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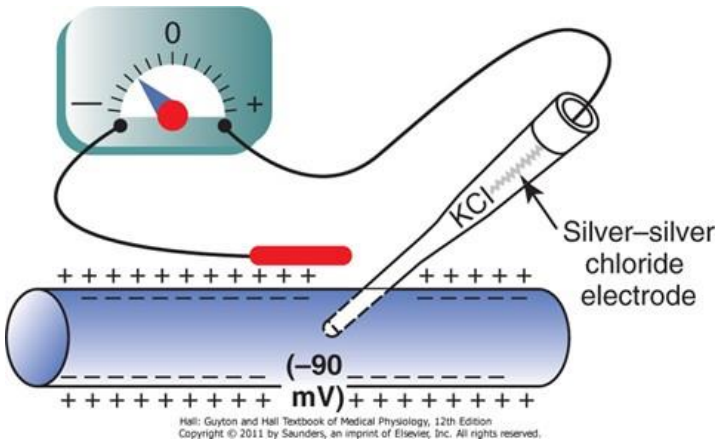
This information was not included in the handout so we mentioned everything new about the e-lecture in these 2 pages …. Please study them after the main handout.

Silver-silver chloride electrode is used to measure membrane potential.



Sodium channels in Action Potential

a) Activation of sodium channel
When the membrane potential approaches -70mv to -50mv
The activation gate undergoes a conformational change and open,
Sodium permeability may increase from 500 to 5000 folds

b) Inactivation of the sodium channel
Same increase in voltage that opens the activation channel closes the inactivation channel
It closes slower than the activation gate opens, the activation gate will no reopen until the membrane potential returns to or near the original resting potential

Initiation of AP
Positive feedback opens Na+ channels
Threshold→ sudden rise by 15-30 mv

Cardiac muscle
Rhythmicity of excitable tissues- repetitive discharge, repetitive self-induced discharge can occur in the heart (rhythmic beat), intestine(peristalsis), and rhythmic control of breathing.
Re-excitation process necessary for spontaneous rhythmicity
The following sequence occurs

1) Some Na+ as Ca2+ ions flow inward
2) This increases the membrane voltage in the +ve direction, which further increases permeability
3) Still more ions flow inward
4) Permeability increases until an Action Potential is generated

Nerve cell
Cellbody= soma
Function of myelin sheath

1) Insulator
2) Increase conduction velocity

Resting membrane potential

1) Rmp is 100 times more permeable to K+ than Na+. K+ tends to leak out of the cell down its concentration gradient, carrying a +ve charge with it. (through Na+ leak channels)

2) Non-diffusible anions (proteins. Sulphate and phosphate ions) cannot leave the cell.

3) Very small amount of Na+ diffuses into the cell down its concentration gradient. The membrane is only slightly permeable to Na+. (through na+ leak channels)

4) Na+-K+ pump maintain concentration gradient of K+, and Na+ between the two sides of the membrane

-oscillioscope to measure rapid changes in membrane potential

Myelinated nerve fibers conserve energy for axon because only nodes depolarize as well as increasing the velocity of conduction of nerve impulses

Multiple sclerosis
-autoimmune disease
-usually young adults
-Blindness, problems controlling muscles ➔ ultimately paralysis
-Immune system attacks myelin sheaths and nerve fibers

- Scar tissue(sclerosis) replaces some damaged cells
- Other now unmyelinated axons sprout Na+ channels

-synaptic gutter has subneural folds to increase surface area. Has Ach gated channels (where Ach blind) at motor end plate in
-post-synaptic membrane
-synaptic cleft (filled with ECF and Ach esterase enzyme)

1)Drugs that act on the neuromuscular junction

Drugs that act on muscle fiber by Ach like action:

Methacholine-Carbacol-Nicotine
They act for minutes to hours since they are not broken down by ach esterase enzyme
2)Drugs that block transmission at neuromuscular junction
Curare and curariform like drugs
Act by competitive inhibition to Ach at its receptors and can not cause Depolarization

3) Drugs that stimulate transmission at neuromuscular junction by inactivation of Ach esterase enzyme:

