Introduction to Neurobiology

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Module 5: Ion Channels Part 2

Diversity of ion channels in central neurons

The simplified account of action potential generation is that the action potential begins with the activation of sodium channels which depolarize the membrane and end when sodium channels close and potassium channels repolarize the membrane. This account is accurate only in a general sense. All neurons in the brain possess many more types of voltage-gated channels which in turn display a staggering variety of properties. This diversity confers different signaling characteristics (threshold for action potential initiation, firing frequency, spike pattern, etc.). In some sense one can regard each neuron type as having an electrical "signature". In this cclass, we will not consider all types of voltage-gated channels. We will focus on a small subset of ion channels that are important for determining the active properties of central neurons. We can capture the essential functional properties of ion channels in many ways, and some of the most important are:

1. *Voltage dependence of activation*: The primary determinant of an ion channel's function is its voltage dependence of activation. This relationship describes what proportion of ion channels is open at each particular membrane voltatge.

2. *Rate of activation*: In addition to knowing how voltage affects the probability of channel opening, it is important to know how quickly a population of channels activate. Activation rates cover a broad range. Some channel types activate within a millisecond, while other channel types can take tens of milliseconds to activate. The activation rate determines how quickly the conductance through that channel type can contribute to changes in membrane potential.

3. *Rate of deactivation*. When a change in membrane voltage causes channels to open, once the membrane voltage returns to a potential where the channels are not activate, it can take some time for all of the channels to close. This process is known as deactivation.

4. Rate of inactivation. After activation, some types of channels will stop conducting ions after a period of time, even when the membrane voltage remains within the channels activation range. This process is known as inactivation and it occurs through a mechanism that is different from deactivation. Inactivation usually involves the movement of part of the channel protein into the channel pore, thus blocking ionic movement through the pore. Sodium channels commonly display inactivation of this type.

5. Recovery from inactivation. Once a channel has inactivated, the only thing that will relieve the inactivation is repolarization of the membrane to a potential low enough to allow recovery from inactivation. Once the membrane reaches a potential that allows recovery, the process of recovery from inactivation takes time to complete. The inactivation and delay associated with recovery from inactivation in sodium channels gives rise to the refractory period following the action potential.

Below is a description of only some of the ion channel types that are expressed by hippocampal neurons, along with descriptions of their relevant functional properties.

Voltage-gated sodium channels

Central neurons express several types of sodium channels. For our purposes, we will lump all of them together in a single category because of their functional similarities. The defining features of sodium channels are that the resulting sodium currents (INa) activate rapidly and inactivate rapidly, with recovery from inactivation requiring delays of a few milliseconds to tens of milliseconds.

Voltage-gated calcium channels

Central neurons also express several types of calcium channels that are activated when the membrane is depolarized. These channels have diverse functional properties, but we shall simplify their actions here quite a bit. All calcium channels open with depolarization and becaiuse E_{Ca} is very high (100+ mV) the driving force on calcium is always into the cell, so open Ca channels allow inward Ca++ current. Importantly, the single-channel conductance for calcium channels is 50-100 times lower than the conductance of sodium channels, so calcium channels typically have much less effect on the membrane potential than sodium channels. The key significance of calcium channels is that even very small influxes of calcium can have dramatic effects on other cellular processes, including other ion channels.

We will focus for now on two categories of calcium channels although many others exist. One category of calcium channels activates only at high membrane potentials (above -30 mV). These high-voltage activated (HVA) Ca channels are distinguished from low-voltage activated (LVA) Ca channels that activate at membrane potentials over -60 mV. One type of LVA calcium channel is known as the **T-type calcium** channel, because the researchers who discovered it found that its conductance was tiny and it produces a transient current (this is an inactivating channel). A second type of calcium channel is an HVA channel known as the **L-type calcium** channel because it has a large conductance and it makes a long lasting current (it shows very little inactivation).

Calcium channels are found in almost all neurons, and the important point here is that calcium channels open during the action potential and allow calcium influx into the cell. The influx of calcium plays a relatively minor role in changing the membrane potential, but calcium entering the cell has a profound effect on other cellular processes, especially other ion currents as you will see below.

