Basic Cellular Neurophysiology

Dr. Brian R. Christie,
Division of Medical Science
University of Victoria
Victoria, BC, Canada

Learning Objectives

- Membrane potentials, types and properties of different ion channels
- Effects of pH on seizure activity
- Neurotransmitters in the brain.
- Gap junctions in the brain.
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Title of Presentation: Basic cellular neurophysiology
Presenter’s Name: Brian R. Christie

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Affiliation/Financial interest: CIHR, NSERC, SRCF, ABMRF
Consultant: Roche (Palo Alto)

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Basic Ion Concentrations

BANANA IN THE OCEAN

Banana: High Potassium inside the cell, 95% of body’s potassium is inside cells!
Ocean: High Sodium and Chloride outside the cell.

The Na-K Pump maintains the difference in ions.

Pump driven by ATP hydrolysis involves reversible phosphorylation of an Aspartate residue on the enzyme, and 2 conformations of the enzyme:

Conformation 1: high affinity for Na+, low affinity for K+, “open” to inside of cell.
Conformation 2: low affinity for Na+, high affinity for K+, “open” to outside of cell.

Transfer of phosphate group from ATP to enzyme (releasing ADP as a product) triggers conformational change in enzyme – phosphorylated enzyme predominantly in conformation II.

Hydrolysis of phosphate group from the enzyme triggers return to original conformation (I).
Steps in the Na-K Pump Operation

The Membrane Potential

Electrical potential difference (voltage) across a cell's membrane (the resistive barrier).

Na/K pump maintains Na and K distributions.

Ion Distribution Across the Neural Membrane

- \( E_{K} \) = -100 mV, resting potential
- \( E_{Na} \) = +60 mV, action potential
- \( E_{Cl} \) = -70 mV, GABA, IPSP
- \( E_{Ca} \) = +90 mV, action potential
Hyperkalemia: (hyper-high; kalium-potassium; emia-in blood)
- fatal arrhythmias can occur with serum K levels >6.5 mmol/L.
Normal serum K is 3.5-5.0 mmol/L. Insulin, bicarbonates, salbutamol can all reduce K+.
- EKG can show reduction in the size of the P wave and development of peaked T waves.

Ion Channels and Carriers

Carriers
proteins that work as transporters that *dissipate ion gradients.*
create pores (channels) through which the material can move.

Carriers
move material from one side of membrane to the other.
i.e. Acyl carrier protein involved in fatty acid synthases.
The Nernst Equation

\[ E_{eq, K^+} = \frac{RT}{zF} \ln \frac{[K^+]_o}{[K^+]_i} \]

Gives us the equilibrium potential for a single ion.

- where
  - \( E_{eq, K^+} \) is the equilibrium potential for potassium, measured in volts
  - \( R \) is the universal gas constant, equal to 8.314 joules/K·mol
  - \( T \) is the absolute temperature, measured in kelvins (= K = degrees Celsius + 273.15)
  - \( z \) is the number of elementary charges of the ion in question involved in the reaction
  - \( F \) is the Faraday constant, equal to 96,485 coulombs/mol
  - \([K^+]_o\) is the extracellular concentration of potassium, measured in mol·m⁻³ or mmol·l⁻¹
  - \([K^+]_i\) is the intracellular concentration of potassium

Potassium equilibrium potentials of around –80 millivolts (inside negative) are common. Differences are observed in different species, different tissues within the same animal, and the same tissues under different environmental conditions. Applying the Nernst Equation above, one may account for these differences by changes in relative K⁺ concentration or differences in temperature.

