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Abstract. Taking advantage of a two-compartment preparation of the frog saccule, we measured the effect of steps or slow ramps of electric current across the sensory tissue on the transduction channels' gating force. Transepithelial currents afforded a means to control the endolymphatic potential in the top compartment with respect to the potential in perilymph that bathed the basal aspect of the hair cells. Under each condition, we estimated the transduction channels' gating force from nonlinearity of the bundle's force-displacement relation. We found that the gating force could nearly double upon application of a negative endolymphatic potential until, beyond a threshold value, the gating force abruptly dropped to a low level, resulting in linear force-displacement relations. The hair-bundle stiffness at large displacements, outside the region of channel gating, was only weakly affected by transepithelial currents. In addition, when descending and ascending ramps of transepithelial currents were applied in succession, the transition between strong and weak gating forces displayed hysteresis. All these effects were fully reversible. We conclude that the endolymphatic potential may serve as a control parameter of the transduction channel's gating force. Our work further indicates that the molecular movement associated to channel gating—the gating swing—can be of variable magnitude in the same hair cell depending on electrical conditions. We propose that modulation of channel gating by the endolymphatic potential results from electric control of the resting calcium influx into the hair cell, which must be large enough to allow for large gating swings and in turn for high mechanosensitivity by the hair cell.

INTRODUCTION

In the inner ear, mechanosensitivity of the hair cells results from direct mechanical activation of ion channels by the tip links of their hair bundle [1,2]. Deflection of the hair bundle modulates tip-link tension, resulting in channel gating and a transduction current. By virtue of mechanical reciprocity, channel gating feeds back on tip-link tension (Fig. 1a), producing an internal force called the "gating force" [3,4]. The gating force is a fundamental determinant of hair-cell mechanosensitivity, for its magnitude sets the maximal slope of the sigmoidal relationship between the transduction current and the hair-bundle displacement. Moreover, gating forces can be large enough to effectively reduce the hair bundle's stiffness [3,5], even sometimes resulting in negative stiffness [4], and increase its friction [6]. Through an active dynamic interplay with adaptation, negative stiffness can foster spontaneous hair-bundle oscillations and amplification of sinusoidal stimuli [2,7–9]. Despite its importance for hair-cell mechanosensitivity, the biophysical determinants of the gating force remain poorly understood. Here, we report that the electric potential in endolymph bathing the hair bundles can serve as a control parameter of the gating force.

METHODS

We took advantage of an excised preparation of the frog saccule (species: Rivan92 [10]) that mimicked *ex vivo* the two-compartment ionic conditions that hair cells experience *in vivo*, with endolymph and perilymph bathing the apical and basal aspects of the sensory tissue, respectively [11]. Under such conditions, the hair bundles routinely displayed

spontaneous oscillations [9]. Using a pair of silver-chloride electrodes, we applied steps or slow ramps of a transepithelial current within a range $\pm 10\ \mu A$. As a result, the potential into the artificial endolymph that bathed the hair bundles—the endolymphatic potential— varied within a range of ± 100 mV with respect to the potential in the perilymphatic compartment, corresponding to a transepithelial resistance of $10 \pm 2\ k\Omega$ ($n = 13$); the potential settled to a steady-state value within a few hundred microseconds after the onset of a current step. We estimated the magnitude of the transduction channels' gating force from mechanical measurements of the force-displacement relation of individual hair bundles and studied how it varied upon application of a transepithelial current.

The mechanics of the hair bundle was characterized using published methods and two different stimulus protocols [6,11]. In short, the tip of a flexible fiber was affixed to the top of a hair bundle at its kinociliary bulb. In a first protocol, a series of step displacements was applied to the fiber's base. The force, F, and the bundle's displacement, X, were estimated $5 - 9\ ms$ after the stimulus onset, a time long enough for the frictional contribution to have vanished and short enough to limit the effect of adaptation. The resulting force-displacement relation displayed a region of lower slope as a result of gating compliance [3]. It was fitted according to the gating-spring model of mechanoelectrical transduction [2] to estimate the whole bundle's gating force, F_G:

