Probing Otoacoustic Emissions in the Budgerigar (*Melopsittacus undulatus*)

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Abstract

Given the complex nature of their vocalizations, birds have become an increasingly attractive model for studying neurophysiological mechanisms underlying the processing of complex sounds. However, much is still unknown about the function of the peripheral auditory system in birds, and there are significant physiological differences between birds and mammals (e.g., hair cell distribution, lower prestin density, structure of the tectorial membrane). The present study explores this issue by characterizing several types of otoacoustic emissions (OAE) in the budgerigar (Melopsittacus undulatus).

Background

- Budgerigar's (parakeets) thresholds for detection of changes in complex sounds, such as amplitude modulations, are comparable to those of human listeners (Dooling et al. 1985, 1987; Dent et al., 2002; Carney et al., ARO 2013). A better characterization of their peripheral auditory system would contribute to modeling efforts that relate physiology to perception.
- Parakeet auditory thresholds are relatively low, but chiefly confined to lower frequencies (Fig.1). The inner ear morphology of birds is quite different relative to mammals (Fig.2) and is known in quantitative detail for the parakeet (Manley et al. 1993): short papilla (~2.5 mm) with ~5400 hair cells. Furthermore, there is evidence for a traveling wave at work in the bird ear (Gummer et al. 1987), however hair cell somatic motility appears absent (He et al. 2003; Koppl et al. 2004). The middle ear consists of a single ossicle (columella).
- Otoacoustic emissions (OAEs) provide a non-invasive strategy for characterizing the tuning of the auditory periphery in both mammals (Shera et al. 2002, 2010; Joris et al. 2011) and non-mammals (Bergevin & Shera, 2010). In addition, detailed descriptions of OAEs in various species provides information relevant to the mechanisms underlying generation of emissions (Bergevin et al. 2008).
- In the current study, we measured stimulus frequency emissions (SFOAEs) and distortion product emissions (DPOAEs) in the anesthetized budgerigar with several questions in mind:

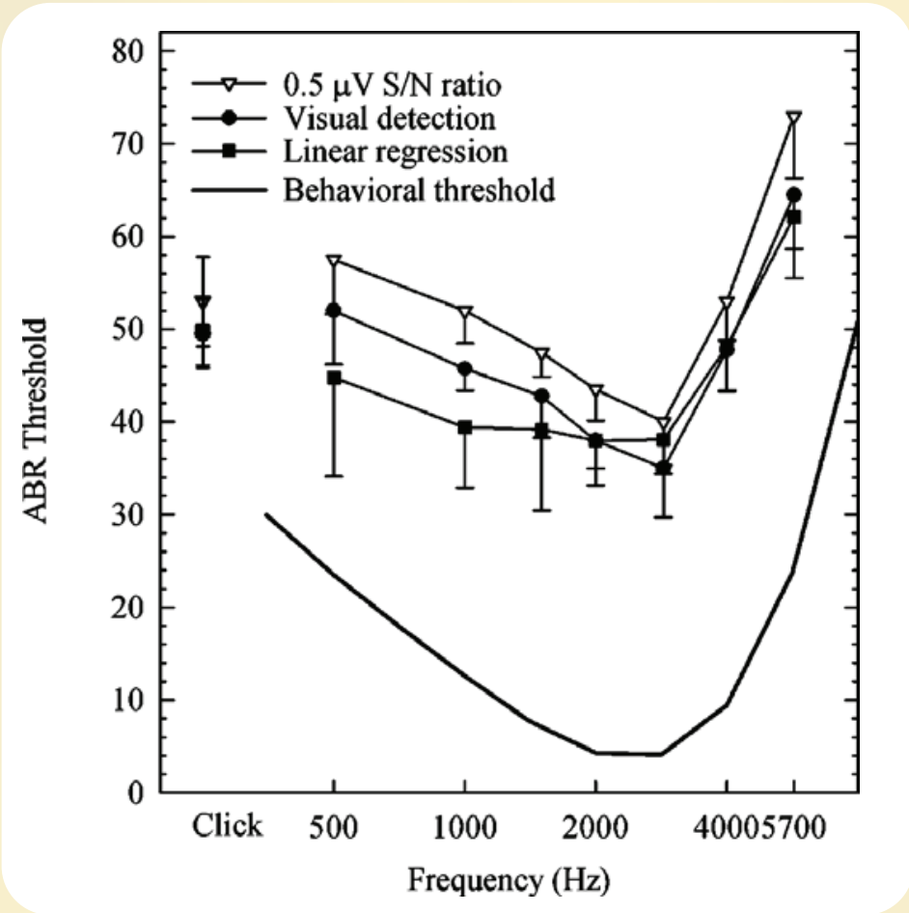

FIGURE 1 - Parakeet audiogram. Data from Brittain-Powell et al. (2002)

