Review

Linear and nonlinear processing in hair cells

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Summary

Mechanosensory hair cells in the ear are exquisitely responsive to minute sensory inputs, nearly to the point of instability. Active mechanisms bias the transduction apparatus and subsequent electrical amplification away from saturation in either the negative or positive direction, to an operating point where the response to small signals is approximately linear. An active force generator coupled directly to the transducer enhances sensitivity and frequency selectivity, and counteracts energy loss to viscous drag. Active electrical amplification further enhances gain and frequency selectivity. In both cases, nonlinear properties may maintain the system close to instability, as evidenced by small spontaneous oscillations, while providing a compressive nonlinearity that increases the cell's operating range. Transmitter release also appears to be frequency selective and biased to operate most effectively near the resting potential. This brief overview will consider the resting stability of hair cells, and their responses to small perturbations that correspond to soft sounds or small accelerations.

Introduction

Hair cells are highly specialized mechanoreceptors in the ears of vertebrate animals that transduce the component of force directed along their sensitive axis into an electrical signal, with sufficient sensitivity to detect Brownian motion of the eardrum. The sensory information is then transmitted across a chemical synapse to the postsynaptic afferent axon, where it is encoded as train of spikes that travel to the brain. Along the way, the signal is filtered to enhance a range of frequencies. Much of the recent work on the cellular physiology of hair cells has been aimed at understanding the input–output relation of each step in this process, with the goal of a complete understanding of the relationship between a time-varying mechanical input and the resulting time-varying spike probabilities in postsynaptic afferent axons.

One important basic question is whether the input–output relation can be usefully regarded as linear over some limited range of stimulus strength, or whether nonlinear properties are essential for the response to even the smallest of stimuli. Linearity has advantages beyond simplicity, especially for separating signal from noise. If the main source of variability in the response to a given input is additive noise applied at the input (e.g. Brownian motion of the eardrum and hair bundle), then a linear sensor is particularly useful because it can be regarded as operating independently on signal and noise. If the separate responses to signal and noise are known, outputs of linear sensors can be summed and filtered to predictably amplify signals relative to noise. On the other hand, the classic description of logarithmic input–output relations in sensory systems (the Weber–Fechner law) emphasizes the multiplicative nature of perceptual uncertainty, and the advantage of a compressive non-linearity for extending the range of non-saturating stimuli.

These two views are not necessarily incompatible, since they employ different measures of the input and output. In the conceptual framework of psychophysics (e.g. the Weber–Fechner law and

Stevens' Power Law), both the stimulus intensity and response amplitude are a non-negative numbers that equal zero when there is no stimulus, whereas mechanosensory transduction by hair cells is directional, with positive and negative stimuli generating opposite responses. Directionality of hair bundle movement may be irrelevant for some tasks, such as judging whether one sound is louder than another, and crucial for others, such as judging the location of a sound source by the time delay between the two ears or for vestibular sensation. The same sensor can be regarded as either linear or nonlinear relative to different sets of measures used to assess its performance in different tasks. It is even possible to have the best of both worlds, as when a memoryless compressive nonlinearity is applied to the output of a linear system. Since a compressive nonlinearity can be unambiguously undone, the linear response can later be recovered from the compressed signal. But this waffling avoids the question of whether it is more useful to emphasize linearities or nonlinearities when thinking about how hair cells respond to small stimuli. In keeping with this theme, it seems fitting that some recent work concludes that hair cells are actively maintained near the boundary between a well-behaved linear system and a spontaneously oscillating non-linear system (Choe et al., 1998; Camalet et al., 2000; Martin et al., 2003; Vilfan and Duke, 2003; Jülicher et al., 2001).

Many hair cells have surprisingly linear responses to small inputs (e.g. Crawford and Fettiplace, 1981; Brandt et al., 2005). Perhaps they do this to best preserve directional information about small signals corrupted by uncorrelated additive noise by responding symmetrically (more or less) to forces applied in opposite directions. Fig. 1 shows an idealized current–displacement curve for a hair cell's mechano-electrical transducer recorded a few milliseconds after the application of displacement steps of various amplitudes from the resting position, after the response has stabilized, but before adaptation sets in. The responses to large stimuli are highly nonlinear
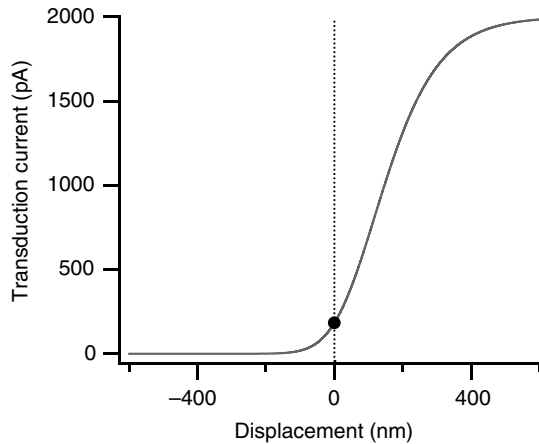
Fig. 1. The relationship between hair bundle displacement and the resulting current through transduction channels. At rest (displacement=0) the transducer in many hair cells lies to the left of center on the curve (filled circle), and therefore not at the point of maximum sensitivity or greatest linearity to small inputs.

in that they saturate in both the positive and negative directions, but the transducer is biased away from positive or negative saturation such that its responses to small displacements from zero are reasonably linear. Over any small range of displacements, the curve looks approximately like the tangent line, which is to say that there is no discontinuity or sharp change in slope that would indicate a threshold nonlinearity. The slope of the tangent line is the transducer's sensitivity to small changes in hair bundle position, which attains a maximum at the center of the curve and declines towards zero in both directions.