Voltage-gated potassium channels

In contrast to sodium and calcium channels, potassium channels are an extremely diverse group, with pronounced differences in their functional characteristics. Dozens of

different potassium channels are expressed in the nervous system. We will consider a small subset of these channels that are often expressed in hippocampal neurons and play prominent roles in shaping their active properties.

1. **Delayed rectifier ($I_{K(DR)}$).** A high voltage-activated (HVA) potassium channel. This channel type is a major component of action potential repolarization in many cells. This channel type activates relatively rapidly and shows little inactivation (Figure 6) . At the molecular level, subunits from many different potassium channel families may give rise to this electrophysiological phenotype.

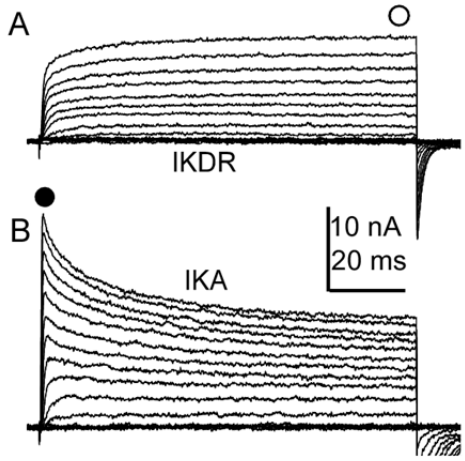

Figure 1. A) Recordings of IKDR in response to depolarizing voltage steps. B) Recordings of IKA in response to depolarizing voltage steps.

3. **A-type K^+ channel (I_{KA})**. A low voltage-activated (LVA) K^+ channel. This channel displays very fast activation and strong inactivation (Figure 6). This type of channel often plays a role in keeping action potentials very brief, and enabling rapid bursts of action potentials by rapidly repolarizing the cell. This channel typically consists of members of the Kv4 family of K^+ channels in mammals, or the*Shal* family in drosophila. Like D-type channels (described below), A-type channel activation occurs at potentials near resting potential and it therefore can result in a transient suppression of action potential firing. However, its inactivating properties render the channel's influence sensitive to the cell's prior level of depolarization (e.g. see Fig. 7).

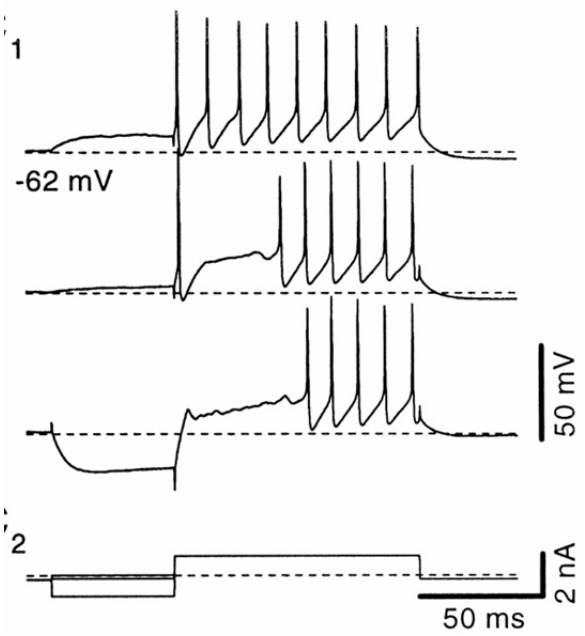

Figure 2. Effect of A-type channels on the firing pattern of a central neuron. With prior membrane depolarization (via a small somatic current pulse), the cell fires a regular train of action potentials in response to a larger depolarization (resting potential=dotted line). However, a pause in action potential firing is observed in response to the same depolarization when inactivation of A-type potassium channels is removed by prior hyperpolarization (lower trace). Adapted from Kanold and Manis (1999)

2. **D-type potassium channel (I_D)**. Another LVA potassium channel. This channel is fast activating and slowly inactivating. It was first named for its tendency to *delay* the time to action potential firing during membrane depolarization. This channel typically consists of members of the Kv1 family of potassium channels in mammals, or the *Shaker* potassium channel family in drosophila.