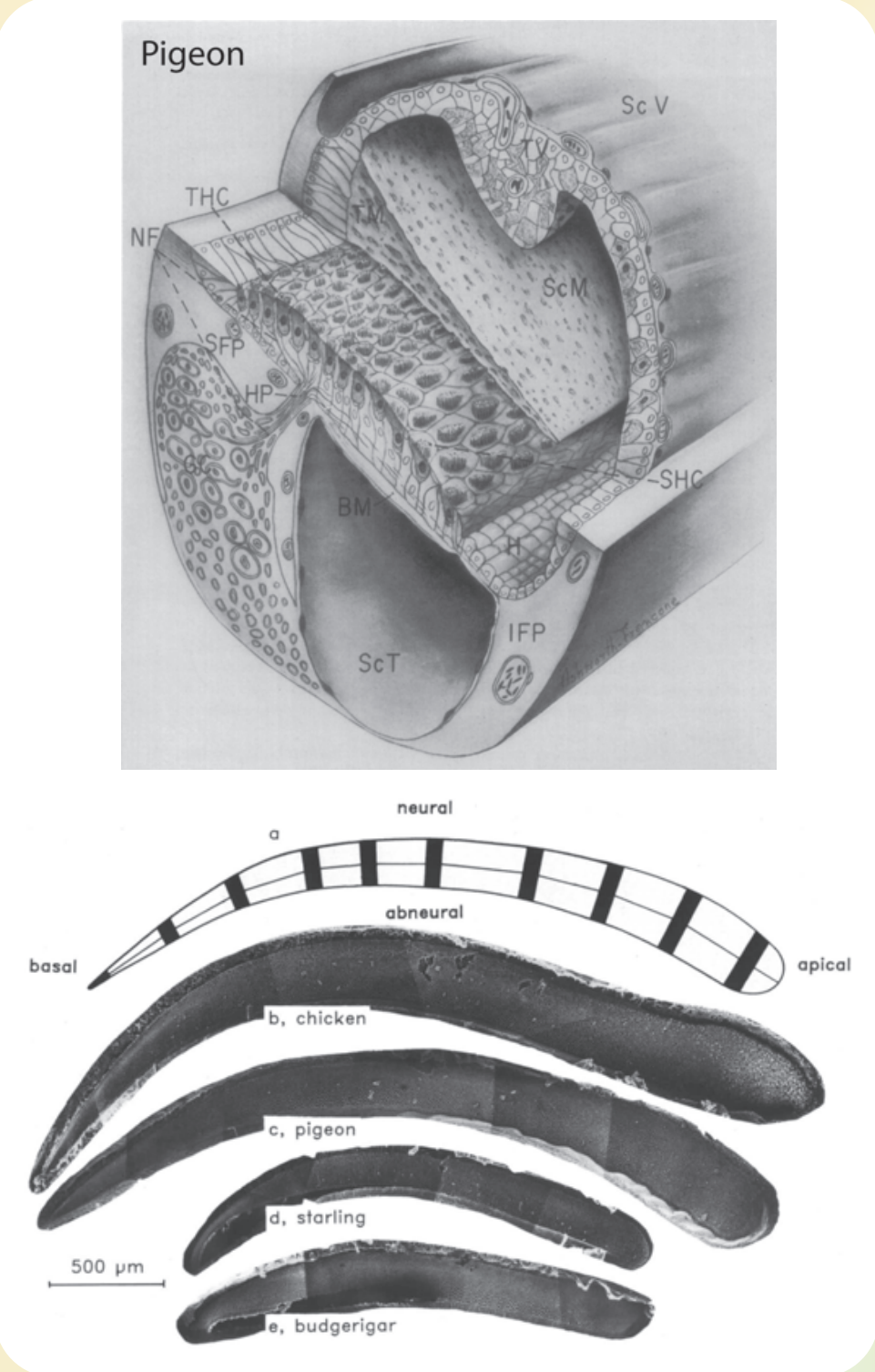

FIGURE 2 - Morphology of the avian basilar papilla. Figures from Takasaka & Smith (1971) and Gleich & Manley (1988).

1. Given that most modeling efforts on OAE generation focus on the mammalian ear, what can we learn by considering ears where significant morphological differences exist (Fig.2)? For example, how do differences in the nature of the traveling wave affect OAE generation?

2. Given that knowledge of the bandwidth of peripheral filters is essential for studies of the neural coding and processing of complex sounds in the auditory periphery, can we use SFOAEs to characterize sharpness of tuning in the parakeet? Previous studies have suggested relatively sharp tuning in the parakeet (Dooling & Searcy, 1985), though a clear morphological basis for such (i.e., a fovea) is absent (Manley et al. 1993). Will SFOAE properties show unique properties in the parakeet?

Methods

- Measurements were performed at the University of Rochester. Data was collected from 8 ears of 8 different birds. The birds were of both sexes and ranged in age from 3.5-10 months.
- Birds were lightly anesthetized using either isoflurane (gas) or ketamine and xylazine or ketamine and dexmedetomidine (injectable, sub-cutaneous). Feathers about the external auditory meatus were gently trimmed and the OAE probe was then sealed to the head using grease. Recording sessions typically lasted 1–3 hours. At the completion of each recording session, animals were then returned to the holding cage and monitored until fully recovered from anesthesia.
- OAEs were measured from ears using either an Etymotic ER-10C or ER-10A/ER2 probe system. Stimuli were generated and recorded digitally using Lynx TWO-A sound cards controlled by custom software.
- SFOAE were obtained using a two-tone suppression paradigm (Shera & Guinan, 1999). Suppressor parameters: fs=fp+40 Hz, Ls=Lp+15 dB. Most SFOAE data were collected using a probe level (Lp) of 40 dB SPL. The DPOAE paradigm used a fixed f2/f1 ratio (discrete tones, sweeping f1 from ~0.5-6 kHz) and equal level primaries (L1=L2).
- SFOAE phase-gradient delays were computed via centered-differences (Shera & Guinan, 2003). Trends (e.g., thick lines in Figs.4, 7 & 8) were computed using locally-weighted regression (loess). Confidence intervals (95 %) for the trends (shaded regions) were computed using bootstrap resampling.

