

Neuroscience 201A (2016)

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Outline for Today

- Course organization and logistics
- “Electricity 101”
- The Electrical Properties of Neurons
- The Determinants of Membrane Potential

Course organization

- 201A Co-Directors:
 - Peter Sargent
 - Jennifer Whistler
- Part of 200, 201A, 201B, 201C sequence
- Overall course director, Eric Huang
- Meets 3-4 mornings a week for lecture or discussion
- Course materials are (should be) on the web site

Schedule (2016)

Week	Date	Day	Room	Time	Topic	Leader
week 1	9/19/16	Mon			Neuroscience Asilomar Retreat	
	9/20/16	Tue			Neuroscience Asilomar Retreat	
	9/21/16	Wed			no class	
	9/22/16	Thu	GH-S261	9a-11a	Membrane Potential	Peter Sargent
	9/23/16	Fri	GH-S261	9a-11a	Action Potentials	Peter Sargent
week 2	9/26/16	Mon	GH-S261	9a-11a	Single Channels	Peter Sargent
	9/27/16	Tue	GH-S261	8a-11a	Biophysics Problem Set	Peter Sargent
	9/28/16	Wed			no class	
	9/29/16	Thu	GH-S261	9a-11a	Structure-Function 1	Yuriy Kirichok
	9/30/16	Fri	BH-209	9a-11a	Structure-Function 2	Yuriy Kirichok
week 3	10/3/16	Mon			Rosh Hashana	
	10/4/16	Tue			Rosh Hashana	
	10/5/16	Wed			no class	
	10/6/16	Thu	GH-S261	8a-11a	Potassium Channels/Paper Discussion	Lily Jan
	10/7/16	Fri	BH-209	9a-11a	Synaptic Transmission 1	Peter Sargent
week 4	10/10/16	Mon	GH-S261	9a-11a	Synaptic Transmission 2	Peter Sargent
	10/11/16	Tue			no class	
	10/12/16	Wed			Yom Kippur	
	10/13/16	Thur	GH-S261	8a-11a	Synaptic Transmission Problem Set & Paper Disc.	Peter Sargent
	10/14/16	Fri	BH-209	9a-11a	Integration 1	Kevin Bender
Exam #1: 45% → week 5	10/17/16	Mon	GH-S261	9a-12n	<u>exam 1, in class</u>	
	10/18/16	Tue	GH-S261	9a-11a	Integration 2	Kevin Bender
	10/19/16	Wed			no class	
	10/20/16	Thu	GH-S261	8a-11a	Integration Paper Discussion	Kevin Bender
	10/21/16	Fri	BH-209	9a-11a	Glutamate Receptors	Roger Nicoll
week 6	10/24/16	Mon	BH-261	9a-11a	Plasticity	Roger Nicoll
	10/25/16	Tue	GH-S261	8a-11a	Plasticity Paper Discussion	Roger Nicoll
	10/26/16	Wed			no class	
	10/27/16	Thu	GH-S261	9a-11a	Neurotransmitter Release	Rob Edwards
	10/28/16	Fri	GH-S261	9a-11a	Neurotransmitter Reuptake	Rob Edwards
week 7	10/31/16	Mon	GH-S261	8a-11a	Release/Reuptake Paper Discussion	Rob Edwards
	11/1/16	Tue	GH-S261	9a-11a	Receptor Pharmacology 1	Jennifer Whistler
	11/2/16	Wed			no class	
	11/3/16	Thu	GH-S261	9a-11a	Receptor Pharmacology 2	Jennifer Whistler
	11/4/16	Fri	BH-209	8a-11a	Receptor Pharmacology Paper Discussion	Jennifer Whistler
Exam #2: 55% → week 8	11/4/16	Fri	BH-209	11a-12n	In class review	
	11/7/16	Mon	GH-S261	9a-12n	<u>exam 2, in class</u>	
	11/8/16	Tue		9a-11a		
	11/9/16	Wed		9a-11a		
	11/10/16	Thu		9a-11a		
	11/11/16	Fri		9a-11a	Veteran's Day, no class	

Assessment

- Two exams: October 17, November 7
 - First exam: in class. 3 hours. Open book, but no web access.
 - Second exam: in class *or* take-home. Class decides.
- The course is letter-graded (A, B, C, etc.)
- You must get a B or better to pass the course (program specific)
- Historically, the course has been graded “B-modal”

Resources

- Texts – on reserve (Rutter Center library)
 - Fain G (2014) *Molecular and Cellular Physiology of Neurons*, 2nd ed., Harvard University Press. (1-day reserve)
 - Hille B (1984) *Ion Channels of Excitable Membranes*, 2nd edition (current edition is the 3rd), Sinauer. (1-day reserve)
 - Johnston D, Wu SM (1995) *Foundations of Cellular Neurophysiology*. MIT Press. (1-day reserve)
 - Kandel ER et al. (2013) *Principles of Neural Science*, 4th edition (current edition is the 5th), McGraw-Hill. (1-day reserve)
 - Nicholls J et al. (2001) *From Neuron to Brain*, 2nd edition (current edition is the 5th), Sinauer. (1-day reserve)
 - Purves D et al. (2001) *Neuroscience*, 2nd edition (current edition is the 5th), Sinauer. (1-day reserve).
- Review Articles
- Me! 8 am – 9 am sessions (bring questions!)
 - Friday, September 23
 - Monday, September 26
 - Thursday, September 29
 - Friday, October 7
 - Monday, October 10
- Your peers.

I. Electricity 101

- Our expectations

What was covered in boot camp?

- Electricity Basics?
- Electrical properties of plasma membranes: capacitance and conductance?
- Neurons as passive conductors of electricity?
- Electrical equivalent of an excitable cell (from Hodgkin Huxley)?

Slides with Green Background are supplemental:

- Provide “Color”
- Provide background
- Provide expectations of familiarity (“you should know this”)

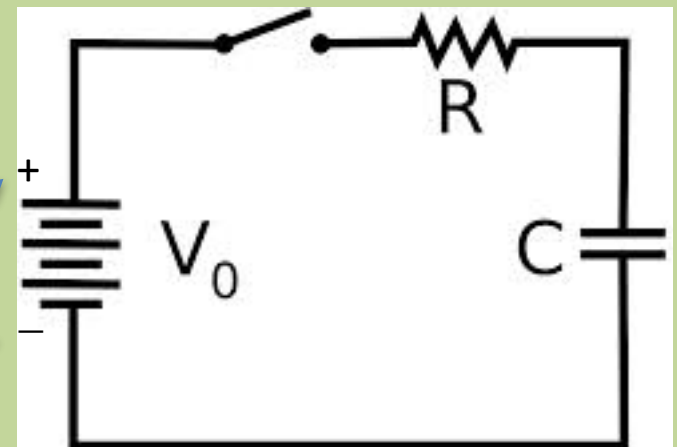
Units

- of voltage or potential (potential difference) V or E – volts (V)
- of charge Q – coulombs (C)
- of current I – amperes (A)
- of capacitance C – Farads (F)
- of resistance R – ohms (Ω)
- of conductance G – Siemens (S)

Symbols

Units

Cathode
Anode



From College Physics



- = a resistor or a conductor; $R=1/G$; $G=1/R$

- Voltage across a resistor: $V=IR$ (Ohm's law)

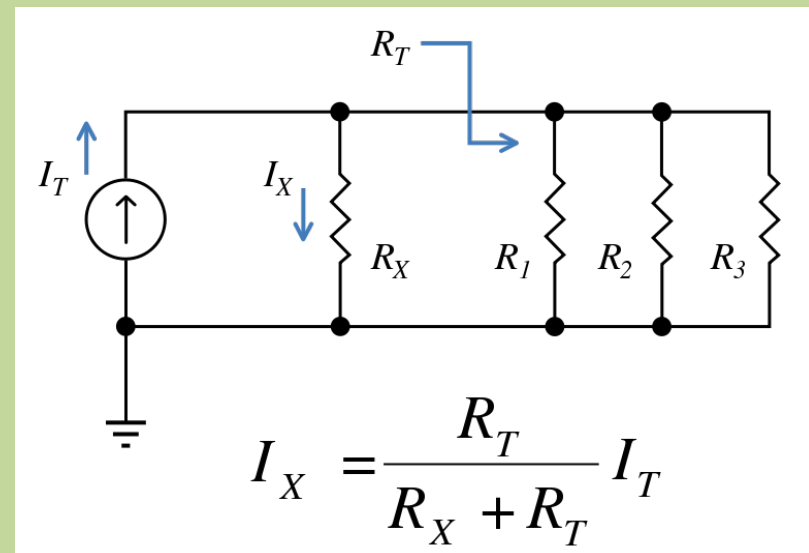
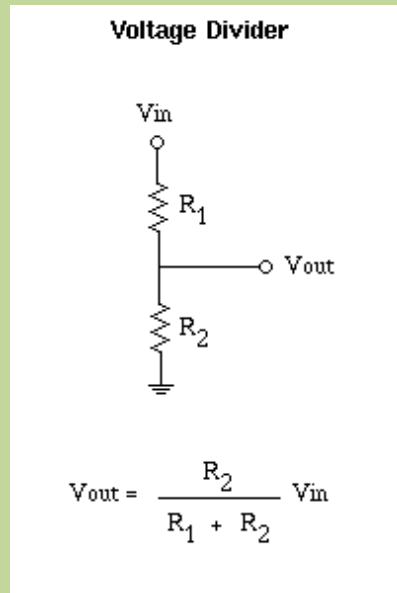


- If thinking of as a conductor: $I=GV$ (Ohm's law)

- Voltage across a capacitor: $V=Q/C$
- In series: resistors add, conductors add inversely, capacitors add inversely
- In parallel: resistors add inversely, conductors add, capacitors add

From college physics (cont.)