It is worth noting that the small signal sensitivity would be greatest and the response more symmetrical if the resting position of the transducer were at the point of maximum slope on the current–displacement curve (Fig. 1). Instead, many hair cells sit on the ascending limb, close to the point of maximum curvature. Mammalian cochlear hair cells make use of the resulting nonlinearity to respond to sounds at high frequencies (>10 kHz), where the unavoidable membrane capacitance greatly attenuates membrane potential oscillations, which constitute the linear part of the response, but leaves a depolarization caused by the amplitude asymmetry of the transduction current. The sensitivity of this non-linear component approaches zero as the amplitude of the sound decreases (i.e. the curve looks locally like a straight line), but responses are seen when high frequency sounds are loud enough to produce significantly asymmetric transduction currents (Russell and Sellick, 1983). High frequency hearing in mammals thus involves an essential nonlinearity, although it remains to be seen whether later stages of signal processing, such as synaptic transmission, are more linear. Recently, a different significance has been proposed for the precise resting position on the current–displacement curve. In these models, feedback causes the transduction apparatus to seek a resting state near (but not at) a Hopf bifurcation (see below), where the gain of its response to small sinusoidal forces approaches infinity. This behavior can only be seen if the stimulus is applied as a force, rather than a displacement. *In vivo*, the input to many hair cells is somewhere in between a pure force and a pure displacement (Martin et al., 2003).

At this point it seems useful to define 'linear', especially because it was misused in the previous paragraphs. Before one can say that

the input–output relation shown in Fig. 1 is approximately linear for small displacements around the resting position (x=0), the y-axis must be offset by subtracting the resting transduction current. This type of output offset of an otherwise linear system is conventionally ignored because it is easily subtracted. More importantly, we have ignored time. In general, a linear system transforms a time-varying input, $g(t)$, into a time-varying output, $h(t)$, and obeys laws of scaling and superposition. An additional requirement of shift-invariance is often an unstated assumption. If a linear system transforms $g(t) \rightarrow h(t)$, the scaling property requires that $a \cdot g(t) \rightarrow a \cdot h(t)$, where a is any constant. The superposition property states that if $g_1(t) \rightarrow h_1(t)$ and $g_2(t) \rightarrow h_2(t)$ then $g_1(t)+g_2(t) \rightarrow h_1(t)+h_2(t)$. Shift invariance requires that shifting the input along the time axis simply shifts the output by the same amount.

Linearity does not require that the shape of the output waveform bear any particular relationship to the shape of the input waveform. Many hair cells respond to a current step with a damped oscillation of the membrane potential. By itself, this says nothing about linearity of the voltage–current relation. Linearity implies that if the step is made twice as large, then the response becomes twice as large while maintaining the same shape, and that steps from rest in opposite directions evoke opposite responses (Crawford and Fettiplace, 1981). The scaling property requires that the response to constant zero input is constant zero output. Shift invariance implies that if $g(t)$ is a constant function of time, then so is $h(t)$. Thus, hair cells that produce large membrane potential oscillations in the absence of any stimulus are fundamentally non-linear because they violate both scaling and shift invariance. Small oscillations might, perhaps, be judiciously ignored.

Finally, linear responses may be particularly important to the auditory and vestibular systems because sounds and accelerations from different sources usually combine linearly, unlike in vision, where nearby objects hide what lies behind. Linearity aids in decomposing additively combined sensory inputs into behaviorally relevant components for the same reason that it aids in separating signal from additive noise. To take advantage of this, the mechanical workings of the outer, middle and inner ears that direct mechanical stimuli arising from different sounds and accelerations in three dimensions onto the appropriate sensory structures in the ear operate nearly linearly over a wide range of stimulus amplitudes.

Stability of linear and nonlinear systems

Recent experiments and mathematical models of hair cell function place the resting hair cell near a point of transition between stable, approximately linear behavior, and instability. The mathematical models used to describe this behavior consist of equations that specify the feedback interactions among a set of state variables {$x_i(t)$, i=1, 2,..., n}, in which each of the states can potentially influence all of the others. The state variables must be chosen such that their values at any time t completely specify the state of the system at that time, regardless of system's history. The interactions among the state variables are specified as the rate of change of each state variable x_i as a function f_i of the instantaneous values of all the other state variables:

$$dx_i / dt = f_i(x_1, x_2,..., x_n). \qquad (1)$$

This type of dynamical model is very general, and therefore conceptually simple. To simulate the system's behavior, one begins with some known initial state [$x_1(t_0)$, $x_2(t_0)$,..., $x_n(t_0)$], divides time into small increments Δt, calculates (dx_i/dt) for each state variable, and extrapolates linearly to the state at time $t+\Delta t$ [$x_1(t_0+\Delta t)$,

$x_2(t_0+\Delta t),..., x_n(t_0+\Delta t)$]. If the time steps are sufficiently small, extrapolation errors can be ignored.