Results

- Evoked emissions were measurable in all birds examined. Emission magnitudes were relatively small when compared to other vertebrates, and similar to chicken (Bergevin et al. 2008). No spontaneous emissions were detected.
- Overall, emission magnitudes matched qualitatively to behavioral thresholds, being confined to the most sensitive portions of the audiogram (Figs.1, 3 & 5). Over the majority of the frequency range tested, SFOAE phase-gradient delays were similar to or smaller than those of chicken (Fig.4).
- Variability across individuals was apparent and possibly due to differences in type of anesthesia and/or depth (Kettembiel et al. 1995). However, this was not explored in detail in the current study.
- Some evidence for dependence upon probe level was observed in the SFOAE phase-gradient delays (typically larger for Lp<40 dB SPL), but this was not explored in detail in the current study nor is it readily apparent in the DPOAE data for those levels tested (Fig.5).
- DPOAE properties (Figs. 5 & 6) are summarized as follows:
  - With f2/f1 = 1.25, Both 2f1–f2 and 2f2–f1 appeared around the most sensitive region of the audiogram, then expanded outwards in frequency with increasing stimulus levels (Fig.5) - similar to that of DPOAEs in mammals
  - 2f1–f2 increased compressively with primary sound pressure level while 2f2–f1 grew almost linearly above f2~3 kHz (Fig.5); non-monotonic growth is apparent
  - 2f1–f2 phase was nearly constant with frequency for larger f2/f1 ratios - similar to that of mammals
  - 2f2–f1 phase depended strongly upon frequency for f2 below ~3 kHz, while that dependence decreased at higher f2 - similar to that of mammals
  - Between f2/f1 ratios of 1.25 and 1.45, characteristics of 2f1–f2 and 2f2–f1 changed little – unlike mammals, parakeet DPOAEs appear less ratio-sensitive
  - At the smallest (1.02) and largest (1.65) ratios, DPOAE magnitudes were greatly reduced

Discussion

- We use SFOAEs here to estimate the sharpness of tuning of the parakeet's peripheral auditory filters by use of *tuning ratios*, defined as Qerb/Nsf (Shera et al. 2010). To do this, we first estimate tuning ratios for the chicken (Fig.7, inset), then apply the chicken ratios to estimate Qerb for the parakeet (Fig.8).
- However, several considerations need to be factored in. First, there is significant variability across (and within) studies characterizing tuning in chicken auditory nerve fibers (ANFs). Second, it is unclear whether it is reasonable that these ratios can be directly applied to the parakeet (e.g., an *apical-basal transition* was not considered here, though is suggested by the 2f2-f1 phase in Fig.6). Nonetheless, given the similarity between SFOAE phase-gradient delays between chicken and parakeet (Fig.4), our results suggest that peripheral sharpness of tuning is similar between these two species. Relatively minimal DPOAE *fine structure* (Figs.5 & 6) and weak ratio-dependence (Fig.6) are consistent with such a conclusion.
- Dooling & Searcy (1988) indicated that peripheral tuning in parakeet is similar, if not sharper, than that of humans. Several lines appear inconsistent with such a conclusion (e.g., Shera et al. 2002; Joris et al. 2011). It still remains to be seen as to how parakeet's remarkable psychophysical abilities can be explained, but morphological aspects of the papilla must factor in (Fig.2; Manley et al. 1993; Gleich & Langemann, 2011).
- DPOAE phase characteristics were qualitatively similar to those of mammals (e.g., flat 2f1-f2, steep 2f2-f1), consistent with chicken and even gecko (Bergevin et al. 2008) but not other lizards. Given the work of Gummer et al. (1987), this observation motivates further study as to what role (if any) traveling waves are playing in the non-mammalian ear.
- Future work should entail a more detailed account of the dependence upon stimulus level, as well as better characterizing the effects of anesthesia (e.g., Kettembeil et al. 1995). Measuring behavioral estimates of tuning (via forward-masking) can also be compared to OAE-based estimates of tuning. Lastly, middle-ear measurements in parakeet can help improve our understanding of how the middle ear influences both OAEs and the audiogram and how emissions make their way out of the inner ear.

