

GAMBA et al.: Individual vocal differences in group-living lemurs

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Modeling individual vocal differences in group-living lemurs using vocal tract morphology

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Abstract

Vocal individuality is widespread in social animals. Individual variation in vocalizations is a prerequisite for discriminating among conspecifics and may have facilitated the evolution of large complex societies. Ring-tailed lemurs *Lemur catta* live in relatively large social groups, have conspicuous vocal repertoires, and their species-specific utterances can be interpreted in light of source-filter theory of vocal production. Indeed, their utterances allow individual discrimination and even recognition thanks to the resonance frequencies of the vocal tract. The purpose of this study is to determine which distinctive vocal features can be derived from the morphology of the upper vocal tract. To accomplish this, we built computational models derived from anatomical measurements collected on lemur cadavers and compared the results with the spectrographic output of vocalisations recorded from *ex-situ* live individuals. Our results demonstrate that the morphological variation of the ring-tailed lemur vocal tract explains individual distinctiveness of their species-specific utterances. We also provide further evidence that vocal tract modeling is a powerful tool for studying the vocal output of non-human primates.

Key words: indexical cues, *Lemur catta*, formants, nasal resonance, ring-tailed lemur.

The capacity to emit and recognize individually distinctive vocalizations is considered to be a crucial prerequisite for the evolution of complex animal and human societies (Tibbetts and Dale 2007). Thus, in recent years, individual recognition mediated by vocal signals has received increasing attention due to the importance of understanding the cognitive and physiological processes underlying these abilities (Pollard and Blumstein 2011). Nonhuman primates (hereafter primates), because of the complexity of their social interactions and the large groups some of them live in, have been targeted as model systems for investigating individual recognition (Rendall et al. 1996; 1998).

The morphology of speech organs is a critical source of inter-speaker variation in the human voice (Titze 1994). In line with the source-filter theory of speech production (Fant 1971; Flanagan 1972), the length of the vocal tract affects the resonance of the signal produced by the larynx (Dellwo et al. 2007). Moreover, the vast amount of information yielded by phonetic studies has shown that vocal tract resonance is crucial in encoding individually distinctive features of the human voice (Daqrouq and Tutunji 2015). Vocal production in nonhuman animals can similarly be interpreted using the source-filter theory (e.g. Taylor and Reby 2010; Gamba et al. 2011; Favaro et al. 2015).

The salience of vocal tract filtering in primate communication has been investigated in several species. Historically, Rendall and colleagues (1998) found that the coos of rhesus monkeys *Macaca mulatta* could be classified by caller identity with high correct classification rates using vocal tract resonances. Evidence that formants play a major role in primate communication can be derived from studies of call production in baboons (Rendall et al. 2005) and macaques

(Fitch 1997), and from the spontaneous abilities of macaques to perceive formants (Fitch and Fritz 2006). However, studies of strepsirhine primates are still limited. Gamba and colleagues (2012a) found that the formants comprising the low-pitched grunts of red-bellied lemurs *Eulemur rubriventer* were markedly more individually specific when compared to larynx-related aspects and were potentially informative as to individual identity. Recently, computational models have been used to simulate resonance in lemur vocal tracts (Gamba et al. 2012b). In particular, the acoustic output of models built using anatomical measurements of the vocal tract showed the same species-specific features observed in wild animal calls, providing evidence that these vocal-tract modeling techniques provide powerful and reliable tools for studying the vocal output of primates (Gamba et al. 2012b).

The fact that vocal tract morphology plays a key role in explaining interspecific and inter-individual acoustic variation invites questions about the degree of inter-individual difference in the vocal apparatus, and the effects that changes in a particular part of the vocal tract may have on the acoustic properties of vocalizations. Because individuality information could be encoded in various acoustic parameters of the vocal signal (e.g. Gustison et al. 2012; Scheumann et al. 2012), we chose to investigate whether variation in vocal tract morphology could yield insights into the sources of variation in resonance. Recent research has demonstrated that nasal sounds play a significant role in the vocal communication of many species (Lapshina et al. 2012; Sibiryakova et al. 2015). The frequencies of short vocalisations released via the nasal regions are also particularly interesting to study the impact of morphology on vocal output, in that these calls are produced without articulation of the vocal apparatus.

Vocal tract modelling (Zhang and Espy-Wilson 2004; Pruthi et al. 2007; Zhou et al. 2008) has proven reliable in describing individual variation in vocal tract morphology and its impact on the formants. Accordingly, this approach holds considerable promise in providing insights into vocal production mechanisms of non-human animals (Gamba and Giacoma 2006; Gamba et al. 2012b).

The ring-tailed lemur *Lemur catta* is a gregarious strepsirhine with a conspicuous vocal repertoire that includes oral and nasal calls (Macedonia 1993). This species lives in large multi-male/multi-female groups and their social interactions involve a complex balance between cooperation and competition (Palagi and Norscia 2014) where individual recognition plays a crucial role (Macedonia 1986; Oda 1996, 1999, 2002; Kulahci et al. 2014). The aim of our study was twofold. First, we aimed to establish whether inter-individual variation in the length and shape of the nasal tract was demonstrable in ring-tailed lemurs. Previous research has indicated that differences in vocal tract length are common among conspecifics, even of the same sex. Fitch (1997) found that nasal tract length in male rhesus macaques could vary between 6.74 and 10.89 cm ($N = 20$). If such variation was present, we predicted that it should be reflected in the acoustic signals emitted by the different tracts. Our second goal was an investigation of the potential of formants to provide clues for individual recognition within small groups of conspecifics (Fitch 2000; Ey et al. 2007). We asked whether individuality in lemurs is encoded exclusively through changes in the length of the nasal tract (which changes in accordance with body size), or whether the shape of the tract may play a role in individual differences in vocalizations. It has been demonstrated that species-specific formant patterns in lemurs (*Eulemur* spp.; Gamba et al. 2012b) are influenced by both shape and length of the vocal tract; hence, we predicted that changes in both of these aspects have the potential to encode individual recognition. Finally, we measured formants of live lemur calls to understand whether formant variation was present in their nasal utterances.

Materials and Methods

Casting and modelling of the vocal tract

We made silicone casts of the vocal tracts of the cadavers of three adult male ring-tailed lemurs *Lemur catta* at the Parc Botanique et Zoologique Tsimbazaza (Antananarivo, Madagascar) that had been frozen shortly after death. Vocal tract casting is an effective means of reproducing in detail the structures of cavities (Riede et al. 2005), and can be performed in Madagascar (Gamba and Giacoma 2006) where no medical imaging is available. We cut the trachea 3–4 cm below the hyoid bone and injected high-speed catalysed silicone rubber (Blue Star Silicones Rhodorsil® RTV) before closing the tracheal tube using surgical clamps. This procedure gave us a flexible cast that we extracted as a single piece through the mouth.

