Frog compound action potential

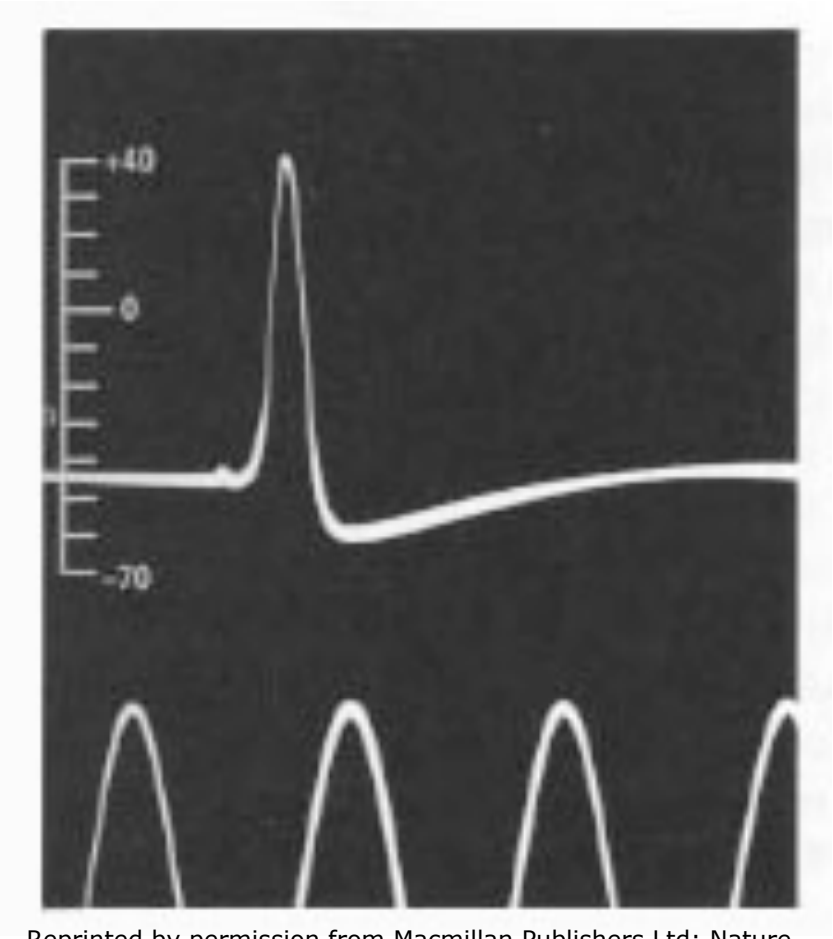
Prof. James DiCarlo

Course 9.17: Brain Laboratory, Brain and Cognitive Sciences

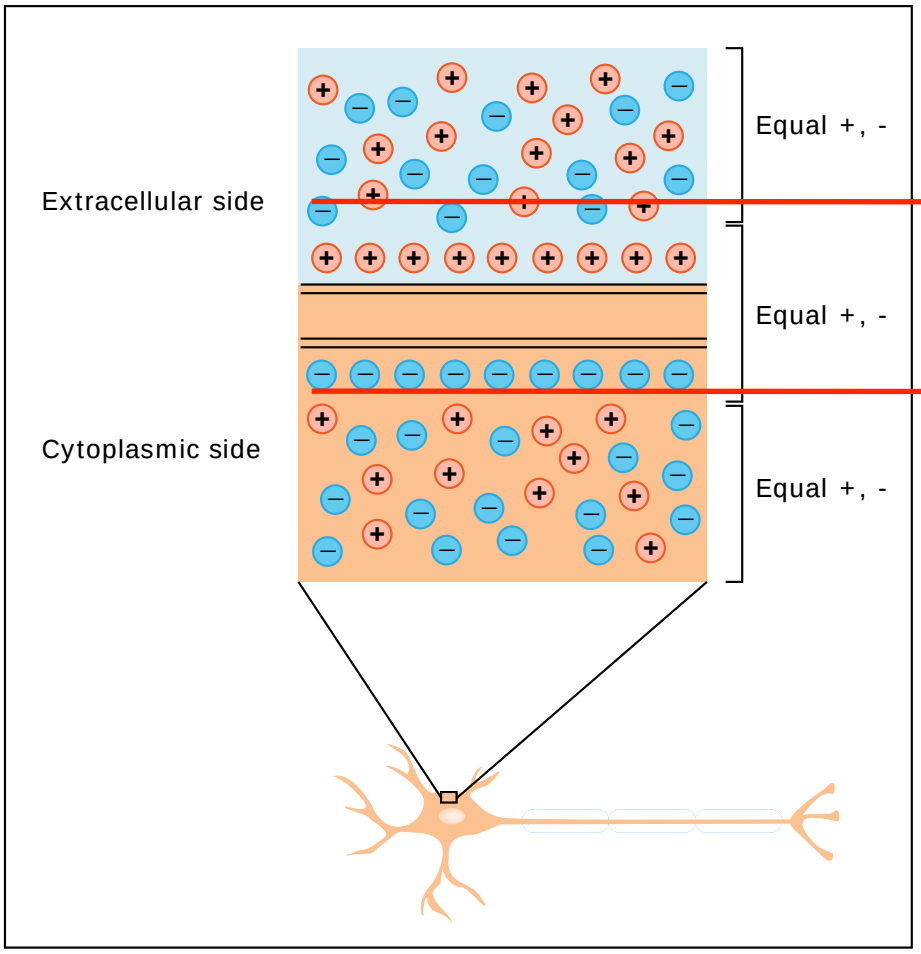
The action potential

Why should we care?

- Nervous system communication
- Time course (~1 ms) and propagation velocity (1-100 m/s) constrain hypotheses on how the brain works
- Understand what we are recording in neurophysiology experiments
- Teach us how we might interact with the nervous system



What “signals” can we measure?



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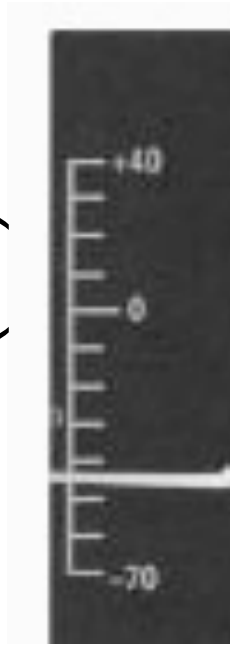
Membrane potential (Vm)



