离子通道生物学

细胞的跨膜信号转导

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Discuss about the definition of this course

“Ion channelology”

Schedules

2/28 细胞膜电学性质、离子通道的分子结构与功能

3/6 Ca^{2+}通道

3/13 Na^{+}通道

3/20 K^{+}通道 (SK, BK channels)

3/27 Cl^{-}通道、配体门控的通道

4/10 细胞内Ca^{2+}释放通道

4/17 离子通道疾病及其分子基础（一）

4/24 离子通道疾病及其分子基础（二）

5/8 膜片钳技术的理论

5/15 膜片钳技术的实践

Score

平时成绩：

课前预习，课堂发言、讨论，课后作业

考试成绩：

- ？闭卷
- ？开卷

References

- Patch clamping: an introductory guide to patch clamp electrophysiology, Areles Molleman, 2003 - 175 pages (ebook)
- Patch-clamp applications and protocols, Alan A. Boulton, Glen B. Baker, Wolfgang Walz - 1995 - 316 pages
- Electrical properties of cells: patch clamp for biologists, Louis J. DeFelice - 1997 - 244 pages
- Patch-clamp analysis: advanced techniques, Wolfgang Walz, Alan A. Boulton, Glen B. Baker - 2002 - 345 pages
- Ion channels: a practical approach, Richard H. Ashley - 1995 - 302 pages
- Ion channels: molecules in action, David J. Aidley, Peter R. Stanfield - 1996 - 307 pages
- Introduction to electrophysiological methods and instrumentation, Franklin Bretschneider, Jan R. De Weille - 2006 - 251 pages

References

- The ion channel factsbook: Voltage-gated channels, Edward C. Conley, W. J. Brammar - 1999 - 860 pages
- Ion channels and genetic diseases, David C. Dawson, Society of General Physiologists. Symposium, Raymond A. Frizzell - 1995 - 178 pages
- Ligand- and voltage-gated ion channels, R. Alan North - 1995 - 349 pages
- Ion channels of excitable membranes, Bertil Hille - 2001 - 814 pages
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- Eur Biophys J 2004, 33, 211-26 ion channels.pdf
- J Mol Cell Cardiol 2010, 48, 65-70 Cardiac Ca channel.pdf
- Pflugers Arch-Eyr J Physiol 2009, Cardiac Na channel.pdf
- Nature Rev Neurosci 2007, 8, 451-65 AP.pdf
- Trends in Neuroscience 2009, 32, 215-24 TRPV channel.pdf

References

- Kartik Venkatachalam, Craig Montell (2007) TRP Channels. Annual Review of Biochemistry, 76: 387-417
- Progress Biophysics Mole Bio 2009, 99, 7-19
- Nat Rev Drug Discov 2009, 8, 153-71
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- 心肌细胞离子通道和通道病, 刘泰槿编著, 人民卫生出版社, 2006年
- 离子通道药理学, 杨宝峰 主编, 人民卫生出版社, 2005年
- 膜片钳实验技术, 陈军, 电子书
- 心肌细胞电生理学：离子通道，离子载体和离子流，刘泰槿编著, 人民卫生出版社, 2005, 252页
- 细胞膜离子通道, 刘安西，陈守同编译, 中央民族学院出版社, 1990年
- 离子通道与疾病, 张宗明, 裘法祖. 世界华人消化杂志, 2005, 13: 585-587

Vital roles of ion channels

- In basic physiological functions
  - generation of electrical activity in nerves and muscle
  - control of cardiac excitability
  - intracellular signaling
  - hormone secretion
  - cell proliferation
  - cell volume regulation
  - many other biological processes
- In a number of pathophysiological conditions
  - epilepsy
  - episodic ataxia
  - long QT syndrome
  - Bartter’s syndrome
  - cystic fibrosis

Vital roles of ion channels

- Ion channels are drug targets for a number of therapeutic agents developed for the treatment of various diseases.
  - Na^+ channel blockers such as lidocaine and lamotrigine are used as local anesthetic and to treat epilepsy, respectively
  - Calcium channel blockers such as verapamil and nifedipine are used for the treatment of cardiac arrhythmias and hypertension
  - ATP-dependent K^+ channel inhibitors such as tolbutamide and glibenclamide are effective in the treatment of type II diabetes

The history of ion channel

- We now know that, ions are not only possible, but a common everyday experience, especially in aqueous solutions such as those which surround living cells. The concept of an *ion channel* had to await the acceptance by the scientific community of the existence of ions as well as the notion that cells are surrounded by a fatty membrane bilayer.
- The late 1700s, Lavisouir and others, physics, chemistry and biology
- The emergence of both ion and cell membrane concepts can be traced back to the late 1800s.

The mystery which remained throughout most of the 1800s

- Were nerve cells analogous to small copper wire conductors?
- Was the situation more complex, such as the way two different chemicals were able to create electricity; as in a battery?

- It would turn out to be the chemical explanation that was the correct one (i.e. ion flow in and out of nerve and muscle cells through ion channels).

Early Ion Channel Work

- In 1759, Michel Adanson was the first to propose that the newly invented Leyden jar capacitor was identical in principle to the way an electric eel stores electricity within itself.
- Not long afterwards, Volta, the inventor of the battery, noticed that there existed similarities in organization between the fish's electric organs and the batteries he was the first to build.
- When Galvani published his results using electrical current in 1791 to make a dead frog's legs twitch, it encouraged many to try and explain how electrical current helps nerve cells transmit messages to muscles, as well as encouraging a cottage industry of medical frauds intent on using electricity to "cure" all types of ills.

Early Ion Channel Work

- The 19th Century, Luigi Galvani publicized to the world that electricity could induce a biological response in a frog's leg, in a way it could be said that he was studying ion channels for the first time, albeit indirectly.
- As early as 1817 the great Swedish chemist Berzelius suggested that the electric eel's current was "elicited by an organic chemical process."
- Italian physicist Leopoldo Nobili, in 1827, using his newly refined galvanometer which corrected for the earth's magnetic field, was the first to report measuring the current in a frog using any kind of an instrument.

Carlo Matteucci (1811–1868)

- An Italian physicist and neurophysiologist who was a pioneer in the study of bioelectricity.
- Using a galvanometer, an instrument that can detect and measure small amounts of currents, Matteucci was able to prove that injured excitable biological tissues generated direct electrical currents.
- He was the first to demonstrate that it was possible to induce muscle contraction by means of an action potential, and that action potentials were associated with depolarization of the muscle resting potential.

Carlo Matteucci (1811–1868)

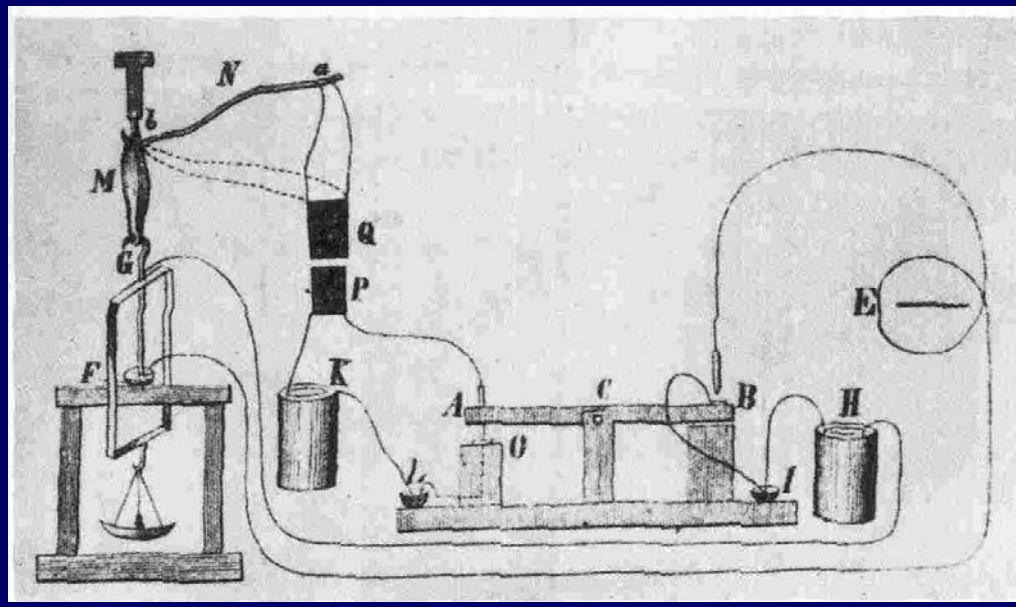
- In 1838 Carlo Matteucci measured current (which we now know to be ions) which leaking out of injured muscles, and by the century's end would form the basis for the important concept of the "resting potential" of both muscle and nerve cells.
- Matteucci also went on to discover that muscle cells, after stimulation by a nerve, produce a current of their own.
  - He used an ingenious detector called the “galvanic frog”, which was simply a severed leg from a frog with the nerve exposed which would twitch each time it detected a current. This muscle-generated current is now known as the "action potential".
- Matteucci would also discover that he could simply change the pH of the solution surrounding the muscle tissue and it would illicit a contraction in the muscle just as the nerve did.

Emil du Bois-Reymond (1818–1896)

- A German physiologist, the founder of modern electrophysiology
- Using a galvanometer, du Bois-Reymond detected the flow of charges through all muscular and nervous tissue.
- He developed the view that a living tissue, such as muscle, might be regarded as composed of a number of "electric molecules" having certain electric properties, and that the electric behavior of the muscle as a whole in varying circumstances was the outcome of the behavior of these native electric molecules.
- His research established electrophysiology as a scientific discipline.

Early Ion Channel Work

- **In Galvani's time**, electricity was still thought of as a kind of "fluid", and this line of reasoning allowed for the ancient Roman belief that hollow nerve cells transmitting some sort of fluid messenger to other parts of the body.
- **The late 1800s:** The first **quantitative measurements** involving electrical activity in animal cells
- **In 1850, Helmholtz** succeeded in determining the **velocity** of the electric signal on a nerve cell and realized that it traveled much too slowly to be due simply to conduction such as occurs in a metal wire.



Sidney Ringer (1836–1910)

- A British clinician and pharmacologist, serendipitously discovered that Ca^{2+} was active in the heart, and performed a completely novel function: it carried the signal that initiated heart contraction.
- Ringer was able to show that adding small amounts of potassium chloride to a normal solution of sodium chloride allowed isolated organs to stay functional for longer periods of time.
- Ringer's papers published in the *Journal of Physiology in the early* 1880s are rightly acknowledged as the starting point for the development of modern understanding of the role of calcium in the contraction of the heart.

Wilhelm Ostwald

- In 1890 Wilhelm Ostwald proposed that the electrical signals measured in living tissue could be caused by ions moving in and out through cell membranes.

- Baltic German chemist
- Sept. 2, 1853 – Apr. 4, 1932
- Nobel Prize in Chemistry 1909

In the 20th century

- **In 1902, a German physiologist named Julius Bernstein (1839–1917) correctly proposed that excitable cells were surrounded by a membrane selective to K^+ ions at rest, and that during excitation the membrane became permeable to other ions. His hypothesis and research laid the foundation for understanding conduction of the nerve impulse and electrical transmission of information in the nervous system.**
- **In 1907, the British physiologist John Newton Langley (1852–1925) introduced the concept of receptor molecules on the surfaces of nerve and muscle tissue, in an attempt to explain the specific and potent actions of certain chemicals on muscle and nerve cells. Langley's theories were much debated at the time, and receptors remained theoretical until their discovery in the 1940s.**

The discovery of ion channels

- The notion of the existence of some type of narrow ion channel arose in the 1920s. The development of the thermionic values and the cathode ray oscilloscope, which would allow greater time-resolution of the experiment as well as greater reproducibility between different experiments.
- As surprisingly accurate as these early results would be, the ability to measure the voltage of a cell membrane directly would remain impossible without electrodes small enough to insert directly inside the cell itself.

In the 20th century

- **In 1937, John Zachary Young (1907–1997) was one of the first to make use of squid neurons to study ionic currents. The ease of working with large neurons made important experiments possible for the first time, including the first intracellular recordings of the nerve cell action potential, as well as the first measurements of the underlying ionic currents that produce them. His discovery and work with giant squid axons eventually led to the award of the Nobel Prize to Alan Hodgkin and Andrew Huxley.**
- **The appearance of true cellular electrophysiology followed the introduction, in 1949, of intracellular glass microelectrodes by Gilbert Ling and Ralf Gerard. With this invention, it became possible to detect and measure the resting potential of a cell by impaling its membrane, without destroying the cell.**

In the 20th century

- Using squid neurons, Alan Lloyd Hodgkin and Bernard Katz (1949) removed sodium ions from outside the neuron and were able to conclude from their data that sodium ions were responsible for the formation of the action potential.
- The next improvement in instrumentation took place around the same period when Kenneth Cole and George Marmont described the concept of the voltage clamp method. This approach consisted of placing a second glass electrode inside the cell in order to stabilize or "clamp" the membrane potential of neurons for experimental purposes.
- Voltage clamping allowed measurements of the effect of changes in membrane potential on the conductance of this membrane to various individual ion species.

Hodgkin and Huxley

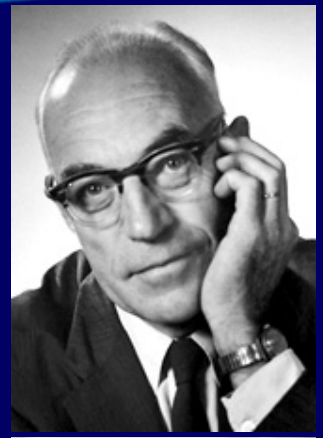
- In the early 1950s, two British scientists Alan Hodgkin and Andrew Huxley characterized the time and voltage dependency of the ionic conductances that underlie an action potential in the squid giant axon, using the voltage clamp technique, and developed a mathematical model that accurately predicted the waveform of the action potential.
- This was a major breakthrough. For this determining work on neuronal excitability, Hodgkin and Huxley received the Nobel Prize in Physiology or Medicine in 1963. They showed how ion transport through nerve cell membranes produces a signal that is conveyed from nerve cell to nerve cell like a relay race baton. It is primarily sodium and potassium ions, Na^+ and K^+, that are active in these reactions.

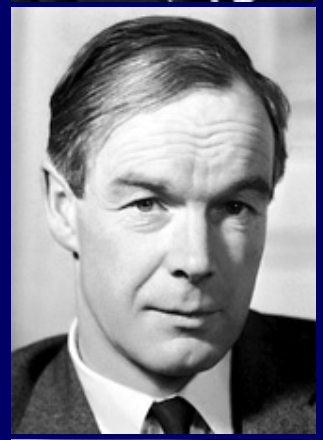
Nobel Prize – Ion channels

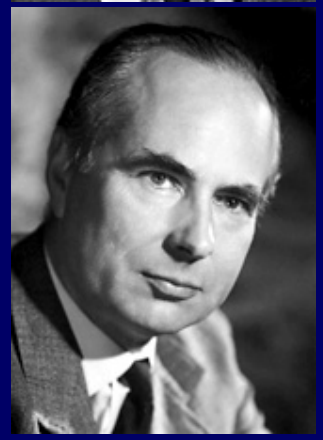
- 1963 Ionic mechanisms of nerve cell membrane
  - Eccles, John Carew
  - 1/27/1903 to 5/2/1997
  - Australian

  - Hodgkin, Alan Lloyd
  - 2/5/1914 to 12/20/1998
  - British

  - Huxley, Sir Andrew Fielding
  - 12/22/1917 to
  - British







Erwin Neher and Bert Sakmann

- in 1976, Erwin Neher and Bert Sakmann pressed a smooth electrode tip on the surface of an isolated skeletal muscle fiber, electrically isolating a patch of membrane and reducing extraneous electrical noise so low that picoampere currents flowing through a single ion channel could be measured directly.
- The patch clamp technique was born, and it quickly became the backbone of modern electrophysiology. It has since been referred to as the single most important development in ion channel research in the last half of the twentieth century.
- In 1991, Neher and Sackmann were rewarded the Nobel Prize for Physiology or Medicine for the development of the patch clamp technique.

Nobel Prize – Ion channels

- 1991 Function of single ion channels in cells
  - Neher, Erwin
  - 3/20/1944 to
  - German

  - Sakmann, Bert
  - 6/12/1942 to
  - German

Breakthroughs in other scientific areas

- **Clay Armstrong** in **1973** proposed that the **structure** of sodium channels in the squid neuron allows for opening and closing of their pores by a "**ball and chain**" model.
- The advent of **recombinant DNA technology** in the mid to late 1970s provided the means for obtaining sequence information about genes, proteins. With the amino acid sequences known, it was possible to begin predictions of what ion channels should look like in three-dimensional space. The sequence could be changed deliberately (site-directed mutagenesis) in order to understand how it worked.
- **Cloning** of the first ion channel (nicotinic acetylcholine receptor, nAChR) occurred in **1982**, and the first channel to be sequenced was the Na^+ channel, followed by the Ca^{2+} channel. Soon thereafter, the first K^+ channel was characterized.