We then measured the silicone cast to build the computational model. Vocal tract modeling (Zhang and Espy-Wilson 2004; Pruthi et al. 2007; Zhou et al. 2008) is a technique for characterizing individual variation in vocal tract morphology and its impact on the formants produced, and has previously provided important insights into animal phonation (Gamba and Giacoma 2006; Gamba et al. 2012b). All length and volume measurements of the casts were taken with Mitutoyo digital calipers (accurate to 0.01 mm) in 0.2 cm increments moving from the vocal folds toward the nostrils. To create the vocal tract models, we calculated cross-sectional area functions, from the glottal constriction to the nostrils, such that the virtual tract consisted of many elliptical segments of equal lengths but different cross-sectional areas (Figure 1). The vocal tract area function serves as the input for a customised version of VTAR (Vocal Tract Acoustic Response) modeling software (Zhang and Espy-Wilson 2004; Zhou et al. 2004) that calculates the acoustic response of each model. Because in primates the column of air passes the glottis and then resonates either solely in the oral or exclusively in the nasal tract (Fitch 2000, 2006), we did not consider resonance in the oral cavity for the lemur nasal grunts. We modelled the double intracranial nasal tract as a singular entity.

According to the aims of the study, we modified the vocal tract models to determine (a) whether lengthening of the vocal tract may affect individual recognition, and (b) to what extent changing length influenced formant structure. To estimate how length variation affected the formant pattern, we generated 180 models (60 per original VT model) where the length of all segments was decreased or increased by 0.5% over a range of $\pm 15\%$. This variation was calculated based on the variation in vocal tract length measured by Fitch (1997) in the Rhesus macaque *Macaca mulatta*. To investigate the effect of changes in cross-sectional areas, we used a different design to accommodate high inter-individual variability. The random module (a set of classes and functions from the *python3* language) was used to generate random cross-sectional areas within a range of variation of 31%, which was based on the variation of the vocal tract cross-sectional areas of the study specimens.

While running the computations, we extracted the length and volume of each model and the first five formants of the acoustic response (F_1 – F_5). All vocal tract geometries were also extracted and we divided the results into three groups. The first group (Anatomical) includes the original models of the three specimens. The second group (Length-variation) presents the results of the models in which vocal tract length was varied, and the third group (Shape-variation) shows the results of those models in which length was kept constant and the cross-sectional areas were modified.

Acoustic recording and analysis of the vocalizations

We recorded nasal click grunts of four male individuals (Figure 2) in captivity at the Parco Natura Viva (Bussolengo, Verona, Italy; July–August 2008) and Zoom Torino (Piscina, Torino, Italy; September–October 2013). All subjects were members of social groups ranging from 6 to 14 individuals. All individuals received a daily diet consisting mainly of vegetables and fruits with water supplied ad libitum and were maintained on a natural light/dark cycle. Vocalizations were recorded in outdoor enclosures using a SoundDevices 702 solid-state recorder and a Sennheiser K6/ME66 microphone (sampling rate of 44.1 kHz, digitized at 16 bits). Lemurs produced grunts in a range of contexts. We

recorded the click grunts while the animals were moving around the enclosure, during solitary exploration, or interactions with conspecifics or visitors.

We estimated formants using Linear Predictive Coding (LPC) in Praat (Boersma 2014; see also Gamba et al. 2015). LPC estimates the frequency values corresponding to the first five formants (Figure 2). In particular, we used the PRAAT function LPC [To Formant (burg)...] all along the signal frames and then extracted the minimum frequency of each formant. Depending on the acoustic characteristics of the click grunt, we used a window length of 0.025–0.055 s. We detected five to six formants in the frequency range 0–8,500 Hz while searching for 5.5 formants (for details, see Gamba and Giacoma 2007). Frequency spectra were then superimposed on the signal spectrogram to verify the goodness of the LPC analysis (Nadhrou et al. 2016). A custom-built script was used to automate file opening and saving of the measurements.

Statistical analyses

We used the *lme4* package (Bates et al. 2015) in R (R Core Team, 2015; version 3.2.0) to run the Linear Mixed Models (LMMs). We used a series of LMMs to investigate formant variation under different values of vocal tract length and volume. We included the log-transformed formant frequency (F_1 – F_5) as the response variable, length or volume as predictors and (individual) model ID as a random factor.

We also used LMMs to explore the relationship between formant variation and the shape of the vocal tract. To reduce the number of predictors, we performed a Principal Components Analysis (PCA) including all the cross-sectional areas of segments that had been increased or decreased randomly within a range of 31% (see Supplementary Materials), while the length was kept constant. The first two PCs represented the final predictors in the LMM models, while the log-transformed formant frequency (F_1 – F_5) was the response variable. Finally, the model ID was included as a random factor. For all models, we verified the assumptions that the residuals were normally distributed and homogeneous (see Gamba et al. 2016 for details) and avoided collinearity among variables (e.g. vocal tract volume was not tested together with PCs). We then tested the full model against a null model comprising only the random factors (see Estienne et al. 2017) and we calculated the P values using the R-function “*drop1*” (Barr et al. 2013).

To evaluate the potential for distinguishing individual identities, we calculated the variation of formants within and among models, and within and among individuals. Individual discrimination using vocalizations is possible when a character shows low intra-individual variation and high inter-individual variation in acoustic parameters. Thus, we calculated within- (CV_i) and between-model (CV_b) coefficients of variation (Sokal and Rohlf 1995) and the potential for individual identity coding (PIC) as the CV_b/CV_i ratio (Mathevon et al. 2003) for the models. We applied the same calculation to the click grunt acoustic parameters (Gamba et al. 2012b).

Results

Original vocal tract transfer-functions and acoustic response

The three specimens measured had nasal tract lengths of 9.2, 9.4 and 9.6 cm, which were divided into to 46, 47 and 48 segments respectively, rendered as a series of concatenated 0.2 cm tubes. The specimens showed clear differences in their cross-sectional area functions (Figure 3). We observed an average coefficient of variation of 31% (\pm 15%) in the

areas of the elliptical cross-sections that we used to discretize the vocal tracts. The actual measurements were used to generate a first set of computational vocal tract models, and the formants predicted from the models showed differences among individuals. The coefficient of variation of F1 was 18.81%, 6.10% for F2, 2.96% for F3, 6.17% for F4 and 0.83% for F5 (Table 1).

Effects of lengthening the vocal tract

The variation in vocal tract length recorded from our three lemur cadavers was 2.13%. It was considerably lower than the 15% reported by Fitch (1997) for his larger sample of 20 Rhesus macaques. Using the three models based on the original casts but standardized to the length of 9.6 cm, we altered the lengths within a range of 15%, generating models of an average length of 9.58 ± 0.84 cm and volumes of 3.52 ± 0.31 cm³, 3.21 ± 0.28 cm³, 2.38 ± 0.21 cm³. The effects produced on the vocal tract acoustics when the length of each segment was changed are shown in Figure 4, for each of the three models. Formants predicted from these computational models showed an average variation of $8.44 \pm 0.14\%$ within model across formants and $8.44 \pm 0.49\%$ between models across formants (Table 1 shows the variation for each model and formant). All formants scaled inversely with length, but not with volume, of the vocal tract model (Table 2). The PICs of formants ranged between 1.88 for F1 and 1.47 for F4, to 1.08 for F3, 1.05 for F2, and 1.01 for F5.

