translocating from the ER to CpG DNA in the lysosome. Nat. Immunol. *5*, 190–198.
11. Brint, E.K., Xu, D., Liu, H., Dunne, A., McKenzie, A.N., O'Neill, L.A., and Liew, F.Y. (2004). ST2 is an inhibitor of interleukin 1 receptor and Toll-like receptor 4 signaling and maintains endotoxin tolerance. Nat. Immunol. *5*, 373–379.
12. DeYoung, B.J., and Innes, R.W. (2006). Plant NBS-LRR proteins in pathogen sensing and host defense. Nat. Immunol. *7*, 1243–1249.

School of Biochemistry and Immunology, Trinity College Dublin, Ireland.
E-mail: laoneill@tcd.ie

DOI: 10.1016/j.cub.2008.02.021

Cochlear Mechanics: No Shout but a Twist in the Absence of Prestin

Mammalian hearing is boosted by mechanically active auditory receptor cells, the outer hair cells which amplify the actions of incoming sounds. Recent evidence indicates that the molecular motor that drives this amplification, prestin, may do more than boogie.

Joseph Santos-Sacchi

About a half century ago, in experiments on the mammalian inner ear, Bekesy [1] observed acoustically driven, frequency-tuned responses of the basilar membrane, the cochlear structure which bears the sound-detecting organ of Corti. These tuned responses underlie tonotopicity, the mapping of characteristic frequency onto anatomical space throughout the auditory system. Modern measurements have shown that the peripheral auditory system response — involving sequentially the basilar membrane, hair cell and eighth nerve fiber — is nonlinear with intensity, at least at low intensities. Measures of basilar membrane motion near threshold are thus several orders of magnitude larger than Bekesy's atomic scale estimates [2]. This nonlinear response is the hallmark of cochlear amplification, and is vulnerable to metabolic insult. Metabolic lability issues from the fragile outer hair cell, one of two receptor cell types in the inner ear.

The outer hair cell employs, as Gold's 'negative resistance' hypothesis predicted [3], voltage-driven mechanical activity which helps counter cochlear fluid viscosity, thus enabling sharply tuned mechanical responses [4–6]. The identification of a unique motor protein [7], termed prestin, which is found exclusively in the outer hair cell, shed light on the molecular basis of this amplificatory process (Figure 1). As expected, elimination of prestin by knocking out its gene produced mice that were deaf, despite having normal stereociliar mechanisms [8,9], attesting to the dominant role of prestin in mammalian cochlear amplification.

In all experiments to date in which normal outer hair cell function or specifically prestin function has been compromised, physiological measures have shown broadened frequency tuning, shifts in characteristic frequency to lower frequencies, and loss in auditory sensitivity. These experiments include studies that have also measured basilar membrane motion [10–12]. But the first measurements of basilar membrane tuning in prestin knockout mice, reported recently in *Current Biology* [13], have shown the story is more complex than suspected.

Mellado Lagarde *et al.* [13] observed that the effect on the peripheral auditory system was different when prestin was silenced by interfering with its function, for example with salicylate or anoxia, compared to when it was molecularly absent from the outer hair cell, as in the knockout mouse. In the former case, the sensitivity of both basilar membrane motion and eighth nerve activity are compromised, whereas in the latter case basilar membrane sensitivity is maintained, albeit at a shifted characteristic frequency with poor tuning, but eighth nerve activity is devastated. This is an interesting finding, as basilar membrane motions and eighth nerve fiber activity are expected to show a tight correspondence, at least in the characteristic frequency region [14]. The authors rightly point out that the dichotomy in response sensitivities between nerve and basilar membrane in the knockout mouse highlights the critical link between basilar membrane motion and inner hair cell stereociliar shear, the drive for forward transduction and auditory sensation [15]. The inner hair cell is the other purely sensory cell that synapses with the majority of eighth nerve afferents. The paper [13] thus presents two interesting findings on the knockout mouse: first, that basilar membrane responses appear as acoustically sensitive as normal; and second, that despite the high sensitivity, coupling between basilar membrane and inner hair cell is lost. How can the absence of prestin do this?

