Membrane Potential

Jim Huettner

The Lipid Bilayer is a Selective Barrier

inside   outside

hydrophobic molecules (anesthetics)

gases (O_2, CO_2)

small uncharged polar molecules

large uncharged polar molecules

Ions

charged polar molecules (amino acids)

water
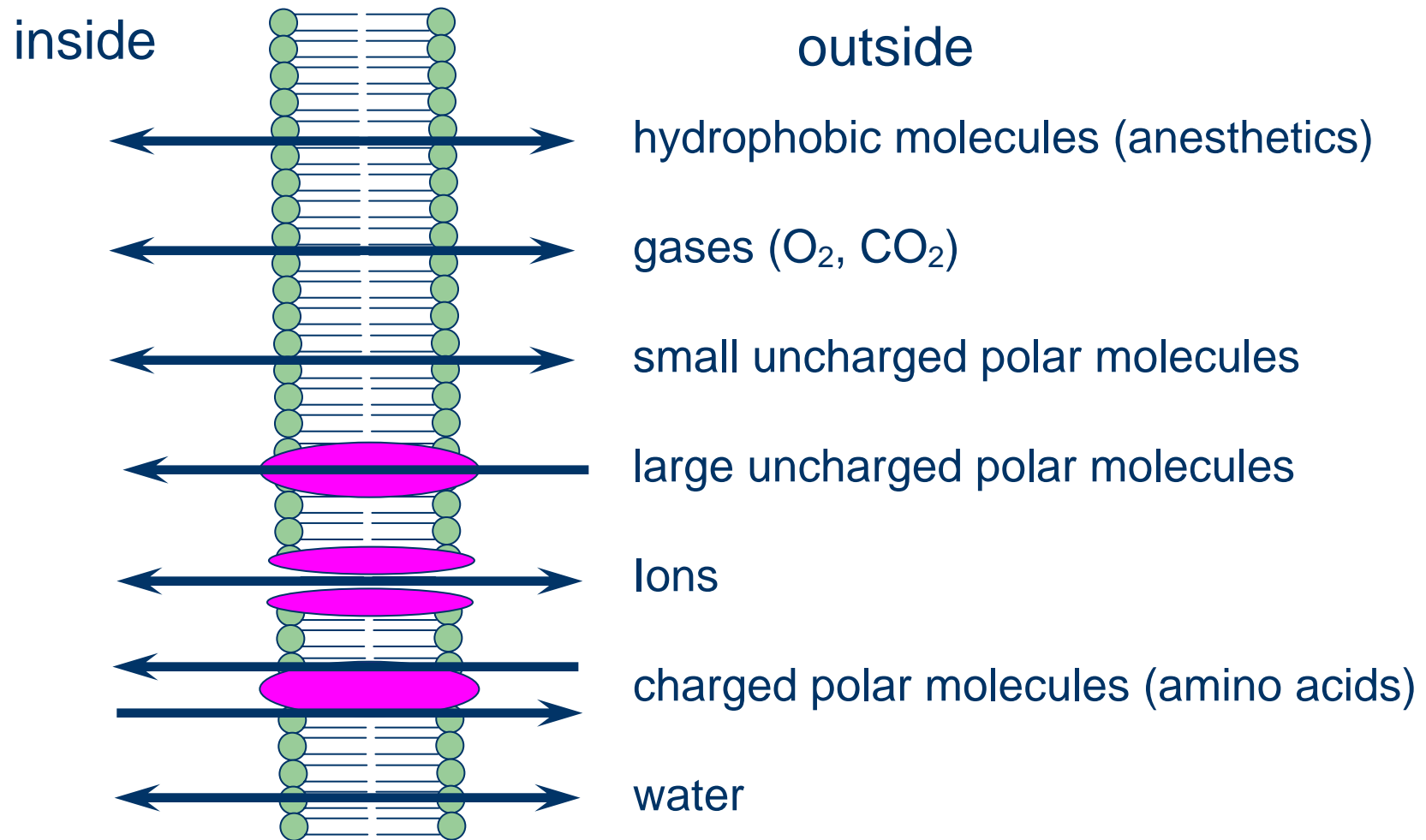
Review of Electricity

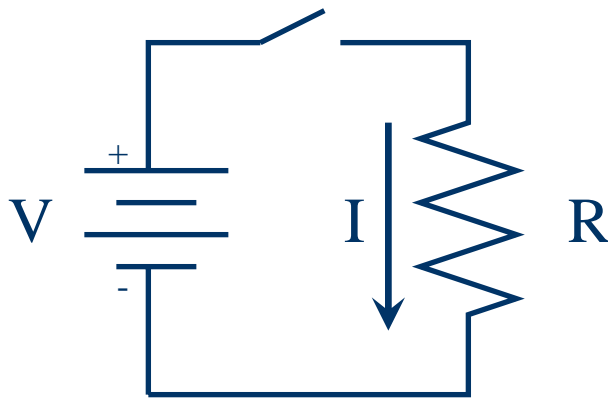|				
Charge	Q		Coulombs	C
Current	I	Coulombs / sec	Amps	A
Voltage	V	Joules / Coulomb	Volts	V
Conductance	G		Siemens	S
Resistance	R		Ohms	Ω

$$G = 1 / R \qquad R = 1 / G$$

Ohm's Law

$$V = I * R = I / G$$

$$I = V / R = V * G$$

R ∝ length

R ∝ 1/area
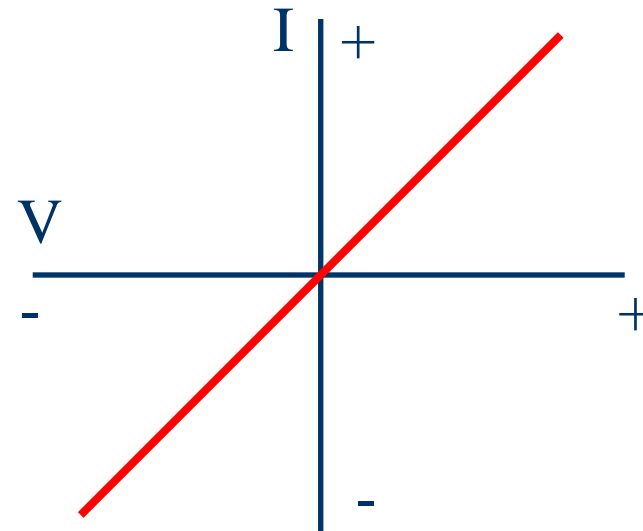
Capacitance

Capacitance   C   Coulombs / Volt   Farads   F

$$C = Q / V \qquad Q = C * V \qquad I = dQ / dt = C * dV / dt$$

$C \propto$ area

I

$C \propto 1 /$ thickness

V

$+ + + + + + + +$

C
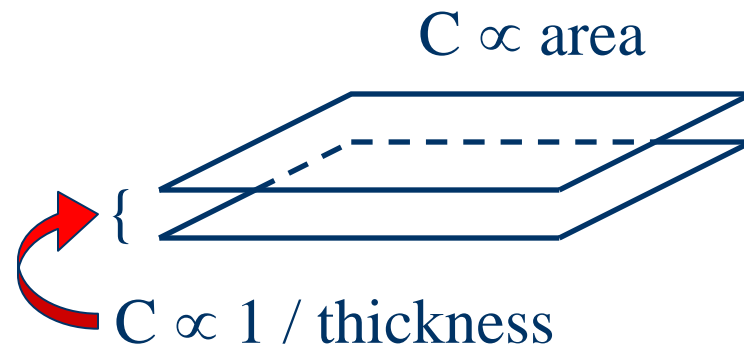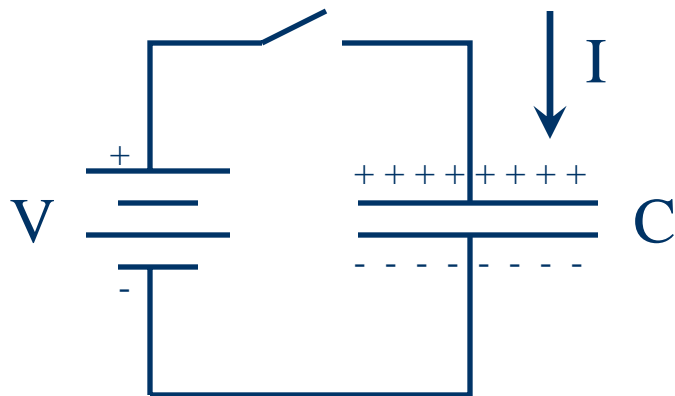
For biological membranes:
Specific Capacitance = 1 µF / cm^2

Recording Membrane Potential

current

voltage

I

V

reference

ground
0 mV

sharp microelectrode

whole-cell "patch" electrode

Ion Concentrations

Na   117
K   3
Cl   120
Anions   0
Total   240

[+ charge] = [- charge]

0 mV

Na   30
K   90
Cl   4
Anions 116
Total   240

[+ charge] = [- charge]