$$F = KX - F_G/[1 + \exp(-(X - X_0)/\delta)] + F_0. \quad (1)$$

We note that F_G corresponds to the force shift between the two linear limbs of the force-displacement relation at large positive and negative displacements, for which the transduction channels are nearly all open and all closed, respectively. The slope of these linear limbs provided the bundle's the linear stiffness, K. We also measured the set-point deflection, X_0, at which the slope of the force-displacement relation is minimal and where the open probability of the transduction channels is $1/2$. Note that we did not use displacement-clamp feedback to stabilize negative stiffness. Thus, the characteristic displacement, δ, in the fit could not be exploited. Parameter F_0 ensures that $F = 0$ at $X = 0$. The force-displacement relation was also measured after disruption of the tip links by iontophoretic application of the Ca^{2+} chelator EDTA; the relation was then linear, with a slope noted K_{SP} [12].

In a second stimulus protocol, we applied a symmetric triangular waveform of motion to the fiber's base, at a frequency of 40 Hz and with a peak-to-peak amplitude of 600 nm. The force applied to the hair bundle was estimated as described in [6] using a detailed description of the fiber's vibrational modes. This protocol resulted in force-displacement cycles with counterclockwise circulation, which were averaged over 10 cycles. Their half height at any position, X, provide an estimate of the average friction force $\phi(X) = [F^+(X) - F^-(X)]/2$ impeding the hair bundle motion at this position in the cycle, where $F^{\pm}(X)$ represents the force on the positive and negative half cycle of stimulation, respectively. As detailed in [6], transduction channels' gating result in a friction force that peaks at the set-point deflection, with a peak value noted $\phi_{MAX} = \max(\phi(X)) = \phi(X_0)$. We thus used ϕ_{MAX} as an index for the gating-force magnitude, although one should keep in mind that its value also depends on the transduction channels' activation time.

RESULTS

We first characterized the mechanical properties of the hair bundle by measuring force-displacement relations in response to a series of step displacements of the stimulus fiber's base (see Methods). Under control conditions, we measured a set-point deflection $X_0 = -2 \pm 7$ nm, a whole hair bundle's gating force $F_G = 17 \pm 4$ pN and a linear stiffness $K = 0.85 \pm 0.2\ \mathrm{pN.nm^{-1}}$ (mean $\pm$ SD, n = 29 in 16 frogs). The procedure was repeated in the presence of a static transepithelial current, I. We start by showing three representative examples (Fig. 1). Applying a positive current, here $I = +6\mu A$, resulted in an increase of the set-point deflection, corresponding to a rightward shift of the force-displacement relation and closure of the transduction channels, a small (-6%) decrease of the linear stiffness, and a large decrease (-50%) of the hair-bundle's gating force (Fig. 1b). Conversely, applying a negative current of moderate magnitude, here $I = -3\ \mu A$, elicited a decrease of the set-point deflection, corresponding to a leftward shift of the force-displacement relation and opening of the transduction channels, a large ($+89\%$) increase in the hair-bundle's gating force and a small increase ($+10\%$) of the linear stiffness (Fig. 1c). Remarkably, applying a large negative current, here $I = -6\ \mu A$, did not result in a further increase or saturation of the hair-bundle's gating force, corresponding to more prominent mechanical nonlinearity, but instead only evoked linear force-displacement relations (Fig. 1d). All these effects were fully reversible.

