# Supplemental Information

# Waves on Reissner's membrane: a mechanism for the propagation of otoacoustic emissions from the cochlea

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# 1. Extended Experimental Procedures

### *A. Fluid dynamics of the waves on Reissner's membrane*

We consider Reissner's membrane within an *x,y,z* coordinate system (Figure 1B) and describe the hydrodynamics in the plane $y = 0$ that lies perpendicular to the membrane and along its midline. For small displacements and incompressible fluids, the pressures $p_1$ and $p_2$, respectively above and below the membrane, satisfy the Laplace equations

$$\Delta p_1 = 0,\ \Delta p_2 = 0. \tag{S1}$$

We consider a high angular stimulation frequency $\omega$ for which the height of the scalae exceeds the wavelength. The pressures must therefore vanish far from the boundaries, which is fulfilled in the ansatz

$$\begin{aligned} p_1 &= -\tilde{p}e^{i\omega t - ikx - kz} + c.c., \\ p_2 &= \tilde{p}e^{i\omega t - ikx + kz} + c.c., \end{aligned} \tag{S2}$$

in which *c.c.* represents the complex conjugate. The pressures satisfy the Laplace relations (Equations S1) and decay exponentially with distance from the membrane with a length scale proportional to the wavelength $\lambda = 2\pi/k$.

Reissner's membrane imposes a boundary condition (Equation 2). Because $\rho\partial_t^2 X_{\mathrm{RM}} = -\partial_z p_1\big|_{z=0} = -\partial_z p_2\big|_{z=0}$, we obtain the dispersion relation (Equation 4).

### *B. Wave propagation on the parallel Reissner's membrane and basilar membrane*

To solve the Laplace relations (Equation 6) together with the boundary conditions (Equations 7 and 8), we employ the ansatz

$$\begin{aligned} p_1 &= \tilde{p}_1(x)\cosh\left[\partial_x b(x)(z-3h)\right]e^{i\omega t - i\omega b(x) - \partial_x b(x)h} + c.c., \\ p_2 &= \left\{\tilde{p}_2^u(x)\cosh\left[\partial_x b(x)(z-2h)\right] + \tilde{p}_2^d(x)\cosh\left[\partial_x b(x)(z-h)\right]\right\}e^{i\omega t - i\omega b(x) - \partial_x b(x)h} + c.c., \\ p_3 &= \tilde{p}_3(x)\cosh\left[\partial_x b(x)z\right]e^{i\omega t - i\omega b(x) - \partial_x b(x)h} + c.c.. \end{aligned} \tag{S3}$$

Because the local wave vector $k(x)$ is related to the phase $b(x)$ by $k(x) = \omega \partial_x b(x)$, the phase may be expressed through the wave vector, $b(x) = \int_0^x dx' k(x')/\omega$.

The pressures yield the velocities of Reissner's membrane and the basilar membrane,

$$\begin{aligned} -\rho \partial_t V_{\mathrm{RM}} &= \partial_z p_1\big|_{z=2h} = \partial_z p_2\big|_{z=2h}, \\ -\rho \partial_t V_{\mathrm{BM}} &= \partial_z p_2\big|_{z=h} = \partial_z p_3\big|_{z=h}. \end{aligned} \tag{S4}$$

Applying the WKB approximation, we consider an expansion in powers of the angular frequency $\omega$ (Steele & Taber, 1979; Lighthill, 1996; Reichenbach & Hudspeth, 2010b). A high frequency implies a small wavelength $\lambda(x)$, a length scale over which the basilar-membrane impedance $Z_{\mathrm{BM}}(x)$ varies little. The spatial variation of the pressure amplitudes, $\partial_x p_n$ ($n$ = 1,2,3), then results predominantly from the derivative of the phase, $\partial_x b(x)$: the corresponding terms are of order $\omega$ whereas terms that involve $\partial_x \tilde{p}_1$, $\partial_x \tilde{p}_2^u$, $\partial_x \tilde{p}_2^d$, $\partial_x \tilde{p}_3$ and $\partial_x^2 b(x)$ are of the smaller order 1. To leading order $\omega^2$ we hence find $\partial_x^2 p_n = -[\partial_x b(x)]^2 p_n$ ($n$ = 1,2,3). Because $\partial_z^2 p_n = [\partial_x b(x)]^2 p_n$, the pressures satisfy the Laplace relations (Equations 6).

