Hidden shifts in allometry scaling between sound production and perception in anurans

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Abstract

Background. Animal communication consists of signal production and perception, which are crucial for social interactions. The main form used by anurans is auditory communication, in most cases produced as advertisement calls. Furthermore, sound perception happens mainly through an external tympanic membrane, and plays an important role in social behavior. In this study, we evaluated the influence of body and tympanic membrane sizes on call frequency across the phylogeny of anurans.

Methods. We use data on snout-vent length, tympanic membrane diameter, and dominant frequency of the advertisement call from the literature and from natural history museum collections. We mapped these traits across the anuran phylogeny and tested different models of

40   diversification. Our final dataset includes data on body size, tympanic membrane size, and call
41   dominant frequency of 735 anuran species.
42   **Results.** The best explanatory model includes body and tympanum size with no interaction term.
43   Although our results show that call frequency is strongly constrained by body and tympanum
44   size, we identify five evolutionary shifts in allometry from that ancestral constraint. We relate
45   these evolutionary shifts to the background noise experienced by populations. We recognize two
46   sources of background noise causing allometric shifts: i) environmental noise due to fast flowing
47   water bodies and ii) biotic noise due to the sound emitted by other syntopic individuals. Body
48   size is important for myriad ecological interactions and tympanum size is strongly associated
49   with female call frequency preferences. Thus, allometric escape in frog calls might arise through
50   environmental selection such as related to breeding in fast flowing or soundscape competition, as
51   well as sexual selection linked to tympanum size.
52
Introduction
54   Communication between animals involves signal production and perception. Signal production is
55   a balance of energy cost and efficiency in information transmission, and it can be highly
56   variable. Communication signal production can be visual (Osorio & Vorobyev, 2008), olfactory
57   (Bossert & Wilson, 1963), tactile (Weber, 1973; Cerrone, 2019), electric (Bratton & Ayers,
58   1987) or acoustic (Suthers et al., 2016). In addition, signal perception is relevant for successfully
59   accomplishing communication, which is related to the efficiency of signal transmission, and
60   consequently influence evolution in communication systems (Endler, 1993).
61   Among different forms of signal production, acoustic signals are one of the most widespread
62   across animals, present in invertebrates (Wenner, 1964) and in all classes of vertebrates (Peters
63   & Ploog, 1973; Ladich, 2019). These signals are shaped by diverse selective pressures, such as
64   species recognition (Claridge & de Vrijer, 1994; Gerhardt & Bee, 2007), predator pressure
65   (Cade, 1975; Tuttle & Ryan, 1981), and sexual selection (Rebouças, Augusto-Alves & Toledo,
66   2020). Among the advantages of acoustic emission are the relatively fast signal transmission,
67   orientation and its complexity. For instance, sounds can be subdivided into components such as
68   frequency, amplitude, duration, and emission rate, which can be decoded into different
69   information (Lopez & Narins, 1991; Morais et al., 2012). However, acoustic communication can
70   be masked by the background noise, jeopardizing communication success (Duarte et al., 2019;
71   Lima et al., 2022). Moreover, conspicuous acoustic signals can attract acoustically oriented
72   predators (Tuttle & Ryan, 1981). In most cases, sound perception is closely related to receiver
73   organs and structures, which are quite diverse and can improve signal sensitivity in such a
74   context. In contrast, anurans can also perceive sounds using novel adaptations, such as in the tiny
75   pumpkin toadlet from the Atlantic Rainforest, *Brachycephalus rotenbergae*. In this species, the
76   inner ear (here the basilar recess) is not connected to its nervous system, suggesting that high
77   frequency sound vibrations (as the sound of their own calls) cannot be recognized. However, low
78   frequency vibrations can be perceived through their arms, which transmit sound to the inner ear
79   through bone vibrations (Goutte et al., 2017).

80   Anurans present a range of communication signals (e.g., Cardoso & Heyer, 1995; Toledo et al.,
81   2015; Narins, 2019), which can be used independently or in combination depending on the
82   behavioral context (Hartmann et al., 2005; Lourenço-de-Moraes et al., 2016; Rebouças, Augusto-
83   Alves & Toledo, 2020; Rebouças, 2022). However, the most used signal in anurans is
84   vocalizations (Toledo et al., 2015; Köhler et al., 2017). Anurans present several vocalization or
85   calls types used in social contexts, such as reproductive, defensive, and aggressive calls.
86   Advertisement calls, one form of reproductive call, are the most widespread communication
87   strategy in anurans, which are generally emitted to attract females and guard territories (Toledo
88   et al., 2015; Köhler et al., 2017). The variation of these calls in both spectral and temporal
89   parameters is also diverse. Although it is well known that temporal parameters of calls, such as
90   call rate and duration, are influenced by the environmental temperature (Lingnau & Bastos,
91   2007; Love & Bee, 2010; Lima et al., 2022), spectral parameters in turn are less so. Spectral
92   parameters of anuran calls are generated by anatomical structures, and consequently constrained
93   by the body size of the calling individual (Rebouças, Augusto-Alves & Toledo, 2020; Tonini et
94   al., 2020). Vocalizations in anurans are produced by the contraction of trunk muscles leading the
95   air passage from the lungs to the buccal cavity, passing through the larynx where it causes the
96   vocal cords to vibrate and, finally, produce sounds (Colafrancesco & Gridi-Papp, 2016). These
97   sounds are further modified by the laryngeal muscles (Gridi-Papp, 2008; Ryan & Guerra, 2014)
98   and other related structures, such as buccal cavity and vocal sac apertures (Kime, Ryan &
99   Wilson, 2013).
100   Besides call emission, call perception also plays a role in anuran social contexts. Anurans use
101   calls to assess other individuals' physical condition and, consequently, respond in terms of
102   territorial defense (Foratto et al., 2021; Rebouças, 2022). Thus, the information contained in calls
103   determines territorial segregation, reproduction, and their fitness (e.g., Giasson & Haddad, 2006;
104   Dautel et al., 2011). For most anurans, the tympanic membrane is the first structure to capture the
105   external sound waves, transmitting acoustic vibrations to their inner ear. In general, it is
106   connected to the otic capsule via extrastapes and stapes, also referred to as extracolumella and
107   columella (Van Dijk et al., 2011; Mason et al., 2015). Some studies have reported that there is a
108   direct relationship between size and acoustic sensitivity, which means that the larger the
109   individuals, with larger tympanic membranes, the better the sound perception (Fox, 1995; James
110   et al., 2022). These relations are physically constrained: larger individuals also have more
111   massive vocals chords, which tends to result in lower call frequencies (Ryan, 1988a); and larger
112   individuals also present larger tympanic membranes, which are more prone to vibrate with
113   sounds with lower amplitudes, which results in a more sensitive ear (Fox, 1995). Thus, an escape
114   from these ancestral relationships must be rare, and probably a consequence of a greater selective
115   pressure resulting from fundamental physical constraints. Also, most studies have concentrated
116   on a few species, and a broader overview of allometric relationships between tympanum, body
117   size, and call frequency across anurans is still lacking.
118   There is a general understanding that the advertisement call frequency of most anuran species is
119   correlated with individual's body size; i.e., the larger the frog, the lower its advertisement call