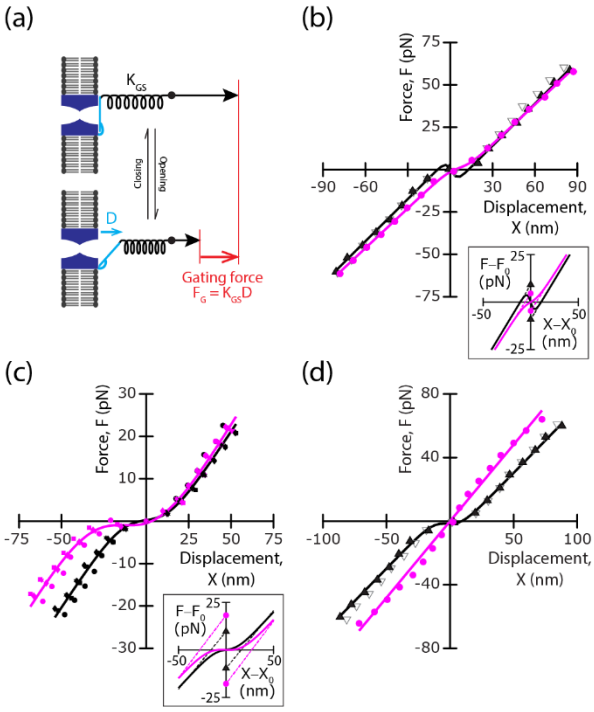

FIGURE 1. (a) Gating-spring model of mechanoelectrical transduction. Opening of the transduction channels reduces tension in the gating springs by an amount given by the gating force, noted F_G, whereas channel closure does the opposite. Here, K_{GS} represents the combined gating-spring stiffness in the hair bundle and D is the conformational change associated to channel gating—the gating swing. (b-d) Representative examples of force-displacement relations under control conditions ($I = 0$ μA; black) and in the presence of a transepithelial current (magenta) of amplitude: $I = +6$ μA (b), $I = -3$ μA (c), $I = -6$μA (d). Each force-displacement relation was fitted by Eq. 1 (solid lines). Insets: the fits are plotted with respect to the origin $(X_0, F(X_0))$. Under each condition, the hair-bundle's gating force, F_G, was given by the vertical distance between the two symbols in the inset ($I = 0$: triangles;$I \neq 0$: disks). In (a), the experiment was repeated 4 times. In (c) and (d), the open black symbols correspond to recovery.

We performed an ensemble of $n = 41$ pairs of measurements in 29 hair bundles from 16 frogs to compare the force-displacement relations under control ($I = 0$) and test conditions ($I \neq 0$). These experiments demonstrated a strong positive correlation between the change $\Delta X_0 = X_0(I) - X_0(I = 0)$ of the set-point deflection and the applied transepithelial current I (Fig. 2a). The change in linear stiffness $\Delta K = K(I) - K(I = 0)$ was only weakly negatively correlated to the transepithelial current (Fig. 2b). Instead, for currents $I > -5$ μA, the change $\Delta F_G = F_G(I) - F_G(I = 0)$ of the hair-bundle's gating force displayed a strong negative correlation with the current: negative currents: negative endolymphatic potentials, evoked an increase of the gating force, while positive currents did the opposite (Fig. 2c). Correspondingly, the transepithelial current ($I > -5$ μA) affected the hair-bundle's gating force F_G much more than the linear stiffness K, with a relative change that could reach $+100\%$ for the former but typically did not go beyond $+25\%$ for the later (Fig. 2d). Strikingly, for all seven hair bundles that experienced a negative current with a magnitude beyond a threshold value, in practice $I \leq I_C = -5$ μA, the gating force was so low that the force-displacement relations were linear, thus with no detectable gating compliance (Fig. 1c; Fig. 2c and d).