We must revisit the effects of the prestin knockout on the outer hair cell itself. It is well know that prestin influences the static and dynamic mechanical properties of the outer hair cell [5,16–18]. There are several million prestin molecules within an outer hair cell membrane [19], and the absence of molecules reduces the membrane surface area and produces shorter cells [9]. This, in turn, affects the cell's mechanical properties. Cell stiffness also relies directly on the activity of prestin in the membrane ([17], but see [20]), possibly through cytoskeletal interactions. Collectively, these changes could alter cochlear partition impedance and account for the observed shifts in best frequency in the passive basilar membrane of the knockout mice; but can they account for the apparent increase in basilar membrane motion over that found in anoxic animals with normal prestin? Can a simple change in basilar membrane impedance and resonance overcome viscous damping effects? Or perhaps prestin did not evolve to overcome viscous damping.

Consider this evolutionary scenario that might help us understand the data. A passive cochlear with outer hair cells lacking prestin will have basilar membrane motion that we consider sensitive, but really it is just how things work in the presence of normal viscous damping. In this case, tuning at the passive characteristic frequency and coupling between basilar membrane

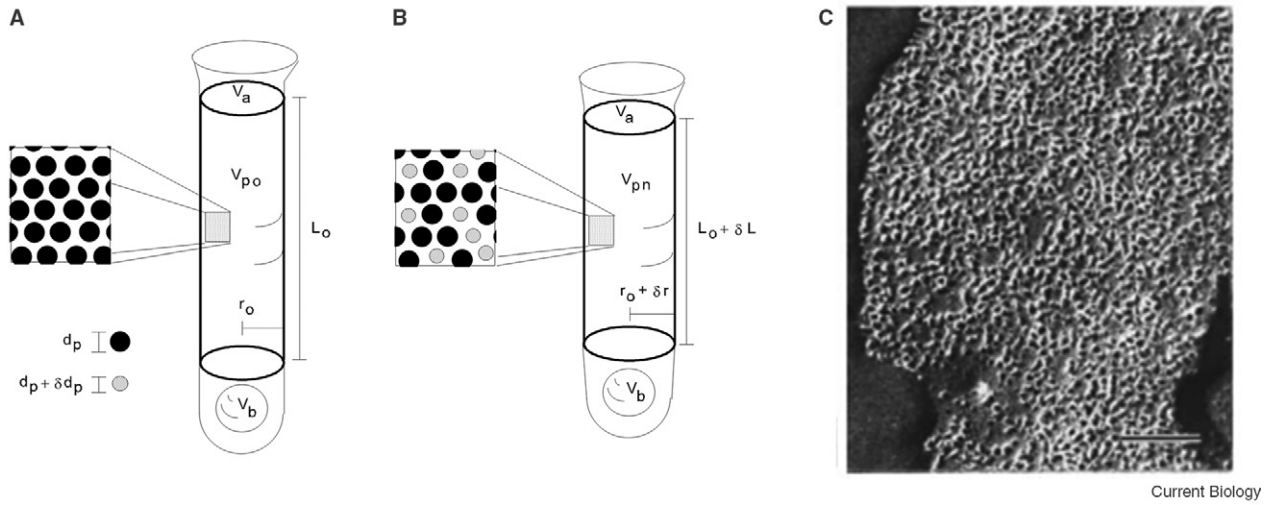

Figure 1. Prestin and outer hair cell function.

The outer hair cell soma is schematized in the fully elongated (A) and partially contracted (B) condition, corresponding to hyperpolarization and depolarization, respectively. (The abbreviations refer to a popular Boltzmann model of motility where motors fluctuate between two states: d_p, diameter of motor; δ d_p, change in diameter when motor contracts; L_o, normal length of the lateral membrane; δ L, change in length when motors contract; r_o, normal radius of the cylindrical cell; δr, change when motors contract; V_a and V_b, constant volume of apex and base, respectively; V_{po} and V_{pn}, volumes of motor-containing cell region during elongation and contraction, respectively.) The outer hair cell motor, prestin, is embedded within the lateral membrane of the cell, forming a dense array. Motors (dark and light circles in the blowup) are depicted in compact or expanded states, corresponding to the two-state model of their activity. Electrical measures of the motor's voltage sensor provide estimates of millions of motor molecules per cell. The lateral membrane (C) when viewed with freeze fracture electron microscopy, displays a dense array of 10 nm particles, which are believed to represent multimers of prestin. The dense packing must have substantial structural consequences beyond the dynamic activity of the protein. (Reproduced with permission from [19].)

and inner hair cell stereociliar shear are normally poor. Now, for mammals, there was a need for auditory sensitivity at higher frequencies. Prestin evolved to improve the coupling of basilar membrane motion to inner hair cell stereociliar shear and simultaneously shift characteristic frequency to higher frequencies, but in the process, the tightened mechanical coupling caused a reduction in the sensitivity of basilar membrane motion, thus necessitating an amplificatory mechanism to reestablish high sensitivity at the new higher characteristic frequency. In this scenario the passive presence of prestin enhances coupling, but is detrimental to basilar membrane motion, and the electrical activity of prestin amplifies the movements of the well coupled basilar membrane.