120 frequency (Ryan, 1988a). However, this pattern was not observed for some anuran lineages (e.g.
121 Southeast Asian ranids, Ranid frogs, Fitzinger Neotropical tree frogs and Poison frogs) that
122 evolved to have divergent allometric relationships (Tonini et al., 2020). Moreover, some recent
123 evidence suggests that anatomical structures closely related to communication, such as tympanic
124 membrane, must be a constraint in the context of the relation between call frequency and sound
125 perception (James et al., 2022). Consequently, an analysis using a phylogenetic approach to test
126 the relation between sound emission and perception should shed light on this relationship and
127 improve the understanding of groups that previously presented allometric escapes. This study
128 aims to evaluate the influence of body and tympanum sizes on advertisement call frequency
129 across the anuran phylogeny.
130

Materials & Methods

131

132 Bioacoustic and morphometric data
133 We assembled data on mean advertisement calls dominant frequency, which is the call frequency
134 with the higher energy, males' snout-vent length (SVL; mm; hereafter, called simply body size)
135 and tympanum diameter (TD; mm; hereafter, called simply tympanum size; Fig. 1) of adult male
136 frogs from literature and complemented with measurements from specimens deposited in the
137 Museu de Diversidade Biológica (MDBio), Universidade de Campinas, Brazil (see
138 Supplementary Material). We used dominant frequency of advertisement call because, among
139 variables in anurans' call, this is stereotyped and not influenced by environmental conditions,
140 such as temperature and humidity (Köhler et al., 2017), and it is commonly used in species
141 description (e.g., Toledo, Ribeiro & Haddad, 2007; Köhler et al., 2017; de Andrade et al., 2020).
142 Also, we only used measurements from male individuals because they are more available in
143 literature than measurements from females, which allowed us to perform the analysis in large
144 scale. Finally, we were not able to include those species which present no visible tympanum (or
145 even no tympanum), since measurement in this case are only possible through anatomical
146 desiccation, which is not commonly available in the literature.
147 Many species present a sexual size dimorphism (review in Monnet & Cherry, 2002); thus, we
148 only considered males for the analysis from type series in species descriptions. We were not able
149 to include the information for those species for which there are only females or juveniles in the
150 type series. In several cases, males presented oval-shaped tympana. In these cases, we considered
151 only tympanum length for our purposes. The dataset used here is available in supplementary
152 material following the current nomenclature available in Frost (2023).
153

154 Phylogenetic comparative analysis
155 We trimmed the amphibian phylogeny (Jetz & Pyron, 2018) to include only species present in
156 our dataset (See Supplementary Material). We log-transformed the data on dominant frequency,
157 snout-vent length, and tympanum size. We estimated the phylogenetic signal of each traits using
158 Bloomberg's K (Blomberg, Garland Jr & Ives, 2003) and Pagel's lambda (Pagel, 1999) in
159 phytools (Revell, 2012). In addition, we tested the fit of three nested models using PGLS, 1) DF

160 ~ SVL, 2) DF ~ SVL + TD and 3) DF ~ SVL + TD + SVL*TD, and compared them using
161 Akaike's Information Criterion (AIC) and Bayesian Information Criterion (BIC). The best fit
162 model was implemented in the R v. 4.0.5 (R Core Team, 2022) package bayou. The bayou
163 package fits Bayesian reversible-jump multi-optima Ornstein-Uhlenbeck (OU) models to
164 phylogenetic comparative data (Uyeda & Harmon, 2014). We used the bayou model to identify
165 the location across the anuran phylogeny, support, and magnitude of shifts in intercept and slope
166 of the scaling relationship between dominant frequency, body size, and tympanum size. Our
167 expectation is that most frog species adhere to a background allometric scaling given the strong
168 constraint imposed by body size on functional and anatomical traits. Here, we ask whether some
169 frog species would represent shifts in the allometric scaling of dominant frequency with body
170 and tympanum sizes. We used as prior a half-Cauchy distribution for a and s2, and normal
171 distribution for b and θ. In addition, we included 0.1 of measurement error to the data. We tuned
172 model parameters to have acceptance ratios between 0.2-0.4. We ran the models four times, each
173 run had 10 million generations, and we used the first 30% as burn, and filtered the results to
174 shifts with 0.75 posterior probability or higher. We check whether all runs would result in similar
175 anuran species identified as having distinct scaling compared to most other frog species. Shifts
176 with less than four species were not considered. Analyses and data visualization were performed
177 in R using packages ape (Paradis & Schliep, 2019), phytools (Revell, 2012), Geiger (Pennell et
178 al., 2014), ggtree (Yu et al., 2017).
179

Results

181 We compiled complete information (advertisement call dominant frequency, body size,
182 tympanum size, and phylogeny) of 735 species. Body (Fig. 2A; r2 = 0.452, p < 0.001) and
183 tympanum (Fig. 2B; r2 = 0.311, p < 0.001) sizes are inversely correlated with the dominant
184 frequency but directly correlated with each other (Fig. 2C; r2 = 0.714, p < 0.001). Thus, large
185 frogs tend to have large tympana and call at lower dominant frequency compared to smaller
186 frogs, confirming the strong allometric relationship between these traits. After phylogeny is
187 taken into account, the influence of tympanum size is attenuated in relation to the model with no
188 phylogeny, which is shown by the difference in slope between regression lines, but it still shows
189 significant correlation (Fig. 3). All three traits have significant phylogenetic signal for both
190 Bloomberg's K (K_{SVL} = 0.27, p = 0.001; K_{TYM} = 0.28, p = 0.001; K_{DF} = 0.13, p = 0.001) and
191 Pagel's lambda (λ_{SVL} = 0.85, p < 0.001; λ_{TYM} = 0.85, p < 0.001; λ_{DF} = 0.72, p < 0.001).
192 Our model comparison results show that models 2 (DF ~ SVL + TD) and 3 (DF ~ SVL + TD +
193 SVL*TD) presented similar marginal likelihood and BIC (Table 1). Although there is a strong
194 allometric relationship between body size and tympanum size, we consider that the simplest
195 model with no interaction between variables provided a better fit to the data (θDf ~ βSVL +
196 βTD, Table 2), and used this model to test shifts in evolutionary allometry across anurans.
197 Despite the great diversity of body and tympanum sizes and dominant frequency across frogs, we
198 confirm the prior expectation that most frog species adhere to a single allometric scaling
199 relationship. However, we identify five shifts from the evolutionary constraint imposed by body