Effects of modifying the cross-sectional shape

The PCA of cross-sectional areas generated two components exceeding an eigenvalue = 1. PC1 and PC2 explained 43% and 15% of the total variance, respectively. All other PCs accounted for less than 2% variation and thus were not included in further analyses. In Figure 5 we report the variation in volume observed when the shape of the segments was modified, and the respective vocal tract acoustics in Table 1.

All formants were significantly influenced by PC1 and PC2, except F4 which showed significant values for PC1 only (Table 2). All formants also scaled inversely with the volume of the vocal tract model ($18.708 < \chi^2 < 1302.573$, all $P_s < 0.001$). Formants predicted from the computational models showed higher PICs for F1 and F4 (3.47 and 4.13, respectively), and lower potential for the other formants (Table 1).

Acoustic analysis of the nasal click grunts

The formants estimated from the click grunts of male ring-tailed lemurs showed a within-individual coefficient of variation of 19.56% for F1, 13.15% for F2, 6.98% for F3, 5.59% and 4.68% for F4 and F5, respectively. When we compared these values with variation among individuals, we obtained PICs ranging from 1.25 (F4) to 0.91 (F5, Table 1).

Discussion

This study presents the first clear evidence linking morphological variation in lemur vocal tracts to formant patterns of the acoustic output and the potential for individual vocal recognition. Research on human vocal tracts has shown considerable variation in length, shape and proportions (Lammert et al. 2011). However, humans themselves have rarely been included in such investigations (Fitch and Giedd 1999) and evidence for vocal tract variation in nonhuman primates is even scarcer. In this study, we quantified morphological differences in the vocal tracts of three adult male ring-tailed lemurs using silicone casts, and derived computational models to understand whether variation observed in the vocal tracts of these specimens could account for acoustical variations we recorded in the vocalizations of this species.

We built anatomical models resembling the vocal tracts of the cadavers from measurements taken from silicone casts. We used actual lengths and realistic cross-sectional areas to estimate differences in the formant patterns likely to have been emitted by the specimens before death. We found 2% variation in vocal tract length among the three specimens, and 20% variation in the volume of the luminal area of the tracts. According to our computational models, these morphological differences correspond to variation in formant frequencies, exceeding 18% for F1 and 6% for F2 and F4. These observations are in agreement with acoustic analyses of *Eulemur fulvus*, *E. macaco* and *E. rubriventer*, in which F1, F2, and F4 were more variable than the other formants making up the nasal grunts (Gamba et al. 2012b). Our results imply that formants contribute not only to the encoding of species identity in vocal emissions, but to individual identification as well. The degree of variation in individual formant patterns estimated from model-derived acoustic responses was similar to that observed in individually distinguishable vocalizations in other lemur species (Gamba et al. 2012b).

Our results demonstrate, in agreement with previous studies involving primates (Fitch 1997), canids (Riede and Fitch 1999), and birds (Fitch and Kelley 2000), that formants in lemur calls are inversely scaled with the length of the vocal tract. Model volume did not show a statistically significant influence on determining the location of formants in the acoustic response.

In the models in which we imposed random variation in the shape of the vocal tract, PC1 significantly influenced variation of F1, F2, F3, F4, and F5. PC2 had a significant effect on all but F4. The model volume, which could not be tested together with PC1 and PC2 in the LMMs to avoid variance inflation, had a significant effect on all formants. In agreement with findings on *Eulemur* spp. (Gamba et al. 2012b), where the shape of the cross-section of the vocal tract had a dramatic effect on the formant pattern. When comparing the effect of length variation (which we set at 15% instead of the 2% observed in the three cadavers) against shape variation (31% but randomized across the cross-sections), we showed that shape changes accounted for a greater degree of variation in formant structure, implying a higher potential for encoding individuality.

Most of the formants estimated from the recorded vocalizations showed higher intra-individual variation than predicted by the models. The PIC of F4 was greater than that of the other formants measured from the click grunts. It is interesting to note that the PIC of F4 was also the highest of those calculated from the models in which we altered vocal tract shape, and one of the highest (with F1) in the models in which length was changed.

The morphology of the larynx and the nasal airways is relevant in determining differences in formant location in ring-tailed lemur click grunts. Using vocal tract models and acoustic analysis, we provide clear evidence that lemur nasal grunts have the potential to allow individual recognition. Accordingly, higher among individual variation is in agreement with previous findings interpreting resonance frequencies as critical clues for individual discrimination (Rendall et al. 1998; Rendall 2003).

We can also interpret our results with respect to the perceptual abilities of lemurs. Within-species formant variation in *Eulemur* spp. has been estimated at 8% (Gamba et al. 2012a). Formant variation in *Lemur catta* appears to be slightly higher on average, around 11% (Table 1). We observed that, according to the audiogram published by Gillette and colleagues (1973, but see also Heffner 2014), the lowest perceptible frequency for *L. catta* is 67 Hz, and the species has a maximum sensitivity range between 5700 Hz and 16000 Hz (Ramsier and Dominy 2010). These values roughly comprise the frequency range of the formants we have estimated from our study of calls and from the computational models.

Our study also has potential limitations. First, recent research suggests that primate vocal tracts are more flexible than what we can reconstruct from investigations of post-mortem anatomy (Fitch et al. 2016). Although we think that this applies in particular to those calls that show oral tract articulation, we cannot rule out the possibility that future

research using digital imaging techniques would reveal more complex vocal tract dynamics involved in the production of lemur click grunts. Second, we assumed that nasal resonance took place without the involvement of the oral cavity, as we have observed the air stream flowing out from nostrils. The fact that click grunts resonate solely in the nasal tract is feasible in the light of the anatomy of the lemur vocal tract. However, we cannot exclude that discrepancy between the formants derived from the vocal tract models and those measured on the click grunts can be due to resonance in the oral tract before the column of air exits from the nose (Riede and Zuberbühler 2006). The difference between formants calculated from the vocal tract models and those measured on the grunts can also result from our choice to model the double intracranial nasal vocal tract as a uniform entity. However, our tests showed that the effect of using this simplification is a shift ranging between 1.4% and 2.0% (Gamba & Friard, unpublished data).

The high intra-individual variation we observed in the recordings may also derive from the fact that the click grunts are short and thus the analysis may be less efficient in estimating formants. This fact, combined with the restriction of our sample to lemur males and that we started from only three vocal tracts, has likely limited the potential to describe the morphological variation at a species level. As a consequence, the study of the individual formant patterns and vocal tract morphology in lemurs deserves further examination, both using larger samples of calls and specimens and considering other vocalization types.

Previous studies suggested that lemur societies are not as sophisticated as those of haplorhine primates (see Byrne and Whiten 1988). However, our results demonstrated that group-living lemurs show similar abilities to identify individuals as social haplorhines do. Such abilities can be associated with the establishment of cohorts and alliances (Soma and Koyama 2013).