Standard ways of thinking about cell biological and biophysical problems easily fit into this framework. For example, the state variables for chemical reactions are the concentrations of the reactants and products, and the rate equations specify their time derivatives. Electrical and mechanical systems are similarly simple to deal with. The problem, of course, comes with using the equations to predict the system's behavior. Simulation provides specific answers but sometimes little insight, which is often better obtained by approximating a complex system by something simpler. Linear approximations are attractive, both because linear systems are relatively easy to understand and because there exists a simple procedure to find the best linear approximation to any dynamical system formulated as described above. The procedure involves evaluating, for all pairs of states (i, j), the effect that perturbing x_j has on f_i while holding all of the other state variables constant. The resulting set of partial derivatives $\partial f_i / \partial x_j$ is called the Jacobian matrix, J, where:

$$J_{ij} = \partial f_i / \partial x_j. \tag{2}$$

Thus, the nonlinear mutual interactions among a large set of state variables is approximated by the sum of pairwise linear interactions. How good an approximation depends on specifics of the problem at hand, but one important general result predicts the behavior of the nonlinear system near a 'stationary' state (i.e. a set of values of the state variables for which all of their time derivatives are zero). A stationary state can be a stable steady state to which the system returns when perturbed, or it can be unstable, like a coin balanced on edge. The important general result is that the eigenvalues of the Jacobian matrix, which are readily calculated numerically for even large matrices, determine whether a stationary state of the nonlinear system is stable. Furthermore, if the system is stable, the eigenvalues predict how the state variables return to the stable state after a perturbation. If the stationary state is unstable the linear approximation explodes, and fails to predict the behavior of the nonlinear system.

One class of unstable stationary states that has received much recent attention in hearing research is called a Hopf bifurcation. The term applies to a nonlinear system in which a control variable μ determines whether the stationary state is stable. For $\mu<0$ the system is stable and behaves approximately linearly near the stationary state. As μ is increased towards zero the system grows increasingly unstable until, for $\mu>0$ the system oscillates, either falling into a limit cycle or spiraling to infinity, depending on other parameter values [for a nice discussion of Hopf bifurcations, see Ospeck et al. (Ospeck et al., 2001)].

Examples of stable and unstable stationary states abound. The constant membrane potential of a neuron at rest is a stable stationary state of the nonlinear interactions among the ion channels that set the resting potential and the membrane capacitance, which is critical for stability. Reducing the membrane capacitance can change the stable stationary state into an unstable state. Some neurons fire a repetitive train of spikes in the absence of any input. The nonlinear interactions between ion channels in these cells generate limit cycles, in which the system's state follows a closed orbit around an unstable stationary state. Limit cycles are common in nonlinear systems, but impossible in linear systems that obey shift invariance. Unless driven by a time-varying input, these systems either settle onto a stable stationary state or explode to infinity.

As a final example, we consider the exponentially damped membrane potential oscillations that many hair cells exhibit when perturbed by current steps or pulses (electrical resonance). This phenomenon is readily understood in terms of the eigenvalues of the Jacobian matrix associated with a simple linear model. When a linear system specified by a Jacobian matrix is perturbed, the system relaxes back to its stable state over a trajectory that is the sum of exponential functions, $A_1\exp(\lambda_1 t)+A_2\exp(\lambda_2 t)+...+A_n\exp(\lambda_n t)$, where the λ_i are the eigenvalues. If λ_i is real, then $\exp(\lambda_i t)$ is a simple exponential function of time. If λ_i is complex, then $\exp(\lambda_i t)$ is product of an exponential with a sinusoid. Complex eigenvalues imply that the system oscillates when perturbed from a stationary state. If the real parts of the λ_i are all negative, then the oscillations are damped, otherwise they grow to infinity. The model of electrical resonance in turtle hair cells (Crawford and Fettiplace, 1981) behaves like an electrical circuit consisting of a resistor, capacitor and inductor, or equivalently a mass attached to a spring damped by friction. In these models the eigenvalues are complex and have a negative real part, such that after the system is perturbed each of its state variables (e.g. membrane potential) relaxes back to its stable state along an exponentially damped sinusoidal time course, which is the observed response of the cell.

Evoked and spontaneous hair bundle movements

The hair bundle is composed of many individual stereocilia, each with its own transduction apparatus, but the bundle moves as a unit (Kozlov et al., 2007), so a single state variable suffices to specify its displacement. An external force, $F(t)$, applied to the tip deflects the hair bundle. The force is transmitted to mechanically gated ion channels (transduction channels) through elastic links called gating springs that pull the channels open, producing a transduction current that changes the hair cell's membrane potential.

