Chapter 1

Hair Cells: The Sensory Transducers of the Inner Ear

Hair cells are specialized cells that transform a mechanical motion into changes in membrane potential. Such changes, whereby one form of energy (mechanical in this example) is changed into another form or energy (changes in membrane potential) is called transduction. The changes in membrane potential caused by a mechanical motion result in the release of transmitter substance which in turn generates action potentials in the nerves which innervate the hair cells. Here we consider how hair cells work and in the following lectures we will discuss the arrangement of hair cells in the cochlea and how they transduce mechanical motion of the cochlea into a neural code that represents the sound waves that are received by the ears.

Fig. 1

Schematic drawing of the constituents of a representative hair bundle. Each stereocilium has at its core a fascicle of cross linked actin filaments, of few of which extend as a rootlet into the fiberous cuticular plate. Three types of contacts interconnect the stereocilia: the longest stereocilia are also attached to the kinocilium at the bundle's tall edge. The hair cell is bound to the supporting cells by a junctional complex with a prominent tight junction.

Hair cells are the receptor cells in all of the many receptor organs in the vertebrate inner ear (semicircular canals, utricle, saccule and cochlea) as well as the lateral line organ of fish and larval amphibians. Exactly what type of mechanical energy the hair cell transduce depends on the organ in which it resides.
Hair cells of all of these organs work basically the same way. The hair cell body is oblong in shape, and numerous hair cells crowd together to form an epithelial sheet. Each hair cell is joined to its neighbors by tight junctions. These junctions ring the top of each hair cell separating its surface membrane into apical and basal surfaces. The apical surface is covered by 50-100 cilia (cilia are the filamentous projections that extend from the apical surface into the extracellular space).

Figure 2.

The hair cells in many organs have two different types of cilia: many short stereocilia arranged in order of descending length, and a single kinocilium asymmetrically placed on one side. (Interestingly, although the cochlear hair cells of mammals have a kinocilium at birth, they lose it shortly thereafter and thus kinocilium are not present in the cilia shown in Fig. 2). The cilia, which are like rigid rods, are attached to each other by protein bridges or cross-links, and move together in a bundle when stimulated. The cilia of hair cells are moved by fluids or associated tissues thereby coupling mechanical energy to the receptor cell for transduction.

The role of cilia in transduction has been studied by A.J. Hudspeth in a series of elegant experiments. He placed a microelectrode into a hair cell and recorded its membrane potential.
(called a receptor potential because the cells are receptors for sensory stimuli) while he manipulated the hairs with a fine glass filament (Hudspeth originally studied the hair cells in the sacculus of the vestibular system of frogs). He found that when he gently detached and removed the kinocilium, there was no decrease in the amplitude of the receptor potential. On the other hand, as he plucked out row after row of stereocilia he found that the receptor potential gradually diminished. These results indicate that the kinocilium is not necessary for transduction per se, while the stereocilia are. The kinocilium, it turns out, is important for attaching the hair cell bundle to overlying structures. Sensory stimulation causes vibrations in those structures which then efficiently delivers energy to the hair cells. Since we will be dealing with hair cells in the cochlea, we will not be concerned with kinocilia and will not mention them again.

Synaptic vesicles are localized in the base of each hair cell. Beneath the hair cell is an afferent nerve ending. As you might expect, the synapse between the hair cell and the afferent fiber is chemical.
If a microelectrode is placed inside a hair cell, in the absence of any stimulation, its resting potential will not be close to the K+ equilibrium potential, $E_k$ (assuming the same intra- and extracellular concentration of K+ as in muscle or squid axon which would give an $E_k$ around -70 mV). Instead, it is about -30 or -40 mV, showing that the hair cell is partly depolarized, even when there is no deflection of the cilia. Another electrode simultaneously placed into the afferent fiber would record spontaneously occurring spikes. If a small probe is then placed onto the ciliary bundle of the receptor cell and is moved back and forth, as in the Hudspeth experiment mentioned previously, a change in membrane potential (a receptor potential) is recorded in the hair cell. When the stereocilia are pushed toward the tallest cilia, the hair cell depolarizes and the number of spikes in the afferent fiber increases; when the stereocilia are pushed away from the tallest cilia, the hair cell hyperpolarizes and the number of spikes in the afferent fiber decreases. Obviously, the movement of the cilia changes the cell's membrane potential and thus transduces mechanical into electrical energy.