200   size on tympanum size and dominant frequency (Table 2, Fig. 3). In Ranidae, we observe two
201   embedded shifts: 1) *Rana* and *Pelophylax*, and 2) *Hylarana*, *Odorrana*, *Babina*, and *Amolops*;
202   thus, we consider, for our purposes, as a single shift shared by their most recent common
203   ancestral. Then, in Ranidae, we observe two regime shifts: a shift comprising *Huia* and
204   *Meristogenys* (hereafter called Asian ranids), and another shift comprising *Rana* and *Pelophylax*,
205   *Hylarana*, *Odorrana*, *Babina* and *Amolops* (hereafter called Ranid frogs).
206   Among other regime shifts, in *Dendropsophus* (hereafter Fitzinger Neotropical Treefrogs) and
207   Leiuperinae (hereafter Neotropical swamp frogs) we observe a negative slope, as most of
208   anurans, but with a different intercept for the allometric relationship. In Fitzinger Neotropical
209   treefrogs, sound frequency is decoupled from body size but still negatively correlated to
210   tympanum size, while in Neotropical swamp frogs the dominant frequency is associated with
211   body and tympanum size (Table 2). In Hylodidae (hereafter Neotropical torrent frogs), Asian
212   ranids, and Ranid frogs, the evolution of dominant frequency is decoupled from the constraint of
213   body and tympanum size, which is shown by the zero slope; whereas in Ranid frogs we observe
214   the inverse situation, in which sound frequency is decoupled from tympanum size but still
215   persists dependent on body size. In Asian ranids, sound frequency is dissociated from tympanum
216   size and positively correlated to body size, which is unique and indicates that large species
217   within those genera tend to call at higher frequency as opposed to what is expected for most
218   other frog species (Fig. 4 and 5).
219
Discussion
221   Frogs have a wide range of body and tympanum size and call frequency, as well as a great
222   diversity of reproductive behaviors (Haddad & Prado, 2005; Nunes-de-Almeida, Haddad &
223   Toledo, 2021). In addition, frogs have colonized a variety of environments across all continents
224   except Antarctica. Despite the vast environmental complexity in terms of biotic and abiotic
225   factors presenting a myriad of selection pressures, the constraint of body size overcomes those
226   selective pressures and strongly constrain the relationship between sound frequency and
227   tympanum size. Our results show that most frog species adhere to a single allometric scaling
228   relationship between advertisement call dominant frequency, body, and tympanum size.
229   Although previously reported for the relationship between call dominant frequency and body size
230   (Tonini et al., 2020; James et al., 2022), we observe here allometric escapes for the relationship
231   between dominant frequency and tympanum size as well.
232   In at least four groups, shifts appear to be independent, considering the great phylogenetic
233   distance between them (Fig. 3) – out of the five shifts observed here, two of them are within
234   ranids. Additionally, our results include allometric shifts for three of four previously reported
235   groups (Asian ranids, Fitzinger Neotropical treefrogs, and ranid frogs) (Tonini et al., 2020). We
236   did not observe any allometric shift including tympanum size for Neotropical poison frogs, as
237   previously reported, and we estimate a shift for Neotropical swamp frogs and Neotropical torrent
238   frogs, which were not observed previously (Tonini et al., 2020). Thus, considering that Tonini et
239   al. (2020) only evaluated variables related to sound emission and James et al. (2022) evaluated

240   the tympanum size allometry (linked to sound perception) of a reduced number of species (81
241   spp., with little overlap for all measurements), our study represents a broader estimation of
242   allometric shifts across the anuran tree of life.
243   Our results show that call frequency is dependent on the size of individuals and correlated with
244   tympanum size. Advertisement calls in anurans, mostly emitted by males, are used both to attract
245   mates and to segregate calling males (Toledo et al., 2015). Accordingly, we suggest two sets of
246   limiting factors: internal, which constrain the vocalization emission, such as body size and other
247   physiological implications (Köhler et al., 2017); and external, which constrain the understanding
248   of social context through the calls of other males, such as inner ear structures, amphibian papilla
249   (for lower frequencies), and basilar papilla (for higher frequencies) (Schoffelen, Segenhout &
250   Van Dijk, 2008). Among species groups representing allometric shifts, Fitzinger Neotropical
251   treefrogs and Neotropical swamp frogs showed a similar negative relation between tympanum
252   size and dominant frequency (i.e., for sound sensitivity). This relationship was distinct from all
253   other groups (Asian ranids, Ranid frogs and Neotropical torrent frogs), which possibly indicates
254   different selective pressures for sound perception. In relation to sound emission (i.e., size and
255   dominant frequency), for Fitzinger Neotropical treefrogs, the sound frequency was decoupled
256   from body size and Neotropical swamp frogs remained size dependent with species calling at
257   lower frequency than expected. For Ranid frogs (referred as *Rana* and *Pelophylax*, *Hylarana*,
258   *Odorrana*, *Babina*, and *Amolops* in Table 1), it is similar to Fitzinger Neotropical treefrogs and
259   Neotropical swamp frogs, but for Neotropical torrent frogs and Asian ranids, the relation was
260   inverse, with larger individuals presenting higher dominant frequencies.
261   Some causes of allometric shifts might be common for all groups of frogs in a certain way. For
262   example, species of Asian ranids, ranid frogs, and Neotropical torrent frogs call near waterfalls
263   and fast flowing water bodies. These environments are highly noisy, and consequently, over the
264   time can limit calls to frequencies higher than the background noise (Tonini et al., 2020). In
265   *Crossodactylus schimidti*, for example, males show a short-term adjustment for dominant
266   frequency in face of background noise frequency (Vidigal et al., 2018). Similar results were
267   found for other species in the genus (e.g., *C. gaudichaudii* and *C. werneri*) and for most of
268   Hylodes species as well (e.g., *H. charadranaetes*, *H. glaber*, and *H. malhagaesi*) (Augusto-
269   Alves, Dena & Toledo, 2021). Among species of the other two groups recovered as a shifts, *Huia*
270   *cavitympanum* for example present most of their communication through ultrasonic calls,
271   ranging from 5 to 25 kHz (Arch, Grafe & Narins, 2008), as do *Wijayarana masonii* (formerly *H.*
272   *masonii*) (Boonman & Kurniati, 2011). Similar results were also found for *Amolops tormotus*
273   (Feng et al., 2006) and *Odorrana graminea* (Shen et al., 2011).
274   The Fitzinger Neotropical treefrogs and Neotropical swamp frogs are not known to call in fast
275   flowing water environments, but our results also indicate them as an allometric scape from the
276   ancestral body size constraint. Species of these two groups use ponds and swamps as
277   reproductive sites. Species in both groups reproduce year around, frequently with hundreds of
278   individuals calling at the same time very closely from each other in lek (Barreto & Andrade,
279   1995; Camargo et al., 2005; Abrunhosa, Wogel & Pombal, 2006; Curi et al., 2014; Pompeu, de

