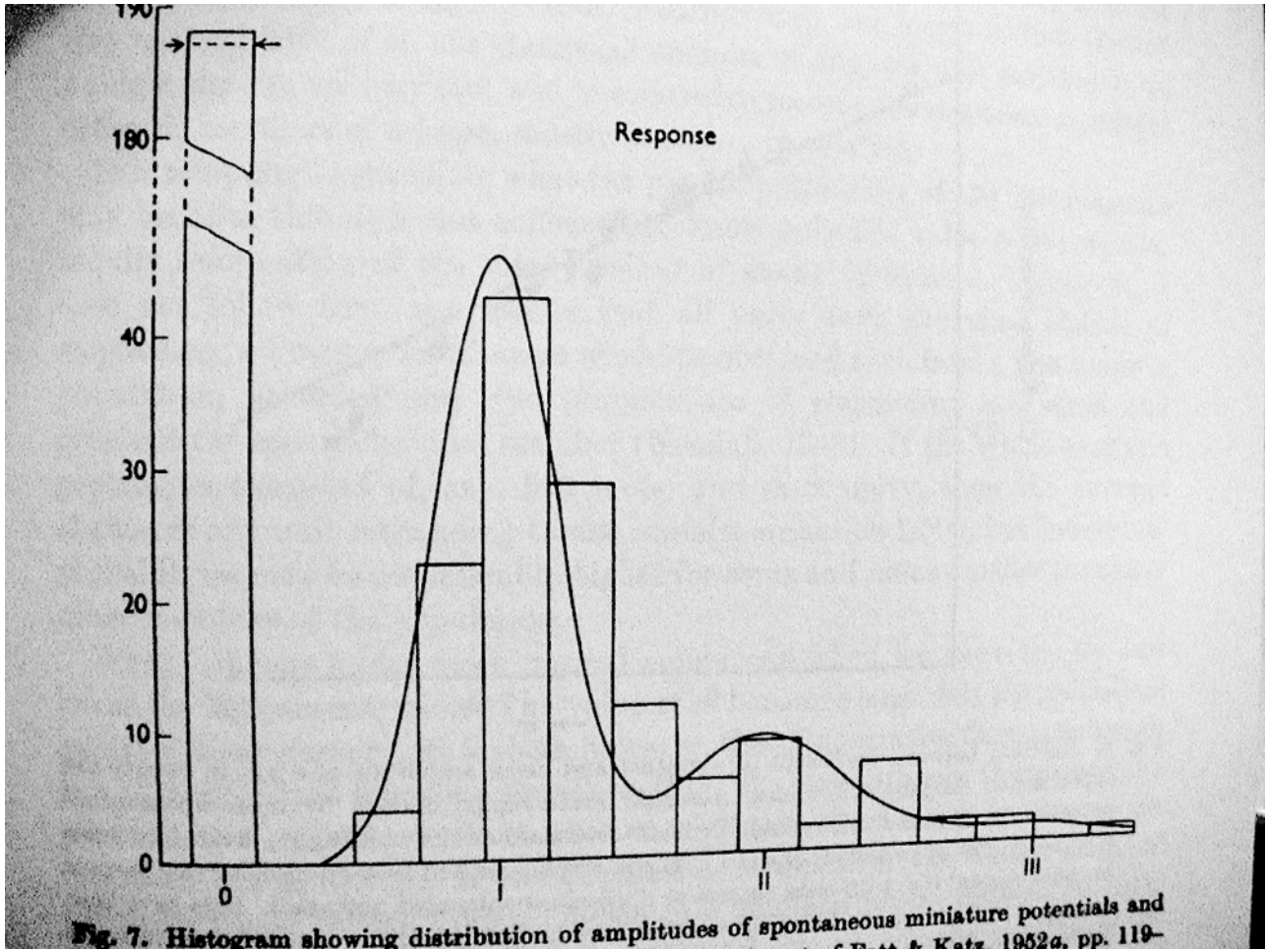
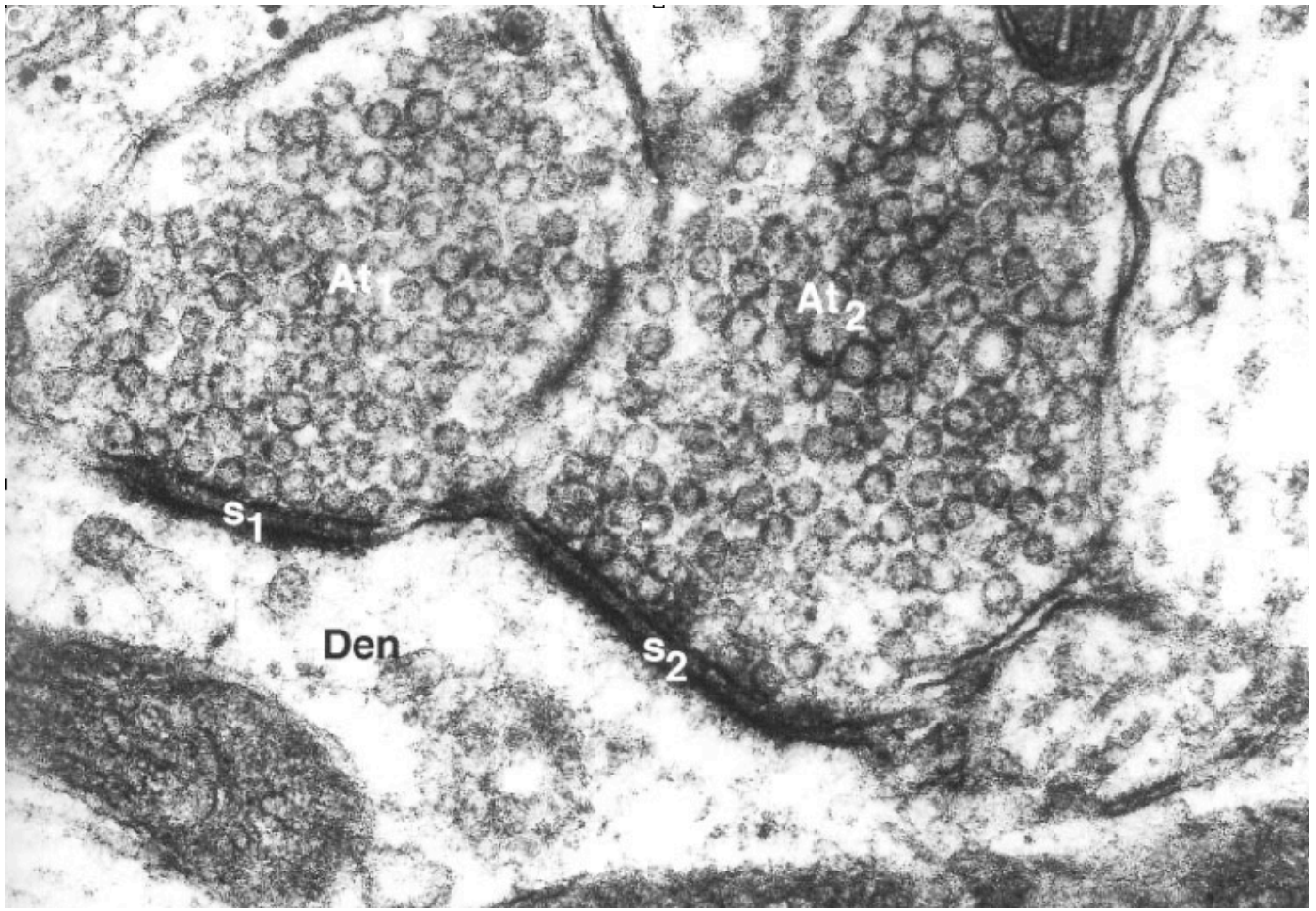


Fig. 7. Histogram showing distribution of amplitudes of spontaneous miniature potentials and

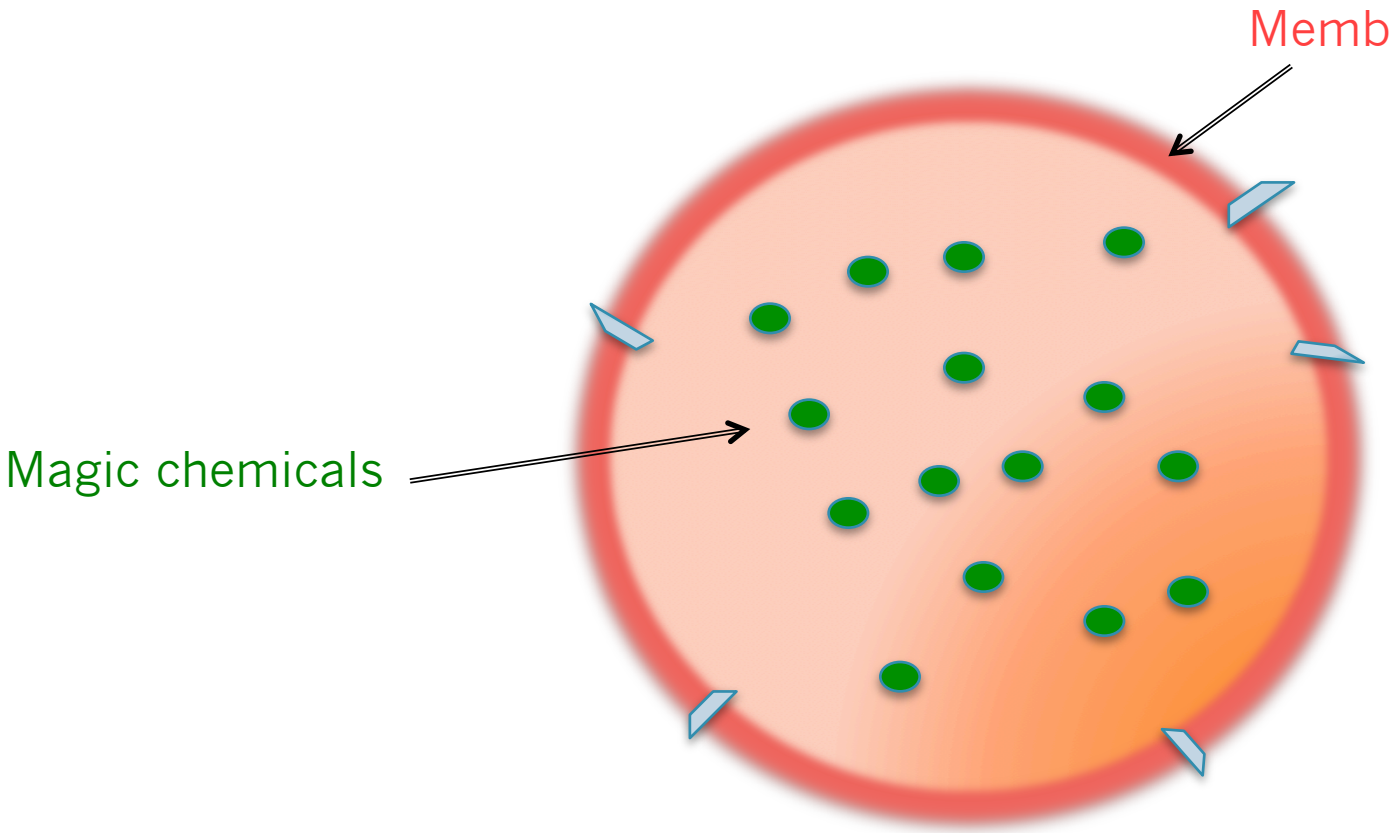
Quantal release!

1 mEPP = 1 vesicle filled with neurotransmitter

Synaptic vesicles



Synaptic vesicles



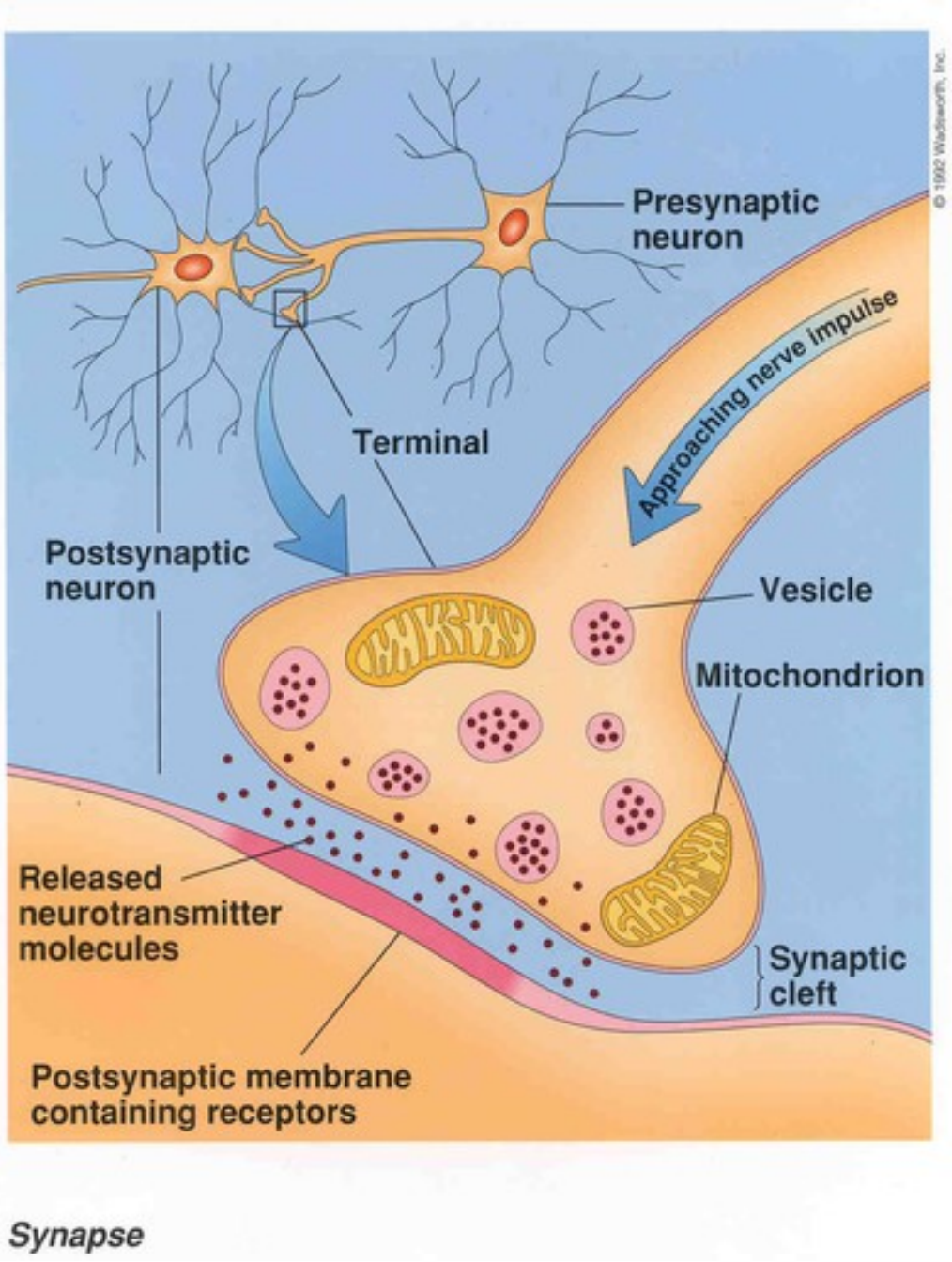
Synaptic vesicles are little packets of chemicals that communicate between neurons

Important point # 1

Synaptic vesicles contain the chemical signalling molecules of neurons

What are these chemicals called?

ne ease



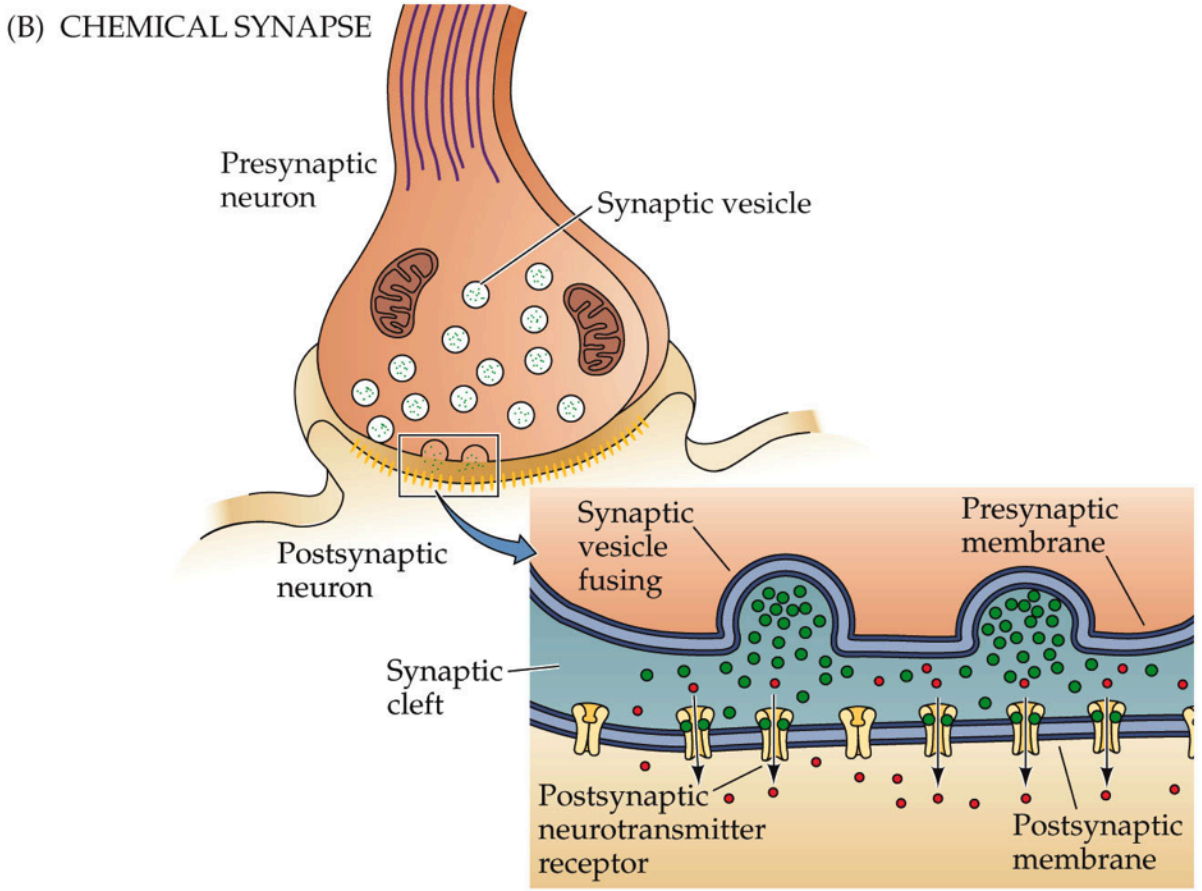
Synapse

Synapse video

Release of neurotransmitter is a highly regulated event

- Fusion of synaptic vesicles and release of neurotransmitter into the synaptic cleft is called **EXOCYTOSIS**
- Exocytosis is regulated by Ca^{++}
- Ca^{++} influx is triggered by the arrival of the action potential