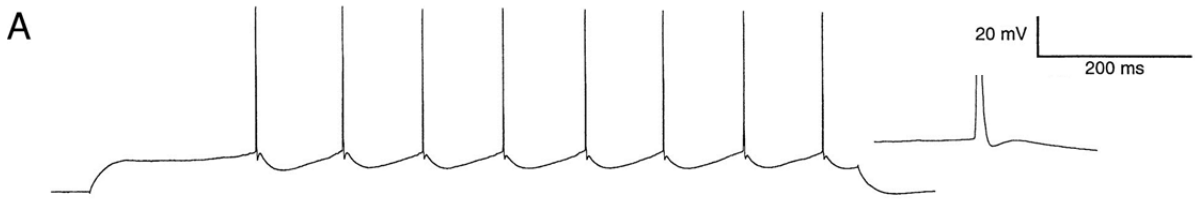

Figure 3. Delayed firing of a pyramidal neuron in the hippocampus. D-type potassium channel contribute to the initial suppression of firing. Note the "shoulder " of the initial depolarization, caused by the activation of potassium channels.

4. **M-type K^+ channel (I_M)**. This LVA K^+ channel activates very slowly, deactivates very slowly, and does not inactivate (Figure 9). This current activates too slowly to shape the action potential waveform. Instead, this current contributes to changes in firing frequency during bursts of action potentials (Gu et al., 2005).

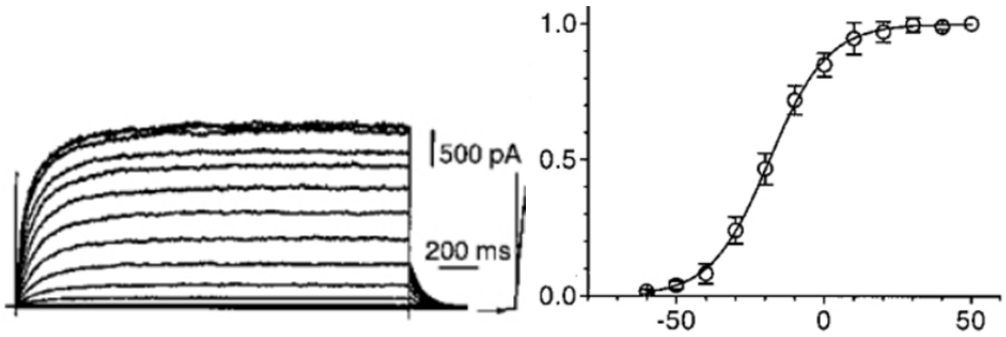

Figure 4. Left panel: A series of currents from M-type K^+ channels elicited by depolarizing voltage steps Right: Voltage dependence of I_M activation. Adapted from Selyanko et al. (Selyanko et al., 2000).

Calcium-activated potassium channels

In addition to the numerous voltage-gated potassium channels, central neurons often express potassium channels that are activated by elevations in intracellular calcium. Thus, calcium that enters the cell through voltage-gated calcium channels during the action potential can, in turn, activate potassium channels. We will consider three types of calcium-activated potassium channels that are often expressed in hippocampal neurons.

SK Channels. The SK channel (small-conductance K^+ channel) is activated by intracellular calcium. Thus, calcium that enters the cell during the action potential activates the K^+ conductance of these channels. Because these channels are activated by internal calcium, the SK conductance will remain active as long as intracellular calcium is elevated. These channels activate too slowly to contribute to AP repolarization, but the current through SK channels (I_{SK}) is responsible for the medium AHP in some cells. In these instances, ISK can contribute to early spike frequency adaptation (Faber and Sah, 2003).

I_{sAHP} Channels. Numerous laboratories have confirmed the presence of a calcium-activated K^+ conductance that develops slowly after a train of action potentials and lasts several from hundreds of milliseconds up to 3 seconds. Surprisingly, the molecular identity of these channels has not yet been determined (Faber and Sah, 2003). This current is responsible for the slow afterhyperpolarization that follows trains of action potentials. This slow AHP contributes to late spike frequency adaptation.

BK channels. The BK channel is unique among ion channels in that it is activated by *both* voltage and calcium (figure 10) (Cui et al., 1997). Under physiological conditions, the activation of BK channels is an interactive function of intracellular calcium levels and membrane potential. One prominent function of BK channels is to accelerate repolarization during the late stages of the action potential. These channels thus keep spike durations short. Some varieties of BK channel show strong inactivation. This inactivation leads to spike broadening during long trains of action potentials.