How could a protein within the lateral membrane of the outer hair cell work to couple basilar membrane motion into inner hair cell stereociliar shear? The organ of Corti is a mass of supporting cells and hair cells bounded apically by the reticular lamina/tectorial membrane and basally by the basilar membrane. Displacements of the basilar membrane, induced acoustically, must be sensed at the apical region where stereocilia of inner hair cells and outer hair cells reside. It is possible that the increase in stiffness provided to the outer hair cells by the immense numbers of prestin protein in the lateral membrane reinforces or stiffens the cellular link between basilar membrane and reticular lamina.

If this is true, we might predict that a knock-in of a nonfunctional prestin into the knock-out mouse would produce a basilar membrane response at lower characteristic frequency, as we see in the knock-out, but with a reduced sensitivity that would reflect an enhanced coupling between basilar membrane and inner hair cell stereocilia. We know how long it takes to get a new mutant mouse. So it may be a while before we get at the deeper significance of the new data reported by Mellado Lagarde *et al.* [13].

References

1. von Bekesy, G. (1960). Experiments in Hearing (New York: McGraw-Hill Book Company).
2. Rhode, W.S. (1971). Observations of the vibration of the basilar membrane in squirrel monkeys using the Mossbauer technique. J. Acoust. Soc. Am. Suppl. *49*, 1218–1231.
3. Gold, T. (1948). Hearing. II. The physical basis of the action of the cochlea. Proc. R. Soc. Lond. B *135*, 492–498.
4. Brownell, W.E., Bader, C.R., Bertrand, D., and de Ribaupierre, Y. (1985). Evoked mechanical responses of isolated cochlear outer hair cells. Science *227*, 194–196.
5. He, D.Z., Zheng, J., Kalinec, F., Kakehata, S., and Santos-Sacchi, J. (2006). Tuning in to the amazing outer hair cell: membrane wizardry with a twist and shout. J. Membr. Biol. *209*, 119–134.
6. Ashmore, J. (2008). Cochlear outer hair cell motility. Physiol. Rev. *88*, 173–210.
7. Zheng, J., Shen, W., He, D.Z., Long, K.B., Madison, L.D., and Dallos, P. (2000). Prestin is the motor protein of cochlear outer hair cells. Nature *405*, 149–155.
8. Cheatham, M.A., Huynh, K.H., Gao, J., Zuo, J., and Dallos, P. (2004). Cochlear function in Prestin knockout mice. J. Physiol. *560*, 821–830.
9. Liberman, M.C., Gao, J., He, D.Z., Wu, X., Jia, S., and Zuo, J. (2002). Prestin is required for electromotility of the outer hair cell and for the cochlear amplifier. Nature *419*, 300–304.
10. Ruggero, M.A., and Rich, N.C. (1991). Furosemide alters organ of corti mechanics: evidence for feedback of outer hair cells upon the basilar membrane. J. Neurosci. *11*, 1057–1067.
11. Murugasu, E., and Russell, I.J. (1995). Salicylate ototoxicity: the effects on basilar membrane displacement, cochlear microphonics, and neural responses in the basal turn of the guinea pig cochlea. Auditory Neurosci. *1*, 139–150.
12. Santos-Sacchi, J., Song, L., Zheng, J., and Nuttall, A.L. (2006). Control of mammalian cochlear amplification by chloride anions. J. Neurosci. *26*, 3992–3998.