Time ->

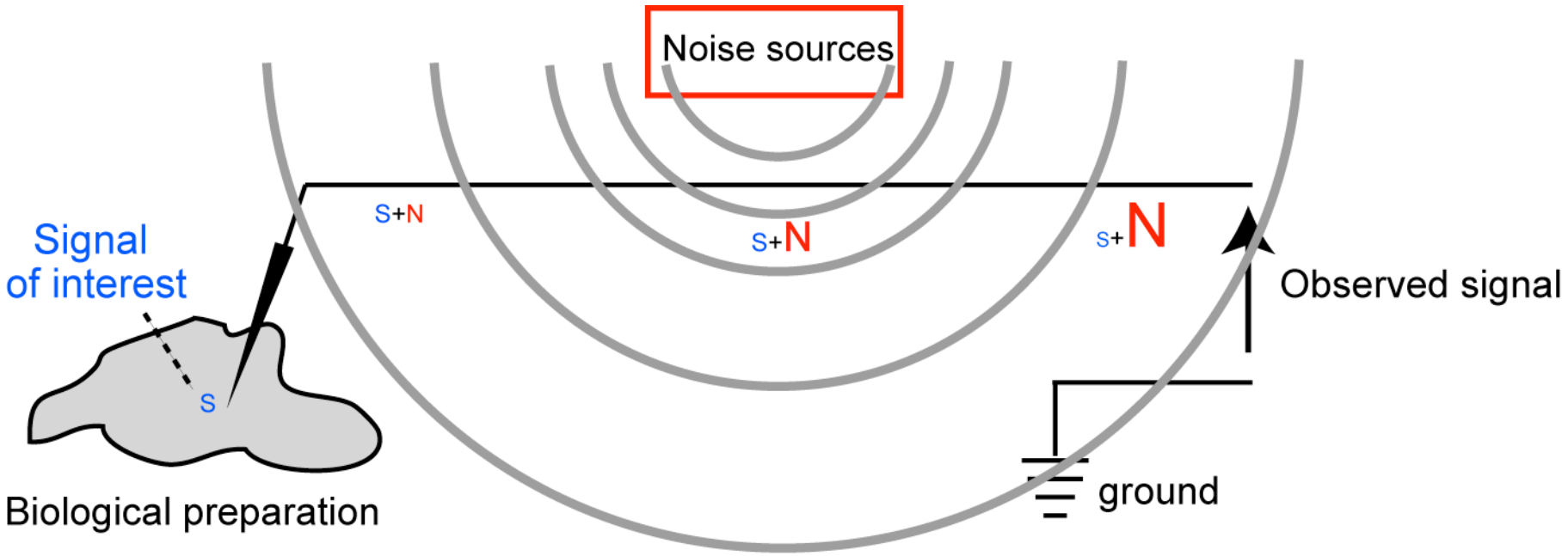
These signals are small

The Action Potential: from inside and out

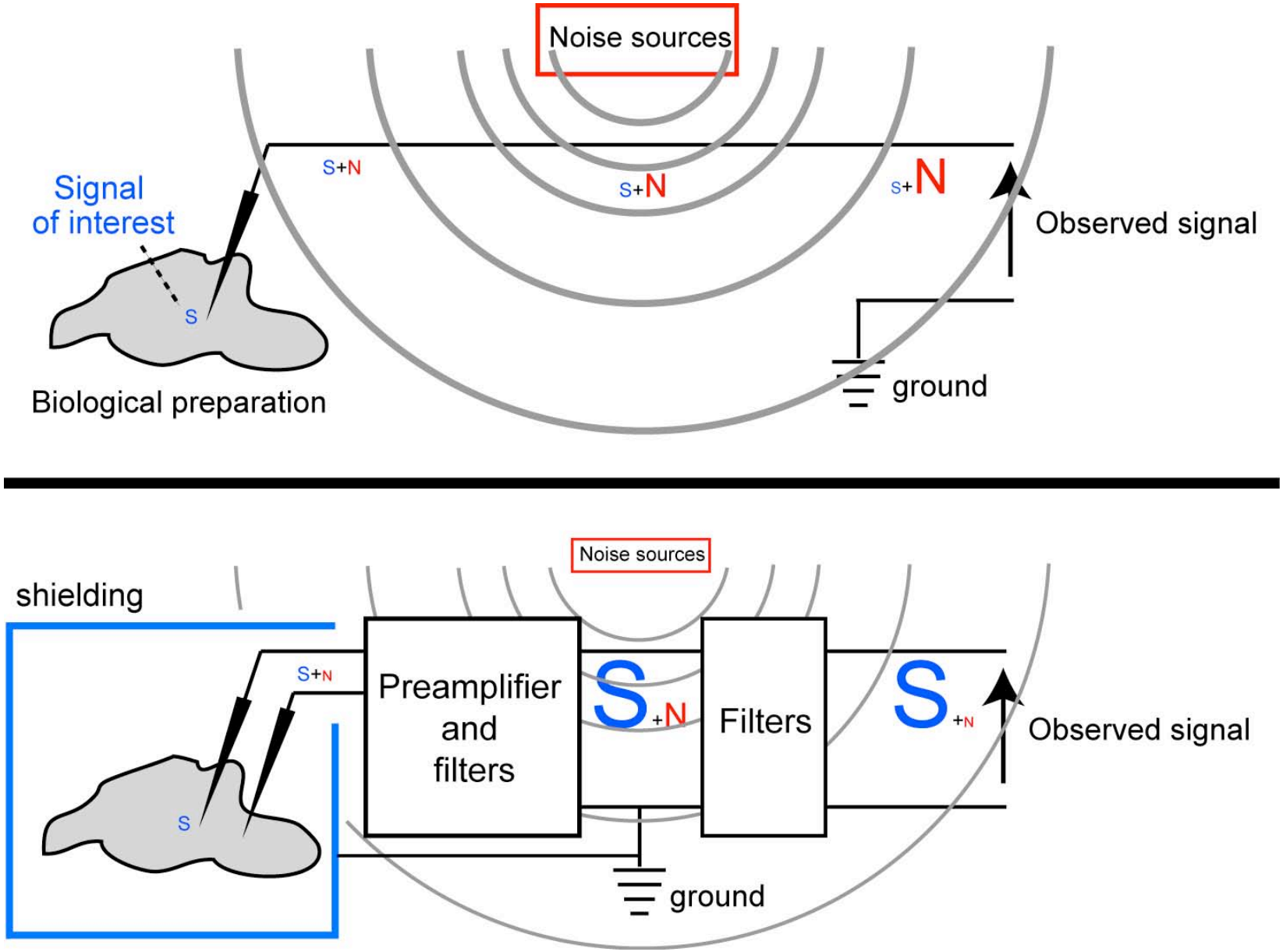
Fig. 1. Simultaneous intracellular and extracellular recording from a CA1 pyramidal cell removed due to copyright restrictions. See Henze, Darrell A., Zsolt Borhegyi, et al. "Intracellular Features Predicted by Extracellular Recordings in the Hippocampus In Vivo." J*ournal of Neurophysiology* 84, no. 1 (2000): 390-400.

Goal: Measure a very small signal (voltage) as a function of time.

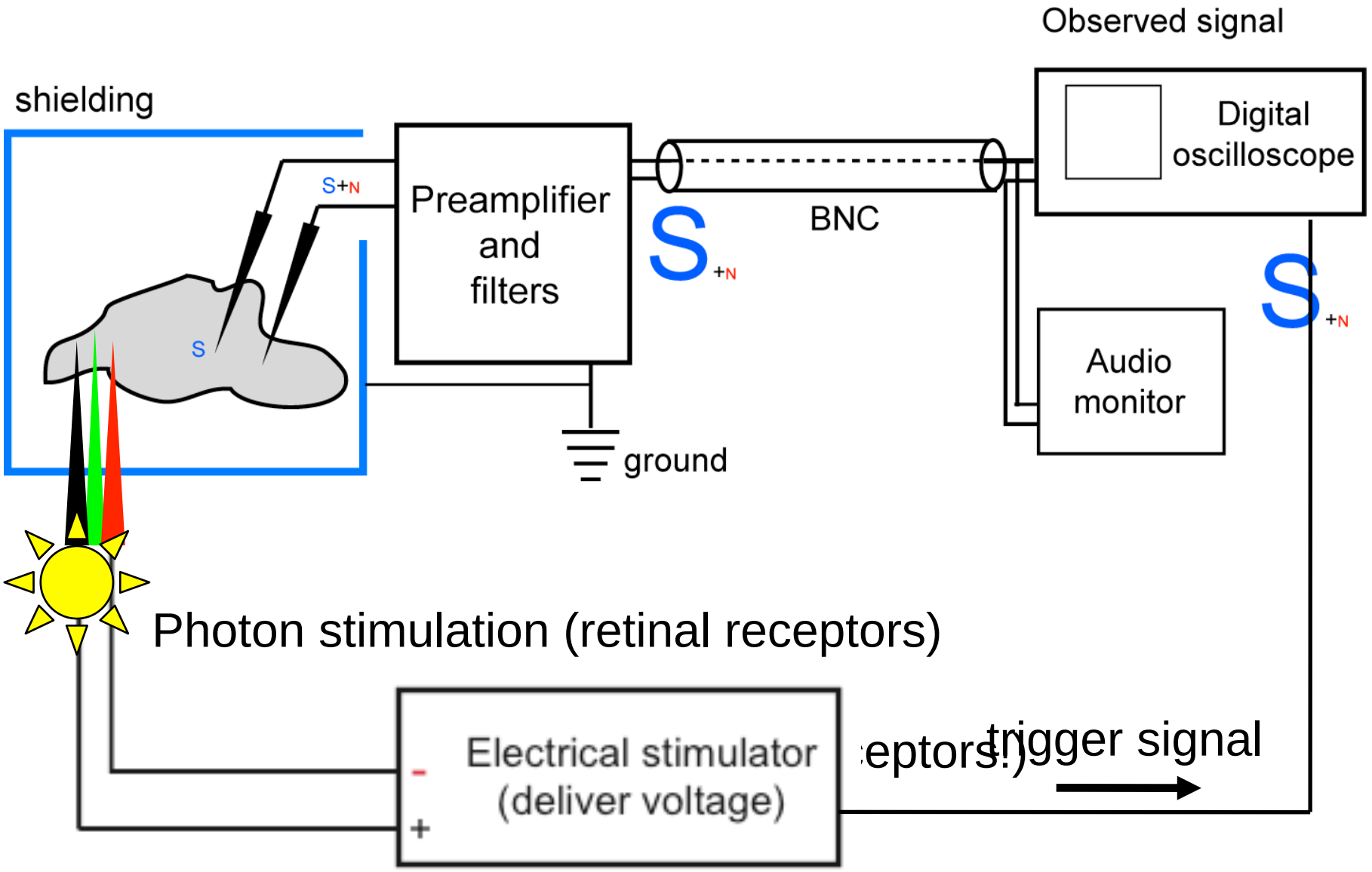
Problem: How do we “see” such a small signal in the presence of inevitable noise ?



Amplifier and filters



Basic electrophysiological setup



Frog lab: The Action Potential

Prof. James DiCarlo

Course 9.17: Brain Laboratory, Brain and Cognitive Sciences

Frog lab: Lecture overview

What I expect you to know before lab

1. What is a compound action potential? (vs. a “regular” action potential)

2. What are the ion channel types, mechanisms, and timings that underlie an action potential ? (REVIEW -- see Kolb article if you need a refresher.)