The synapse

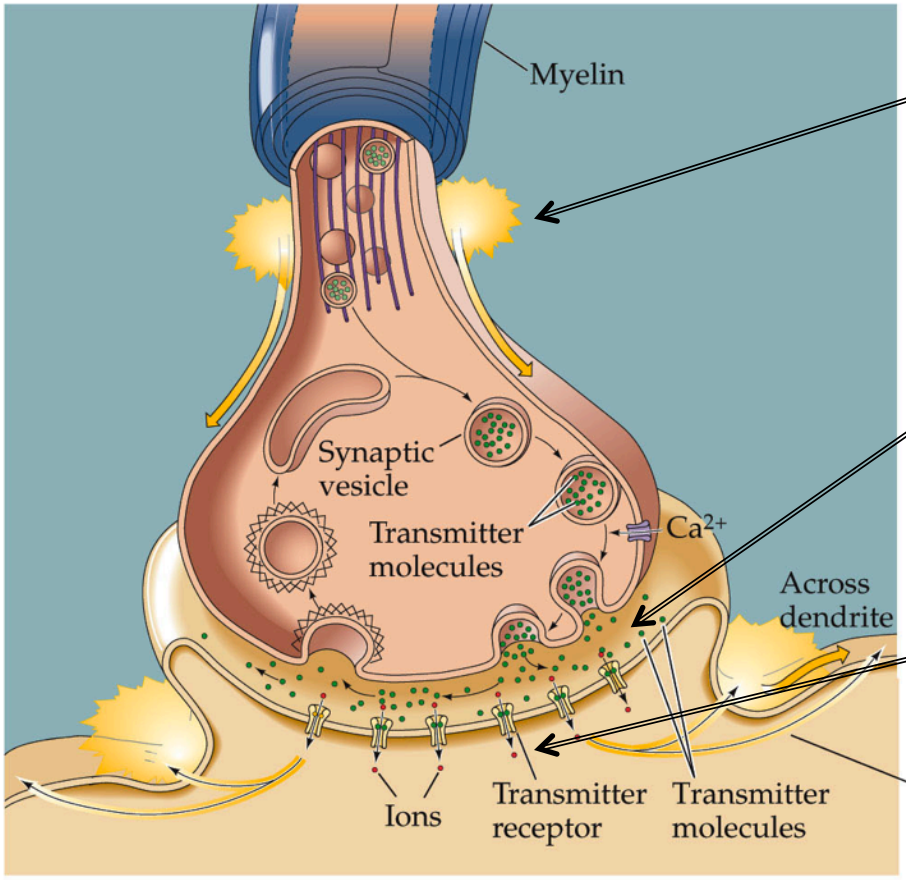


***NEUROSCIENCE, Fourth Edition,* Figure 5.1 (Part 2)**

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- **The synapse is the area of communication between two connected neurons**

Chemical neurotransmission



1) Electric signal: action potential

2) Chemical signal: neurotransmitter binds to receptor

3)Electrical signal: Excititory post-synaptic potential

***NEUROSCIENCE, Fourth Edition,* Figure 5.3**

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How to you make sure exocytosis only happens when an action potential arrives?

- A multi-step process involving dozens, maybe hundreds of proteins!

Steps to vesicle release

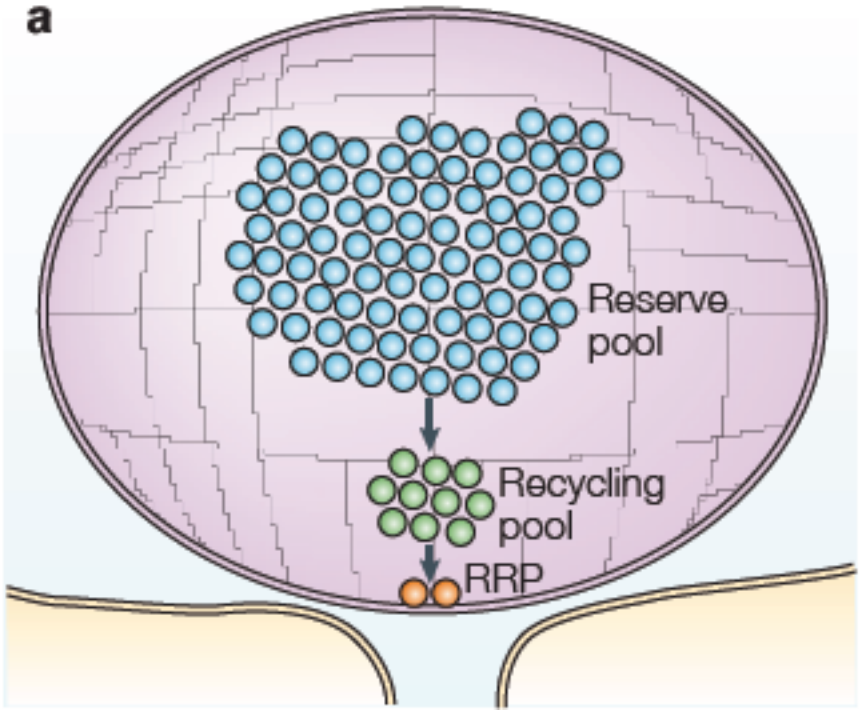
1)Targeting: Retain vesicles close to the plasma membrane – but watch out for spontaneous fusion

2)Docking and priming: Dock vesicles to active zone –

3) Fusion: Vesicle merges with the plasma membrane aka Exocytosis

4)Vesicle retrieval - endocytosis

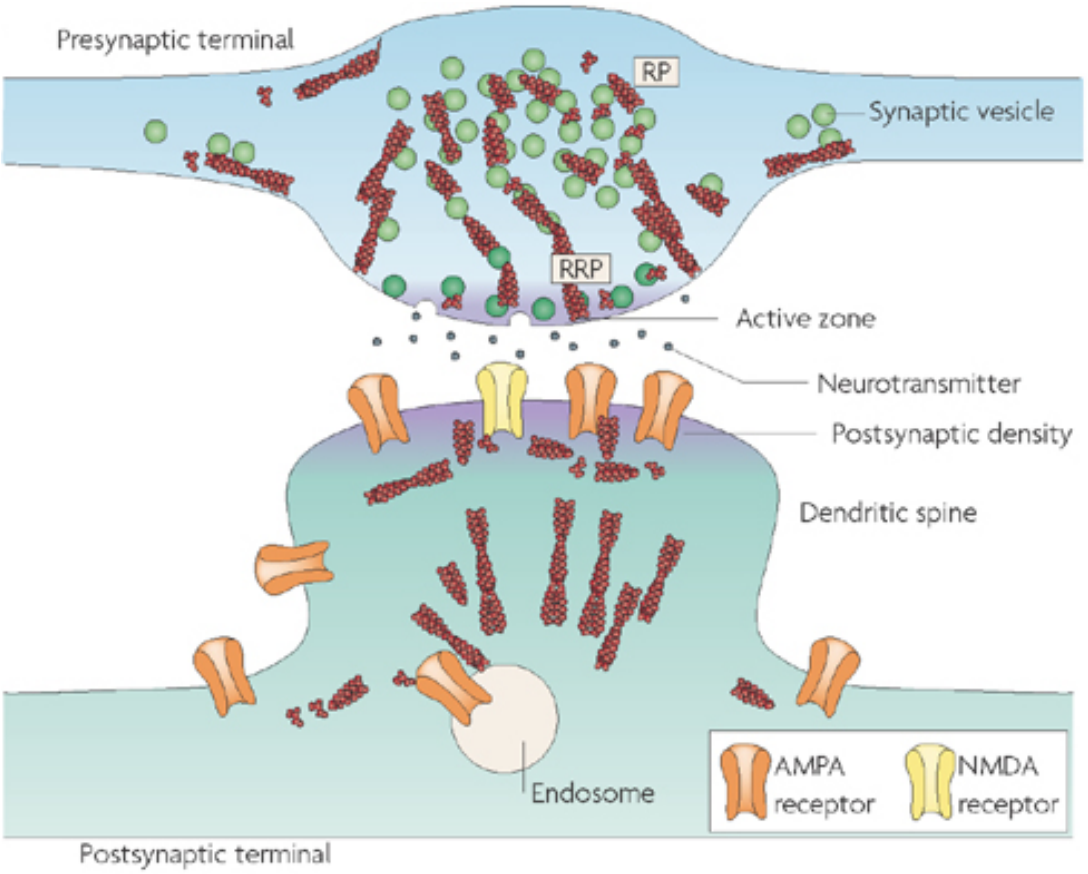
Step 1: The readily releasable pool and vesicle targeting

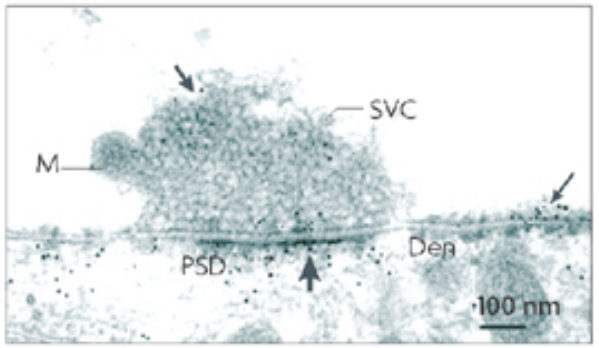


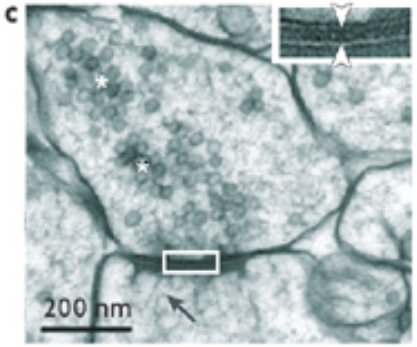
Only a small percentage of all synaptic vesicles are sitting, docked and primed, at the active zone ready for rapid fusion

The active zone:

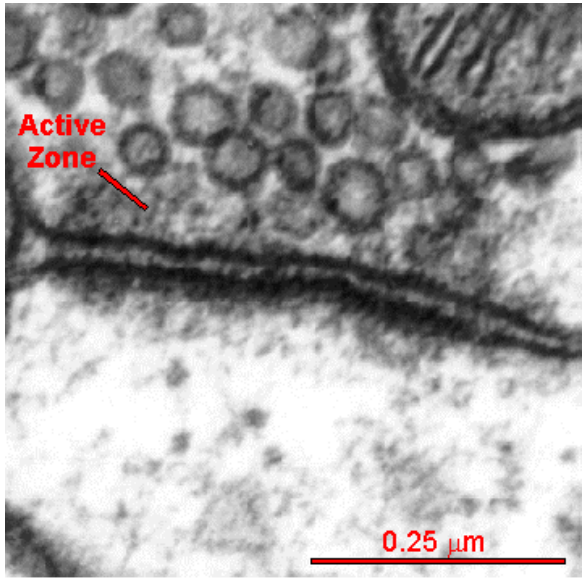
The part of the synapse where vesicles are ready to fuse upon arrival of Ca^{++}





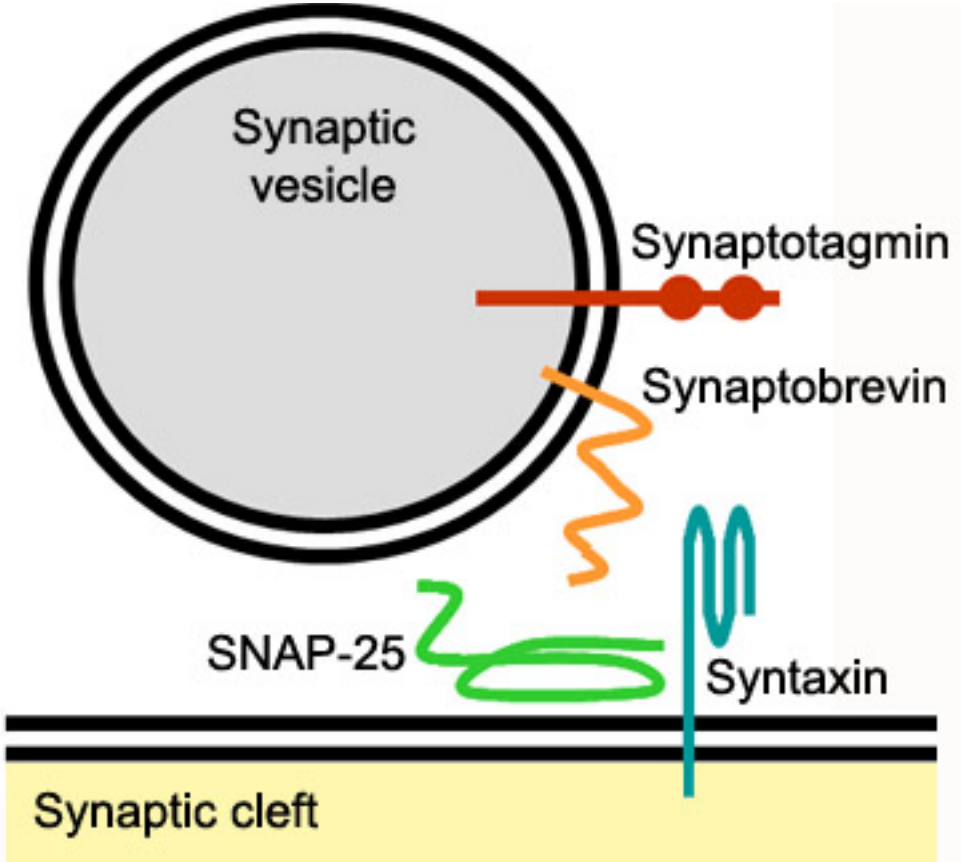


Nature Reviews | Neuroscience



Step 2: Docking and priming

The SNARE hypothesis

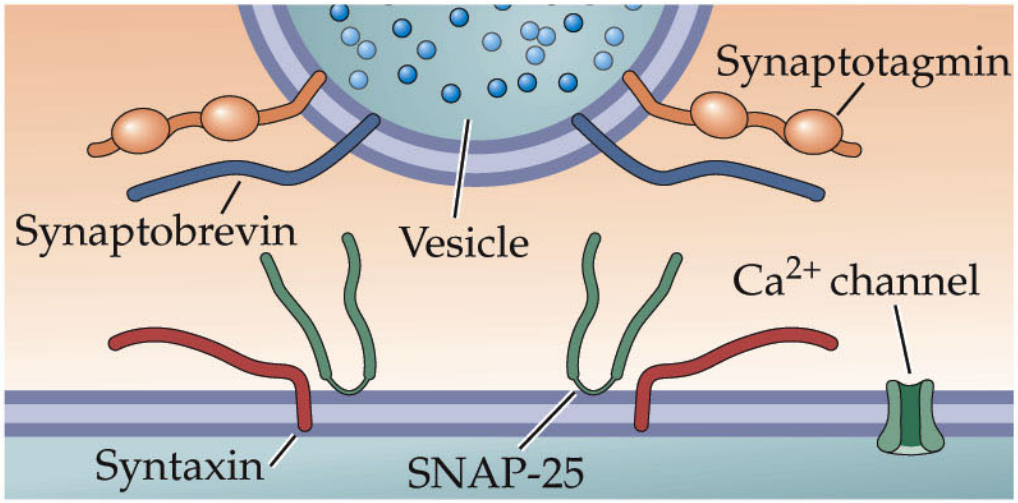


Three proteins come together:

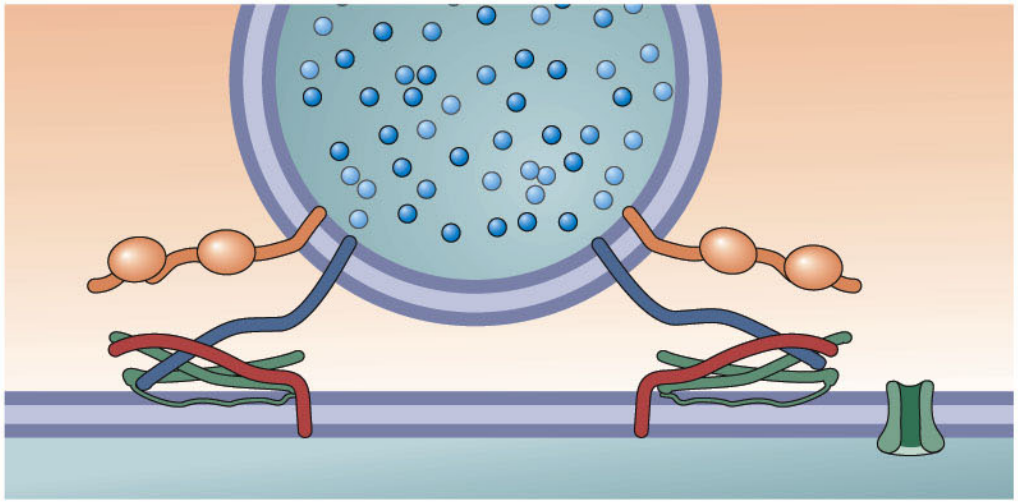
1 from the vesicle:
synaptobrevin (or VAMP)