A-Neostigmine, prostigmine and physostigmine ➔ inactivates Ach esterase enzyme temporarily
B- di-idopropyl florophosphate (nerve gas poison) inactivates Ach esterase enzyme for days and weeks --------death because of respiratory muscle spasm
Myasthenia Gravis
-Diseases of adult females affects eyelid, extra ocular. Bulbar and proximal limb muscles
-presents with ptosis, dysarthria, dysphagia, and proximal limb weakness in hands and feet.
- an autoimmune disorder
- the body forms antibodies against Ach receptors. Patients have 20% of number of Ach receptors.
-the EEPs are too small to trigger action potentials and muscles can not contract.
----Treatment----
Administration of an inhibitor of acetyl cholinesterase temporarily
-prostigmine or neostigmine
-allowing more Ach to remove at the neuromuscular junction to bind to the remained Ach receptors. And allow contraction

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Medical students, 2018/2019
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Review: Membrane physiology and the basis of excitability

Ref: Guyton, 13th ed. pp: 61-71. 12th ed. pp: **57-69**, 11th ed: **p57-71**, 10th Edition, **p52-66.**

MEMBRANE POTENTIALS AND ACTION POTENTIALS:

MEMBRANE POTENTIAL:

If we assume that a cellular membrane is permeable **only** to K+, which is found in a very high concentration inside the cell. K+ will diffuse to the extracellular fluid because of the concentration gradient. The diffusion of K+ will result in a movement of positive charges outside the cell and leaving behind negative charges inside the cell. This will create an electrical potential difference across membrane (positive outside and negative inside). Creation of this potential difference will oppose diffusion of K+ to the outside at a certain concentration difference. When you reach a point at which diffusion of K+ is completely opposed by the potential difference created across membrane and the net diffusion for K+ is zero even though you still have a concentration gradient, you have reached the equilibrium potential for K+ (E_K). The equilibrium potential for any univalent ion at normal temperature can be calculated by Nernest equation:

E (mV) = - 61.log (Ci/Co)

E = equilibrium potential for a univalent ion
Ci = concentration inside the cell.
Co = concentration outside the cell.

When more ions are involved in creating the potential, we can calculate the potential according to Goldman-Hodgkin-Katz equation.

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

P = permeability of the membrane to that ion.

In this equation, Goldman and his colleagues considered that these ions are mostly involved in the development of membrane potential.

According to this equation the permeability of the membrane to an ion is very important in determining the membrane potential. If the membrane is permeable only to K+ and not permeable to Cl- and Na+, the membrane potential will be equal to E_{K+}.

Resting membrane potential:

In excitable cells the membrane potential is not constant. When the cell is stimulated, the membrane potential changes. These changes in membrane potential are due to changes in permeability of plasma membrane to different ions. For example, when neuron is stimulated, this will result in increased permeability to Na+. This will bring the membrane potential closely to E_{Na}. The recorded membrane potential for a cell under resting conditions when no stimulus is involved is known as **resting membrane potential**. For neurons the recorded resting membrane potential is about (– 90 mV). This represents a potential difference between the inside to the outside when neuron is not active.

Origin of resting membrane potential:

Contribution of K+ diffusion:

As mentioned earlier, if the membrane is permeable only for K+ the calculated E_{K+} is about (-94mV).

Co_{K+} = 4meq/l , Ci_{K+} = 140meq/l

E_{K+} = -61. log 140/4 = -94mV

Which is not far from the recorded membrane potential but not exactly.

The contribution of Na+ diffusion:

Membrane is also permeable to Na+. The permeability of the plasma membrane for Na+ is much less than that of K+. If the membrane is permeable only to Na+, the calculated E_{Na+} = + 61mV.

…….. (Co_{Na+} = 142meq/l , Ci_{Na+} = 14meq/l).

Because of the permeability of the membrane for the two ions, the E would be between (-94mV and +61mV). The calculated E for the two ions is –86mV, which is not far from the E_{K+} because of the higher permeability of membrane for K+ than for Na+ (100 times more).