When a transduction channel opens its gating spring relaxes, and when it closes the spring stretches, applying a force that adds to $F(t)$ and causing the hair bundle to move (Crawford and Fettiplace, 1985; Howard and Hudspeth, 1988). Hair bundle movements and other force generators associated with auditory transduction (e.g. outer hair cell motility in the mammalian cochlea) can propagate back towards the stimulus, causing the eardrum to vibrate and emit sounds (otoacoustic emissions). This bidirectional coupling between the sensory stimulus and the transduction mechanisms is a unique feature of mechanosensory transduction that can lead to instability, as evidenced by spontaneous otoacoustic emissions.

Maintained displacement of the hair bundle causes transduction to adapt in two phases (Howard and Hudspeth, 1988; Ricci and Fettiplace, 1998; Martin and Hudspeth, 1999; Wu et al., 1999; Ricci et al., 2000; Holt and Corey, 2000; Eatock, 2000; Holt et al., 2002; Ricci et al., 2002; Vilfan and Duke, 2003; Vollrath and Eatock, 2003; Martin et al., 2003; Ricci et al., 2005; Cheung and Corey, 2006) (reviewed by LeMasurier and Gillespie, 2005; Fettiplace, 2006). The slow phase operates on a time scale of tens to hundreds of milliseconds and is responsible for establishing resting tension in the gating springs, although it has not been reported in mammalian cochlear hair cells. Slow adaptation is accomplished by a myosin motor that stretches the gating springs until the channels begin to open. The open transduction channels admit Ca^{2+} into the cell. Ca^{2+} inhibits the adaptation motor, allowing it to slip back, establishing a dynamic equilibrium at which each transduction channel is open part of the time. This partial activation of the transduction machinery moves the system away from positive or negative saturation, into a more linear range where small positive or negative deflections of the hair bundle produce symmetrical positive or negative transduction currents. Two state variables per transduction channel are required to describe slow adaptation: the position of the

adaptation motor and the Ca^{2+} concentration that regulates it. The 50–100 transduction channels per hair cell can be modeled as individual elements (Beurg et al., 2008), but have usually been combined into a single element to reduce the number of state variables.

When a force applied to the hair bundle opens transduction channels, the resulting relaxation of the gating springs causes the hair bundle to move farther than it would have if the channels had remained closed. When the hair bundle is allowed to move freely, this gating compliance can create a region of apparent negative bundle stiffness. If the set point for slow adaptation is within this region, the hair bundle oscillates spontaneously as the adaptation motor attempts the Sisyphean task of returning the bundle to an unstable position (Crawford and Fettiplace, 1985; Howard and Hudspeth, 1987; Martin et al., 2003). The resulting oscillation can be large enough to cause the transduction channels' open probability to alternate between 15% and 70% (Martin et al., 2003). Such large oscillations would nullify any assumption of linear transduction, were it not for stabilizing factors such as the tectorial membrane or similar structures that are attached to hair bundles *in vivo*. In mammalian inner hair cells either this mechanism does not operate or other factors must be present to prevent large spontaneous oscillation of the cells' free-standing hair bundles.

The fast phase of adaptation, which in some hair cells can take place in a fraction of a millisecond (Ricci et al., 2005), operates quite differently from slow adaptation, and also appears to be involved in mechanical amplification. It is caused by Ca^{2+} binding directly the transduction channel or some closely associated molecule, and has the opposite mechanical effect from slow adaptation (reviewed by Fettiplace, 2006). Transduction channels exist in two conductance states (open or closed), but to describe the fast component of adaptation requires adding a third, Ca^{2+}-bound state. Ca^{2+} binding promotes the closure of an open transduction channel, which then pulls upon the gating spring, causing the hair bundle to move against the applied force, generating a 'twitch' that can cause the hair bundle to oscillate briefly in response to a force step (Hudspeth et al., 2000). The energy required to produce this damped oscillation comes from the steep electrochemical gradient to Ca^{2+}, which drives Ca^{2+} into the cell when transduction channels open. It has been proposed that the myosin motor also contributes to fast adaptation (Martin et al., 2003; Stauffer et al., 2005). Several models of hair bundle mechanics have incorporated fast adaptation (Choe et al., 1998; Camalet et al., 2000; Vilfan and Duke, 2003; Ricci et al., 2005; Beurg et al., 2008). Differences in the speed of fast adaptation in outer hair cells may contribute to frequency selectivity (Ricci et al., 2005; Kennedy et al., 2005).

Spontaneous hair bundle oscillations have led several investigators to propose that adaptation serves to position the unstimulated hair bundle close to a Hopf bifurcation (Choe et al., 1998; Camalet et al., 2000; Martin et al., 2003; Vilfan and Duke, 2003). In the neighboring stable region the Jacobian matrix describes the system's linear behavior. Near the bifurcation the system has an extremely high-gain, approximately linear response to small, applied forces at a preferred frequency (Martin and Hudspeth, 2001). Larger forces engage a compressive nonlinearity that obeys a power law with exponent=1/3 (Martin and Hudspeth, 2001). If the system is to maintain an approximately linear small-signal response, it is important that the hair cell's operating point remains on the linear side of the Hopf bifurcation (Eguíluz et al., 2000). The large spontaneous oscillations that have been recorded indicate that, under the experimental conditions studied, the hair bundle does not stay in the linear region, but *in vivo* the mechanical load of structures attached to the hair bundle are expected to prevent or greatly reduce spontaneous oscillation (Martin et al., 2003). As with all feedback systems, taking the system apart can result in behaviors that are radically different from the behavior of the intact system, which has led some to question the usefulness of Hopf bifurcation models of cochlear function (Zweig, 2003).