Breakthroughs in other scientific areas

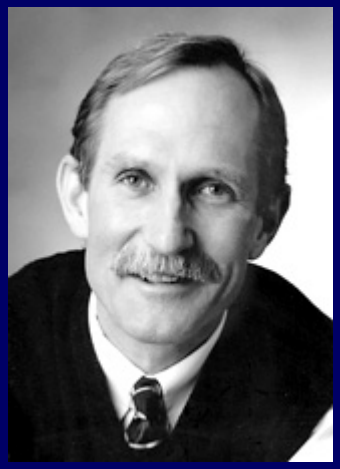
- Introduction of recombinant expression of ion channels provided a well-defined, replenishable source of cells expressing a variety of human ion channels, opening the way to a number of biochemical assays to study ion channel function.
- There is little doubt that combining the information and knowledge acquired from electrophysiology, molecular biology, crystallography, and other techniques has greatly improved the understanding of ion channel structure and function, and heightened the interest in developing novel ion channel modulators.

Roderick MacKinnon

- The first high resolution crystal structure of an ion channel (3.2Å resolution), the potassium channel *KscA from a bacteria, was provided by* MacKinnon and his group. This meant that it was possible to determine the exact positions of nearly all of the individual atoms in the protein.
- His work revealed for the first time the inner workings of an ion channel at the atomic level. It provided tremendous insight into the selectivity of potassium channels by revealing rings of carboxyl oxygen atoms in the narrow selectivity filter that stabilize the potassium ions as they travel in the water-filled cavity in the center of the channel, and shed their water molecules.
- For this astonishing breakthrough, MacKinnon was awarded the Nobel Prize in Chemistry in 2003.

Nobel Prize – Ion channels

- 2003 Structural and mechanistic studies of ion channels
  - Agre, Peter
  - 1/30/1949 to
  - American



  - MacKinnon, Roderick
  - 2/16/1956 to
  - American

Planar patch electrophysiology

- **In 2003, the first automated parallel patch clamp system rolled off the assembly line, revolutionizing the high throughput screening (HTS) approach to ion channel drug discovery. Solutions to the problem of internal perfusion in patch clamping led to the first prototype of planar electrodes.**
- **Planar patch electrophysiology uses a planar arrangement of micron-sized holes in a glass or plastic substrate to replace the glass electrode or pipette used in conventional patch clamping. This prompted regulatory agencies around the world to require preclinical testing of all new drugs to determine their propensity to block channels.**
- **Only a decade after the initial concept, planar patch clamping is already an established technology with numerous commercially available devices producing high quality data in HTS format.**

Top ion channel laboratories

- Dr. WA Catterall
- Dr. R MacKinnon
- Dr. M Lazdunski
- Dr. DE Clapham
- Dr. RA Nicoll
- Dr. SG Waxman
- Dr. B Nilius
- Dr. RL Huganir
- Dr. B Sakmann
- Dr. LY Jan
- Dr. MJ Welsh
- Dr. RW Tsien
- Dr. FJ Sigworth
- Dr. TJ Jentsch
- Dr. JP Changeux
- Dr. RC Malenka
- Dr. AM Brown
- Dr. M Sheng
- Dr. SH Snyder
- Dr. OP Hamill

Top ion channel companies

The Ion Channel Experts
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High throughput patch clamping for ion channel drug screening
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Includes experiments from 1932 that discusses theories of conditioning
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Protein & Pab for PDGFRB
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SinoBiological.com/PDGFRB_Protein

Label-free Ion Channel
Cell-based Functional Assays for Ion Channels, 96 - 1536-well Format
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Channelrhodopsin Optical switching by UHP-Fiber-Coupled-LED
prizmatix.com/Ultra-High-PwrLED-460

Custom Antibodies
High-Quality Research Antibodies. Monoclonals Antibody Service $3,900
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Antibodies
Covance, The Leading Provider of Quality Immunological Reagents
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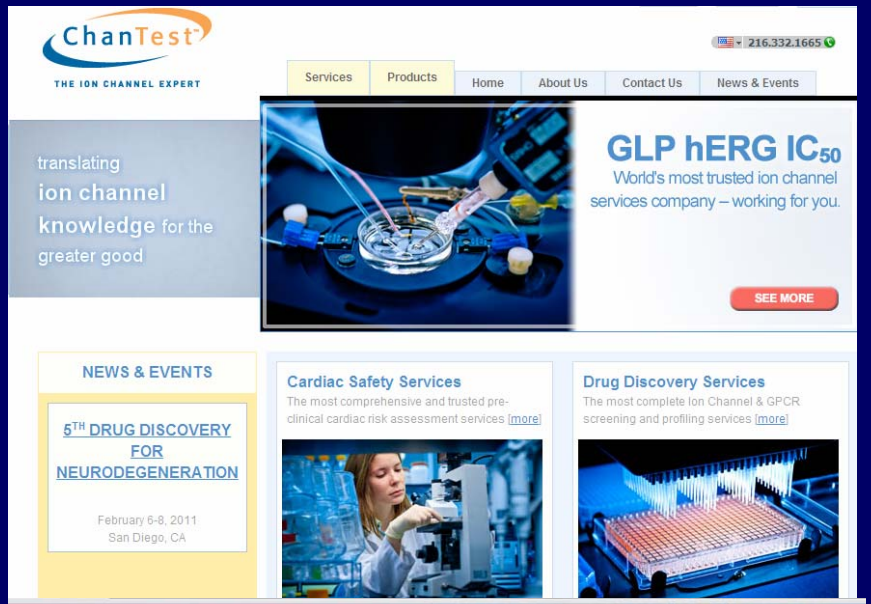


The outline…

Required readings:

Boron & Boulpaep “Medical Physiology” (2nd Edition), 2009 Chapter 6

Physiol Rev 2008, 88(4), 1407–1447 CNS Ion channels

Trimmer JS, Rhodes KJ. (2004) Localization of voltage-gated ion channels in mamalian brain. Annu Rev Physiol. 66, 477-519

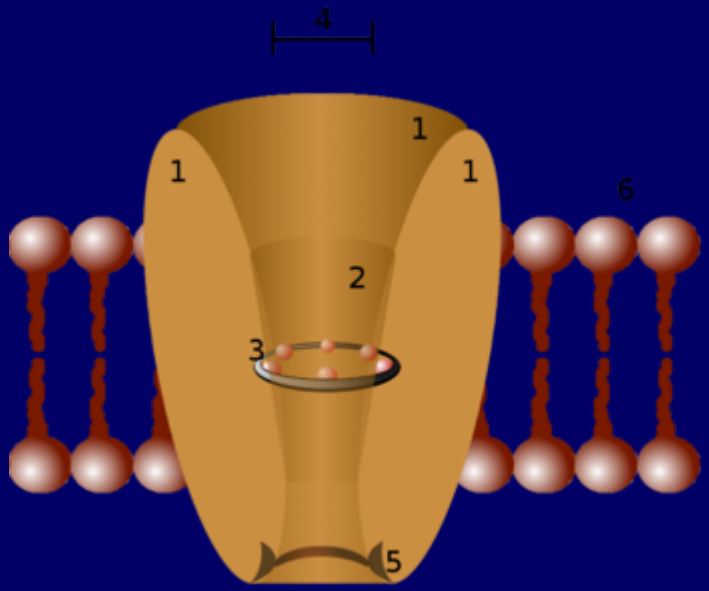
Fermini B (2008). Recent advances in ion channel screening technologies. Top Med Chem. 3: 1–25

This class will cover:

1. Basic principles of electricity
2. Essentials of electrophysiology
3. Molecular biology of ion channels
4. Overview of channel structure and function

Ion Channels

- **Pore-forming proteins** that help establish and control the small voltage gradient across the plasma membrane of all living cells by allowing the flow of ions down their electrochemical gradient
- Present in the membranes that surround all biological cells
- The study of ion channels involves many scientific techniques such as patch clamp, immunohistochemistry, and RT-PCR



Basic features

- Ion channels regulate the flow of ions across the membrane in all cells.
- Ion channels are integral membrane proteins; or, more typically, an assembly of several proteins. Such "multi-subunit" assemblies usually involve a circular arrangement of identical or homologous proteins closely packed around a water-filled pore through the plane of the membrane or lipid bilayer.
- For most voltage-gated ion channels, the pore-forming subunit(s) are called the α subunit, while the auxiliary subunits are denoted β, γ, and so on. Some channels permit the passage of ions based solely on their charge of positive (cation) or negative (anion).

Basic features

- The archetypal channel pore is just one or two atoms wide at its narrowest point and is selective for specific species of ion, such as sodium or potassium. These ions move through the channel pore single file nearly as quickly as the ions move through free fluid.
- In some ion channels, passage through the pore is governed by a "gate," which may be opened or closed by chemical or electrical signals, temperature, or mechanical force, depending on the variety of channel.

Biological role

- Because "voltage-activated" channels underlie the nerve impulse and because "transmitter-activated" channels mediate conduction across the synapses, channels are especially prominent components of the nervous system.
- Most of the offensive and defensive toxins that organisms have evolved for shutting down the nervous systems of predators and prey (e.g., the venoms produced by spiders, scorpions, snakes, fish, bees, sea snails and others) work by modulating ion channel conductance and/or kinetics.
- Ion channels are key components in a wide variety of biological processes that involve rapid changes in cells, such as cardiac, skeletal, and smooth muscle contraction, epithelial transport of nutrients and ions, T-cell activation and pancreatic β-cell insulin release.

Diversity

- Of the more than 400 ion channel genes currently identified in the human genome, about 170 encode potassium channels, 38 encode calcium channels, 29 encode sodium channels, 58 encode chloride channels, and 15 encode glutamate receptors.
- Ion channels may be classified by the nature of their gating, the species of ions passing through those gates, and the number of gates (pores).

By gating

-- what opens and closes the channels

- Voltage-gated
- Ligand-gated
- Stretch or Pressure-gated
- Phosphorylation gated
- Mechano-sensitive channels
- K^+ leak channels

Classification of the two major groups of ion channels

Ion channel	Selectivity	Activator
Voltage-gated channels		
Potassium	K^+	Membrane potential
Sodium	Na^+	Membrane potential
Calcium	Ca^{2+}	Membrane potential
Chloride	Cl^-	Membrane potential
HCN	Na^+, K^+	Membrane potential
Ligand-gated channels		
nAChR	Na^+, K^+, Ca^{2+}	Ach, nicotine
$GABA_{A,C}$	Cl^-	GABA
Glycine	Cl^-	Glycine, strychnine
$5\text{-}HT_3$	Na^+, K^+, Ca^{2+}	Serotonin
AMPA	Na^+, K^+, Ca^{2+}	Glutamate, AMPA
Kainate	Na^+, K^+, Ca^{2+}	Glutamate
NMDA	Na^+, K^+, Ca^{2+}	Glutamate, NMDA
CNG	Na^+, K^+, Ca^{2+}	cAMP
IP_3R	Ca^{2+}	IP_3
P2X, P2Z	Na^+, Ca^{2+}, Mg^{2+}	ATP

By ions

- Calcium channels
- Potassium channels
  - Voltage-gated potassium channels
  - Calcium-activated potassium channels
  - Inward-rectifier potassium channels
  - Two-pore-domain potassium channels
- Sodium channels
  - voltage-gated sodium channels
  - epithelial sodium channels (ENaC)
- Chloride channels
- Proton channels
  - Voltage-gated proton channels
- Non-selective cation channels
- Most transient receptor potential channels
- Other classifications

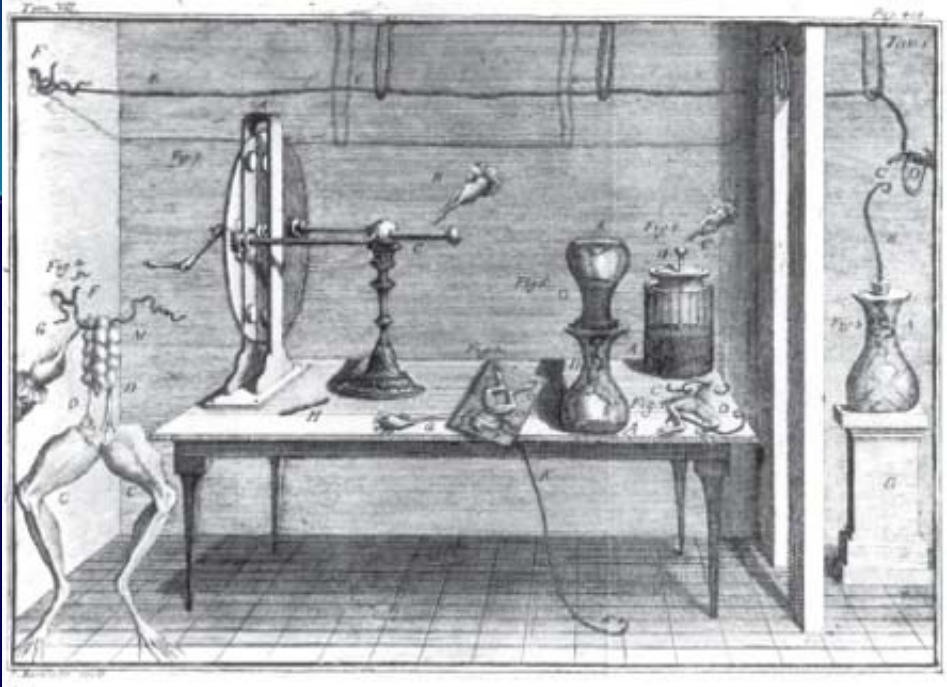
By pore

- **One single pore channels: almost all ion channels**
- **Two-pore channels**
  - **are 2 members**
  - **form cation-selective ion channels**
  - **contain two KV-style six-transmembrane domains, suggesting they form a dimer in the membrane**
  - **are related to catsper channels and, more distantly, TRP channels**

By the duration of the response to stimuli

- Transient receptor potential (TRP) channels:
  - is named after their role in Drosophila phototransduction.
  - group (family), containing at least 28 members
  - subdivided into 6 subfamilies based on homology: canonical (TRPC), vanilloid receptors (TRPV), melastatin (TRPM), polycystins (TRPP), mucolipins (TRPML), and ankyrin transmembrane protein 1 (TRPA)
  - incredibly diverse in its method of activation. Some TRP channels seem to be constitutively open, while others are gated by voltage, intracellular Ca^{2+}, pH, redox state, osmolarity, and mechanical stretch.
  - vary according to the ion(s) they pass, some being selective for Ca^{2+} while others are less selective, acting as cation channels

Electrophysiology of the Cell Membrane



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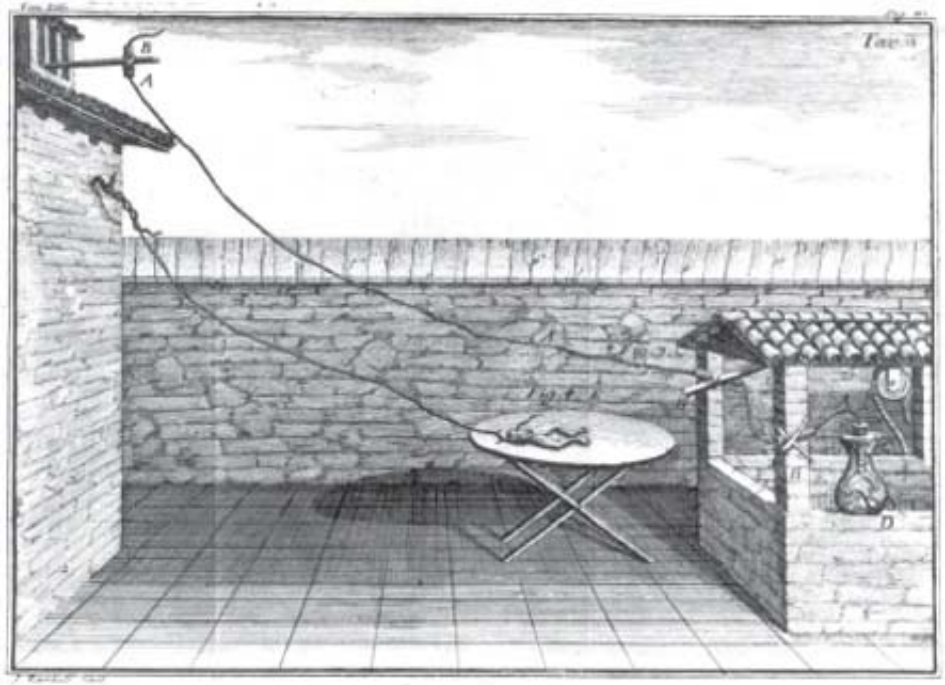


B

Early electrophysiological experiments of Galvani

In the late 1700s, Luigi Galvani:

frog legs vigorously contracted when electrical stimulation was applied either directly to the leg muscle or to the nerves leading to the muscle

Animal electricity

- Electrical currents in a metal wire are conducted by the flow of electrons
- Electrical currents across cell membranes are carried by the major inorganic ions of physiological fluids: Ca^{2+}, Na^{+}, K^{+}, Cl^{-}, and $[HCO_3]^{-}$.
- The flow of ions through specific types of channels is the basis of electrical signals that underlie neuronal activity and animal behavior.
- Opening and closing of such channels is the fundamental process behind electrical phenomena such as the nerve impulse, the heartbeat, and sensory perception.