To leading order $\omega^2$ the boundary conditions yield

$$\begin{aligned} \tilde{p}_2^d(x) &= -\tilde{p}_1, \\ \tilde{p}_2^u(x) &= -\tilde{p}_3, \end{aligned} \tag{S5}$$

as well as

$$\begin{aligned} \frac{\tilde{p}_3}{\tilde{p}_1} &= \frac{ik(x)Z_{\mathrm{RM}}}{\rho\omega} \sinh[k(x)h] - 2\cosh[k(x)h], \\ \frac{\tilde{p}_1}{\tilde{p}_3} &= \frac{ik(x)Z_{\mathrm{BM}}(x)}{\rho\omega} \sinh[k(x)h] - 2\cosh[k(x)h]. \end{aligned} \tag{S6}$$

The last two equations for the ratio of the pressure amplitudes $\tilde{p}_1$ and $\tilde{p}_3$ must agree, which gives the dispersion relation (Equation 10).

Because the basilar-membrane mode exhibits a large wavelength, $|k(x)|h \ll 1$, we can approximate $\sinh[k(x)h] \approx k(x)h$ and $\cosh[k(x)h] \approx 1$ to obtain

$$k_{1/2}^2(x) = \frac{\rho\omega}{hZ_{\mathrm{RM}}Z_{\mathrm{BM}}(x)} \left\{ -i[Z_{\mathrm{RM}} + Z_{\mathrm{BM}}(x)] \pm i\sqrt{Z_{\mathrm{RM}}^2 - Z_{\mathrm{RM}}Z_{\mathrm{BM}}(x) + Z_{\mathrm{BM}}^2(x)} \right\} \tag{S7}$$

When the impedance of Reissner's membrane is much below that of the basilar membrane, $Z_{\mathrm{RM}} << Z_{\mathrm{BM}}(x)$, we find that

$$k^2(x) = -\frac{3i\rho\omega}{2hZ_{\mathrm{BM}}(x)} \tag{S8}$$

and the wave vector $k(x)$ depends on only the basilar membrane's impedance. Because the wavelength of this mode is much greater than the height of the channels, the system can be regarded as one-dimensional and the WKB approximation yields an amplitude of basilar-membrane vibration that varies in proportion to $\sqrt{k(x)}$ (Steele and Taber, 1979; Lighthill, 1996; Reichenbach and Hudspeth, 2010b).

The impedances of Reissner's membrane and the basilar membrane are comparable near the cochlear apex. Because stimulation at frequencies below 1 kHz elicits large wavelengths for both modes, the respective wave vectors follow from Equation S7. The impedance of Reissner's membrane is dominated by the membrane's transverse flexion, $Z_{\mathrm{RM}} \approx -8iT/(\omega w^2)$, and the wavelength of the corresponding wave mode is thus inversely proportional to the frequency, $\lambda \sim f^{-1}$.