280 Sa & Haddad, 2020). In this case, the selective force causing is not fast flowing water but
281 hypothesized to be the noise of other organisms that produce sounds, including vertebrates and
282 invertebrates, that may impose similar selective pressure to individuals as fast flowing streams
283 (e.g., Both & Grant, 2012; Lima et al., 2022). The environment in which these species tend to
284 live might also influence the optimal call frequency (Marten, Quine & Marler, 1977; Fricke,
285 1984). For a given body size, individuals of Fitzinger Neotropical treefrogs and Neotropical
286 swamps frogs call at a lower frequency than expected, which might represent an advantage in
287 territorial dispute and female attraction. The fitness of males is commonly evaluated through call
288 for both males and females (Rebouças, Augusto-Alves & Toledo, 2020; Rebouças, 2022), since
289 lower-frequency calls indicate larger males (Tonini et al., 2020), which may present an
290 advantage in disputes (Ryan, 1988b). Consequently, larger males, which spend less energy
291 calling in lower frequencies, present a higher probability to win disputes, and consequently better
292 protect the reproductive territory. In this scenario, females tend to be more attracted by those
293 males through lower-frequency calls (Ryan, 1988b). In some cases, males could even present a
294 call with a frequency lower than predicted by its size, which effort the sexual selection role of
295 calls (Rebouças, Augusto-Alves & Toledo, 2020). In these two groups, the selective constraint of
296 body size on sound frequency and tympanum size is weaker compared to other frogs, which is
297 shown by the lower slope value relative to the background regime. Once sound frequency and
298 tympanum size are less constrained by body size, call frequency not necessarily would indicate
299 larger sizes. Therefore, selection on tympanum size and male sound frequency could result from
300 other parts of the male advertisement call, such as temporal parameters or behavioral interactions
301 that include territorial or mating displays. However, an intraspecific relationship between call
302 frequency / body size / tympanum size could still be present, and further analysis should evaluate
303 it considering an within species variation, mainly in these groups that presented allometric
304 escapes.
305

Conclusions
307 This study evaluated allometric escape across the anuran phylogeny using parameters of call
308 emission (i.e., males' call frequency) and parameters of call sensitivity (i.e., tympanum size). We
309 showed that the inclusion of tympanum size allows the identification of new acoustic allometric
310 shifts across anurans. Also, we hypothesize that shifts might result from selective pressure of
311 background noise and those that reproduce in high species-rich or overpopulated ponds. Finally,
312 our observations provide insights for future studies which aim to evaluate sound communication
313 in anurans, and additional conclusions could be reached with measurements of females in the
314 analysis.
315

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322

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324 **References**

325

326 Abrunhosa PA, Wogel H, Pombal JP. 2006. Anuran temporal occupancy in a temporary pond

327   from the Atlantic Rain Forest, South-Eastern Brazil. *The Herpetological Journal* 16:115–

328   122.

329 de Andrade FS, Haga IA, Ferreira JS, Recco-Pimentel SM, Toledo LF, Bruschi DP. 2020. A new

330   cryptic species of *Pithecopus* (Anura, Phyllomedusidae) in north-eastern Brazil.

331   *European Journal of Taxonomy* 723:108–134.

332 Arch VS, Grafe TU, Narins PM. 2008. Ultrasonic signalling by a Bornean frog. *Biology Letters*

333   4:19–22.

334 Augusto-Alves G, Dena S, Toledo LF. 2021. Acoustic allometry, background stream noise and

335   its relationship with large-bodied and voiceless rheophilic frogs. *Zoologischer Anzeiger*

336   295:156–162.

337 Barreto L, Andrade GV. 1995. Aspects of the reproductive biology of *Physalaemus cuvieri*

338   (Anura: Leptodactylidae) in northeastern Brazil. *Amphibia-Reptilia* 16:67–76.

339 Blomberg SP, Garland Jr T, Ives AR. 2003. Testing for phylogenetic signal in comparative data:

340   behavioral traits are more labile. *Evolution* 57:717–745.

341 Boonman A, Kurniati H. 2011. Evolution of high-frequency communication in frogs. *Evolutionary*

342   *Ecology Research* 13:197–207.

343 Bossert WH, Wilson EO. 1963. The analysis of olfactory communication among animals.

344   *Journal of Theoretical Biology* 5:443–469. DOI: 10.1016/0022-5193(63)90089-4.

345 Both C, Grant T. 2012. Biological invasions and the acoustic niche: the effect of bullfrog calls on

346   the acoustic signals of white-banded tree frogs. *Biology letters* 8:714–716.

347 Bratton BO, Ayers JL. 1987. Observations on the electric organ discharge of two skate species

348   (Chondrichthyes: Rajidae) and its relationship to behaviour. *Environmental Biology of*

349   *fishes* 20:241–254.

350 Cade W. 1975. Acoustically orienting parasitoids: fly phonotaxis to cricket song. *Science*

351   190:1312–1313.

352 Camargo A, Naya DE, Canavero A, da Rosa I, Maneyro R, Naya D. 2005. Seasonal activity and

353   the body size—fecundity relationship in a population of *Physalaemus gracilis*

354   (Boulenger, 1883)(Anura, Leptodactylidae) from Uruguay. *Annales Zoologici Fennici*

355   42:513–521.

356 Cardoso AJ, Heyer WR. 1995. Advertisement, aggressive, and possible seismic signals of the

357   frog *Leptodactylus syphax* (Amphibia, Leptodactylidae). *Alytes* 13:67–76.

358 Cerrone M. 2019. Keepers as social companions: Tactile communication and social enrichment

359   for captive apes. *Sign Systems Studies* 47:453–479.

360 Claridge MF, de Vrijer PW. 1994. Reproductive behavior: the role of acoustic signals in species

361   recognition and speciation. In: *Planthoppers*. Springer, 216–233.

362 Colafrancesco KC, Gridi-Papp M. 2016. Vocal Sound Production and Acoustic Communication

363   in Amphibians and Reptiles. In: Suthers RA, Fitch WT, Fay RR, Popper AN eds.