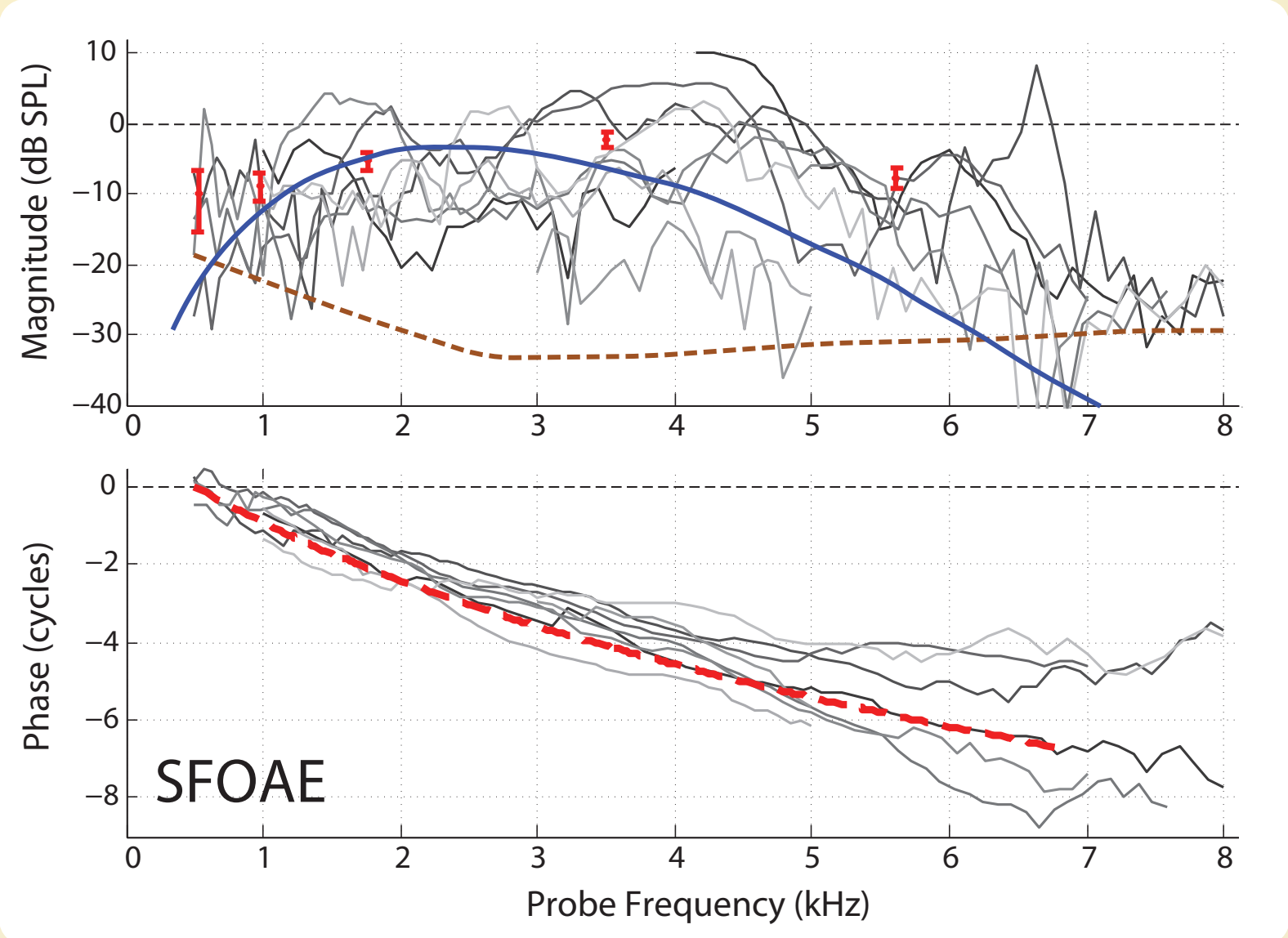

FIGURE 3 - Parakeet SFOAEs (magnitude and phase) measured at Lp= 40 dB SPL. Data are from 8 ears from 8 different birds. ***Dashed brown curve*** shows the approximate magnitude noise floor. ***Red points*** indicate average magnitudes in octave wide bins (including the SEM). Also included in the magnitude is a relative indication of parakeet behavior thresholds (Brittain-Powell et al. 2002; ***solid blue curve*** shows threshold multiplied by -1). Error bars have been omitted from individual curves for clarity. The ***dashed red curve*** in the phase indicates the integrated phase-gradient trend (see Fig.4). Some phase curves have been offset vertically (by an integral number of cycles) for clarity.