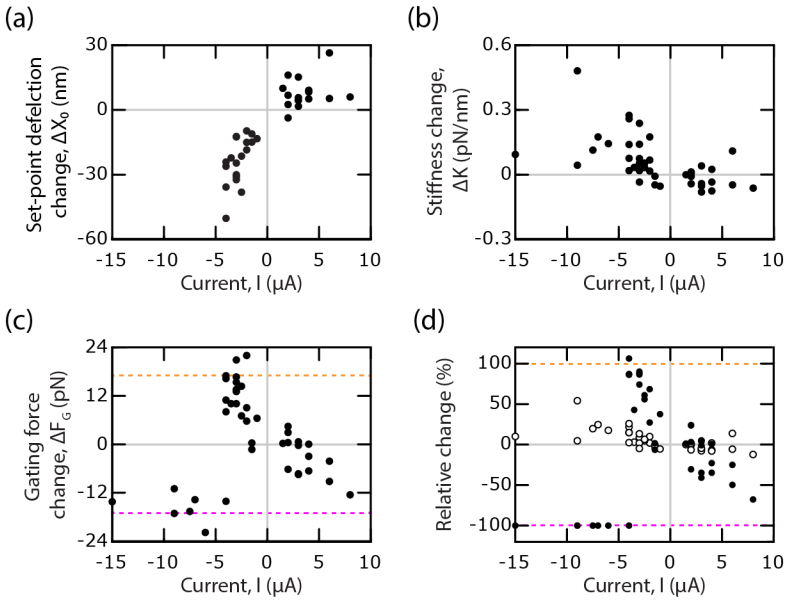

FIGURE 2. Mechanical changes evoked by a transepithelial current, I, over an ensemble of hair bundles with respect to control conditions ($I = 0$). (a) Change, ΔX_0, of the set-point deflection. ΔX_0 is positively correlated to I (Pearson correlation $r = 0.87$, p value $= 4 \cdot 10^{-11}$, $n = 34$). (b) Change, ΔK, of the linear stiffness. ΔK is weakly negatively correlated to I (Pearson correlation $r = -0.57$, p value $= 10^{-4}$, $n = 41$). (c) Change, ΔF_G, of the hair-bundle's gating force. For $I > -4$μA, ΔF_G is strongly negatively correlated to I (Pearson correlation $r = -0.86$, p value $= 8 \cdot 10^{-11}$, $n = 34$). In contrast, for $I < -5$μA, all the force displacement relations were linear ($F_G(I) = 0$) and thus $\Delta F_G(I) = -F_G(I = 0)$ is centered around the average value $F_G = 17 \pm 4$ pN (mean ± SD, $n = 29$) measured under control conditions (magenta dashed line). The gating force could nearly double upon application of a negative current of moderate magnitude (orange dashed line). (d) Relative change of the hair-bundle's gating force, $\Delta F_G/F_G(0)$, (solid disks) and of linear stiffness, $\Delta K/K(0)$, (open disks).

We further explored the transition from a nonlinear to a linear force-displacement relation by applying ramps of transepithelial current at a slow rate, $r = 0.9\ \mu A.s^{-1}$ (Fig. 3a), while the hair bundle was subjected to continuous triangular stimulation (see Methods and [6]). The time required to measure a smooth force-displacement cycle was short enough to consider that the transepithelial current was stationary. Under control conditions ($I = 0$), the force-displacement cycle displayed a region of gating compliance on the positive and negative half cycles (Fig. 3b; black line), corresponding to channel opening and closure, respectively [6]. In addition, the height of the force-displacement cycle was larger within the region of channel gating. This is because gating of the transduction channels contributes to friction on the hair bundle—a phenomenon called gating friction [6]. The half-height of the cycle provides an estimate of the friction force, $\phi(X)$ (see Methods), on the hair bundle and reached a maximal value $\phi(X_0) = \phi_{MAX}$ at a position that has been identified [6] as the set-point deflection X_0 (disks in Fig. 3b).

As the transepithelial current decreased from zero to negative values, the region of gating compliance and gating friction in the force-displacement cycle shifted toward more negative displacements (Fig. 3b), corresponding to a decrease of the set-point deflection X_0. In addition, the height of the force-displacement cycles, and thus the maximal friction force ϕ_{MAX} increased: gating friction became larger at negative transepithelial currents. Again, when the absolute magnitude of the negative current became large enough, corresponding to $I \leq I_C = -6.5\ \mu A$, the nonlinearity vanished (Fig. 3b, red) and the two limbs of the force-displacement cycles were then linear. The polarity of the current ramp was then reversed. As the negative current increased, the signatures of gating compliance and gating friction, associated to nonlinear force-displacement relations, were retrieved. Remarkably, however, the nonlinearity reappeared at a smaller absolute current, i.e. a current that was less negative, than that necessary to conceal the signatures of channel-gating forces on the descending ramp. Thus, the nonlinear-to-linear transition displayed hysteresis, as shown in the relation $\phi_{MAX}(I)$ (Fig. 3c).