The mechanisms responsible for the exquisite sensitivity and frequency selectivity in the mammalian cochlea are not fully understood. Outer hair cells in the cochlea possess force generators along their lateral walls that are hypothesized to underlie the mammalian 'cochlear amplifier' (Dallos et al., 2006; Ashmore, 2008). This mechanism appears to be unique to mammals, and has therefore received much attention as a possible source of the unique properties of the mammalian cochlea, particularly high sensitivity and frequency selectivity above 10 kHz. However, this force is controlled by membrane potential, which is attenuated by the membrane's electrical time constant, such that the amplitude of high-frequency oscillations falls off in proportion to 1/frequency, which poses difficulties for amplification at frequencies above 15 kHz (Ospeck et al., 2003; Dallos et al., 2006). The lateral wall force generator cannot, by itself, explain frequency tuning in the mammalian cochlea because it does not appear to exhibit tonotopic differences, but by counteracting viscous damping it could enhance the tuning from other sources. There is also evidence that mammalian inner hair cells employ the same hair bundle amplification mechanism found in other vertebrates (Chan and Hudspeth, 2005; Ricci et al., 2005). This mechanism may be better suited to operate at high frequencies because it relies on changes in local Ca^{2+} concentration within nanometers of a source (the transduction channel), which can rise and fall on a microsecond time scale.

Evoked and spontaneous membrane potential oscillations

The next step in sensory reception involves excitability of the hair cell basolateral membrane. Unlike nearly all neurons, most adult hair cells in the auditory system do not produce all-or-none voltage changes. They do express voltage-gated Ca^{2+} channels and several types of voltage-gated and Ca^{2+}-gated K^+ channels that are capable of producing highly nonlinear responses, but as with transduction the system appears to be designed to generate nearly linear responses to small perturbations from the resting state. Some vestibular hair cells generate large spontaneous membrane potential fluctuations that are better described as limit cycles of a nonlinear system.

The auditory hair cells of fish, amphibians, reptiles and birds typically respond to a current step with a damped oscillation of the membrane potential (Crawford and Fettiplace, 1981; Ashmore, 1983; Lewis and Hudspeth, 1983; Fuchs and Evans, 1988; Fuchs et al., 1988; Sugihara and Furukawa, 1989; Steinacker and Romero, 1992) at a frequency that varies with the cell's position along a tonotopic axis (reviewed by Fettiplace and Fuchs, 1999). This phenomenon has been studied most extensively in the turtle cochlea, where small positive and negative current steps evoke symmetrical voltage oscillations around the resting potential that scale with the step amplitude. The resting transduction current caused by the slow mechanical adaptation described above may be an important factor in depolarizing V_m into this linear operating range (Farris et al., 2006). V_m also influences adaptation through its effect on Ca^{2+} influx.

The simplest model of electrical oscillations in hair cells is a linear electrical circuit consisting of a capacitor, resistor and inductor (Art and Fettiplace, 1987). In this model, the 'phenomenological

inductance' is provided by K channels, which behave like an inductor in series with a resistor. More detailed models based on the voltage- and Ca^{2+}-dependent gating properties of several classes of ion channels that interact with the membrane capacitance have also been used (Hudspeth and Lewis, 1988; Ospeck et al., 2001; Jørgensen and Kroese, 2005). The number of variables needed to specify the states of these channels can be quite large, but there is no difficulty constructing models that reproduce the observed behavior.

Some auditory hair cells from amphibians, reptiles and birds have step responses that undergo many cycles of damped oscillation, but not all hair cells are so sharply tuned. In many fish and amphibians, the sacculus is an important auditory organ. The frog sacculus is sensitive to sounds in the 5–200 Hz range, with much greater sensitivity to vibrations conducted through the ground than through the air (Christensen-Dalsgaard and Narins, 2003). Studies of goldfish (Sugihara and Furukawa, 1989) and frog (Armstrong and Roberts, 1998) sacculus have shown that hair cells in these organs typically respond to large current steps with highly damped membrane potential oscillations, suggesting that they each respond over a broad range of frequencies.

In some species, 'electrical resonance' is a major factor in establishing the frequency selectivity and the tonotopic organization of the auditory organ. Like transduction, the hair cell's electrical response is linearized by maintaining a fraction of the cell's ion channels open at the resting potential. Transduction currents perturb the membrane potential by opening or closing a fraction of these channels. As described above for mechanical transduction, nonlinearities may serve to place the system close to a point of instability (a Hopf bifurcation), where amplification of small signals is large (Ospeck et al., 2001; Jørgensen and Kroese, 2005), but the response to small perturbations remains nearly linear. As with transduction, the nonlinearity generates a small spontaneous membrane potential oscillation, and a compressive nonlinearity for larger stimuli.