-89 mV
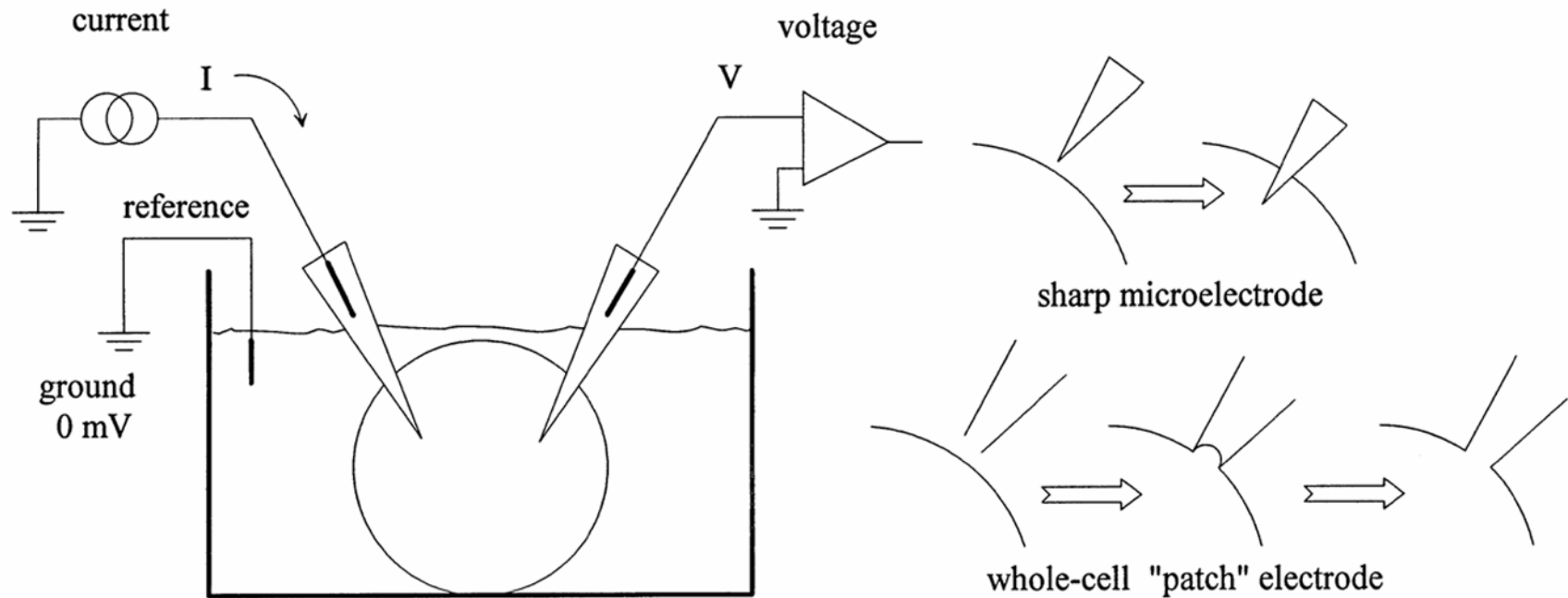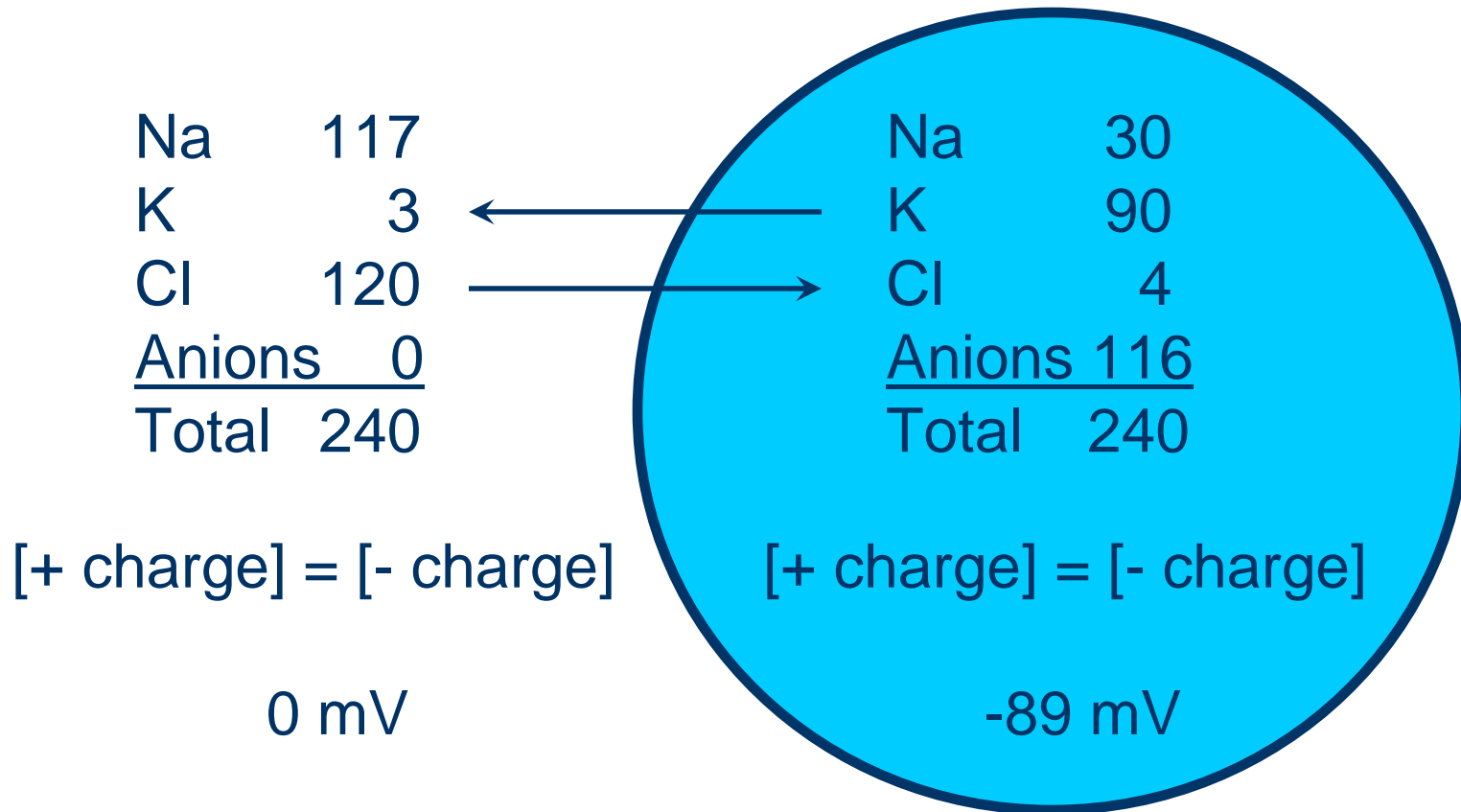
Movement of Individual K$^+$ ions

Na   117
K   3
Cl   120
Anions   0
Total   240

[+ charge] = [- charge]

0 mV

Na   30
K   90
Cl   4
Anions 116
Total   240

[+ charge] = [- charge]

0 mV
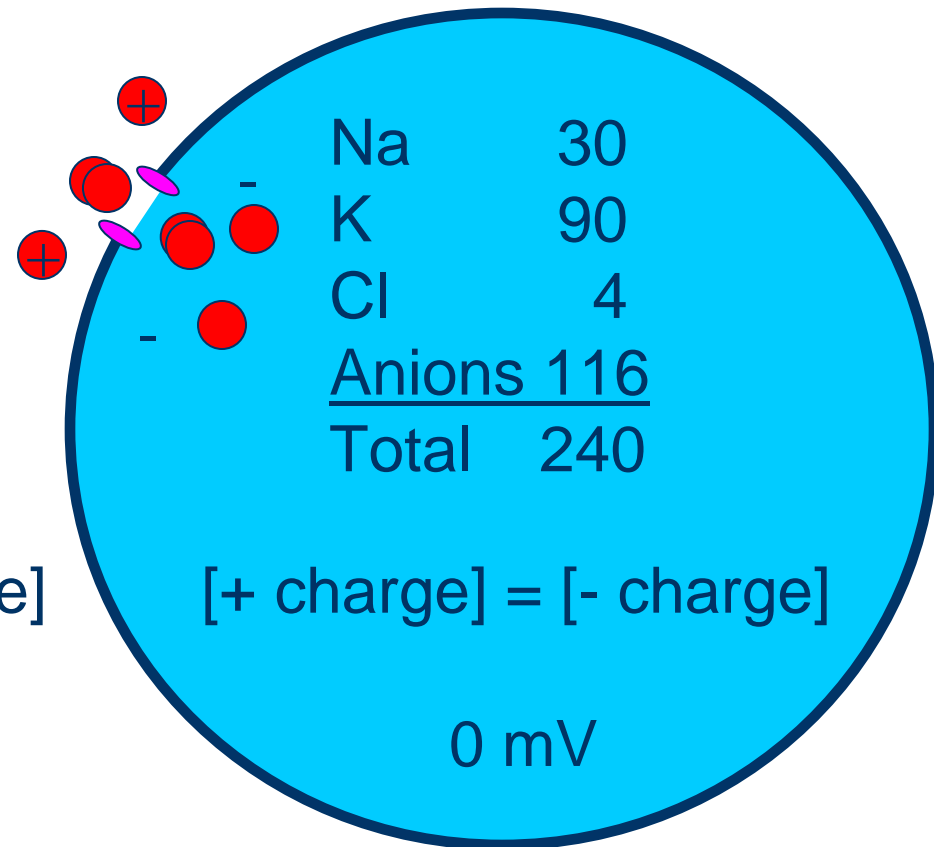
Movement of Individual Cl⁻ ions

Na   117
K   3
Cl   120
Anions   0
Total   240

[+ charge] = [- charge]

0 mV

Na   30
K   90
Cl   4
Anions 116
Total   240
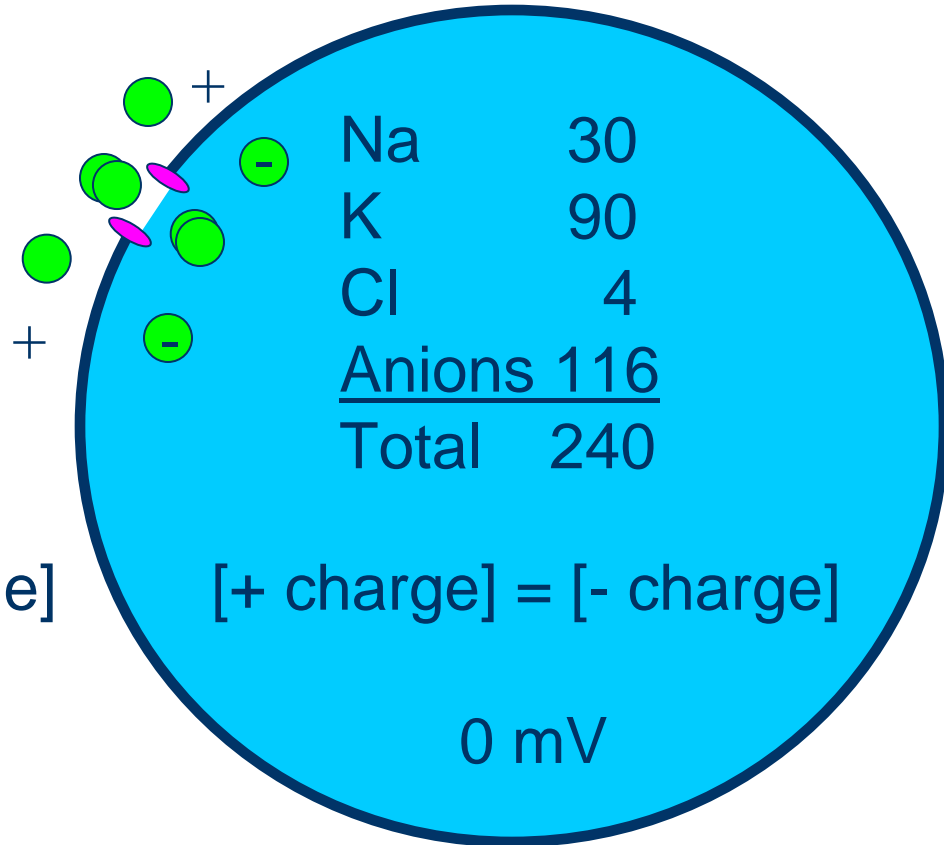
[+ charge] = [- charge]

0 mV

Sample Problem

- How many ions must cross the membrane of a spherical cell 50 µm in diameter (r = 25 µm) to create a membrane potential of −89 mV?