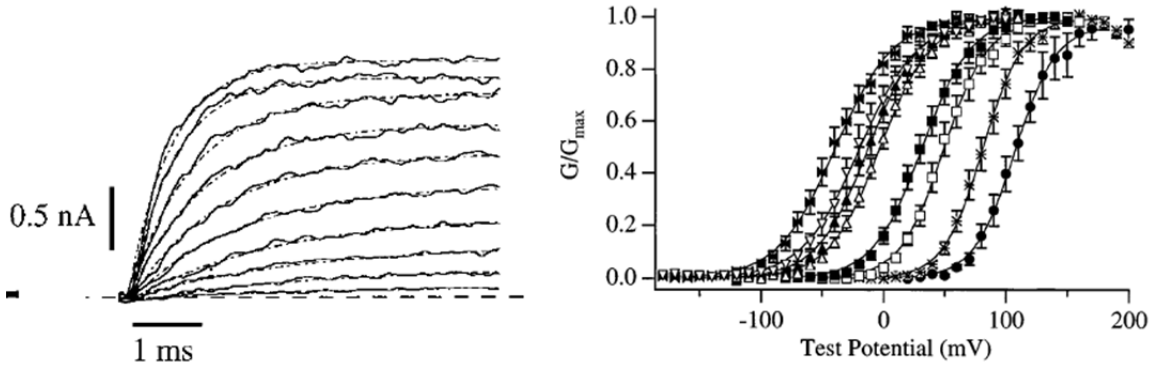

Figure 5. Left panel: A series of currents from BK channels elicited by depolarizing voltage steps in 10 uM Ca++. Right: Voltage dependence of BK activation at different intracellular Ca++ concentrations ranging from 1000 μM (filled bow-ties at left) to 0.84 μM (filled circles at right). Adapted from (Cui et al., 1997).

Biophysical steps underlying action potential generation.

The action potential is generated through the coordinated activation of both voltage-gated sodium and potassium channels. Should depolarization of the membrane during synaptic activity be sufficient to activate a significant local population of sodium channels, the flow of events leading to action potential generation is as follows:

1. Action potential upstroke: sodium channel activation.
Remember that the equilibrium potential for sodium in neurons is approximately +40-50 mV. When voltage-gated sodium channels open, the electrochemical driving force on sodium is to flow inside the cell, producing a regenerative depolarization that makes up the upstroke of the action potential. During the upstroke, more and more sodium channels are recruited.

2. Action potential repolarization. Without a mechanism to oppose sodium channel activation, the membrane potential would be locked permanently at the equilibrium potential for sodium, at about +40-50 mV. However, two mechanisms terminate membrane depolarization.

2A. Sodium channel inactivation. Following activation, sodium channels rapidly enter an inactivated state. You should remember from your coursework that inactivation is biophysically distinct from channel deactivation. During inactivation, a charged, intracellular portion of the channel protein becomes attracted to charges now exposed in the open pore, and plugs the channel (the “ball and chain” hypothesis). By contrast, deactivation requires repolarization of the membrane, whereupon the transmembrane portion of the channel changes conformation and no longer permits the flow of sodium ions.

2B. Potassium channel activation. During the upstroke of the action potential, voltage-gated potassium channels are activated in addition to sodium channels. The equilibrium potential for potassium is about –90 mV, far away from the peak of the action potential (around +40 mV). Thus the electrochemical driving force strongly drives potassium out of the cell, leaving net negative charges and hyperpolarizing the cell membrane. Fortunately for all living things possessing nervous systems, the speed at which potassium channels activate is slightly slower than that of sodium channels. This allows sodium channels a brief window of time in which to generate the upstroke of the action potential. Without this mismatch in activation timing, the sodium and potassium currents would cancel out, and the neuron would be inexcitable!

In addition to the activation of voltage gated potassium channels, the entry of calcium during the action potential often activates BK channels, which contribute to the late phase of action potential repolarization.

3. The afterhyperpolarization (AHP). Following the action potential or a train of action potentials, AHPs occur on at least three timescales. The fast AHP is produced by K^+ currents through BK channels. The medium AHP is regulated by M-type K^+ channels or SK channels, and the slow AHP is produced by I_{sAHP}, a slow calcium-activated K^+ current.