Ionic Basis of Membrane Potentials

A basic review of electrical energy

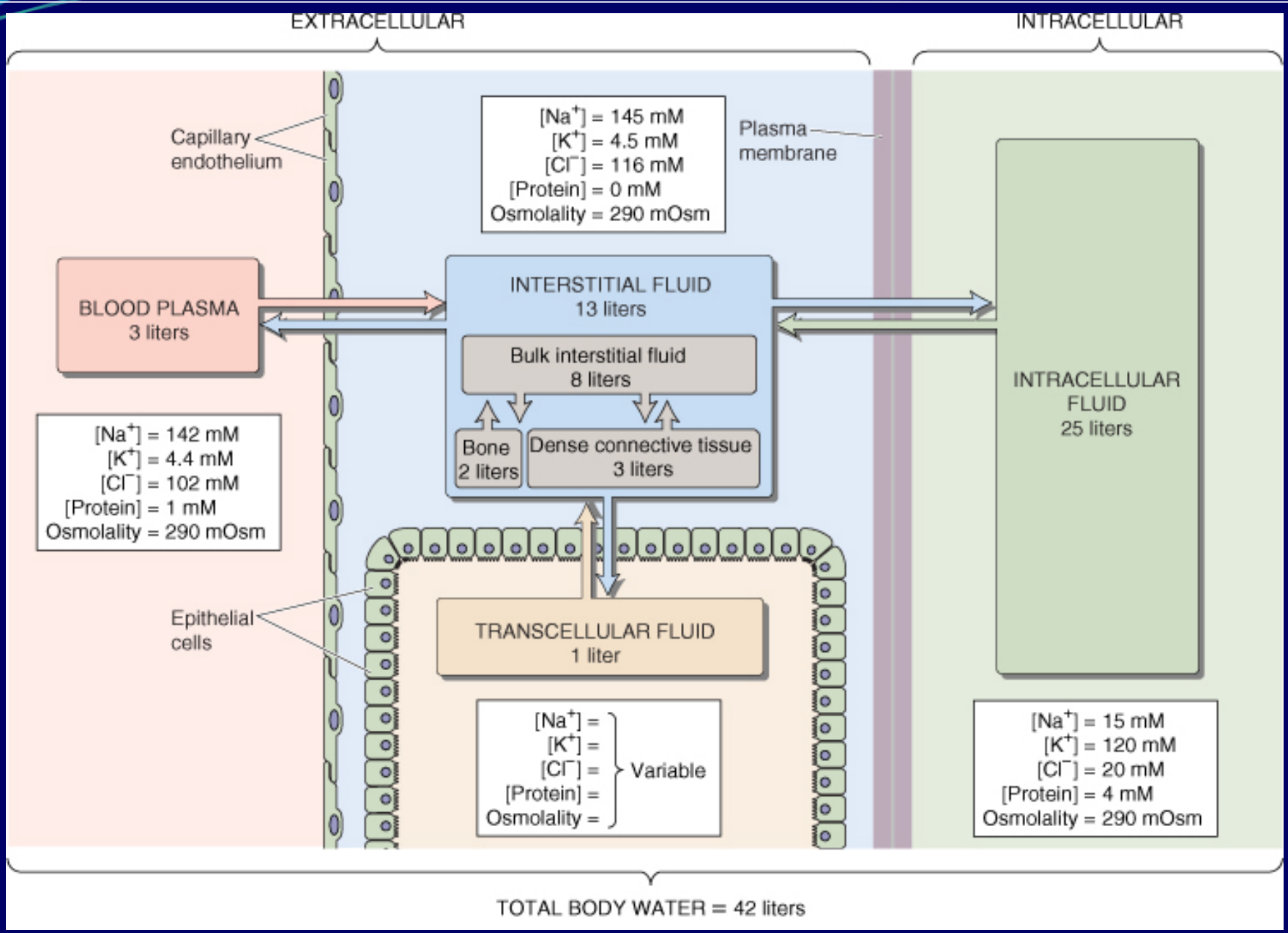
- Atoms consist of negatively (-) and positively (+) charged elementary particles, such as electrons (e^-) and protons (H^+), as well as electrically neutral particles (neutrons).
- Charges of the same sign repel each other, and those of opposite sign attract.
- Charge is measured in units of coulombs (C). The unitary charge of one electron or proton is denoted by e_0 and is equal to 1.6022×10^{-19} C.
- Ions in solution have a charge valence (z) that is an integral number of elementary charges; for example, $z = +2$ for Ca^{2+}, $z = +1$ for K^+, and $z = -1$ for Cl^-.
- The charge of a single ion (q_0), measured in coulombs, is the product of its valence and the elementary charge:

$$q_0 = ze_0$$

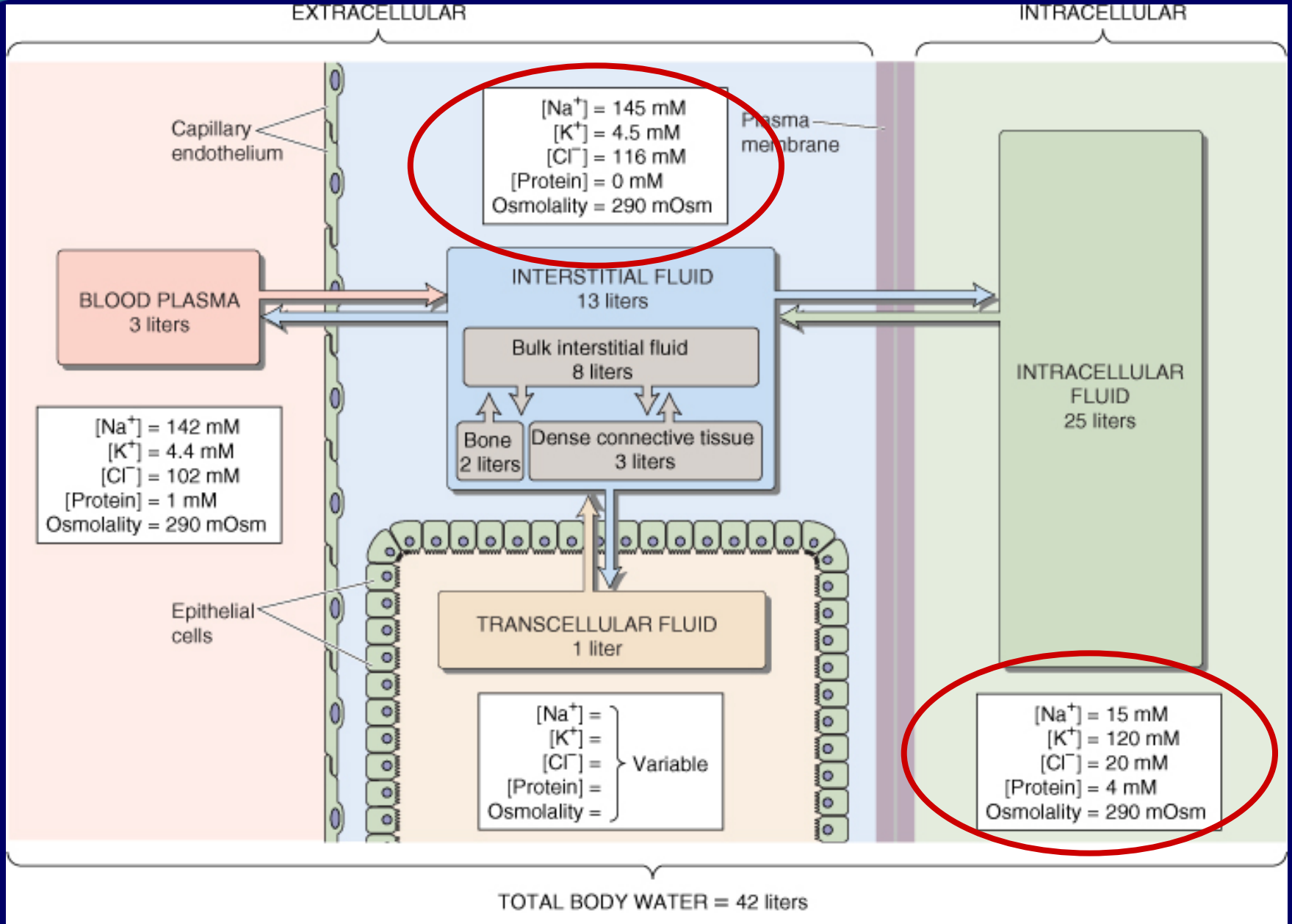
- **The attractive electrostatic force (*F*) between two ions that have valences of z_1 and z_2 can be obtained from Coulomb's law.**
- **This force (*F*) is proportional to the product of these valences and inversely proportional to the square of the distance (*r*) between the two.**
- **The force is also inversely proportional to a dimensionless term called the dielectric constant (ε):**

$$F \propto \frac{z_1 \cdot z_2}{\varepsilon r^2}$$

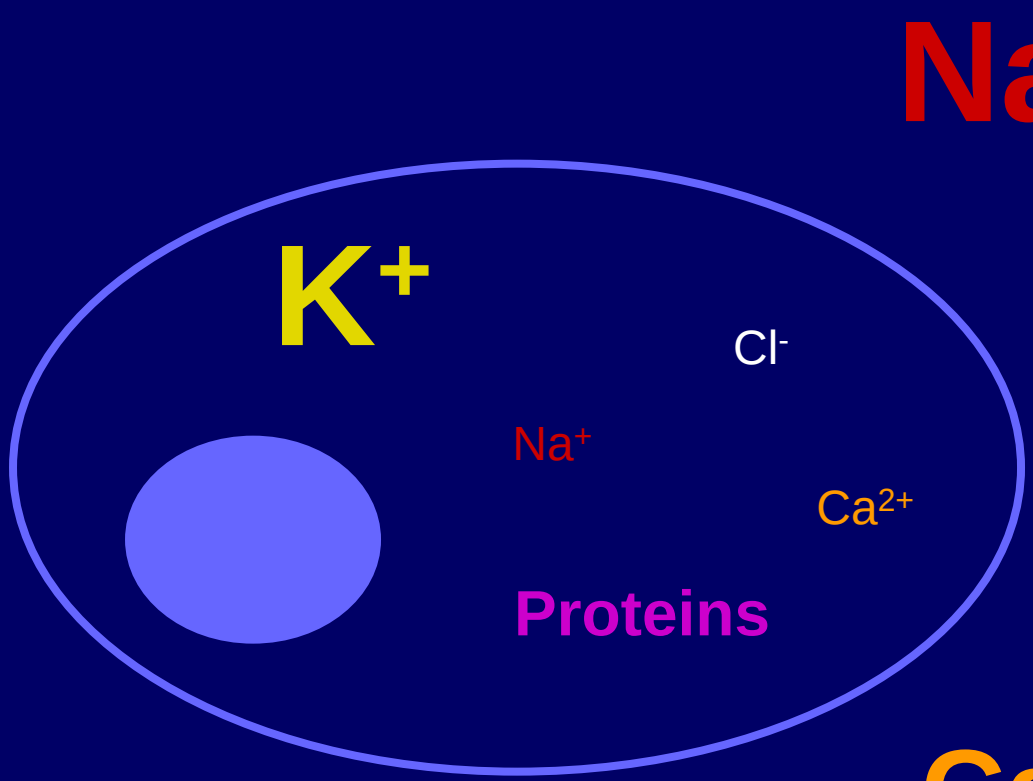
Extra- and intracellular fluids



Extra- and intracellular fluids



First rule of thumb



Na+

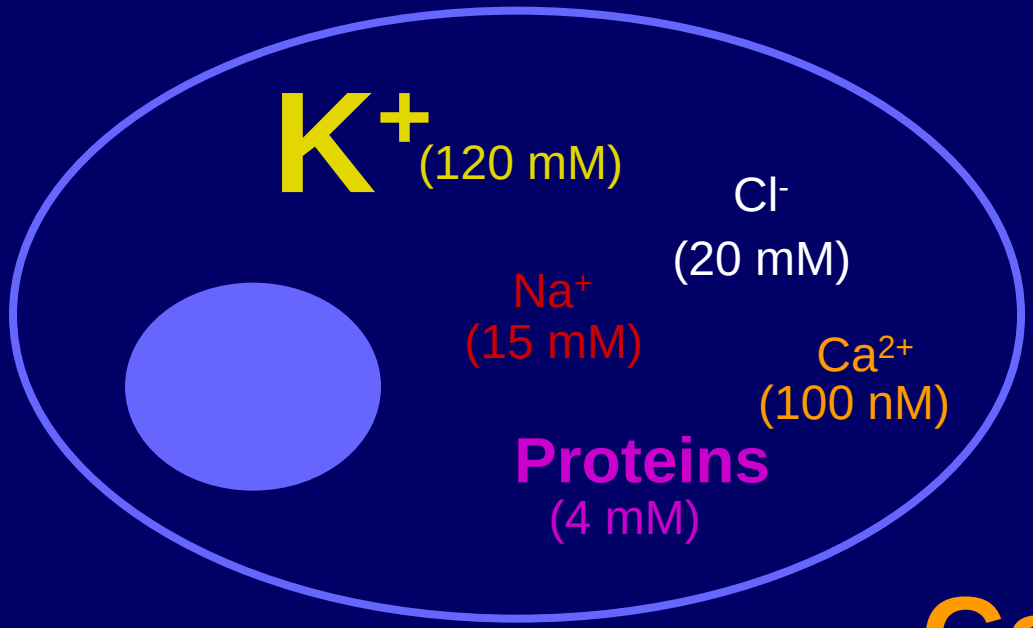
K+

Cl-

Proteins

Ca2+

First rule of thumb



Na^+ (150 mM)

K^+ (5 mM)

Cl^- (115 mM)

Proteins
(negligible)

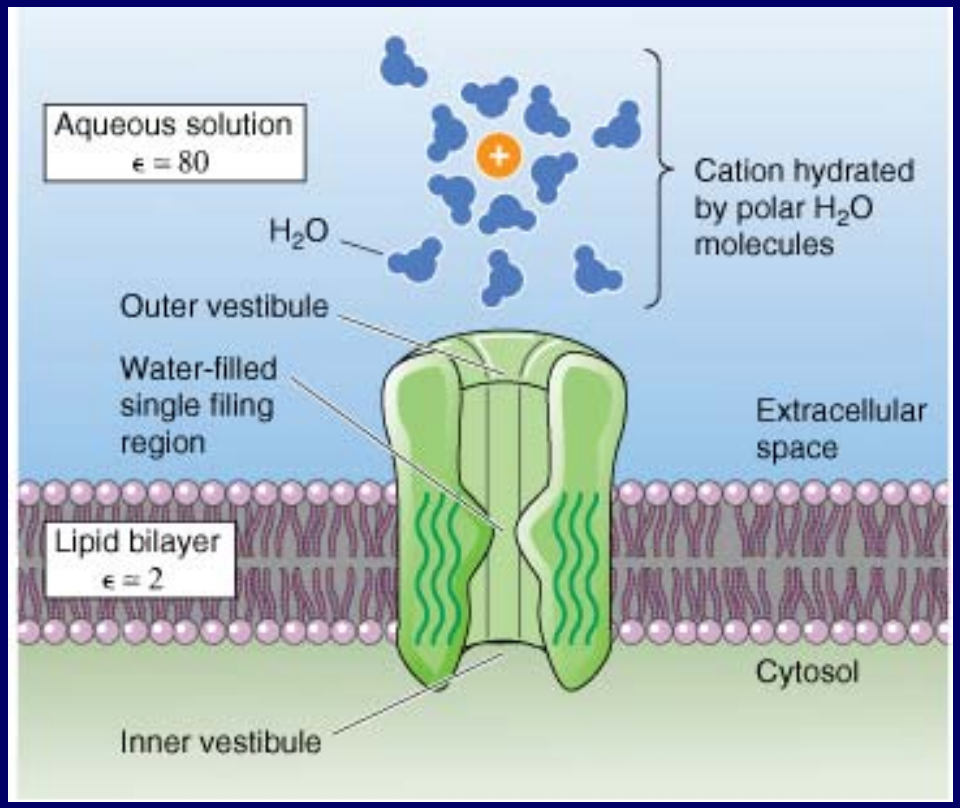
Ca^{2+} (1-2 mM)

Integral membrane proteins do the trick

These proteins create “holes” in the bilayer

These “holes” can be open all the time or some of the time

=> Permeability to a particular ion will be given by a particular protein

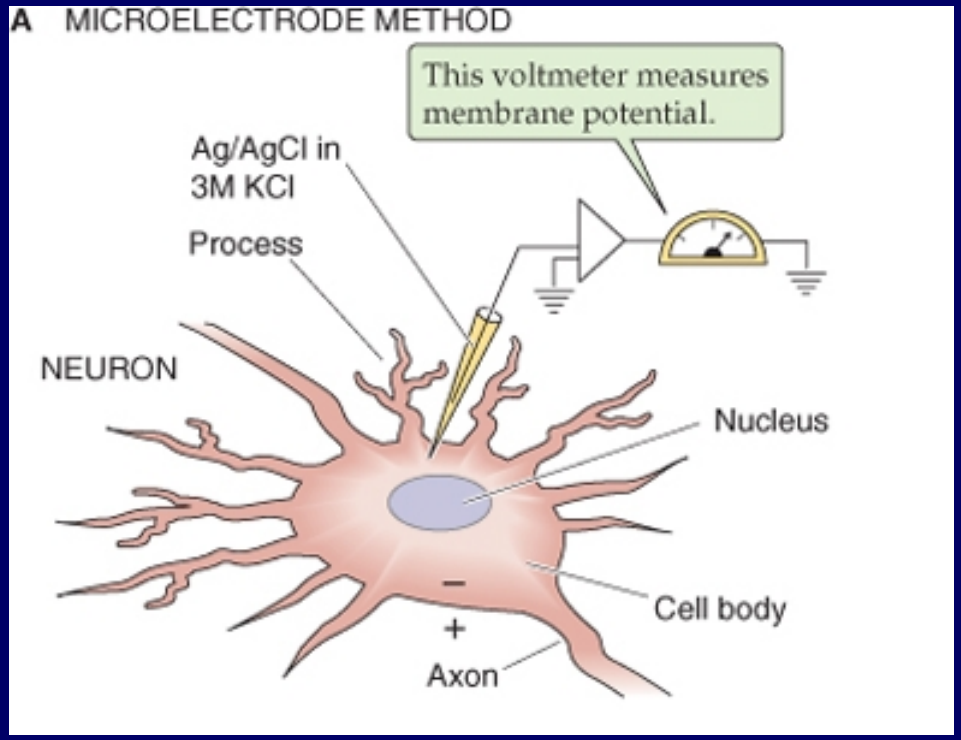


The voltage difference across the cell membrane, or the **membrane potential (V_m)**, is the difference between the electrical potential in the cytoplasm (Ψ_i) and the electrical potential in the extracellular space (Ψ_o).