3. What is conduction velocity? Why do we care about conduction velocity? What axon properties affect it?

4. How are you going to setup your frog nerve and measure conduction velocity? (Lab notebook)

Sciatic nerve of the Bullfrog

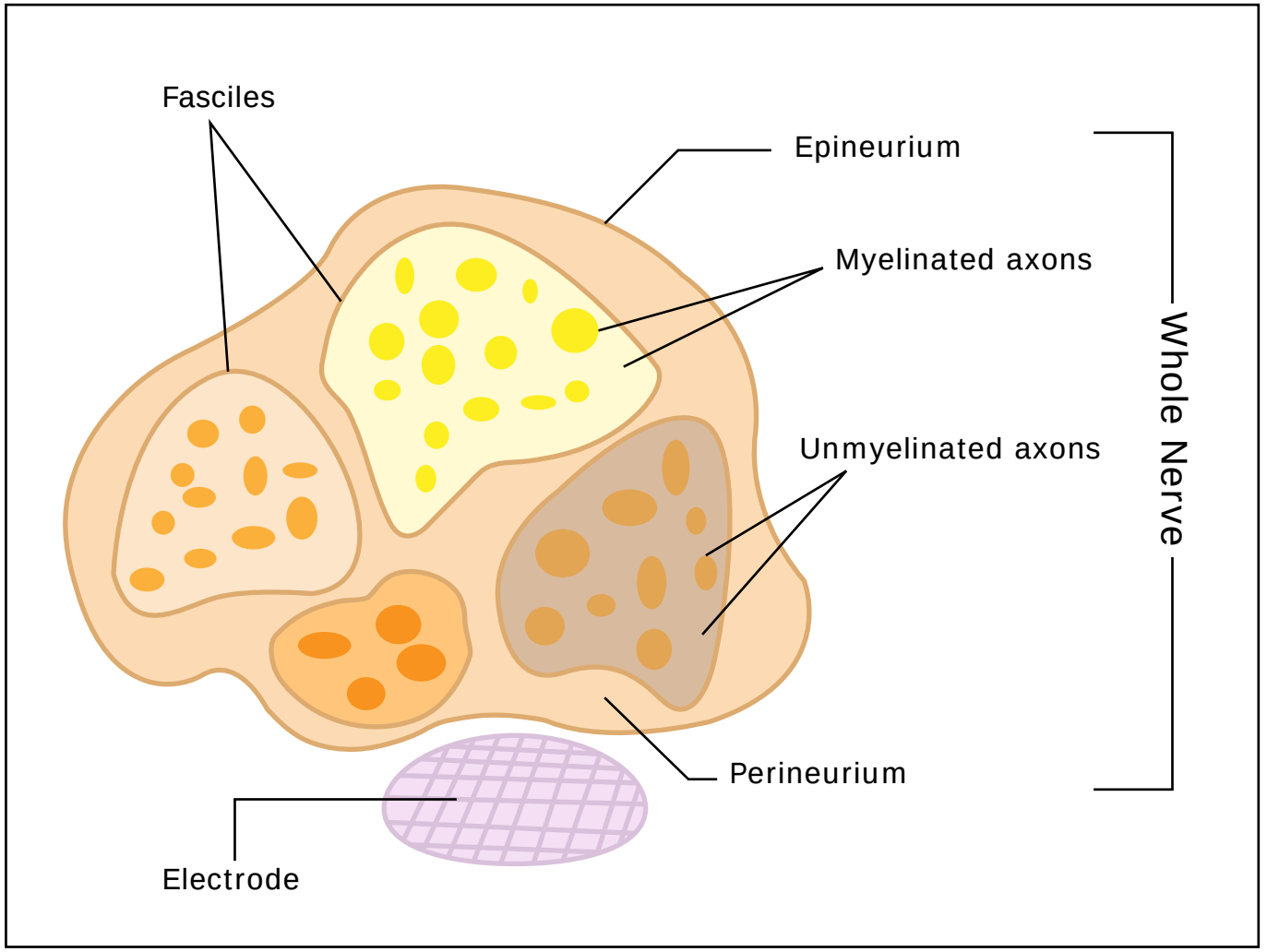
Sensory and motor signals

Illustration of dissecting out the frog sciatic nerve removed due to copyright restrictions.
See: http://www.medicine.mcgill.ca/physio/vlab/cap/prep.htm.

Review lab handbook on how to do the dissection

Figure 4.2-2 Recording arrangement removed due to copyright restrictions. See Oakley, Bruce, and Rollie Schafer. "Compound Action Potential." Chapter 4.2 in *Experimental Neurobiology: a Laboratory Manual*. University of Michigan Press, 1978, pp. 87.

The **compound action potential** is the combined* response resulting from many* individual action potentials



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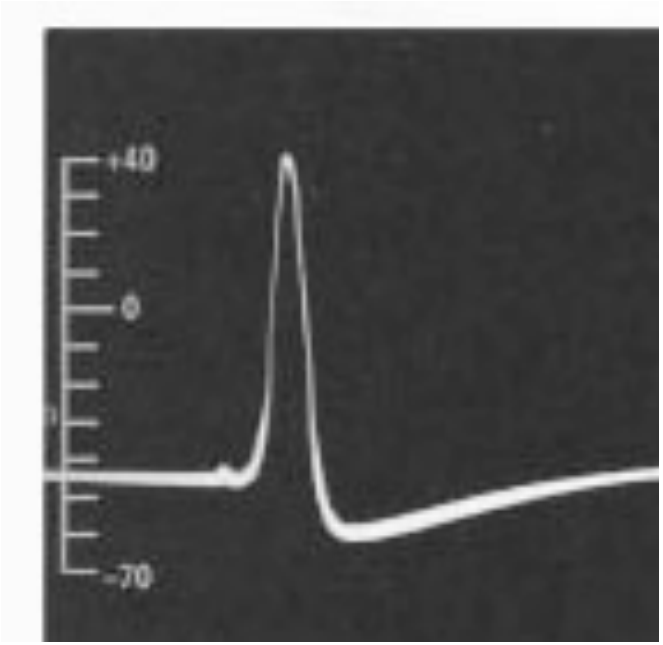
Textbook action potential description

?

Compound action potential observations



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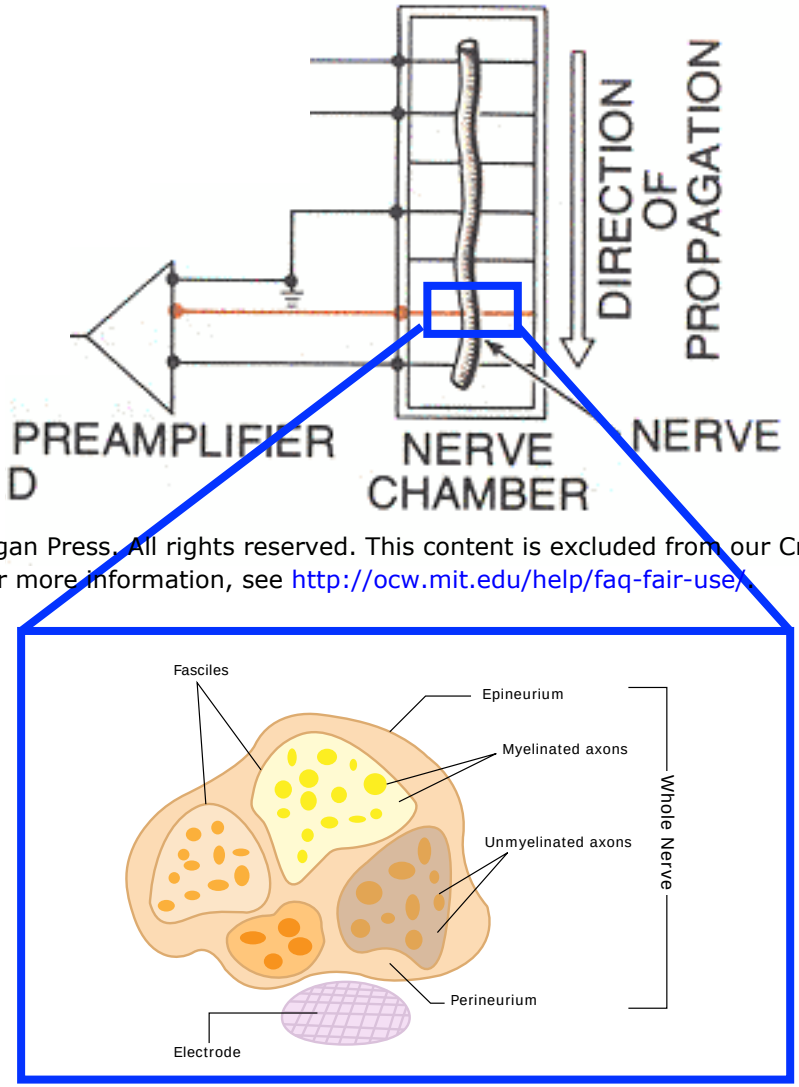
Source: Hodgkin, A. L., and A. F. Huxley. "Action Potentials Recorded from Inside a Nerve Fibre." *Nature* 144, (1946) 710-1. © 1946.