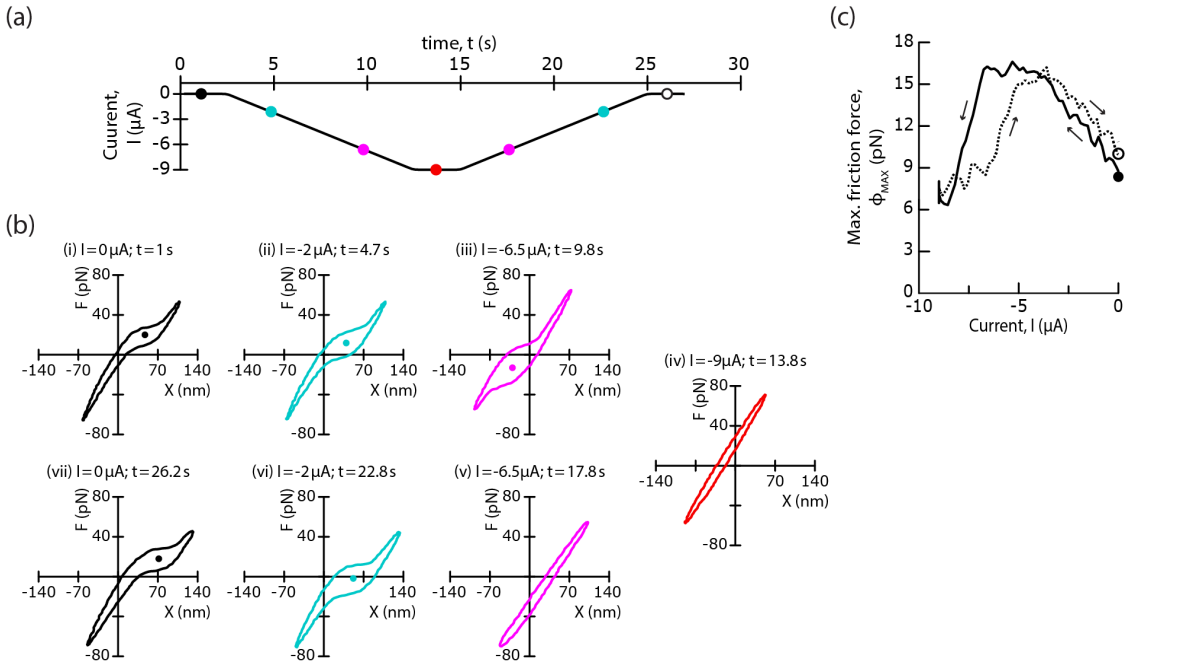

FIGURE 3. Effects of ramps of transepithelial current on hair-bundle mechanics. (a) Descending and then ascending ramps of a negative transepithelial current were applied at an absolute rate of $0.9\ \mu A.s^{-1}$ (minimal current:$-9\ \mu A$), while a hair bundle was subjected to triangular stimulation, with a peak-to-peak amplitude of 600 nm and a frequency of 40Hz, applied at the base of a stimulus fiber. The colored disks indicate the time points of the analysis shown in (b). (b) Force-displacement cycles for the current values and times indicated at the top of each sub-panel, corresponding to the colored disks in (a). (c) Maximal friction force, $\phi_{MAX}(I)$, as a function of the transepithelial current, I. Black and white disks indicate the start and end of the experiment, respectively. The solid and dotted lines correspond to the descending and ascending ramps of transepithelial current, respectively. Arrows indicate counterclockwise circulation around the hysteretic cycle.

DISCUSSION

Within the framework of the gating-spring model, the gating force for the whole hair bundle, $F_G = K_{GS}D$, is the product of the combined stiffness of the gating springs, K_{GS}, by the gating swing, D, of a transduction channel, here estimated along the horizontal axis of mechanical stimulation. A major finding of the present study is that the gating force could increase by up to twofold upon application of a negative endolymphatic potential, whereas the change in linear hair-bundle stiffness was comparatively much smaller (Fig. 2d). Because the gating springs are expected to contribute a significant fraction of the total hair-bundle stiffness K [13], we reasoned that the change in gating force (Fig. 2c) resulted mainly from a change in the gating swing. We could confirm this inference by measuring the hair-bundle stiffness, $K_{SP} = 0.17 \pm 0.03\, pN/nm$ $(n = 14) \simeq 0.2\, K$, after disruption of the tip links. By subtracting the average value of K_{SP} to individual measurements of K, we could then estimate, for each hair bundle, the gating-spring stiffness $K_{GS} = K - K_{SP}$, and in turn the gating swing, D. After projection along the oblique axis of the tip links using the projection factor $\gamma = 0.14$ [14], we found a gating swing $d = \gamma D = 3.7 \pm 1.4\, nm$ $(n = 29)$ under control conditions (Fig. 4a). Replicating the behavior of the gating force, the gating-swing change displayed a strong negative correlation with the endolymphatic potential (Pearson test: $r = -0.86; p = 1 \cdot 10^{-10}$) (Fig. 4b). Correspondingly, the gating swing d could reach a value of up to $10\, nm$ in the presence of a negative endolymphatic potential or, conversely, be as small as $2\, nm$ upon application of a potential of opposite polarity (Fig. 4a). The gating swing corresponds to the decrease in gating-spring extension upon channel opening. Understanding how this conformational change can be up to tenfold as large as the size of the channel's pore, a necessary condition for high mechanosensitivity, remains an outstanding problem for the field.