Cell membrane potentials depend on ionic **concentration gradients**

How to measure V_m with an intracellular electrode?

1. Use of Microelectrodes
2. Use of Voltage-Sensitive Dyes



The sharp tip of a microelectrode is gently inserted into the cell and measures the transmembrane potential with respect to the electrical potential of the extracellular solution, defined as ground (i.e., $\Psi_o = 0$). If the cell membrane is not damaged by electrode impalement and the impaled membrane seals tightly around the glass, this technique provides an accurate measurement of V_m. Such a voltage measurement is called an **intracellular recording**.

There is a simple relationship between the electrical potential difference across a membrane and another parameter, the **electrical field (*E*)**:

$$E = \frac{V_m}{a}$$

Vm: Electrical potential difference across the membrane

α: Distance across the membrane

Typical resting membrane potentials

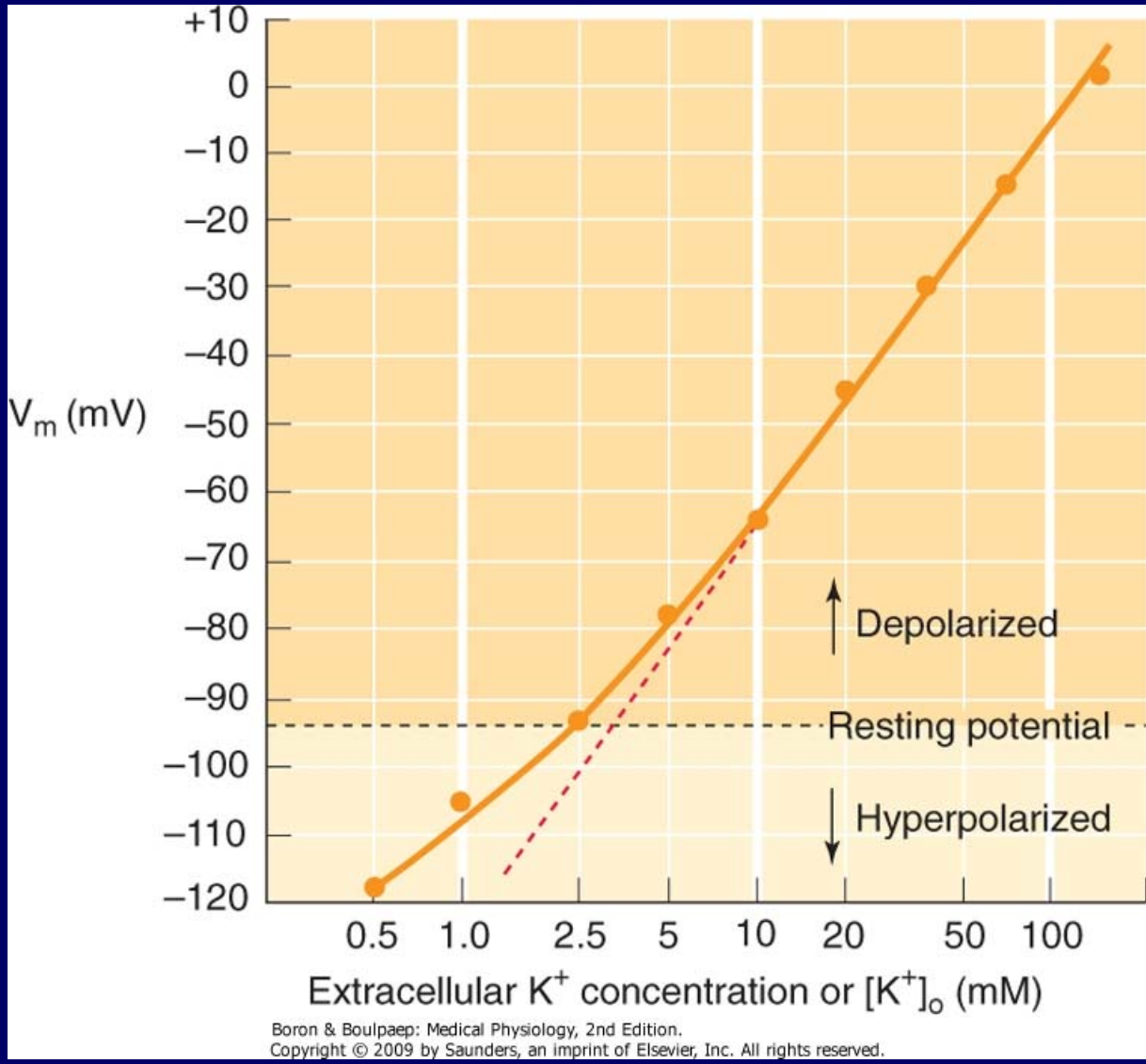
- Skeletal muscle cells, cardiac cells, and neurons: approximately -60 to -90 mV;
- Smooth muscle cells: ~ -55 mV;
- Human erythrocyte: about -9 mV;
- Certain bacteria and plant cells: as large as -200 mV;
- Very small cells (such as erythrocytes), small intracellular organelles (such as mitochondria), and fine processes (such as the synaptic endings of neurons): cannot be directly measured with a microelectrode.

Resting potential

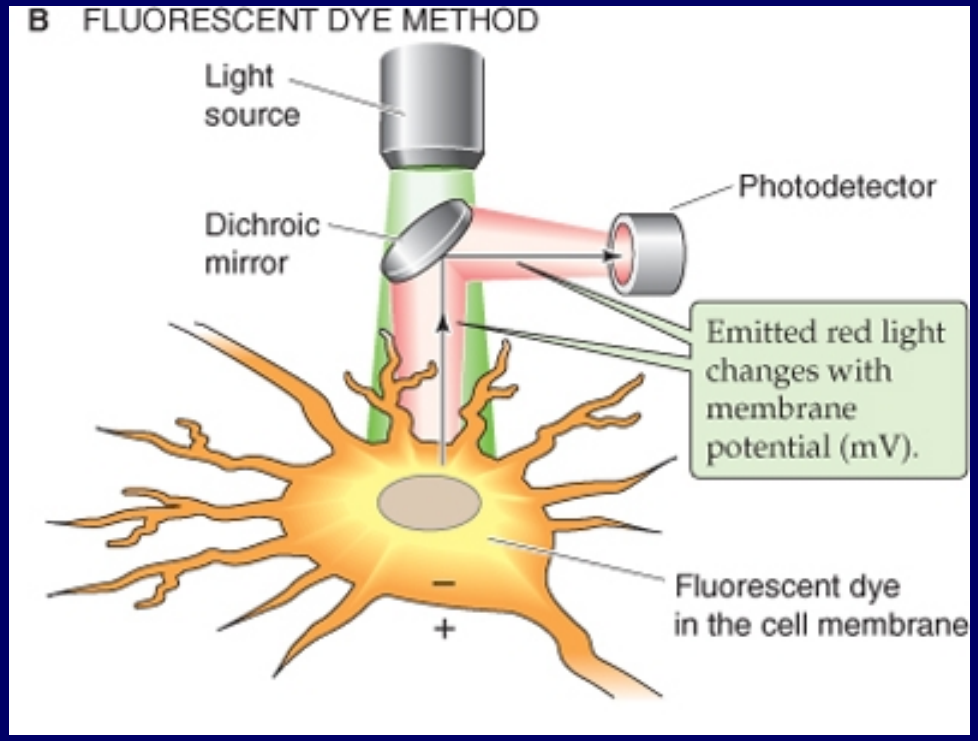
- The resting potential of large cells-whose surface-to-volume ratio is so large that ion gradients run down slowly-is maintained for a long time even when metabolic poisons block ATP-dependent energy metabolism.
- An ATP-dependent pump is not the immediate energy source underlying the membrane potential. Indeed, the squid giant axon normally has a resting potential of -60 mV.
- When the Na-K pump in the giant axon membrane is specifically inhibited with a cardiac glycoside, the immediate positive shift in V_m is only 1.4 mV. In most cases, the direct contribution of the Na-K pump to the resting V_m is very small.

Paul Horowicz and Alan Hodgkin's experiment

- Frog muscle fiber, intracellular microelectrode
- $(SO4)^{-2}$ replaced Cl^-, $[K^+]_o$ = 2.5 mM and $[Na^+]_o$ = 120 mM ➔ V_m ~ -94 mV.
- $[K^+]_o$ <2.5 mM, V_m becomes more negative
- $[K^+]_o$ > 10 mM, linear function of the *logarithm* of $[K^+]_o$

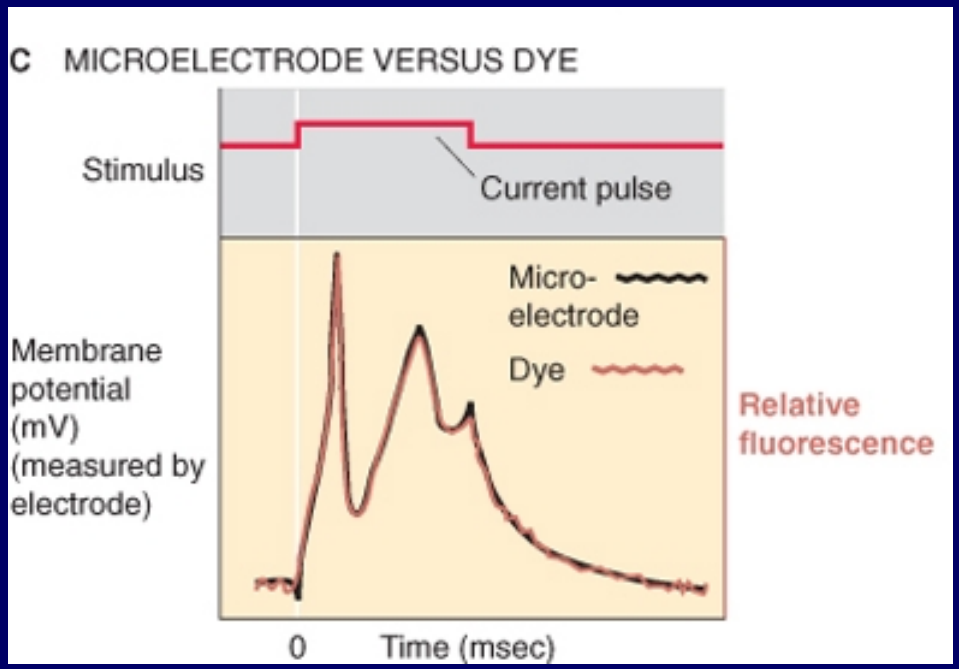


Spectroscopic techniques



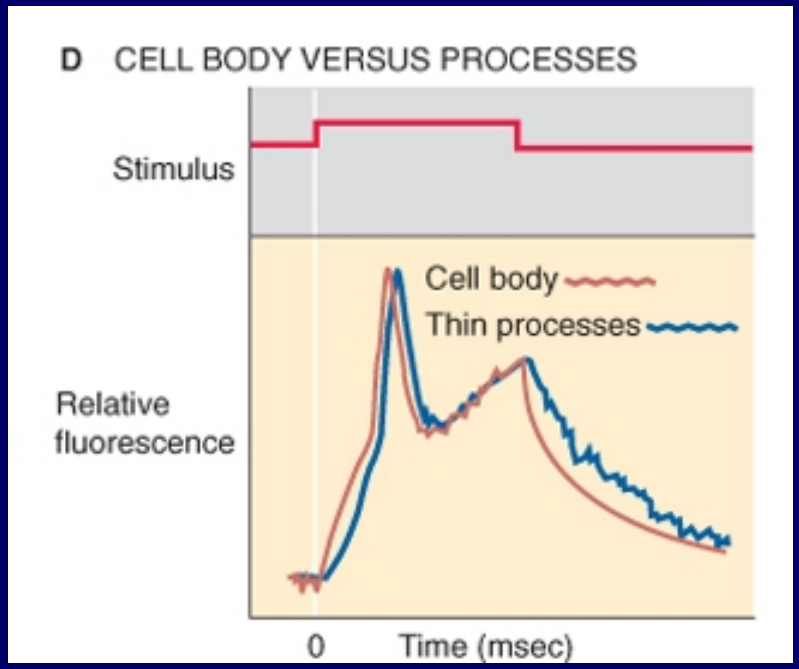
- This technique involves labeling of the cell or membrane with an appropriate organic dye molecule and monitoring of the absorption or fluorescence of the dye.
- The optical signal of the dye molecule can be independently calibrated as a function of V_m.

Spectroscopic techniques



- Electrically excitable
- V_m exhibits characteristic **time-dependent changes** in response to electrical or chemical stimulation.
- When the cell body, or soma, of a neuron is electrically stimulated, electrical and optical methods for measuring V_m detect an almost identical response at the cell body.

Spectroscopic techniques



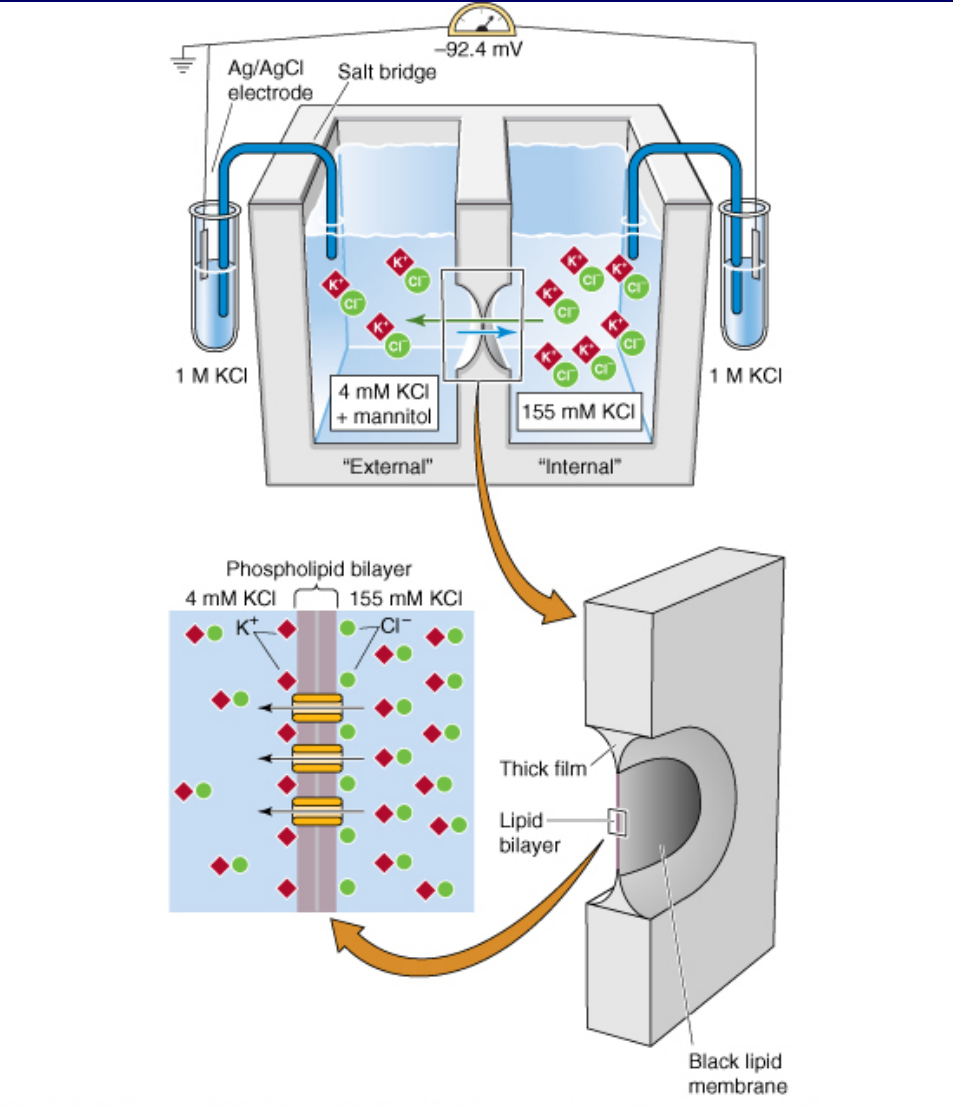
- The optical method provides the additional insight that the V_m changes are similar but **delayed** in the more distant neuronal processes that are inaccessible to a microelectrode.
- When the cell is not undergoing such active responses, V_m usually remains at a steady value that is called the **resting potential**.