2 from the plasma membrane:
SNAP-25 and syntaxin

(B) (1) Vesicle docks



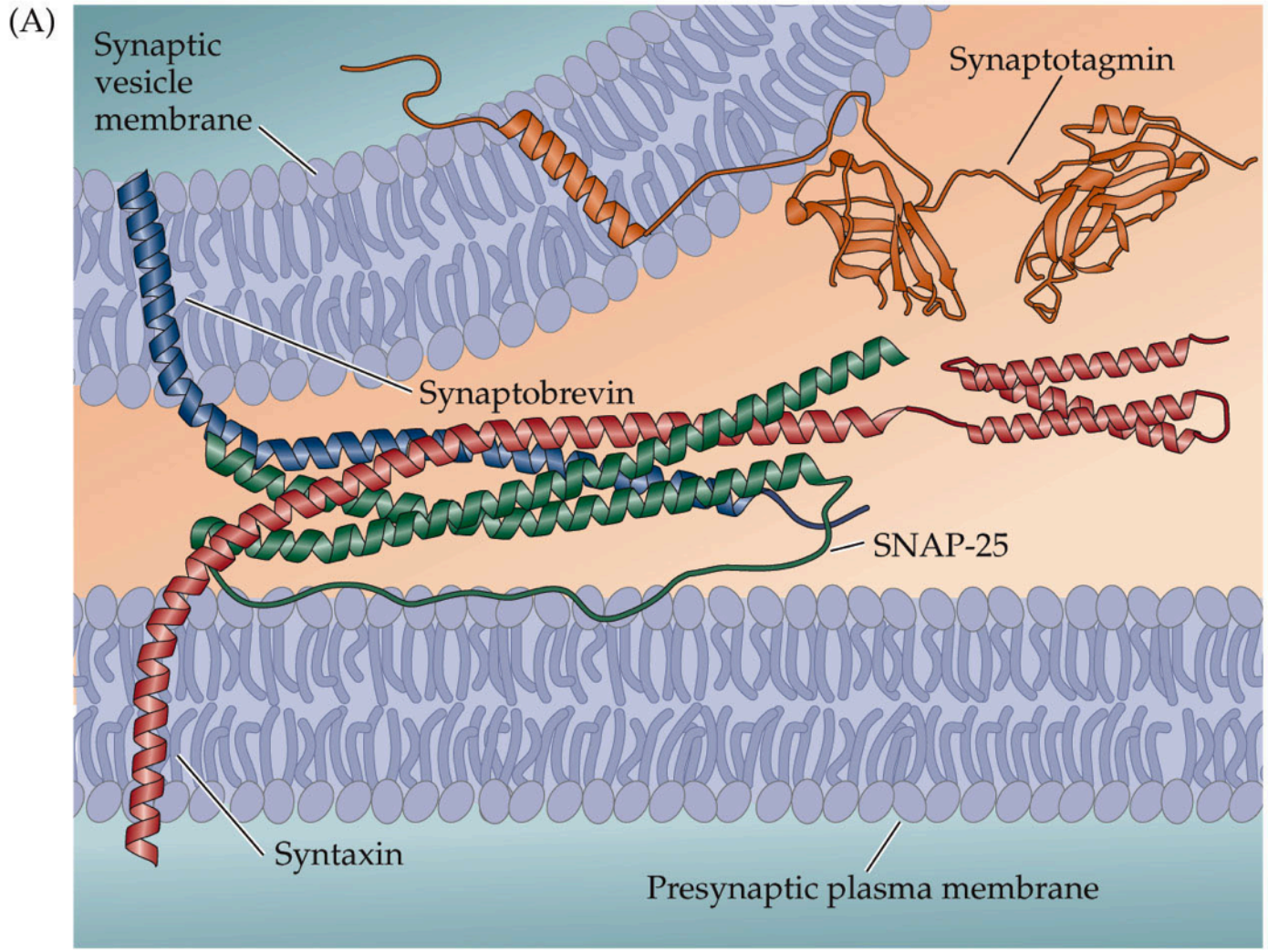
(2) SNARE complexes form to pull membranes together



NEUROSCIENCE, *Fourth Edition,* Figure 5.14 (Part 2)

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The SNAREs form a coiled-coil



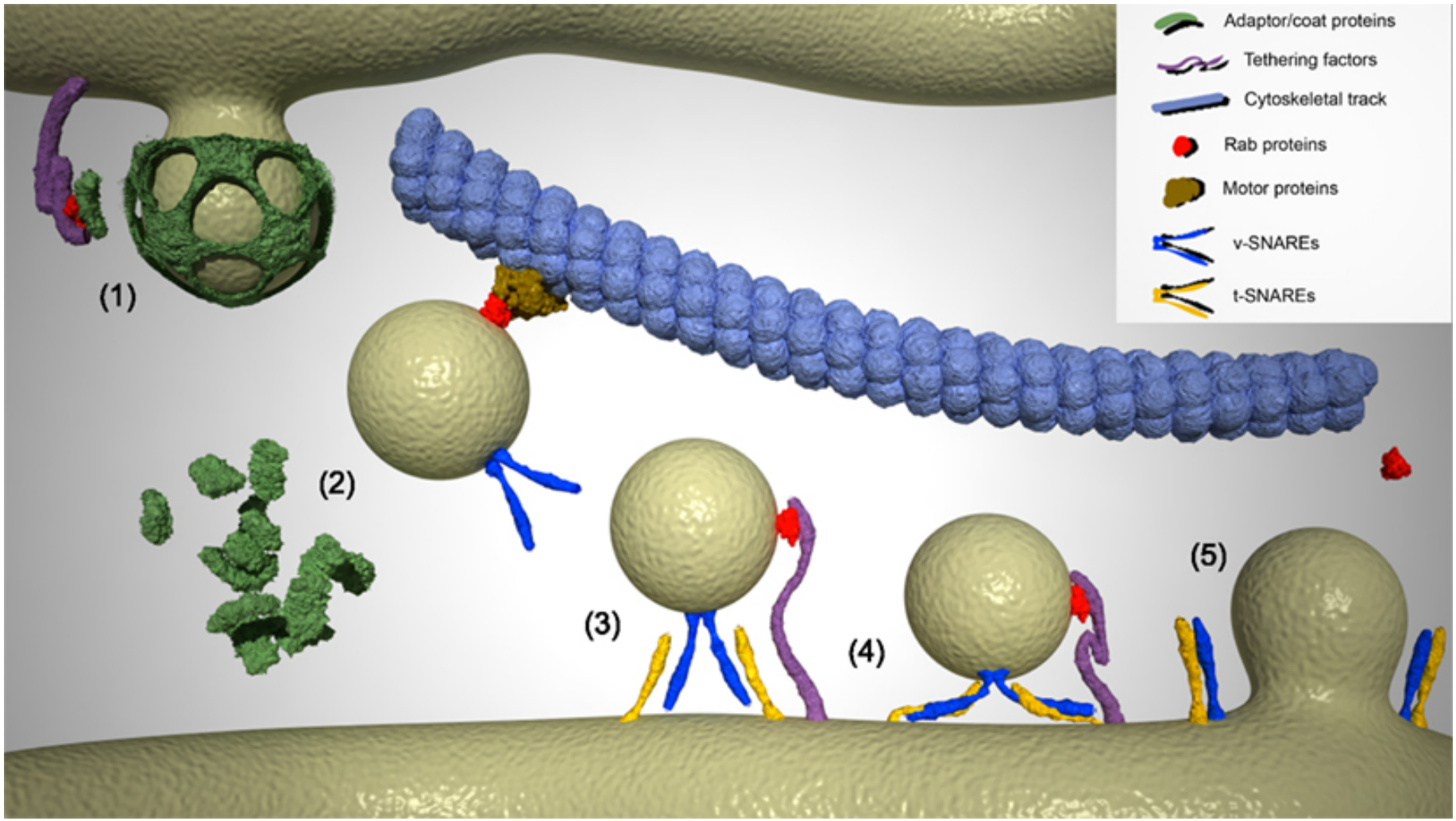
***NEUROSCIENCE, Fourth Edition,* Figure 5.14 (Part 1)**

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Soluble NSF *(N-Ethylmaleimide-Sensitive Factor)* Attachment Protein Receptor)

SNAREs also play a role in targeting

Rab3: -Small proteins that bind to GTP and to synaptic vesicles



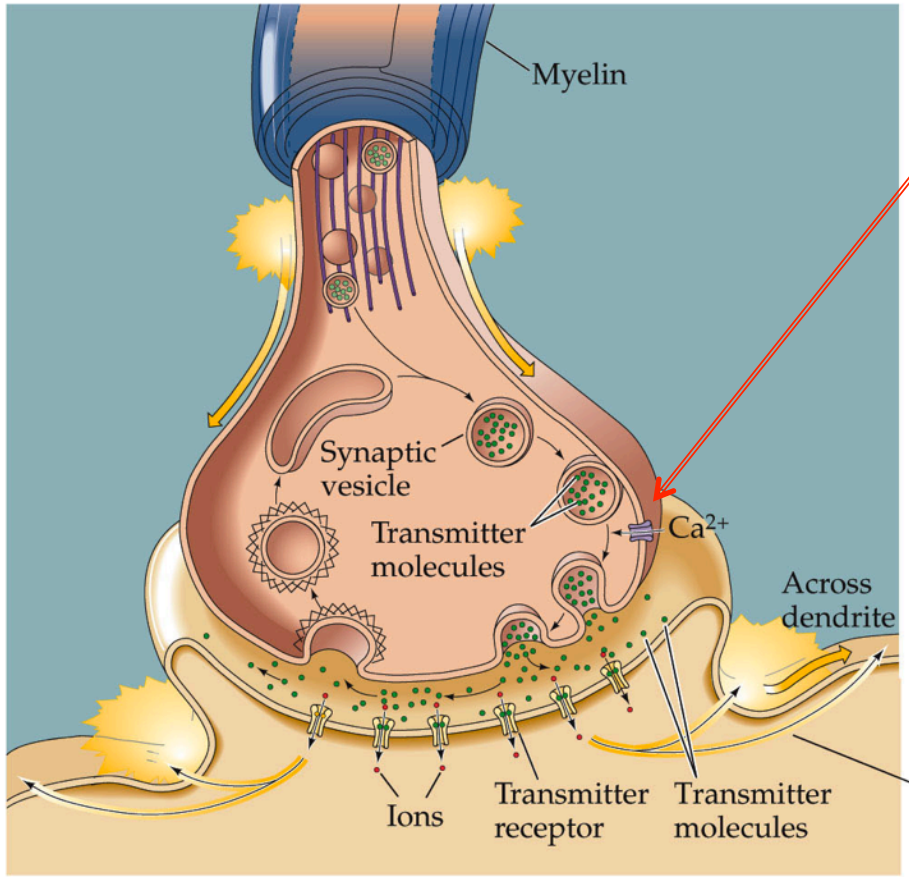
Important Point # 3

The SNAREs target vesicles to the right location and hold them in close proximity to the membrane

Vesicles ready to go....now what?

Step 3: Exocytosis!

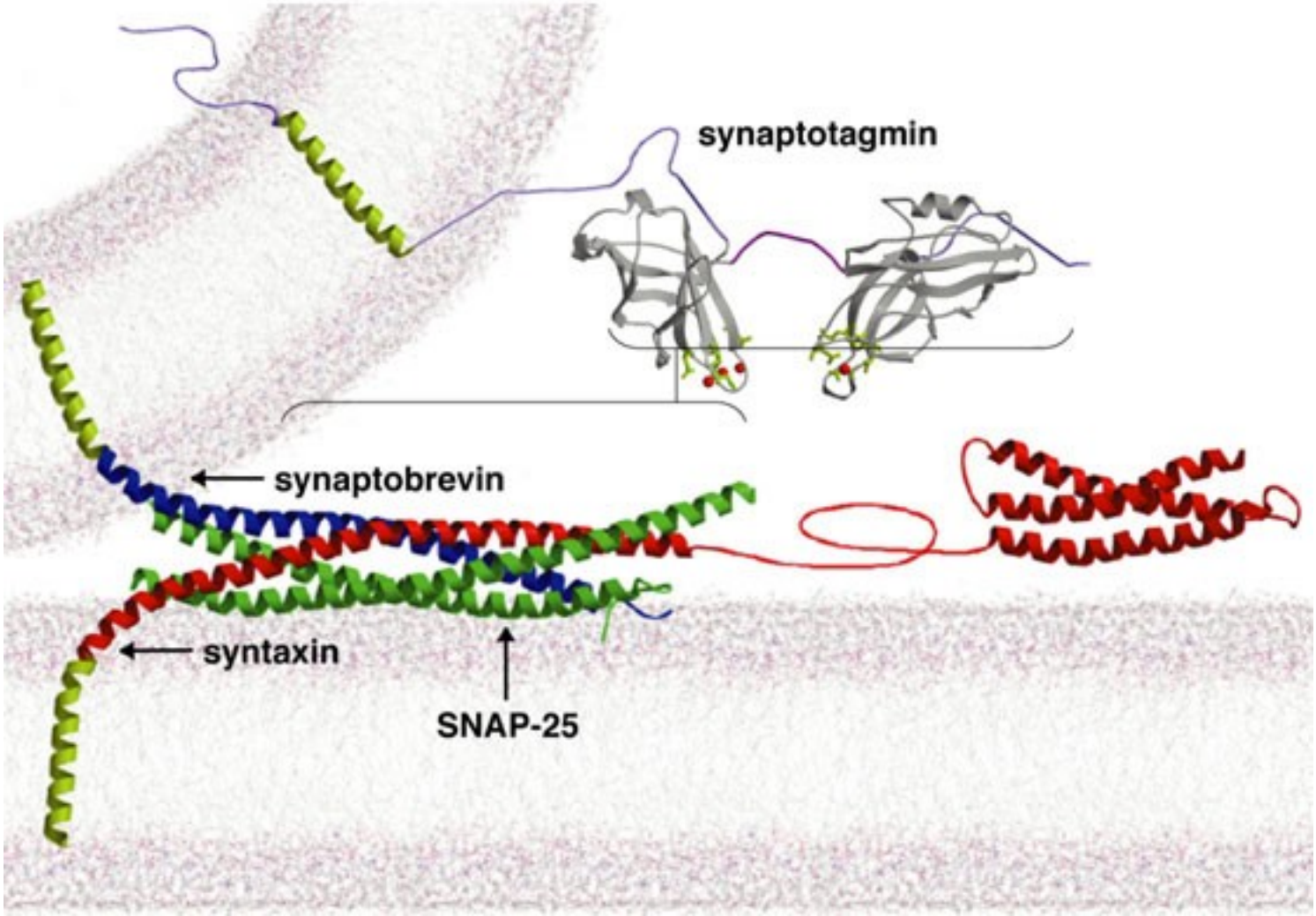
Calcium-dependent!



***NEUROSCIENCE, Fourth Edition,* Figure 5.3**

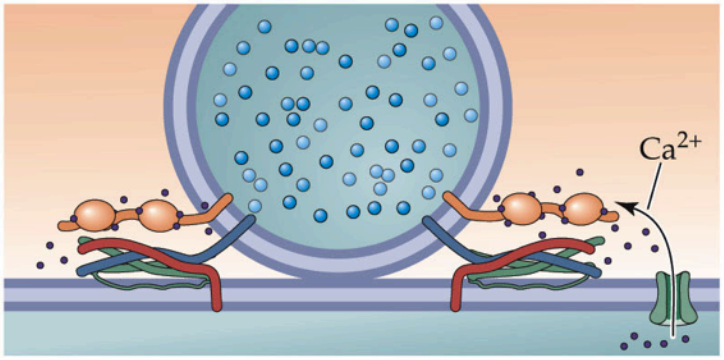
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Exocytosis is regulated by calcium

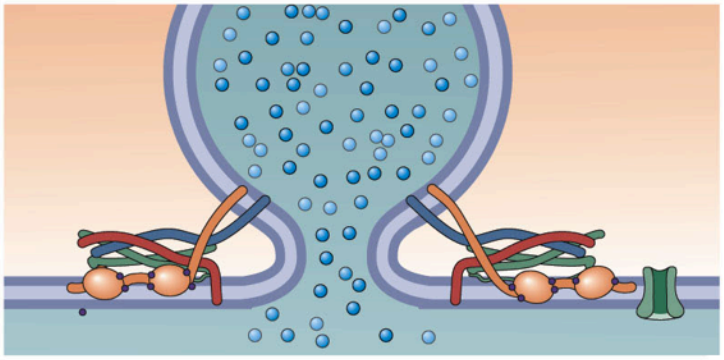


Vesicle fusion is triggered by Calcium

(3) Entering Ca^{2+} binds to synaptotagmin



(4) Ca^{2+}-bound synaptotagmin catalyzes membrane fusion



What is the Ca^{++} sensor?