So the Na+ contribution in resting potential is by bringing the membrane potential to a lower value than the calculated E_{K+}.

Contribution of Na+ - K+ pump:

As mentioned earlier, this pump is electrogenic. It moves more positive charges outside the cell (3 for 2). This will induce loss of positive charges from the cell and bring the membrane potential to a higher negativity (about –4mV additional negativity).

Therefore all these factors, during **rest**, will give a net membrane potential of –90mV (called **Resting Membrane Potential**).

ACTION POTENTIAL:

As we have seen, the plasma membrane is **polarized** (has ability to separate opposite charges) during resting state. When the membrane potential decreases (becomes less negative), the membrane is in **depolarization** stage. While the change in membrane potential in opposite direction (becomes more negative than resting potential) is known as **hyperpolarization**.

When a cell is depolarizing, it reaches a maximum according to stimulus, then the membrane potential returns to its resting state. The phase of returning from depolarized state to resting state is known as **repolarization**. These changes in membrane potential can be recorded by placing one electrode inside the cell and the other out side the cell. By recording of whole action potential in this way, we will obtain a **monophasic action potential**.

Let us consider the changes in membrane potential of an excitable cell to understand the events that appear during changes of membrane potential. To induce a change, a stimulus must be applied to change activity of channels at the membrane. Any increase in permeability of membrane to Na+ will result in diffusion of (+) charges inward. This event will decrease the membrane potential (becomes less negative). And conversely any increase in K+ diffusion (movement outward) will result in an increase in membrane potential (becomes more negative). The diffusion of these ions depends on the activity of Na+ and K+ channels that are found on the membrane. Activation of Na+ channels will induce depolarization, while activation of K+ channels will increase the potential difference across membrane.

Action potential and the role of Na+ channels:

On the membrane, most Na+ channels during resting state are inactive (closed). According to channel type, these channels can be activated by a chemical stimulus (in case of chemical gated channels), electrical stimulus (in case of voltage gated channels), or mechanical stimulus. In the case of chemical gated channels, binding of ligand to its receptor will induce activation of chemical gated Na+ channels. Once activated, the membrane potential will decrease (becomes less negative). Which means that the membrane depolarizes. The voltage changes in the membrane will cause the other type of channels (Na+ voltage gated channels) to be activated. Activation of these channels will cause more changes in membrane potential (more depolarization). More and more depolarization will occur in the membrane by a positive feed back mechanism. If we reach a point at which most voltage gated Na+ channels are activated, this will cause a sudden increase in Na+ permeability. This increase in Na+ permeability will even reverse the membrane potential (becomes positive inside and negative outside) (this is known as the **overshot** in the action potential), because Na+ is trying to approach its equilibrium potential (E_{Na}). At this point membrane has reached maximal changes in membrane potential (a peak of an action potential).

As we have seen, during depolarization there is a point at which a sudden increase in Na+ influx which induces rapid and maximal change in membrane potential. This point is known as **threshold** of an action potential. The rapid change in membrane potential during the raising phase of an action potential is known as **firing stage**. When a stimulus causes a depolarization that brings the membrane potential to the threshold, the membrane will respond by the firing stage of an action potential. If depolarization in the membrane has not reached threshold, the membrane will not enter firing stage, and instead, the potential returns to its resting level. Therefore the response in the membrane will be either by an action potential when threshold is achieved or no appearance of an action potential when the membrane potential has not reached threshold. For that reason induction of an action potential in excitable cells follows the **NONE OR ALL PRINCIPLE**.

The voltage changes in membrane potential not only activate voltage dependent Na+ channels, but also inactivate these channels at certain potential difference. This inactivation appears because channels have changed their state from opened channels to closed channels due to voltage changes. The closing event of Na+ channels does not make these

channels as the only responsible for bringing membrane potential to its resting level. But also, activation of voltage dependent K+ channels is the main player in returning the membrane potential to its resting level.

Action potential and K+ channels:

Although there is some leakage of K+ during resting state, which maintains the resting membrane potential close to $E_{K+,}$ depolarization causes activation of voltage gated K+ channels. The activation of these channels is much slower than activation of Na+ channels. This results in a delay in the maximal activation of K+ channels.