- Kirchoff's current law (KCL) – the sum of currents at a node is zero.
- Kirchoff's voltage law (KVL) – sum of potential differences around a closed network is zero.
- “Voltage divider”
- “Current divider”



Equivalencies in the SI system of units

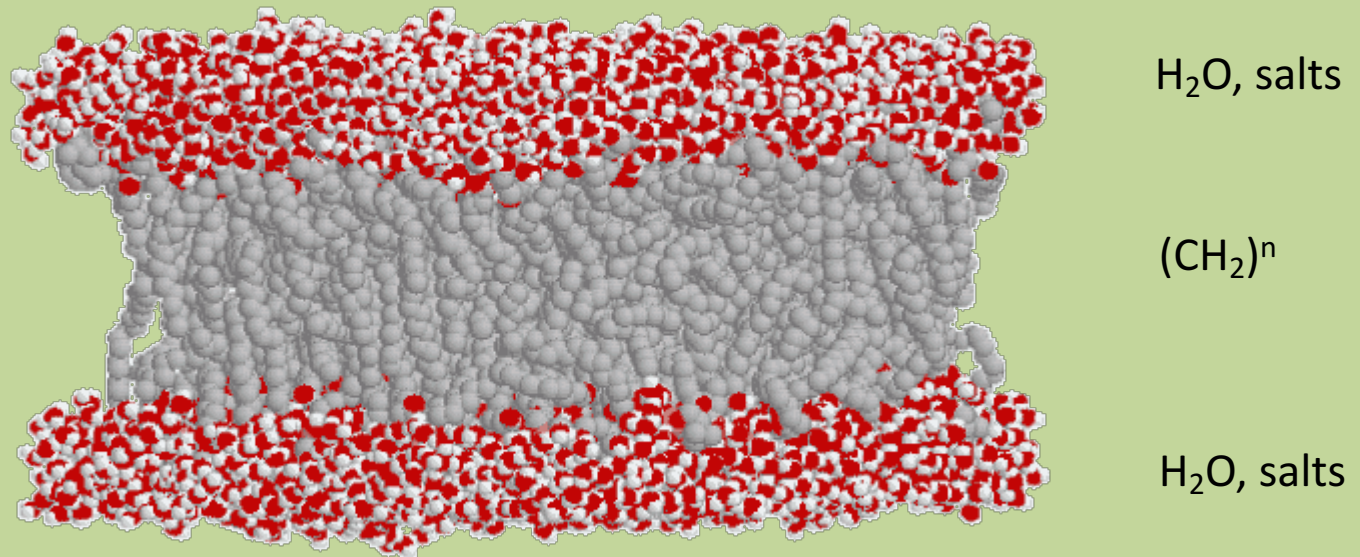
- Ampere (A) = 1 C/s
- Volt (V) = unit of electromotive force required to drive 1 A of current across 1 ohm of resistance.
- Farad (F) = 1C/1V
- Ohm (Ω) = 1V/1A
- Faraday (F) – charge per mole of unitary ions. = elementary charge x Avogadro's number
= $\sim 1.6 \times 10^{-19} \text{ C} \times \sim 6.0 \times 10^{23} = 9.6 \times 10^4 \text{ C}$.

II. Electrical Properties of Neurons

- Properties of biological membranes
- The consequences of capacitance on time-variant properties of membrane potential
- When cells are “isopotential”
- When cells are not “isopotential”

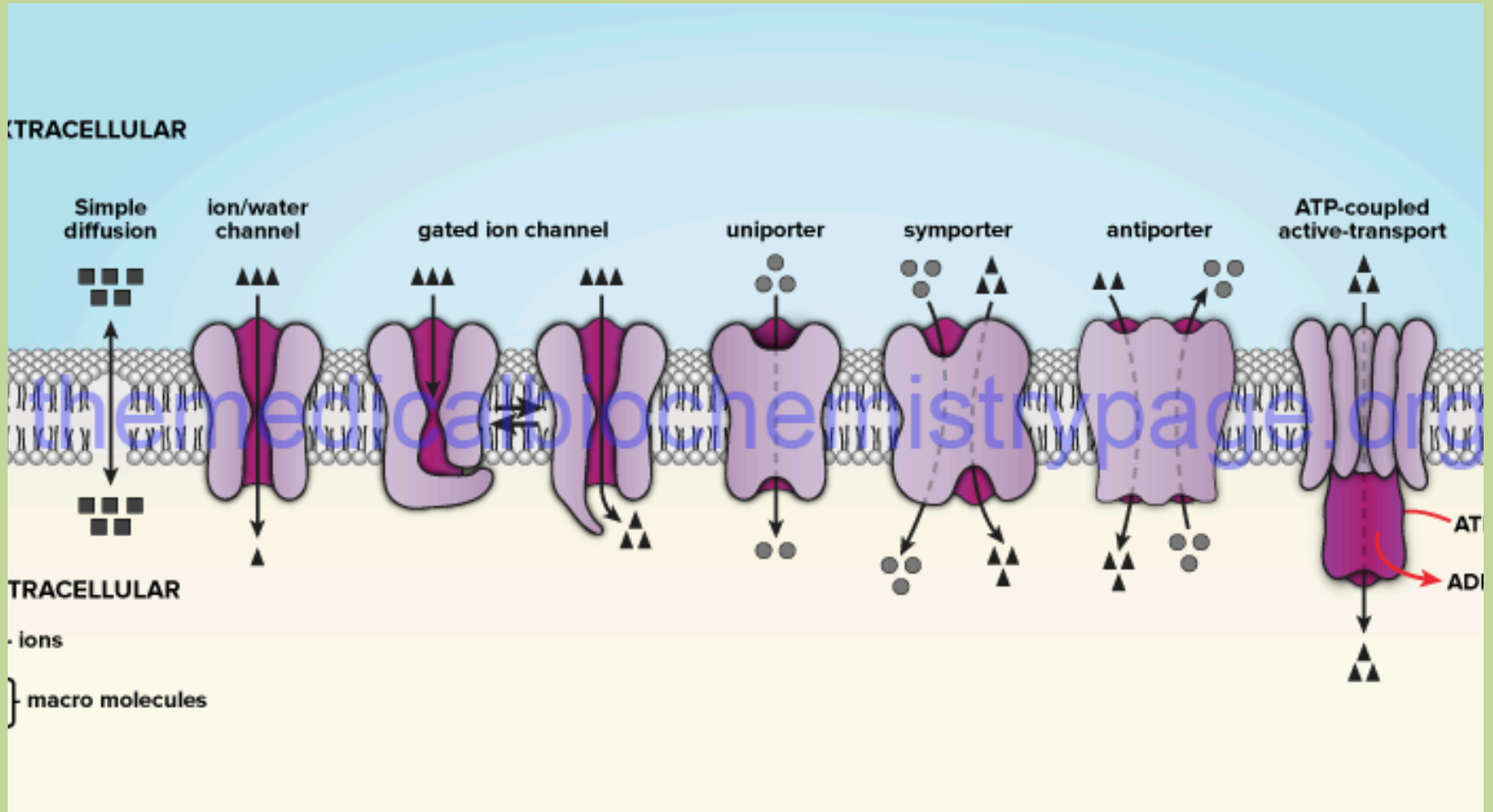
Membrane properties

- The hydrocarbon interior of membranes is an insulator sandwiched between conductors (saline): a capacitor
- The dielectric constant, ϵ , of hydrocarbon is low (≈ 2);
 $\epsilon_{\text{vacuum}}=1$; $\epsilon_{\text{water}}=78$)
- Ions and polar molecules cross *very* rarely.



Dielectric constant (*Encyclopedia Britannica*): property of an electrical insulating material (a dielectric) equal to the ratio of the capacitance of a capacitor filled with the given material to the capacitance of an identical capacitor in a vacuum without the dielectric material

Transport mechanisms



Ion movement across membranes is mediated by proteins.

