Generation of the Endocochlear Potential: A Biophysical Model

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ABSTRACT The generation and maintenance of the endocochlear potential (EP) by the stria vascularis is essential for proper function of the cochlea. We present a mathematical model that captures the critical biophysical interactions between the distinct cellular layers that generate the EP. By describing the relationship between the K^+ concentration in the intrastrial space and the intermediate cell transmembrane potential, we rationalize the presence of a large intermediate cell K^+ conductance and predict that the intrastrial $[K^+]$ is ~4 mM at steady state. The model also predicts that the stria vascularis is capable of buffering the EP against external perturbations in a manner modulated by changes in intrastrial $[K^+]$, thus facilitating hearing sensitivity across the broad dynamic range of the auditory system.

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The stria vascularis is a metabolically active epithelium in the lateral wall of the inner ear cochlear duct. It is responsible for two of the unusual properties of the endolymphatic fluid that bathes the stereocilia of auditory hair cells: a high K^+ concentration (over 150 mM) and a large electrical potential relative to other extracellular spaces (80–90 mV) (1,2). The strial epithelium consists of two functional layers, each of which forms a tight junction barrier (Fig. 1 *A*). The layer facing the endolymph consists of marginal cells, which use the Na^+, K^+-ATPase, and the NKCC1 cotransporter to synergistically transport K^+ into the cochlear duct (3). A second layer is composed of flattened basal cells and stellate intermediate cells that are interconnected by gap junctions and further connected to a fibrocyte network in the cochlear spiral ligament. Between these layers is a space isolated from the high potassium cochlear duct and the low potential extracellular fluid in the spiral ligament referred to as the intrastrial space.

Double-barreled electrode recordings suggest that the large cochlear duct potential, called the endocochlear potential (EP), is generated across the membrane of intermediate and basal cells (4). This concept was established by evidence that the intrastrial space had a potential similar to the EP, but a lower K^+ concentration (at ~20 mM) than the endolymph. The measured concentration has been questioned (5) because double-barreled electrodes are large and potentially destructive, and the volume of the intrastrial space is small.

To address this issue, Takeuchi and Ando performed whole-cell patch-clamp recordings on isolated intermediate cells (6). They found that these cells have a large K^+ permeability and further demonstrated that an extracellular K^+ concentration of 1.2 mM was necessary to develop a membrane potential similar to the EP (5). This estimated concentration is extremely low; given the K^+ affinity of their basolateral transporters, the marginal cells would function well below their capacity (3). Moreover, the estimate was based upon the assumption that the intermediate cell membrane was at equilibrium and the measurements were made under zero current clamp conditions in isolated intermediate cells. However, the cochlea has a significant standing current that makes this assumption invalid (7). Thus, the precise value and physiological significance of the intrastrial K^+ concentration remain elusive.

We have developed a mathematical description of the intrastrial space that allows us to estimate the intrastrial K^+ concentration and determine whether marginal cells are capable of maintaining K^+ at a level sufficient to generate the EP. To understand the relationship between the intermediate cell potential and intrastrial K^+ as a function of marginal cell activity, we considered the simplified model shown in Fig. 1 *B*. The only pathways included are the two transporters in the marginal cell and a lumped, constant K^+ conductance in the intermediate cell. For convenience, the transport rates are expressed as currents: p is the net outward current of the pump and n is the inward K^+ or Na^+ current of the NKCC transporter. The marginal cell K^+ current is therefore $-2p - n$. To ensure osmolar balance in the intrastrial space, at steady state the Na^+ current must be zero ($3p - n = 0$), so $n = 3p$. Across the intermediate cell membrane, there is a membrane potential of V and a linear K^+ current of G_K (V–E_K), where E_K is the Nernst potential for K^+.

At steady state, the K^+ current leaving the intermediate cells should match the K^+ current entering the marginal cells. Thus, if r is the ratio of marginal cells to intermediate cells, then G_K (V–E_K) = 5 rp. The intermediate cell transmembrane potential is then