Electrogenic transporters

- Some integral membrane proteins are **electrogenic transporters** in that they generate an electrical current that sets up an electrical potential across the membrane.
- One class of electrogenic transporters includes the adenosine triphosphate **(ATP)-dependent ion pumps**. These proteins use the energy of ATP hydrolysis to produce and to maintain concentration gradients of ions across cell membranes. In animal cells, the Na-K pump and Ca^{2+} pump are responsible for maintaining normal gradients of Na^{+}, K^{+}, and Ca^{2+}.
- The reactions catalyzed by these ion transport enzymes are electrogenic because they lead to separation of charge across the membrane. enzymatic turnover of the Na-K pump results in the translocation of **three Na^{+} ions out** of the cell and **two K^{+} ions into** the cell, with a net movement of one positive charge out of the cell.

The role of ion gradients

- One way to investigate the role of ion gradients in determining V_m is to study this phenomenon in an in vitro (cell-free) system.
- An artificial model of a cell membrane called a planar lipid bilayer.
- V_m originates from the diffusion of K^+ down its concentration gradient.



- **The model system of a planar bilayer (impermeable membrane), unequal salt solutions (ionic gradient), and an ion-selective channel (conductance pathway) contains the minimal components essential for generating a membrane potential.**
- **The hydrophobic membrane bilayer is a formidable barrier to inorganic ions and is also a poor conductor of electricity.**
- **Poor conductors are said to have a high resistance to electrical current, in this case, ionic current. On the other hand, ion channels act as molecular conductors of ions. They introduce a conductance pathway into the membrane and lower its resistance.**

- Membrane potentials that arise by this mechanism are called **diffusion potentials**.
- At equilibrium, the diffusion potential of an ion is the same as the **equilibrium potential** (***E*X**).

$$E_X = -\frac{RT}{z_X F} \ln \frac{[X]_i}{[X]_o}$$

- ***E*X is often simply referred to as the Nernst potential.**
- The Nernst equation predicts the equilibrium membrane potential for any concentration gradient of a particular ion across a membrane. The Nernst potentials for K^+, Na^+, Ca^{2+}, and Cl^-, respectively, are written as E_K, E_{Na}, E_{Ca}, and E_{Cl}.

Practice I

- insert the appropriate values
  - R is the universal gas constant, equal to 8.314 joules·K^{-1}·mol^{-1}
  - T is the absolute temperature, measured in kelvins (= K = degrees Celsius + 273.15)
  - F is the Faraday constant, equal to 96,485 coulombs·mol^{-1} or $J \cdot V^{-1} \cdot mol^{-1}$
- convert the logarithm base *e* (ln) to the logarithm base 10 ($\log_{10}$) ➔ a coefficient of -58.1 mV
- the Nernst equation becomes

$$E_K = (-58.1\text{mV}) \log_{10} \frac{[K^+]_i}{[K^+]_o}$$

Practice II

- For a negative ion such as Cl^-, where $z = -1$, the sign of the slope is positive:

$$E_{Cl} = (+58.1\,mV)\log_{10}\frac{[Cl^-]_i}{[Cl^-]_o}$$

- For Ca^{2+} ($z = +2$), the slope is half of -58.1 mV, or approximately -30 mV.

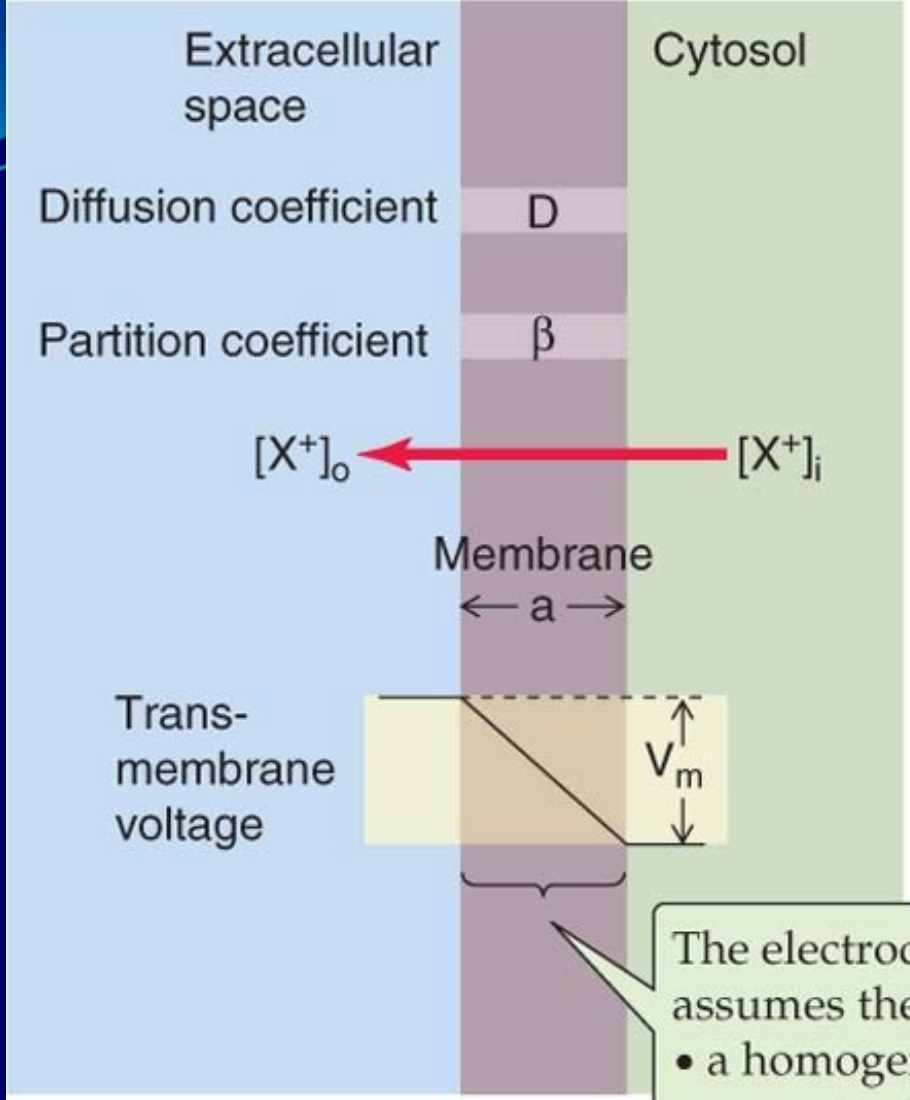
- **Notes:**
  - A Nernst slope of 58.1 mV is the value for a univalent ion at 20° C.
  - For mammalian cells at 37° C, this value is 61.5 mV.

Ion Concentration Gradients in Mammalian Cells

Ion (X)	$[X]_{out}$ (mM)	$[X]_{in}$ (mM)	$[X]_{out}/[X]_{in}$	V_x* (mV)
Skeletal muscle				
K^+	4.5	155	0.026	-95
Na^+	145	12	12	+67
Ca^{2+}	1.0	10^{-4}	10,000	+123
Cl^-	116	4.2	29	-89
[HCO3]-	24	12	2	-19
Most other cells				
K^+	4.5	120	0.038	-88
Na^+	145.4	15	9.67	+61
Ca^{2+}	1.0	10^{-4}	10,000	+123
Cl^-	116	20	5.8	-47
[HCO3]-	24	15	1.6	-13

- **Both electrical and concentration gradients are responsible for the ionic current.**
- **The process of ion permeation through the membrane is called electrodiffusion.**
- **To a first approximation, the permeation of ions through most channel proteins behaves as though the flow of these ions follows a model based on the Nernst-Planck electrodiffusion theory, which was first applied to the diffusion of ions in simple solutions.**
- **This theory leads to an important equation in medical physiology called the constant-field equation, which predicts how V_m will respond to changes in ion concentration gradients or membrane permeability.**

Electrodiffusion model of the cell membrane



The electrodiffusion model assumes the following conditions:

- a homogeneous membrane slab
- a constant electric field
- ions moving independently of one another
- a constant permeability coefficient of $P = D\beta/a$.

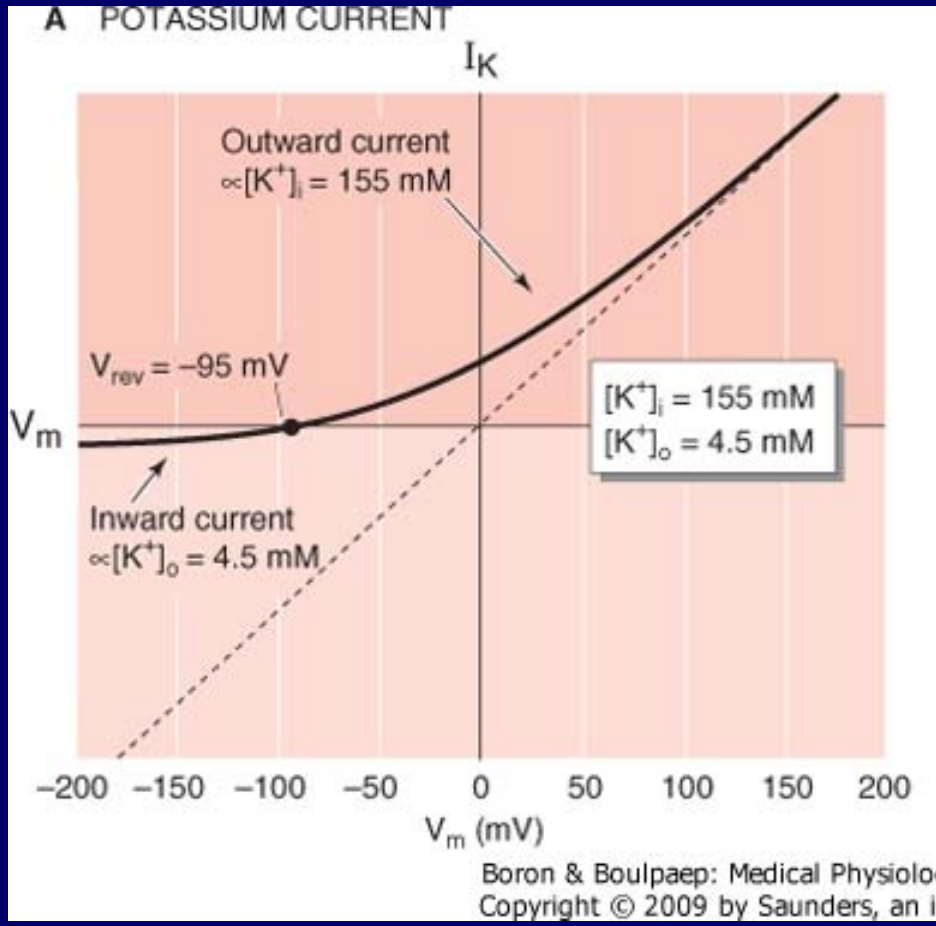
Goldman-Hodgkin-Katz (GHK) *current* equation

- To calculate the current carried by a single ion X (I_X) through the membrane by using the basic physical laws
  - the movement of molecules in solution (Fick's law of diffusion)
  - the movement of charged particles in an electrical field (electrophoresis),
  - the direct proportionality of current to voltage (Ohm's law)

$$I_X = \frac{z^2 F^2 V_m P_X}{RT} \left(\frac{[X]_i - [X]_o e^{(-zFV_m/RT)}}{1 - e^{(-zFV_m/RT)}} \right)$$

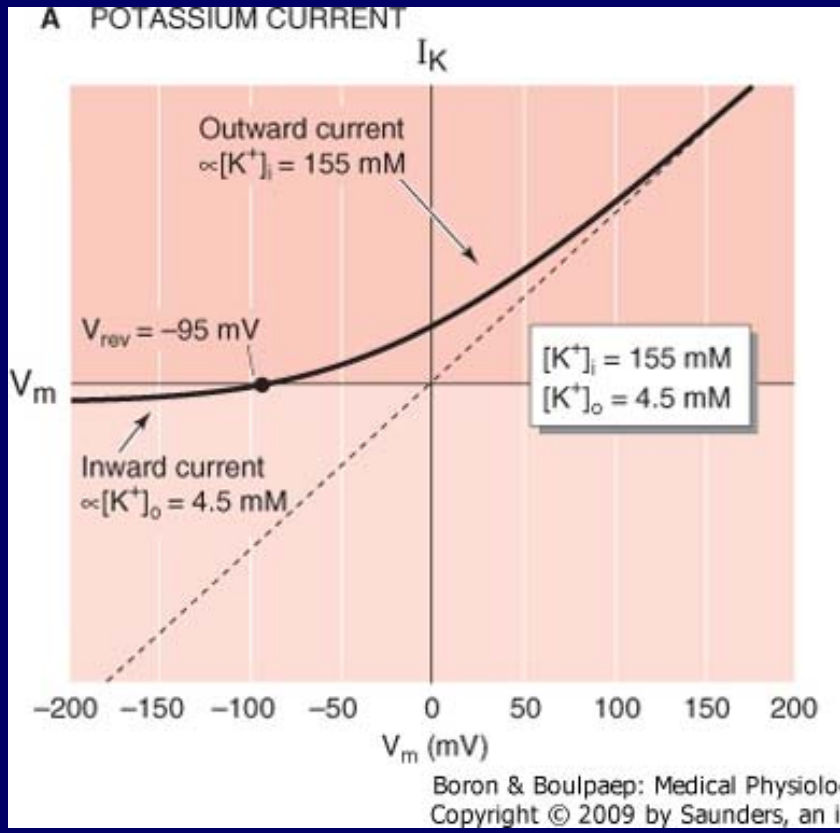
Current-voltage relationships predicted by the GHK current equation

- assuming that the membrane is perfectly selective for K^+- for a $[K^+]i$ of 155 mM and a $[K^+]o$ of 4.5 mM
- how the K^+ current (I_K) depends on *V*m
- a current of ions flowing into the cell (inward current) is defined in electrophysiology as a negative-going current, and a current flowing out of the cell (outward current) is defined as a positive current.



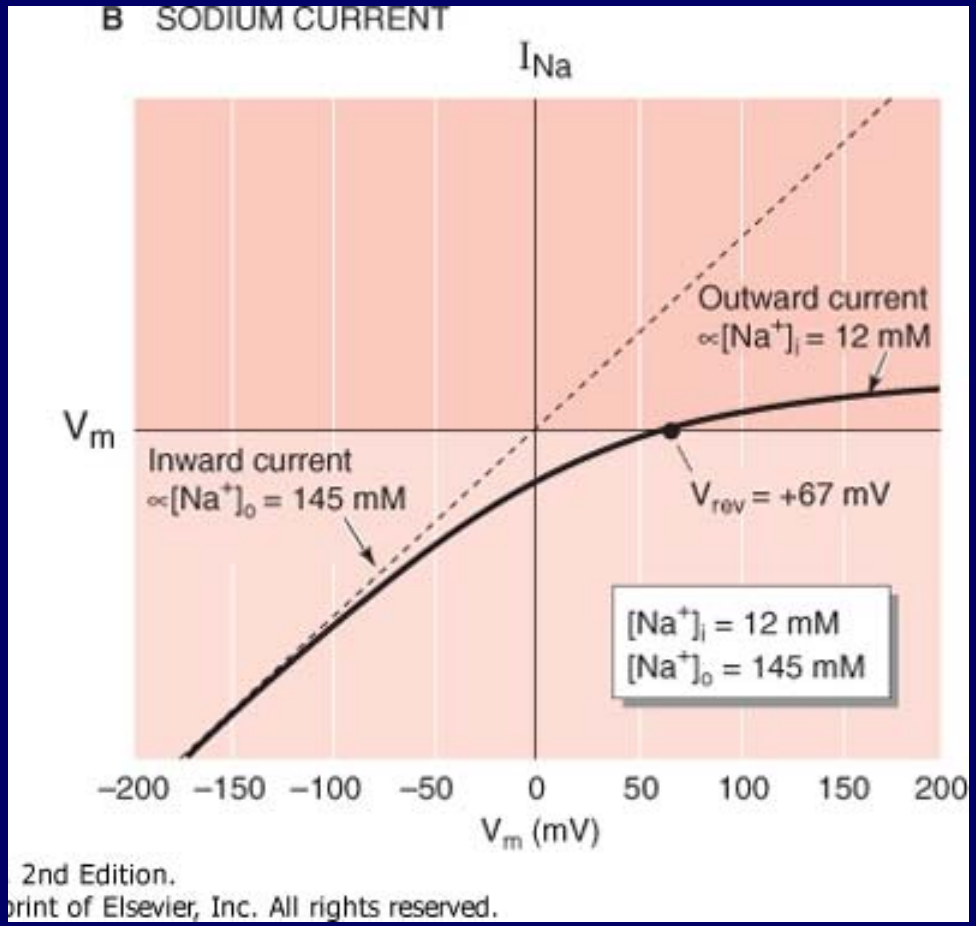
Current-voltage relationships predicted by the GHK current equation

- The value of -95 mV is called the reversal potential (V_{rev}) because it is precisely at this voltage that the direction of current reverses (i.e., the net current equals zero).
- The GHK current equation for an ion X predicts a reversal potential (V_{rev}) equal to the Nernst potential (E_X) for that ion; that is, the current is zero when the ion is in electrochemical equilibrium.
- At *V*m values more negative than V_{rev}, the net driving force on a cation is inward; at voltages that are more positive than V_{rev}, the net driving force is outward.