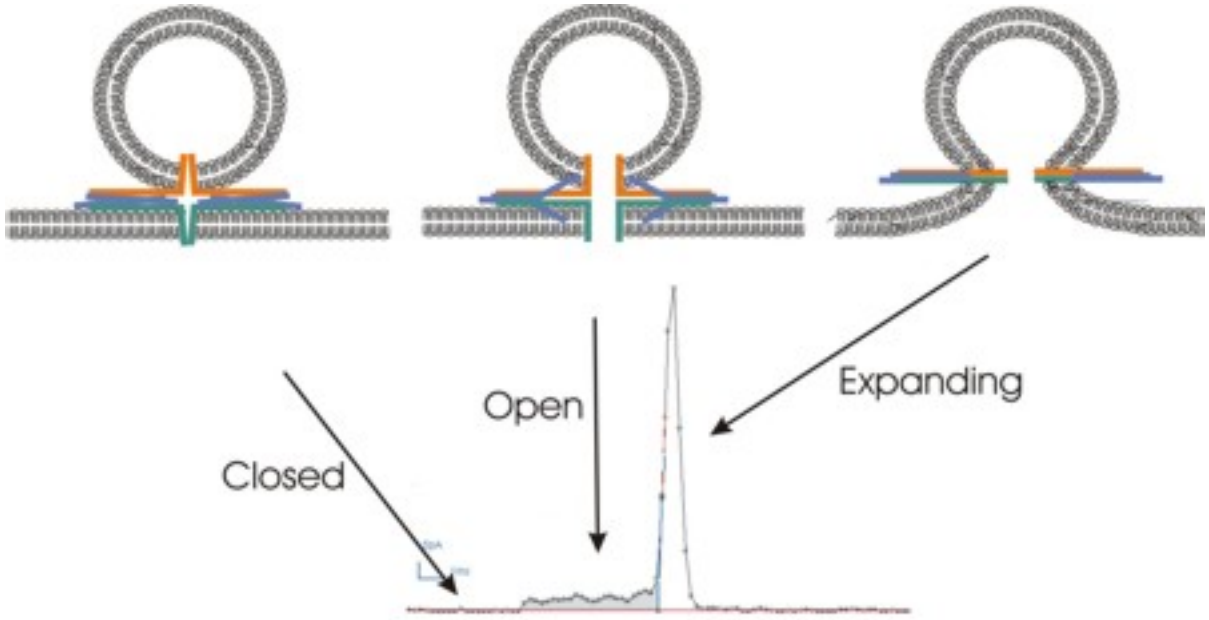
***NEUROSCIENCE, Fourth Edition,* Figure 5.14 (Part 3)**

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Important Point

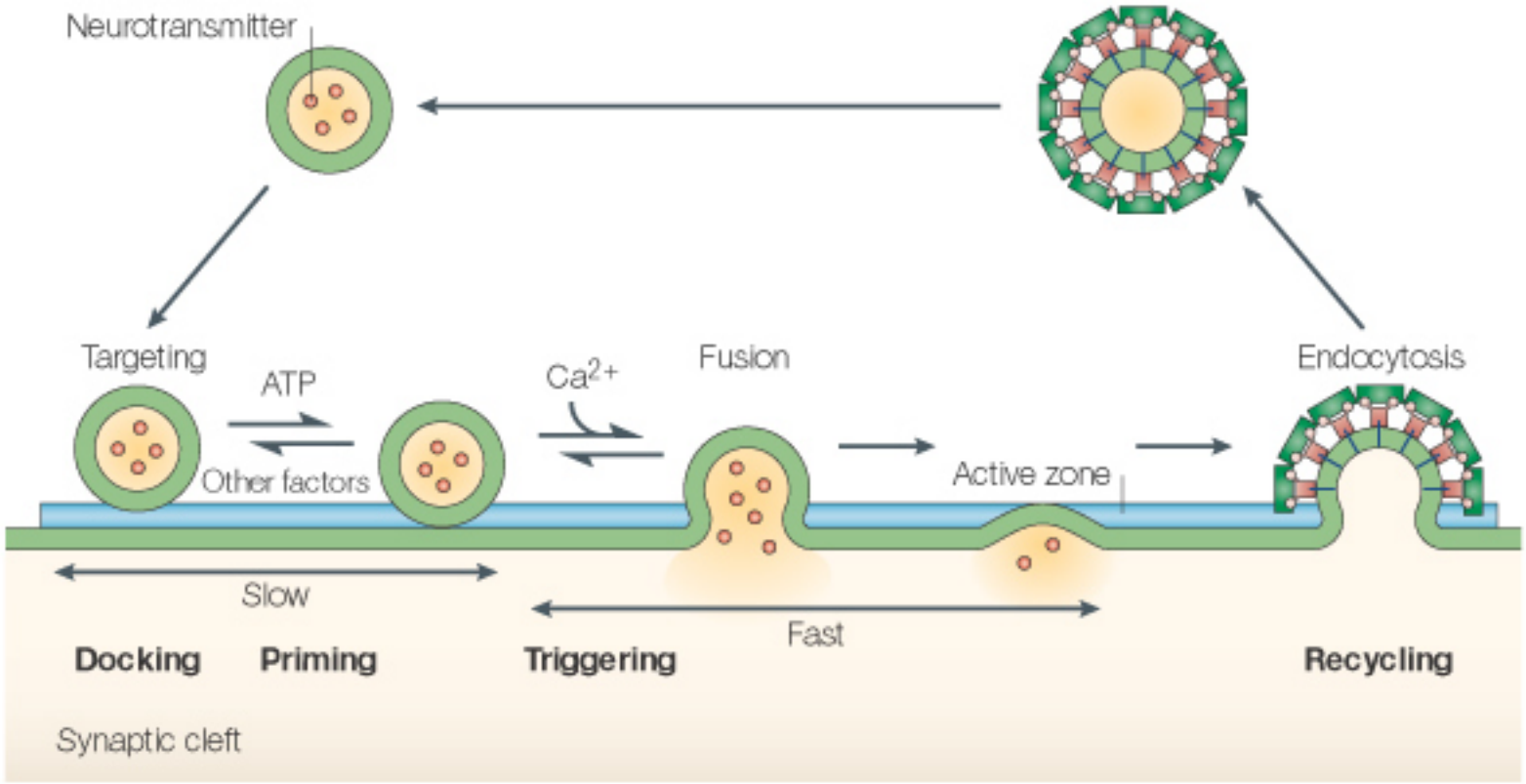
Exocytosis (vesicle fusion) is triggered by calcium

A fusion pore opens and neurotransmitter is released into the synaptic cleft



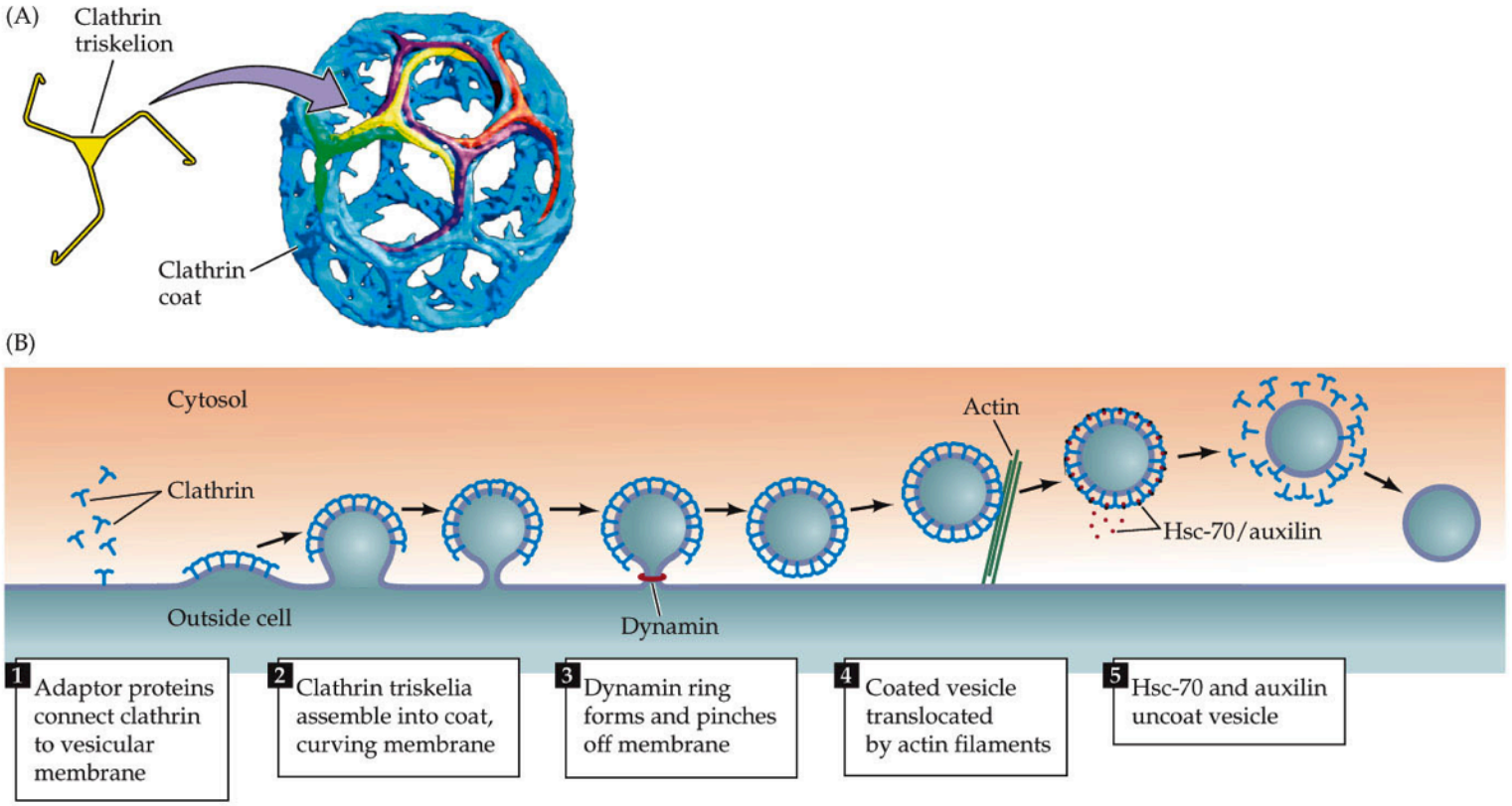
video time!

Endocytosis- vesicle retrieval



Nature Reviews | Molecular Cell Biology

Clathrn is in charge of endocytosis



NEUROSCIENCE, Fourth Edition, Figure 5.15

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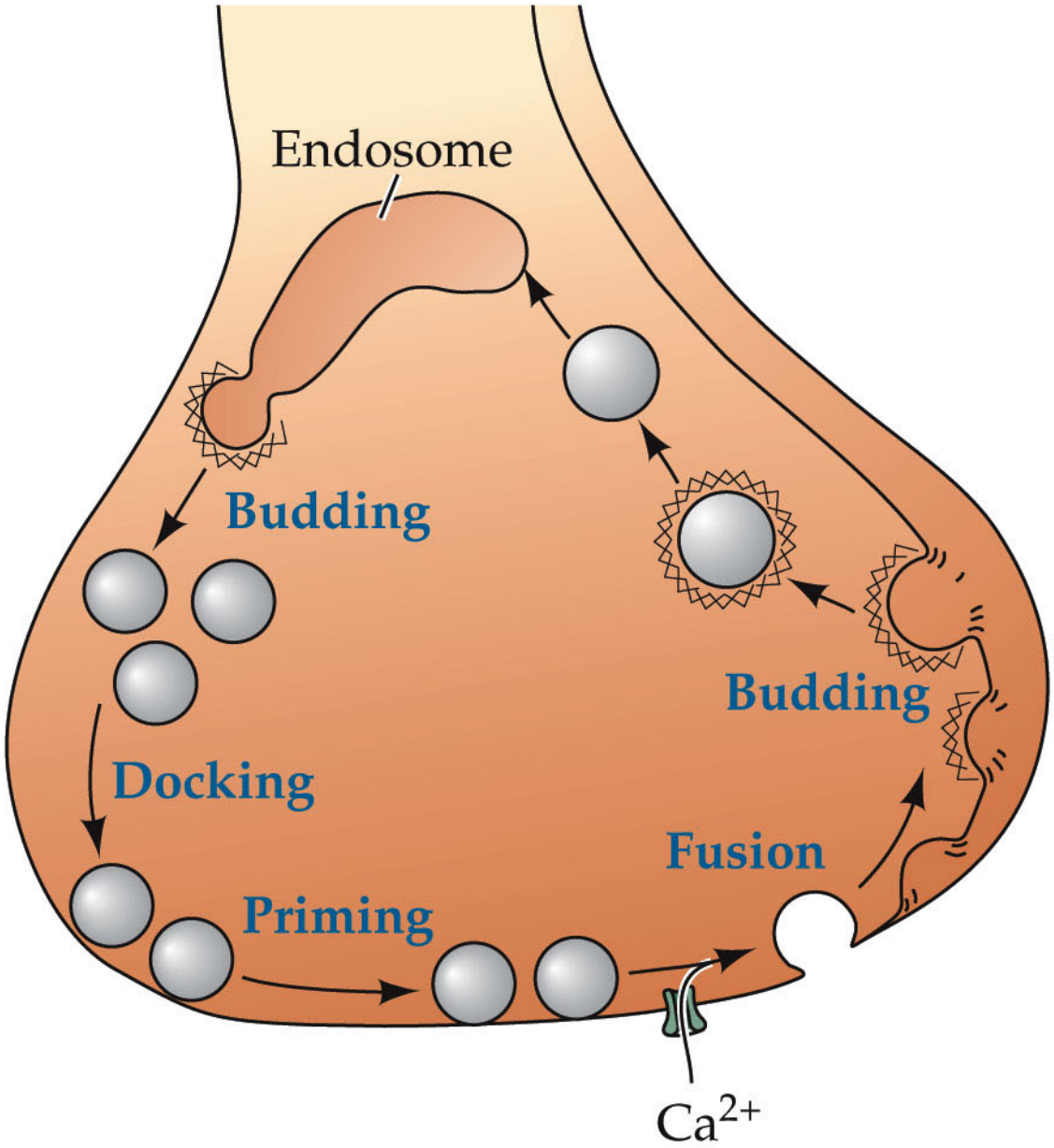
Review of synaptic vesicle exocytosis

1) Target the vesicles to the active zone: Rab3

2) Dock the vesicle to the active zone of the membrane and prime for vesicle fusion: The SNAREs

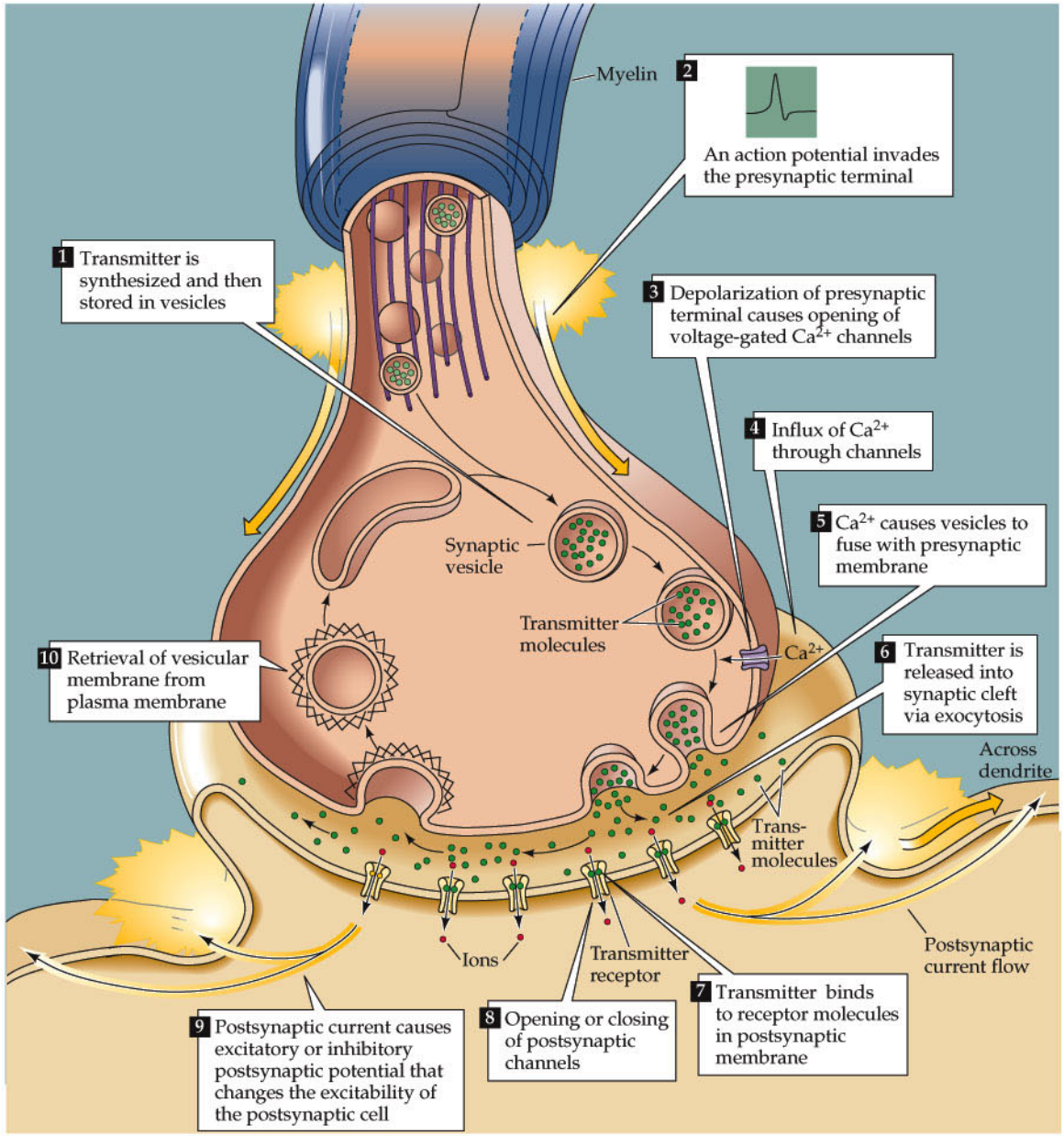
3) Ca^{++}- mediated exocytosis: fusion of the vesicle with the plasma membrane: synaptotagmin is the calcium sensor