Work from Hudspeth's lab has elucidated the transduction process in some detail. The main points are summarized here. The transduction channels are located near the tips of the stereocilia. Each stereocilium is thought to have only a few channels so that each hair cell has no more than a few hundred channels.
A model for mechoelectrical transduction by hair cells. In the absence of any stimulation, at any instant each transduction channel at a stereocilium's tip may be either closed (A and F) or open (E). The greater probability is that the channel is closed. When the hair bundle is deflected with a positive stimulus, the spring or tip link, exerts a force on the trap door and opens the channel (B, C, and D). The influx of K+ ions into the hair cell causes it to depolarize. Pushing the hair bundle in the opposite direction compresses the spring (tip link), ensuring that the channel remains closed (G). This prevents the influx of K+ ions and causes the cell to hyperpolarize.

The transduction channels are covered with little "trap doors" and are associated with the protein cross-bridges near the tips of the stereocilia. The protein cross bridges act as springs that connect the transduction channels on the top of one cilium with a point on the shaft of the neighboring, taller cilium. As shown in the above figure, bending of the cilia in one direction tends to either stretch the spring and open the transduction channel whereas bending the cilia in the other direction tends to compress the spring and close the channel.

At rest, some of the channels (trap doors) are open but most are closed. When the stereocilia are slid toward the tallest cilium (or kinocilium), more channels are opened as their trapdoors are pulled open; when the cilia are moved in the opposite direction, the strain on the cross-bridges is relaxed and the trap doors close. The number of open channels depends largely on the position of the ciliary bundle and not on its velocity or acceleration of movement.
The trap doors are not tightly shut, but rather only loosely cover the pores. They therefore "rattle" back and forth between the open and closed states, and under normal conditions are sometimes open, but more often closed. Deflections of the stereocilia tend to increase the probability of opening or closing. If the cilia are bent towards the tallest cilia (or kinocilium in vestibular hair cells), the probability of opening is greatly enhanced, while bending away from the kinocilium decreases the probability of opening. An analogy would be an opening covered by a thin, lightweight sheet of metal. A small breeze would sometimes lift the sheet, thereby exposing the opening, but as soon as the breeze dies away, the sheet would fall back and again cover the opening. The breeze would be analogous to the normal movements of fluids within the inner ear that cause the trap doors to rattle back and forth between their open and closed states.

The conductance of the transduction channels is \textit{not voltage dependent} but depends largely on the position of the stereocilia. Interestingly, the channels are not specific to a particular ion but, like the acetylcholine channel at the neuromuscular junction which is permeable to both Na\(^+\) and K\(^+\), the transduction channels will pass most monovalent ions. (even Ca\(^{++}\) with a little difficulty). However, for reasons explained below, the transduction current is normally carried by K\(^+\).

In order to know how the hair cell works, it is important to know about the ionic composition of the fluids bathing their apical and basal surfaces. Although the fluid bathing the basal (bottom) face of the receptor (called perilymph) is similar to normal extracellular fluid (high Na\(^+\) concentration and a low K\(^+\) concentration), the fluid bathing the apical face (called endolymph) is very special. The endolymph contains high levels of K\(^+\) ions (150 mM) and low levels of Na\(^+\) (5 mM). In other words, the K\(^+\) concentration of the endolymph is about the same as the inside of the cell. Thus, if only the apical membrane of the hair cell were permeable to K\(^+\) ions, one would expect a resting potential of 0 mV. However, since the basal membrane faces a normal extracellular fluid with low K\(^+\), Ek ought to be about -70 mV. Thus, if the basal membrane alone were permeable to K\(^+\) ions, then the resting potential of the hair cell would be about -70 mV. It is easy to see that if the conductances of both the apical and basal membranes were activated, then the hair cell resting potential would be somewhere between -70 and 0 mV, depending on the relative strengths of each conductance. Also, note that if conductances were activated on both faces of the hair cell, this would result in K\(^+\) current going into the apical face.
(drawn in not by a concentration difference, but rather because the inside of the cell is more negative than the endolymph) and out of the basal face.