Individual variation in lemur calls can also play a role in the non-invasive monitoring of wild populations and, more generally, contribute to the conservation of these endangered primates (Terry et al. 2005). The integration between acoustic analysis and computational modeling of the vocal apparatus is a promising field of modern zoology and bioacoustics. Increasing knowledge on the anatomical diversity of the lemur vocal tracts would be of interest to facilitate understanding of the evolution of lemur communication and the importance of acoustic cues in the process of lemur speciation.

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References

- Barr DJ, 2013. Random effects structure for testing interactions in linear mixed-effects models. *Frontiers in Psychology* 4: 328.
- Bates, D, Maechler, M, Bolker, B, and Walker, S, 2015. *lme4: Linear Mixed-Effects Models Using Eigen and S4*. R package version 1.
- Boersma, P, 2014. *The use of Praat in corpus research*. In: Durand J Gut U, Kristoffersen G eds. *The Oxford Handbook of Corpus Phonology*. Oxford: Oxford University Press.

- Byrne RW, Whiten A, 1989. *Machiavellian Intelligence: Social Expertise and the Evolution of Intellect in Monkeys, Apes, and Humans*. Oxford: Clarendon Press.
- Daqrouq K, Tutunji TA, 2015. Speaker identification using vowels features through a combined method of formants, wavelets, and neural network classifiers. *Applied Soft Computing* 27: 231–239.
- Dellwo V, Huckvale M, Ashby M, 2007. How is individuality expressed in voice? An introduction to speech production and description for speaker classification. *Lecture Notes in Computer Science* 4343: 1–20.
- Efremova KO, Volodin IA, Volodina EV, Frey R, Lapshina EN, Soldatova NV, 2011. Developmental changes of nasal and oral calls in the goitred gazelle *Gazella subgutturosa*, a nonhuman mammal with a sexually dimorphic and descended larynx. *Naturwissenschaften* 98: 919–931.
- Estienne V, Mundry R, Kühl HS, Boesch C, 2017. Exploitation of underground bee nests by three sympatric consumers in Loango National Park, Gabon. *Biotropica* DOI: 10.1111/btp.12354
- Ey E, Pfeufferle D, Fischer J, 2007. Do age- and sex-related variations reliably reflect body size in non-human primate vocalizations? A review. *Primates*, 48, 253–267.
- Fant G, 1971. *Acoustic Theory of Speech Production: with Calculations Based on –Ray Studies of Russian Articulations*. Vol. 2. Mouton: de Gruyter.
- Favaro L, Gamba M, Alfieri C, Pessani D, McElligott AG, 2015. Vocal individuality cues in the African penguin *Spheniscus demersus*: a source-filter theory approach. *Scientific Reports* 5: 17255.
- Fitch WT, 1997. Vocal tract length and formant frequency dispersion correlate with body size in rhesus macaques. *The Journal of the Acoustical Society of America* 102: 1213–1222.
- Fitch WT, 2000. The phonetic potential of nonhuman vocal tracts: comparative cineradiographic observations of vocalizing animals. *Phonetica* 57: 205–218.
- Fitch WT, 2006. *Production of vocalizations in mammals*. In: Brown K eds. *Encyclopedia of Language and Linguistics*. Oxford: Elsevier, 115–121.
- Fitch WT, Giedd J, 1999. Morphology and development of the human vocal tract: a study using magnetic resonance imaging. *The Journal of the Acoustical Society of America* 106: 1511–1522.
- Fitch WT, Kelley JP, 2000. Perception of vocal tract resonances by whooping cranes *Grus americana*. *Ethology* 106, 559–574.
- Fitch WT, Fritz JB, 2006. Rhesus macaques spontaneously perceive formants in conspecific vocalizations. *The Journal of the Acoustical Society of America* 120: 2132–2141.
- Fitch WT, de Boer B, Mathur N, Ghazanfar AA, 2016. Monkey vocal tracts are speech-ready. *Science Advances* 2: e1600723.
- Flanagan JL, 1972. *Speech Analysis: Synthesis and Perception*. 2nd edn. Berlin: Springer.
- Gamba M, Giacoma C, 2006. Vocal tract modeling in a prosimian primate: the black and white ruffed lemur. *Acta Acustica United with Acustica* 92: 749–755.
- Gamba M, Giacoma C, 2007. Quantitative acoustic analysis of the vocal repertoire of the crowned lemur. *Ethology Ecology & Evolution* 19: 323–343.
- Gamba M, Favaro L, Torti V, Sorrentino V, Giacoma C, 2011. Vocal tract flexibility and variation in the vocal output in wild indris. *Bioacoustics* 20: 251–266.
- Gamba M, Colombo C, Giacoma C, 2012a. Acoustic cues to caller identity in lemurs: a case study. *Journal of Ethology* 30: 191–196.
- Gamba M, Friard O, Giacoma C, 2012b. Vocal tract morphology determines species-specific features in vocal signals of lemurs *Eulemur*. *International Journal of Primatology* 33: 1453–1466.
- Gamba M, Friard O, Riondato I, Righini R, Colombo C et al., 2015. Comparative analysis of the vocal repertoire of *Eulemur*: a dynamic time warping approach. *International Journal of Primatology* 36: 894–910.
- Gamba M, Torti V, Estienne V, Randrianarison RM, Valente D et al., 2016. The indris have got rhythm! Timing and pitch variation of a primate song examined between sexes and age classes. *Frontiers in Neuroscience* 10: 249.
- Gillette RG, Brown R, Herman P, Vernon S, Vernon J, 1973. The auditory sensitivity of the lemur. *American Journal of Physical Anthropology* 38: 365–370.