Summary: multiple ion currents interact to shape the AP waveform and AHP characteristics .

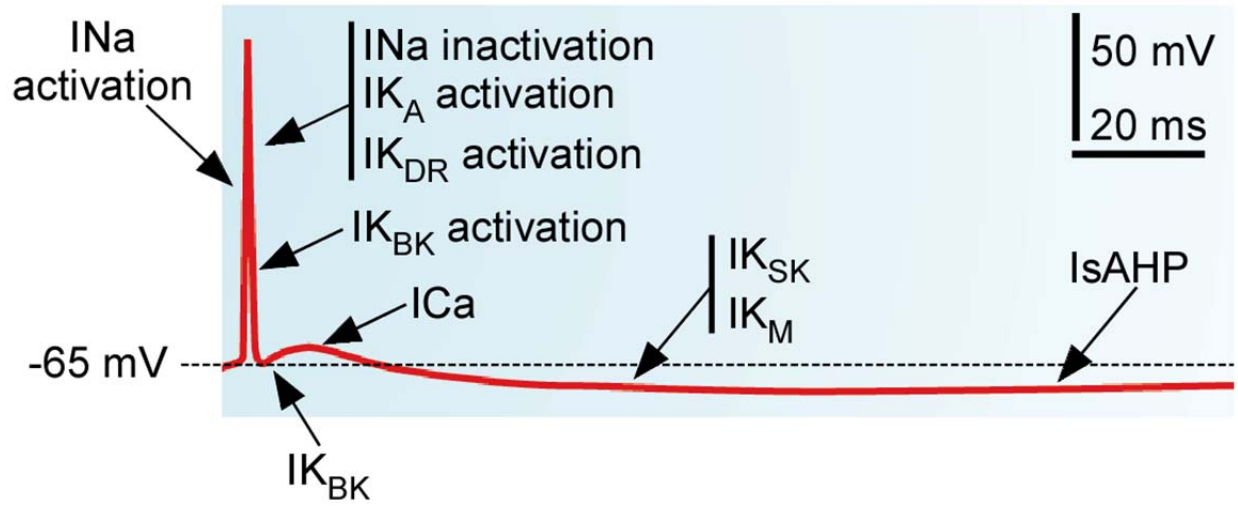

Figure adapted from Bean (1997).

V. Mechanisms controlling spike frequency and burst patterning

Just as the particular combination of ion currents in each cell determines its AP waveform, the complement of ion currents in the cell will shape spike frequency and burst patterning for that cell.

Spike frequency depends on the maintenance of very brief APs. In general, rapidly activating K^+ currents contribute to the rapid repolarization necessary for a short action potential. These include fast voltage-gated K^+ channels such as IK_{DR} and IK_A channels, as well as BK channels.

Controlling spike frequency adaptation is most typically a function of the slower calcium-activated K^+ currents. In particular, early spike frequency adaptation is produced by the currents that contribute to the medium AHP: SK channels or M-type K^+ channels. Late spike frequency adaptation is due to the inhibitory K^+ currents associated with the slow AHP. At very high firing frequencies, the fast AHP associated with BK channels facilitates rapid spiking and causes spike frequency adaptation very early in the spike train (Gu et al., 2007).

Modulation of any of these currents can produce functionally important changes in the neuron's dynamic range and spike-frequency adaptation. These changes are far from trivial. Modulations of AHP properties contribute to task learning (Matthews et al., 2008; Oh et al., 2009), and dysfunctions in these properties are associated with disorders such as epilepsy (Rogawski, 2000).

VI. Summary

The active properties of neurons give rise to the action potential. The action potential is a dynamic interaction of voltage, current, and conductance. Driving force, or voltage, is produced by the unequal distribution of ions across the cell membrane. Changes in conductance are a function of the opening and closing of ion channels selective for particular ions. These ion channels have a wide range of functional characteristics. The driving force in conjunction with changing ion conductances gives rise to ion currents across the membrane, and resulting changes in membrane potential.

The active functional characteristics of neurons are shaped by the particular complement of ion channels expressed by each neuron. This determines the AP threshold, AP waveform, and firing properties of each cell that enable it to function appropriately within its network.

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