### *C. Green's functions*

The Green's functions, the pressures $p_1^{(G;x_0,\omega)}$, $p_2^{(G;x_0,\omega)}$, and $p_3^{(G;x_0,\omega)}$, fulfill the Laplace relations (Equations 6) together with the boundary conditions (Equations 7 and 8), but the boundary condition at the basilar membrane is given by Equation 13. The WKB approximation again facilitates the solution. As shown above, a wave's local wave vector follows from the local impedance alone, irrespective of putative impedance changes. We therefore start by considering a uniform basilar-membrane impedance $Z_{\mathrm{BM}}$ and make the ansatz

$$
\begin{aligned}
p_1^{(G;x_0,\omega)} &= \int_{-\infty}^{\infty} dk G_1(k)\cosh\left[k(z-3h)\right]e^{i\omega t-ik(x-x_0)-kh} + c.c.,\\
p_2^{(G;x_0,\omega)} &= \int_{-\infty}^{\infty} dk\left\{G_2^u(k)\cosh\left[k(z-2h)\right]+G_2^d(k)\cosh\left[k(z-h)\right]\right\}e^{i\omega t-ik(x-x_0)-kh} + c.c.,\\
p_3^{(G;x_0,\omega)} &= \int_{-\infty}^{\infty} dk G_3(k)\cosh\left[kz\right]e^{i\omega t-ik(x-x_0)-kh} + c.c..
\end{aligned}
\tag{S9}
$$

From the boundary conditions $\partial_z p_1^{(G;x_0,\omega)}\big|_{z=2h} = \partial_z p_2^{(G;x_0,\omega)}\big|_{z=2h}$ and $\partial_z p_2^{(G;x_0,\omega)}\big|_{z=h} = \partial_z p_3^{(G;x_0,\omega)}\big|_{z=h}$, we obtain $G_2^d(k) = -G_1(k)$ and $G_2^u(k) = -G_3(k)$. Because the Dirac d□□□□□□□□□□□□□ can be represented as

$$
\delta(x-x_0) = \frac{1}{2\pi}\int_{-\infty}^{\infty} dk e^{-ik(x-x_0)}, \tag{S10}
$$

we compute

$$
\begin{aligned}
G_1(k) &= \frac{p_F e^{kh}}{2\pi L(k)},\\
G_3(k) &= \left[\frac{ikZ_{\mathrm{RM}}}{\rho\omega}\sinh(kh) - 2\cosh(kh)\right]\frac{p_F e^{kh}}{2\pi L(k)},
\end{aligned}
\tag{S11}
$$

in which $L(k)$ is defined as

$$
L(k) = \left[\frac{ikZ_{\mathrm{RM}}}{\rho\omega}\sinh(kh) - 2\cosh(kh)\right]\left[\frac{ikZ_{\mathrm{BM}}}{\rho\omega}\sinh(kh) - 2\cosh(kh)\right] - 1. \tag{S12}
$$

With this notation the dispersion relation (Equation 10) reads $L(k) = 0$.

In considering the propagation of distortion products, we are interested in waves far from their generation site $x_0$. The integrals in Equations S9 can then be calculated by closing the contour of integration in the complex plane (Figure S1). Complex analysis informs us that only the poles of the integrand contribute to an integral along such a closed path. Poles occur at those values $k$ for which $L(k)$ vanishes, and hence at the values $\pm k_a$ and $\pm k_b$ with $k_a, k_b > 0$ that describe the two wave modes in the cochlea. If the impedances $Z_{\mathrm{BM}}$ and $Z_{\mathrm{RM}}$ involve friction, the solutions $\pm k_a$ and $\pm k_b$ possess small imaginary parts and are located in the second and fourth quadrants of the complex plane (Figure S1). For $x < x_0$ we can close the integration contour in the upper half plane, and obtain contributions from $-k_a$ and $-k_b$ that describe retrograde waves. In the opposite case, when $x > x_0$, the contour can be closed in the lower half plane to yield contributions from $k_a$ and $k_b$ and forward-traveling waves.