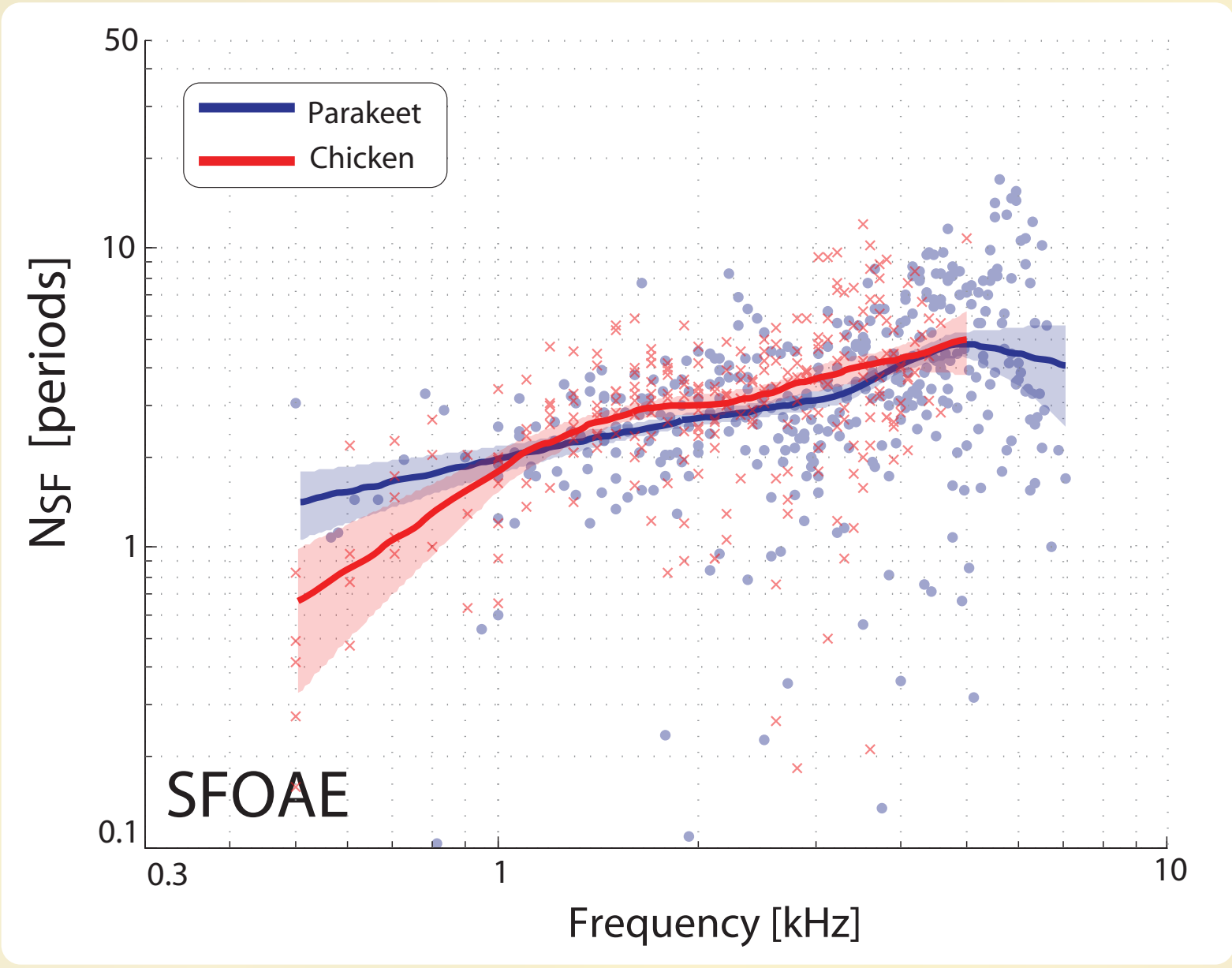

FIGURE 4 - Cross-species comparison of SFOAE phase-gradient delays in stimulus periods (Nsf). Data points and trends are shown for both parakeet (blue) and chicken (red). Chicken data are from Bergevin et al. 2008 and were measured using the same paradigms and stimulus level (Lp = 40 dB SPL). These delays are slightly smaller than those for mammals such as cat and guinea pig (Shera & Guinan, 2003) and comparable to those of gecko (Bergevin et al. 2008).

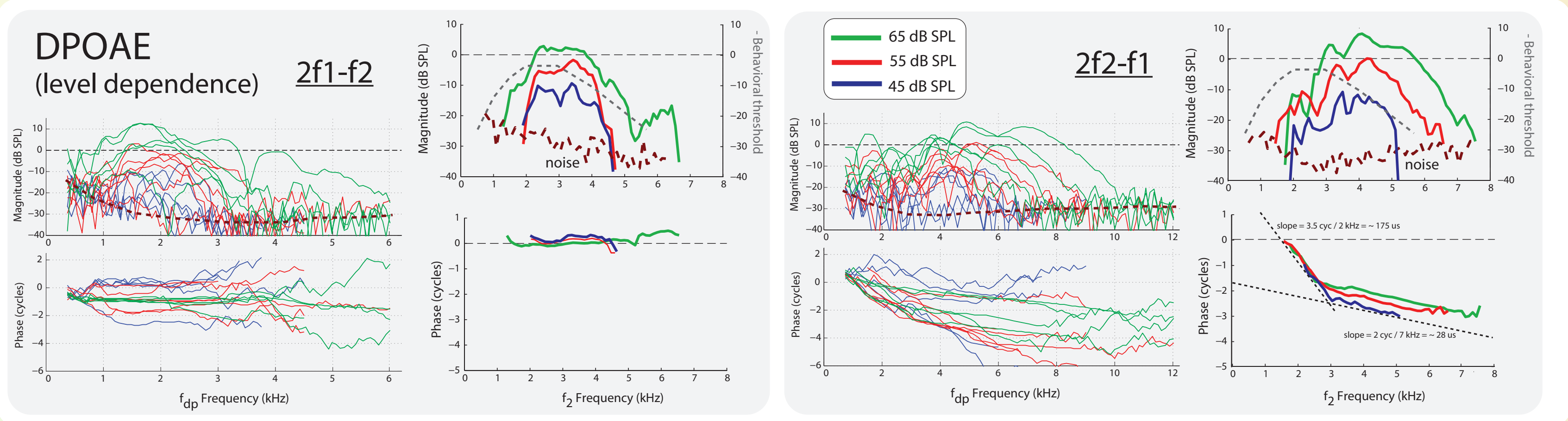

FIGURE 5 - Parakeet DPOAE level dependence. Data are shown for both a representative individual (right) and compiled data across all ears (left). Left plots show 2f1-f2 and right plots 2f2-f1. For all data, primary levels were equal (**L1=L2**) and the primary ratio was fixed at **f2/f1=1.25**. Brown dashed curve shows approximate noise floor. Note different abscissae.