Frequency selectivity of transmitter release at the afferent synapse

The final stage of signal processing by hair cells takes place at the afferent synapse, where glutamate is released onto afferent terminals. Our understanding of the synaptic transfer function is far from complete, particularly for small voltage perturbations around the resting potential, but there is evidence for ongoing transmitter release at the resting potential (Glowatzki and Fuchs, 2002; Keen and Hudspeth, 2006), as expected if the synapse is biased to be active at rest. One recent study (Rutherford and Roberts, 2006) suggests that afferent synaptic transmission in the frog sacculus possesses an intrinsic 'tuning' mechanism that accentuates transmitter release when the hair cell membrane potential oscillates at ~50 Hz. Like other tuning mechanisms in hair cells, it operates most effectively for small stimuli near the resting potential.

Recent studies of auditory hair cells from mouse (Brandt et al., 2005; Johnson et al., 2007), turtle (Schnee et al., 2005) and frog (Keen and Hudspeth, 2006) have shown a nearly linear relationship between Ca^{2+} influx and transmitter release, in contrast to the 3rd or 4th power dependence of transmitter release on cytoplasmic Ca^{2+} that has been reported at most synapses. A detailed mechanistic explanation will require more experimentation, but one possible explanation is that a single open Ca^{2+} channel may provide all the Ca^{2+} needed to trigger release of a nearby synaptic vesicle, perhaps due to an unusually close spatial coupling of Ca^{2+} channels and

Ca^{2+} sensors for release (Brandt et al., 2005). The approximately linear relationship between Ca^{2+} influx and transmitter release is seen experimentally under conditions where the current through each open Ca^{2+} channel is nearly constant. Transmitter release is thus interpreted to reflect the number of open Ca^{2+} channels near sites of exocytosis. It is also possible that the Ca^{2+} sensor that controls synaptic exocytosis in auditory hair cells, which is reported to be otoferlin (Roux et al., 2006), has a more linear dependence on cytoplasmic Ca^{2+} than does synaptotagmin II, which controls transmitter release at conventional synapses. In many types of hair cells the resting transduction current depolarizes the membrane potential to a level where many of the Ca^{2+} channels are open at rest which, in conjunction with a linear relationship between the number of open Ca^{2+} channels and transmitter release, is expected to linearize the overall relationship between small forces applied to the hair bundle and transmitter release.

Conclusion

Several converging lines of evidence suggest that hair cells attain high sensitivity to small inputs at a preferred frequency by operating approximately linearly, but close to instability, both at the level of mechoelectrical transduction and in their resonant electrical properties. In both cases, nonlinearities may serve to reduce the gain when oscillations grow, thereby preventing the system from falling into instability, and provide a compressive nonlinearity that maintains a large operating range. These mechanisms provide a resting level of activation of the transduction channels as well as the ion channels responsible for electrical resonance and transmitter release, ensuring that these systems respond nearly linearly to small perturbations. Larger stimuli evoke a compressive nonlinearity that greatly expands the dynamic range. There is evidence (Martin and Hudspeth, 2001) that nonlinear hair bundle responses appear only when the stimulus exceeds some threshold. It is possible that hair cells are specialized to maintain a linear response to very small stimuli to help the CNS extract periodic signals from additive noise by averaging in time and across inputs, and by other linear operations. For louder sounds, where thermal noise is relatively unimportant, maximizing dynamic range may be more important. Further work is needed to understand how the results of this linear processing of small sensory signals in hair cells are translated into a spike train in the postsynaptic afferent axons, and whether the time-varying probability of an afferent spike is linearly related to the time-varying force applied to the hair bundle.

References

Armstrong, C. E. and Roberts, W. M. (1998). Electrical properties of frog saccular hair cells: distortion by enzymatic dissociation. *J. Neurosci.* **18**, 2962-2973.

Art, J. J. and Fettiplace, R. (1987). Variation of membrane properties in hair cells isolated from the turtle cochlea. *J. Physiol.* **385**, 207-242.

Ashmore, J. F. (1983). Frequency tuning in a frog vestibular organ. *Nature* **304**, 536-538.

Ashmore, J. (2008). Cochlear outer hair cell motility. *Physiol. Rev.* **88**, 173-210.

Beurg, M., Nam, J.-H., Crawford, A. and Fettiplace, R. (2008). The actions of calcium on hair bundle mechanics in mammalian cochlear hair cells. *Biophys. J.* **94**, 2639-2653.

Brandt, A., Khimich, D. and Moser, T. (2005). Few CaV1.3 channels regulate the exocytosis of a synaptic vesicle at the hair cell ribbon synapse. *J. Neurosci.* **25**, 11577-11585.

Camalet, S., Duke, T., Jülicher, F. and Prost, J. (2000). Auditory sensitivity provided by self-tuned critical oscillations of hair cells. *Proc. Natl. Acad. Sci. USA* **97**, 3183-3188.

Chan, D. K. and Hudspeth, A. J. (2005). Ca^{2+} current-driven nonlinear amplification by the mammalian cochlea *in vitro*. *Nat. Neurosci.* **8**, 149-155.