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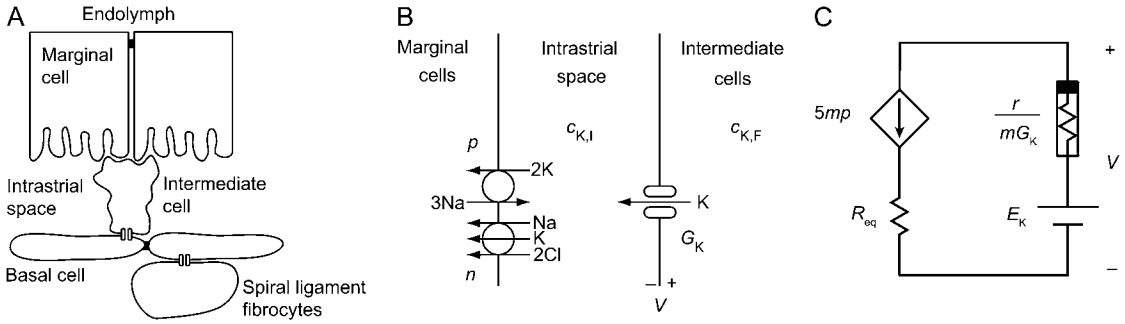

FIGURE 1 (*A*) The two cell layers of the stria vascularis are separated by an extracellular space that is isolated from the rest of the cochlea. (*B*) Simplified model of K^+ transport in the intrastrial space. Potassium enters the space through the intermediate cell membranes and is cleared by two transporters in the marginal cell basolateral membrane. (*C*) The stria vascularis operates against an external load resistance, R_{eq}, which represents the effective resistance of the pathways for cochlear potassium recirculation.

$$V = E_K + \frac{5rp}{G_K}. \tag{1}$$

The EP is approximately equal to $-V$, offset by the transepithelial potential of the marginal cell layer and the membrane potential of the spiral ligament fibrocytes, each of which is small (2).

We can immediately see the relationship between V and intrastrial $[K^+]$, since E_K is a function of intrastrial $[K^+]$. A low $[K^+]$ is needed because the second term reduces the EP. If $p = 0$, there is no current and $V = E_K$, as Takeuchi et al. used in their approximation (5). However, if $p > 0$, as it is in vivo, then $V > E_K$.

How large is this extra term? The steady-state, inwardly rectified K^+ conductance is ~110 nS per intermediate cell at negative voltages (6). The resting marginal cell current has been estimated to be ~235 pA per cell (7). According to stereological measurements, there are 2.5 marginal cells per intermediate cell (8). With these values, $5rp/G_K = 5.3$ mV. This value is low relative to E_K. Thus, one reason for having a large intermediate cell K^+ conductance G_K is to prevent depolarizing currents from other ions. This calculation also suggests that the stria vascularis uses a large intermediate cell conductance to keep the marginal cell-driven offset low to maintain the EP.

The above analysis assumes that p and G_K are constants, but they actually depend on voltage and substrate concentration. Making p an increasing function of $[K^+]$, for example, may make it easier to maintain a very negative V, because the second term in Eq. 1 will be smaller (there will be less current). To make this more explicit, consider a concentration-dependent pump rate (9):

$$p([K^+]) = p_{max}\left(\frac{[K^+]}{[K^+] + 1.16\,\text{mM}}\right)^2. \tag{2}$$

We model the voltage dependence of G_K by fitting the measured whole cell current-voltage relationship for the inwardly rectified intermediate cell K^+ conductance (3) with a Boltzmann model:

$$G_K(V) = \frac{126\,\text{nS}}{1 + \exp\left(\frac{V + 46.8\,\text{mV}}{29.3\,\text{mV}}\right)}. \tag{3}$$

The intermediate cells also express an outwardly rectifying conductance (6), but it is not activated at physiologically relevant voltages.

The resulting intermediate cell voltage, as a function of the extracellular $[K^+]$, is shown in Fig. 2 for a range of pump activities. As shown, a large marginal cell activity decreases the potassium concentration necessary to maintain a given voltage. To maintain the voltage at ~−90 mV, a large pump-current lowers the necessary extracellular $[K^+]$ by ~1 mM. A concentration near 4 mM with $p \le 100$ pA would produce a voltage of ~90–105 mV, near the EP.