  Q (Coulombs) = C (Farads) * V (Volts)

  Specific Capacitance = 1.0 µFarad / cm^2

  Surface area = 4 π r^2

  Faraday's Constant = 9.648 x 10^4 Coulombs / mole

  Avogadro's # = 6.022 x 10^{23} ions / mole

Calculations

Area = $4 \pi r^2$ ①

$= 4 \pi (25 \times 10^{-4} \text{ cm})^2$

$= 7.85 \times 10^{-5} \text{ cm}^2$

$= 78.5 \times 10^{-6} \text{ } \mu\text{Farads}$

$= 78.5 \times 10^{-12} \text{ Farads}$

Q = C * V ②

$= 78.5 \times 10^{-12} \text{ Farads} * 0.089 \text{ Volts}$

$= 7 \times 10^{-12} \text{ Coulombs}$

③

$\Rightarrow 7 \times 10^{-12} \text{ Coulombs} / 9.65 \times 10^4 \text{ Coulombs per mole}$

$= 7.3 \times 10^{-17} \text{ moles of ions must cross the membrane}$

$= 0.073 \text{ femptomoles} \quad \text{or} \quad \sim 44 \times 10^6 \text{ ions}$

The Nernst Equation

Calculates the membrane potential at which an ion will be in <u>electrochemical equilibrium</u>.

At this potential: total energy inside = total energy outside

Electrical Energy Term: z * F * V

Chemical Energy Term: R * T * ln [Ion]

Z is the charge, 1 for Na^+ and K^+, 2 for Ca^{2+} and Mg^{2+}, -1 for Cl^-

F is Faraday's Constant = 9.648×10^4 Coulombs / mole

R is the gas constant = 8.315 Joules / °Kelvin * mole

T is the temperature in °Kelvin

Nernst Equation Derivation

$$zF * V_{in} + RT * \ln [K^+]_{in} = zF * V_{out} + RT * \ln [K^+]_{out}$$

$$zF (V_{in} - V_{out}) = RT (\ln [K^+]_{out} - \ln [K^+]_{in})$$

$$E_K = V_{in} - V_{out} = (RT / zF) \ln ([K^+]_{out} / [K^+]_{in})$$

$$E_K = 2.303 (RT / F) * \log_{10} ([K^+]_{out} / [K^+]_{in})$$

In General:

$$E_{ion} = (60 \text{ mV} / z) * \log ([ion]_{out} / [ion]_{in}) \quad @ \quad 30°$$

Nernst Potential Calculations

- **First K and Cl**

  E_K = 60 mV log (3 / 90) = 60 * -1.477 = -89 mV

  E_{Cl} = (60 mV / -1) log (120 / 4) = -60 * 1.477 = -89 mV

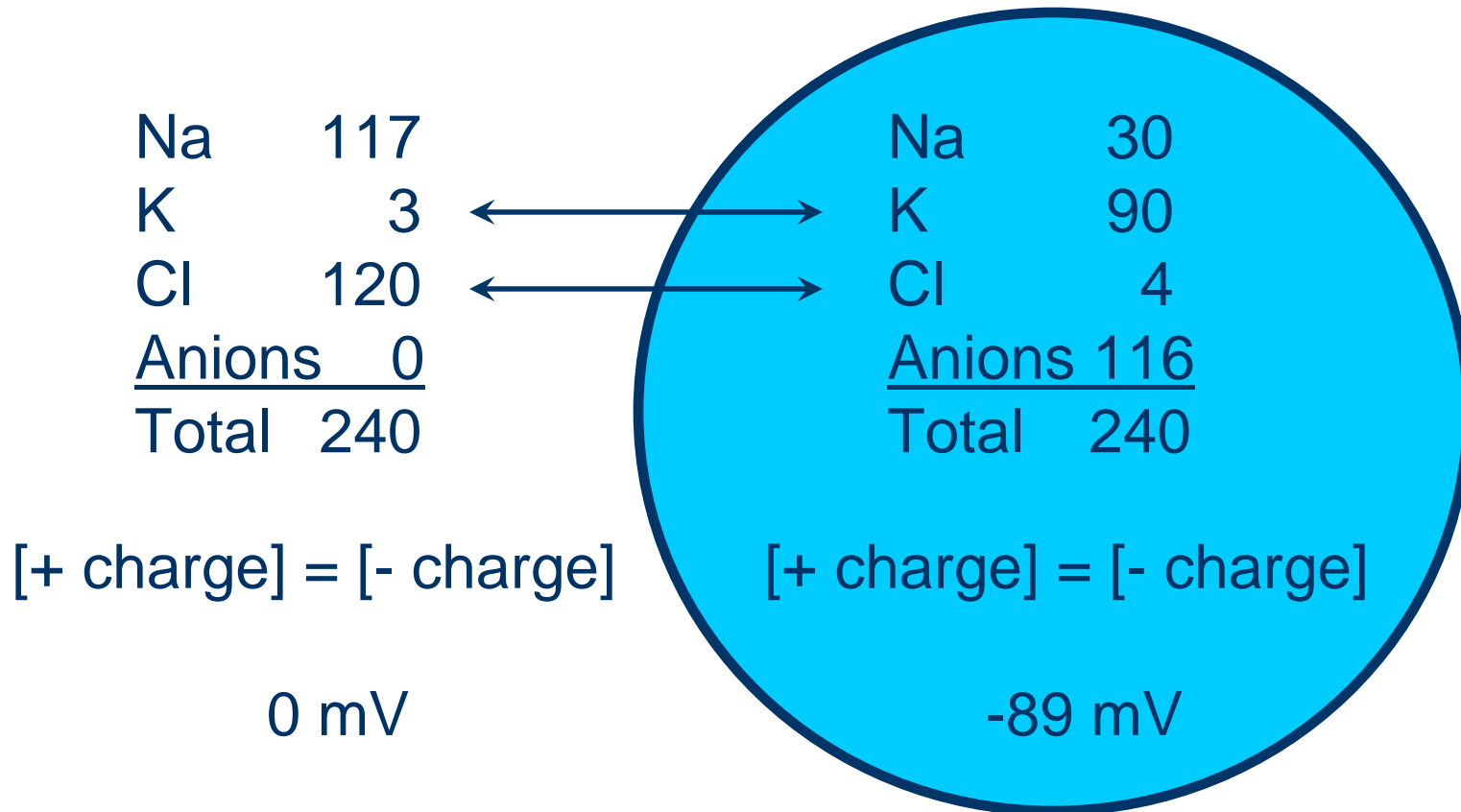 Both Cl and K are at electrochemical equilibrium at -89 mV

- **Now for Sodium**

  E_{Na} = 60 mV log (117 / 30) = 60 * 0.591 = +36 mV

  When Vm = -89 mV, both the concentration gradient and electrical gradient for Na are from outside to inside

At Electrochemical Equilibrium:

- The concentration gradient for the ion is exactly balanced by the electrical gradient

- There is no net flux of the ion

- There is no requirement for any sort of energy-driven pump to maintain the concentration gradient

Ion Concentrations

Na   117
K   3
Cl   120
Anions   0
Total   240

[+ charge] = [- charge]

0 mV

Na   30
K   90
Cl   4
Anions 116
Total   240

[+ charge] = [- charge]

-89 mV

Environmental Changes: Dilution

Add water

Na	58.5
K	1.5
Cl	60
Anions	0
Total	120

E_K = -107 mV
E_{Cl} = -71 mV

H_2O

Na	30
K	90
Cl	4
Anions	116
Total	240

0 mV

-89 mV

Environmental Changes: ↑K⁺ or ↓Cl⁻

Starting Conditions

	Out	In
Na	117	30
K	3	90
Cl	120	4
Anions	0	116

$E_K = E_{Cl} = $ -89 mV

Relative Volume = 1

↑K⁺
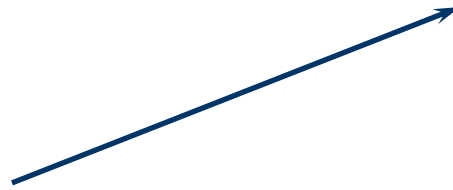
Na	114	29
K	6	91
Cl	120	7.9
Anions	0	112.1

$E_K = E_{Cl} = $ -71 mV

Relative Volume = 1.034

↓Cl⁻
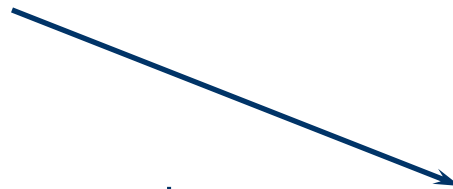
Na	117	30.5
K	6	89.5
Cl	60	2.1
Anions	60	117.9

$E_K = E_{Cl} = $ -88 mV

Relative Volume = 0.984

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Deviation from the Nernst Equation

Resting membrane potentials in real cells deviate from the Nernst equation, particularly at low external potassium concentrations.

The Goldman, Hodgkin, Katz equation provides a better description of membrane potential as a function of potassium concentration in cells.

