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Supporting Material

Tectorial Membrane Morphological Variation: Effects Upon Stimulus Frequency Otoacoustic Emissions

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NOTE: The material below supplements the article entitled *Tectorial Membrane Morphological Variation: Effects Upon Stimulus Frequency Otoacoustic Emissions* by Bergevin et al. (Biophysical Journal, 2010).

I – Spontaneous Emissions

As indicated in the main document, SOAEs were also measured during the course of these experiments. To summarize, there appears to be a clear correlation between SOAE and SFOAE activity. While further study is needed to better understand the inter–relationship between these two emission types in lizards, this correlation is presumably relevant to the primary thesis of the main document (i.e., peripheral mechanisms for tuning and the role of the TM in such).

Methods

For SOAEs, 60 waveforms (32768 sample window, SR= 44.1 kHz) were acquired and the FFT magnitudes averaged, either with or without a suppressor tone present. Despite the presence of external noise, SOAE activity could be readily distinguished in that it was both temperature–dependent as well as suppressible by a nearby external tone (1), (2).

Results

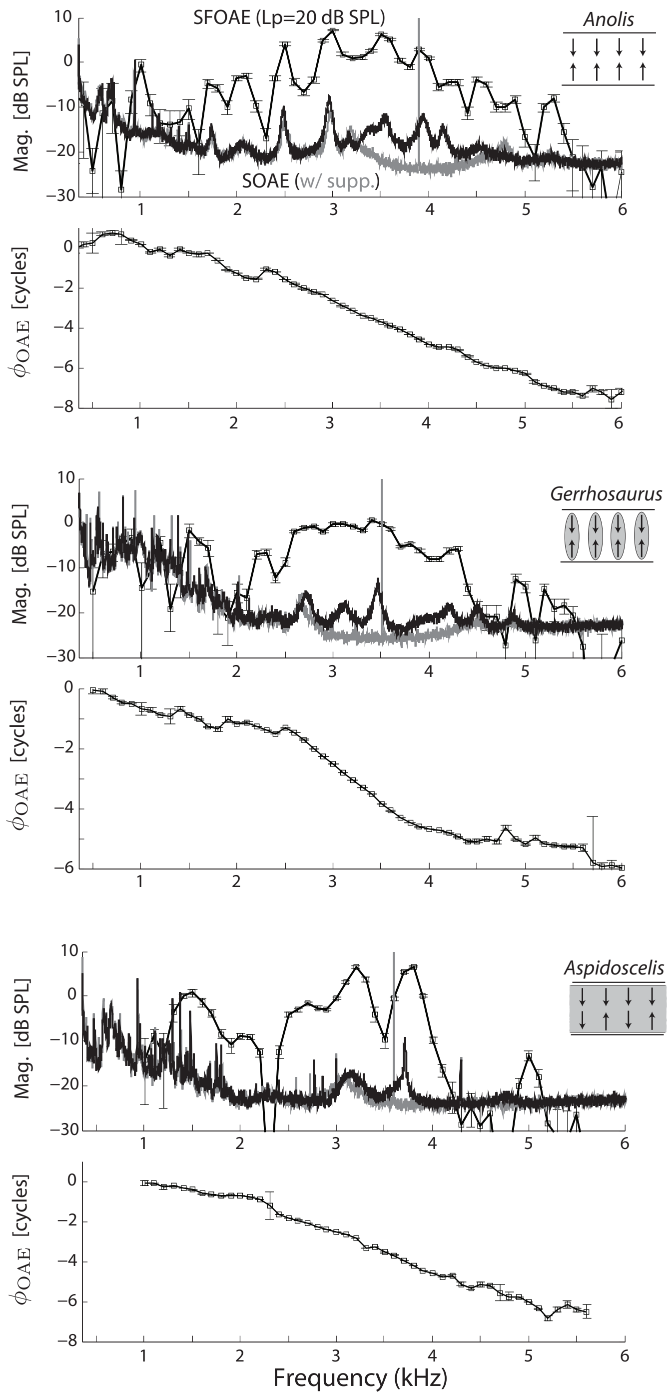
Spontaneous activity, as identified via temperature dependence and suppressibility due to external tones (1), (2), was apparent in the vast majority of ears examined (bottom black trace in Supp. Fig.1). Supplemental Fig. 1 also includes an SOAE spectrum with a 40 dB SPL tone present (grey trace) to demonstrate the resulting region of localized suppression due to the external tone (e.g., (1)). Spontaneous emissions commonly consist of *baseline activity* (a broad, suppressible plateau, (1), (2)) with several distinct, narrowband peaks atop it. However with the exception of the anoles, SOAE activity in the non–TM species (e.g., iguanids, anguids) comprised a suppressible baseline emission with only one or two (if any) distinct and relatively wideband peaks. Furthermore, SOAEs and low-level SFOAE magnitudes were correlated (Supp. Fig.1). A rise in SOAE activity at a given frequency correlated with an increase in SFOAE magnitude. However the converse is not always true: significant SFOAEs could be measured where no SOAE activity was detected.