Because we are interested in the retrograde waves that reach the stapes, we consider $x < x_0$. Denote as $p_n^{(G,a;x_0,\omega)}$ ($n$ = 1,2,3) the contribution to the pressure $p_n^{(G;x_0,\omega)}$ from the pole at $-k_a$ and denote as $p_n^{(G,b;x_0,\omega)}$ ($n$ = 1,2,3) the contribution from the pole at $-k_b$. The pressures $p_n^{(G,a;x_0,\omega)}$ therefore represent the pressures of the Reissner's membrane mode and the pressures $p_n^{(G,b;x_0,\omega)}$ those of the basilar-membrane mode. We find $p_n^{(G;x_0,\omega)} = p_n^{(G,a;x_0,\omega)} + p_n^{(G,b;x_0,\omega)}$ with

$$
\begin{aligned}
p_1^{(G,a;x_0,\omega)} &= 2\pi i \left[ \partial_k G_1^{-1}(k)\Big|_{k=-k_a} \right]^{-1} \cosh\left[k_a(z-3h)\right] e^{i\omega t + ik_a(x-x_0)+k_a h} + c.c., \\
p_2^{(G,a;x_0,\omega)} &= -2\pi i \left\{ \left[ \partial_k G_3^{-1}(k)\Big|_{k=-k_a} \right]^{-1} \cosh\left[k_a(z-2h)\right] \right. \\
&\qquad \left. + \left[ \partial_k G_1^{-1}(k)\Big|_{k=-k_a} \right]^{-1} \cosh\left[k_a(z-h)\right] \right\} e^{i\omega t + ik_a(x-x_0)+k_a h} + c.c., \\
p_3^{(G,a;x_0,\omega)} &= 2\pi i \left[ \partial_k G_3^{-1}(k)\Big|_{k=-k_a} \right]^{-1} \cosh\left[k_a z\right] e^{i\omega t + ik_a(x-x_0)+k_a h} + c.c.,
\end{aligned}
\tag{S13}
$$

and the pressures $p_n^{(G,b;x_0,\omega)}$ follow analogously.

In the actual cochlea the basilar-membrane impedance $Z_{\mathrm{BM}}(x)$ varies with the longitudinal position $x$. As elaborated above, the local wave vectors $k_a$ and $k_b$ also depend on the position $x$. In the WKB approximation the pressure amplitudes vary as $1/\sqrt{k(x)}$ (Steele and Taber, 1979; Lighthill, 1996; Reichenbach and Hudspeth, 2010b). Because in the WKB approximation, and to leading order, only the local wave vector $k(x)$ contributes to the derivatives of the pressures, one verifies that adjusting the pressures in Equation S13 in proportion to $1/\sqrt{k(x)}$ solves the Laplace relations (Equation 6) with the stated boundary conditions (Equations 7, 8, and 13). For the retrograde waves at $x < x_0$ we find

$$p_1^{(G,a;x_0,\omega)} = 2\pi i\sqrt{\frac{k_a(x_0)}{k_a(x)}}\left[\partial_k G_1^{-1}(k)\Big|_{k=-k_a(x_0)}\right]^{-1}\cosh\left[k_a(x)(z-3h)\right]e^{i\omega t+i\int_x^{x_0}dx' k_a(x')+k_a(x)h}+c.c.,$$
$$p_2^{(G,a;x_0,\omega)} = -2\pi i\sqrt{\frac{k_a(x_0)}{k_a(x)}}\left\{\left[\partial_k G_3^{-1}(k)\Big|_{k=-k_a(x_0)}\right]^{-1}\cosh\left[k_a(x)(z-2h)\right]\right.$$
$$\left.+\left[\partial_k G_1^{-1}(k)\Big|_{k=-k_a(x_0)}\right]^{-1}\cosh\left[k_a(x)(z-h)\right]\right\}e^{i\omega t+i\int_x^{x_0}dx' k_a(x')+k_a(x)h}+c.c., \tag{S14}$$
$$p_3^{(G,a;x_0,\omega)} = 2\pi i\sqrt{\frac{k_a(x_0)}{k_a(x)}}\left[\partial_k G_3^{-1}(k)\Big|_{k=-k_a(x_0)}\right]^{-1}\cosh\left[k_a(x)z\right]e^{i\omega t+i\int_x^{x_0}dx' k_a(x')+k_a(x)h}+c.c..$$

The pressures $p_n^{(G,b;x_0,\omega)}$ as well as the case $x > x_0$ follow analogously.