The delayed activation of K+ channels combined with inactivation of Na+ channels will result in a rapid returning of the membrane potential to its resting level causing the **falling phase** in the action potential. The membrane potential may go for a while to more negative potential than during resting potential, which is known as **positive afterpotential** (**after hyperpolarization**). Followed by a full recovery in membrane potential (returns completely to its resting level). The positive after potential is probably due to an excess in K+ efflux, which causes more deficit of positive ions inside the cell.

Action potential and Ca++:

As discussed before, the raising phase of an action potential results by fast activation of Na+ channels. These are called *fast channels*. In some excitable cells, like cardiac muscle and uterine muscle, cells are equipped with another type of channels known as *slow* Na+ – Ca++ *channels*. These channels are activated at slower rate than Na+ channels. The slow and prolonged opening of slow channels will cause mainly Ca++ to enter the cell and prevents the rapid fall induced by activation of K+ channels, and the membrane potential is maintained for a while then the potential falls to its resting level. This is known as **plateau** in action potential. The presence of plateau in this type of cells is important in prolonging the time of an action potential, giving more time for the cell to be able to respond to another stimulus, because the cell remains longer time in **refractory period**.

Refractory periods of an action potential:

During action potential the cell is not able to respond to another stimulus. From the firing stage to the end of first third of falling phase the cell will not respond at all even by a stronger stimulus. In this stage the cell is said to be in **absolute refractory period**. From the beginning of the second phase until the resting membrane potential is achieved the cell cannot respond the usual stimulus, but a stronger stimulus can change the

membrane potential. In this period the cell is in **relative refractory period**.

The periods depend on the activity of Na+ channels. These channels pass three states during action potential. During resting potential, Na+ channels are **closed but capable for opening** when stimulated. During the raising phase (firing), almost all Na+ channels are **opened**. And any other stimulus (even stronger one) will not cause activation of more Na+ channels. During this period the membrane is in absolute refractory period.

In the third state, when voltage dependent Na+ channels become closed after the membrane potential has reached positive values. At this state Na+ channels are not capable for opening. During all the falling phase of an action potential, these channels remain **closed and not capable for opening**. They can pass to the first state (closed and capable for opening) when the membrane potential returns to its normal level or to a more negative potential than resting potential. During this period, the membrane is in relative refractory period. This means that a stronger (suprathreshold) stimulus may activate the closed channels that are not capable for opening by normal stimulation. In addition to the role of voltage gated Na+ channels in establishing the relative refractory period, the presence of widely opened K+ channels during falling phase, which cause excess flow of positive charges to the outside, may also play a role by opposing stimulating signals.

Na+ -K+ pump and action potential:

This pump has **no** role in the electrical activity that are taking place during action potential. But it plays an important role in restoring ionic composition that has been altered during action potential. This role is important in maintaining the ionic composition of the intra-and the extra-cellular fluids.

Nerve Cells (Neurons)

The nervous system is formed of neurons and supportive cells. A neuron, typically consists of 3 basics parts: **cell body, dendrites, and axon** (or nerve fiber). Dendrites are short projections from the cell body, which receive inputs from neighboring neurons. Axon is a long tubular like structure which projects from cone-shaped elevation in the cell body known as **axon hillock** (means small hill). The impulse begins at the junction between axon hillock and the initial segment of axon. Axon ends

into fine processes called axon terminals. Some of these terminals end with a bulb-shape structure called **synaptic end bulb (synaptic knob)**, where neurotransmitter is stored in vesicles and ready for the release.

Many classifications for neurons are known, according to shape, function, neurotransmitter they release, myelination, location…etc.

Supportive cells and function (NEUROGLIA):

Many types of supportive cells around neurons have been described (at least 6). Microglia, Astrtocytes, oligodendrogliocytes have been shown around neurons from the CNS. And glial cells which are similar to astrocytes from the CNS have been described in the neural network of the GI tract.

These cells perform the following functions:

*Maintenance of neural environment.