- Gustison ML, Le Roux A, Bergman TJ, 2012. Derived vocalizations of geladas *Theropithecus gelada* and the evolution of vocal complexity in primates. *Philosophical Transactions of the Royal Society B* 367: 1847–1859.
- Heffner RS, 2004. Primate hearing from a mammalian perspective. *The Anatomical Record Part A: Discoveries in Molecular, Cellular, and Evolutionary Biology* 281: 1111–1122.
- Kulahci IG, Drea CM, Rubenstein DI, Ghazanfar AA, 2014. Individual recognition through olfactory-auditory matching in lemurs. *Proceedings of the Royal Society B*, 281: 20140071.
- Lammert AC, Proctor MI, Katsamanis A, Narayanan SS, 2011. Morphological variation in the adult vocal tract: A modeling study of its potential acoustic impact. *INTERSPEECH* 2813.
- Lapshina EN, Volodin IA, Volodina EV, Frey R, Efremova KO et al., 2012. The ontogeny of acoustic individuality in the nasal calls of captive goitred gazelles *Gazella subgutturosa*. *Behavioural Processes* 90: 323–330.
- Macedonia JM, 1986. Individuality in a contact call of the ringtailed lemur *Lemur catta*. *American Journal of Primatology* 11: 163–179.
- Macedonia JM, 1993. The vocal repertoire of the ringtailed lemur *Lemur catta*. *Folia Primatologica* 61: 163–179.
- Mathevon N, Charrier I, Jouventin P, 2003. Potential of individual recognition in acoustic signals: a comparative study of two gulls with different nesting patterns. *Cr Acad Sci III-Vie* 326: 329–337.
- Nadhrourou B, Gamba M, Andriaholinirina NV, Ouledi A, Giacoma C, 2016. The vocal communication of the mongoose lemur *Eulemur mongoz*: phonation mechanisms, acoustic features and quantitative analysis. *Ethology Ecology & Evolution* 28: 241–260.
- Oda R, 1996. Effects of contextual and social variables on contact call production in free-ranging ringtailed lemurs *Lemur catta*. *International Journal of Primatology* 17: 191–205.
- Oda R, 1999. Scent marking and contact call production in ring-tailed lemurs *Lemur catta*. *Folia Primatologica* 70: 121–214.
- Oda R, 2002. Individual distinctiveness of the contact calls of ring-tailed lemur. *Folia Primatologica* 72: 132–136.
- Palagi E, Norscia I, 2015. The season for peace: reconciliation in a despotic species *Lemur catta*. *PLoS ONE*, 10: e0142150.
- Pollard KA, Blumstein DT, 2011. Social group size predicts the evolution of individuality. *Current Biology* 21: 413–417.
- Pruthi T, Espy-Wilson CY, 2007. Acoustic parameters for the automatic detection of vowel nasalization. *INTERSPEECH*, 1925.
- Ramsier MA, Dominy NJ, 2010. A comparison of auditory brainstem responses and behavioral estimates of hearing sensitivity in *Lemur catta* and *Nycticebus coucang*. *American Journal of Primatology* 72: 217–233.
- Rendall D, Kollias S, Ney C, Lloyd P, 2005. Pitch, F0) and formant profiles of human vowels and vowel-like baboon grunts: the role of vocalizer body size and voice-acoustic allometry. *The Journal of the Acoustical Society of America* 117: 944–955.
- Rendall D, Owren MJ, Rodman PS, 1998. The role of vocal tract filtering in identity cueing in rhesus monkey *Macaca mulatta* vocalizations. *The Journal of the Acoustical Society of America* 103: 602–614.
- Rendall D, Rodman PS, Emond RE, 1996. Vocal recognition of individuals and kin in free-ranging rhesus monkeys. *Animal Behaviour* 51: 1007–1015.
- Rendall D, 2003. Acoustic correlates of caller identity and affect intensity in the vowel-like grunt vocalizations of baboons. *The Journal of the Acoustical Society of America* 113: 3390–3402.
- Riede T, Fitch WT, 1999. Vocal tract length and acoustics of vocalization in the domestic dog *Canis familiaris*. *Journal of Experimental Biology* 202: 2859–2867.
- Riede T, Bronson E, Hatzikirou H, Zuberbühler K, 2005. Vocal production mechanisms in a non-human primate: morphological data and a model. *Journal of Human Evolution* 48: 85–96.
- Riede T, Zuberbühler K, 2003. The relationship between acoustic structure and semantic information in Diana monkey alarm vocalization. *The Journal of the Acoustical Society of America* 114: 1132–1142.
- Scheumann M, Roser AE, Konerding W, Bleich E, Hedrich HJ et al., 2012. Vocal correlates of sender-identity and arousal in the isolation calls of domestic kitten *Felis silvestris catus*. *Frontiers in Zoology* 9: 1.
- Sibiryakova OV, Volodin IA, Matrosova VA, Volodina EV, Garcia AJ et al., 2015. The power of oral and nasal calls to discriminate individual mothers and offspring in red deer, *Cervus elaphus*. *Frontiers in Zoology* 12: 1.
- Sokal R, Rohlf FJ, 1995. *Biometry*. New York: WH Freeman.

- Soma T, Koyama N, 2013. Eviction and troop reconstruction in a single matriline of ring-tailed Lemurs *lemur catta*: what happened when “grandmother” died? In: Masters J, Gamba M, Génin F eds. *Leaping Ahead*. Springer, 137–146.
- Taylor, AM, and Reby, D, 2010. The contribution of source-filter theory to mammal vocal communication research. *Journal of Zoology* 280: 221–236.
- Terry AM, Peake TM, McGregor PK, 2005. The role of vocal individuality in conservation. *Frontiers in Zoology* 2:1.
- Tibbetts EA, Dale J, 2007. Individual recognition: it is good to be different. *Trends in Ecology and Evolution* 22: 529–537.
- Titze IR, 1994. *Principles of Voice Production*. Englewood Cliffs: Prentice Hall.
- Volodin IA, Sibiryakova OV, Kokshunova LE, Frey R, Volodina EV, 2014. Nasal and oral calls in mother and young trunk-nosed saiga antelopes *Saiga tatarica*. *Bioacoustics* 23: 79–98.
- Zhang Z, Espy-Wilson CY, 2004. A vocal-tract model of American English/l. *The Journal of the Acoustical Society of America* 115: 1274–1280.
- Zhou X, Espy-Wilson CY, Boyce S, Tiede M, Holland C et al., 2008. A magnetic resonance imaging-based articulatory and acoustic study of “retroflex” and “bunched” American English/r. *The Journal of the Acoustical Society of America* 123: 4466–4481.

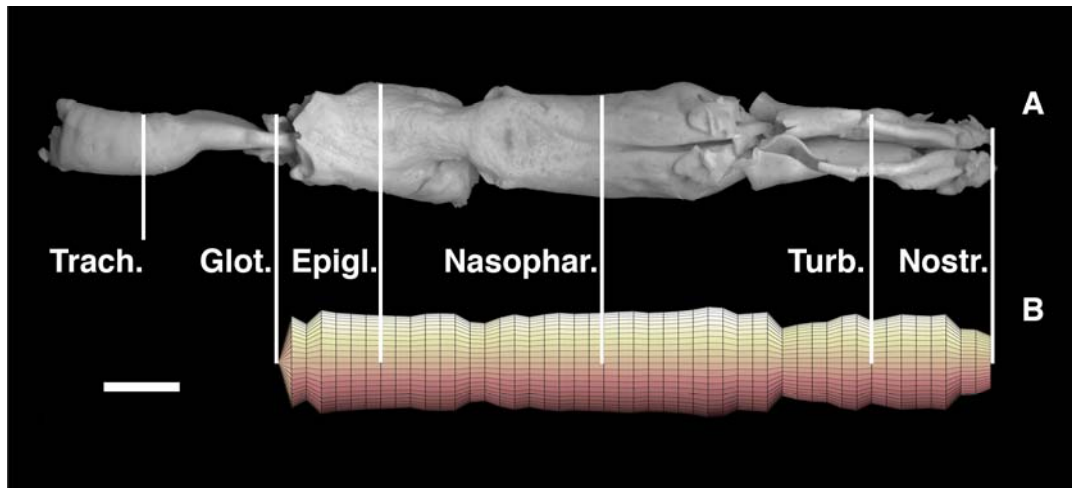


Figure 1. An old silicone cast of the nasal tract of a male *Lemur catta* and its 3D model. (A) Dorsal view of the silicone cast stretched on a plane. (B) A 3D rendering of the concatenated tubes model. The luminal areas of the tract are labelled with the corresponding anatomical parts: Trach., trachea; Glot., glottis; Epigl., epiglottis; Nasophar., nasopharynx; Turb., turbinates; Nostr., nostril. Scale bar, 10 mm.