Squid Axon
Curtis and Cole, 1942

$P_K : P_{Na} : P_{Cl}$
1 : 0.04 : 0.05

E_K (Nernst)

Resting Potential (mV)

External [K] (mM)
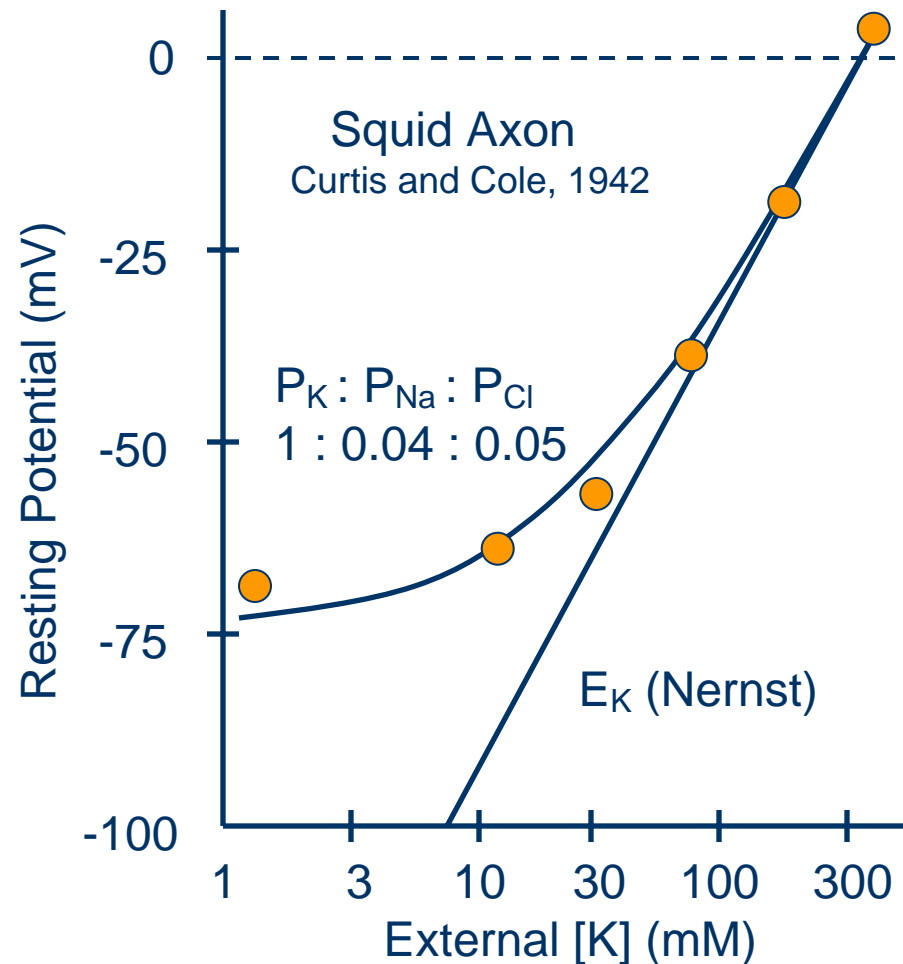
The Goldman Hodgkin Katz Equation

$$V_m = 60mV * \log\left(\frac{P_K * [K]_{out} + P_{Na} * [Na]_{out} + P_{Cl} * [Cl]_{in}}{P_K * [K]_{in} + P_{Na} * [Na]_{in} + P_{Cl} * [Cl]_{out}}\right)$$

- Resting Vm depends on the concentration gradients <u>and</u> on the relative permeabilities to Na, K and Cl. The Nernst Potential for an ion does not depend on membrane permeability to that ion.

- The GHK equation describes a steady-state condition, <u>not</u> electrochemical equilibrium.

- There is net flux of individual ions, but no net charge movement.

- The cell must supply energy to maintain its ionic gradients.

GHK Equation: Sample Calculation
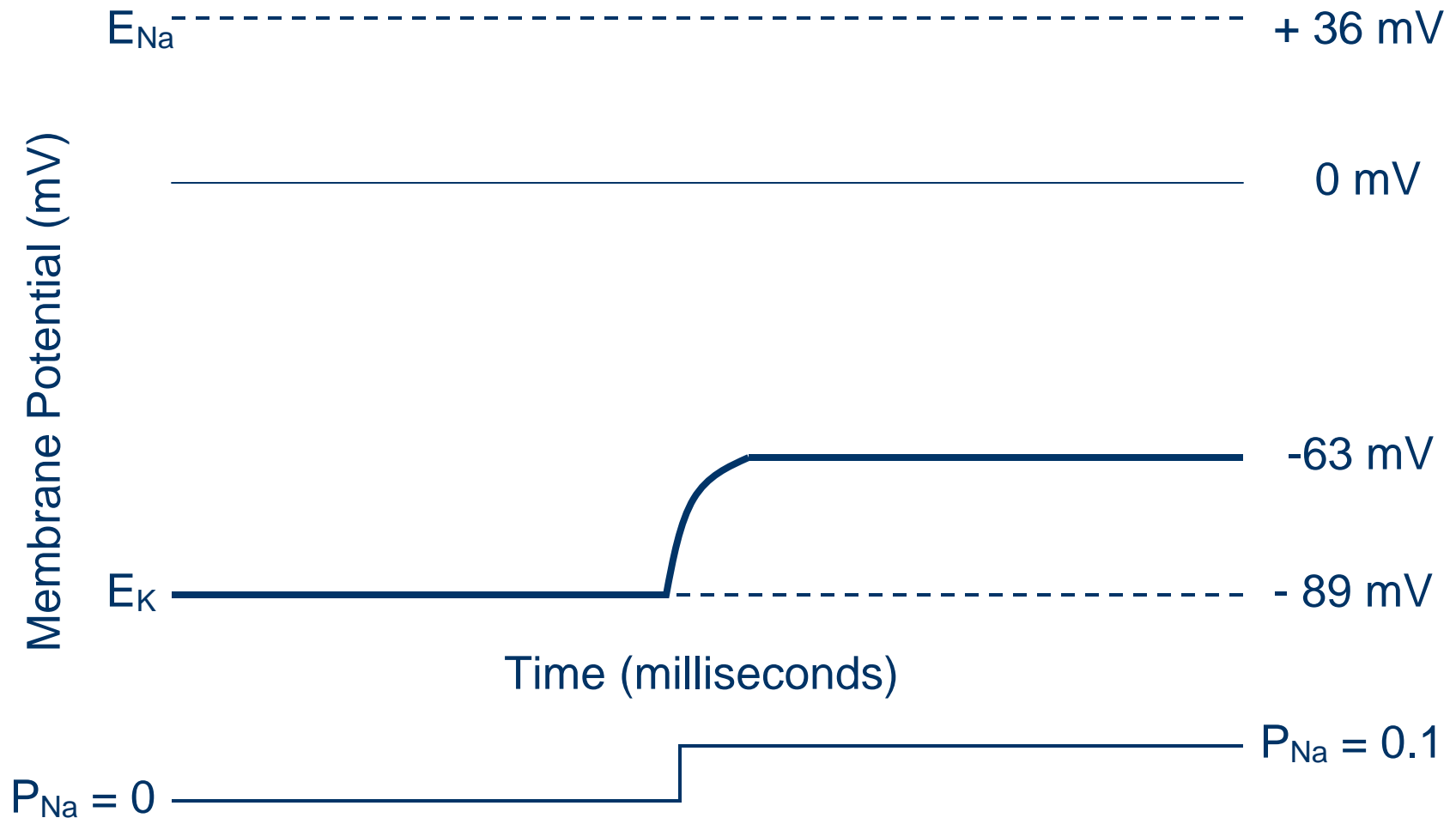
Suppose $P_K : P_{Na} : P_{Cl} = 1 : 0.1 : 1$

$$V_m = 60\text{mV} * \log\left(\frac{3 + 11.7 + 4}{90 + 3 + 120}\right)$$
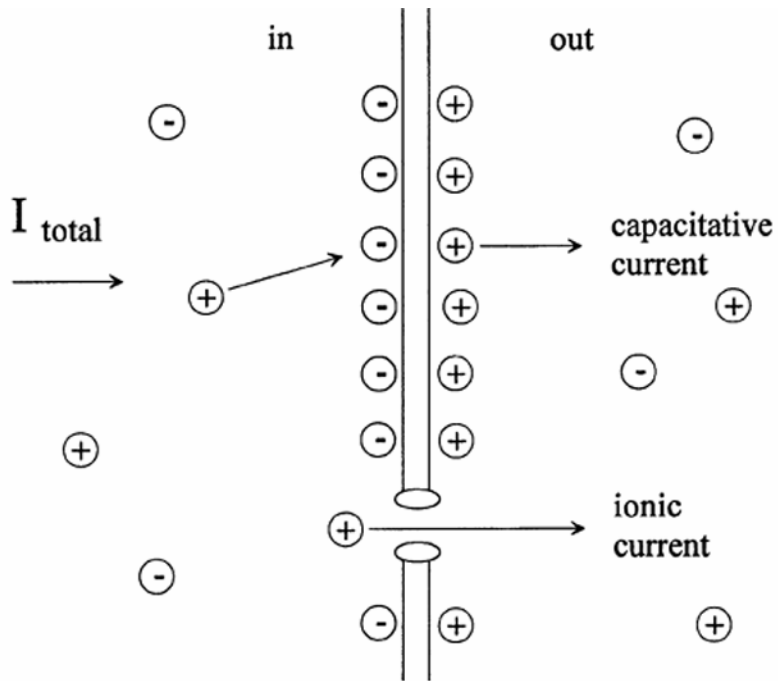
= 60 mV * log (18.7 / 213)

= 60 mV * -1.06

= -63 mV

Membrane Potential as a Function of Time
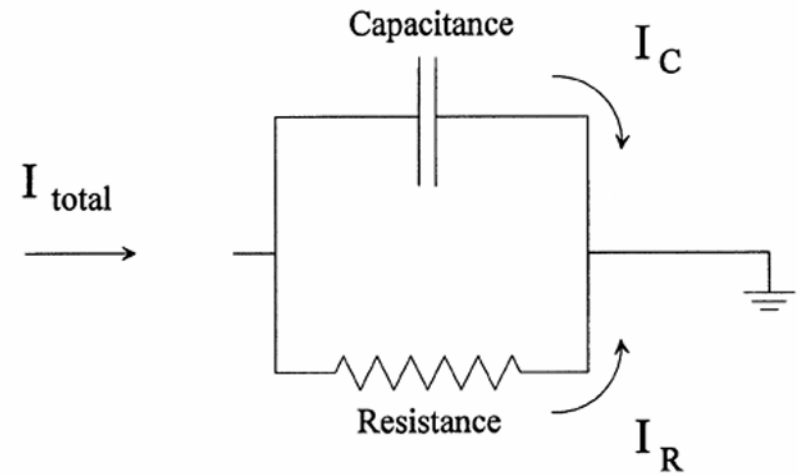

E_{Na} .... + 36 mV

0 mV

Membrane Potential (mV)

-63 mV

E_K .... - 89 mV

Time (milliseconds)

$P_{Na} = 0.1$

$P_{Na} = 0$

Changing Membrane Potential

in   out

I_{total}

capacitive
current

ionic
current

Capacitance I_C

I_{total}

Resistance I_R

Physical Model

Electrical Model

A simple model cell

A membrane sphere with conducting pores. The sphere contains, and is bathed in saline. The equivalent circuit is a capacitor and resistor in parallel. Inject a square pulse of current with a microelectrode, some of it will charge the capacitance and some will go through the resistance of the conducting pores.