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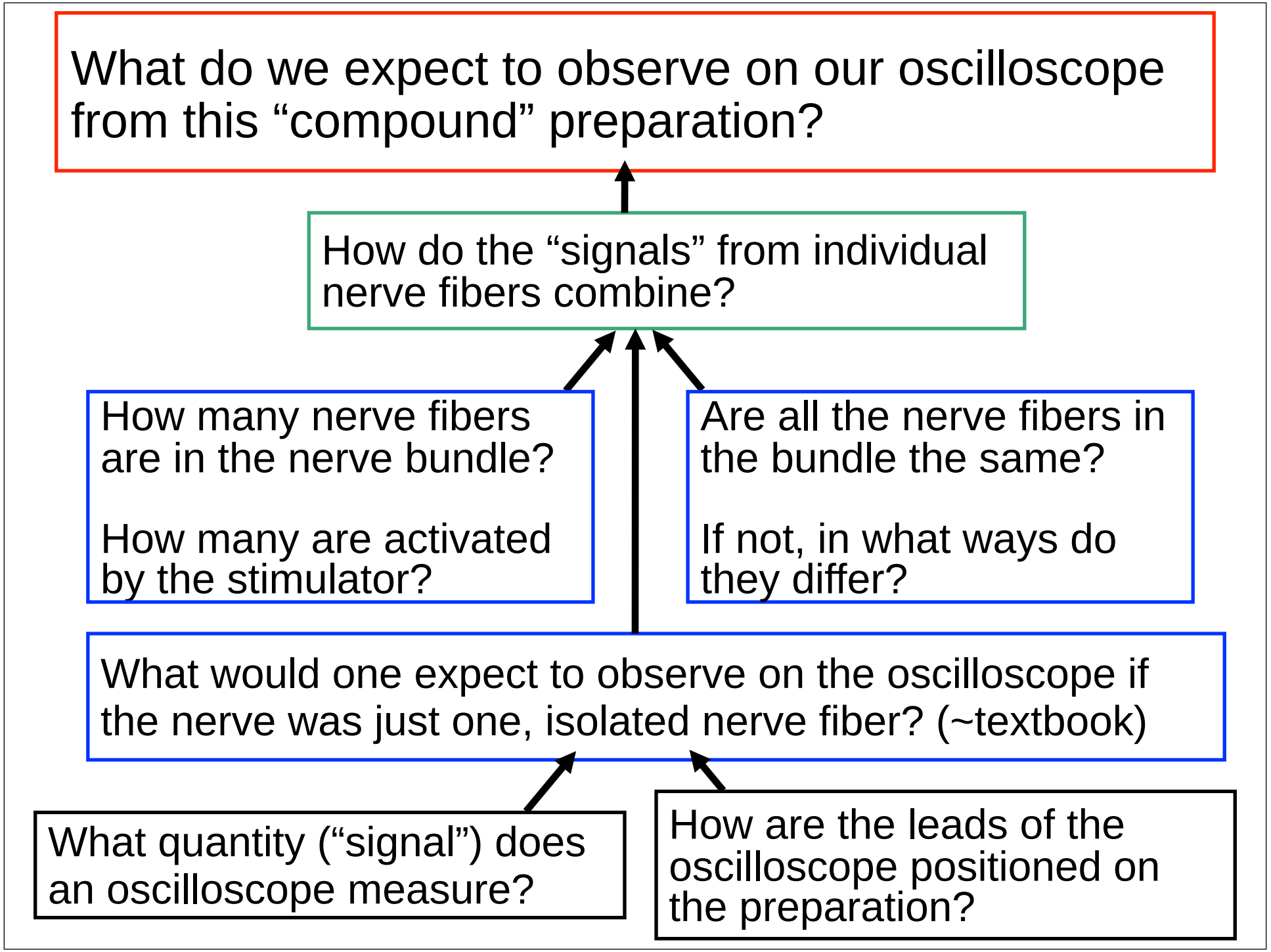
What do we expect to observe on our oscilloscope from this “compound” preparation?
How do the “signals” from individual nerve fibers combine?
How many nerve fibers are in the nerve bundle?
How many are activated by the stimulator?
Are all the nerve fibers in the bundle the same?
If not, in what ways do they differ?
What would one expect to observe on the oscilloscope if the nerve was just one, isolated nerve fiber? (~textbook)
What quantity (“signal”) does an oscilloscope measure?
How are the leads of the oscilloscope positioned on the preparation?

An action potential is a traveling wave

(How fast does it travel?)

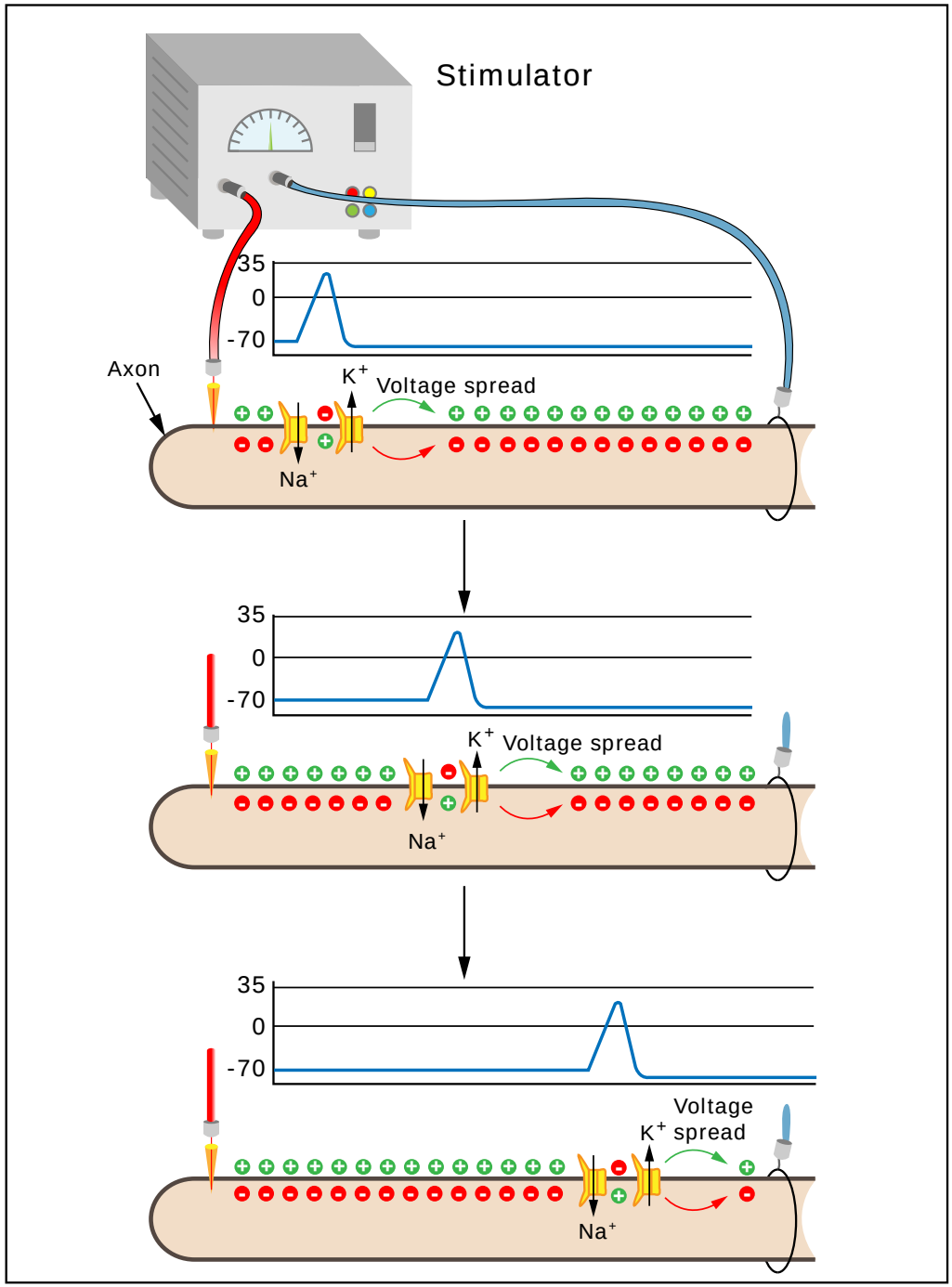

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Figure 2.19B Channel Openings and Local Circuits removed due to copyright restrictions. See Hille, Bertil. "Classical Biophysics of the Squid Giant Axon" Chapter 2 in *Ionic Channels of Excitable Membranes*. Sinauer Associates, Inc., 2001.

The Action Potential: from inside and out

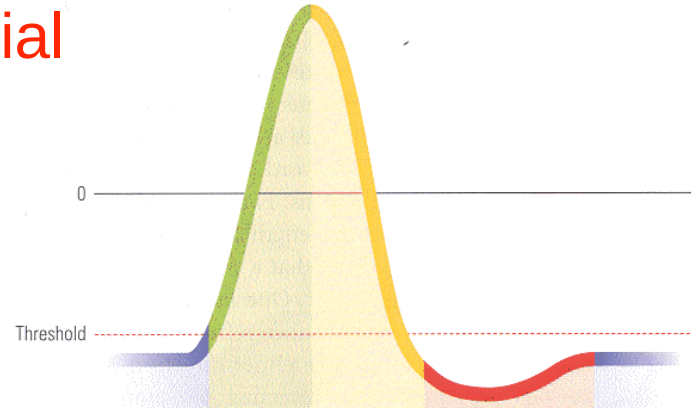
Fig. 1. Simultaneous intracellular and extracellular recording from a CA1 pyramidal cell removed due to copyright restrictions. See Henze, Darrell A., Zsolt Borhegyi, et al. "Intracellular Features Predicted by Extracellular Recordings in the Hippocampus In Vivo." J*ournal of Neurophysiology* 84, no. 1 (2000): 390-400.

All of the neuronal signals recorded in 9.02 Brain lab are recorded from OUTSIDE the cell (or axon)

1) The magnitude (i.e. voltage) of the recorded action potential signals will typically be much less than the magnitude of the INTRACELLULAR changes in membrane potential that occur with an action potential.
2) The polarity of the recorded signals will typically be opposite of the intracellular polarity.
3) The temporal shape of the recorded signals ("voltage waveform") will typically be similar in duration, but will differ from the shape of the intracellular membrane potential.

The CAP will look biphasic, but not for the same reason that the the membrane voltage for a single action potential looks biphasic.