13. Mellado Lagarde, M.M., Drexl, M., Lukashkin, A.N., Zuo, J., and Russell, I.J. (2008). Prestin's role in cochlear frequency tuning and transmission of mechanical responses to neural excitation. Curr. Biol. *18*, 200–202.
14. Narayan, S.S., Temchin, A.N., Recio, A., and Ruggero, M.A. (1998). Frequency tuning of basilar membrane and auditory nerve fibers in the same cochleae. Science *282*, 1882–1884.
15. Nowotny, M., and Gummer, A.W. (2006). Nanomechanics of the subtectorial space caused by electromechanics of cochlear outer hair cells. Proc. Natl. Acad. Sci. USA *103*, 2120–2125.
16. Spector, A.A., Deo, N., Grosh, K., Ratnanather, J.T., and Raphael, R.M. (2006). Electromechanical models of the outer hair cell composite membrane. J. Membr. Biol. *209*, 135–152.
17. He, D.Z., Jia, S., and Dallos, P. (2003). Prestin and the dynamic stiffness of cochlear outer hair cells. J. Neurosci. *23*, 9089–9096.
18. Jensen-Smith, H., and Hallworth, R. (2007). Lateral wall protein content mediates alterations in cochlear outer hair cell mechanics before and after hearing onset. Cell Motil. Cytoskel. *64*, 705–717.
19. Santos-Sacchi, J., Kakehata, S., Kikuchi, T., Katori, Y., and Takasaka, T. (1998). Density of motility-related charge in the outer hair cell of the guinea pig is inversely related to best frequency. Neurosci. Lett. *256*, 155–158.
20. Hallworth, R. (2007). Absence of voltage-dependent compliance in high-frequency cochlear outer hair cells. J. Assoc. Res. Otolaryngol. *8*, 464–473.

Department of Surgery (Otolaryngology), Cellular and Molecular Physiology, and Neurobiology, Yale University School of Medicine, BML 244, 333 Cedar Street, New Haven, Connecticut 06510, USA.
E-mail: joseph.santos-sacchi@yale.edu

DOI: 10.1016/j.cub.2008.02.019

Chemical Coevolution: Host–Parasite Arms Race Runs Hot and Cold

Coevolution is a major process operating across biological communities at a range of spatial scales. Rapid ecological change makes it vital that we understand how coevolution proceeds if we are to conserve genetic diversity, combat disease and predict the effects of species invasions.

Duncan E. Jackson

Coevolution is a reciprocal evolutionary change in the genetic composition of one species in response to change in another interacting species. We can only invoke coevolution as an explanation when the observed traits of two species have evolved through ongoing interaction between the two species. Research has shown that coevolution is a dynamic process that can continually change the nature of inter-specific interactions over broad geographic ranges [1]. The current rapid environmental change makes it essential that we understand coevolutionary processes if we are to predict the changing dynamics of biological communities, conserve biodiversity and combat disease [2]. Some of the best examples of coevolution are found in mutualisms, especially flowers and their pollinators, but several 'coevolutionary arms races' are well-documented in the context of predator–prey and host–parasite relationships. Nash *et al.* [3] have recently shed fresh light on dynamic coevolutionary processes with a long-term study of the relationship between the parasitic Alcon blue butterfly (*Maculinea alcon*) and its *Myrmica* ant host. They have shown that Alcon blue caterpillars adopt a chemical disguise, matching that of ant larvae, to mediate their 'adoption' into *Myrmica* nests, and that local variations in ant chemistry are closely matched by their caterpillar parasites.

Unlike the familiar examples of mutualistic coevolution seen in flowering plants and their pollinators, coevolving parasites and their hosts are engaged in an antagonistic process. The most important property of a parasite that is attributed to coevolution is its virulence. A more virulent parasite will reduce a host's fitness more quickly, relative to an unparasitised host. Clearly selection must favour highly virulent parasites, those which more quickly exploit host resources, but this process might lead to the death of all potential hosts. However, if a parasite is so virulent that it kills its host before transmission of the parasite's offspring then this will select for a reduction in parasitic virulence, as well as more resistant hosts. The best-cited example of how parasitic virulence can decline is that of myxomatosis among European and Australian rabbits during the 1950s. An initially maximally virulent myxoma virus (100% kill) declined rapidly, accompanied by sharp increases in host resistance, such that the disease was seldom seen after less than 20 years. The myxoma virus still persists in populations in an attenuated form, although on occasions more virulent strains appear causing short-lived, localised outbreaks.

Antagonistic coevolution can lead to static equilibria where an optimal situation is reached, such as may be the case with the myxoma virus, but also dynamic equilibria where adaptive improvement is always possible. Dynamic equilibria are known as the 'Red Queen hypothesis' because, as

Figure 1. An Alcon blue butterfly (*Maculinea alcon*) lays her eggs on marsh gentian (*Gentiana pneumonanthe*) flower heads.

After 2–3 weeks of feeding on flower heads the caterpillar lowers itself to the ground using a silken thread and awaits discovery by foraging *Myrmica* ants. Alcon blue caterpillars are chemical mimics of *Myrmica* ant larvae, an attribute which tricks ants into carrying caterpillars back to their nest ('adoption') where they become social parasites. (Photo: David Nash.)