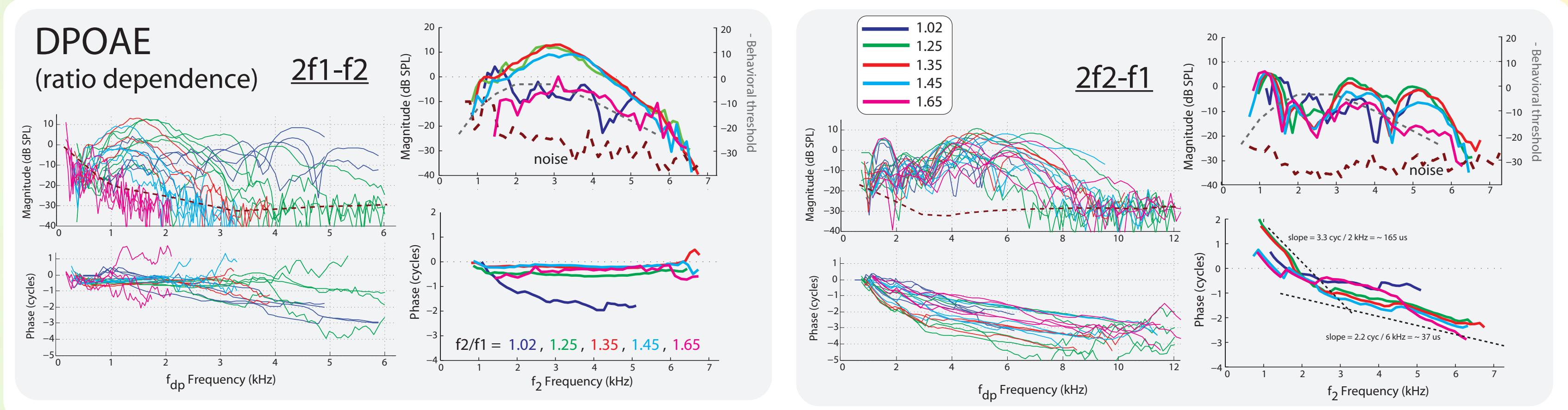

FIGURE 6 - Parakeet DPOAE primary ratio dependence. Data are shown for both a representative individual (right) and compiled data across all ears (left). Left plots show 2f1-f2 and right plots 2f2-f1. For all data, primary levels were equal (**L1=L2= 65 dB SPL**) and the primary ratio was fixed as the tones were swept. Brown dashed curve shows approximate noise floor. Note different abscissae.