Differences in SOAEs between TM and non–TM species are also apparent, consistent with previous studies (3), (4). While all species exhibit some degree of baseline activity, species with a continuous TM tend to exhibit fewer and more sharply tuned SOAE peaks while salletal species commonly exhibit more numerous and wider–band peaks. Non-TM species commonly produced one (and sometimes several) broad, smaller peaks, with anoles appearing to be an exception (as described below).

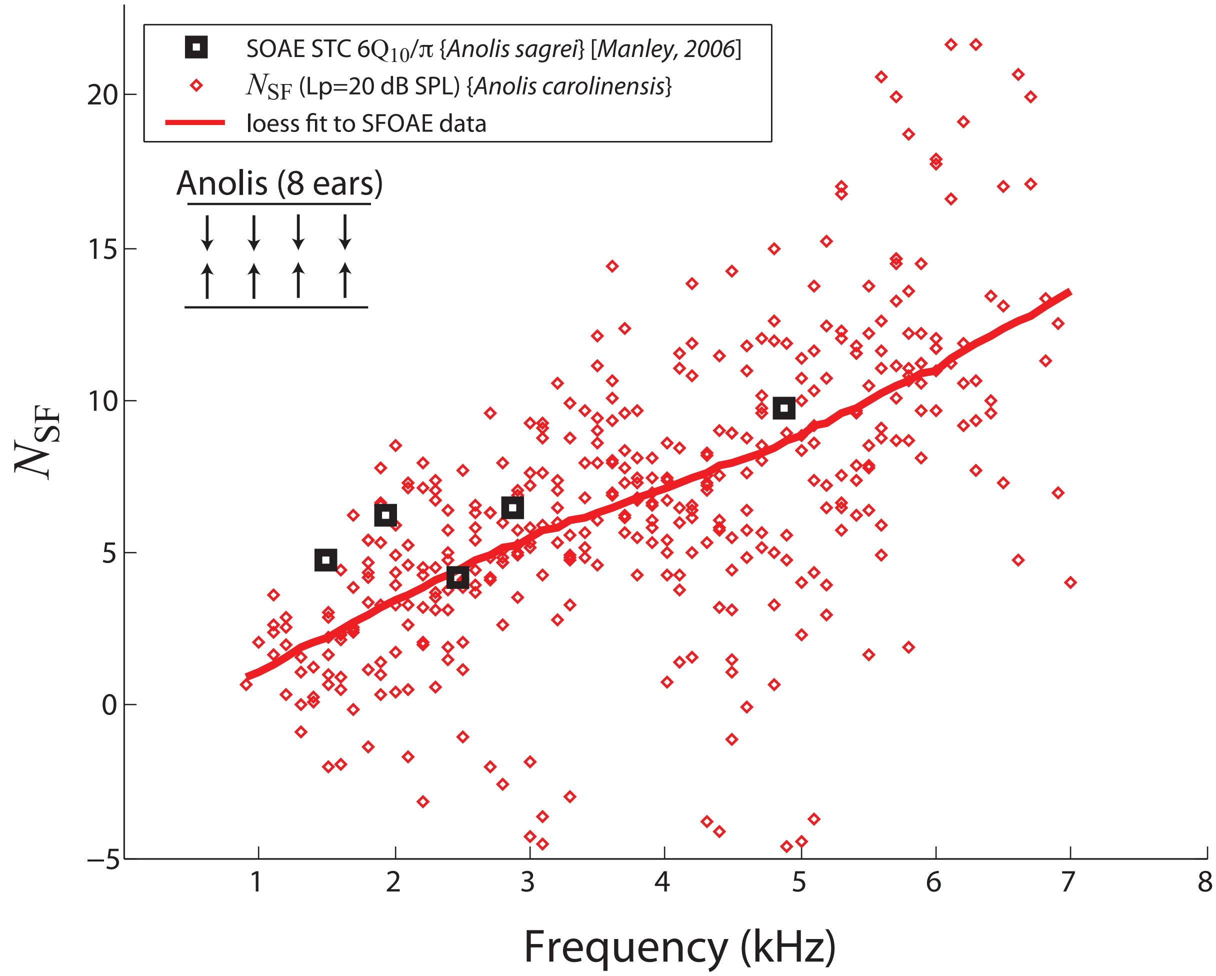
Shown in Supp. Fig.2 is two different otoacoustic estimates of tuning from a non–TM genus (*Anolis*). The SOAE data come from a different study (3), as well as from a different species (but the same genus).