### *D. Distortion products*

The pressure waves produced by nonlinear distortion can be computed through Equation 14 from the Green's functions (Equations S14). This equation contains the Fourier component $\overline{V_{\mathrm{BM}}^3}(x_0,\omega)$ from which $V_{\mathrm{BM}}^3(x_0,t)$ follows as

$$V_{\mathrm{BM}}^3(x_0,t) = \int_0^\infty d\omega\, \overline{V_{\mathrm{BM}}^3}(x_0,\omega)e^{i\omega t}+c.c.. \tag{S15}$$

The Fourier component $\overline{V_{\mathrm{BM}}^3}(x_0,\omega)$ can be expressed through the Fourier component $\tilde{V}_{\mathrm{BM}}(x_0,\omega)$ of $V_{\mathrm{BM}}(x_0,t)$:

$$\overline{V_{\mathrm{BM}}^3}(x_0,\omega) = \left[\tilde{V}_{\mathrm{BM}}(x_0)*\tilde{V}_{\mathrm{BM}}(x_0)*\tilde{V}_{\mathrm{BM}}(x_0)\right](\omega) \tag{S16}$$

in which $*$ denotes the convolution defined by

$$(f*g)(\omega) = \int_{-\infty}^{\infty} d\omega' f(\omega')g(\omega-\omega') = \int_0^\infty d\omega' \left[f(\omega')g(\omega-\omega')+f^*(\omega')g(\omega+\omega')\right]. \tag{S17}$$

The last equality holds when $f(\omega) = f^*(-\omega)$, as is the case when $f(\omega)$ represents the Fourier component of a real-valued function.

To compute the retrograde waves at the cubic distortion frequencies $2f_1 - f_2$ and $2f_2 - f_1$, we consider stimulation of the cochlea at the two primary frequencies $f_1$ and $f_2$. In the linear, passive cochlea the basilar-membrane response then contains only those two frequencies:

$$\tilde{V}_{\mathrm{BM}}(x_0,\omega) = V_{\mathrm{BM}}^{(1)}(x_0)\delta(\omega-\omega_1)+V_{\mathrm{BM}}^{(2)}(x_0)\delta(\omega-\omega_2). \tag{S18}$$

Upon inserting Equation S18 into Equation S16 we find that the basilar-membrane inputs at $f_1$ and $f_2$ produce responses at linear combinations, specifically at frequencies $f \in I$ for which the set $I$ is $I = \left\{ f = \pm f_i \pm f_j \pm f_k \right\}$ with $i,j,k \in \{1,2\}$ and $f > 0$:

$$\overline{V_{\mathrm{BM}}^{3}}\left(x_0,\omega\right) = \sum_{\substack{\omega'=2\pi f \\ f\in I}} S^{(\omega')}(x_0)\delta(\omega-\omega'). \tag{S19}$$

The amplitudes at the distortion frequencies $2f_1 - f_2$ and $2f_2 - f_1$ are

$$\begin{aligned} S^{(2\omega_1-\omega_2)}(x_0) &= 3\left[V_{\mathrm{BM}}^{(1)}(x_0)\right]^2 \left[V_{\mathrm{BM}}^{(2)}(x_0)\right]^*, \\ S^{(2\omega_2-\omega_1)}(x_0) &= 3\left[V_{\mathrm{BM}}^{(2)}(x_0)\right]^2 \left[V_{\mathrm{BM}}^{(1)}(x_0)\right]^*. \end{aligned} \tag{S20}$$

This distortion elicited by the linear, passive basilar-membrane velocity represents the Born approximation to the full, nonlinear Equation 14.