Intracellular membrane potential



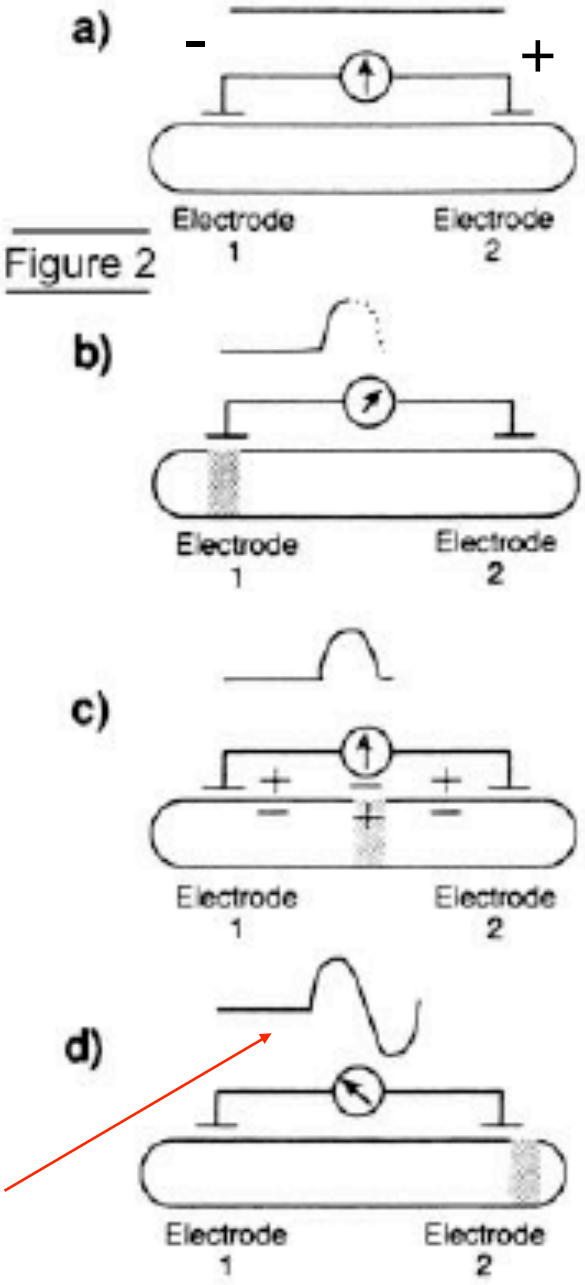
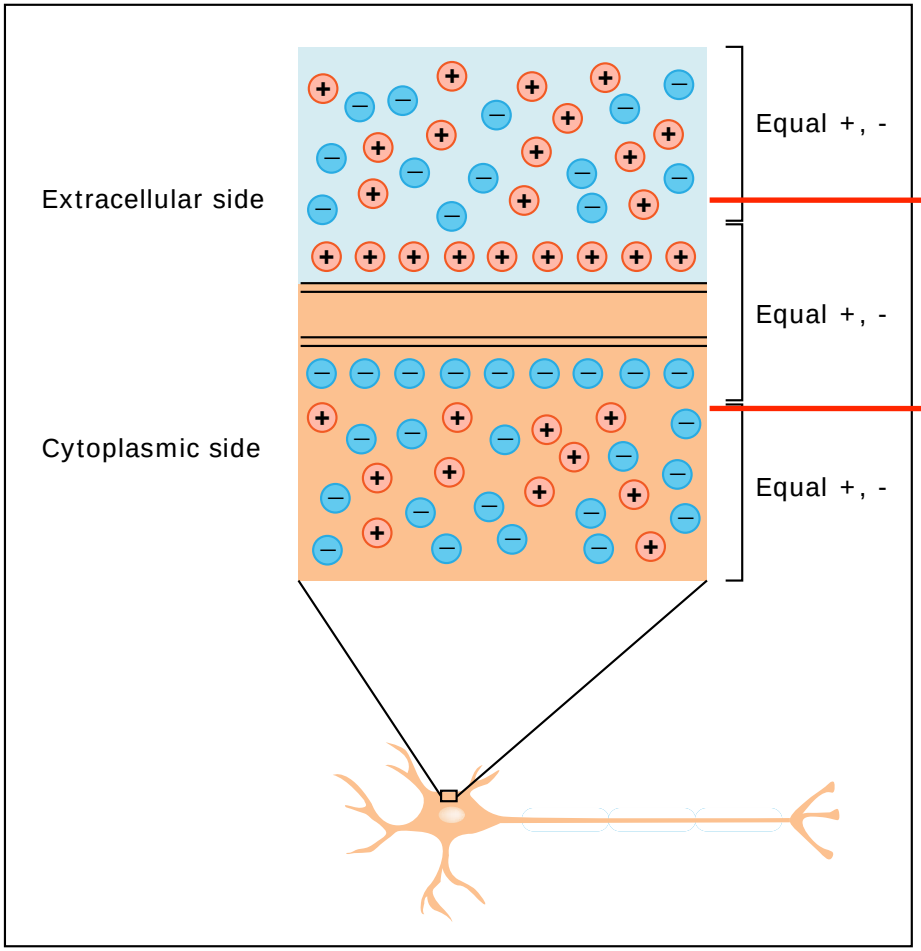


Figure 2

Extracellular CAP signal

- **Fundamentals of the action potential**
  - **Resting potential**
  - **Threshold**
  - **Refractory period**
  - **Conduction velocity**



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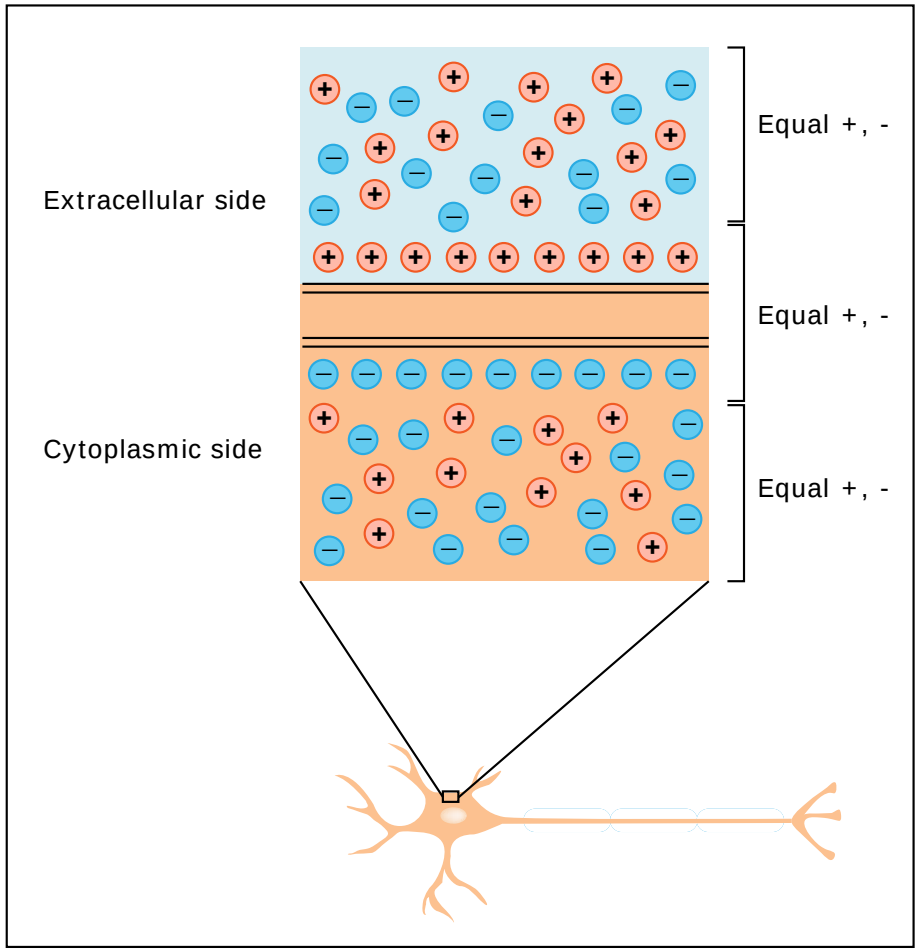
Membrane potential:

Due to a separation of positive and negative charges across the membrane.

Convention: Potential is measured as in relative to out.

$$V_m = V_{in} - V_{out}$$

"Resting" membrane potential ~ -60mV



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High [Na+], High [Cl-]

High [K+], High [A-]

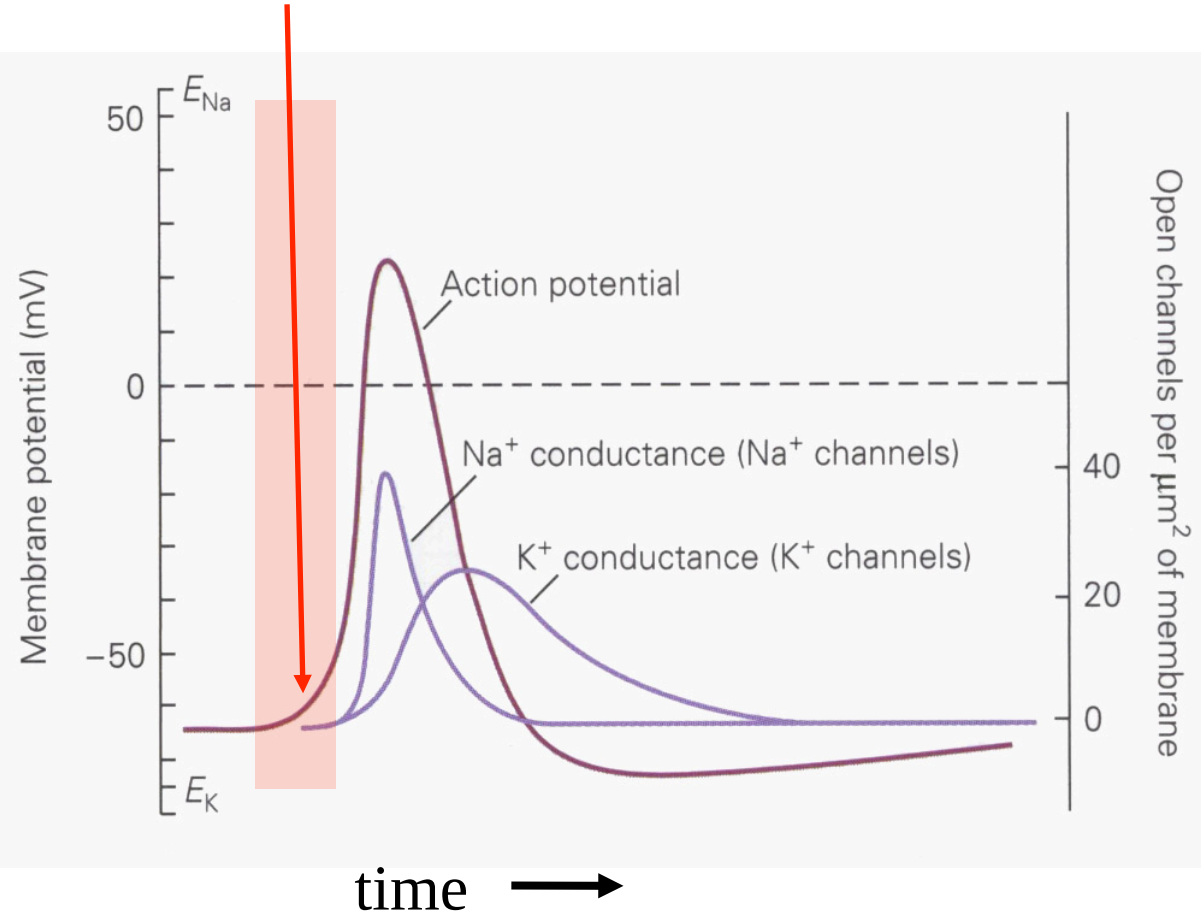
Concentration gradients:

Concentrations of ionic species are not equal on both sides of the membrane.

“Salt water outside”

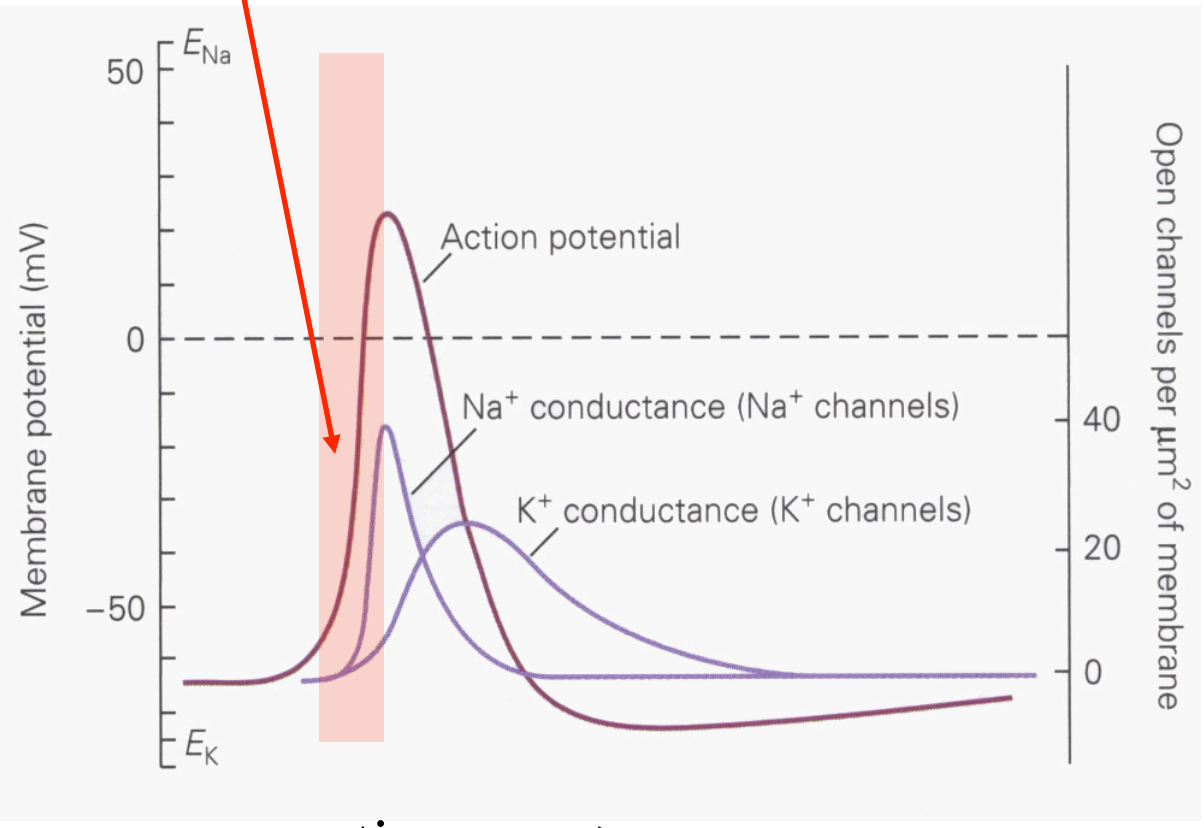
Getting things started…

An action potential is triggered by an increase in membrane potential (Vm)



Fundamental functional property of an action potential: **Threshold --> all or none (binary)**

The rising phase of the action potential is due to a rapid increase in Na+ conductance



Voltage-gated Na+ channels open more readily when the membrane potential increases (depolarization).

→Na+ flows in

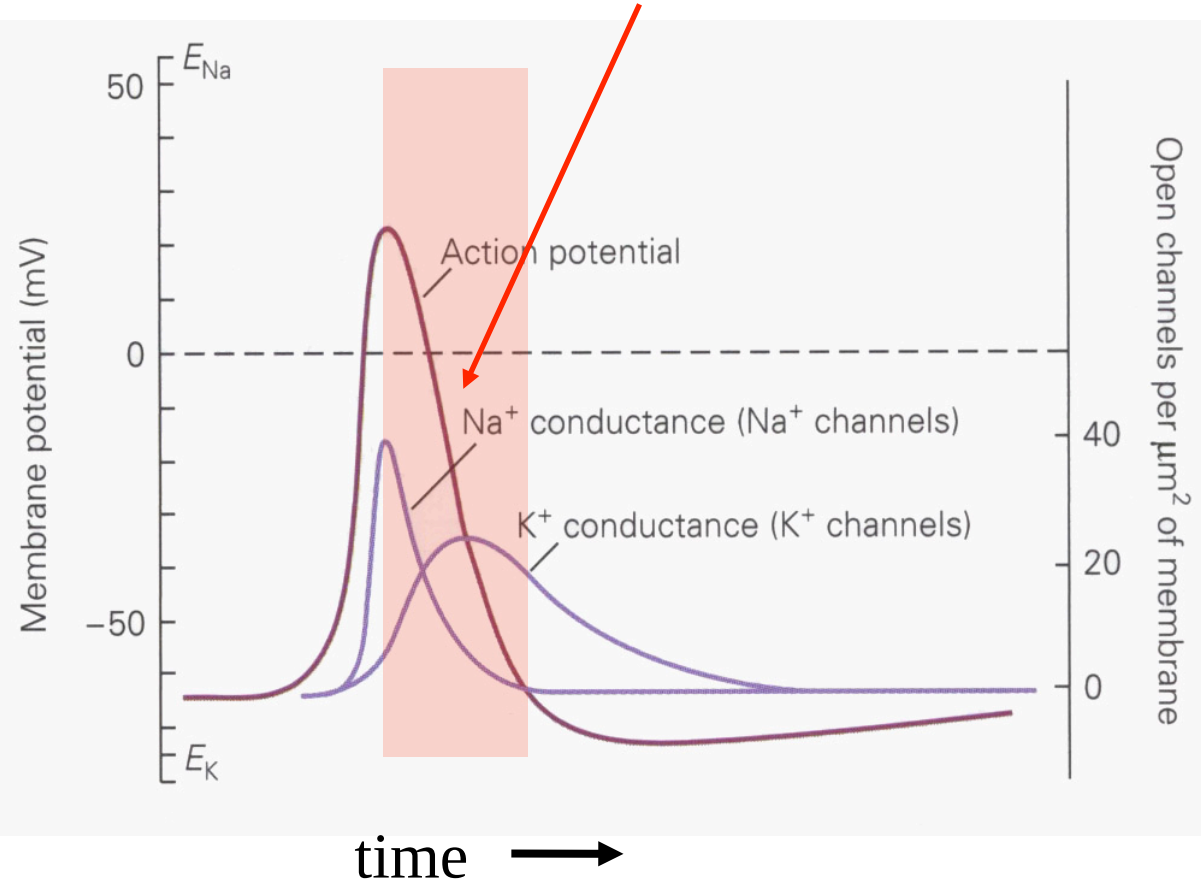
→ membrane potential increases (toward E_{Na})

→ voltage-gated Na+ channels open …

Concept: **threshold results from positive feedback on voltage gated Na+ channels**

Fundamental functional property of an action potential: **Short duration** (<1 ms)

The falling phase of the action potential is due to a decrease in Na+ conductance and an increase in K+ conductance