Consequences of adding channels to a lipid bilayer

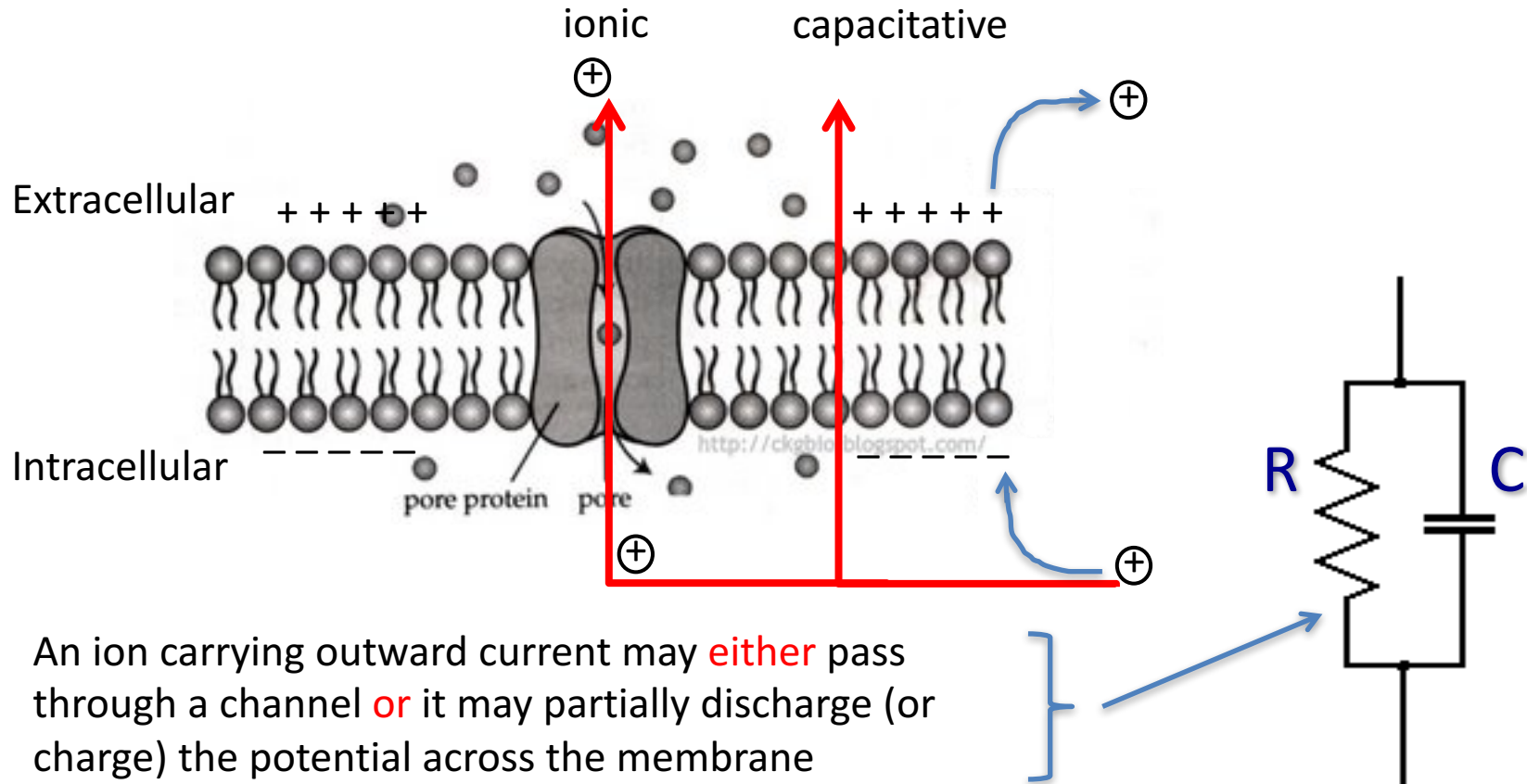
- Resistance of a pure lipid bilayer: $\approx 10^9$ ohm cm^2 (high!)
- Q: What is the conductance of a $1 \mu\text{m}^2$ patch of bilayer?
- Q: What is the consequence of adding 100 channels, each with conductance of 3 pS, to the membrane?
 - Calculate this!
 - A: It increases by a factor of $\approx 10^7$.
- Q: What happens to the capacitance of the membrane when you add these 100 channels?
 - A: very little!
 - Q: Why?
 - A: because the protein occupies only about 1% of the surface area.

R_m = specific membrane resistance $\Omega \cdot \text{cm}^2$

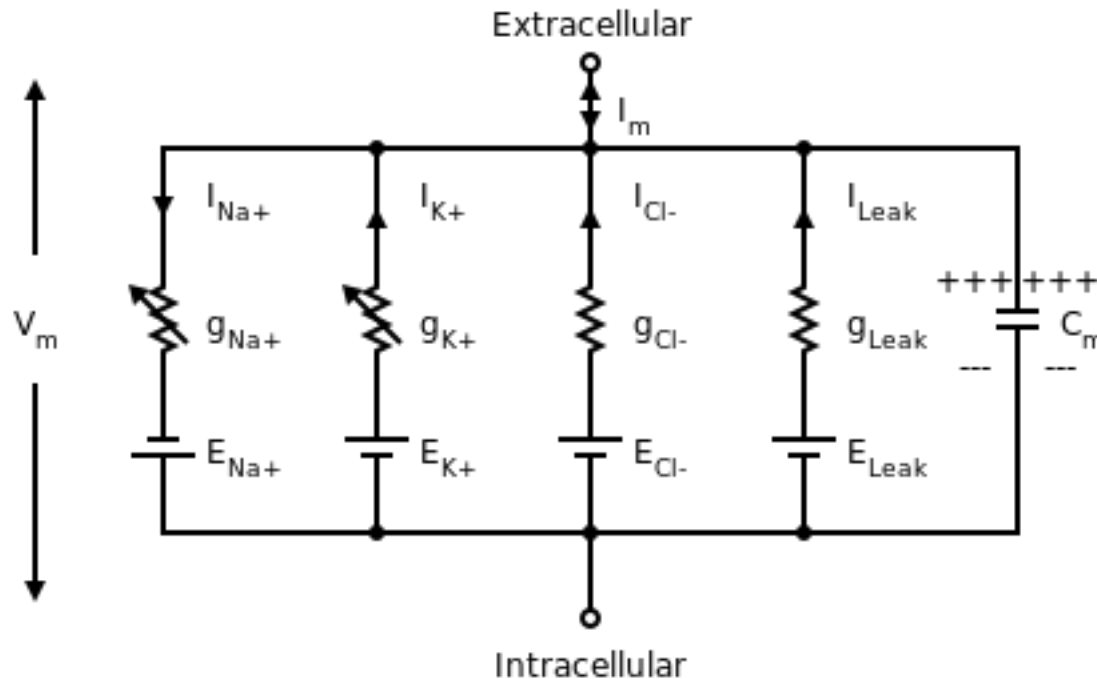
C_m = specific membrane capacitance F/cm^2

Plasma membrane = G(R) and C in parallel

- >99% hydrocarbon – capacitance
- < 1% channels – conductance

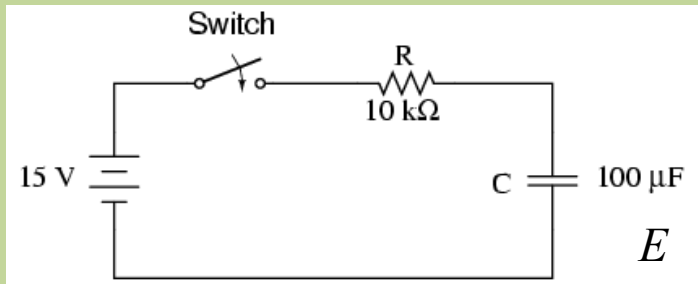


Each population of channels is equivalent to a conductor (resistor) and a battery in series



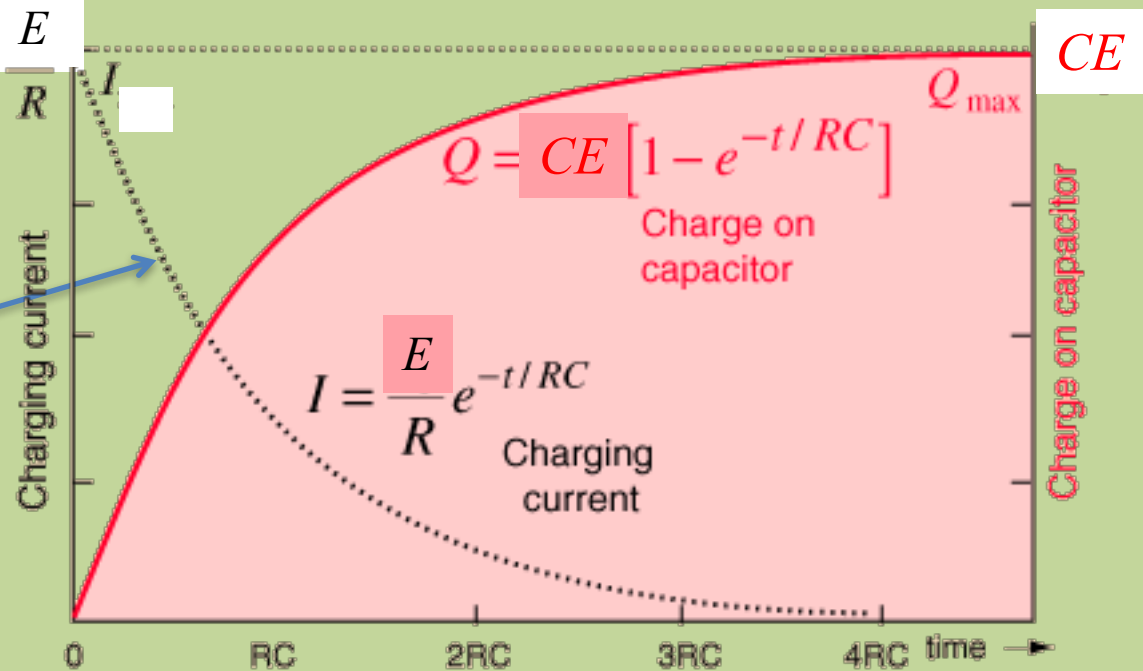
- The conductor represents the channels and their summed conductance (but remember that channels are never entirely selective for individual ions)
- The battery represents the electrochemical gradient acting on the ion