Discussion

A novel feature indicated here is a degree of correlation between SOAEs and low–level SFOAEs in lizards (Supp. Fig.1), suggestive that the underlying emission generation mechanisms for the



Supplemental Figure 1: Correlation between SOAE and low–level SFOAEs. For each individual, SOAEs (with and without a 40 dB SPL suppressor tone, indicated by the grey and black traces respectively) are also shown with the SFOAE magnitude and phase (ϕ_{OAE}) evoked using a 20 dB SPL tone. SOAE measurements were stable before and after the SFOAE measurements. Error bars for SFOAEs denote the standard error of the mean. *Anolis* and *Gerrhosaurus* data shown here were at body temperatures ∼26–27° C (where SOAE activity in these species was observed to be more robust) while the *Aspidoscelis* data were obtained at ∼32–33° C (heating pad on). Spectral artifacts due to external acoustic noise (e.g., 4.2 kHz peak for *Aspidoscelis*) are distinguished from SOAEs by virtue of lack of temperature–dependence and suppressibility.



Supplemental Figure 2: Correlation between tuning measures derived from SOAEs and low–level SFOAEs from two different species of *Anolis*. The SOAE data is from *Anolis sagrei* (3). From that study, which reported $Q_{10\mathrm{dB}}$ derived from SOAE suppression tuning curves, the plotted points (black squares) indicate the corresponding tuning estimate relative to that of the SFOAE via the model prediction (5) as indicated in the figure legend (Eqn.2 in main document).

two OAE types are related (if not identical). Such a correlation between the two is remarkable in that it appears consistent with predictions from a standing model for the mammalian SOAEs (6), despite the absence of BM traveling waves in the lizard (7). Additional study is warranted of these interrelations between lizard SOAEs and SFOAEs to elucidate how generation mechanisms might be similar and different between mammals and non–mammals (e.g., (5)). Clearly the presence of a TM has some influence on SOAEs, (e.g., focusing baseline activity into distinct peaks), but little overall effect upon whether OAEs are ultimately present or not. As previously pointed out (3), this observation speaks to the robustness of the underlying generation mechanisms (e.g., hair cell bundle motility (8)).

It is worth noting that, as indicated previously (9), the external tone is probably not really suppressing per se. Clearly in the spectra there is a frequency–dependent region about the external tone where the SOAE amplitudes are reduced. However, the underlying generators are unlikely to simply stop oscillating (i.e., they are suppressed), but more likely shift their frequency to match that of the external tone (i.e., they are entrained). Thus, to some extent, SOAE *suppression* is likely a bit of a misnomer when specifically considering the dynamics of the underlying generators.

As shown in Supp. Fig.2, tuning estimates from both SOAEs (3) and SFOAEs appear to match up well via the model predictions (5). However, further study is warranted given that the comparison is made across two different species (which may manifest differences in peripheral tuning) and because of the relatively limited SOAE data shown here. Preliminarily, these data demonstrate that SOAE and SFOAE measures may reasonably be expected to yield similar estimates of tuning sharpness.

II – Additional SFOAE Data

Similar in nature to Figs.1 and 2, data from several other species is included for comparison in Supp. Fig.3.