4) Endocytosis: recycling of the vesicle membrane and reformation of empty vesicle



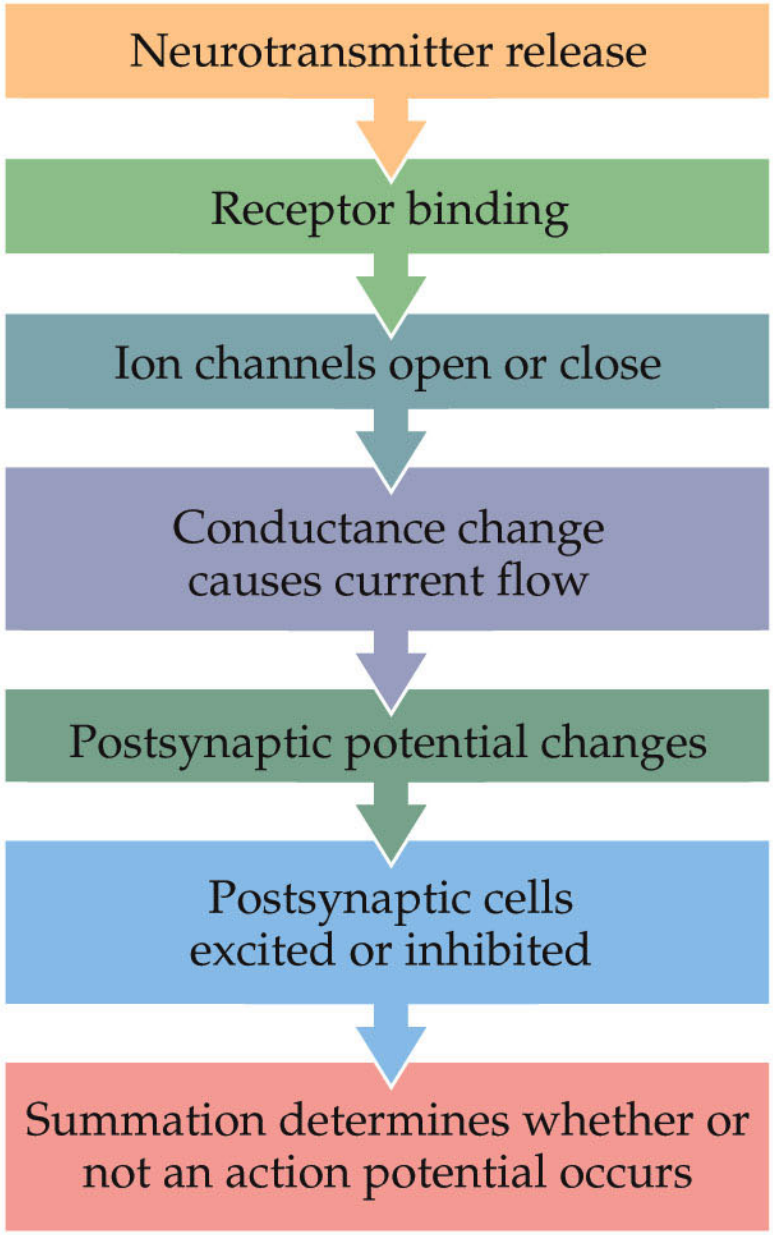
***NEUROSCIENCE, Fourth Edition,* Box 5B**

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***NEUROSCIENCE, Fourth Edition,* Figure 5.3**

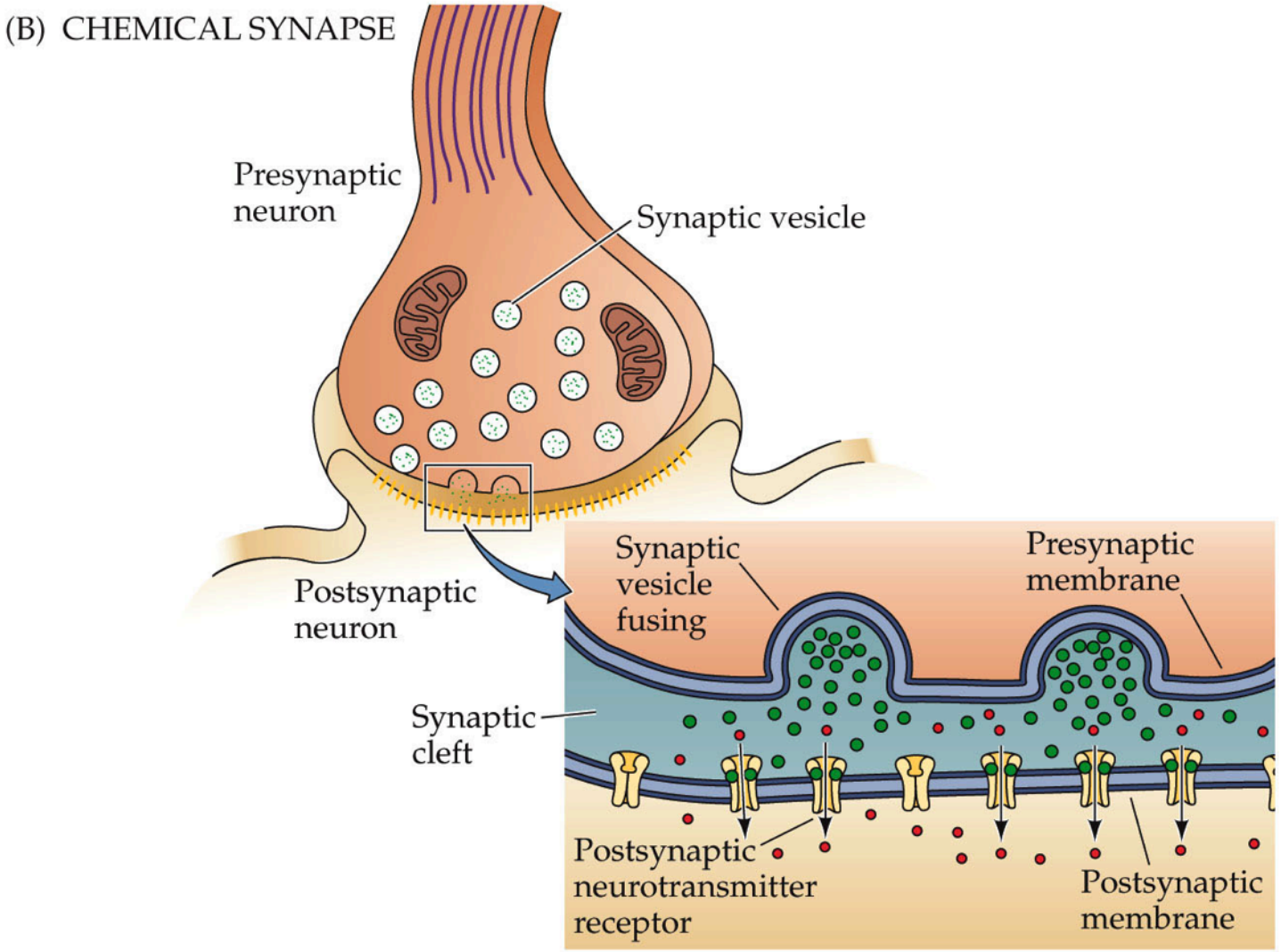
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***NEUROSCIENCE, Fourth Edition,* Figure 5.22**

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The post-synaptic response: binding of the neurotransmitter



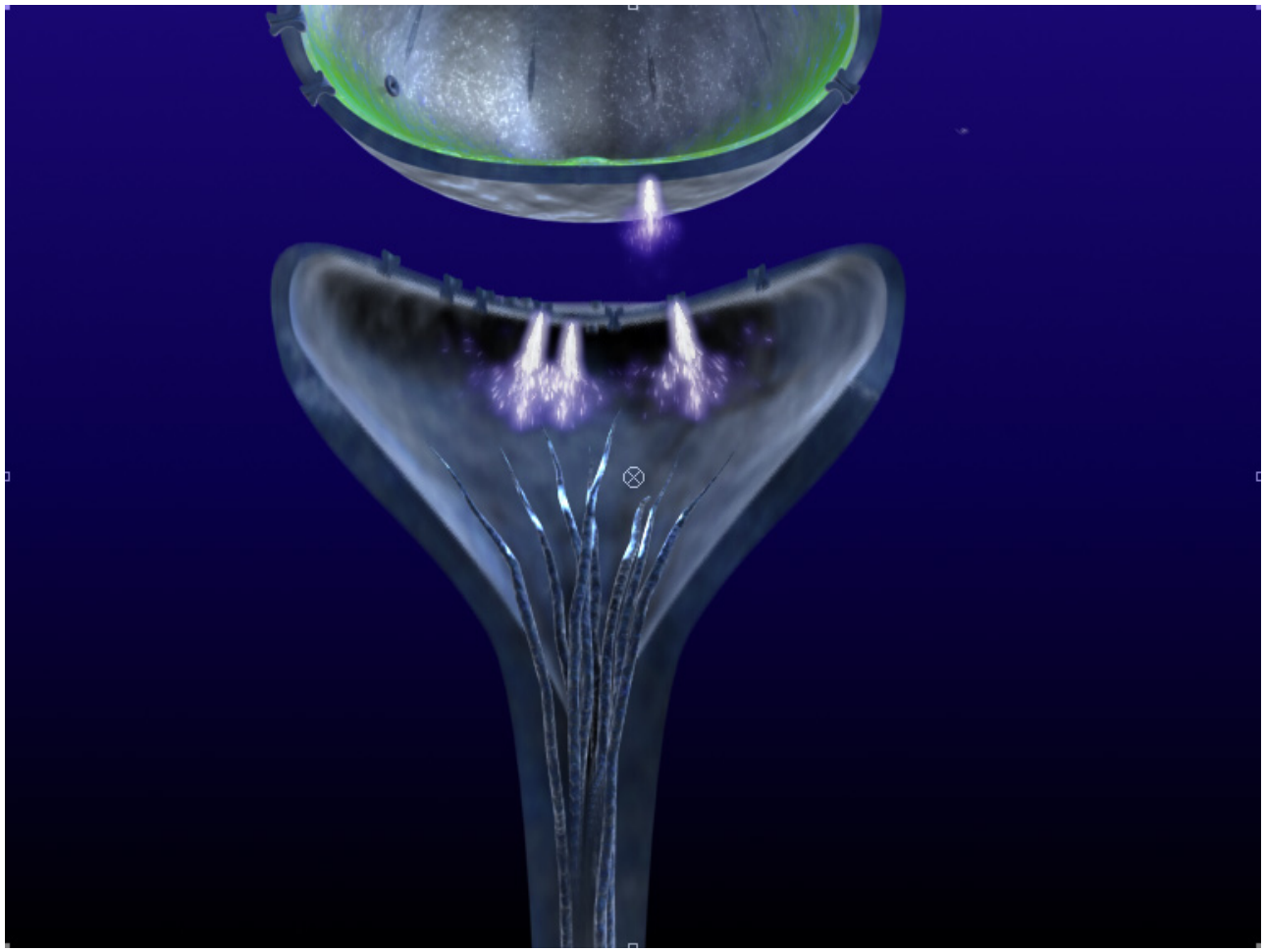
***NEUROSCIENCE, Fourth Edition,* Figure 5.1 (Part 2)**

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The post-synaptic response

What happens to the neurotransmitter after it is released into the synapse?

Note: This picture is cool looking, but biologically goofy.



Ion Channel Review

-Transmembrane passes- usually 6

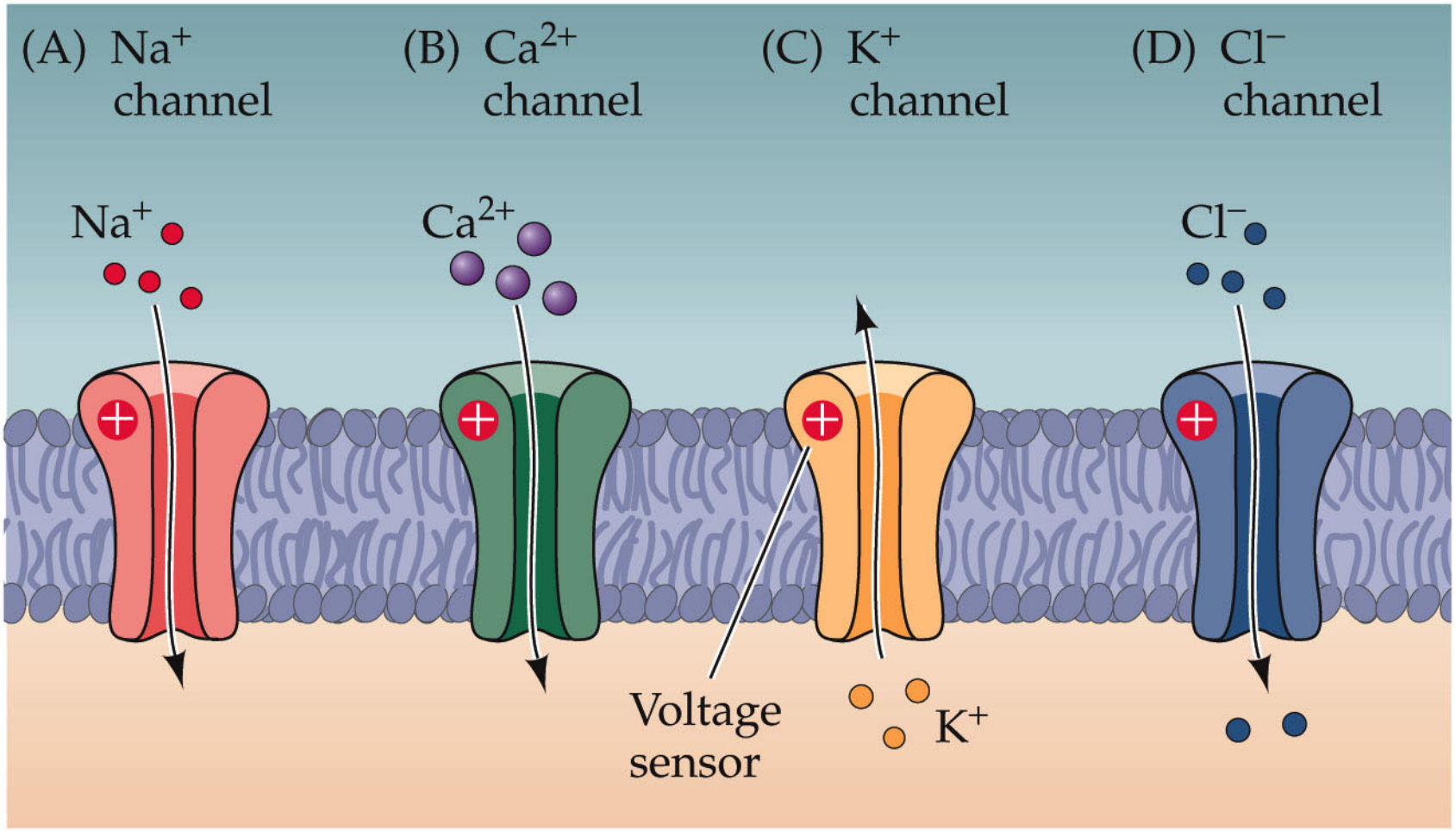
-Pore domain

-Selectivity filter

-Voltage sensor – think back to Marco's lecture

-Allows ions to pass through the membrane according to their electrochemical gradients