The presence of capacitance in the membrane results in time-varying responses

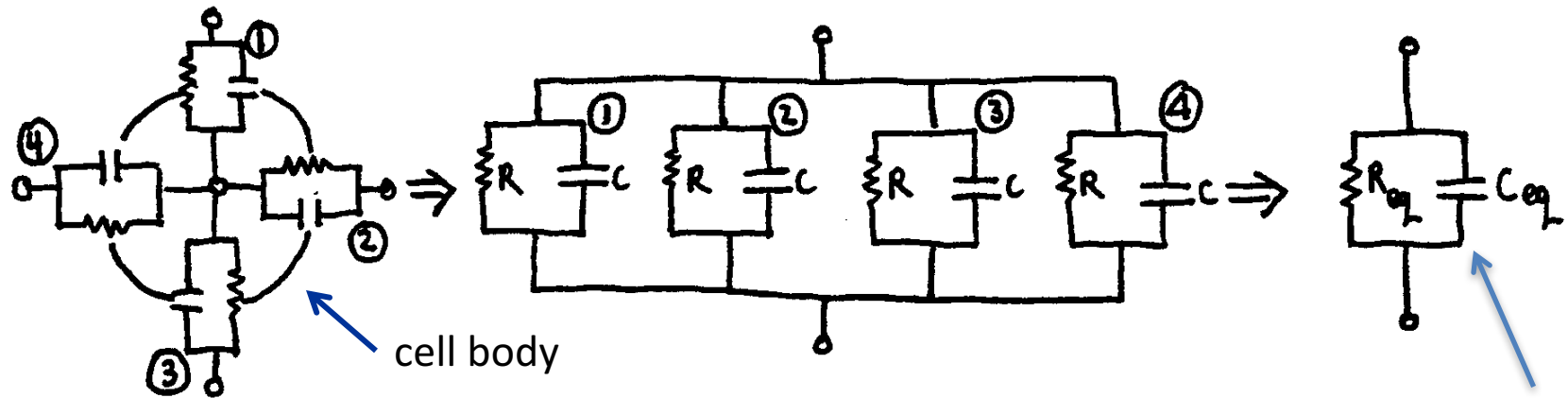


Charging of the capacitor

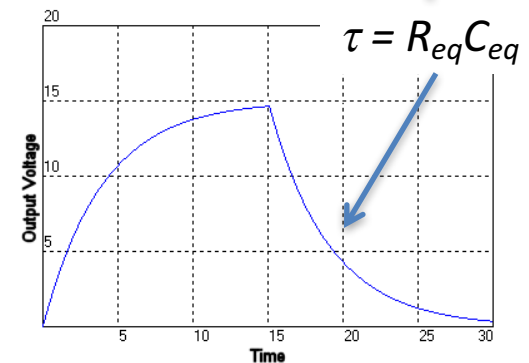
You can calculate $V_{R(t)}$, $V_{C(t)}$, from I .



The geometry of the cell determines its passive electrical properties: (1) sphere



Spheres (cell bodies) are isopotential, since the resistance between any two points within the sphere is small compared to the membrane resistance. Thus, the membrane potential is everywhere the same and the action potential occurs everywhere at once



Why does farad*ohm=second (units of τ)?

Farad \equiv coulomb/volt, but

Volt \equiv ampere*ohm=coulomb*ohm/sec

Therefore farad \equiv sec/ohm, and

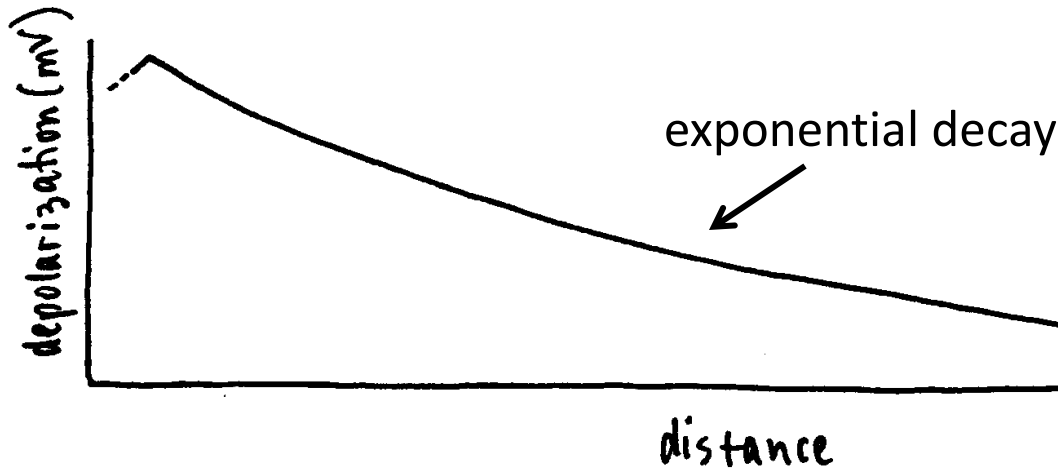
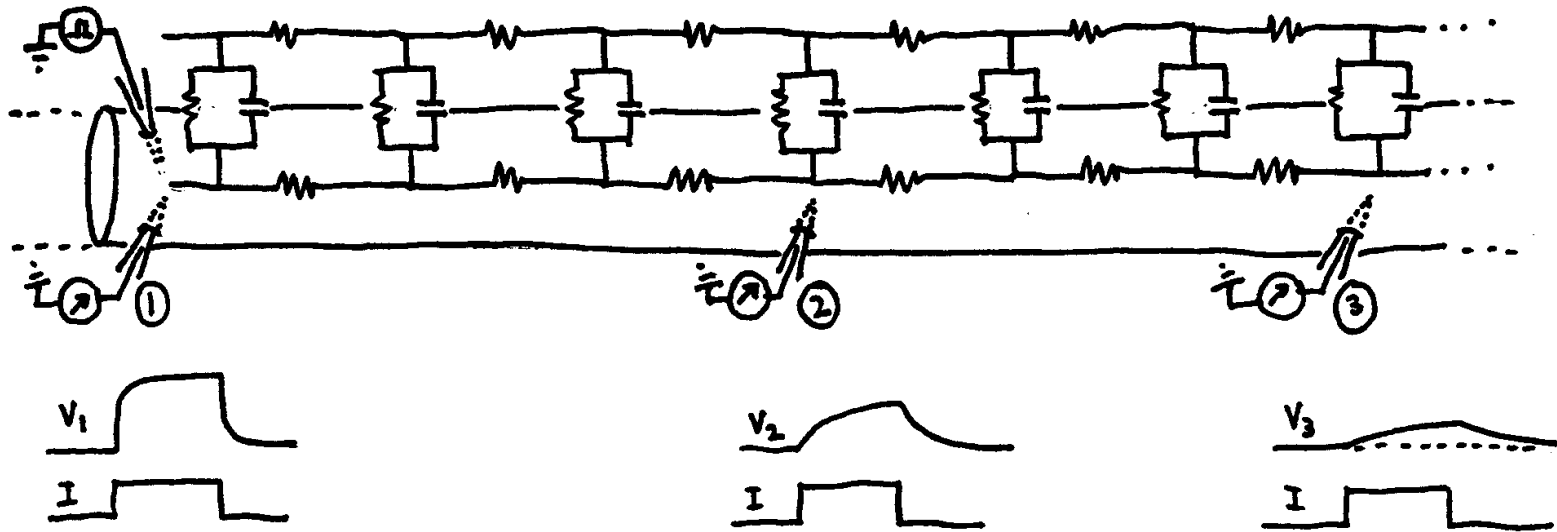
farad*ohm \equiv sec

Rising phase $V_m = I_m R_m (1 - e^{-t/\tau}) = I_m R_m (1 - \exp(-t/\tau))$

Falling phase $V_m = I_m R_m (e^{-t/\tau}) = I_m R_m (\exp(-t/\tau))$

The geometry of the cell determines its passive electrical properties: (2) axon or dendrite

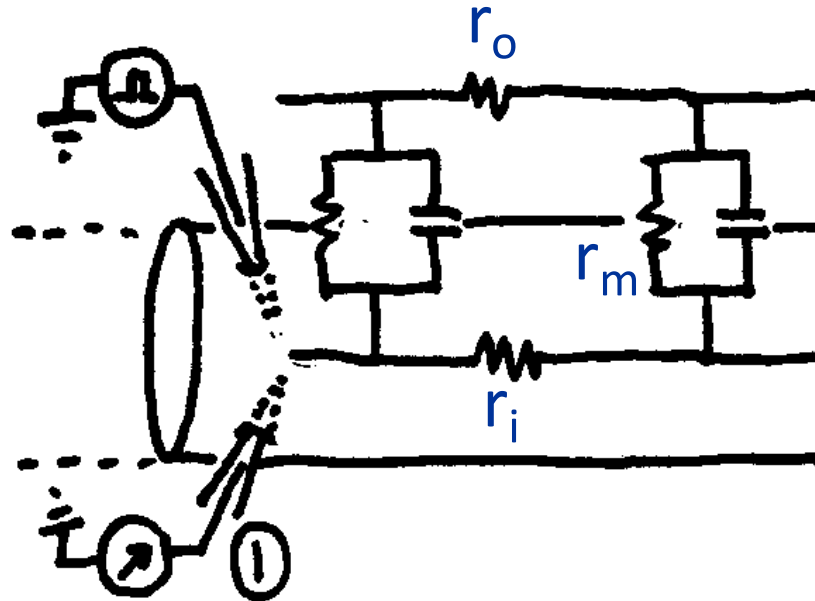
Axons and dendrites are not isopotential. There is significant axial resistance.



What determines the rate of decay of local potentials with distance along cylinders?

The relative value of the "escape" and axial resistances.