364   *Vertebrate Sound Production and Acoustic Communication*. Cham: Springer

365   International Publishing, 51–82. DOI: 10.1007/978-3-319-27721-9_3.

366 Curi LM, Olea G, Álvarez BB, Céspedez JA, Lombardo DM, others. 2014. Reproductive aspects

367   of *Dendropsophus sanborni* (Anura, Hylidae) in northeastern Argentina. *Iheringia. Série*

368   *Zoologia* 104:432–438.

369 Dautel N, Maldonado ALS, Abuza R, Imba H, Griffin K, Guayasamin JM. 2011. Advertisement

370   and combat calls of the glass frog *Centrolene lynchi* (Anura: Centrolenidae), with notes

371   on combat and reproductive behaviors. *Phyllomedusa: Journal of Herpetology* 10:34–43.

372   Duarte MH, Caliari EP, Scarpelli MD, Lobregat GO, Young RJ, Sousa-Lima RS. 2019. Effects of

373   mining truck traffic on cricket calling activity. *The Journal of the Acoustical Society of*

374   *America* 146:656–664.

375   Endler JA. 1993. Some general comments on the evolution and design of animal

376   communication systems. *Philosophical Transactions of the Royal Society of London.*

377   *Series B: Biological Sciences* 340:215–225.

378   Feng AS, Narins PM, Xu C-H, Lin W-Y, Yu Z-L, Qiu Q, Xu Z-M, Shen J-X. 2006. Ultrasonic

379   communication in frogs. *Nature* 440:333–336. DOI: 10.1038/nature04416.

380   Foratto RM, Llusia D, Toledo LF, Forti LR. 2021. Treefrogs adjust their acoustic signals in

381   response to harmonics structure of intruder calls. *Behavioral Ecology* 32:416–427.

382   Fox JH. 1995. Morphological correlates of auditory sensitivity in anuran amphibians. *Brain,*

383   *behavior and evolution* 45:327–338.

384   Fricke F. 1984. Sound attenuation in forests. *Journal of Sound and Vibration* 92:149–158. DOI:

385   10.1016/0022-460X(84)90380-8.

386   Frost DR. 2023.Amphibian Species of the World: an Online Reference. Version 6.1. *Available at*

387   *https://amphibiansoftheworld.amnh.org/index.php.* (accessed April 3, 2021).

388   Gerhardt HC, Bee MA. 2007. Recognition and localization of acoustic signals. In: *Hearing and*

389   *sound communication in amphibians*. Springer, 113–146.

390   Giasson LO, Haddad CF. 2006. Social interactions in *Hypsiboas albomarginatus* (Anura:

391   Hylidae) and the significance of acoustic and visual signals. *Journal of Herpetology*

392   40:171–180.

393   Goutte S, Mason MJ, Christensen-Dalsgaard J, Montealegre-Z F, Chivers BD, Sarria-S FA,

394   Antoniazzi MM, Jared C, Sato LA, Toledo LF. 2017. Evidence of auditory insensitivity to

395   vocalization frequencies in two frogs. *Scientific Reports* 7:12121.

396   Gridi-Papp M. 2008. The structure of vocal sounds produced with the mouth closed or with the

397   mouth open in treefrogs. *The Journal of the Acoustical Society of America* 123:2895–

398   2902.

399   Haddad CF, Prado CP. 2005. Reproductive modes in frogs and their unexpected diversity in the

400   Atlantic Forest of Brazil. *BioScience* 55:207–217.

401   Hartmann MT, Giasson LO, Hartmann PA, Haddad CF. 2005. Visual communication in Brazilian

402   species of anurans from the Atlantic forest. *Journal of Natural History* 39:1675–1685.

403   James LS, Taylor RC, Hunter KL, Ryan MJ. 2022. Evolutionary and allometric insights into

404   anuran auditory sensitivity and morphology. *Brain, Behavior and Evolution* 97:138–148.

405   Jetz W, Pyron RA. 2018. The interplay of past diversification and evolutionary isolation with

406   present imperilment across the amphibian tree of life. *Nature ecology & evolution* 2:850–

407   858.

408   Kime NM, Ryan MJ, Wilson PS. 2013. A bond graph approach to modeling the anuran vocal

409   production system. *The Journal of the Acoustical Society of America* 133:4133–4144.

410   Köhler J, Jansen M, Rodriguez A, Kok PJ, Toledo LF, Emmrich M, Glaw F, Haddad CF, Roedel

411   M-O, Vences M. 2017. The use of bioacoustics in anuran taxonomy: theory, terminology,

412   methods and recommendations for best practice. *Zootaxa* 4251:1–124.

413   Ladich F. 2019. Ecology of sound communication in fishes. *Fish and Fisheries* 20:552–563.

414   DOI: 10.1111/faf.12368.

415   Lima N de AP, Rebouças R, Toledo LF, Lisboa CS. 2022. Influence of urban noise in call traits

416   of the Atlantic Forest treefrog *Bokermannohyla hylax*. *Zoologischer Anzeiger* 300:41–46.

417   Lingnau R, Bastos RP. 2007. Vocalizations of the Brazilian torrent frog *Hylodes heyeri* (Anura:

418   Hylodidae): Repertoire and influence of air temperature on advertisement call variation.

419   *Journal of Natural History* 41:1227–1235. DOI: 10.1080/00222930701395626.

420   Lopez PT, Narins PM. 1991. Mate choice in the neotropical frog, Eleutherodactylus coqui.

421   *Animal Behaviour*.

422 Lourenço-de-Moraes R, Ferreira RB, Mira-Mendes CV, Zocca CZ, Medeiros T, Ruas DS,

423   Rebouças R, Toledo LF, Brodie ED, Solé M. 2016. Escalated antipredator mechanisms

424   of two neotropical marsupial treefrogs. *The Herpetological Journal* 26:237–244.

425 Love EK, Bee MA. 2010. An experimental test of noise-dependent voice amplitude regulationin

426   Cope's grey treefrog, *Hyla chrysoscelis*. *Animal Behaviour* 80:509–515. DOI:

427   10.1016/j.anbehav.2010.05.031.

428 Marten K, Quine D, Marler P. 1977. Sound transmission and its significance for animal

429   vocalization: II. Tropical forest habitats. *Behavioral ecology and sociobiology*:291–302.

430 Mason MJ, Segenhout JM, Cobo-Cuan A, Quiñones PM, van Dijk P. 2015. The Frog Inner Ear:

431   Picture Perfect? *Journal of the Association for Research in Otolaryngology* 16:171–188.

432   DOI: 10.1007/s10162-015-0506-z.

433 Monnet J-M, Cherry MI. 2002. Sexual size dimorphism in anurans. *Proceedings of the Royal*

434   *Society of London. Series B: Biological Sciences* 269:2301–2307.