References

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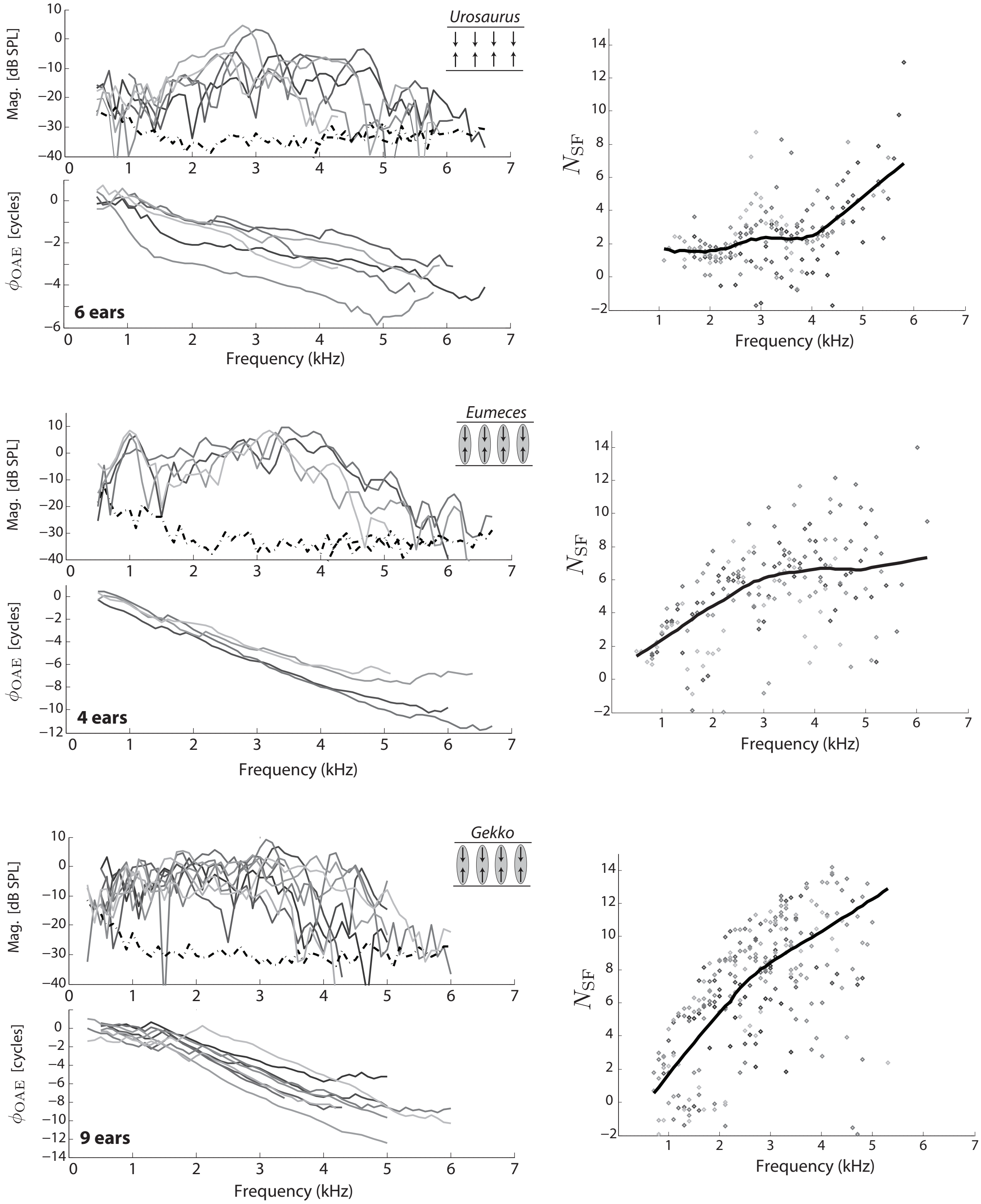

Supplemental Figure 3: Comparison across individuals similar for three additional species: *Urosaurus ornatus* (top), *Eumeces schneideri* (middle), and *Gekko gecko* (bottom). Same parameters as described in the captions for Figs.1 and 2.