Voltage-gated Na+ channels close shortly after opening

→ less Na+ flows in

Voltage-gated K+ channels open after a delay

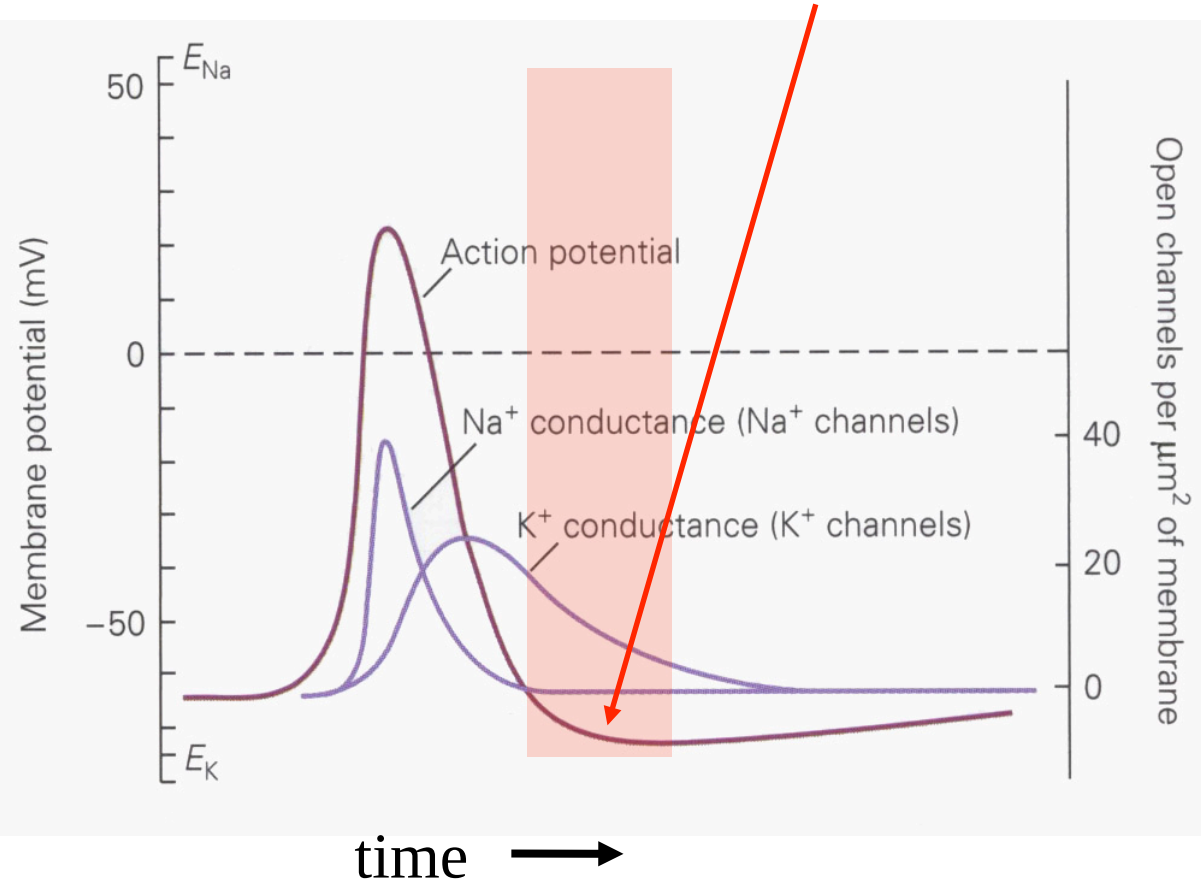
→ K+ flows out

→ membrane potential rapidly decreases (moves toward E_K)

Concept: **short duration**

Fundamental functional property of an action potential: **Refractory period**

The hyperpolarization phase of the action potential is due to a continued increase in K+ conductance



Voltage-gated K+ channels do not close immediately

→ K+ continue to flow out (at a lower rate)

→ membrane potential continues to <u>decrease</u> (moves toward E_K)

Many Na+ channels are now inactivated.

Concept: **refractory period**

Take home intuitions

The membrane potential is determined by who is winning the conductance ‘war.’

- Fundamentals of the action potential
  - Resting potential
  - Threshold
  - Refractory period
  - **Conduction velocity**

Conduction velocity of action potentials:

Determines the how fast information can be communicated from one part of the nervous system to another.

Can you think of situations where you want this to be very fast? Can you think of situations where you want the information to travel slowly?

Ballpark guess at conduction velocity?

Time from toe to spinal cord?

So… fast is good!

Why not make all action potentials travel as fast as possible?

Conduction velocity of action potentials:

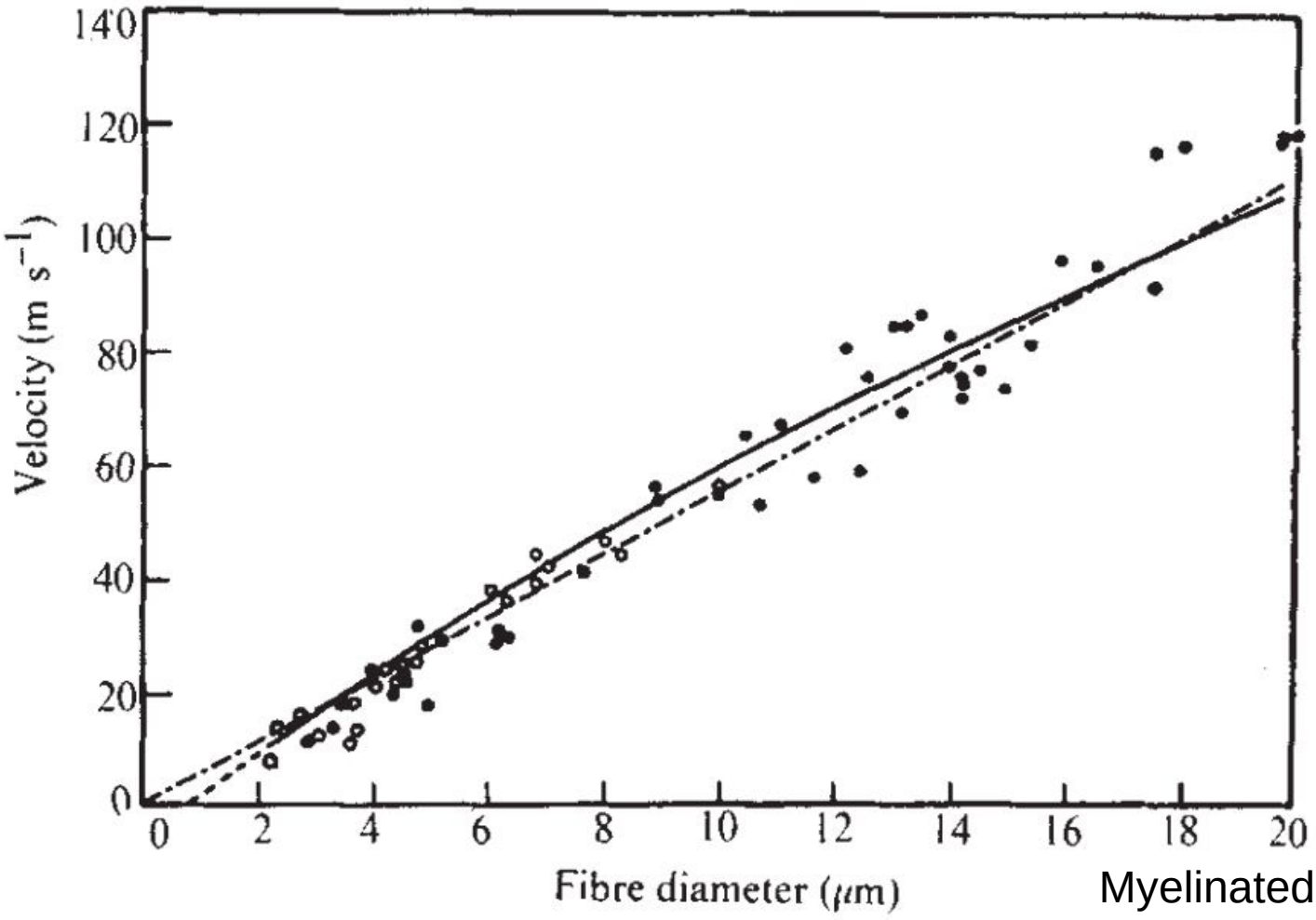
How do we build an axon so that an action potential travels fast? (I.e. Which axon properties determine the conduction velocity?)

- Axon diameter
- Membrane capacitance

Conduction velocity of action potentials:

- Axon diameter

(bigger diameter --> faster conduction velocity)



Myelinated fibers

Conduction velocity of action potentials:

Membrane capacitance

(smaller capacitance --> faster cond velocity)
("thicker" membrane --> smaller capacitance)
(THUS: "thicker" membrane --> faster cond vel)

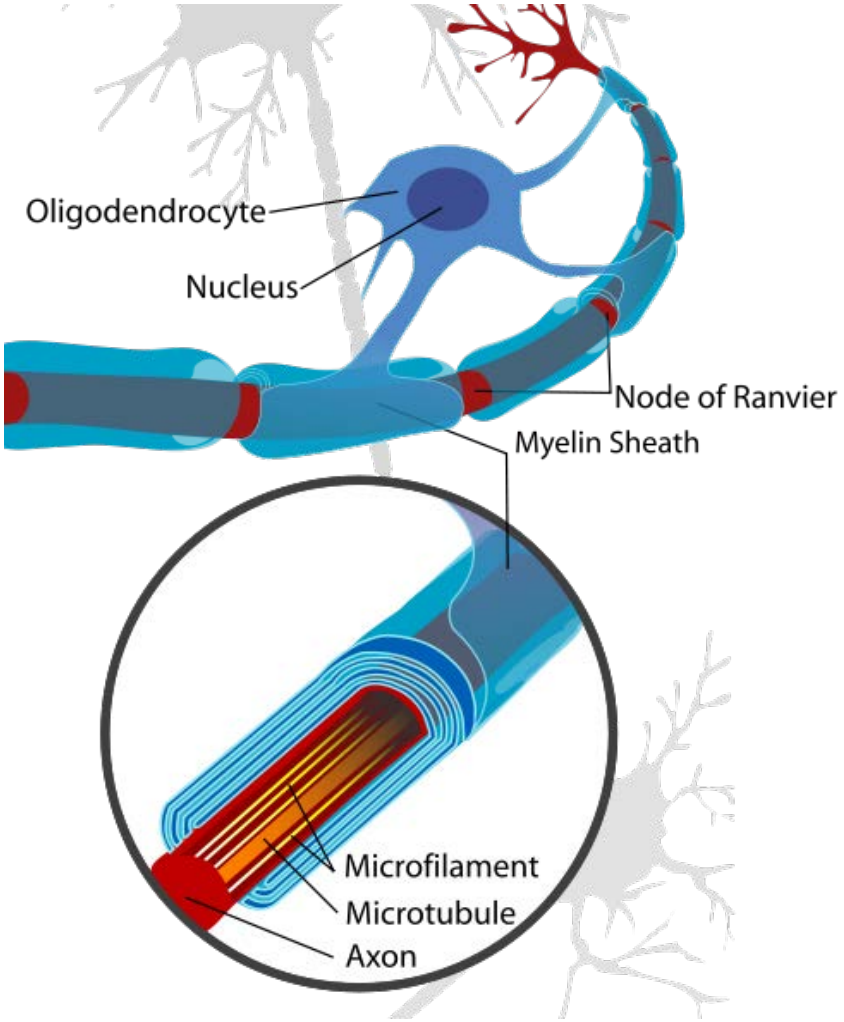
Capacitance is the "capacity" to store charge