435 Morais AD, Bastos RP, Annunziata BB, ... 2012. Description of the advertisement call of

436   Rhinella mirandaribeiroi (Gallardo, 1965)(Anura: Bufonidae). *Zootaxa*.

437 Narins PM. 2019. Seismic Communication in the Amphibia with Special Emphases on the

438   Anura. In: Hill PSM, Lakes-Harlan R, Mazzoni V, Narins PM, Virant-Doberlet M, Wessel

439   A eds. *Biotremology: Studying Vibrational Behavior*. Cham: Springer International

440   Publishing, 277–292. DOI: 10.1007/978-3-030-22293-2_14.

441 Nunes-de-Almeida CHL, Haddad CFB, Toledo LF. 2021. A revised classification of the

442   amphibian reproductive modes. *Salamandra* 57:413–427.

443 Osorio D, Vorobyev M. 2008. A review of the evolution of animal colour vision and visual

444   communication signals. *Vision Research* 48:2042–2051. DOI:

445   10.1016/j.visres.2008.06.018.

446 Pagel M. 1999. Inferring the historical patterns of biological evolution. *Nature* 401:877–884.

447   Paradis E, Schliep K. 2019. ape 5.0: an environment for modern phylogenetics and evolutionary

448   analyses in R. *Bioinformatics* 35:526–528.

449   Pennell MW, Eastman JM, Slater GJ, Brown JW, Uyeda JC, FitzJohn RG, Alfaro ME, Harmon

450   LJ. 2014. geiger v2. 0: an expanded suite of methods for fitting macroevolutionary

451   models to phylogenetic trees. *Bioinformatics* 30:2216–2218.

452   Peters M, Ploog D. 1973. Communication Among Primates. *Annual Review of Physiology*

453   35:221–242. DOI: 10.1146/annurev.ph.35.030173.001253.

454   Pompeu CCL, de Sa FP, Haddad CF. 2020. Seasonal reproductive dynamics of a lek-breeding

455   Neotropical treefrog is not organized by male size (Anura, Hylidae). *South American*

456   *Journal of Herpetology* 18:33–41.

457   R Core Team. 2022. *R: A Language and Environment for Statistical Computing*. Vienna,

458   Austria: R Foundation for Statistical Computing.

459   Rebouças R. 2022. White-edged cowards: high-pitched treefrogs will be attacked by those with

460   orange legs. *Behaviour*:1–25. DOI: https://doi.org/10.1163/1568539X-bja10187.

461   Rebouças R, Augusto-Alves G, Toledo LF. 2020. Evolution of treefrogs' calls in tropical islands

462   might be under directional selection. *Journal of Zoology* 312:43–52. DOI:

463   https://doi.org/10.1111/jzo.12792.

464   Revell LJ. 2012. phytools: An R package for phylogenetic comparative biology (and other

465   things). *Methods in Ecology and Evolution* 3:217–223.

466   Ryan MiJ. 1988a. Constraints and patterns in the evolution of anuran acoustic communication.

467   In: *The evolution of the amphibian auditory system*. 637–677.

468   Ryan MJ. 1988b. Energy, calling, and selection. *American Zoologist* 28:885–898.

469   Ryan MJ, Guerra MA. 2014. The mechanism of sound production in túngara frogs and its role in

470   sexual selection and speciation. *Current opinion in neurobiology* 28:54–59.

471   Schoffelen RL, Segenhout JM, Van Dijk P. 2008. Mechanics of the exceptional anuran ear.

472   *Journal of Comparative Physiology A* 194:417–428.

473   Shen J-X, Xu Z-M, Yu Z-L, Wang S, Zheng D-Z, Fan S-C. 2011. Ultrasonic frogs show

474   extraordinary sex differences in auditory frequency sensitivity. *Nature Communications*

475   2:1–5.

476   Suthers RA, Fitch WT, Fay RR, Popper AN (eds.). 2016. *Vertebrate Sound Production and*

477   *Acoustic Communication*. Cham: Springer International Publishing : Imprint: Springer.

478   DOI: 10.1007/978-3-319-27721-9.

479   Toledo LF, Martins IA, Bruschi DP, Passos MA, Alexandre C, Haddad CF. 2015. The anuran

480   calling repertoire in the light of social context. *Acta Ethologica* 18:87–99.

481   Toledo LF, Ribeiro RS, Haddad CFB. 2007. Anurans as prey: an exploratory analysis and size

482   relationships between predators and their prey. *Journal of Zoology* 271:170–177.

483   Tonini JFR, Provete DB, Maciel NM, Morais AR, Goutte S, Toledo LF, Pyron RA. 2020.

484   Allometric escape from acoustic constraints is rare for frog calls. *Ecology and Evolution*

485   10:3686–3695.

486   Tuttle MD, Ryan MJ. 1981. Bat predation and the evolution of frog vocalizations in the

487   Neotropics. *Science* 214:677–678.

488   Uyeda JC, Harmon LJ. 2014. A novel Bayesian method for inferring and interpreting the

489   dynamics of adaptive landscapes from phylogenetic comparative data. *Systematic*

490   *biology* 63:902–918.

491   Van Dijk P, Mason MJ, Schoffelen RL, Narins PM, Meenderink SW. 2011. Mechanics of the frog

492   ear. *Hearing research* 273:46–58.

493   Vidigal I, De Carvalho TR, Clemente-Carvalho RB, Giaretta AA. 2018. Vocalizations, tadpole,

494   and natural history of *Crossodactylus werneri* Pimenta, Cruz Caramaschi, 2014 (Anura:

495   Hylodidae), with comments on distribution and intraspecific variation. *Zootaxa* 4388:61–

496   75.

497   Weber I. 1973. Tactile communication among free-ranging langurs. *American Journal of*

498   *Physical Anthropology* 38:481–486. DOI: 10.1002/ajpa.1330380251.

499   Wenner AM. 1964. Sound Communication in Honeybees. *Scientific American* 210:116–125.

500   Yu G, Smith DK, Zhu H, Guan Y, Lam TT-Y. 2017. ggtree: an R package for visualization and

501   annotation of phylogenetic trees with their covariates and other associated data.

502   *Methods in Ecology and Evolution* 8:28–36.

503

Output of models

Model selection considering only tympanum (TD, model 1), only snout-vent length (SVL, model 2), considering both (model 3) or considering both variables and the interaction between them (model 4) in relation to dominant frequency (Df). Values of p refer to specific comparison between models 2 and 3 and 3 and 4, and significant values are in bold.