Current-voltage relationships predicted by the GHK current equation

- V_{rev} of Na: +67 mV
- The Na^+ current (I_{Na}) is inward at V_m values more negative than V_{rev} and outward at voltages that are more positive than this reversal potential.
- V_{rev} is the same as the Nernst potential, in this case, E_{Na}.



Membrane Potential Depends on Ionic Concentration Gradients and Permeabilities

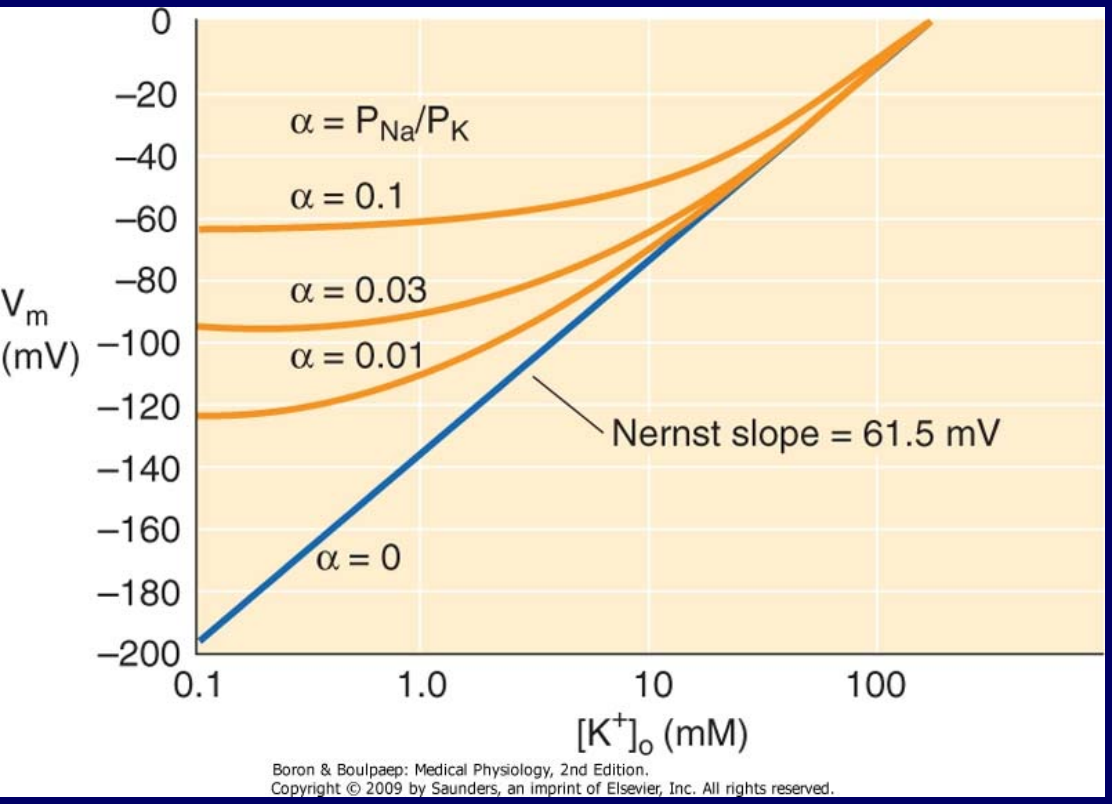
GHK *voltage* equation (constant-field equation)

- At the resting membrane potential (i.e., V_m is equal to V_{rev}), the sum of all ion currents is zero (i.e., $I_{total} = 0$).
- When we set I_{total} to zero in the expanded Equation and solve for V_{rev}, we get an expression:

$$V_{rev} = \frac{RT}{F} \ln \left(\frac{P_K[K^+]_o + P_{Na}[Na^+]_o + P_{Cl}[Cl^-]_i}{P_K[K^+]_i + P_{Na}[Na^+]_i + P_{Cl}[Cl^-]_o} \right)$$

- indicating that resting V_m depends on the concentration gradients and the permeabilities of the various ions. However, resting V_m depends primarily on the concentrations of the most permeant ion.

Dependence of the resting membrane potential on $[K^+]_o$ and on the P_{Na}/P_K ratio



- The *blue line* describes an instance in which there is no Na^+ permeability (i.e., $P_{Na}/P_K = 0$).
- The three *orange curves* describe the V_m predicted by Equation 6-10 for three values of α greater than zero and assumed values of $[Na^+]_o$, $[Na^+]_i$, and $[K^+]_i$ for skeletal muscles.

- The deviation of these orange curves from linearity is greater at low values of $[K^+]_o$, where the $[Na^+]_o$ is relatively larger.

Simplified equation

Assuming

- the sum of $[K^+]_o$ and $[Na^+]_o$ is kept fixed at its physiological value of 4.5 + 145 = 149.5 mM
- the membrane permeability to Cl^- is very small (i.e. $P_{Cl} \cong 0$).
- at 37° C
- rearrange Equation by dividing the numerator and denominator by P_K and representing the ratio P_{Na}/P_K as α.

$$V_{rev} = (61.5\,mV) \times \log_{10}\left(\frac{[K^+]_o + \alpha[Na^+]_o}{[K^+]_i + \alpha[Na^+]_i}\right)$$

Relative Permeabilities

- Reflect the ability of an ion channel protein to pull an ion from solution into the "capture volume" within the pore vestibule.
- It may therefore be highly dependent on hydration energy.
- Some permeability sequences reflect low-affinity ion-pore interactions (ex: I > Br > Cl > F). The reverse sequence often indicates a high affinity interaction.
- CFTR has a more complex sequence which indicates a combinatin of both low and high field strength interactions (Br > Cl > I > F).
- For most anions, the relative permeability is determined by the relative hydration energies of the ions.

Summary

- In general, the resting potential of most vertebrate cells is dominated by high permeability to K^+, which accounts for the observation that the resting V_m is typically close to E_K. The resting permeability to Na^+ and Ca^{2+} is normally very low.
- Skeletal muscle cells, cardiac cells, and neurons typically have resting membrane potentials ranging from -60 to -90 mV.
- Excitable cells generate action potentials by transiently increasing Na^+ or Ca^{2+} permeability and thus driving V_m in a positive direction toward E_{Na} or E_{Ca}.
- A few cells, such as vertebrate skeletal muscle fibers, have high permeability to Cl^-, which therefore contributes to the resting V_m. This high permeability also explains why the Cl^- equilibrium potential in skeletal muscle is essentially equivalent to the resting potential.

Electrical Model of a Cell Membrane

- Various Ionic Conductances
- Electromotive Forces in Parallel with a Capacitor

- The current carried by a particular ion varies with membrane voltage, as described by the *I-V* relationship for that ion.
- Suggesting that the contribution of each ion to the electrical properties of the cell membrane may be represented by elements of an electrical circuit.
- The various ionic gradients across the membrane provide a form of stored electrical energy, much like that of a battery. In physics, the voltage source of a battery is known as an **emf** (**e**lectro**m**otive **f**orce).
- The equilibrium potential of a given ion can be considered an emf for that ion.
- Each of these batteries produces its own ionic **current** across the membrane, and the sum of these individual ionic currents is the total ionic current.
- According to **Ohm's law**, the emf or voltage (*V*) and current (*I*) are directly related to each other by the **resistance** (*R*)-or inversely to the reciprocal of resistance, **conductance** (*G*):

$$V = IR$$

$$= I/G$$

- In a membrane, we can represent each ionic permeability pathway with an electrical conductance.
  - Ions with high permeability or conductance move through a low-resistance pathway;
  - Ions with low permeability move through a high-resistance pathway.
- For cell membranes, V_m is measured in millivolts, membrane current (I_m) is given in amps per square centimeter of membrane area, and membrane resistance (R_m) has the units of ohms $\times$ square centimeter.
- Membrane **conductance** (G_m), the reciprocal of membrane resistance, is thus measured in units of ohms^{-1} per square centimeter, which is equivalent to siemens per square centimeter.

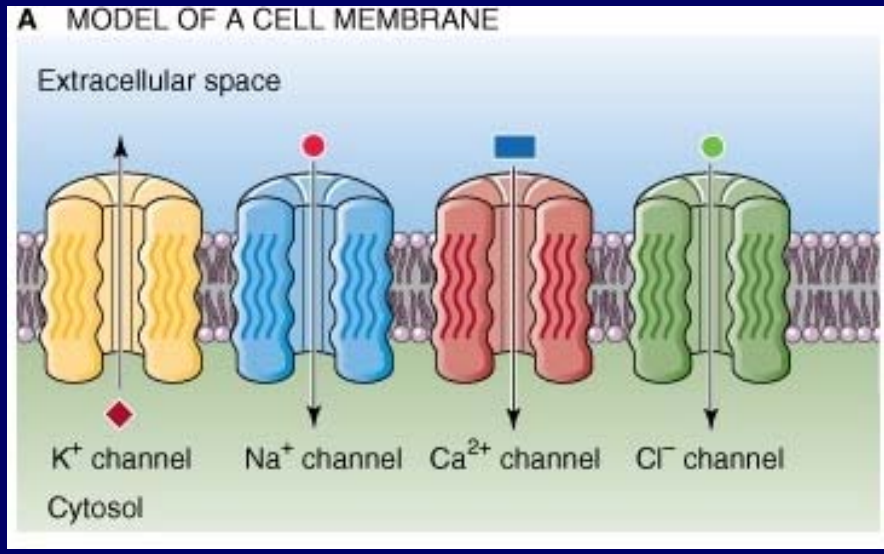
- **The GHK voltage equation predicts steady-state V_m, provided the underlying assumptions are valid. We can also predict steady-state V_m (i.e., when the net current across the membrane is zero) with another, more general equation that assumes channels behave like separate ohmic conductances:**

$$V_m = \frac{G_K}{G_m} E_K + \frac{G_{Na}}{G_m} E_{Na} + \frac{G_{Ca}}{G_m} E_{Ca} + \frac{G_{Cl}}{G_m} E_{Cl} \cdots$$

- **Thus, V_m is the sum of equilibrium potentials (E_X), each weighted by the ion's fractional conductance (e.g., G_X/G_m).**

Electrical model of a cell membrane

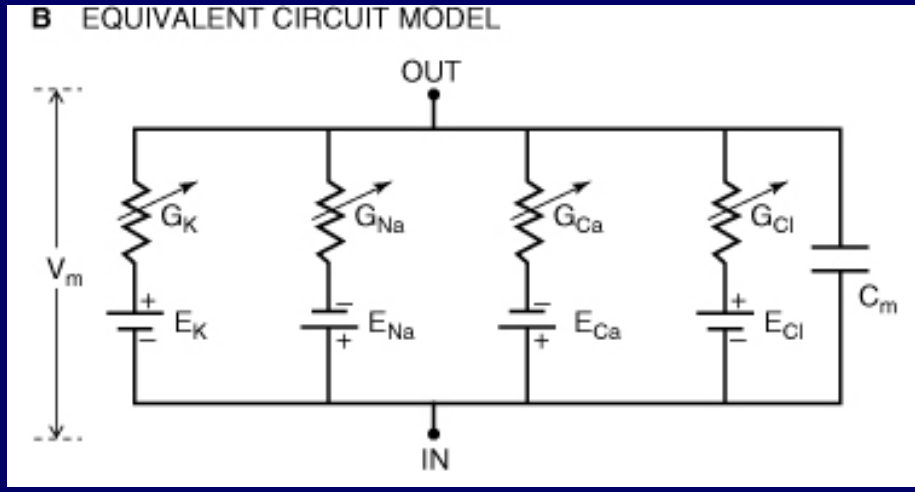
- Currents of Na^+, K^+, Ca^{2+}, and Cl^- generally flow across the cell membrane through distinct pathways. At the molecular level, these pathways correspond to specific types of ion channel proteins.



The lipid bilayer acts as a capacitor (device capable of storing current) in parallel to resistances (channels)

Electrical model of a cell membrane

- It is helpful to model the electrical behavior of cell membranes by a circuit diagram. The electrical current carried by each ion flows through a separate parallel branch of the circuit that is under the control of a variable resistor and an emf.



- For instance, the variable resistor for K^+ represents the conductance provided by K^+ channels in the membrane (G_K). The emf for K^+ corresponds to E_K.
- Similar parallel branches of the circuit above represent the other physiologically important ions. Each ion provides a component of the total conductance of the membrane, so $G_K + G_{Na} + G_{Ca} + G_{Cl}$ sum to G_m.

Total membrane current (I_m)

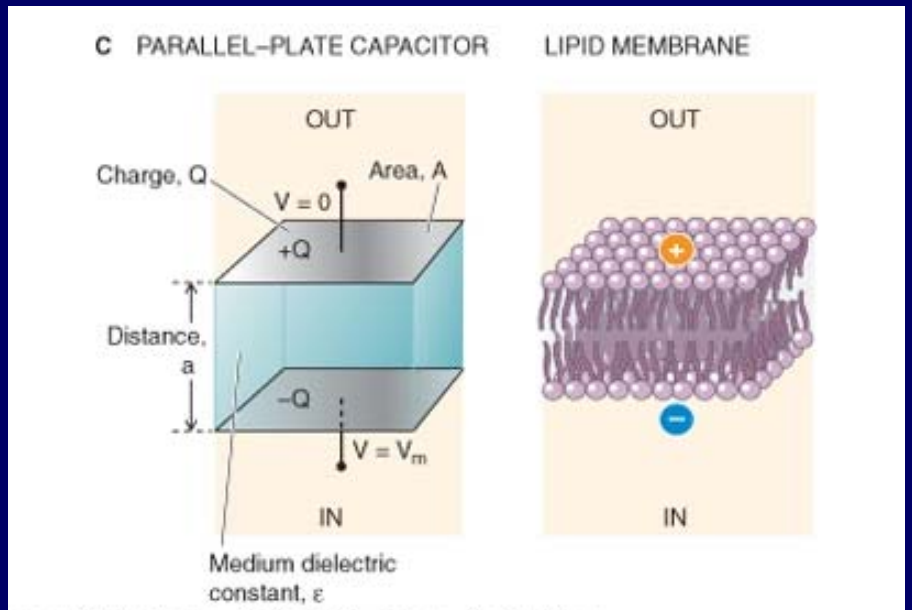
- I_m is the sum of the individual currents through each of the parallel branches of the circuit.
- For a simple case in which only one type of ionic current (I_X) flows through the membrane, I_m is simply the sum of the capacitative current and the ionic current

$$I_m = \underbrace{I_C}_{\text{Capacitative current}} + \underbrace{I_X}_{\text{Ionic current}}$$
$$= I_C + G_X(V_m - E_X)$$

- a powerful way to analyze how ionic conductance (G_X) changes with time
- directly monitor changes in G_X because this conductance parameter is directly proportional to I_m when V_m is constant (i.e., clamped).

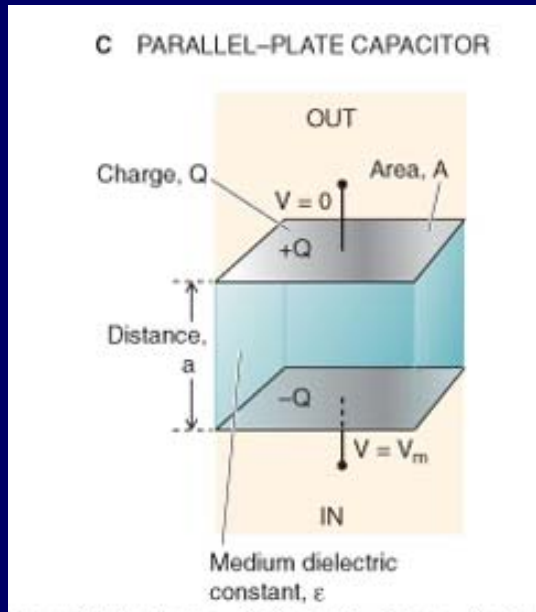
An idealized capacitor

- One more parallel element, a capacitor, is needed to complete our model of the cell membrane as an electrical circuit.
- On the left is an idealized capacitor, which is formed by two parallel plates, each with an area *A* and separated by a distance *d*.
- On the right is a capacitor that is formed by a piece of lipid membrane. The two *plates* are, in fact, the electrolyte solutions on either side of the membrane.