The nervous system's way to decrease capacitance: myelin

Conduction Velocity:

An elegant solution: myelin sheaths:

- Decrease membrane capacitance --> faster conduction velocity
- unmyelinated sections (nodes of Ranvier) allow Na channels to strengthen the action potential



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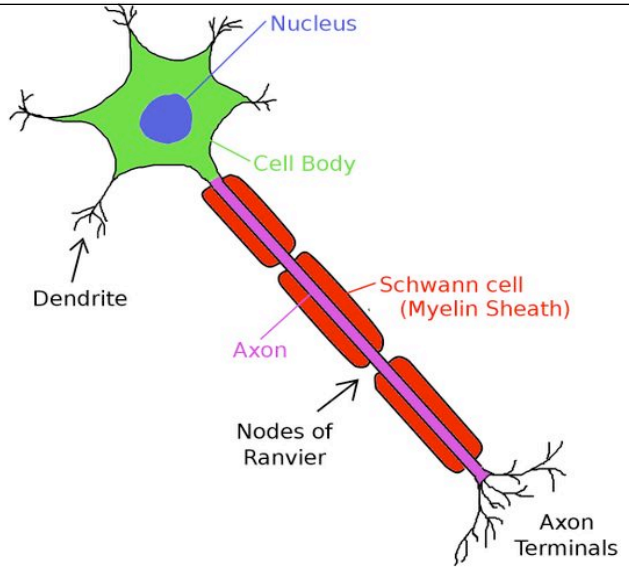
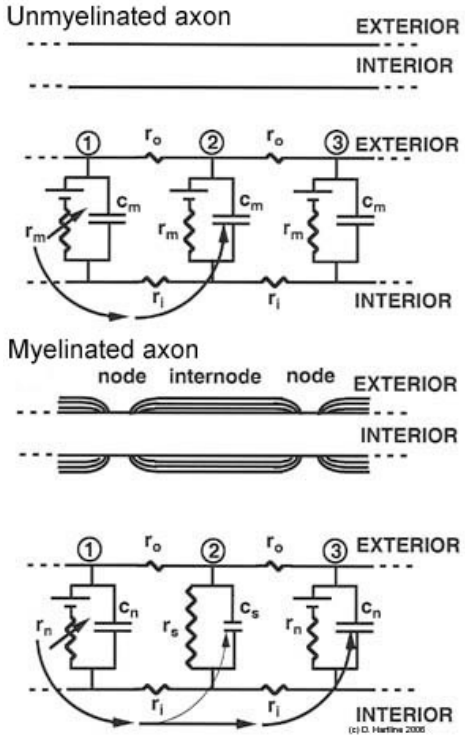
Myelin to increase conduction velocity





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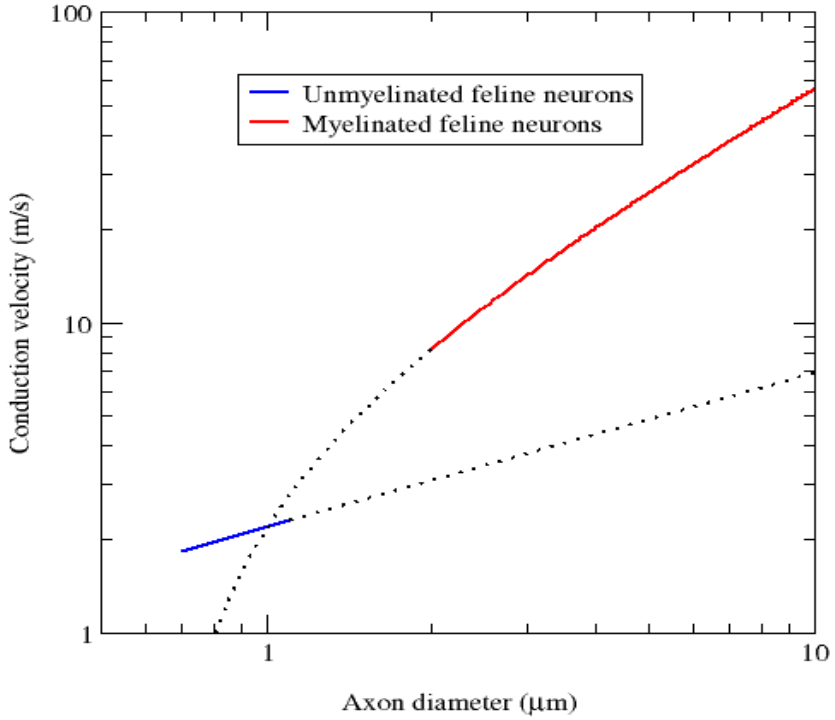

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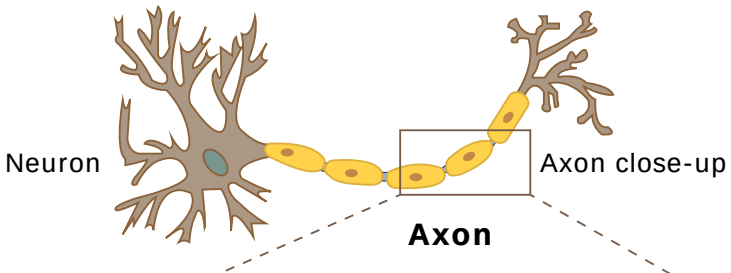
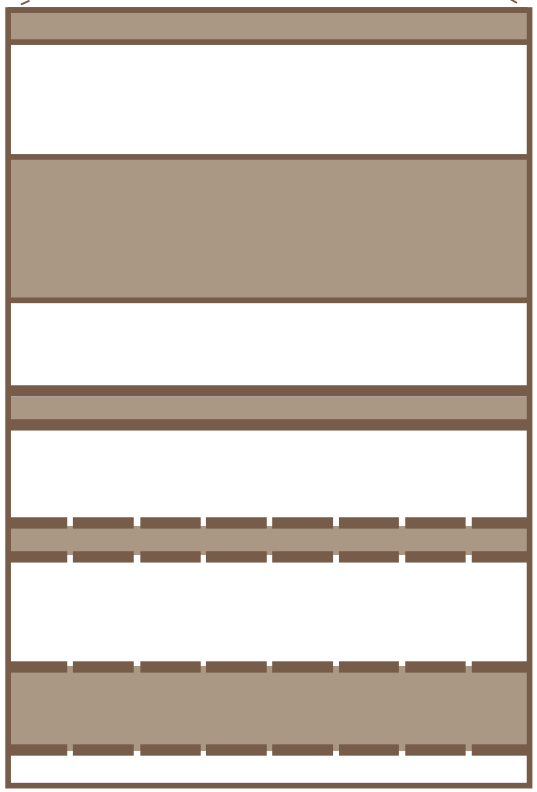
Design trade-offs!





Pro	Con
Faster (lower axial R)	Too big
Faster (lower membrane capacitance)	Transmission fails!
Less energy	
Faster	
Fastest!	Big more energy

Inside (Cytoplasm) Outside (extracellular space)

Image by MIT OpenCourseWare.

A compromise on axon diameter: use it where you most need it!

Mammalian Axon Properties

Fiber Types	Fiber Diameter (μm)	Conduction Velocity (m/sec)	Action Potential Duration (msec)	Absolute Refractory Period (msec)	Functions
Aα motoneurones	12-22	70-100	0.4-0.5	0.2-1.0	Efferent alpha Afferent muscle spindles, tendon organs
Aβ	5-13	30-70	0.4-0.5	0.2-1.0	Afferent, cutaneous, Touch, pressure
Aγ	3-8	15-40	0.4-0.7	0.2-1.0	Gamma motoneurons
Aδ	1-5	12-30	0.2-1.0	0.2-1.0	Afferent, fast Pain, temperature
B	1-3	3-15	1.2	1.2	Efferent, autonomic preganglionic
C (unmyelinated)	0.2-1.2	0.2-2.0	2	2	Afferent, “slow” Pain, Efferent Autonomic postganglionic

Note: amphibian Aα fibers at room temperature are slower than shown here.

This week's quiz:

- Components of an electrophysiological setup
- Layout of the frog nerve setup
- Action potential basics (review)
- Factors that affect conduction velocity
- Recitation (Thorpe et al.)

9.17 Systems Neuroscience Lab

Spring 2013