1   Table 1. Model selection considering only tympanum (TD, model 1), only snout-vent length

2   (SVL, model 2), considering both (model 3) or considering both variables and the interaction

3   between them (model 4) in relation to dominant frequency (Df). Values of *p* refer to specific

4   comparison between models 2 and 3 and 3 and 4, and significant values are in bold.

Model	call	df	AIC	BIC	logLik	Test	L. Ratio	*p*
1	log(Df) ~ log(TD)	3	1765.00	1778.80	-879.50		–	–
2	log(Df) ~ log(SVL)	3	1603.63	1617.43	-798.82		–	–
3	log(Df) ~ log(SVL) + log(TD)	4	1583.61	1602.01	-787.81	2 vs 3	22.02	< **0.001**
4	log(Df) ~ log(SVL) + log(TD) + log(SVL) * log(TD)	5	1577.24	1600.24	-783.62	3 vs 4	8.37	**0.004**

Regime shifts

Model estimates of slope and intercept for the evolutionary regime shifts in dominant frequency (DF), tympanum (TD), and body size (SVL).

1 Table 2. Model estimates of slope and intercept for the evolutionary regime shifts in dominant

2 frequency (DF), tympanum (TD), and body size (SVL).

Taxa	θ_{DF}	β_{TD}	β_{SVL}	Posterior probability
Root	9.338	-0.369	-0.4034	
Fitzinger Neotropical treefrogs	8.507	-0.179	-0.0412	0.81
Neotropical swamp frogs	7.821	-0.185	-0.1371	0.96
Neotropical torrent frogs	8.078	0.025	0.0999	0.95
Asian ranids	8.543	0.048	0.1383	0.86
Ranid frogs				
Rana	7.235	0.001	-0.1159	0.76
Amolops, Babina, Hylarana, Pelophylax, and *Odorrana*	8.402	-0.073	-0.0828	0.68

Figure 1

Diversity of tympana in Anura

Concave tympanum of *Huia cavitympanum* (photo by Ulmar Grafe) (A); enlarged tympanum in *Thoropa megatympanum* (photo by Carlos Henrique Luz Nunes-de-Almeida) (B); regular tympanum in *Hylodes cardosoi* (photo by Luís Felipe Toledo) (C); tympanum whit external apparatus in *Petropedetes vulpiae* (photo by Václav Gvoždík) (D); reduced tympanum in *Megaelosia apuana* (photo by João Luiz Gasparini) (E); and tympanum not externally visible in *Cycloramphus rhyakonastes* (photo by Luís Felipe Toledo) (F) (individuals present different sizes).
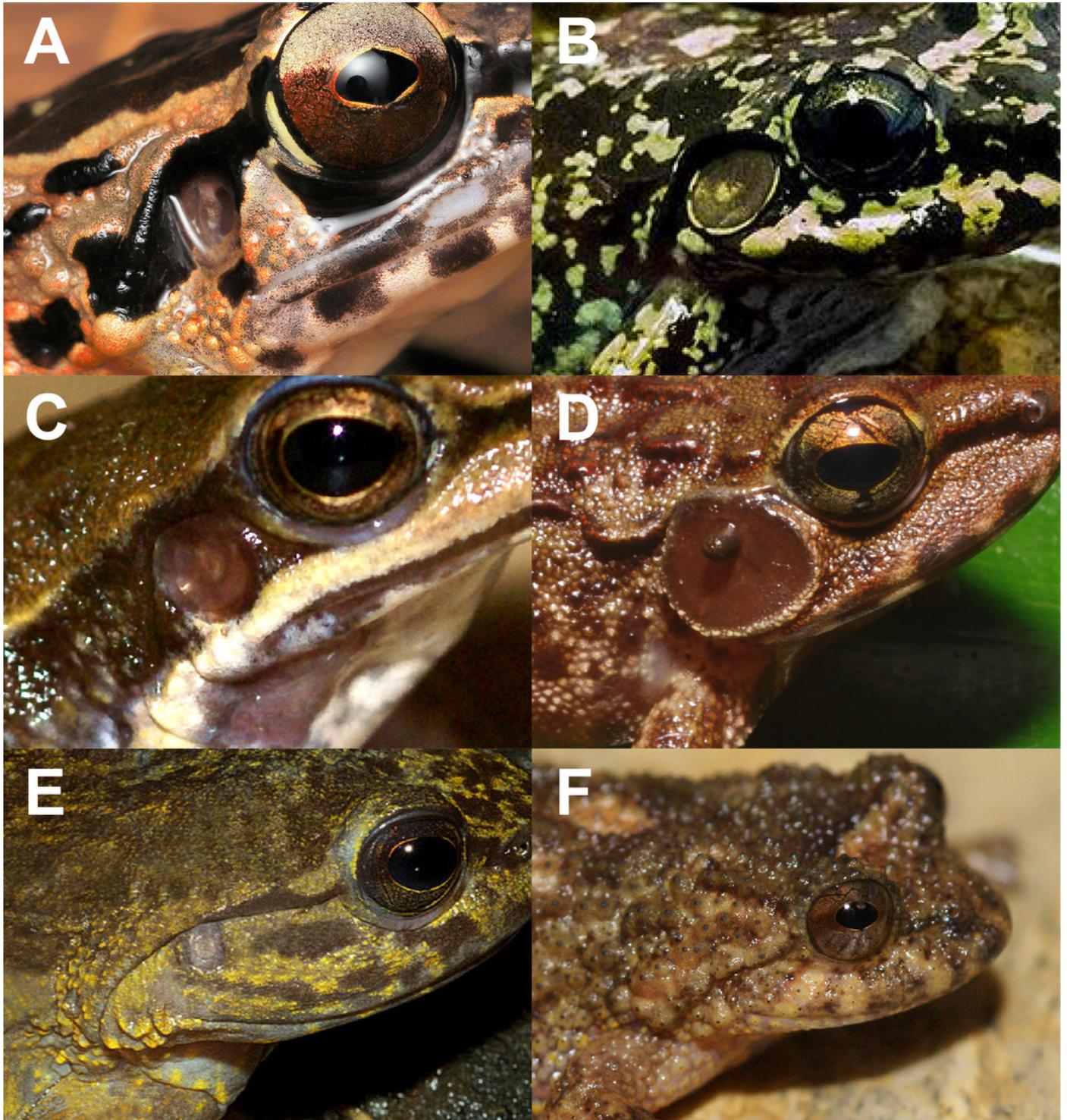
Figure 2

Linear regression (solid line) and phylogenetic generalized linear squared models (dotted line).