$I_{Tot} = I_R + I_C$ where $I_R = V_m / R$ and $I_C = C * dV_m / dt$

$\Rightarrow dV_m / dt = (I_{Tot} / C) - (V_m / (R * C))$

The solution of this equation is: $V_m = I_{Tot} * R * (1 - \exp(-t / \tau))$
where $\tau = R * C$ is the membrane time constant

At equilibrium, when $t \gg \tau$ $V_m = I_{Tot} * R$
Where $R = 1 / \Sigma G_{pores}$ is called the Input Resistance

When $t = \tau$ then $(1 - \exp(-t / \tau))$ becomes $(1 - \exp(-1)) = 0.632$

Sample Calculation

A spherical cell, 100 µm in diameter, that has 200 open channels, each with a conductance of 10 pS.

Surface area of a sphere is $A = 4 * \pi * r^2$

Area = $4 * 3.1416 * (50 \times 10^{-4} \text{ cm})^2$ = $3.1 \times 10^{-4} \text{ cm}^2$

C = 3.1×10^{-4} µFarads

G_{Tot} = $200 * 10$ pS so $R_{in} = 1 / G_{Tot}$ = $5 \times 10^8 \ \Omega$ and

τ = $R_{in} * C = 155$ msec

A square pulse of current, 5 pA for 1 second will produce a change in membrane potential that will reach a steady state value of

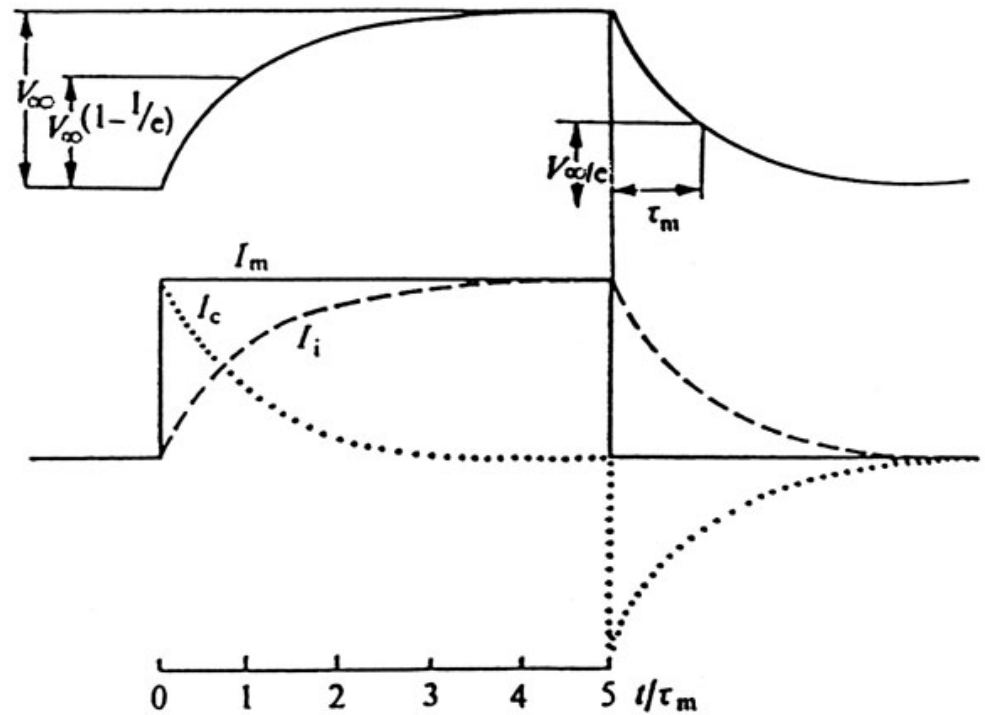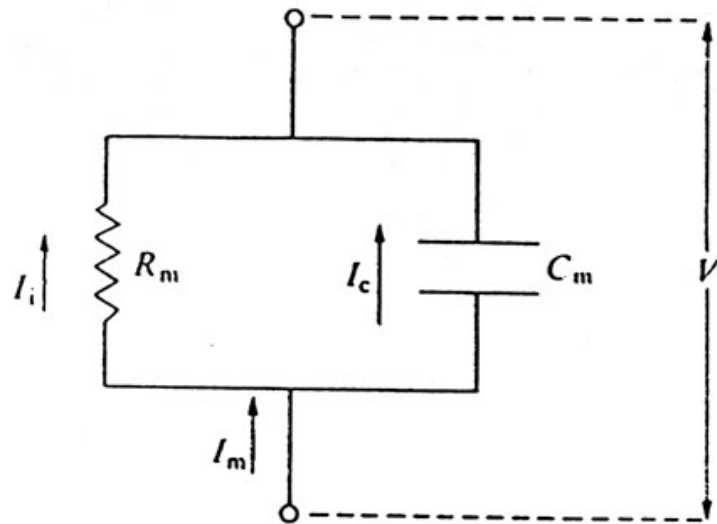$V = I * R$ = $5 \times 10^{-12} \text{ A} * 5 \times 10^8 \ \Omega$ = 2.5 mV
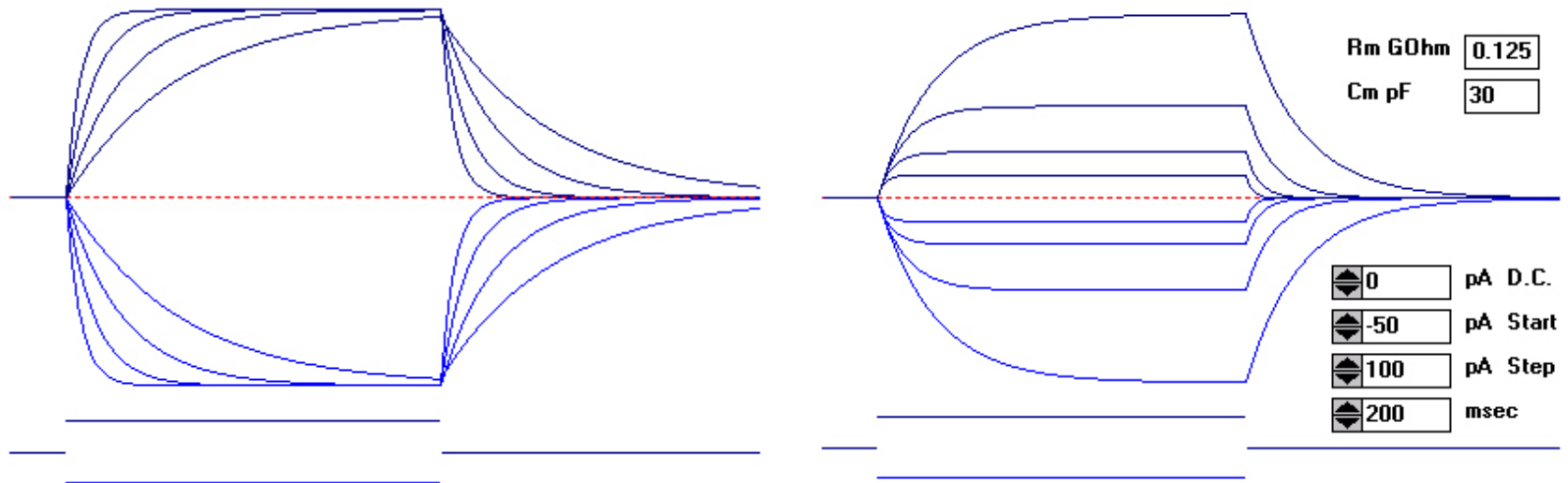
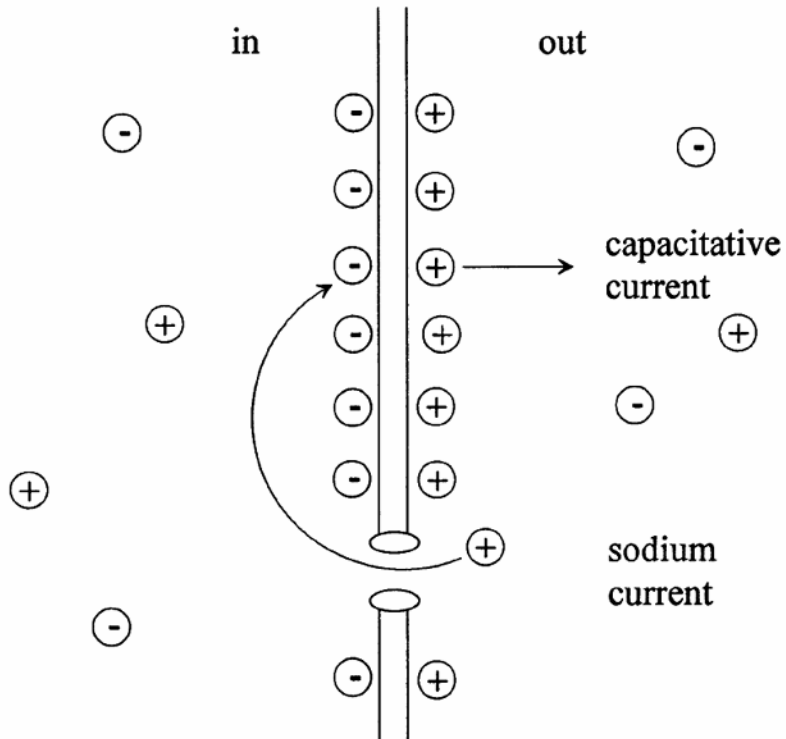# Plot of Potential versus Time
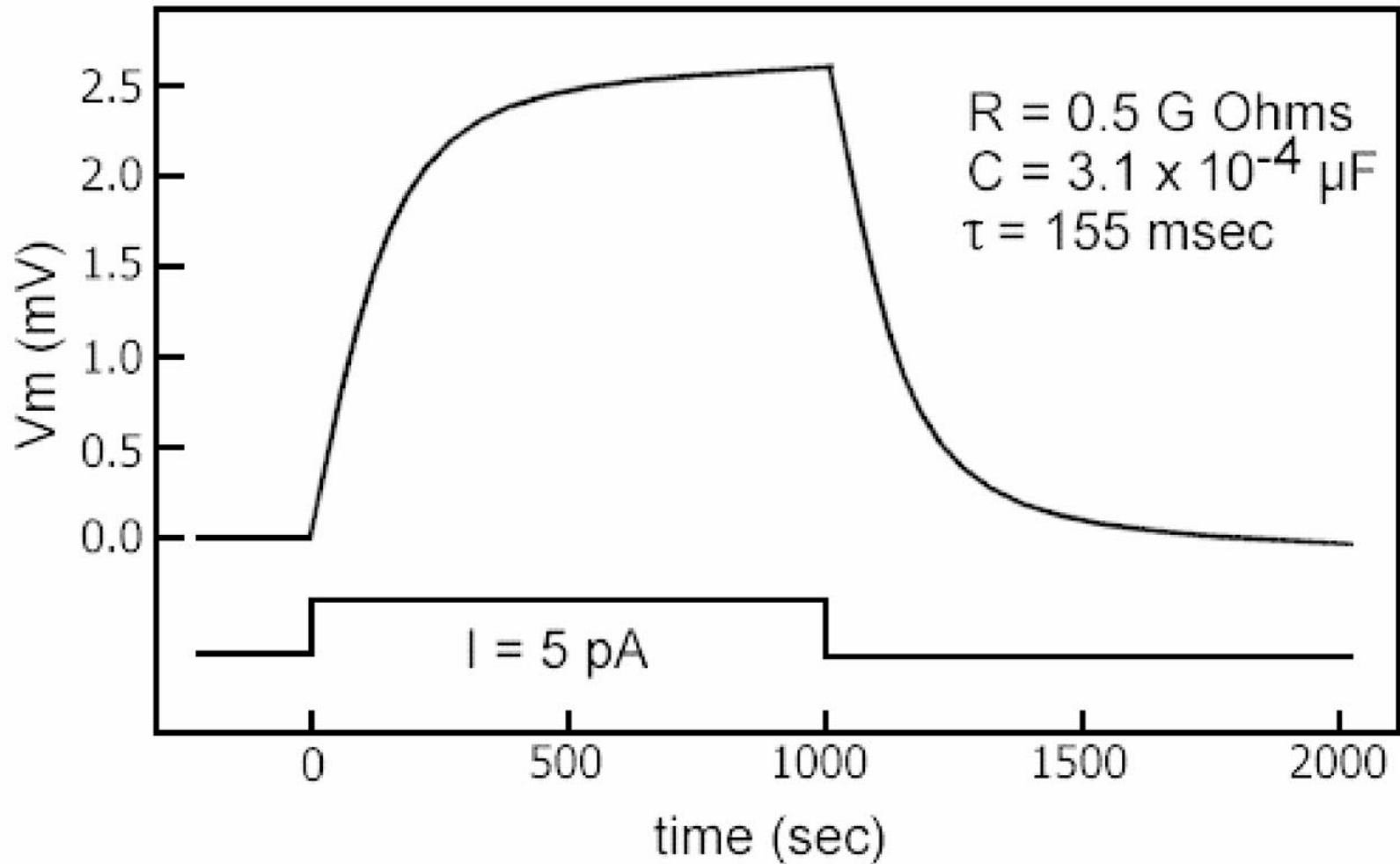
Ionic and capacitative current