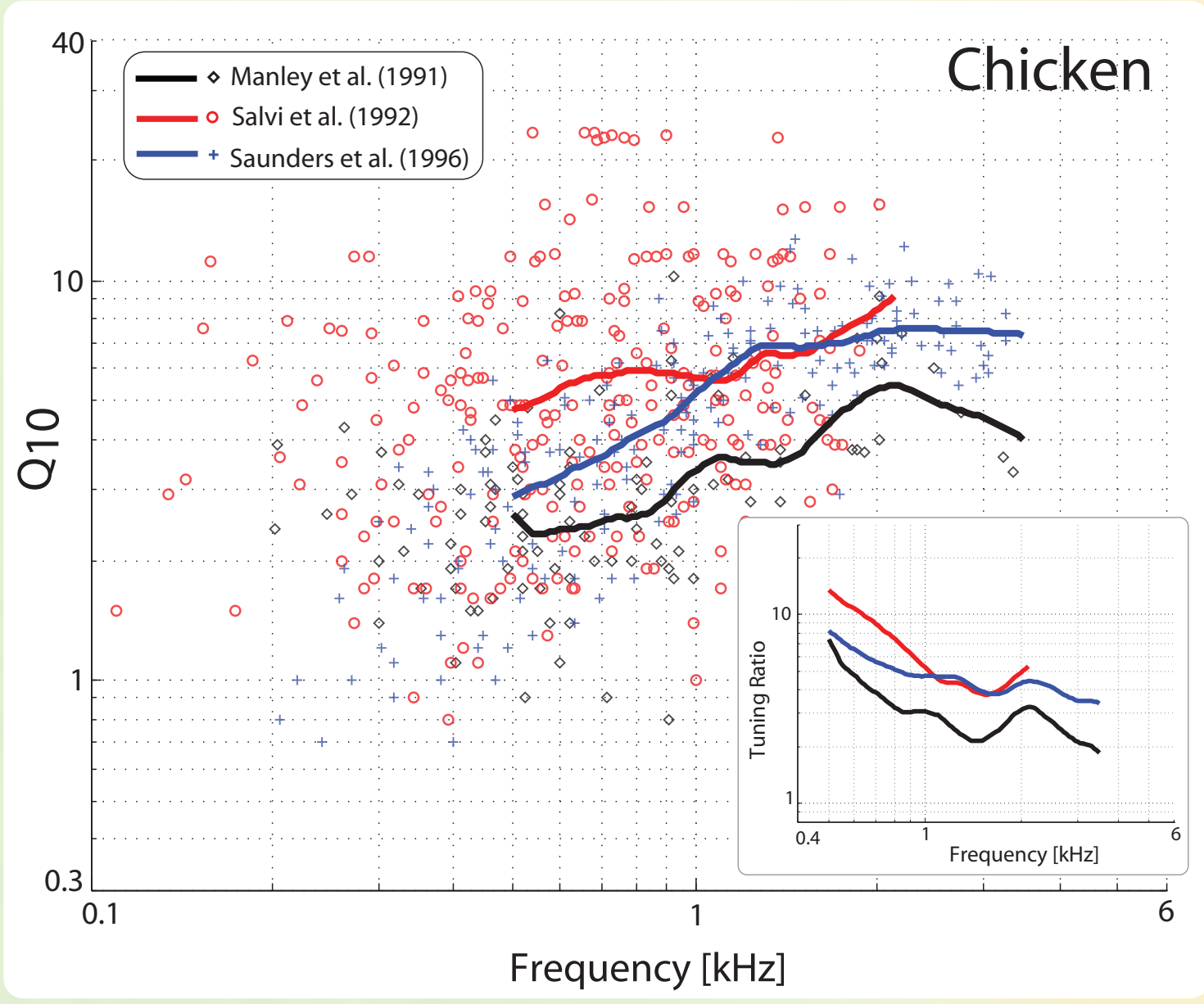

FIGURE 7 - Chicken ANF quality factors and estimates of the 'tuning ratio'. Auditory nerve data taken from studies noted in legend. Loess fits are shown for regions where SFOAE were measurable and sufficient ANF data existed. Estimates of the chicken 'tuning ratio' (Qerb/Nsf; inset) used Nsf values from Fig.4 (Lp=40 dB SPL). It was assumed that Qerb ~1.75 • Q10 (Shera & Guinan, 2003).

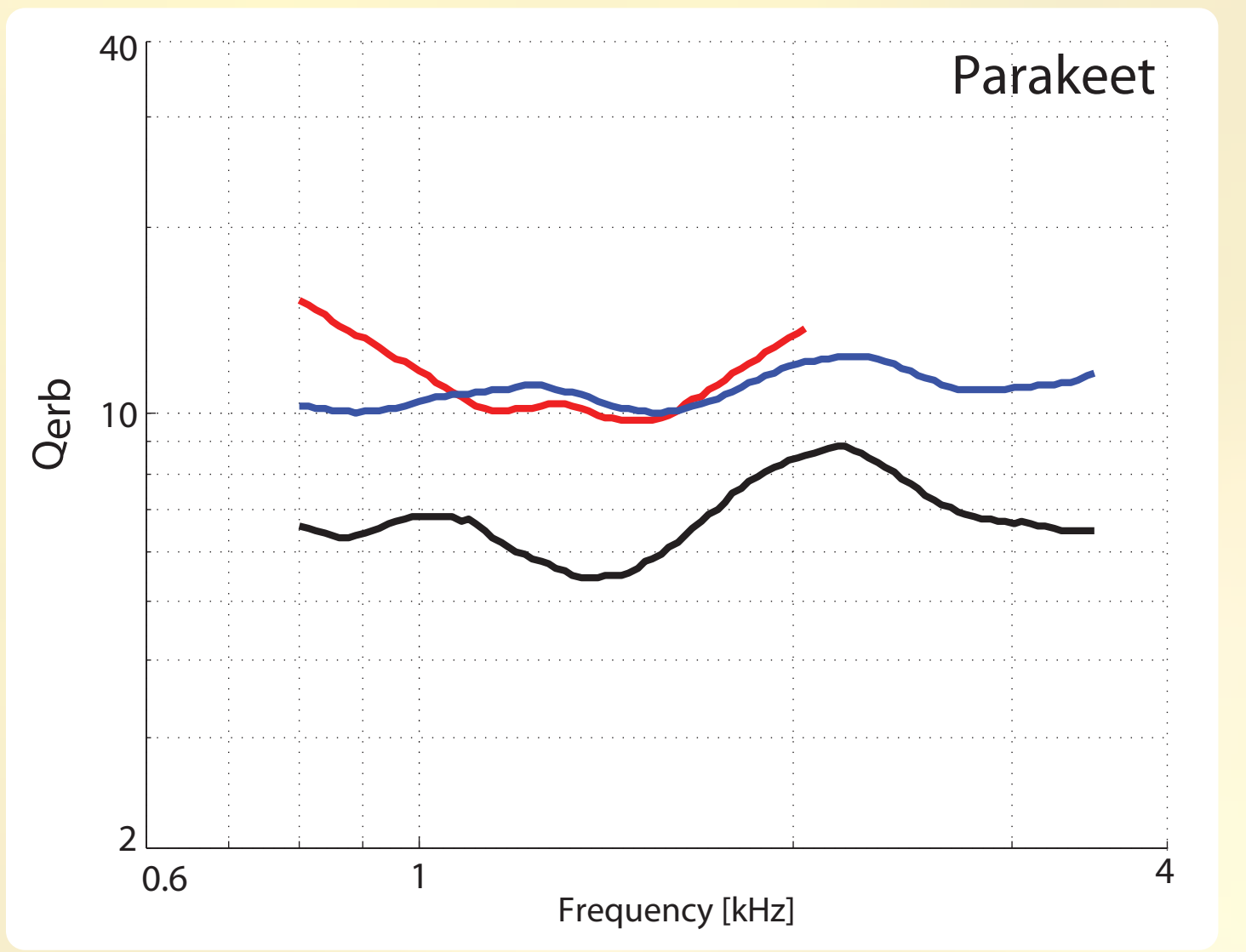

FIGURE 8 - Estimate of parakeet Qerb. Rough estimates based upon the assumption that the parakeet tuning ratio is similar to that of the chicken. Estimates are confined to regions where there was both sufficient SFOAE and (chicken) ANF data available. No uncertainty is shown, but it is large given the scatter in the ANF data. Note that unlike Shera et al. (2010), no correction was made for an 'apical-basal' transition.