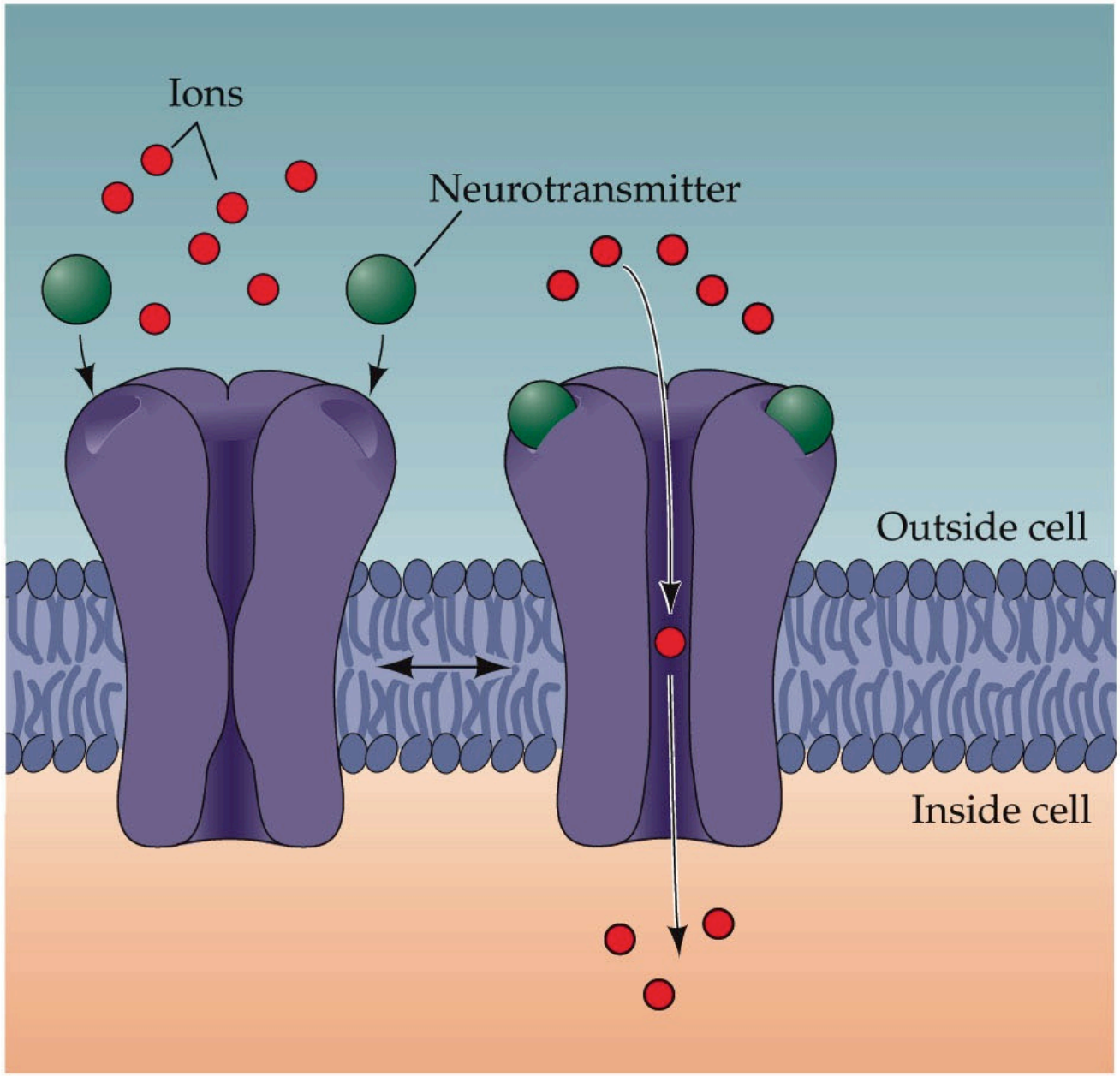
VOLTAGE-GATED CHANNELS



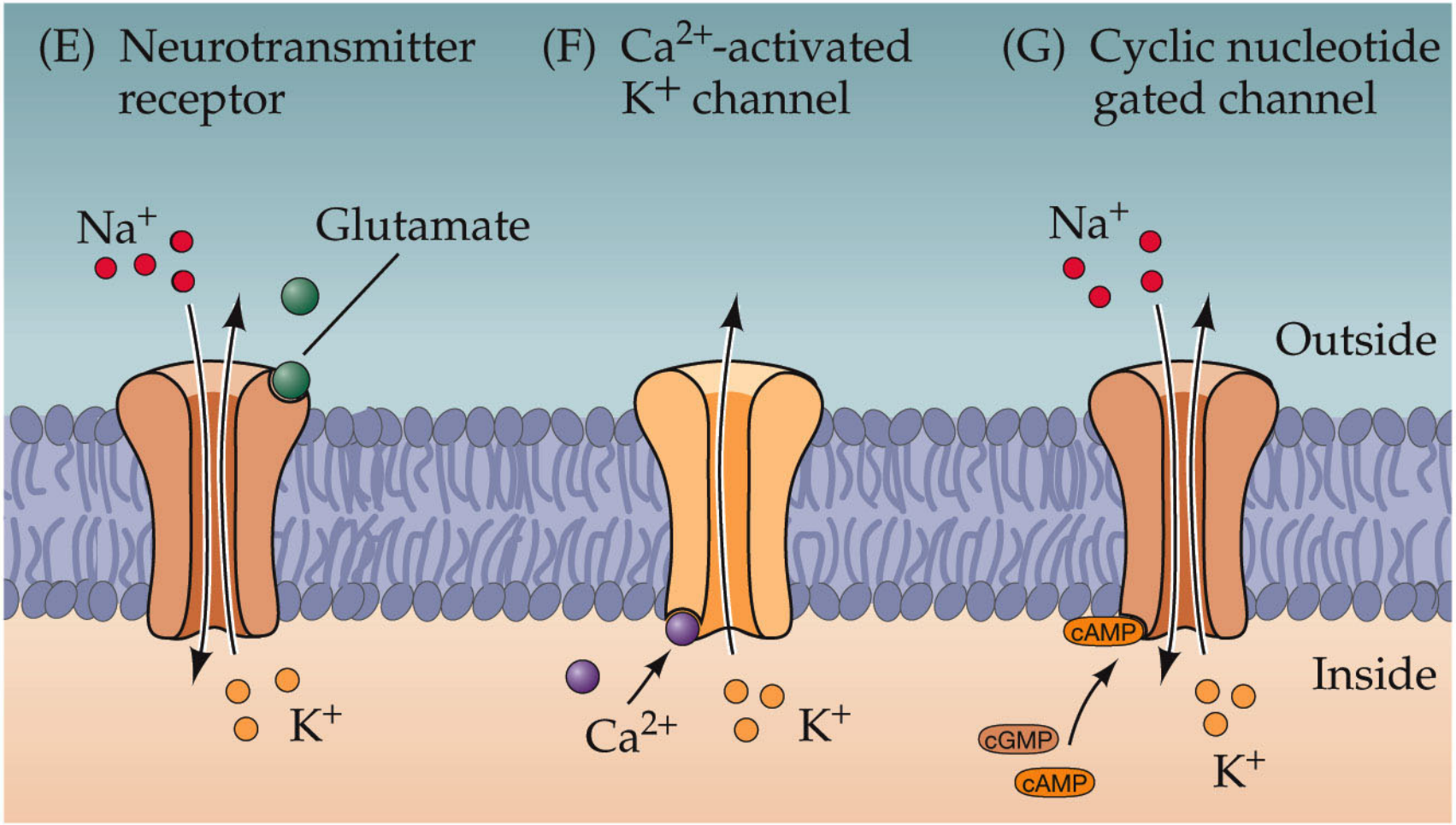
***NEUROSCIENCE, Fourth Edition,* Figure 4.4 (Part 1)**

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(A) LIGAND-GATED ION CHANNELS



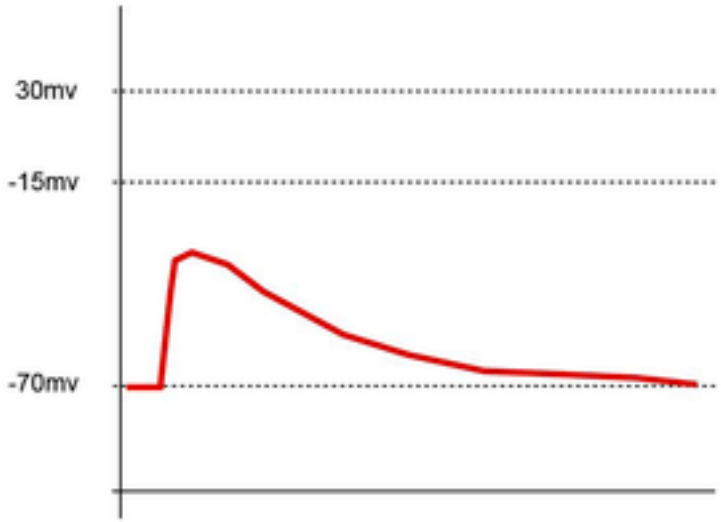
LIGAND-GATED CHANNELS



***NEUROSCIENCE, Fourth Edition,* Figure 4.4 (Part 2)**

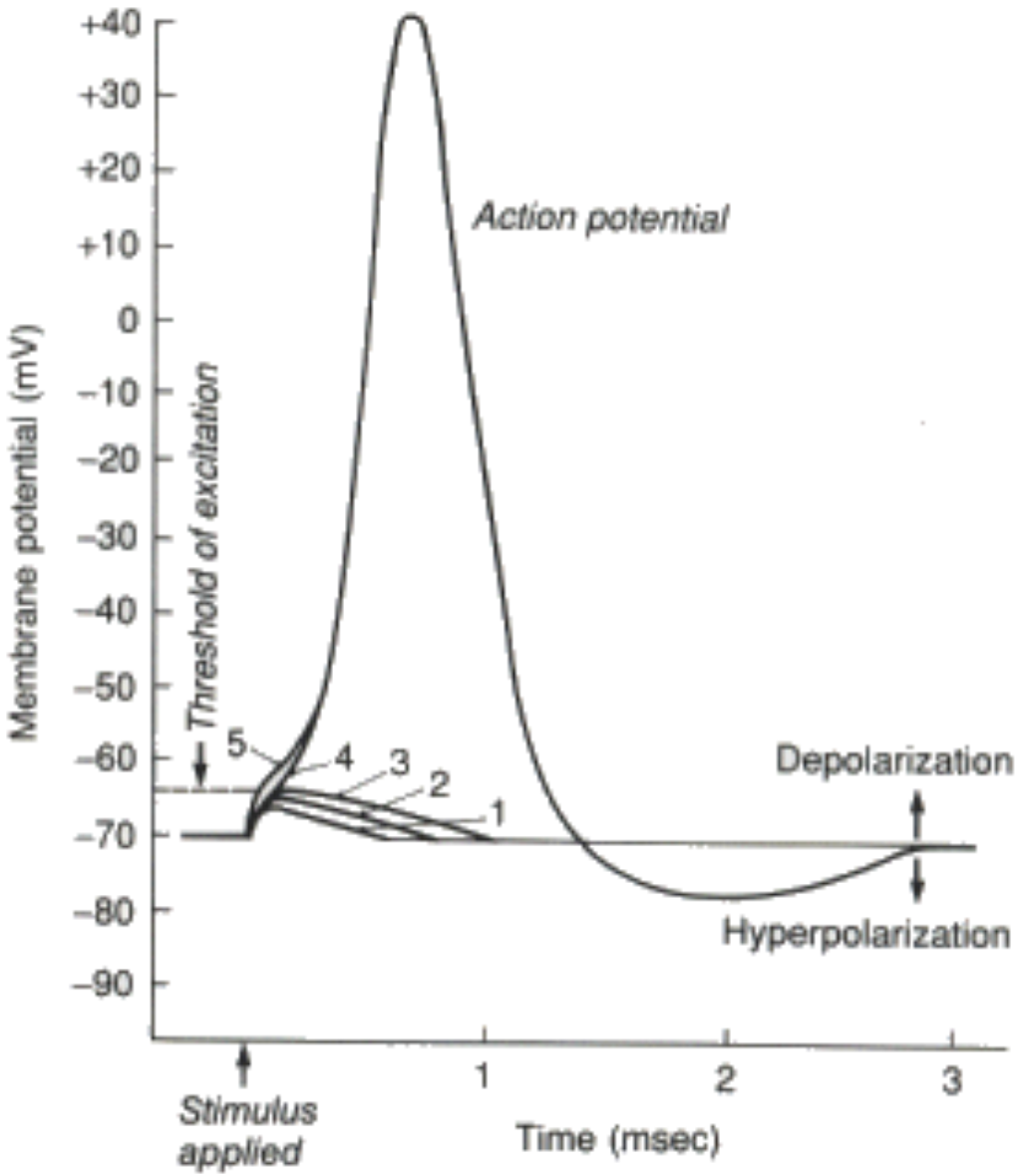
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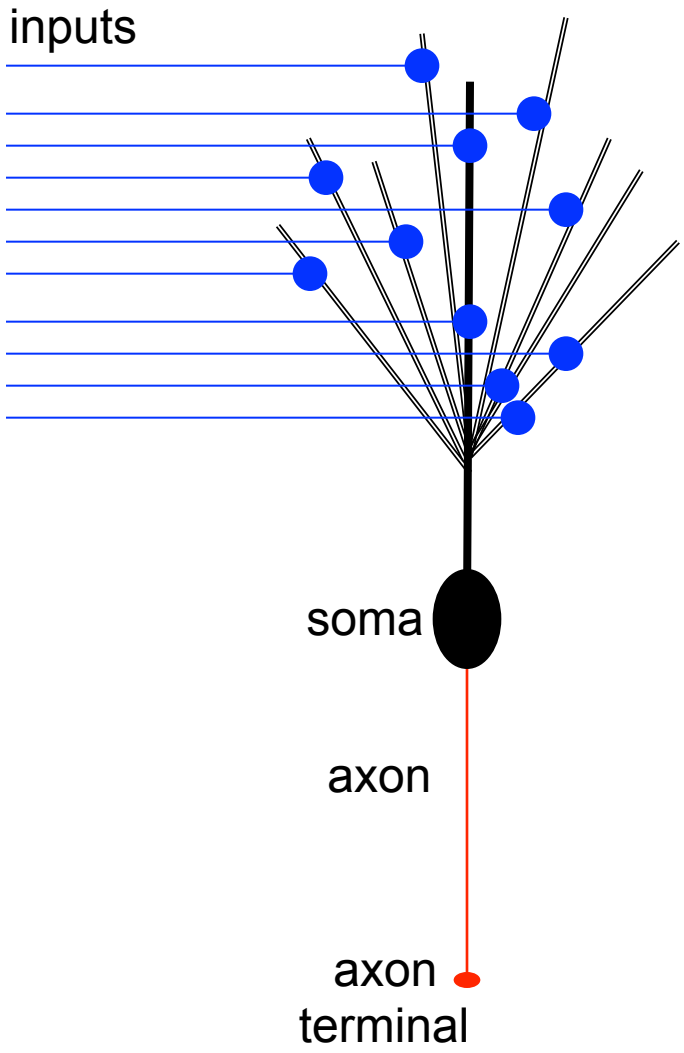
The change in potention (caused by the entry of ion channels) is called an excititory post-synaptic potential



The potential travels down the dendrite passively – no active transmission like during an action potential

Many inputs may be needed to cause the cell to spike





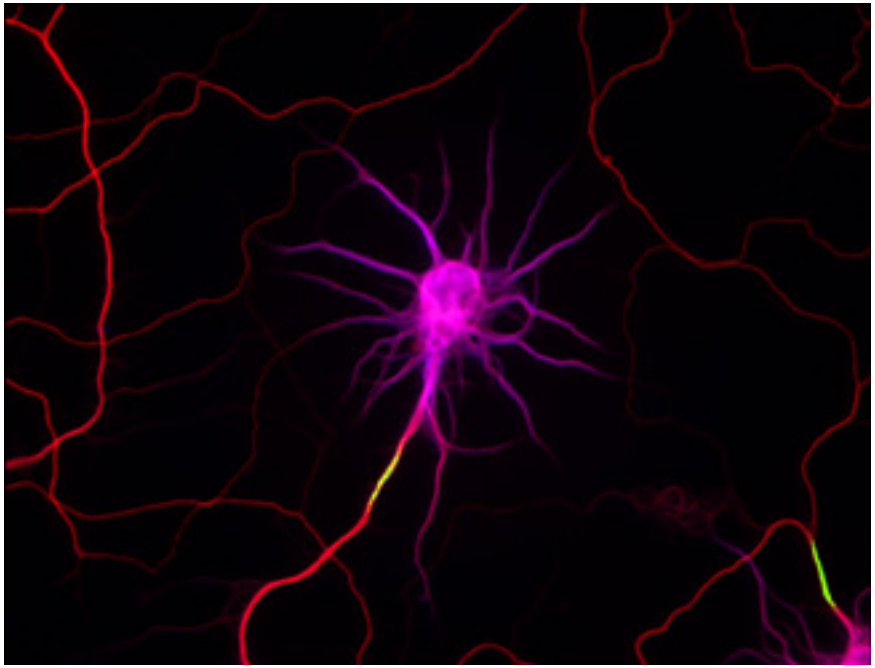
simple computation by a neuron:

if $\sum_{\substack{\text{(sum)} \\ \text{inputs}}} > V_{\text{threshold}}$

then,

fire action potential

Triggering an action potential



If the EPSP is large enough or if multiple EPSPs reach the soma at the same time, they will encounter a dense concentration of sodium channels. This region is called the **axon initial segment**

What happens next?

Putting it all together

neurotransmission

Let's talk about neurotransmitters

What are the criteria to be called a neurotransmitter?

1) Must be produced in a neuron and released at the synapse following action potential arrival
2) Must produce an effect in the post-synaptic cell
3) Must be rapidly removed from the cleft
4) Must have the same effect when applied experimentally as when secreted by the pre-synaptic neuron

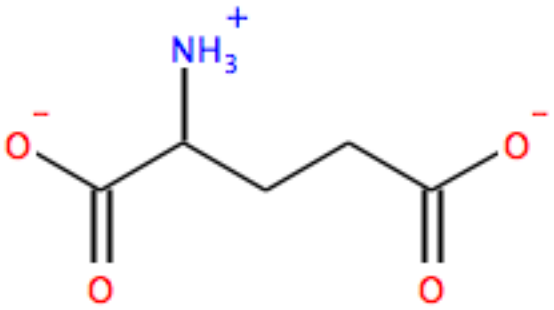
Over 60 molecules meet these criteria!

To qualifty as a neurotransmitter

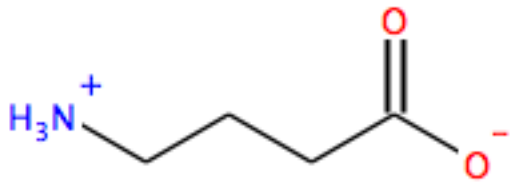
- Precursors and/or **synthesis enzymes** located in the presynaptic side of the synapse.
- The chemical is present in the presynaptic element.
- It is available in sufficient quantity in the presynaptic neuron to affect the postsynaptic neuron.
- There are **postsynaptic receptors** and the chemical is able to bind to them.
- A biochemical mechanism for **inactivation** is present.

What is a neurotransmitter

- For now, three main groups:
- 1) Excititory neurotransmitter: Glutamate
- 2) Inhibitory neurotransmitter: GABA
- 3) Modulatory neurotransmitters: Dopamine, serotonin, acetylcholine, et al.