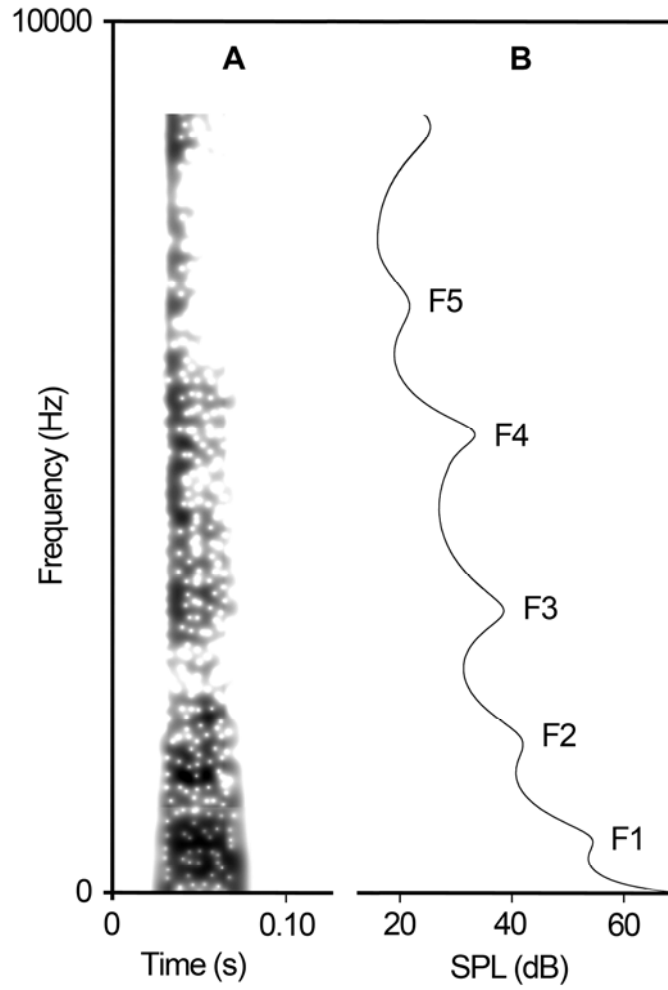


Figure 2. Spectrogram (A) and linear predictive coding analysis (B) of a ring-tailed lemur click grunt. Formants F1-F5 are indicated on the respective LPC peaks.

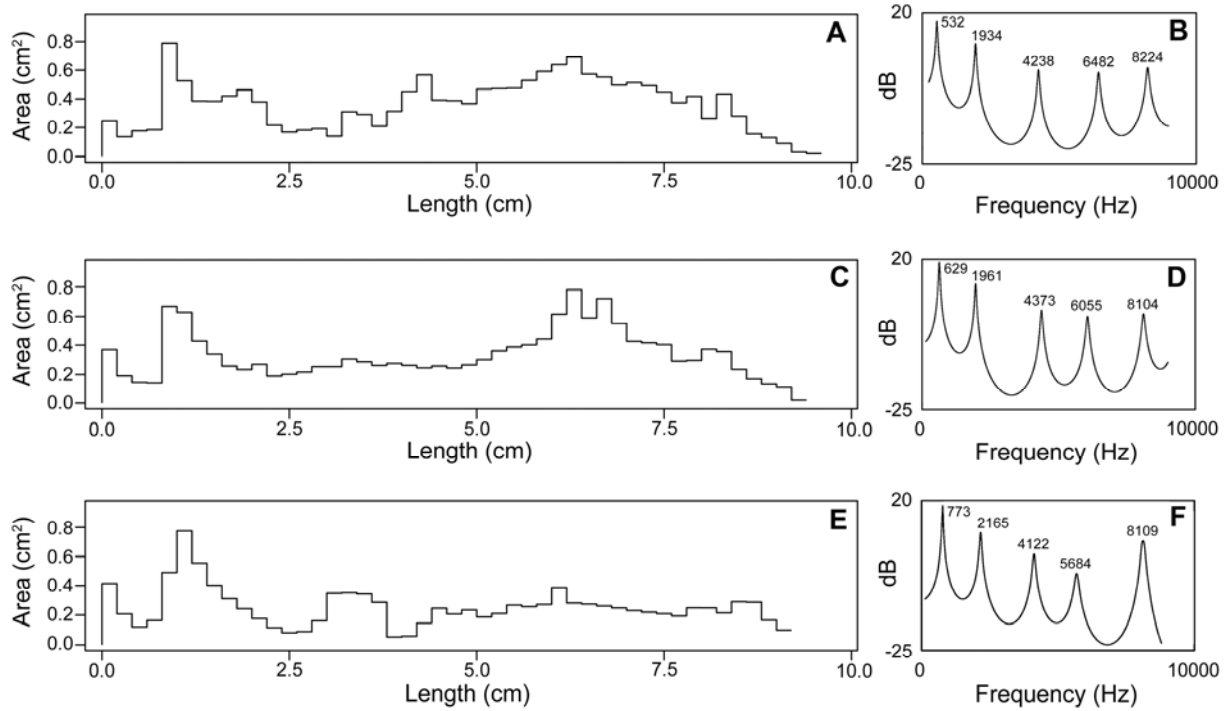


Figure 3. Cross-sectional area functions of the discretized vocal tracts of three male ring-tailed lemurs (A - PBZTX1, C - PBZTX2, E - PBZTA1). We took area measurements in 46 to 48 0.2 cm steps from the glottis to the nostrils. The two nostrils were modelled as one. The corresponding vocal tract transfer functions estimated from the acoustic response of the computational models are shown on the right (B, D, F). The numbers above the frequency peaks are the values of the formants F1–F5.