changing C versus changing R

R=1; C=7.5, 15, 30, 60   C=30; R = 0.125, 0.25, 0.5, 1
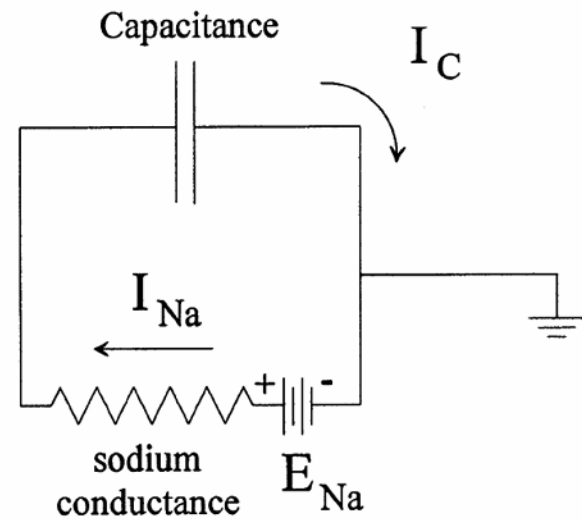

Rm GOhm 0.125
Cm pF 30

0 pA D.C.
-50 pA Start
100 pA Step
200 msec

Equivalent Circuit for Sodium Entry

in   out

capacitative
current

sodium
current

Capacitance I_C

I_{Na}

sodium
conductance E_{Na}

Physical Model   Electrical Model
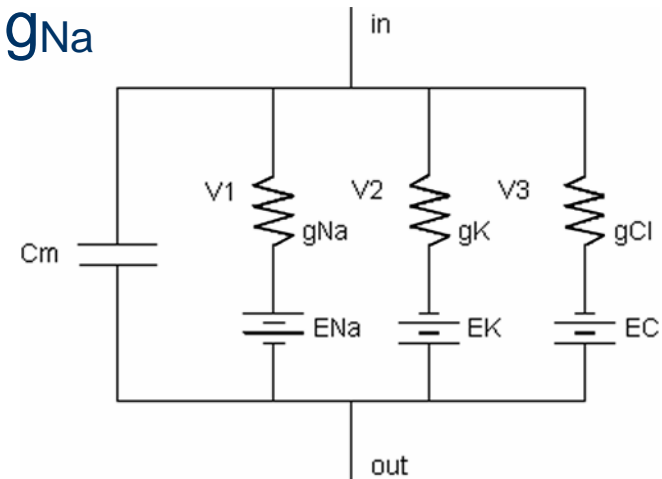
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Competing Batteries Model

Ohm's Law: $V = I * R$, so $I_{Na} = V_1 * g_{Na}$

$I_K = V_2 * g_K \qquad I_{Cl} = V_3 * g_{Cl}$

At rest there is no net current

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

$$\Rightarrow V_1 * g_{Na} + V_2 * g_K + V_3 * g_{Cl} = 0$$

V_m or $V_{rest} = V_1 + E_{Na} = V_2 + E_K = V_3 + E_{Cl}$

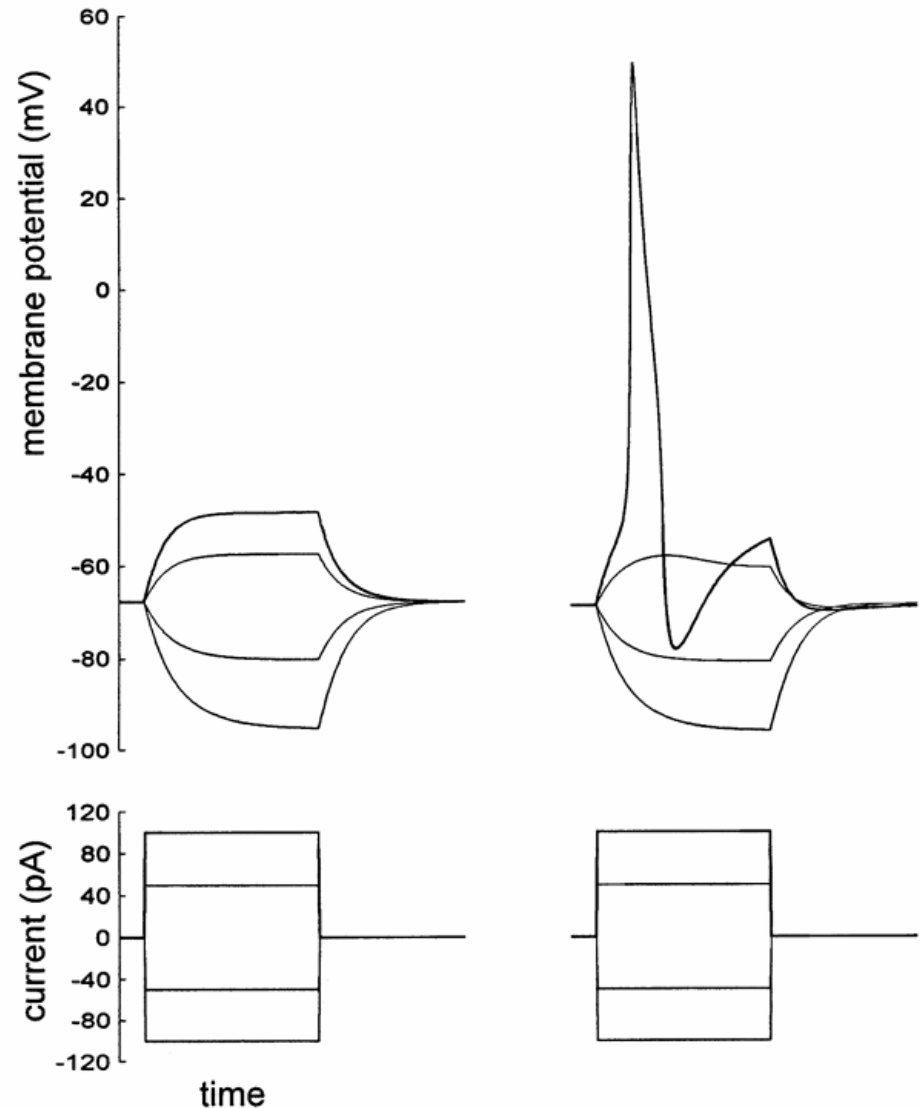$\Rightarrow V_1 = V_m - E_{Na}$ and $V_2 = V_m - E_K$ and $V_3 = V_m - E_{Cl}$