-uptake of K+ and neurotransmitters from the interstitial fluid around the neurons.

*Synthesize and release neurotrophic factors → maintain the survival and protection of neurons

* Other specialized supportive cells are responsible for myelination of axons. In the CNS these cells are oligodendrogliocytes. In the peripheral nervous system, these cells are known as **Schwan cells**. These cells wrap around axon segments and secrete myelin sheath (a protein lipid complex that insulates nerve fiber). There are gaps in myelin sheaths known as **nodes of Ranvier**, which appear at intervals along axon. These gaps are used for transmission of impulse along myelinated nerve fiber.

TANSMISSION OF ACTION POTENTIAL ALONG NERVE FIBERS:

Once action potential is generated at the axon hillock, no more triggering events are needed to activate the whole nerve fiber (axon). The generated impulse is conducted along the nerve fiber by one of the followings 2 methods of propagation:

1. Continuous conduction (conduction by local current flow): occurs in unmyelinated fibers. Local currents flow between the active area, which is at the peak of action potential and inactive area, which is still in resting potential. This flow will cause activation of Na+ channels in inactive area and reduce the

membrane potential to the threshold, which triggers an action potential in this area (that was previously inactive).
This process is repeated all along the nerve fiber until the impulse has reached nerve terminals.

2. Saltatory conduction: In myelinated fibers the impulse skips the myelinated regions in the axon and jumps from one node of Ranvier to the adjacent node. This process ensures faster propagation of an action potential along the myelinated axons (50 times faster than in unmyelinated fibers of the same size). The conduction also involves current flow between two adjacent nodes of Ranvier, which results in activation of Na+ channels in the adjacent node, which is still in resting potential. The process is repeated until the impulse activates the axon terminals.

Note: current flow in both types of conduction is from the **positively charged to the negatively charged regions at both sides of the membrane**, and the membrane has high resistance to the passage of current flow across it (**no** current flow can pass through the membrane).

Not only myelination can influence the velocity of conduction, but also the diameter of nerve fibers. Larger fibers conduct impulse with higher velocity.
Nerve fibers have been classified in (A, which includes as subtypes (α, β, γ, δ) fibers, B, C). The diameter and the velocity of conduction is the highest in Aα, and is the lowest in C fibers.

The importance of refractory periods in conduction:

The presence of refractory periods during action potential is very important in the conduction of impulse. The refractory periods ensures the **one-way (unidirectional)** propagation of action potential. Once an area has developed an action potential, the previous region is still under refractory period (unresponsive area). This area will not develop another action potential. But the following area that is at resting potential is capable to initiate an action potential.

SYNAPSES AND INTEGRATION OF RESPONSES:

Synapses:

Neuron may terminate at one of three structures: a neuron, a muscle, or a gland. The junction between 2 neurons is known as synapse. The first neuron ends with end bulb (**synaptic knob**), where neurotransmitters are stored in vesicles and ready for the release. The membrane of the synaptic knob is known as **presynaptic membrane**. When secretory vesicles fuse with presynaptic membrane, they release their content into a small space between two membranes known as **synaptic cleft**. The released transmitters act on the second neurons by binding to their receptors at the second membrane, which is called **postsynaptic membrane** (**subsynaptic membrane**).

Synapses operate in one direction. Transmit signals from one neuron to adjacent neuron. When the impulse from the presynaptic neuron reaches the synaptic knob, this will cause activation of voltage dependent Ca++ channels. This will result in Ca++ diffusion into the synaptic knob. The increase in Ca++ concentration inside axon terminal will trigger the release of neurotransmitter from vesicles into synaptic cleft by a process of exocytosis. Inactivation of synaptic knob by inhibitory inputs that may synapse with the membrane at the nerve terminal may induce inhibition of the release of transmitter. This inhibition that appears at this site reduces the effectiveness of transmission in the synapse. This type of inhibition is known as presynaptic inhibition.