Cheung, E. L. and Corey, D. P. (2006). Ca^{2+} changes the force sensitivity of the hair-cell transduction channel. *Biophys. J.* **90**, 124-139.

Choe, Y., Magnasco, M. O. and Hudspeth, A. J. (1998). A model for amplification of hair-bundle motion by cyclical binding of Ca^{2+} to mechanoelectrical-transduction channels. *Proc. Natl. Acad. Sci. USA* **95**, 15321-15326.

Christensen-Dalsgaard, J. and Narins, P. M. (1993). Sound and vibration sensitivity of VIIIth nerve fibers in the frogs *Leptodactylus albilabris* and *Rana pipiens pipiens. J. Comp. Physiol. A* **172**, 653-662.

Crawford, A. C. and Fettiplace, R. (1981). An electrical tuning mechanism in turtle cochlear hair cells. *J. Physiol. Lond.* **312**, 377-412.

Crawford, A. C. and Fettiplace, R. (1985). The mechanical properties of ciliary bundles of turtle cochlear hair cells. *J. Physiol. Lond.* **364**, 359-379.

Dallos, P., Zheng, J. and Cheatham, M. A. (2006). Prestin and the cochlear amplifier. *J. Physiol.* **576**, 37-42.

Eatock, R. A. (2000). Adaptation in hair cells. *Annu. Rev. Neurosci.* **23**, 285-314.

Eguíluz, V. M., Ospeck, M., Choe, Y., Hudspeth, A. J. and Magnasco, M. O. (2000). Essential nonlinearities in hearing. *Phys. Rev. Lett.* **84**, 5232-5235.

Farris, H. E., Wells, G. B. and Ricci, A. J. (2006). Steady-state adaptation of mechanotransduction modulates the resting potential of auditory hair hells, providing an assay for endolymph [Ca^{2+}]. *J. Neurosci.* **26**, 12526-12536.

Fettiplace, R. (2006). Active hair bundle movements in auditory hair cells. *J. Physiol.* **576**, 29-36.

Fettiplace, R. and Fuchs, P. A. (1999). Mechanisms of hair cell tuning. *Annu. Rev. Physiol.* **61**, 809-834.

Fuchs, P. A. and Evans, M. G. (1988). Voltage oscillations and ionic conductances in hair cells isolated from the alligator cochlea. *J. Comp. Physiol.* **164**, 151-163.

Fuchs, P. A., Nagai, T. and Evans, M. G. (1988). Electrical tuning in hair cells isolated from the chick cochlea. *J. Neurosci.* **8**, 2460-2467.

Glowatzki, E. and Fuchs, P. A. (2002). Transmitter release at the hair cell ribbon synapse. *Nat. Neurosci.* **5**, 147-154.

Holt, J. R. and Corey, D. P. (2000). Two mechanisms for transducer adaptation in vertebrate hair cells. *Proc. Natl. Acad. Sci. USA* **97**, 11730-11735.

Holt, J. R., Gillespie, S. K., Provance, D. W., Shah, K., Shokat, K. M., Corey, D. P., Mercer, J. A. and Gillespie, P. G. (2002). A chemical-genetic strategy implicates myosin-1c in adaptation by hair cells. *Cell* **108**, 371-381.

Howard, J. and Hudspeth, A. J. (1987). Mechanical relaxation of the hair bundle mediates adaptation in mechanoelectrical transduction by the bullfrog's saccular hair cell. *Proc. Natl. Acad. Sci. USA* **84**, 3064-3068.

Howard, J. and Hudspeth, A. J. (1988). Compliance of the hair bundle associated with gating of mechanoelectrical transduction channels in the bullfrog's saccular hair cell. *Neuron* **1**, 189-199.

Hudspeth, A. J. and Lewis, R. S. (1988). A model for electrical resonance and frequency tuning in saccular hair cells of the bullfrog, *Rana catesbeiana. J. Physiol.* **400**, 275-297.

Hudspeth, A. J., Choe, Y., Mehta, A. D. and Martin, P. (2000). Putting ion channels to work: mechanoelectrical transduction, adaptation, and amplification by hair cells. *Proc. Natl. Acad. Sci. USA* **97**, 11765-11772.

Johnson, S. L., Adelman, J. P. and Marcotti, W. (2007). Genetic deletion of SK2 channels in mouse inner hair cells prevents the developmental linearization in the Ca^{2+} dependence of exocytosis. *J. Physiol.* **583**, 631-646.

Jørgensen, F. and Kroese, A. B. (2005). Ion channel regulation of the dynamical instability of the resting membrane potential in saccular hair cells of the green frog (*Rana esculenta*). *Acta Physiol. Scand.* **185**, 271-290.

Jülicher, F., Andor, D. and Duke, T. (2001). Physical basis of two-tone interference in hearing. *Proc. Natl. Acad. Sci. USA* **98**, 9080-9085.

Keen, E. C. and Hudspeth, A. J. (2006). Transfer characteristics of the hair cell's afferent synapse. *Proc. Natl. Acad. Sci. USA* **103**, 5537-5542.