### *E. Parameter values*

We model a cochlea 35 mm in length with a maximal best frequency of $f_{\mathrm{max}}$ = 30 kHz at its base and a minimal best frequency of $f_{\mathrm{min}}$ = 50 Hz at its apex. The longitudinal position $x$ is measured in units of the cochlear length such that $x = 0$ denotes the base and $x = 1$ the apex. The maximal and minimal frequencies define an exponential map $f_0(x)$ of best frequencies in the cochlea in which $f_0(x)$ matches $f_{\mathrm{max}}$ at the base and $f_{\mathrm{min}}$ at the apex.

The specific acoustic impedance $Z_{\mathrm{BM}}(x)$ of the basilar membrane follows from the stiffness, viscosity, and mass. We consider a strip of the basilar membrane with a width of 8 μm, the width of one hair cell. This strip has an area of $A_{\mathrm{BM}}(x) = w_{\mathrm{BM}}(x)\cdot 8$ μm, in which $w_{\mathrm{BM}}(x)$ denotes the membrane's width as a function of the longitudinal position $x$. The impedance follows as

$$Z_{\mathrm{BM}} = A_{\mathrm{BM}}^{-1}(x)\left[-iK(x)/\omega + \mu(x) + i\omega m(x)\right], \tag{S21}$$

in which $K(x)$ is the stiffness, $\mu(x)$ the drag coefficient, and $m(x)$ the mass of the basilar-membrane strip.

At each longitudinal position in the cochlea, the mass and stiffness define a resonant frequency $f_{\mathrm{res}}(x) = (2\pi)^{-1}\sqrt{K(x)/m(x)}$. We assume that, in the basal region of the cochlea, this resonant frequency equals the best frequency $f_0(x)$ and hence consider a stiffness $K(x)$ proportional to $f_0(x)$ and a mass $m(x)$ inversely proportional to $f_0(x)$. We choose a maximal stiffness $K(x=0) = 1\ \mathrm{N \cdot m^{-1}}$ at the base and a mass according to $f_{\mathrm{res}}(x) = f_0(x)$. To represent viscous damping we assume that the drag coefficient $\mu(x)$ is proportional to the membrane's width $w_{\mathrm{BM}}(x)$ with a proportionality coefficient of $0.015\ \mathrm{N \cdot s \cdot m^{-2}}$.

The nonlinearity that produces distortion results from an active process that counteracts viscous damping. We assume that the active process produces a force that is proportional to the basilar-membrane displacement. The coefficient $A$ in Equation 12 is thus inversely proportional to $\omega^3$ as well as to $A_{\mathrm{BM}}(x)$: $A = 5 \cdot 10^{11} \cdot \omega^{-3} \cdot A_{\mathrm{BM}}^{-1}\ \mathrm{kg \cdot m^{-2} \cdot s^{-2}}$.

The remaining parameter values are summarized in Table S1.

## 2. Supplemental Figure Titles and Legends

**Figure S1. Computation of the Green's functions, related to Figure 4.** The functions $p_1^{(G;x_0,\omega)}$, $p_2^{(G;x_0,\omega)}$, and $p_3^{(G;x_0,\omega)}$ can be evalulated through integration of Equations S9 in the complex plane. When $x < x_0$ the integration contour can be closed in the upper half plane and yields contributions from $–k_a$ and $–k_b$ that describe retrograde waves. In the opposite case, when $x > x_0$, the contour can be closed in the lower half plane to provide contributions from $k_a$ and $k_b$ and hence forward-traveling waves.

## 3. Supplemental Movie Titles and Legends

**Movie S1. Interferometric recordings of waves propagating on Reissner's membrane, related to Figure 2.** The images portray the movement along the midline of a segment about 1.5 mm in length near the apex of the guinea pig's cochlea. As quantified in Figure 2, the wavelength decreases with increasing stimulus frequency.

## 4. Supplemental Tables

**Supplemental Figure S1**