An idealized capacitor

- A capacitor is a device that is capable of storing separated charge. Because the lipid bilayer can maintain a separation of charge (i.e., a voltage) across its ~4-nm width, it effectively functions as a capacitor. In physics, a capacitor that is formed by two parallel plates separated by a distance *a* can be represented by the diagram.
- When the capacitor is charged, one of the plates bears a charge of +*Q* and the other plate has a charge of -*Q*.
- Such a capacitor maintains a potential difference (*V*) between the plates. Capacitance (*C*) is the magnitude of the charge stored per unit potential difference:



Capacitance

- Capacitance (*C*) is the magnitude of the charge stored per unit potential difference

  $C = Q / V$

- Capacitance is measured in units of farads (F); 1 farad = 1 coulomb/volt.
- For the particular geometry of the parallel-plate capacitor, capacitance is directly proportional to the surface area (*A*) of one side of a plate, to the dielectric constant of the medium between the two plates (ε), and to the permittivity constant (ε_o), and it is inversely proportional to the distance (*a*) separating the plates.

  $C = AE\varepsilon_o / a$

- **Because of its similar geometry, the cell membrane has a capacitance that is analogous to that of the parallel-plate capacitor.**
- **The capacitance of 1 cm^2 of most cell membranes is ~1 μF; that is, most membranes have a specific capacitance of 1 $\mu F/cm^2$.**
- **We can use above Equation to estimate the thickness of the membrane. If we assume that the average dielectric constant of a biological membrane is $\varepsilon = 5$ (slightly greater than the value of 2 for pure hydrocarbon), Equation gives a value of 4.4 nm for *a*-that is, the thickness of the membrane. This value is quite close to estimates of membrane thickness that have been obtained by other physical techniques.**

The Separation of Relatively Few Charges Across the Bilayer

Capacitance Maintains the Membrane Potential

- Use the capacitance of the cell membrane to estimate the amount of charge that the membrane actually separates in generating a typical membrane potential.
- Consider a spherical cell with a diameter of 10 μm and a $[K^+]_i$ of 100 mM. This cell needs to lose only 0.004% of its K^+ to charge the capacitance of the membrane to a voltage of -61.5 mV.
- This small loss of K^+ is clearly insignificant in comparison with a cell's total ionic composition and does not significantly perturb concentration gradients.
- In general, cell membrane potentials are sustained by a very small separation of charge.
- Total membrane current has two components: one carried by ions through channels, and the other carried by ions as they charge the membrane capacitance.

Ionic Current Is Directly Proportional to the Electrochemical Driving Force

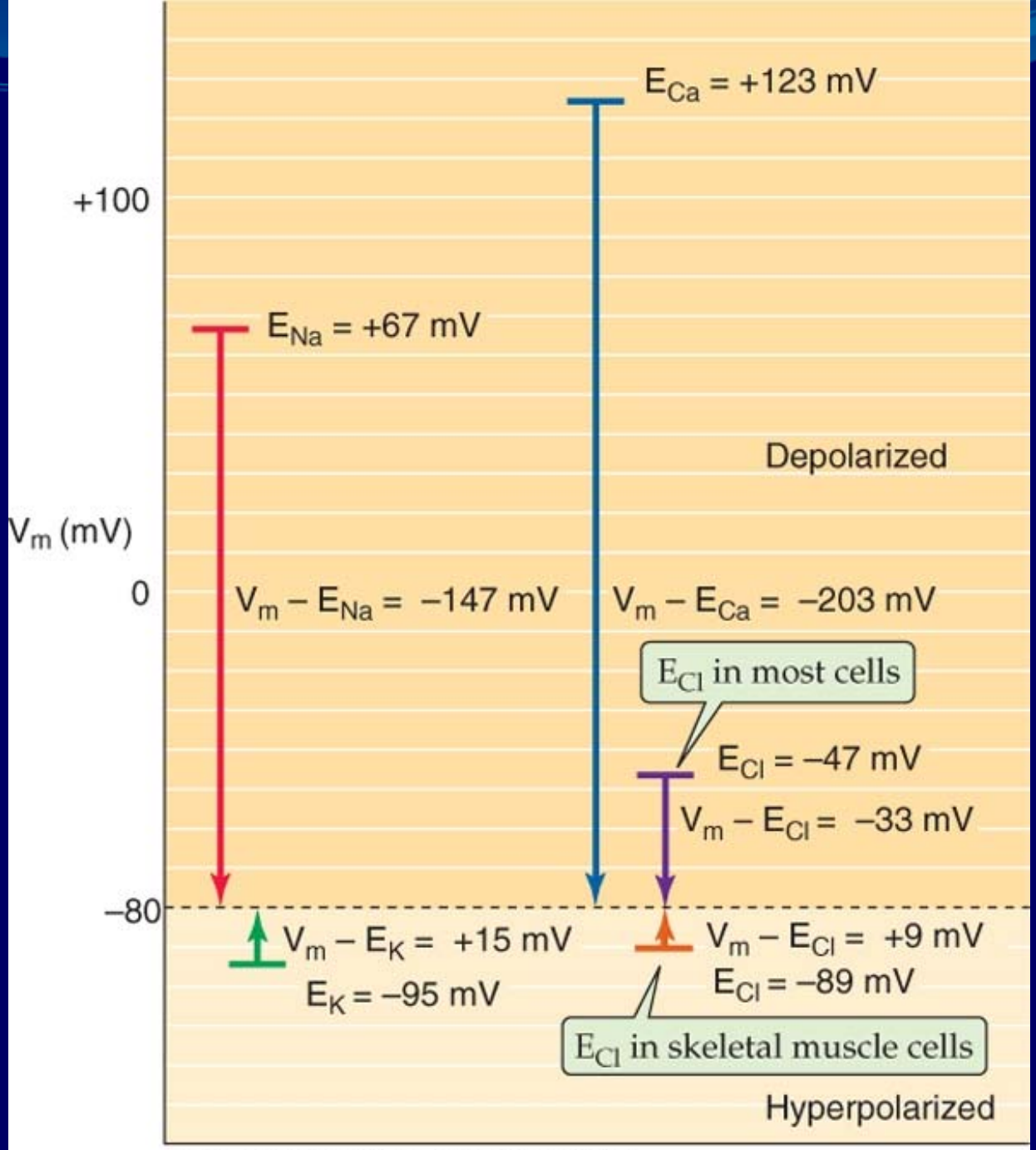
- the ionic current through a given conductance pathway is proportional to the difference (V_m - E_X)
- the proportionality constant is the ionic conductance (G_X)

$$I_X = G_X (V_m - E_X)$$

- The term (V_m - E_X) is often referred to as the driving force in electrophysiology.
- The larger the driving force, the larger the observed current.
- Returning to the *I-V* relationship for K^+, when V_m is more positive than E_K, the driving force is positive, producing an outward (i.e., positive) current.

Electrochemical driving forces acting on various ions

- the arrows represent the magnitudes and directions of the driving forces for the various ions

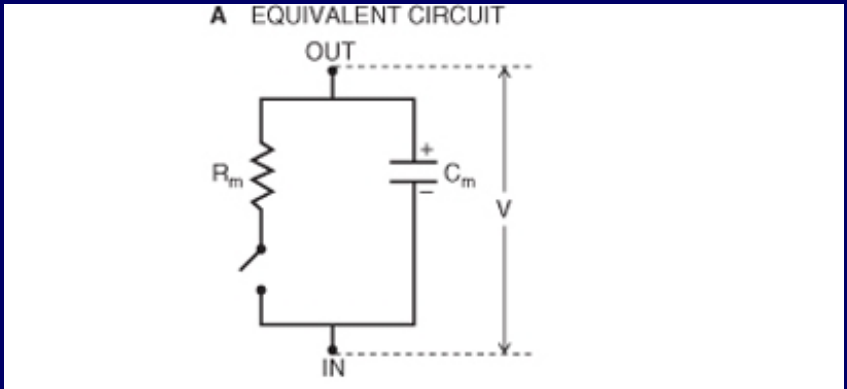


Capacitative Current Is Proportional to the Rate of Voltage Change

- Current carried by inorganic ions flows through open channels according to the principles of electrodiffusion and Ohm's law, as explained above.
- However, when V_m is changing-as it does during an action potential-another current due to the membrane capacitance also shapes the electrical responses of cells. This current, which flows only while *V*m is changing, is called the **capacitative current**.
- How does a capacitor produce a current? When voltage across a capacitor changes, the capacitor either gains or loses charge. This movement of charge onto or off the capacitor is an electrical (i.e., the capacitative) current.

A simple membrane circuit

- Compose
  - a capacitor (C_m)
  - a resistor (R_m)
  - a switch

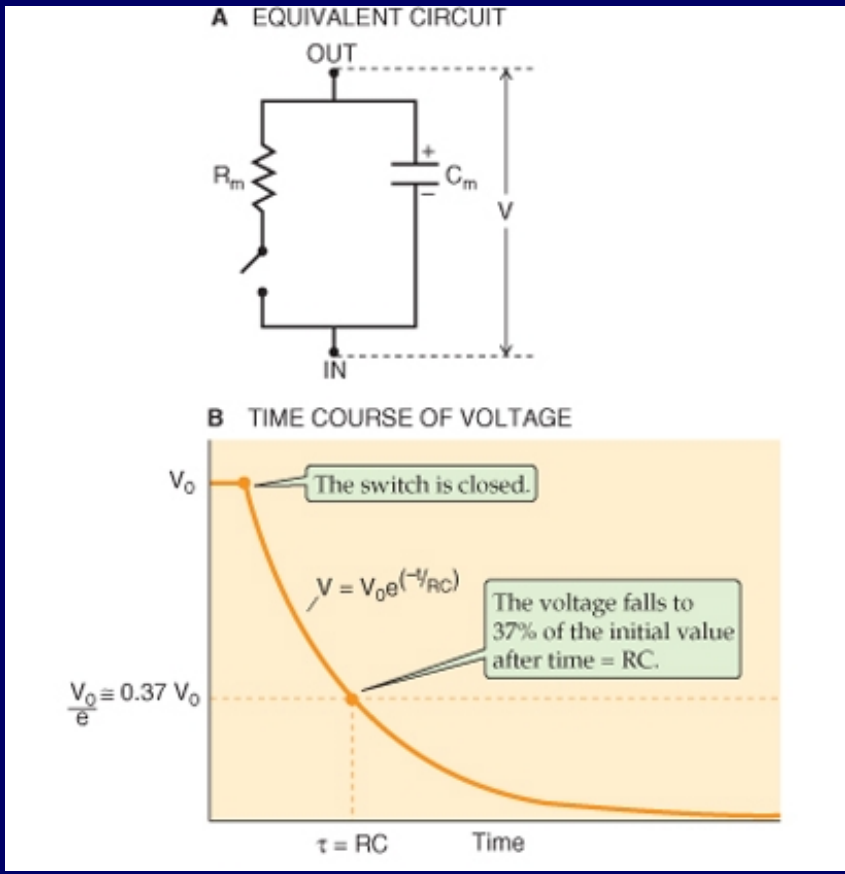


- Assume that the switch is open and that the capacitor is initially charged to a voltage of V_0, causing a separation of charge (Q) across the capacitor.
- According to the definition of capacitance, the charge stored by the capacitor is a product of capacitance and voltage.

$$Q = C_m V_0$$

The simple membrane circuit

- As long as the switch in the circuit remains open, the capacitor maintains this charge.
- when the switch is closed, the charge on the capacitor discharges through the resistor, and the voltage difference between the circuit points labeled "In" and "Out" decays from V_0 to a final value of zero.



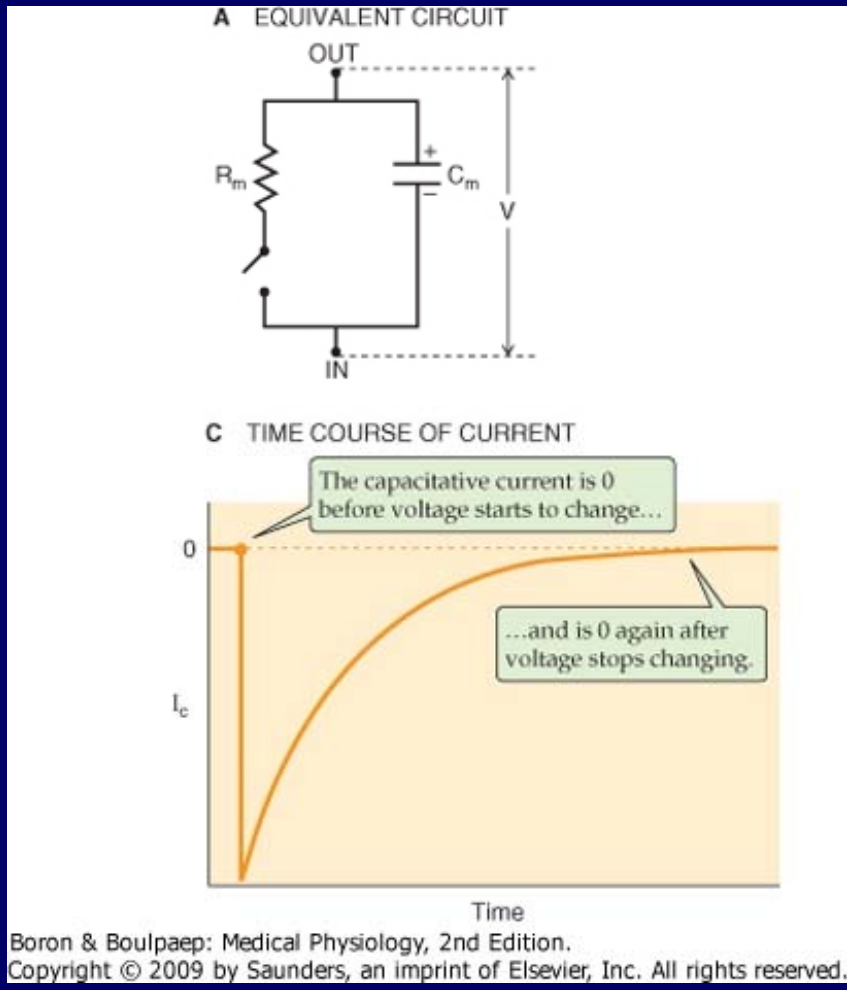
- This voltage decay follows an exponential time course. The time required for the voltage to fall to 37% of its initial value is a characteristic parameter called the **time constant** (τ), which has units of time.

$$\tau = R_m \cdot C_m$$

$$V = V_0 e^{-t/RC}$$

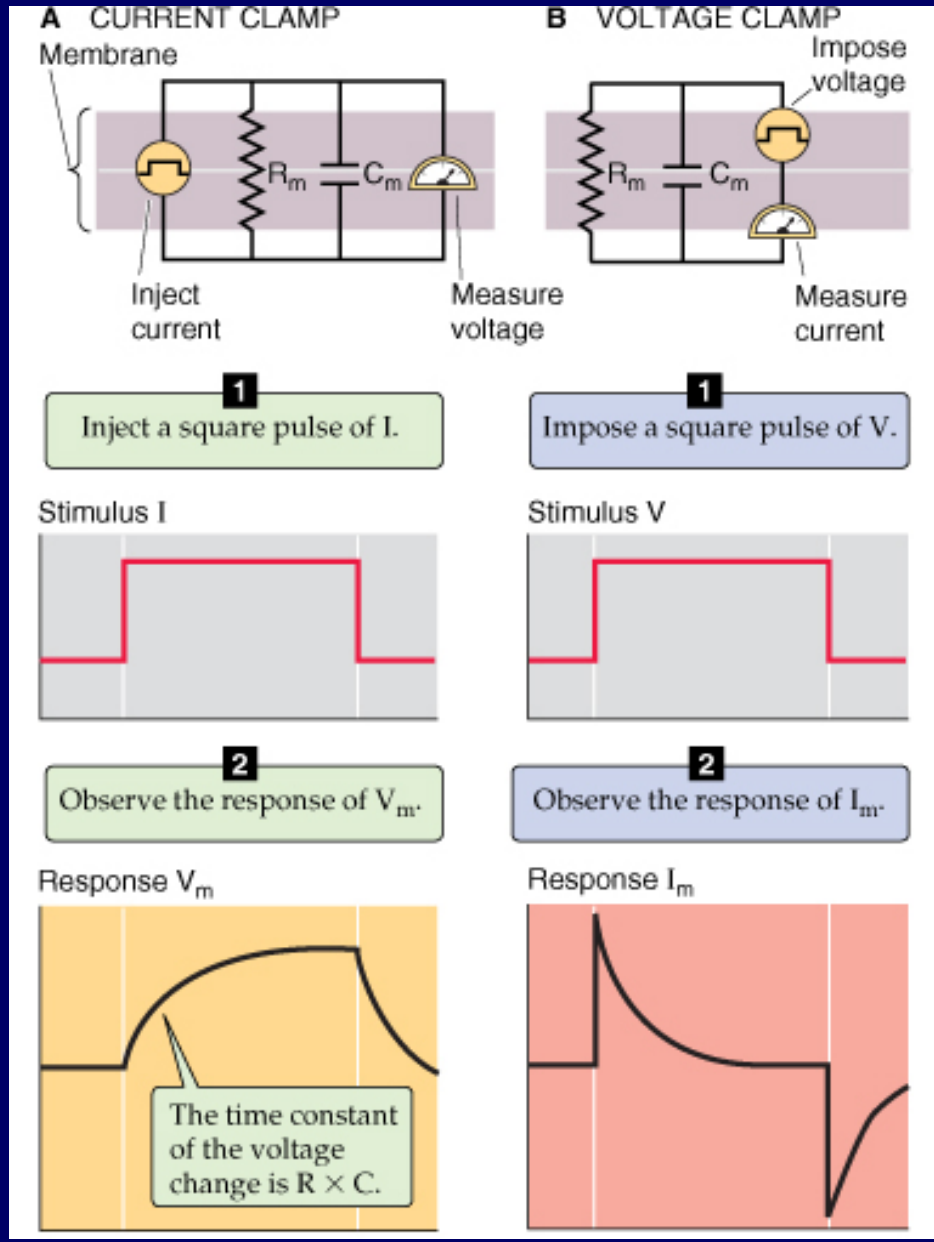
Capacitative current

- The **capacitative current** (I_C) is zero before the switch is closed, when the voltage is stable at V_0.
- When we close the switch, charge begins to flow rapidly off the capacitor, and the magnitude of I_C is maximal.
- As the charge on the capacitor gradually falls, the rate at which charge flows off the capacitor gradually falls as well until I_C is zero at "infinite" time.
- Note, however, that V and I_C relax with the same time constant.