**Rest:** Small amount of $K^+$ leaks into cell from rattling channels of stereocilia. The leaking $K^+$ depolarizes cell thereby opening voltage sensitive $Ca^{++}$ channels resulting in spontaneous release of transmitter and excitation of afferent nerve fibers (not shown). The depolarization causes $K^+$ to leave cell in basal region.

**Excitation:** Stereocilia are bent thereby opening channels which results in larger entry of $K^+$ into cell. The influx of $K^+$ causes additional depolarization thereby opening more voltage gated $Ca^{++}$ channels. The additional $Ca^{++}$ channels that are now open cause a larger influx of $Ca^{++}$ in the base. The influx of $Ca^{++}$ causes more transmitter to be released and thus a greater excitation of the afferent fibers (not shown).

**Inhibition:** Stereocilia are bent in opposite direction thereby closing channels. This results in less influx of $K^+$ at channels in the tips of stereocilia. $K^+$ efflux occurs at base and is not replenished by influx at stereocilia. Consequently, the hair cell hyperpolarizes thereby closing voltage sensitive $Ca^{++}$ channels at the base resulting in a smaller release, or even no release of transmitter. Discharge of afferent fiber is therefore reduced to a rate even below the resting level.

The full story is that, at rest, $K^+$ leaves the cell across its basal membrane driven by the higher concentration of $K^+$ inside than outside the cell. Since positive charges leave the cell, the efflux of $K^+$ produces a negative resting potential inside the cell, which tends to drive the cell towards the $K^+$ equilibrium potential. However, the transduction channels are partially open at rest as well, so that the net internal negativity attracts $K^+$ from the endolymph, resulting in an inward $K^+$ current across the apical surface of the cell, which tends to depolarize the membrane potential. Thus, while the efflux of $K^+$ drives the membrane potential towards a more negative value (towards $Ek$), the influx of $K^+$ through the partially opened channels in the stereocilia acts in the opposite direction, and tends to depolarize the membrane potential. This explains why the resting potential of the hair cell is around -30 mV. Since the hair cell is partially depolarized,
calcium currents are turned on at the base of the cell causing transmitter release even in the absence of sound. This constant release of transmitter drives the spontaneous activity in auditory and vestibular nerve fibers.

Upon mechanical stimulation in the excitatory direction (when the stereocilia are bent toward the kinocilium) the pores in the tips of the stereocilia open resulting in an enhanced inward $K^+$ current which depolarizes the cell towards 0 mV and thus increases transmitter release (and increases firings in auditory nerve fibers). When the ciliary bundle is moved in the opposite direction, the conductance across the apical membrane is lessened as the pores close. Due to the decrease of $K^+$ influx, as well as efflux of $K^+$ in basal membrane, the membrane potential approaches the $E_k$ (-70 mV) and transmitter release decreases.

It is important to realize that, like rods in the retina, hair cells work close to the limit imposed by physics; at their extreme limit of sensitivity they may respond to deflections of ten or few angstroms, and thus is one the order of dimensions of atoms. This is because the channels or pores are opened (or closed) by only minute movements of the cilia. Even slight channel openings can cause changes in the membrane potential that, in turn, modulates or regulates the amount of transmitter released. Thus, the hair cell acts both as a transducer and an amplifier. It can transduce small movements of the cilia, on the order of tens to hundreds of angstroms, into sizable changes in membrane potential that may then be signaled to the central nervous system by changes in the spike activity of the afferent nerve fibers. Thus, a small stimulus modulates a large electrochemical potential (the membrane potential).