Kennedy, H. J., Crawford, A. C. and Fettiplace, R. (2005). Force generation by mammalian hair bundles supports a role in cochlear amplification. *Nature* **433**, 880-883.

Kozlov, A. S., Risler, T. and Hudspeth, A. J. (2007). Coherent motion of stereocilia assures the concerted gating of hair-cell transduction channels. *Nat. Neurosci.* **10**, 87-92.

LeMasurier, M. and Gillespie, P. G. (2005). Hair-cell mechanotransduction and cochlear amplification. *Neuron* **48**, 403-415.

Lewis, R. S. and Hudspeth, A. J. (1983). Voltage- and ion-dependent conductances in solitary vertebrate hair cells. *Nature* **304**, 538-541.

Martin, P. and Hudspeth, A. J. (1999). Active hair-bundle movements can amplify a hair cell's response to oscillatory mechanical stimuli. *Proc. Natl. Acad. Sci. USA* **96**, 14306-14311.

Martin, P. and Hudspeth, A. J. (2001). Compressive nonlinearity in the hair bundle's active response to mechanical stimulation. *Proc. Natl. Acad. Sci. USA* **98**, 14386-14391.

Martin, P., Bozovic, D., Choe, Y. and Hudspeth, A. J. (2003). Spontaneous oscillation by hair bundles of the bullfrog's sacculus. *J. Neurosci.* **23**, 4533-4548.

Ospeck, M., Eguíluz, V. M. and Magnasco, M. O. (2001). Evidence of a Hopf bifurcation in frog hair cells. *Biophys. J.* **80**, 2597-2607.

Ospeck, M., Dong, X. X. and Iwasa, K. H. (2003). Limiting frequency of the cochlear amplifier based on electromotility of outer hair cells. *Biophys. J.* **84**, 739-749.

Ricci, A. J. and Fettiplace, R. (1998). Calcium permeation of the turtle hair cell mechanotransducer channel and its relation to the composition of endolymph. *J. Physiol. Lond.* **506**, 159-173.

Ricci, A. J., Crawford, A. C. and Fettiplace, R. (2000). Active hair bundle motion linked to fast transducer adaptation in auditory hair cells. *J. Neurosci.* **20**, 7131-7142.

Ricci, A. J., Crawford, A. C. and Fettiplace, R. (2002). Mechanisms of active hair bundle motion in auditory hair cells. *J. Neurosci.* **22**, 44-52.

Ricci, A. J., Kennedy, H. J., Crawford, A. C. and Fettiplace, R. (2005). The transduction channel filter in auditory hair cells. *J. Neurosci.* **25**, 7831-7839.

Roux, I., Safieddine, S., Nouvian, R., Grati, M., Simmler, M.-C., Bahloul, A., Perfettini, I., Le Gall, M., Rostaing, P., Hamard, G. et al. (2006). Otoferlin, defective in a human deafness form, is essential for exocytosis at the auditory ribbon synapse. *Cell* **127**, 277-289.

Russell, I. J. and Sellick, P. M. (1983). Low-frequency characteristics of intracellularly recorded receptor potentials in guinea-pig cochlear hair cells. *J. Physiol.* **338**, 179-206.

Rutherford, M. A. and Roberts, W. M. (2006). Frequency selectivity of synaptic exocytosis in frog saccular hair cells. *Proc. Natl. Acad. Sci. USA* **103**, 2898-2903.

Schnee, M. E., Lawton, D. M., Furness, D. N., Benke, T. A. and Ricci, A. J. (2005). Auditory hair cell-afferent fiber synapses are specialized to operate at their best frequencies. *Neuron* **47**, 243-254.

Stauffer, E. A., Scarborough, J. D., Hirono, M., Miller, E. D., Shah, K., Mercer, J. A., Holt, J. R. and Gillespie, P. G. (2005). Fast adaptation in vestibular hair cells requires Myosin-1c activity. *Neuron* **47**, 541-553.

Steinacker, A. and Romero, A. (1992). Voltage-gated potassium current and resonance in toadfish saccular hair cells. *Brain Res.* **574**, 229-236.

Sugihara, I. and Furukawa, T. (1989). Morphological and functional aspects of two different types of hair cells in the goldfish sacculus. *J. Neurophysiol.* **62**, 1330-1343.

Vilfan, A. and Duke, T. (2003). Two adaptation processes in auditory hair cells together can provide an active amplifier. *Biophys. J.* **85**, 191-203.

Vollrath, M. A. and Eatock, R. A. (2003). Time course and extent of mechanotransducer adaptation in mouse utricular hair cells: comparison with frog saccular hair cells. *J. Neurophysiol.* **90**, 2676-2689.

Wu, Y.-C., Ricci, A. J. and Fettiplace, R. (1999). Two components of transducer adaptation in auditory hair cells. *J. Neurophysiol.* **82**, 2171-2181.

Zweig, G. (2003). Cellular cooperation in cochlear mechanics. In *Biophysics of the Cochlea: From Molecules to Models* (ed. A. W. Gummer), pp. 315-329. New Jersey, London, Singapore, Hong Kong: World Scientific.