Glutamate



Gamma-aminobutryic acid (GABA)

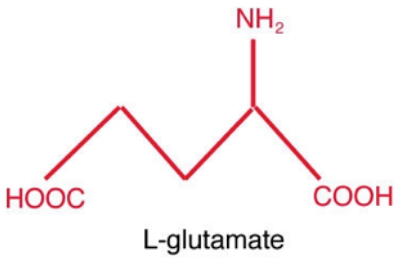
Important point

Neurotransmitters are the chemical signaling molecules of the brain

The two main neurotransmitters are glutamate and GABA

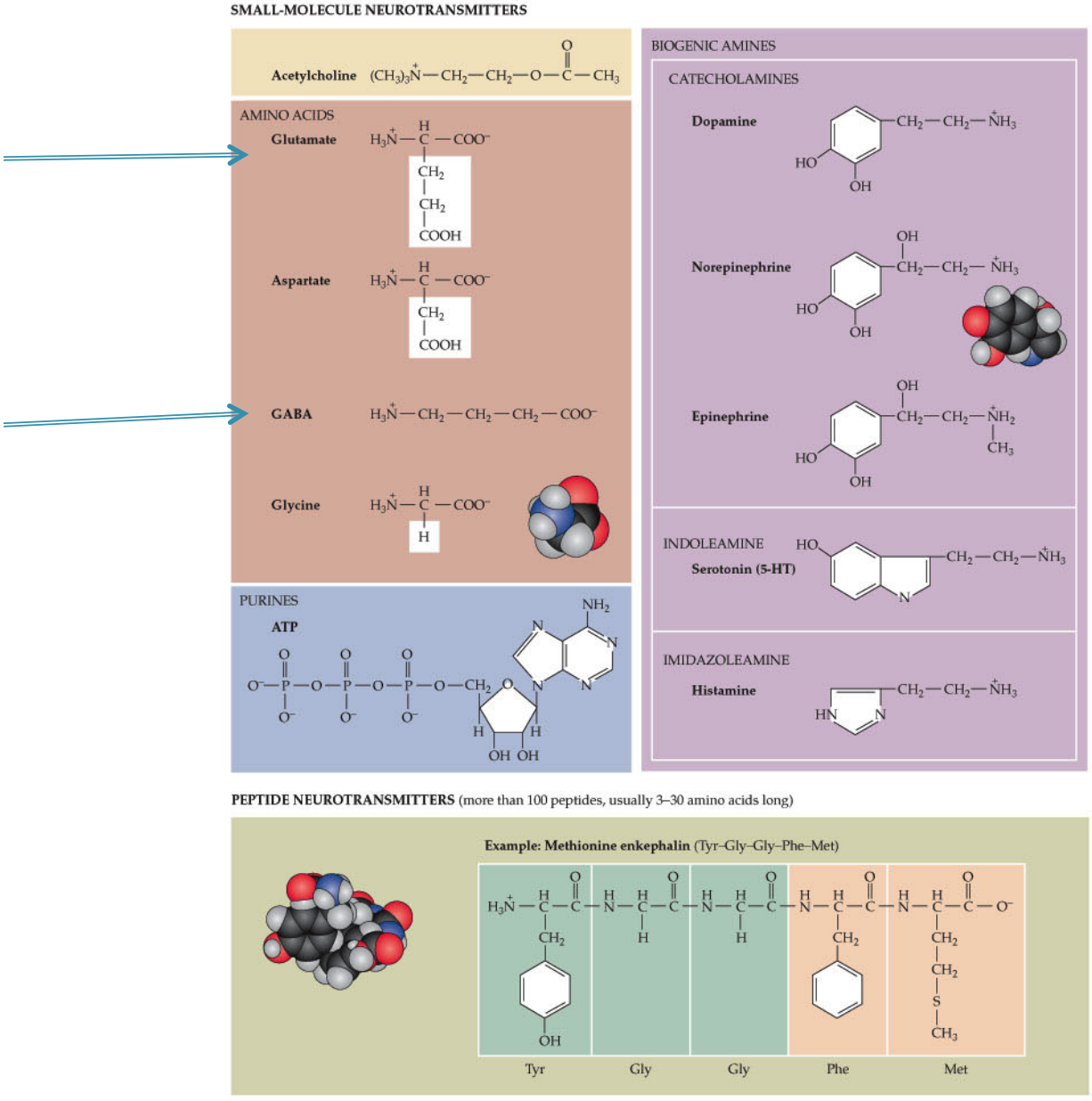
Glutamate: The main excitatory neurotransmitter in the brain

- Produced from alpha-keoglutarate
- After release, taken up from the synaptic cleft by VGLUTs et al



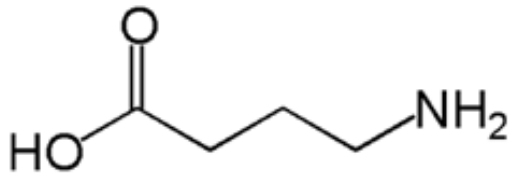
L-glutamate

Neurotransmitters



GABA

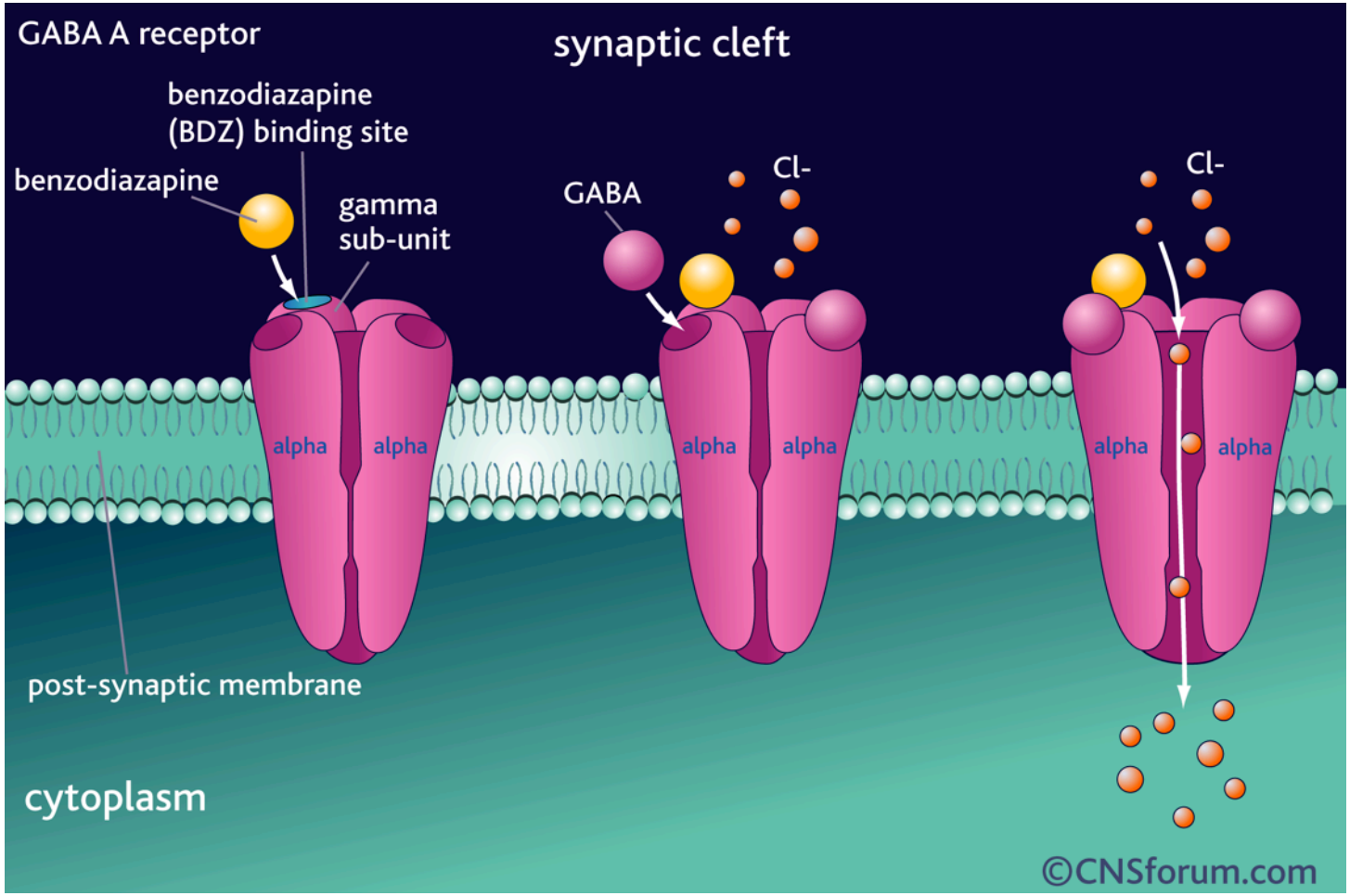
The main **inhibitory** neurotransmitter



excitatory = *depolarizing* = closer to firing action potential

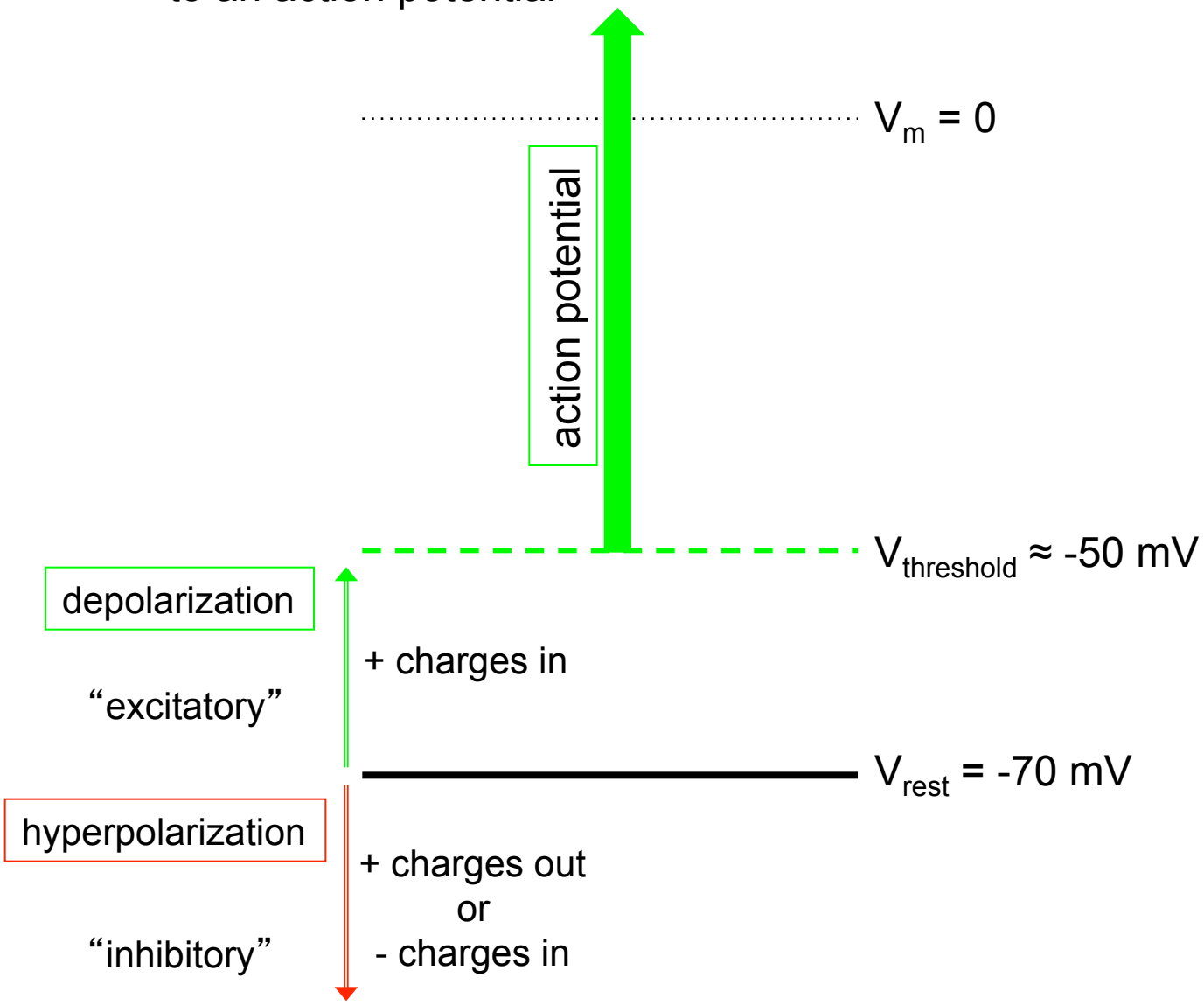
inhibitory = *hyperpolarizing* = further from firing action potential

GABA receptor

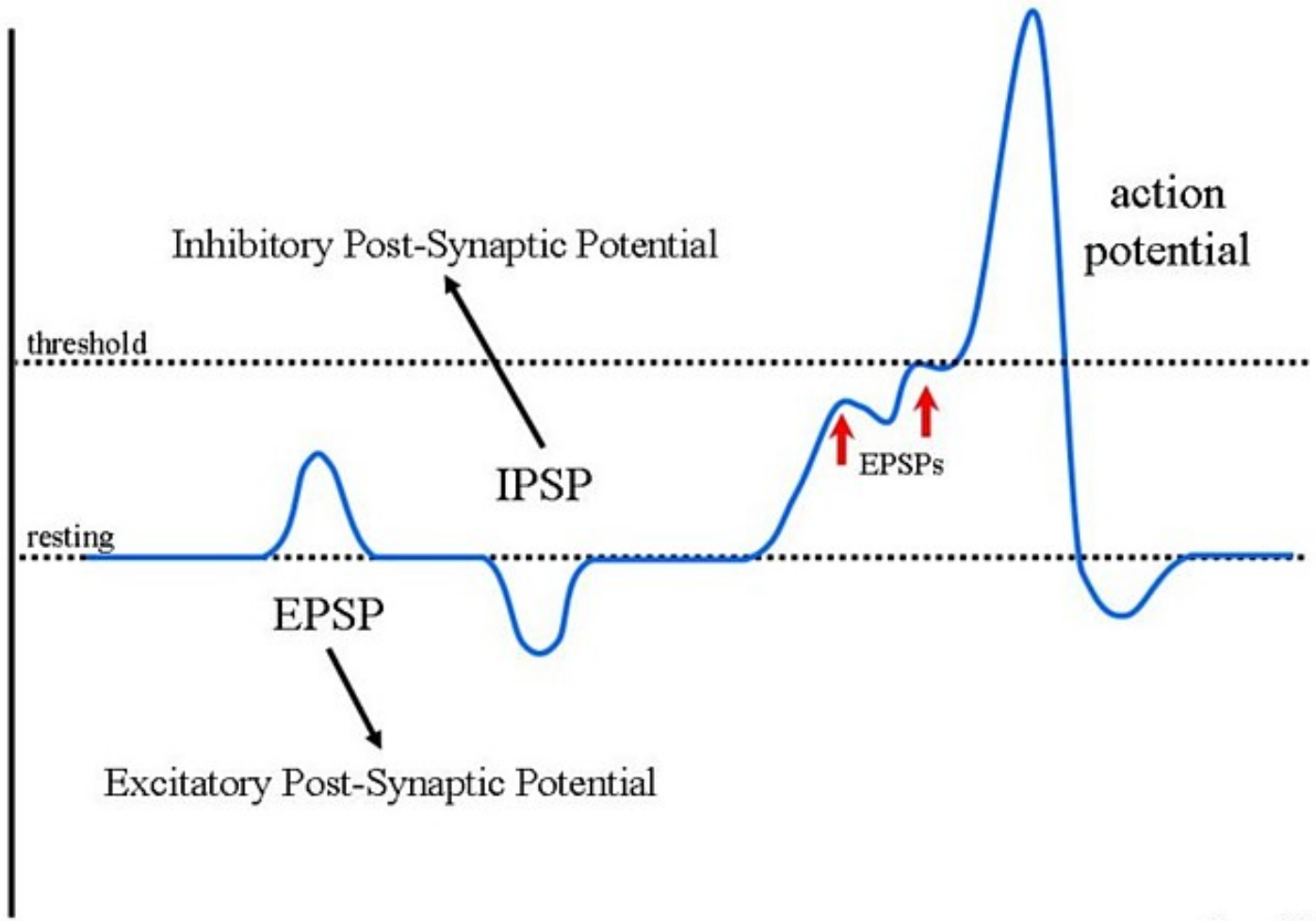


The GABA receptor is a chloride channel

Principle #5: ion flow that moves cell closer to threshold (depolarizes) can lead to an action potential