The “length constant” is a measure of the degree to which a potential decays with distance along an axon



- Length constant, λ , = distance over which the steady state signal decays to $1/e$ (37%) of its original value

$$\lambda = \sqrt{\frac{r_m}{(r_i + r_o)}} \approx \sqrt{\frac{r_m}{r_i}} \quad \text{since usually } r_i \gg r_o$$

The length constant is defined at steady state, when $dV/dt=0$

- It's more precise to calculate what would happen as a function of distance to transient signals (like EPSPs)
- The cable equation (http://en.wikipedia.org/wiki/Cable_theory)

$$\frac{1}{r_i} \frac{\partial^2 V}{\partial x^2} = c_m \frac{\partial V}{\partial t} + \frac{V}{r_m}$$

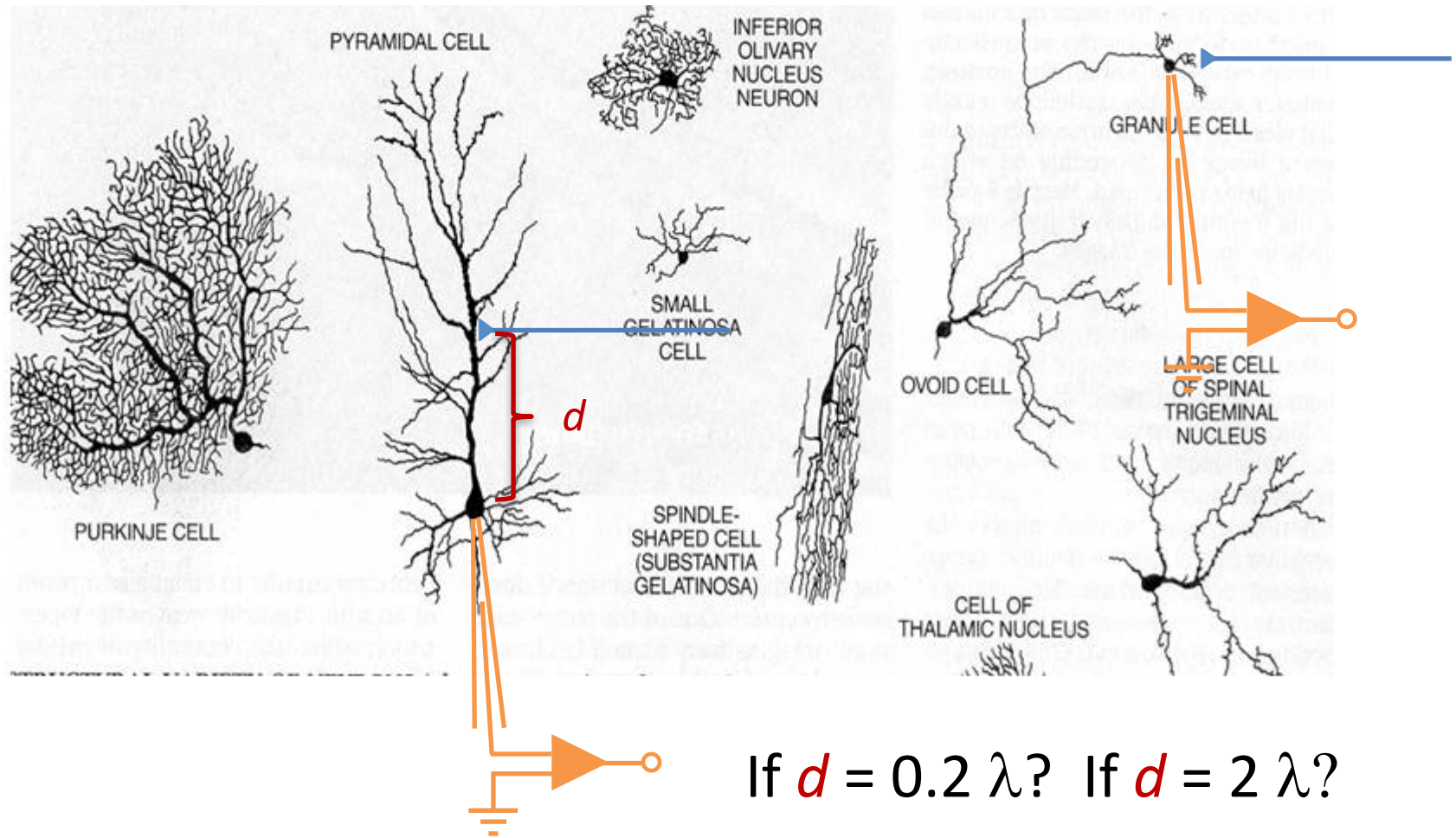
- Substituting in length constant (λ) and time constant (τ)

$$\lambda^2 \frac{\partial^2 V}{\partial x^2} = \tau \frac{\partial V}{\partial t} + V$$

← The Cable Equation

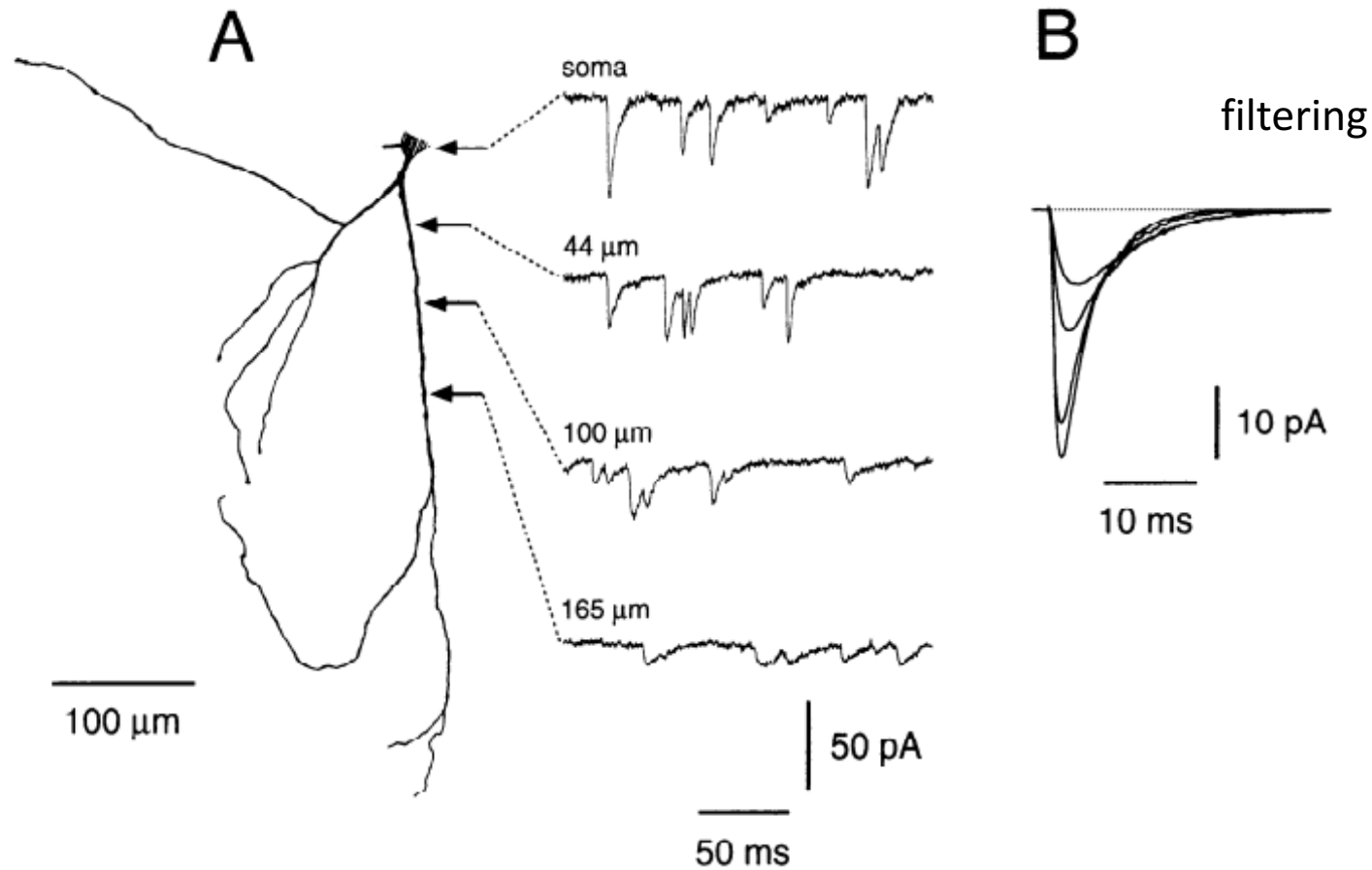
Useful for calculating how signals will decay as a function of time and distance from their point of origin.

Actual distance/electrotonic distance



Electronic distance, expressed as a fraction of λ

Synaptic responses are slower and smaller the greater their electronic distance from the recording site



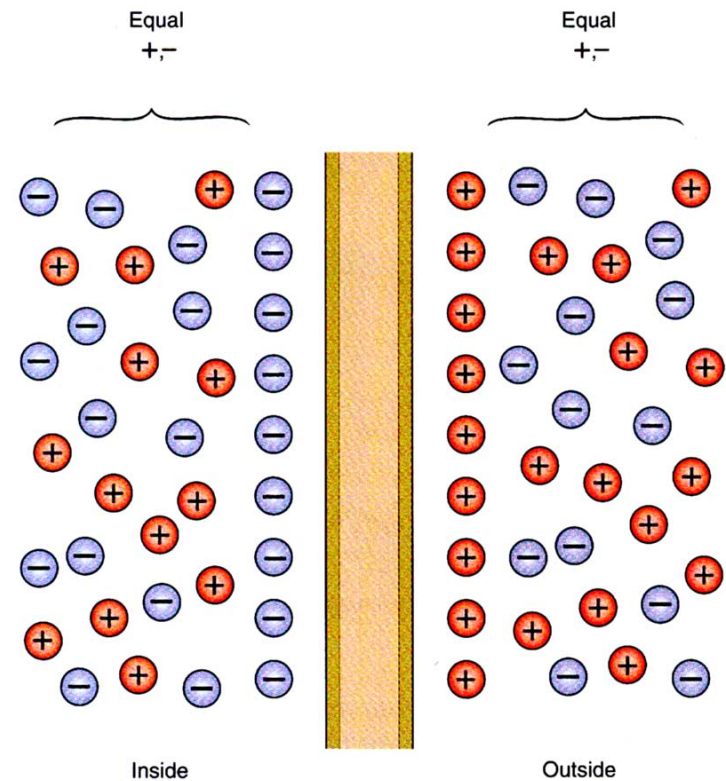
From Bekkers and Stevens (1996)

III. The Determinants of Membrane Potential

- Two factors determine the membrane potential of a cell:
 1. The permeability (conductance) ratios of its membrane to permeant ions, and
 2. The concentration gradients of those ions across the membrane.
- The sodium pump determines, directly or indirectly, the concentration gradients of ions (#2 above) and they also make a small contribution to membrane by being electrogenic.