For common usage the Nernst equation is often given in a simplified form by assuming typical human body temperature (37°C), reducing the constants and switching to Log base 10. (The units used for concentration are unimportant as they will cancel out into a ratio). For Potassium at normal body temperature one may calculate the equilibrium potential in millivolts as:

\[ E_{eq, K^+} = 61.54 \log \frac{[K^+]_o}{[K^+]_i} \]

Likewise the equilibrium potential for sodium (Na⁺) at normal human body temperature is calculated using the same simplified constant. For chloride ions (Cl⁻) the sign of the constant must be reversed (–61.54 mV). If calculating the equilibrium potential for calcium (Ca²⁺) the 2⁺ charge halves the simplified constant to 30.77 mV. If working at room temperature, about 21°C, the calculated constants are approximately 58 mV for K⁺ and Na⁺, –58 mV for Cl⁻ and 29 mV for Ca²⁺.
Goldman-Hodgkin-Katz Equation: Like the Nernst, but has a term for each permeant ion. **Gives us the equilibrium potential for multiple ions!**

Note Chloride, being negative, is i/o rather than o/i.

Do Cardiac pacemaker cells have a resting potential?

Not really, as the membrane potential is always changing and there is never any real rest for these cells. Thus a resting membrane potential is a theoretical concept at best for these cells.
The Synapse

Calcium Channels

Display Selective permeability to calcium. Most commonly thought of as voltage-gated, but there are ligand gated calcium channels as well.

<table>
<thead>
<tr>
<th>Type</th>
<th>Gated by</th>
<th>Protein</th>
<th>Gene</th>
<th>Location</th>
<th>Function</th>
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<tbody>
<tr>
<td>L-type</td>
<td>High voltage</td>
<td>C₉₂,1.1</td>
<td>CACNA1S</td>
<td>Neurons, Skeletal Muscle, ventricular myocytes, bone</td>
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<td>CACNA1C</td>
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<td>C₉₂,1.3</td>
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<td>C₉₂,1.4</td>
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<td>P-type/Q-type</td>
<td>High voltage</td>
<td>C₉₂,2.1</td>
<td>CACNA1A</td>
<td>Neurons</td>
<td>Familial Hemiplegic Migraine, Episodic Ataxia associated with primary generalized epilepsy.</td>
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<td>N-type</td>
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<td>CACNA1B</td>
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<tr>
<td>R-Type</td>
<td>Intermediate voltage</td>
<td>C₉₂,2.3</td>
<td>CACNA1E</td>
<td>Neurons</td>
<td>unknown</td>
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<tr>
<td>T-type</td>
<td>Low-voltage</td>
<td>C₉₂,3.1</td>
<td>CACNA1G</td>
<td>Neurons, Cardiac Myocytes</td>
<td>Are enhanced in several animal models of epilepsy, no monogenetic defects reported yet in humans.</td>
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<td></td>
<td></td>
<td>C₉₂,3.2</td>
<td>CACNA1H</td>
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<tr>
<td></td>
<td></td>
<td>C₉₂,3.3</td>
<td>CACNA1H</td>
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</tr>
</tbody>
</table>
Febrile Seizures

- Convulsion in young children (6 months to 6 years) caused by a sudden spike in body temperature, often from an infection.
- Most common – middle ear infection, roseola (viral infection, rash around neck and swollen lymph nodes).
- Can also be caused by meningitis, encephalitis, and sometimes is seen after some common childhood immunizations.
- Usually occurs on the first day of fever due to the rapid rise in temperature, but can also occur as the fever declines.
- Rapid changes in temperature can impair GABA mediated inhibition and allow the brain to become more prone to seizure activity.
- Febrile Seizures are not indicative of longer term epileptic disorders and occur in as much as 4-6% of the population.

pH and febrile seizures

- pH is a measure of the acidity or alkalinity of a solution.
- Acidosis is said to occur when arterial pH falls below 7.35, while its counterpart (alkalosis) occurs at a pH over 7.45.
- Extra- and intracellular pH (pHo and pHi) are important physiological variables that both reflect and, in turn, influence neuronal function.
- A hyperthermia induced rise in brain pH, alkalosis, enhances neuronal excitability and can precipitate febrile seizures.
- Suppressing alkalosis with 5% CO2 can abolish seizures. This appears to block changes in a potassium conductance (I\text{\text{H}}) that underlie the increase in neuronal excitability.
The Synapse and Neurotransmission