So what's different about an



see fig. 48.12

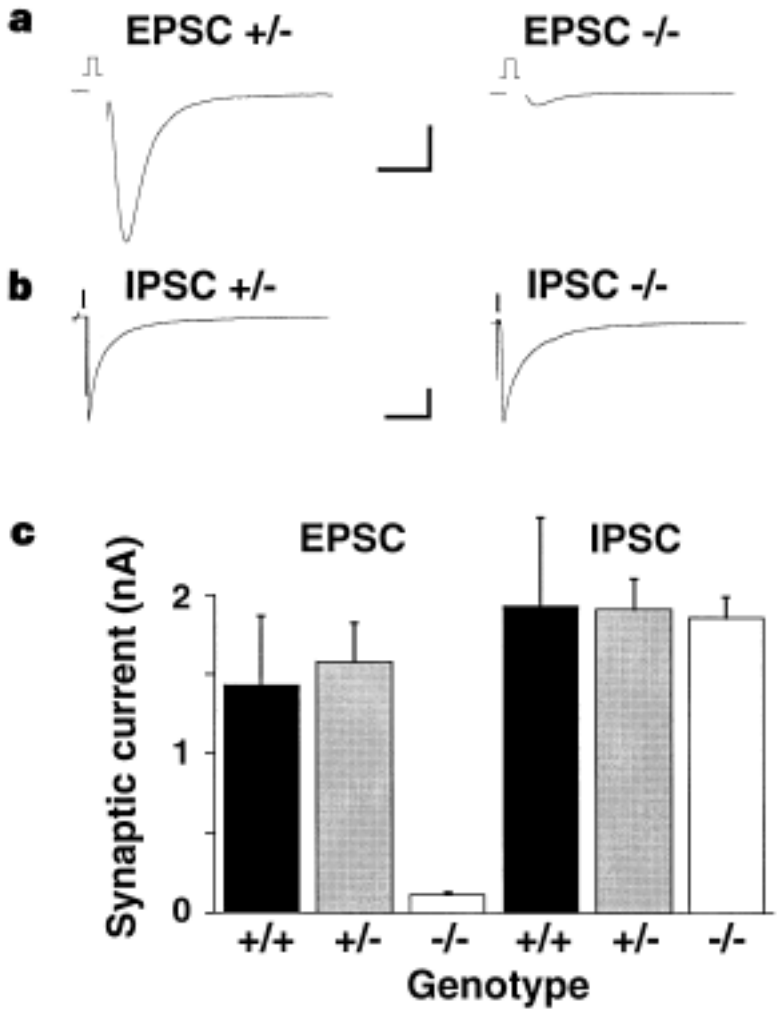
Important points summary

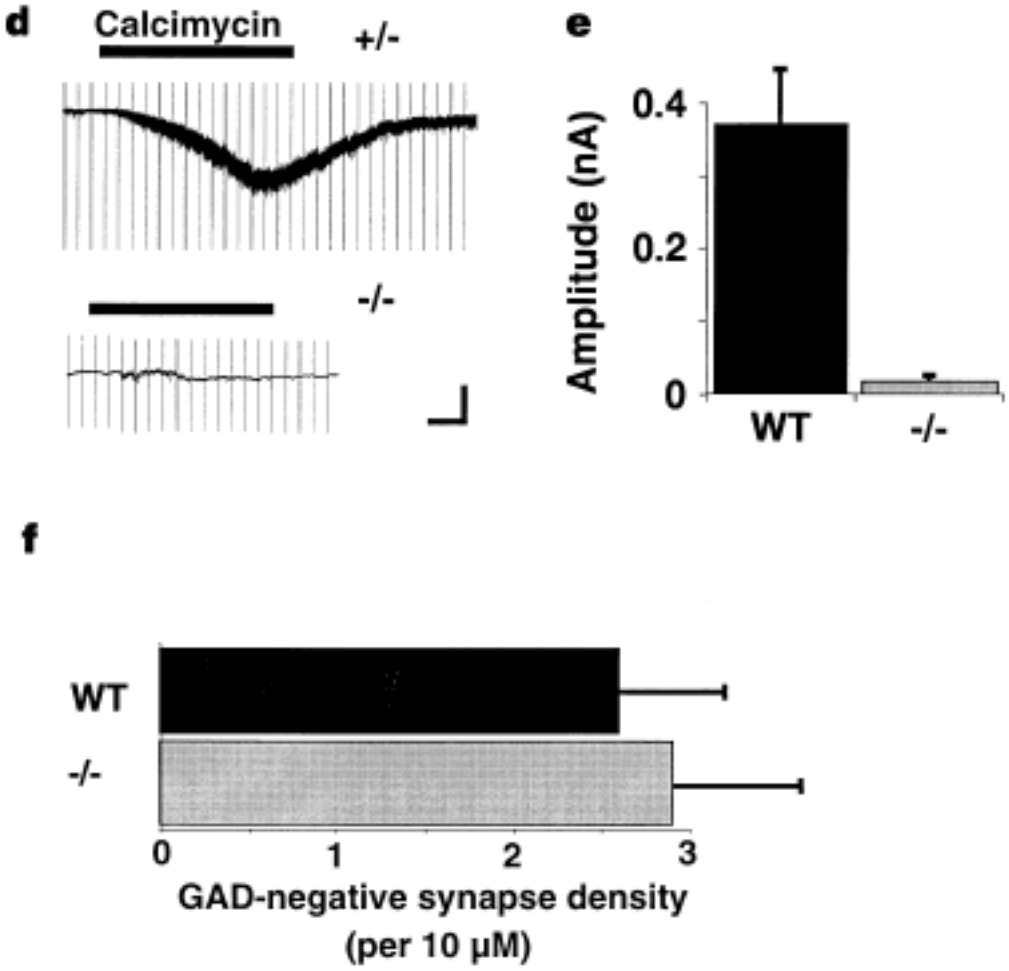
- 1) Neurotransmitters are the chemical signalling molecules of the brain
- 2) Neurotransmitters are packaged into synaptic vesicles
- 3) Exocytosis is the fusion of these vesicles with the plasma membrane and results in the release of neurotransmitter into the synaptic cleft
- 4) Exocytosis is triggered by calcium

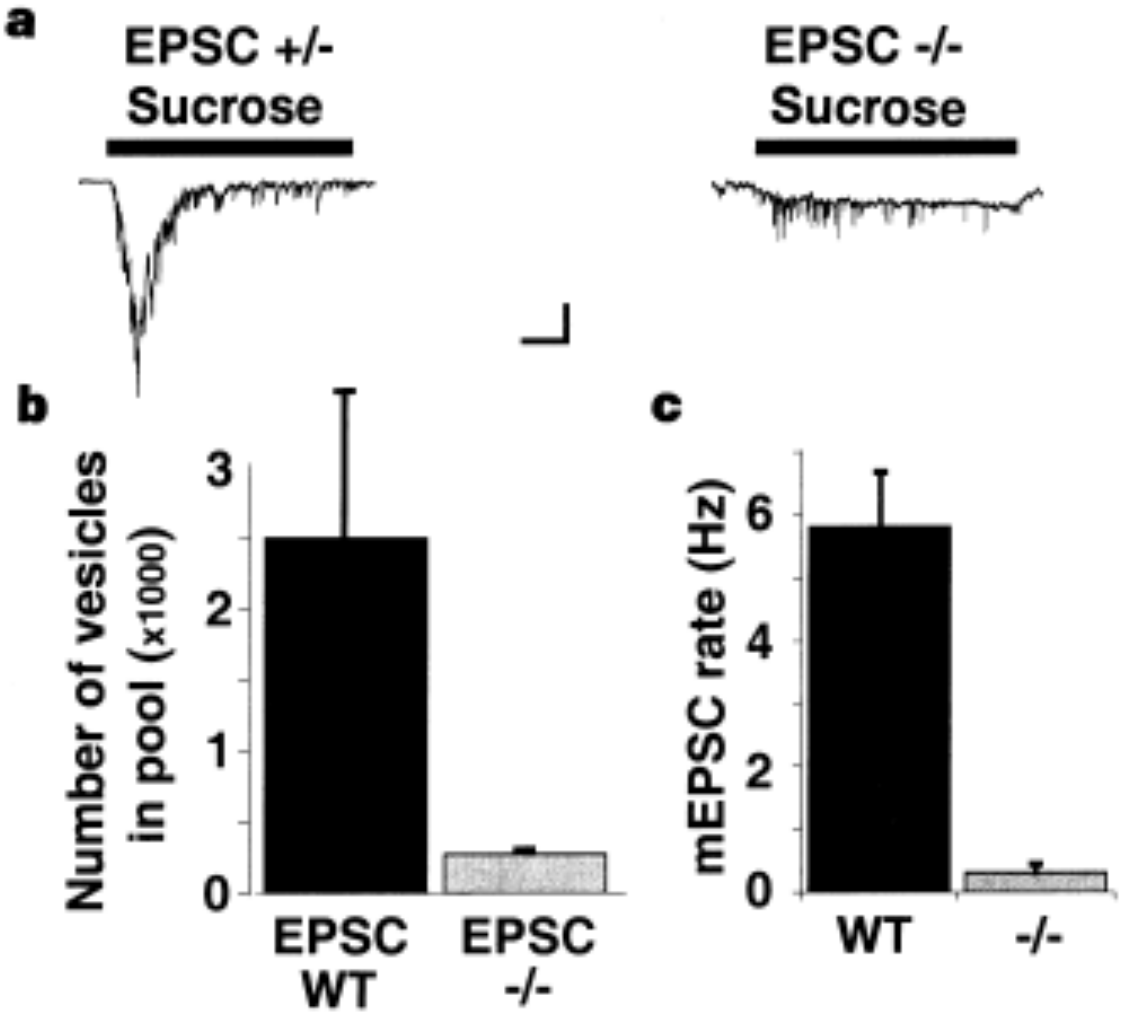
Journal Club

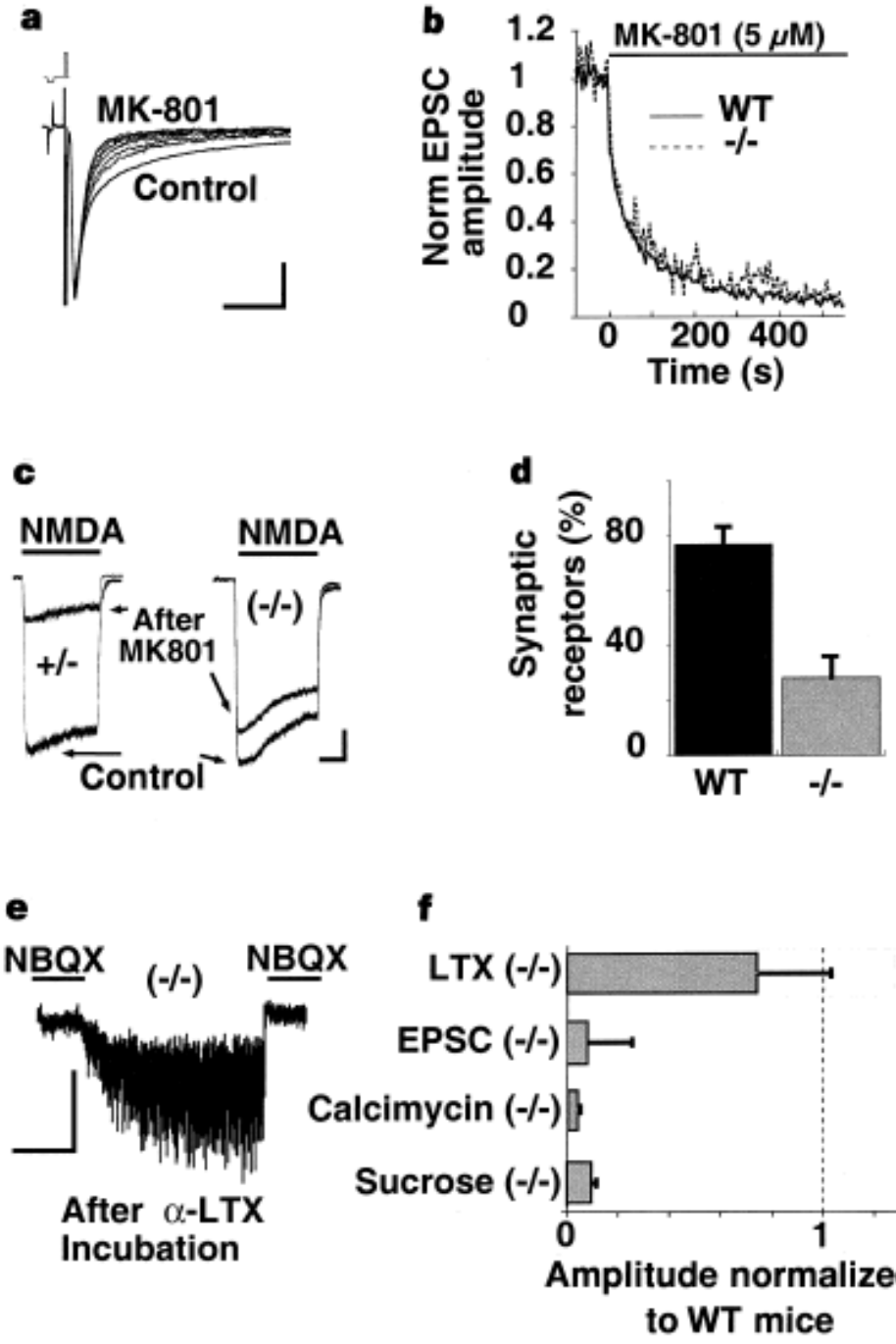
Munc13-1 is essential for fusion competence of glutamatergic synaptic vesicles

Iris Augustin*, Christian Rosenmund†, Thomas C. Südhof‡ & Nils Brose*


a
EPSC +/-
EPSC -/-
b
IPSC +/-
IPSC -/-
c
EPSC
IPSC
Synaptic current (nA)
2
1
0
+/+
+/-
-/-
+/+
+/-
-/-
Genotype


d
Calcimycin
+/-
-/-
e
Amplitude (nA)
0.4
0.2
0
WT
-/-
f
WT
-/-
0
1
2
3
GAD-negative synapse density
(per 10 μM)


a
EPSC +/-
Sucrose
EPSC -/-
Sucrose
b
Number of vesicles in pool (x1000)
3
2
1
0
EPSC WT
EPSC -/-
c
mEPSC rate (Hz)
6
4
2
0
WT
-/-


a
MK-801
Control
b
MK-801 (5 μM)
WT
-/-
Norm EPSC amplitude
1.2
1
0.8
0.6
0.4
0.2
0
0
200
400
Time (s)
c
NMDA
NMDA
+/-
After MK801
(-/-)
Control
d
Synaptic receptors (%)
80
40
0
WT
-/-
e
NBQX
(-/-)
NBQX
After α-LTX Incubation
f
LTX (-/-)
EPSC (-/-)
Calcimycin (-/-)
Sucrose (-/-)
0
1
Amplitude normalized to WT mice

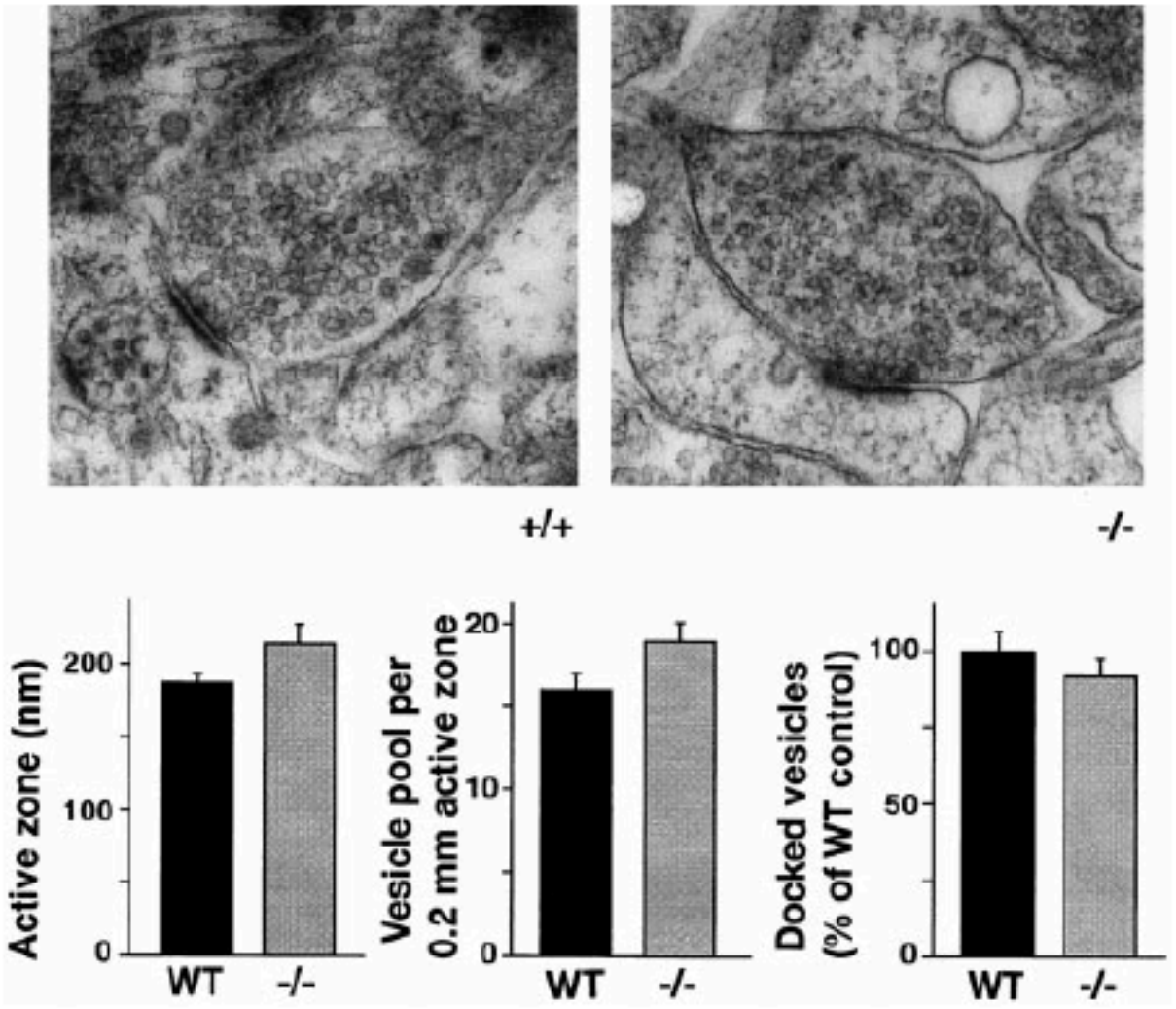


Figure 4 Munc13-1-deficient neurons form ultrastructurally normal synapses. Hippocampal neurons were analysed after 11 days in culture (122 synapses from four wild-type animals, and 54 synapses from two knockout animals).