Once released, neurotransmitter binds to its receptor at the postsynaptic membrane. According to transmitter – receptor combination, this will induce either a decrease in membrane potential (depolarization) or increase in membrane potential (hyperpolarization). When there is a decrease in membrane potential, the developed postsynaptic potential is called **EPSPs** (**E**xcitatory **P**ost **S**ynaptic **P**otentials), while the increase in membrane potential is called **IPSPs** (**I**nhibitory **P**ost **S**ynaptic **P**otentials).

After inducing the appropriate response at the postsynaptic membrane, the transmitter is inactivated or removed leaving the postsynaptic membrane ready to receive additional messages from the same presynaptic membrane. The inactivation of transmitter takes place by postsynaptic membrane bound enzymes. An example of these enzymes is *acetylcholine esterase*, which destroys acetylcholine (Ach) into acetyl and choline molecules, which then transported back to the synaptic knob, where they combine again to form new Ach molecules. Some types of transmitters are transported back, without inactivation, into

synaptic knob. Conditions that alter the activity of destroying enzyme, uptake of transmitter by nerve terminal, or induce release of high concentration of transmitter (presynaptic facilitation) alter the activity of synapse by prolonging the activation of receptors at the postsynaptic (subsynaptic) membrane. In addition to that, some drugs may combine with receptor and prevents binding of transmitter to its receptor. These drugs are known as **blockers**. An example of these is hexamethonium, which can bind to acetylcholine (Ach) receptor at postsynaptic membrane and prevents Ach from binding. This will inhibit transmission induced by Ach neurons.

The EPSPs are not action potentials. They are small depolarization (subthreshold potential) that can be induced by activation of few Na+ channels.

The IPSPs are usually induced by activation of K+ channels. Which result in efflux of K+ and change in the membrane potential to more negative potential. Some transmitters activates Cl- channels, the activation of these channels will not induce hyperpolarization (during rest, neural cell is near the E_{cl}, and the opening of Cl- channels will not induce inward diffusion of Cl-). But this activation is inhibitory on neural activity. This inhibition is achieved by holding the membrane at its resting potential and preventing depolarization.

The time it takes for a signal from the first neuron to induce changes in membrane potential in the second neuron is known as **synaptic delay**.

Integration of responses at postsynaptic membrane:

Usually, the complexity of neural network connections permit synapsing of many axonal terminals from different neurons to one neural cell body (called **convergence**), and branching of one nerve fiber to many terminals that synapse to different neurons (**divergence**). This complexity results in converting the signal from one neuron to many postsynaptic neurons in the case of divergence, and many inputs from presynaptic neurons can be received by single postsynaptic neuron in the case of convergence.

As mentioned before, one stimulus may induce depolarization or hyperpolarization at the postsynaptic membrane. The induced depolarization is not an action potential, but it is a subthreshold potential. The action potential will develop only when threshold is achieved. In neural network, to have subthreshold potentials eliciting an action potential, **summation** (two depolarizations can sum to elicit a higher

depolarization) must take place between responses at the postsynaptic membrane.

Two types of summation are known at the postsynaptic membrane. **Spatial summation** appears when 2 or more responses from 2 or more different neurons have appeared simultaneously (at the same time) at the same site of postsynaptic membrane, which result in summing of these responses into a final response. This summation can take place between 2 or more IPSPs to elicit more hyperpolarization, two or more EPSPs to elicit more depolarization in the membrane, or between excitatory and inhibitory potentials which results in cancellation of potentials and induce postsynaptic inhibition.

The second type of summation is called **temporal summation**. Appears when 2 or more postsynaptic potentials, which were elicited by **one** presynaptic neuron at different times, sum to induce more depolarization in the membrane potential. In this case, the repetitive excitation of postsynaptic membrane from a single input induces a higher depolarization that may elicit an action potential at the postsynaptic membrane.

Recordings of action potential:

Recording of **monophasic action potential** is by placing one electrode inside the cell and the other electrode outside the cell. While a different configuration of an action potential can be obtained by placing the two electrodes outside the cell membrane. The later recording is known as **biphasic action potential**. Two waves are obtained in the recording of biphasic action potential, the first represents depolarization, and the second is in the reverse direction of the first and represents repolarization.