Glutamate

- Most abundant excitatory neurotransmitter.
- Causes cell to depolarize.
- Ionotropic Receptors
  - NMDA receptor
  - Kainate receptor
  - AMPA receptor
- Metabotropic receptor
  - mGluR

Allow Na+/Ca2+ in and K+ out.
Glutamate Channels (ionotropic)

Non-NMDA
AMPA, kainate

NMDA
N-methyl-D-aspartate

AMPA = α-amino-3-hydroxy-5-methyl-isoxazole-4-propionate

Gly = glycine
PCP = phencyclidine

Glutamate receptors and domoic acid intoxication

- Domoic acid (DA) is a naturally occurring marine toxin (from algae *Psuedonitzschia*).
- Often produced in algae/plankton blooms.
- DA binds to and activates glutamate receptors (NMDA and Kainate).
- Increased activity can produce seizures and increased intracellular calcium leads to cell death, particularly in limbic structures.
**GABA: γ-aminobutyric acid**

Drugs that act as agonists of GABA receptors (known as GABA analogues or GABAergic drugs) or increase the available amount of GABA typically have relaxing, anti-anxiety and anti-convulsive effects. Many of the substances below are known to cause anterograde amnesia and retrograde amnesia (i.e. ethanol, barbiturates, benzodiazepines).

Antagonists block inhibition and can promote seizure activity. Sub-convulsive doses may have benefits for Parkinson’s, motion sickness, improving memory, etc.

GABA<sub>α</sub>: the fast response of neurons to GABA that is blocked by bicuculline and picrotoxin is due to direct activation of an anion channel.

GABA<sub>β</sub>: A second type of ionotropic GABA receptor, insensitive to typical allosteric modulators of GABAA receptor channels such as benzodiazepines and barbiturates.

GABA<sub>γ</sub>: A G-protein coupled receptor that mediates the slow response to GABA.

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**Acetylcholine**

- Acetylcholine has functions both in the peripheral nervous system (PNS) and in the central nervous system (CNS) as a neuromodulator.
- In the PNS, acetylcholine activates muscles, and is a major neurotransmitter in the autonomic nervous system.
- In the CNS, acetylcholine and the associated neurons form a neurotransmitter system, the cholinergic system, which tends to cause excitatory actions.
- Cortical acetylcholine (ACh) release is low during non-rapid-eye-movement (NREM) sleep and highest during the electroencephalographic (EEG) activation of wakefulness and REM sleep.
- The volatile anaesthetic halothane decreases cortical ACh and induces 7- to 14-Hz spindles in the cortical EEG.
- Nicotinic acetylcholine receptors (nAChR) are "ionotropic" receptors particularly responsive to nicotine.
- Muscarinic acetylcholine receptors (mAChR) are "metabotropic" receptors particularly responsive to muscarine. They are G-protein coupled receptors.
Rebound Excitation

- Alcoholism is associated with increased inhibition (GABA agonist) and decreased excitation (NMDA antagonist)
- Withdrawal -> GABA inhibition decreased and excitatory tone increased -> can lead to withdrawal seizures.

Ach Receptors

- Nicotinic Receptors:
  - \( \alpha_4, \alpha_7, \) and \( \beta_2 \) subtypes in CNS.
  - Carbachol, nicotine are agonists
  - \( \alpha \)-bungarotoxin, \( \alpha \)-conotoxin, tubocurarine are antagonists.

- Muscarinic receptors: 5 subtypes (M1-M5)
  - M1 and M5 most common in CNS.
  - Atropine and derivatives are antagonists
  - Carbachol, Oxytremorine are agonists
ACh and Human EEG

- Cortical Ach release is highest during wakefulness and REM sleep.
- Lowest during NREM sleep when see sleep spindles.
- Anaesthetics suppress EEG activation and cortical Ach release. i.e. Halothane induces 7-14 Hz spindles, similar to those seen in NREM sleep, and reduces cortical Ach.