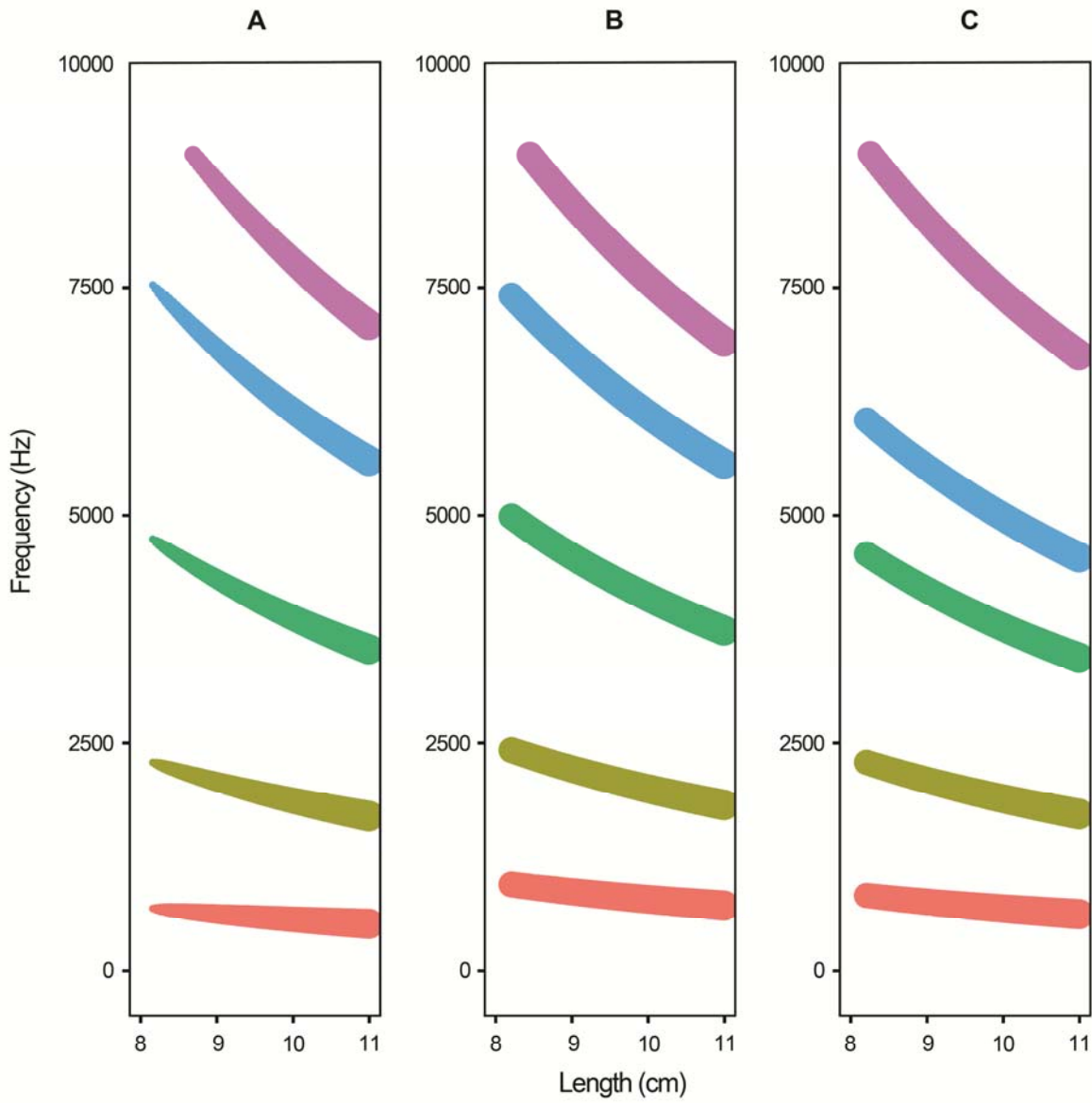


Figure 4. Variation of formant frequencies F1 to F5 in the models corresponding to PBZTXY1 (A), PBZTXY2 (B), and PBZTA1 (C). Vocal tract length was standardized across models and then gradually increased and decreased within a 15% range of variation.

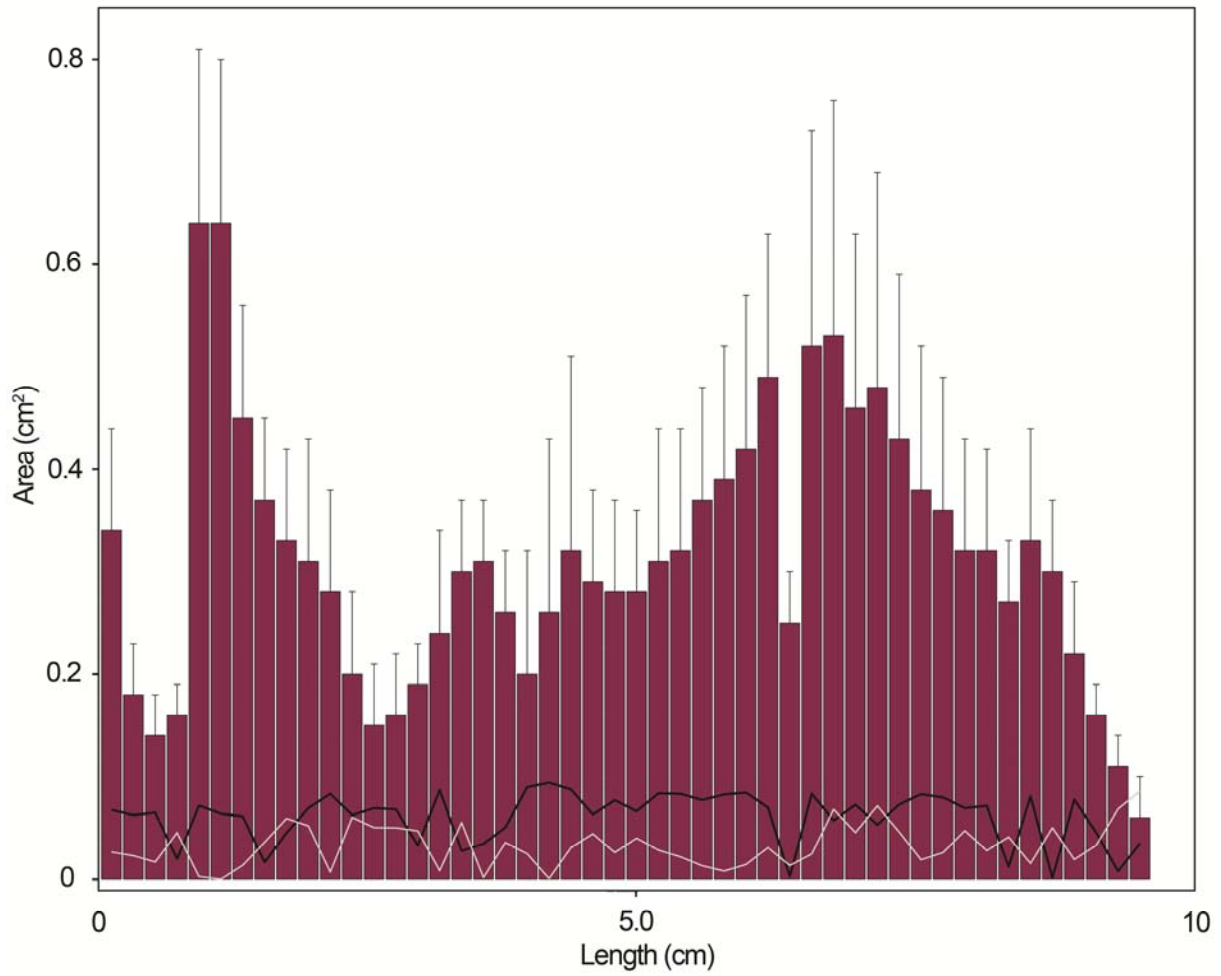


Figure 5. Average (+ standard deviation) shape variation (bars) estimated from 15000 computational vocal tract models. The traces below represent the loading of each segment on the Principal Components (PC1 in black, PC2 in grey).