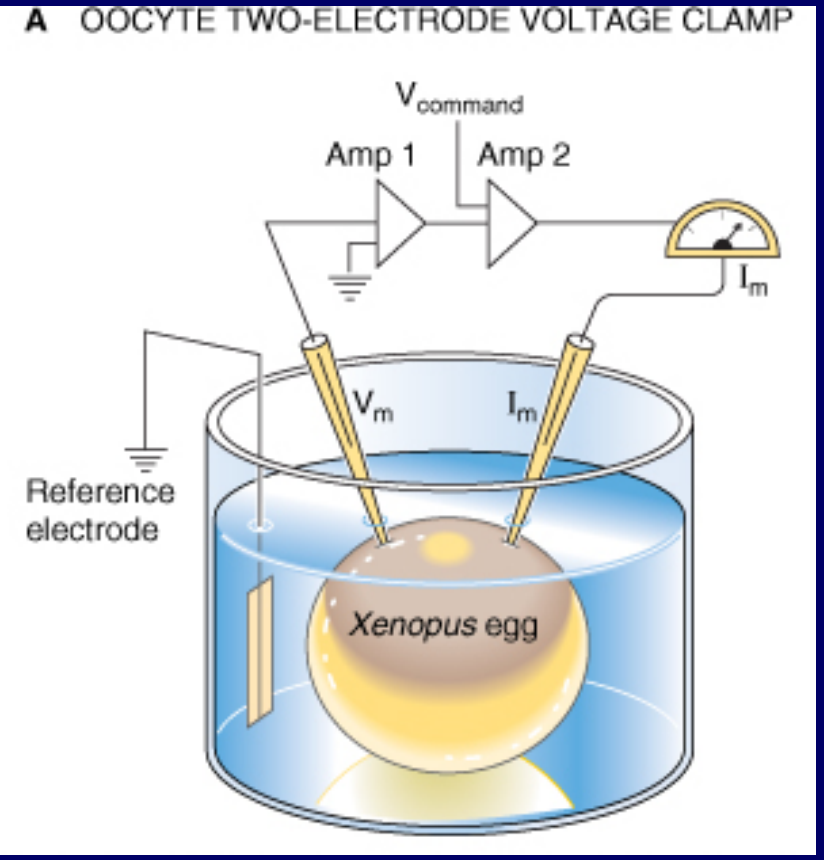


Consequences of membrane capacitance

- current or voltage is abruptly changed to a fixed value, held constant for a certain time, and returned to the original value. This pattern is called a square pulse.
- suddenly change voltage to a new value, a transient capacitative current flows as charge flows onto the capacitor
- The capacitative current is maximal at the beginning of the square pulse, when charge flows most rapidly onto the capacitor, and then falls off exponentially with a time constant of *RC*
- suddenly decrease the voltage to its original value, I_C flows in the direction opposite that observed at the beginning of the pulse. Thus, I_C appears as brief spikes at the beginning and end of the voltage pulse



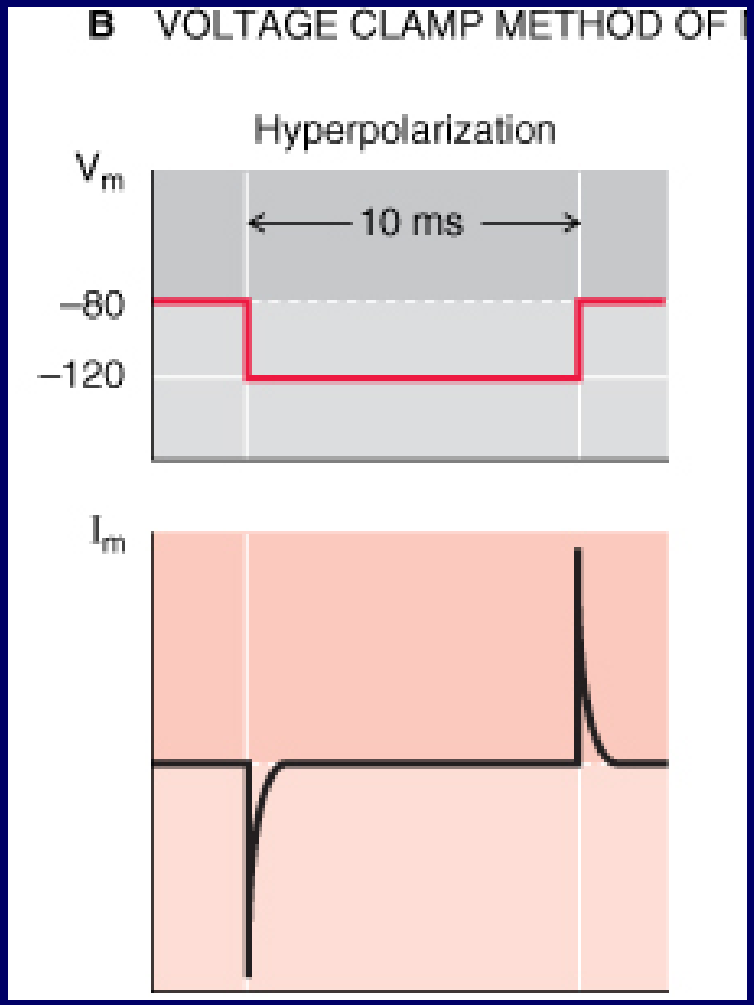
Voltage-clamping



- When the voltage-sensing electrode detects a difference from the intended voltage, called the command voltage, a feedback amplifier rapidly injects opposing current to maintain a constant V_m.
- The magnitude of the injected current needed to keep V_m constant is equal, but opposite in sign, to the membrane current and is thus an accurate measurement of the total membrane current (I_m)

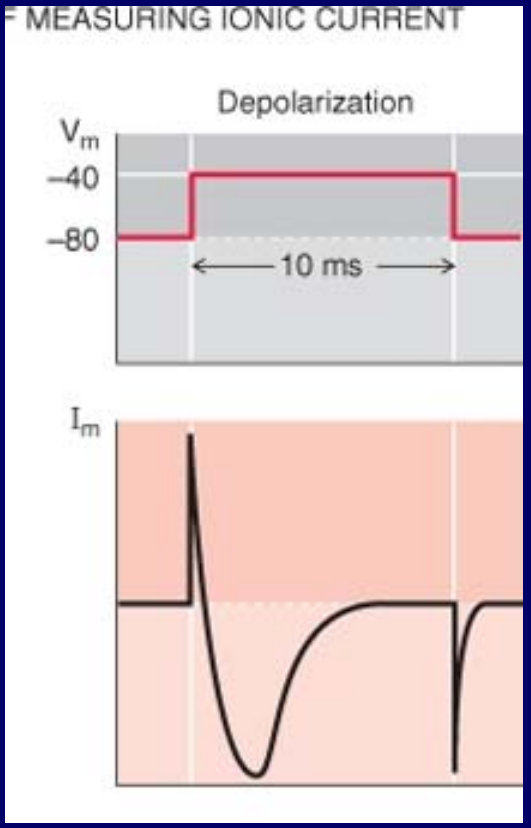
Hyperpolarization

- A cell membrane is initially clamped at a resting potential of -80 mV. V_m is then stepped to -120 mV for 10 ms (a step of -40 mV) and finally returned to -80 mV.
- Such a negative-going V_m change is called a **hyperpolarization**.
- Only brief spikes of current are observed at the beginning and end of the voltage step and are due to the charging of membrane capacitance. No current flows in between these two spikes.



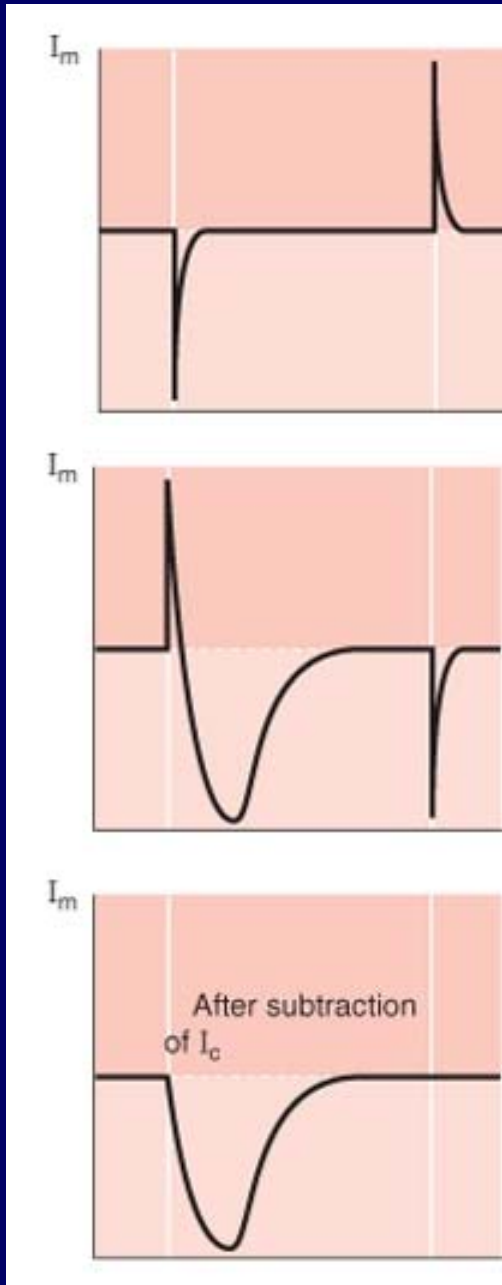
Depolarization

- What happens if we rapidly change V_m in the opposite direction by shifting the voltage from -80 to -40 mV (a step of +40 mV)?
- Such a positive-going change in V_m from a reference voltage is called a **depolarization**.
- In addition to the expected transient *capacitative* current, a large, inward, time-dependent current flows. This current is an *ionic* current and is due to the opening and closing kinetics of a particular class of channels called voltage-gated Na^+ channels, which open only when V_m is made sufficiently positive.



Macroscopic current

- We can remove the contribution of the capacitative current to the total current by subtracting the inverse of the rapid transient current recorded during the hyperpolarizing pulse of the same magnitude.
- The remaining slower current is inward (i.e., downward) and represents I_{Na}, which is directly proportional to G_{Na}.
- The ionic current in the lowest panel is called a **macroscopic current** because it is due to the activity of a large population of channels sampled from a whole cell.



Diseases of ion channels: Chemicals

- Tetrodotoxin (TTX), used by puffer fish and some types of newts for defense. It blocks sodium channels.
- Saxitoxin, is produced by a dinoflagellate also known as "red tide". It blocks voltage dependent sodium channels.
- Conotoxin, is used by cone snails to hunt prey.
- Lidocaine and Novocaine belong to a class of local anesthetics which block sodium ion channels.
- Dendrotoxin is produced by mamba snakes, and blocks potassium channels.
- Iberiotoxin is produced by the Buthus tamulus and blocks potassium channels.
- Heteropodatoxin is produced by Heteropoda venatoria and blocks potassium channels.

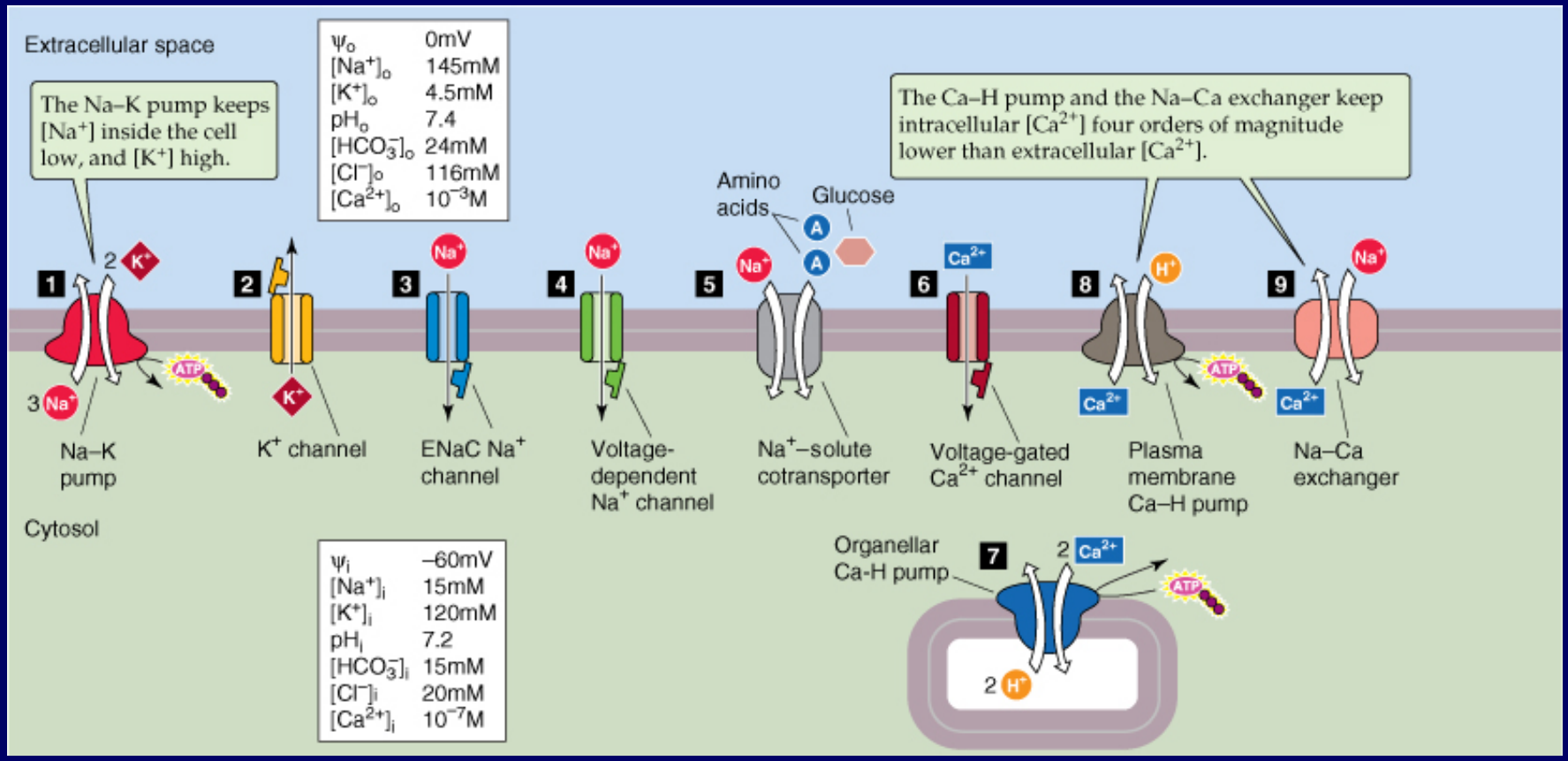
Diseases of ion channels: Genetic

- Shaker gene mutations cause a defect in the voltage gated ion channels, slowing down the repolarization of the cell.
- Equine hyperkalaemic periodic paralysis as well as Human hyperkalaemic periodic paralysis (HyperPP) are caused by a defect in voltage dependent sodium channels.
- Paramyotonia congenita (PC) and potassium aggravated myotonias (PAM)
- Generalized epilepsy with febrile seizures plus ($GEFS^{+}$)
- Episodic Ataxia (EA), characterized by sporadic bouts of severe discoordination with or without myokymia, and can be provoked by stress, startle, or heavy exertion such as exercise.
- Familial hemiplegic migraine (FHM)

Diseases of ion channels: Genetic

- Spinocerebellar ataxia type 13
- Long QT syndrome is a ventricular arrhythmia syndrome caused by mutations in one or more of presently ten different genes, most of which are potassium channels and all of which affect cardiac repolarization.
- Brugada syndrome is another ventricular arrhythmia caused by voltage-gated sodium channel gene mutations.
- Cystic fibrosis is caused by mutations in the CFTR gene, which is a chloride channel.
- Mucolipidosis type IV is caused by mutations in the gene encoding the TRPML1 channel.

Compartmentalization of ions is a net outcome



See you next week!