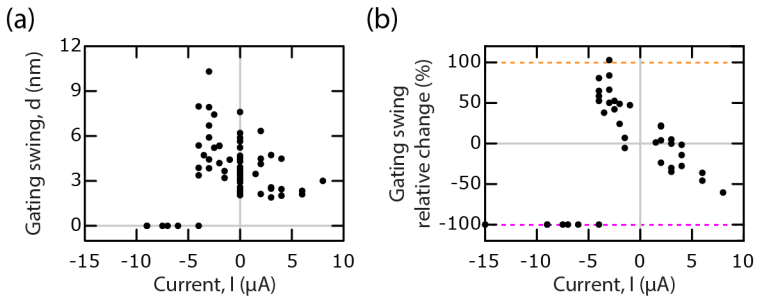

FIGURE 4. Effects of a transepithelial current on the gating swing. We plot the gating swing, d, (a) and its relative change, $\Delta d(I)/d(0)$, (b) as a function of the transepithelial current, I for the same dataset as that used in Fig. 2.

We can think of two mechanisms that may explain a modulation of the gating swing by the endolymphatic potential. First, the endolymphatic potential modulates the magnitude of the electric field across the lipid bilayer that houses the transduction channels. A movement of some of the channels' charged components may affect how the transduction complex then responds to external mechanical force. In support for this mechanism, a voltage-evoked movement of the hair bundle—sometimes termed the flick—has been reported, even under circumstances for which there is no gating because the channels were pharmacologically blocked [15–17]. Second, the endolymphatic potential also controls the magnitude of the electrochemical force that drives cations across open transduction channels, in particular Ca^{2+} ions. The Ca^{2+} concentration on the intracellular side of the transduction channels may affect channel gating, either by binding to a channel component [18] or by altering lipid-bilayer properties in the vicinity of the transduction channel [19,20]. We note that the observed variations of the set-point deflection, X_0, upon application of an endolymphatic potential (Fig. 2a) are consistent with Ca^{2+}-mediated adaptation: a negative endolymphatic potential reduces the intracellular Ca^{2+} concentration, which results in opening of the transduction channels, and conversely [21].

We observed a remarkable hysteretic transition between a regime of strong gating forces and a regime where the gating force is so low, that there is no sign of gating compliance in the force-displacement relation (Fig. 2c, Figs. 3c-d, and Fig. 4b). To account for linear force-displacement relations at large negative potentials, we must assume that the gating swing dropped to sub-nanometer levels. This transition happened at an endolymphatic potential of about $-50\, mV$, perhaps when the intracellular Ca^{2+} concentration fell below a threshold value.

Overall, our work demonstrates that the endolymphatic potential may serve as a control parameter of the transduction channel's gating force by modulating the size of the molecular movement associated to channel gating—the gating swing. Although applying an endolymphatic potential is somewhat artificial in frog, the finding that the gating swing can be of variable magnitude in the same hair cell depending on electrical conditions is expected to have broad implications for our general understanding of transduction-channel gating.

ACKNOWLEDGMENTS

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COMMENTS & QUESTIONS

[*Online Forum*]

Elisabeth Olson : Dear Pascal and Achille -- Nice paper and interesting findings. It is well written and the figures are clear. I just have a couple comments/ suggestions.

(1) It would be nice to include a stick figure of the channel opening and gating spring, but not necessary since this sort of figure is in the literature.