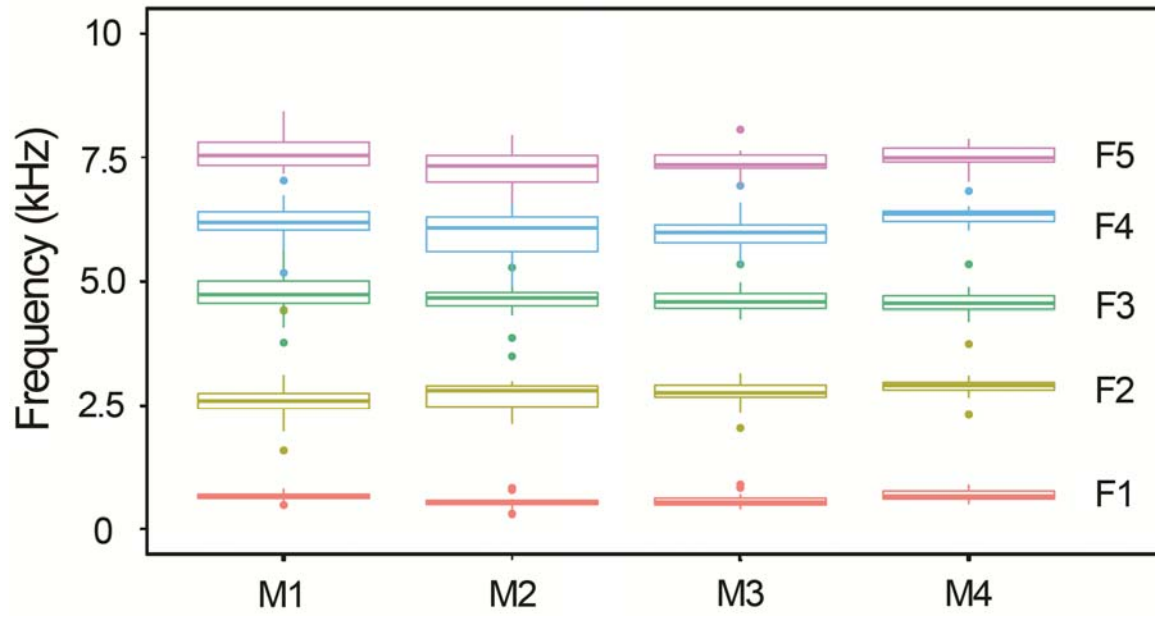


Figure 6. Individual variation of formants F1-F5 in four male ring-tailed lemurs (M1-M4) recorded in captivity. Box-and-Whisker plots show the lower and upper quartiles. Solid lines represent medians and dots are outliers.

Table 1. First five formant frequencies for the different vocal tract models and vocalizations. Lengths and cross-sectional areas measured from the cadavers (Anatomical) were then standardized to a length of 9.60 cm (*) to simulate changes in length (Length variation) and shape (Shape-variation). Formants measured from click grunts are presented for each male individual we recorded. The potential for individuality coding (PIC) is also reported.

Method	Vocal tract model	Length (cm)	Volume (cm ³)	F1 (Hz)	F2 (Hz)	F3 (Hz)	F4 (Hz)	F5 (Hz)	N
Anatomical	g1	9.6	3.481	532	1943	4238	6432	8224	1
	g2	9.4	3.117	629	1961	4373	6055	8104	1
	g3	9.2	2.322	773	2165	4122	5684	8109	1
Length variation (±15%)	g1*	9.58±0.84	3.52±0.310	595±490	1967±172	4078±357	6472±568	7955±555	60
	g2*	9.58±0.85	3.21±0.280	828±700	2102±181	4317±378	6422±556	7846±607	60
	g3*	9.58±0.86	2.38±0.210	721±620	1987±170	3966±345	5242±453	7763±652	60
	PIC			1.88	1.05	1.08	1.48	1.01	
Shape variation (±31%)	g1*	9.6	3.49±0.110	576±400	1913±510	3964±940	6293±136	7964±180	5000
	g2*	9.6	3.19±0.100	807±200	2048±480	4200±920	6248±132	7742±196	5000
	g3*	9.6	2.36±0.070	701±200	1933±54	3862±105	5108±139	7538±210	5000
	PIC			3.47	1.53	1.77	4.13	1.34	
Click grunts	Male 1			657±710	2623±381	4790±359	6169±437	7592±233	44
	Male 2			542±113	2665±286	4588±287	5875±200	7271±229	16
	Male 3			564±161	2734±434	4612±298	5999±198	7383±314	8
	Male 4			693±114	2907±305	4580±330	6321±516	7518±592	10
	PIC			1.05	1.03	1.15	1.25	0.91	

Table 2. Influences of the predictors on formant frequencies; results of the full models (full vs. null).

Length	χ^2	<i>df</i>	<i>P</i>		Estimate	<i>SE</i>	<i>df</i>	χ^2	<i>P</i>
	F1	1493.238	3	<0.001	Intercept	8.772	0.145	^a	^a
				length	-1.009	0.092	1	93.546	<0.001
				volume	0.057	0.092	1	0.344	0.558
F2	1664.793	3	<0.001	Intercept	9.778	0.066	^a	^a	^a
				length	-0.949	0.054	1	169.226	<0.001
				volume	-0.029	0.054	1	0.212	0.646
F3	1976.838	3	<0.001	Intercept	10.554	0.038	^a	^a	^a
				length	-0.992	0.024	1	421.019	<0.001
				volume	0.003	0.024	1	0.036	0.849
F4	1762.378	3	<0.001	Intercept	10.891	0.089	^a	^a	^a
				length	-0.960	0.045	1	231.12	<0.001
				volume	-0.024	0.045	1	0.216	0.642
F5	1804.203	3	<0.001	Intercept	11.216	0.027	^a	^a	^a
				length	-0.990	0.02	1	453.72	<0.001
				volume	0.001	0.02	1	0.025	0.874
Shape	χ^2	<i>df</i>	<i>P</i>		Estimate	<i>SE</i>	<i>df</i>	χ^2	<i>P</i>
F1	709.1559	3	<0.001	Intercept	6.533	0.069	^a	^a	^a
				PC1	-0.015	0.003	1	24.675	<0.001
				PC2	0.042	0.002	1	671.730	<0.001
F2	387.5482	3	<0.001	Intercept	7.582	0.018	^a	^a	^a
				PC1	-0.021	0.002	1	158.327	<0.001
				PC2	0.013	0.001	1	211.118	<0.001
F3	724.1647	3	<0.001	Intercept	8.295	0.025	^a	^a	^a
				PC1	0.039	0.002	1	656.934	<0.001
				PC2	0.008	0.001	1	92.726	<0.001
F4	411.8387	3	<0.001	Intercept	8.675	0.05	^a	^a	^a
				PC1	0.030	0.001	1	411.169	<0.001
				PC2	0.001	0.001	1	0.039	0.844
F5	1036.343	3	<0.001	Intercept	8.955	0.018	^a	^a	^a
				PC1	0.011	0.002	1	49.338	<0.001
				PC2	-0.025	0.001	1	844.259	<0.001

^a Not shown as not having a meaningful interpretation.