References

- Bergevin, C., Freeman, DM., Saunders, JC. & Shera, CA. Otoacoustic emissions in humans, birds, lizards, and frogs: evidence for multiple generation mechanisms. J. Comp. Physiol. A. 194:665-683 (2008)
- Bergevin, C. & Shera, CA. Coherent reflection without traveling waves: On the origin of long-latency otoacoustic emissions in lizards. J. Acoust. Soc. Am. 127:2398-2409 (2010)
- Brittain-Powell, EF., Dooling, RJ. & Gleich, O. Auditory brainstem responses in adult budgerigars (Melopsittacus undulatus). J. Acoust. Soc. Am. 112:999-1008 (2002)
- Carney, LH., Ketterer, AD., Abrams, KS., Koch, KJ., Schwarz, DM. & Idrobo, F. Detection thresholds for amplitude modulations of tones in budgerigar. ARO 2013 abstract
- Dent, ML., Klump, GM. & Schwenzfeier, C. Temporal modulation transfer functions in the barn owl (Tyto alba). J. Comp. Physiol. A. 187(12):937-943 (2002)
- Dooling, RJ. & Searcy MH. Nonsimultaneous auditory masking in the budgerigar (Melopsittacus undulatus). J. Comp. Psychol.. 99(2):226-230 (1985)
- Dooling, RJ.Brown, SD., Park, TJ.,Okanoya, K. & Soli, SD. Perceptual Organization of Acoustic Stimuli by Budgerigars (Melopsittacus-Undulatus): 1. Pure-Tones. J. Comp. Psych. 101(2)-139-149 (1987)
- Gleich, O. & Manley, GA.. Quantitative morphological analysis of the sensory epithelium of the starling and pigeon basilar papilla. Hear. Res. 34:69-85 (1988)
- Gleich, O. & Langemann, U. Auditory capabilities of birds in relation to the structural diversity of the basilar papilla. Hear. Res. 273:80-88 (2011)
- Gummer, AW., Smolders, JW. & Klinke, R. Basilar membrane motion in the pigeon measured with the Mössbauer technique. Hear. Res. 29:63-92 (1987)
- He, DZZ. et al. Chick hair cells do not exhibit voltage-dependent somatic motility. J. Physiol. 546:511-520 (2003)
- Joris, PX., Bergevin, C., Kalluri, R., Mc Laughlin M., Michelet, P., van der Heijden, M. & Shera, CA. Frequency selectivity in Old World monkeys corroborates sharp cochlear tuning in humans. PNAS 108(42): 17516-17520 (2011)
- Kettembeil, S., Manley, GA. & Siegl, E. Distortion-product otoacoustic emissions and their anaesthesia sensitivity in the European Starling and the chicken. Hear. Res. 86:47-62 (1995)
- Koppl, C., Forge, A. & Manley, GA. Low density of membrane particles in auditory hair cells of lizards and birds suggests an absence of somatic motility. J. Com. Neurol. 479:149-155 (2004)
- Manley, GA., Kaiser, A., Brix, J. & Gleich, O. Activity patterns of primary auditory-nerve fibres in chickens: Development of fundamental properties. Hear. Res. 57:1-15 (1991)
- Manley, GA., Schwabedissen, G. & Gleich, O. Morphology of the Basilar Papilla of the Budgerigar, Melopsittacus-Undulatus. J. Morphol. 218:153-165 (1993)
- Salvi, RJ., Saunders, SS., Powers, NL. & Boettcher, FA. Discharge patterns of cochlear ganglion neurons in the chicken. J. Comp. Physiol. A 170:227-241 (1992)
- Saunders, JC., Doan, DE., Poje, CP. & Fisher, KA. Cochlear nerve activity after intense sound exposure in neonatal chicks. J. Neurophysiol. 76(2):770-787 (1996)
- Shera, CA. & Guinan, JJ. Evoked otoacoustic emissions arise by two fundamentally different mechanisms: A taxonomy for mammalian OAEs. J. Acoust. Soc. Am. 105(2):782-798 (1999)
- Shera, CA., Guinan, JJ. & Oxenham, AJ. Revised estimates of human cochlear tuning from otoacoustic and behavioral measurements. Proc. Natl. Acad. Sci. USA 99:3318-3323 (2002)
- Shera, CA. & Guinan, JJ. Stimulus-frequency-emission group delay: A test of coherent reflection filtering and a window on cochlear tuning. J. Acoust. Soc. Am. 113:2762-2772 (2003)
- Shera, CA., Guinan, JJ. & Oxenham, AJ. Otoacoustic Estimation of Cochlear Tuning: Validation in the Chinchilla. J. Assoc. Res. Otolaryngol. 11:343-365 (2010)
- Takasaka, T. & Smith, CA. The structure and innervation of the pigeon's basilar papilla. J. Ultrastructure Res. 35:20-65 (1971)

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