In principle, this model allows us to examine how the stria vascularis responds to changes in the EP. However, an additional equation is required to solve for the equilibrium intrastrial $[K^+]$ and V independently. The simplest possible equation is

$$V = -I_K R_{eq}, \tag{4}$$

where I_K is the length-specific K^+ current and R_{eq} is the length-specific resistance of the cochlear partition and represents the components that make up the remainder of the K^+ circulation pathway (1,2). The resulting circuit is shown in Fig. 1 *C*. Identifying I_K with the currents described in Fig. 1 *B* and Eq. 2, the voltage is

$$V = -5mp([K^+]_{IS})R_{eq}, \tag{5}$$

where $[K^+]_{IS}$ is the intrastrial $[K^+]$ and m is the marginal cell density (number of cells per unit length of the cochlea). This allows us to solve for the intrastrial potassium concentration,

$$[K^+]_{IS} = [K^+]_{IC} e^{-\frac{5p([K^+]_{IS})}{RT}\left(mR_{eq} + \frac{r}{G_K(V)}\right)}, \tag{6}$$

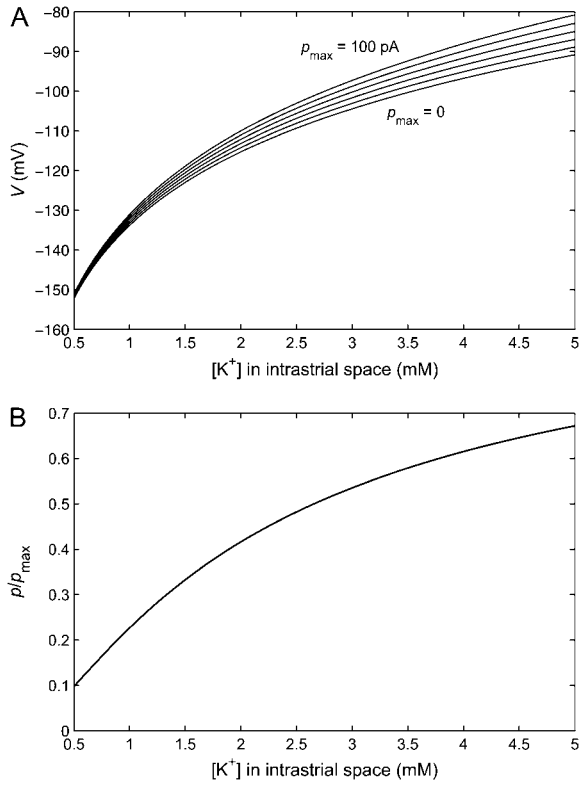

FIGURE 2 (*A*) Relation between intrastrial $[K^+]$ and intermediate cell voltage at various levels of marginal cell activity (*p*). (*B*) Dependence of marginal cell pump activity on intrastrial $[K^+]$. Note that the total K^+ current across the stria is proportional to *p*.

where $[K^+]_{IC}$ is the potassium concentration in the intermediate cell (~150 mM). The second equation states that the pump (p) reduces $[K^+]_{IS}$ and the intermediate cell conductance (G_K) increases $[K^+]_{IS}$. Given an estimate for R_{eq}, Eqs. 5 and 6 together describe the system.

We can evaluate the response of the system to changes in voltage with these equations. For example, if the EP falls due to a drop in R_{eq}, as occurs with increased hair cell activity, intrastrial $[K^+]$ rises (Eq. 6). The rise in $[K^+]_{IS}$ causes p to increase (Eq. 2), which acts to restore the EP. If the rise in $[K^+]_{IS}$ is substantial, it will cause G_K to increase, as the conductance of Kir channels rises with extracellular $[K^+]$ (10). This would result in a further increase in $[K^+]_{IS}$, which would stimulate the pump, helping to restore the EP. These features would buffer the EP against changes. On the other hand, G_K is expected to decrease after a rise in $[K^+]_{IS}$ as a result of intermediate cell depolarization, because the conductance is an inward rectifier. In that case, p will decrease and the EP will drop further. However, intermediate cells possess an outward rectified conductance as well (3). This conductance has not been fully characterized but conceivably functions to maintain G_K in the face of membrane depolarization.

The reason an inward rectifier is located in intermediate cells is unclear (11). The above considerations suggest the channel's sensitivity to extracellular $[K^+]$ as a possible explanation. Otherwise, at steady state, a sufficiently negatively activating outward rectifier would be able to provide the same conductance, and the response of an outward rectifier to depolarization would actually be more effective at maintaining the EP.

This relatively straightforward analysis provides a framework for understanding how interactions between the components of the stria vascularis function to generate and maintain the endocochlear potential. We conclude that the intrastrial space is physiologically active and maintains a K^+ concentration of 3.5–4.5 mM (Fig. 2 *A*). As further details on ion channel expression and function in the stria vascularis become available, this model can be refined to yield a more accurate understanding of the generation of the endocochlear potential.

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