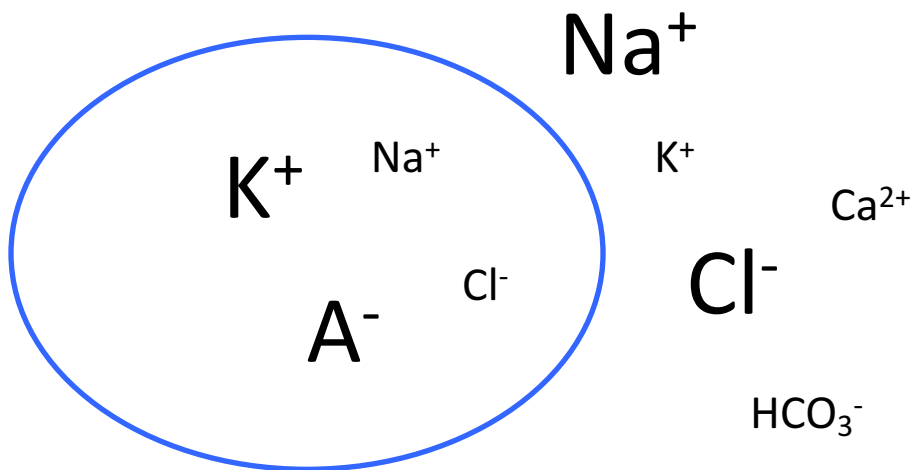
Resting Potential and the Determinants of Membrane Potential

- Neurons (and most other cells) have a standing/resting potential that is negative inside with respect to the outside (defined as 0 mV). This potential is typically in the range of -40 mV to -90 mV.
- The negative potential arises because resting cells have a high permeability to potassium, which is more concentrated inside the cell than out, and a low permeability to sodium, which is more concentrated outside the cell than inside.

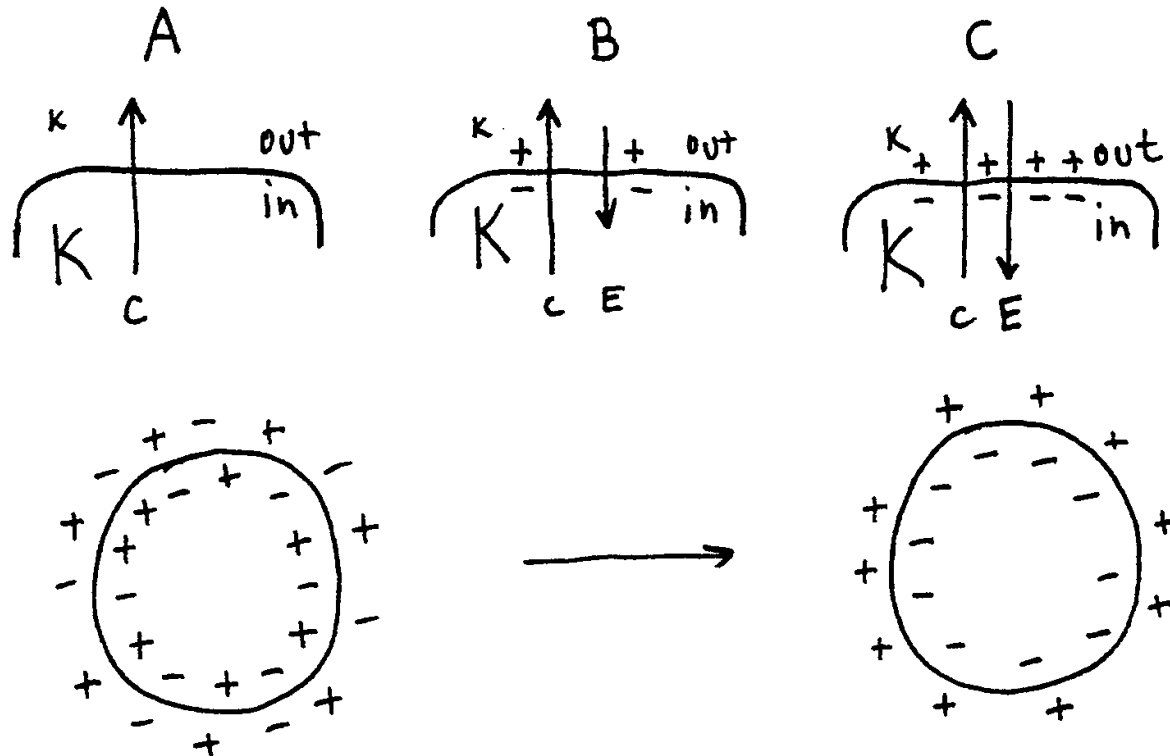


Ions are asymmetrically distributed across the membrane

Ion	Intracellular (mM)	Extracellular (mM)
Potassium	135	4
Sodium	18	145
Chloride	8	105
Calcium	0.00005	1.5
Magnesium	0.2	2
Bicarbonate and other organic anions	10	25
Large anions	135	



Building a cell, part 1: Start with K⁺ channels only



- Potassium continues to leave the cell until the concentration “force” is balanced by the opposing electrical potential.

$$E_K = \frac{RT}{zF} \ln \frac{[K]_o}{[K]_i} = 58mV \log \frac{[K]_o}{[K]_i}$$

The cell, and potassium, are at equilibrium.

Movement of ions in solution (*Hille, 2011*)

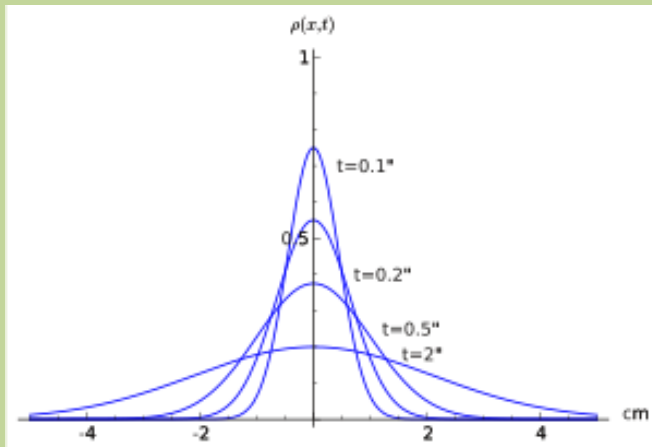
$$M_s = -D_s \frac{dc_s}{dx} \quad 1.$$

- Diffusion flux, Fick (1855)
- D_s is the diffusion coefficient, which has the units of cm^2/s .
- Mean square displacement (one dimension) $\overline{r^2} = 2Dt$

$$t \approx \frac{r^2}{2D}$$

For 2 and 3 dimensions, the denominator is $4D$ and $6D$, respectively.

For glucose
 $D = 0.5 \times 10^{-5} \text{ cm}^2/\text{s}$



Time	Distance
0.01 μm	100 ns
0.1 μm	10 μs
1 μm	1 ms
10 μm	100 ms
100 μm	10 s
1 mm	1000 s

Movement of ions in solution (cont.)

- Under the influence of an electric field,

Molar flux $M_S = -u_S c_S \frac{d\psi}{dx}$ 2.

Converting to current $I_S = z_S F M_S = -z_S F u_S c_S \frac{d\psi}{dx}$

Replacing $d\psi/dx$ with E/d ... $I_S = \frac{-z_S F u_S c_S}{d} E$

What is this?

u_S electrical mobility

c_S concentration

z_S valence

ψ potential



Nernst-Planck equation



- Nernst (1888) and Planck (1890) realized that the two terms describing the influence of electric fields and Brownian motion on ions could be combined into a single expression.