(2) What is the EP normally in bullfrog sacculus?

(3) Do you have reason to believe that these findings would also occur in mammalian hair cells?

(4) Finally, we have some studies that seem related, in gerbil cochlea — their findings are along the same lines as yours, in a way. Both started with IV furosemide, which rapidly reduces EP to even negative values, then it recovers pretty quickly, in about 40 mins to a sub-normal level (about 40-60mV).

The first study (Wang et al 2019, BPJ) found that recovery of cochlear tuned responses (measured with local cochlear microphonic (LCM) in that case) occurred many minutes AFTER EP had recovered to a sub-normal level. Another important finding was that recovery in the LCM tuning coincided in time with the hair bundle MET channel apparently returning to a relatively centered operating point. (This determined by LCM waveform and nonlinearity). We therefore speculated that the recovery of cochlear tuning / cochlear amplification had something to do with hair bundle mechanics.

In the second study (Strimbu et al 2020 BPJ) we repeated the IV furosemide study but this time monitored recovery with vibration responses and DPOAEs. The basic findings were the same as with the Wang et al paper – the recovery of tuned motion responses occurred at a time AFTER EP would have recovered to a sub-normal level, and the recovery of tuned motions occurred along with DPOAE increasing, which sort of represents operating-point recentering b/c DPOAEs are odd distortions, which are larger with centered operating point.

Authors:

(1) Thank you for the suggestion. We have added a schematic in Fig. 1.

(2) In vivo, the frog saccule experiences an endolymphatic potential around ~3 mV (Schmidt RS and Fernandez C, Comp Biochem Physiol 1962). The electrical condition that we imposed on the hair cells in our experiments is thus somewhat artificial. It can nevertheless be used to ask, in general terms, whether the endolymphatic potential can control hair-bundle mechanics, in particular the magnitudes of the gating force and of the gating swing—the conformational change associated to opening of the transduction channels.

(3) Whether or not our findings also apply to mammals remains to be demonstrated. Available gating-force estimates in mammals show relatively low values and, accordingly, the force-displacement relations of individual hair bundles are linear (e.g. Beurg, Schwalbach, and Fettiplace, PNAS 121:e2318270121 (2024)). However, force measurements are usually performed under ex-vivo conditions for which the cochlea is fully immersed in artificial perilymph and with no transepithelial potential. It is thus tempting to speculate that the gating force and the gating swing might be much larger under more physiological conditions. In support of this working hypothesis, ex-vivo preparations of isolated cochlear segments, with reconstituted endolymphatic-perilymphatic compartments and an endolymphatic potential, have already demonstrated a mechanical nonlinearity (see Chan and Hudspeth, Nat. Neurosci. 8:149-155 (2005) and Francesco Gianoli's presentation at this workshop). Although direct measurements of hair-bundle mechanics are still lacking, this nonlinearity likely results from channel-gating forces, implying that the gating force is larger in these preparations.

(4) Thank you for this information. It would be nice if one could discriminate effects of the endolymphatic potential on the operating point of the transducer—where the hair bundle operates in its sigmoidal current-displacement relationship—and on the maximal mechanosensitivity—how steep is the current-displacement relationship, a property under the control of the gating force. Our experiments imply that both are affected.

Raul Araya-Secchi: It is a very interesting, compelling and well-presented work, well written and figures are clear.

I only have a few comments:

(1) I think that more context for the significance of the gating force and gating swing in the broader scope of auditory mechanics should be added. In its current version it´s too specific an focused on a narrow specialized audience. It is perhaps a consequence of the length constraints required for this manuscripts but it improve a final version of the manuscript.

(2) A discussion of the potential effects of fluctuating endolymphatic potentials in living organisms, and how might these affect hearing sensitivity and adaptation could be interesting.

(3) How does the hysteresis observed in the gating force transitions might affect real-time auditory processing in fluctuating environments?

(4) How does the presented results relate to our current understanding of the structure of the transduction channel. How do the changes in gating swing relate to the physical structure of the channels and tip links? At least in terms of predicted structures of TMC candidates and current knowledge of tip-link structure.