$\Rightarrow 0 = (V_m - E_{na}) * g_{Na} + (V_m - E_K) * g_K + (V_m - E_{cl}) * g_{Cl}$

$$V_m = \frac{g_{Na}E_{Na} + g_K E_K + g_{Cl}E_{Cl}}{g_{Na} + g_K + g_{Cl}} \qquad I_{ion} = g_{ion} * (V_m - E_{ion})$$
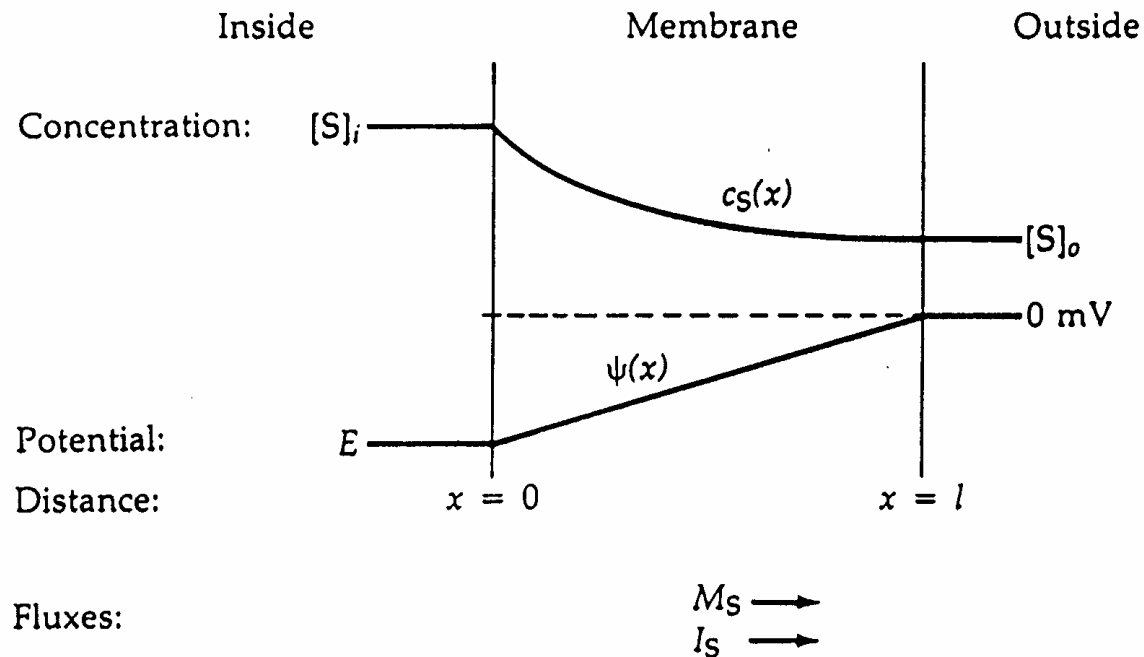
Passive v.s. Active

- All cells exhibit passive changes in membrane potential when stimulated

- Only excitable cells fire action potentials

- Excitability depends on specialized channels

GHK constant field theory

Nernst-Plank electrodiffusion equation

$$I = -z\,F\,D\,((dc/dx)+(F\,z\,c\,/\,(R\,T)))\,/\,(R\,T)\,(d\psi\,/dx)$$

Inside	Membrane	Outside

Concentration: $[S]_i$

$c_S(x)$

$[S]_o$

0 mV

$\psi(x)$

Potential: E

Distance: $x = 0$ $x = l$

Fluxes: $M_S \longrightarrow$

$I_S \longrightarrow$

2 ELECTRODIFFUSION IN A MEMBRANE

The flow of ion S across a hypothetical membrane is associated with a chemical flux M_S and an electric current I_S. The ionic concentration profile in the membrane is $c_S(x)$ and the electrical potential profile is $\psi_S(x)$.
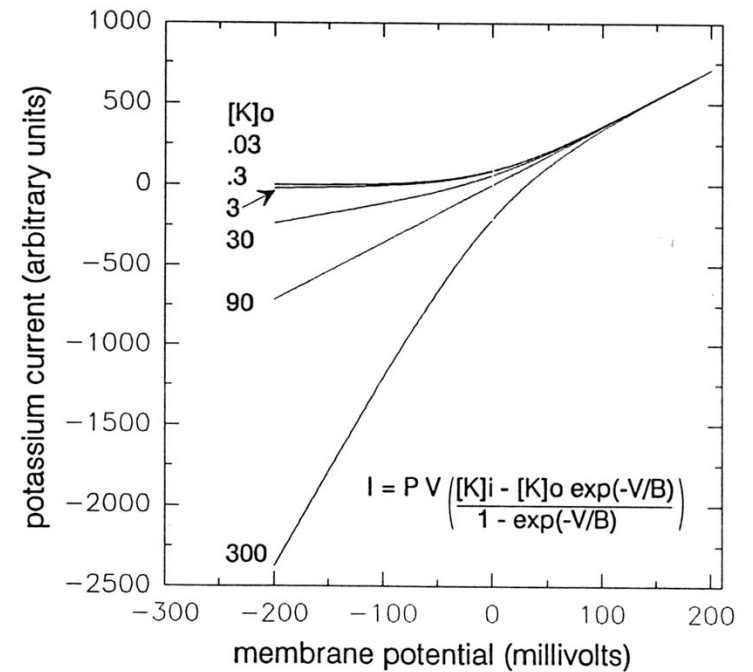
GHK constant field equation

(A) UNIDIRECTIONAL FLUXES

(B) CONCENTRATION DEPENDENCE
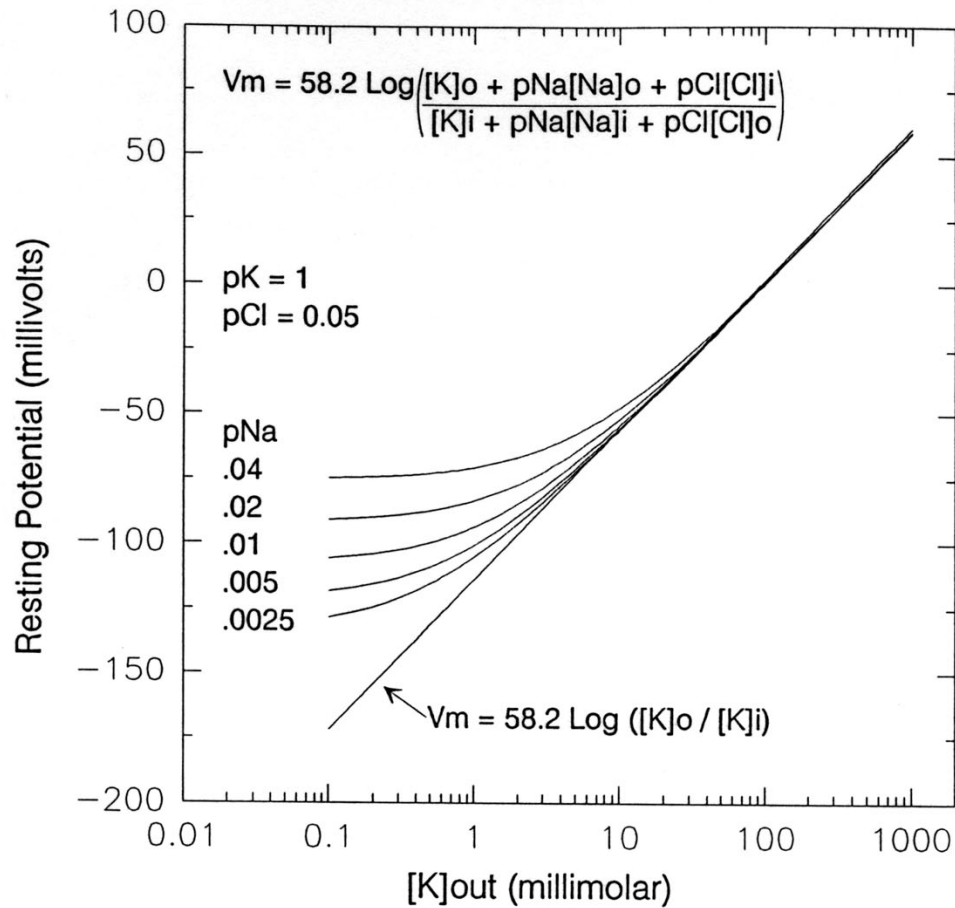
$\dfrac{[S]_o}{[S]_i} = 8$