Combining 1. and 2.
$$M_S = -D_S \frac{dc_S}{dx} - u_S c_S \frac{d\psi}{dx} \quad 3.$$

- We need to convert molar flux into current, where $I_S = z_S F M_S$
- Multiply equation #3 by $z_S F$ to produce

$$I_S = -z_S F D_S \frac{dc_S}{dx} - z_S F u_S c_S \frac{d\psi}{dx}$$

- Substitute for u_S from the Einstein relation (1905), relating diffusion and electrical mobility

$$D_S = \frac{u_S RT}{z_S F}, \text{ to get:}$$

$$I_S = -z_S F D_S \frac{dc_S}{dx} - z_S F \frac{D_S z_S F}{RT} c_S \frac{d\psi}{dx}$$

Nernst-Planck

- Rearranging terms ...

$$I_S = -z_S f D_S \left(\frac{dc_S}{dx} + \frac{Fz_S c_S}{RT} \frac{d\psi}{dx} \right)$$

- When $I_S = 0$,

$$\frac{d\psi}{dx} = -\frac{RT}{z_S F} \frac{1}{c_S} \frac{dc_S}{dx} = -\frac{RT}{z_S F} \frac{d}{dx} (\ln c_S)$$

NPE: Nernst
Planck equation

- Integrating over x,

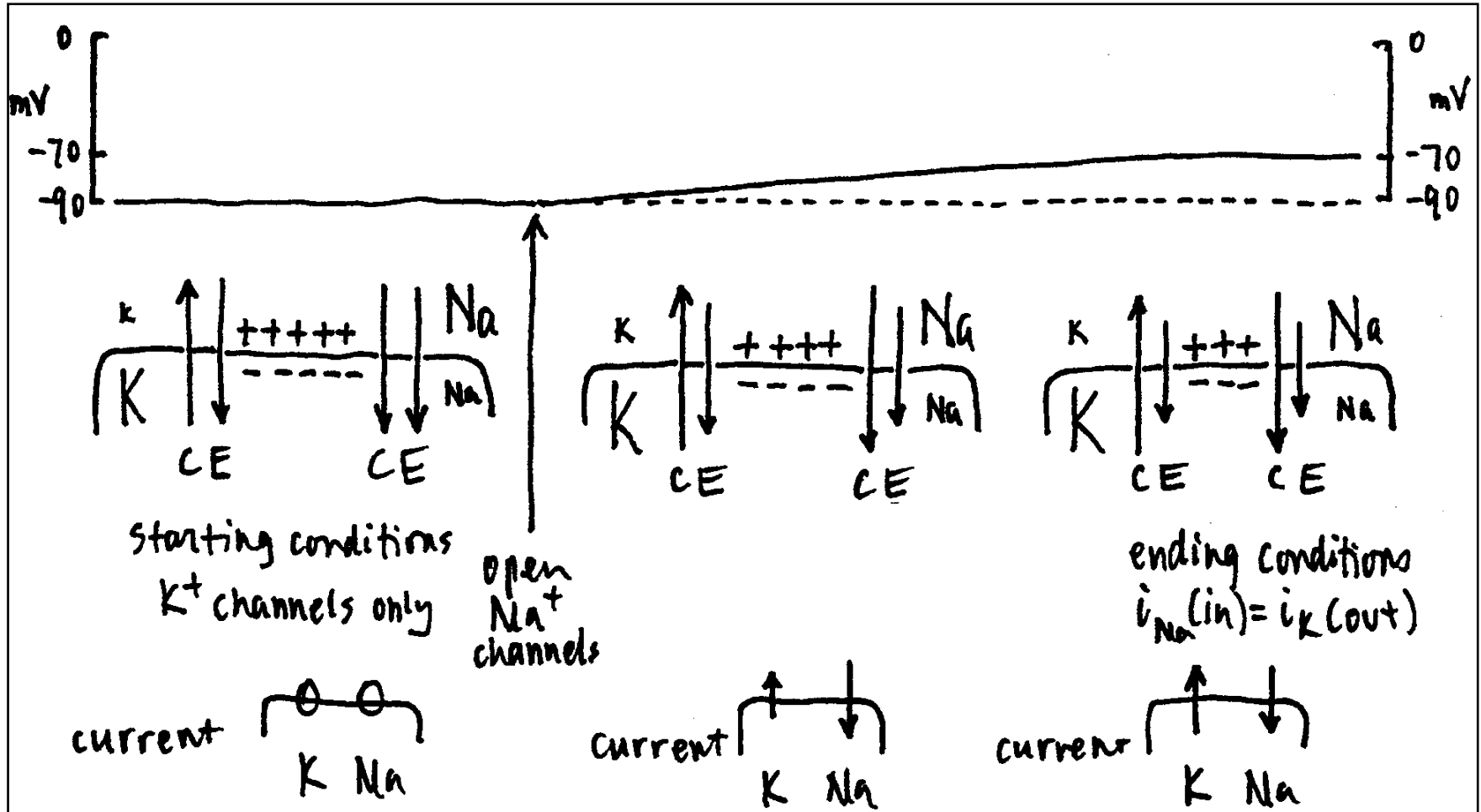
$$E_1 - E_2 = \Delta E = \frac{RT}{z_S F} \ln \frac{[S]_2}{[S]_1}$$

For our purposes,
2 is “outside” and
 $E_2 = 0$.

Model cell, permeable only to K⁺

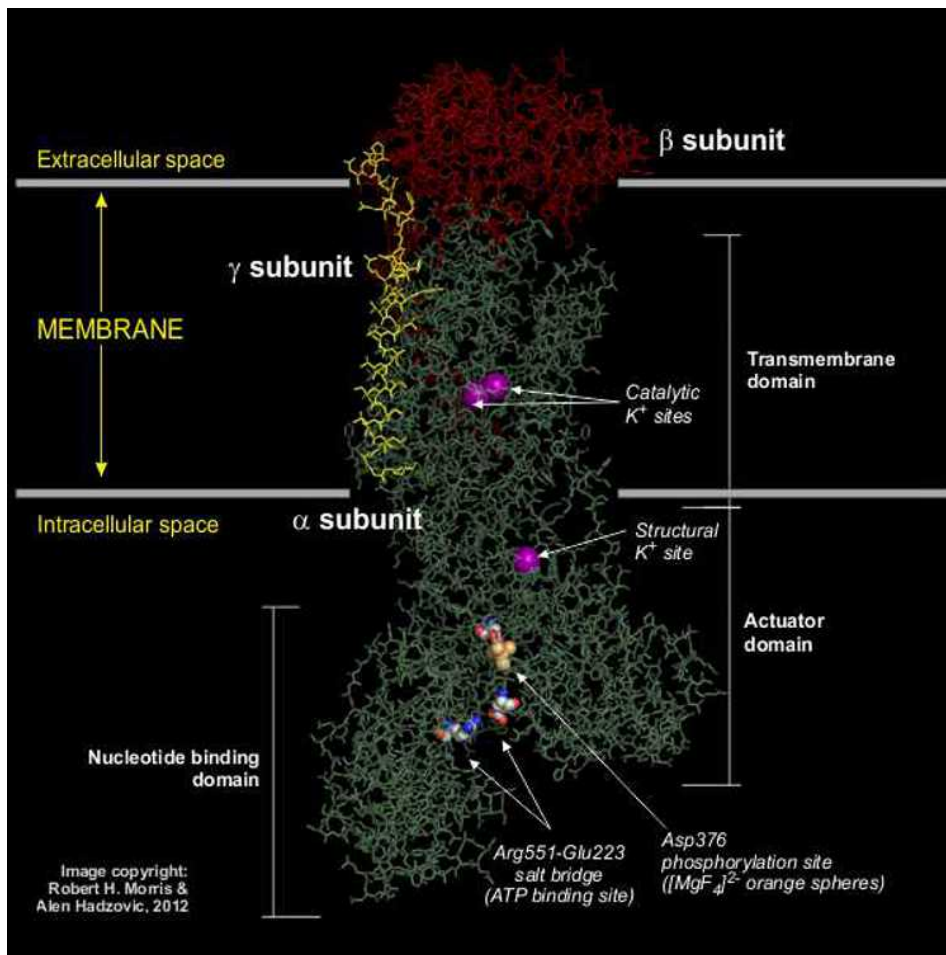
- If K_o is 4 mM and K_i is 135 mM, V_m will go to ~-89 mV.
$$E_K = \frac{RT}{zF} \ln \frac{[K]_o}{[K]_i} = 58mV \log \frac{[K]_o}{[K]_i}$$
- But wait! Is it OK to assume that the concentrations have remained constant?
- Consider a cell that is a sphere with a 25 μm diameter.
 - vol. = 8 x 10⁻¹² l.; surface area = 2 x 10⁻⁵ cm²
- What is the capacitance of the membrane?
 - Q = CV, V = Q/C
 - Capacitance of biological membranes ≅ 10⁻² F/m² = 1 μF/cm²; therefore C = 2 x 10⁻¹¹ F
- Calculate charge (Q) needed to charge the membrane to 90 mV
 - Q = CV = 2 x 10⁻¹¹ F * 0.09 V = 1.8 x 10⁻¹² coulombs
- This much charge carried by 1.8 x 10⁻¹⁷ moles of monovalent ion, which, in this cell, corresponds to about 2 μM (1 part in 70,000).

Building a cell, part 2: Add Na⁺ channels

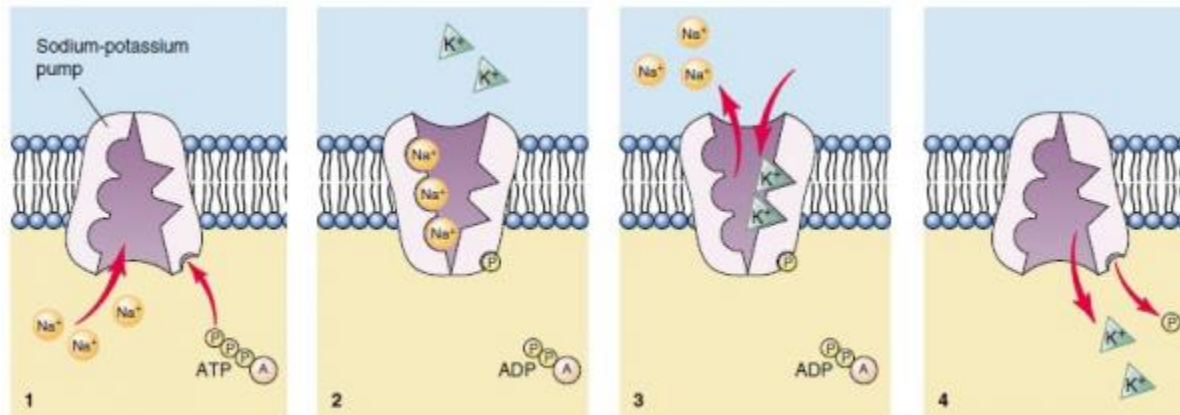


- Neither potassium nor sodium is at equilibrium. Although net current is zero, this situation is not sustainable.
- The sodium pump (Na⁺/K⁺-dependent ATPase) comes to the rescue.