Auhors:

(1) We have added a schematic to the manuscript (Fig. 1) to recall the gating-model of mechanoelectrical transduction. We hope that this will help the reader to grasp how channel gating affect hair-bundle mechanics and, in return, how hair-bundle mechanics provides useful biophysical information about channel gating, here the size of the gating force and of the gating swing.

(2) The condition we impose here on the hair cells is artificial. We do aim to suggest that the endolymphatic potential "fluctuates" in vivo. Varying the endolymphatic potential nevertheless affords a means to probe the properties and physiology of channel gating. A major finding of the work is that the channels can exist, in the same cell and depending on the endolymphatic potential, in a state where channel gating produces strong gating forces as the result of a large gating swing—as revealed by the presence of gating compliance—or in a state where channel gating has no effect on the mechanics (small gating swing), corresponding to linear force-displacement relations. These findings shed light on our limited understanding of the molecular substrate for the gating swing, a fundamental issue that remains to be elucidated.

(3) Hysteresis with clockwise circulation around a force-displacement cycle betrays energy dissipation, which limits the sensitivity and frequency selectivity of the hair bundle to external stimuli. As previously studied in our group (Bormuth et al PNAS 2014), channel gating contributes to energy dissipation by the whole hair bundle.

(4) Our finding that the gating swing can be reversibly modulated over a large range of magnitudes (1-10 nm) by the endolymphatic potential raise important fundamental questions regarding the mechanism of transduction-channel gating and the exact molecular nature of the conformational change associated to channel gating. There is a gap between what we know about the molecular identity of the constituents of the protein complex that mediates mechanoelectrical transduction—a rapidly evolving field—and the coarse-grained information we get from biophysical experiments about fundamental parameters such as the gating force and the gating swing.

Note that mechanosensitivity is favored by large conformational changes associated to channel gating (up to 10 nm in our ex-vivo preparation of the frog saccule) whereas ionic selectivity instead calls for a small channel pore (here about 1nm), thus imposing seemingly opposite requirements on channel gating. Implementing a molecular "lever arm" to magnify the opening of the channel pore is a problem faced by hair cells of all species to achieve high mechanosensitivity.

[*Post-Talk Q&A*]

Jont Allen: (1) This threshold [*Author clarificaton: the value of the endolymphatic potential that elicits a transition between linear and nonlinear hair bundle mechanics or, equivalently, between states of strong and weak gating force*] seems to be related to the Hopf bifurcation. If you agree, how? (2) Can we call this the cochlea amplifier?

Author: (1) It's related because to get a Hopf bifurcation you need to destabilize the system and you destabilize the system if you get negative stiffness, which happens if the gating forces are large enough. (2) No, because everything I described is totally passive. The mechanical properties of the transduction channels are nonlinear but the channels must be coupled to an active motor, which we believe, in the case of hair bundles from the frog, comes from myosin motors and calcium feedback.

Robert Fettiplace: I thought that the link you brought at the end is very important: the link between the (gating-spring) model with a gate moving and what the molecule is really like. The question is that if you can get a movement, and this has always mystified me, of 8 to 12 nm, what does that mean in terms of conformational change in a molecule? It's huge.

Author: Yes, it's huge. But there is some avenue for hope there. You may know about this paper on Piezo by Rob McKinnon's group (Guo YR, MacKinnon R (2017) eLife 6:e33660). Piezo is a mechanosensitive channel and the same problem arises for this channel, which is well described in this paper. There too, there is a relatively change in the surface occupancy of the channel in the membrane when the channel opens and yet the channel pore is very small. These authors showed that, in that case, when the channel is inside the membrane, the membrane is curved and forms a dome which may magnify the conformational change associated to channel opening by having this dome flattening or even bulging out. This mechanism provides one example where a small conformational change in a channel protein can result in a bigger motion overall, here via the membrane. Another possibility, which was proposed by Andryi Kozlov and Francesco Gianoli (Gianoli, Risler and Kozlov PNAS 2018), is that cooperative channel gating of two channels, each connected at the end of one molecule of PCDH15 in a tip link, would effective lead to a large gating swing. There are thus mechanisms that may account for a large gating swing but we don't know precisely which one is at work in the hair cell. Nature has already found ways to decouple the opening of the channel pore to the effective change in conformation happening at the scale of the transduction machinery as a whole.