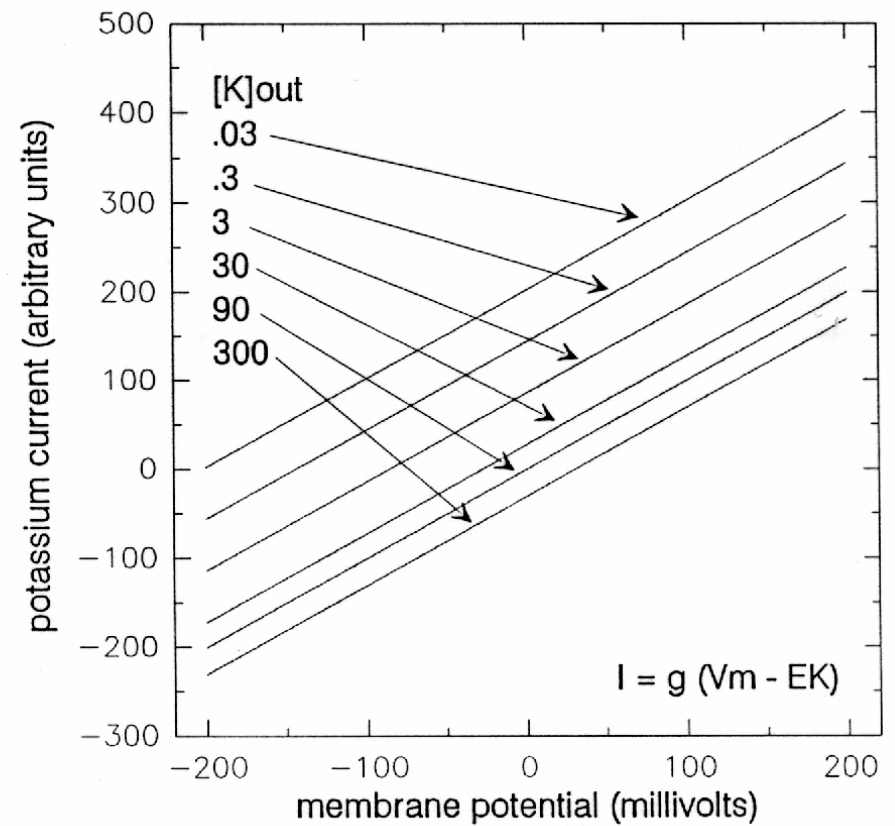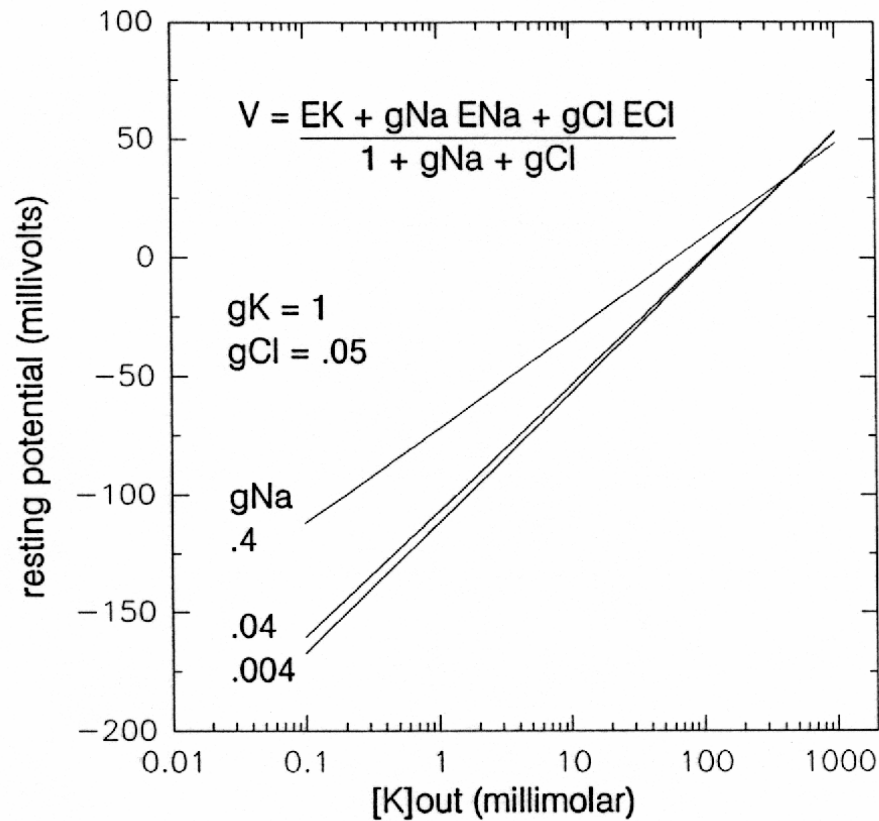$\dfrac{[S]_o}{[S]_i} = 16$
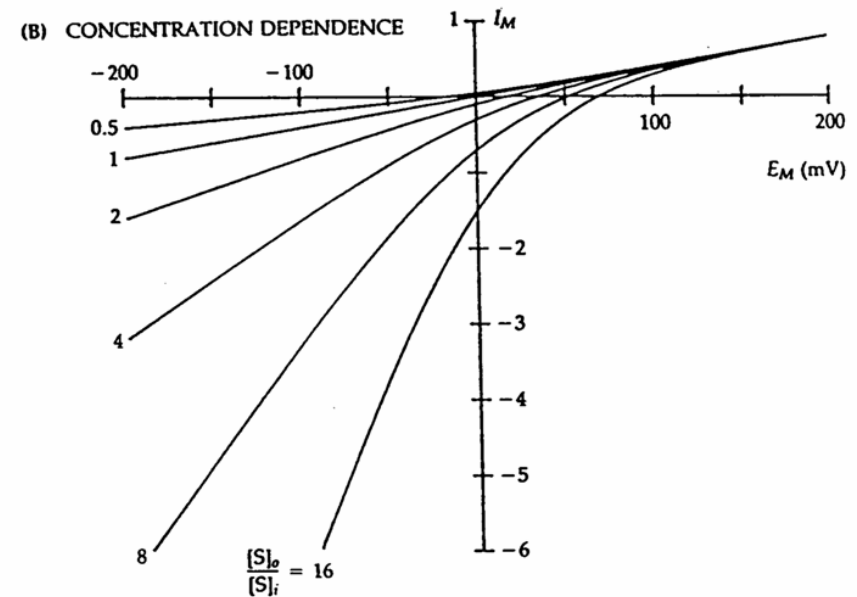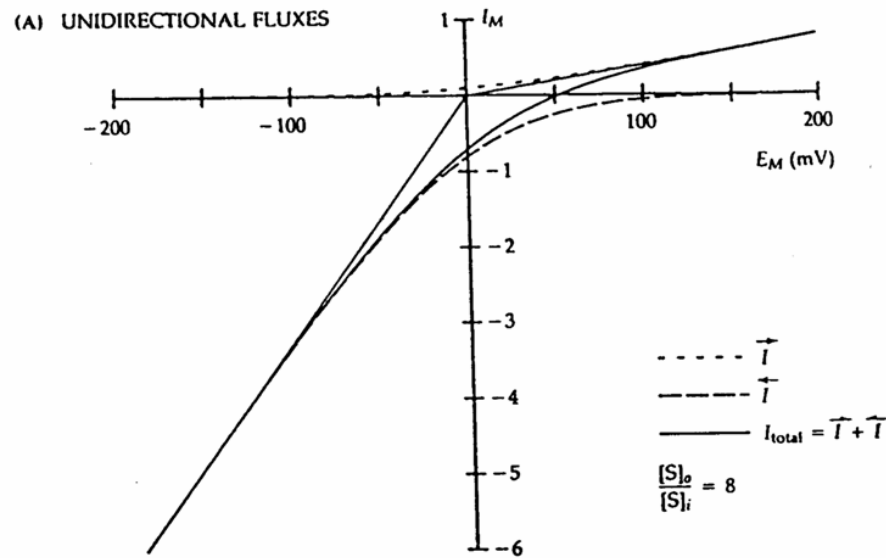
2   CURRENT-VOLTAGE CURVES OF GHK THEORY

Theoretical I–E relations for a homogeneous membrane obeying the Goldman (1943) and Hodgkin and Katz (1949) current equation for a single permeant, univalent cation. (A) Eightfold rectification with an eightfold concentration gradient, showing how asymptotes extrapolate to the origin and showing the underlying unidirectional efflux and influx making up the total current. (B) Change of curvature and of reversal potential as the external concentration is varied from 0.5 to 16 while the internal concentration is kept constant at 1. (Current and concentration in arbitrary units.)

$$\text{Current} = \frac{P\, z^2\, E\, F^2}{R\,T}\ \frac{([S]i - [S]o\, \exp(-z\,F\,E\,/\,R\,T))}{(1-\exp(-z\,F\,E\,/\,R\,T)}$$

GHK Equations

Left figure axes: Resting Potential (millivolts) vs [K]out (millimolar)

$$V_m = 58.2 \, \text{Log}\left(\frac{[K]o + pNa[Na]o + pCl[Cl]i}{[K]i + pNa[Na]i + pCl[Cl]o}\right)$$

pK = 1
pCl = 0.05

pNa
.04
.02
.01
.005
.0025

$$V_m = 58.2 \, \text{Log} \, ([K]o \, / \, [K]i)$$

Right figure axes: potassium current (arbitrary units) vs membrane potential (millivolts)

[K]o
.03
.3
3
30
90
300

$$I = P \, V \left(\frac{[K]i - [K]o \exp(-V/B)}{1 - \exp(-V/B)}\right)$$

Linear Competing Batteries

$$V = \frac{EK + gNa\ ENa + gCl\ ECl}{1 + gNa + gCl}$$

gK = 1
gCl = .05

gNa
.4

.04
.004

$I = g\ (Vm - EK)$

[K]out
.03
.3
3
30
90
300

9/6/2005

Summary:

I. Cell membranes form an insulating barrier that acts like a parallel plate capacitor (1 µF /cm^2)

II. Only a small number of ions must cross the membrane to create a significant voltage difference

   $\Rightarrow$ bulk neutrality of internal and external solution

III. Cells need channels to regulate their volume

IV. Permeable ions move toward electrochemical equilibrium

   - E_{ion} = (60 mV / z) * log ([Ion]$_{out}$ / [Ion]$_{in}$) @ 30°C
   - Electrochemical equilibrium does not depend on permeability, only on the concentration gradient

Summary (continued):

V. The Goldman, Hodgkin, Katz equation gives the steady-state membrane potential when Na, K and Cl are permeable

$$V_m = 60mV * \log\left(\frac{P_K * [K]_{out} + P_{Na} * [Na]_{out} + P_{Cl} * [Cl]_{in}}{P_K * [K]_{in} + P_{Na} * [Na]_{in} + P_{Cl} * [Cl]_{out}}\right)$$

- In this case, V_m does depend on the relative permeability to each ion and there is steady flux of Na and K

  $\Rightarrow$ The cell must supply energy to maintain its ionic gradients