Sodium-potassium ATPase



- Coupled K^+ entry)
- Driven by mass action under physiological conditions, meaning that substrate availability is limiting
- Electrogenic, meaning that it produces a current



Goldman Hodgkin Katz (GHK) equation

$$E_{rev} = \frac{RT}{F} \ln \frac{P_K [K]_o + P_{Na} [Na]_o + P_{Cl} [Cl]_i}{P_K [K]_i + P_{Na} [Na]_i + P_{Cl} [Cl]_o}$$

- This applies to non-equilibrium situations when total membrane current is zero. It's thus a more general expression than the Nernst equation, which refers to an equilibrium situation.
- Goldman (1943) and Hodgkin and Katz (1949)
- Assumptions:
 - ion flux is influenced both by the concentration gradient and by the electric field, according to the Nernst-Planck Equation
 - ions cross the membrane independently of one another (no physical or electrostatic interaction)
 - the electric field is constant (constant field).

Some of the assumptions used to derive the GHK:

- Ions don't interact with one another.
- No saturation
- No interaction of the ion with the channel (which would violate the constant field assumption)
- No channel block

are wrong!

- Nonetheless the equation has proven to be fairly accurate. Some of the assumptions (e.g., block, saturation) have little effect on the estimate of reversal potential.

An electrical approach (*g*, not *P*)

driving force = extent to which an ion is out of equilibrium

$$I_i = I_{Na} + I_K + I_{Cl} = 0$$

$$I_i = g_i(V_m - E_i)$$

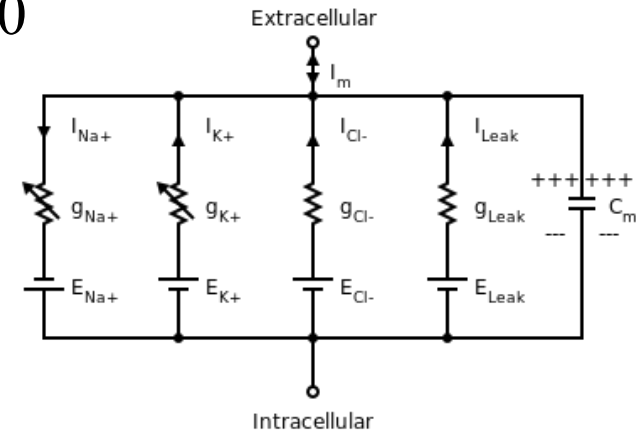
Ohm's law: current = conductance * **driving force**

$$g_{Na}(V_m - E_{Na}) + g_K(V_m - E_K) + g_{Cl}(V_m - E_{Cl}) = 0$$

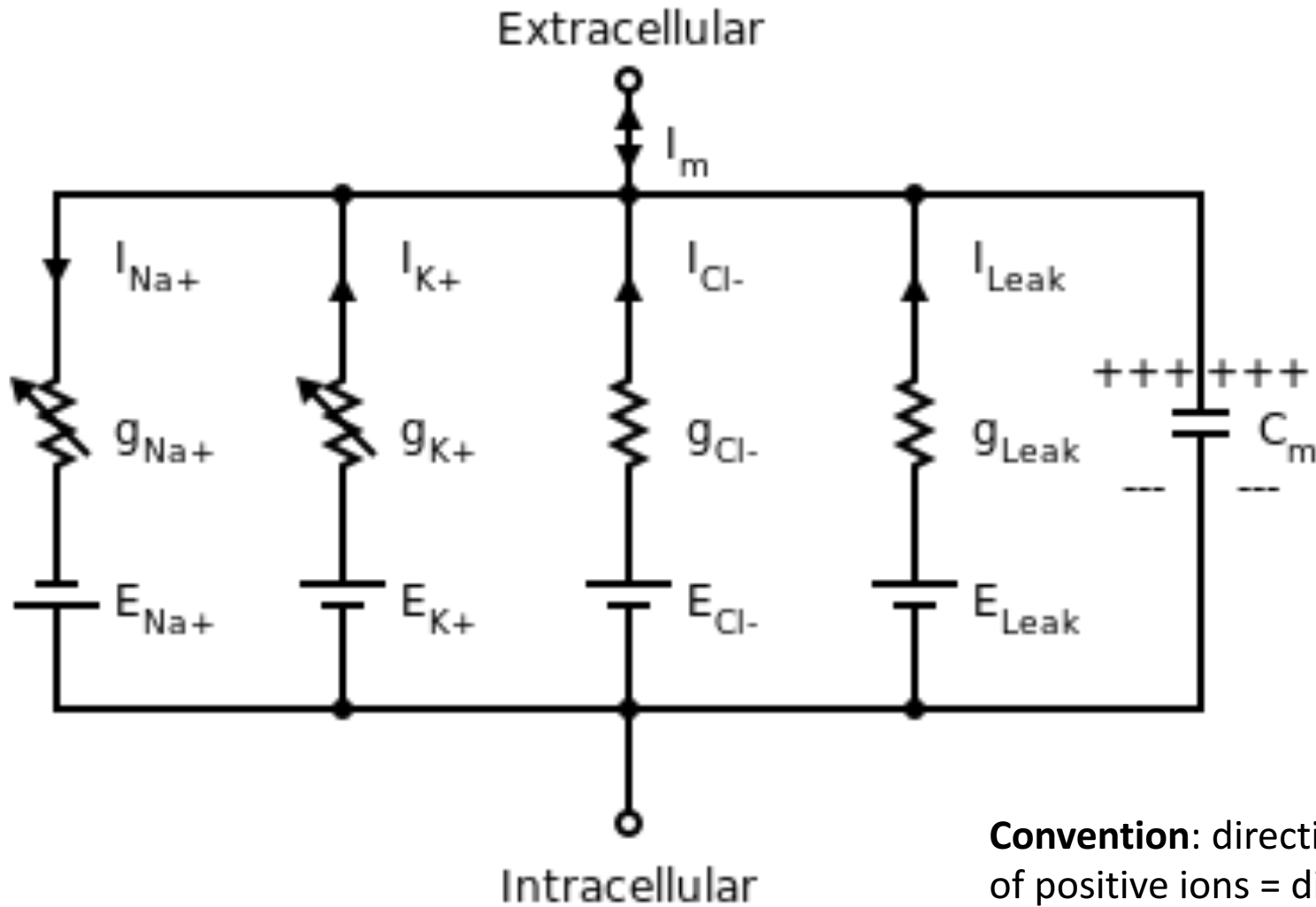
Solve for V_m

$$V_m = \frac{g_{Na}}{\Sigma g_i} E_{Na} + \frac{g_K}{\Sigma g_i} E_K + \frac{g_{Cl}}{\Sigma g_i} E_{Cl}$$

- Not as general as GHK
 - If you change the concentration or permeant ions outside the cell, *g* is likely to change, but not *P*
- Nonetheless useful
- Both this expression and GHK reduce to Nernst when only one ion is considered, assuming that *g* and *P* are equivalent.



Electrical Equivalent of a Neuron



Convention: direction of travel of positive ions = direction of current flow

Convention: inward current is negative in sign (Franklin B)

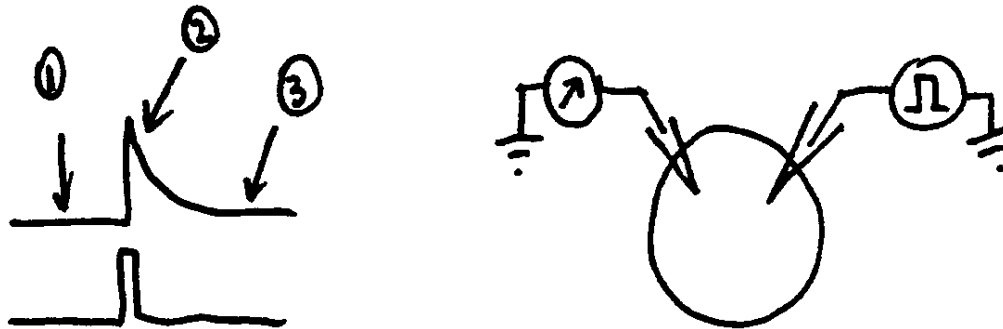
How does the pump contribute to the resting potential?

1. By generating gradients of permeant ions
 2. By passing current. GHK does not account for this contribution.
- In a model with only sodium and potassium currents, what is the ratio of i_{Na} and i_K flux through channels in the absence of the pump?
 - In the presence of the pump?

Leak current and the resting cell

- What are the channels that account for the resting cell's "K-like" potential? Are these simply a few of the voltage-dependent potassium channels that happen to be open at rest?
- No.
 - HCN (hyperpolarization activated cyclic nucleotide gated), cation channels, which produce the I_h current
 - 2P K channels
 - M current (K)
 - BK and SK channels (calcium activated)
 - NALCN channel (Na Leak Channel Non-selective), cation

The properties of leak channels, acting together



- What is the influence of leak channels on signaling?
- What is the difference between the influence of leak channels and that of chloride channels, when chloride is at equilibrium?

- End of day 1