body and tympanum sizes explain 45% and 31% of the diversity of dominant frequency, respectively. Moreover, body size explains 71% of the variation in tympanum size.
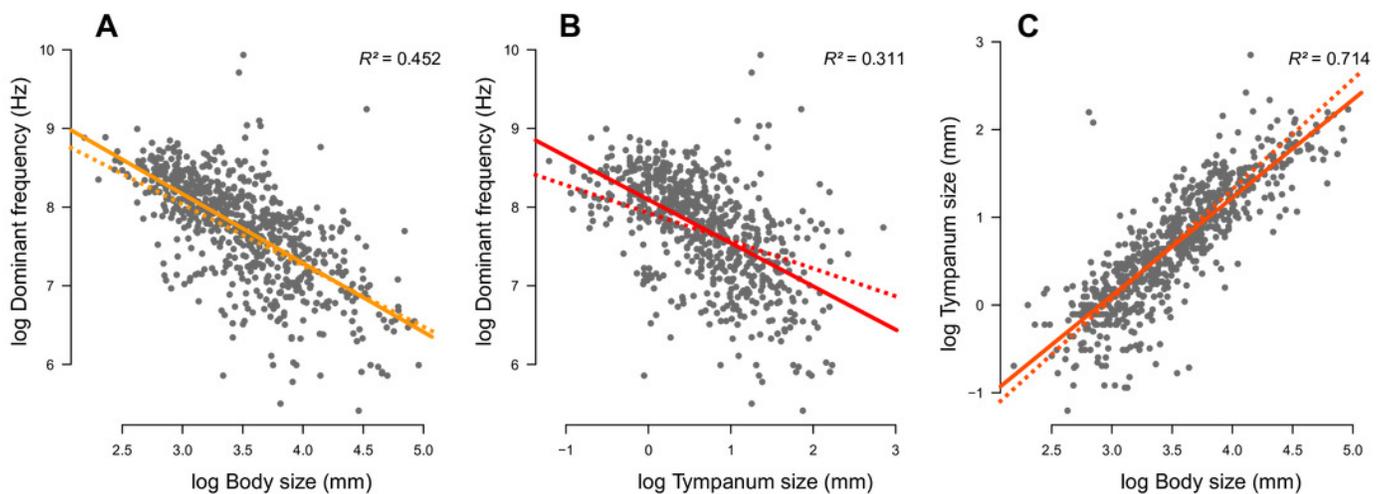
Figure 3

Measured variables on the phylogeny

Barplot of measured variables of the 735 species included in our estimatives. Inner circle represent the Snout-vent length (SVL), mid circle represent dominant frequency and outer circle represent tympanum size. Values are log-transformed.
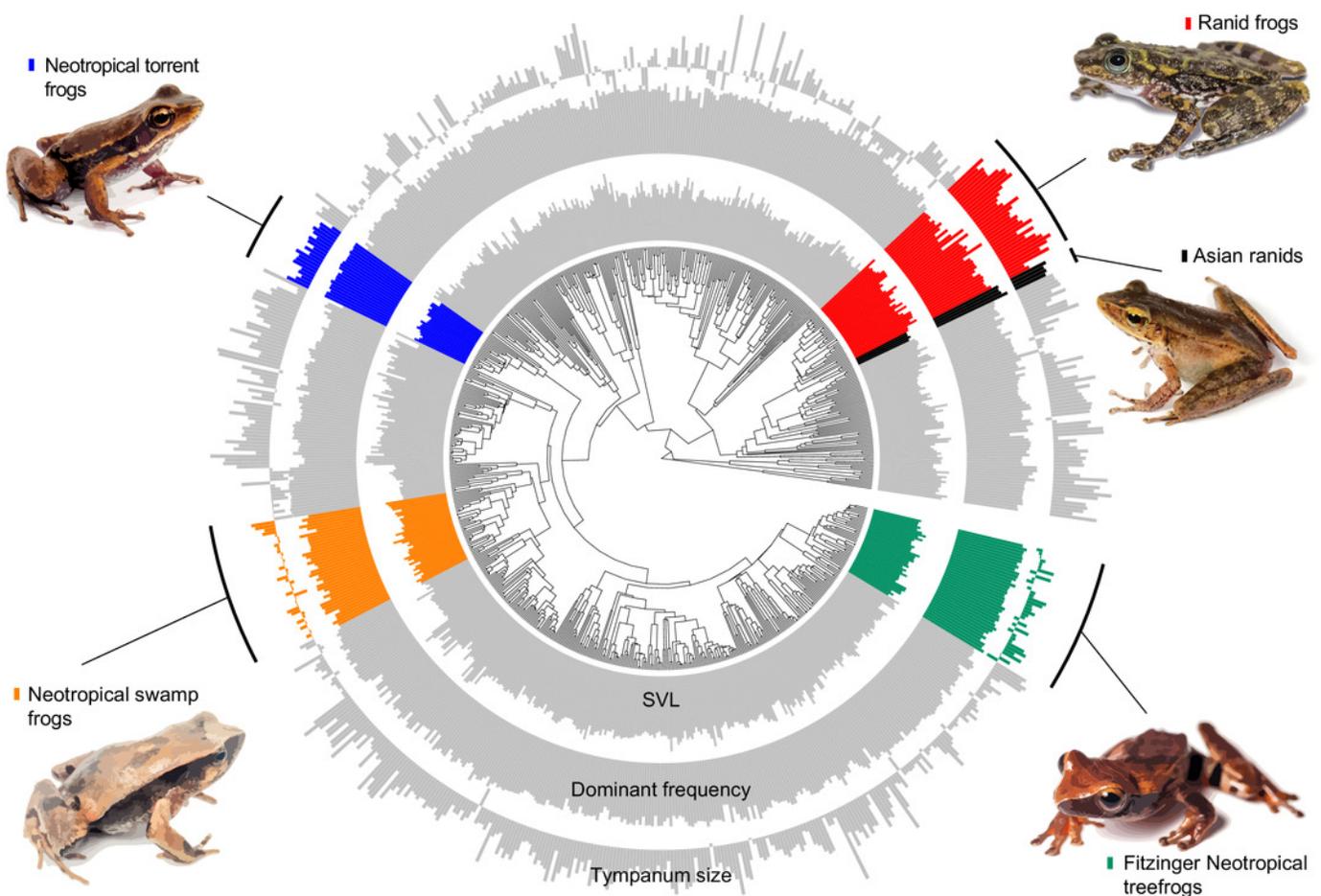
Figure 4

Allometric shifts across anuran phylogeny

Relation between dominant frequency and body size (left chart) and between advertisement call dominant frequency and tympanum size (right chart). The general relationship for all sampled species is in grey, and specific relation are coloured as follows: Asian ranids (black), Ranid frogs (red), Neotropical torrent frogs (blue), Fitzinger Neotropical treefrogs (green) and Neotropical swamp frogs (orange).
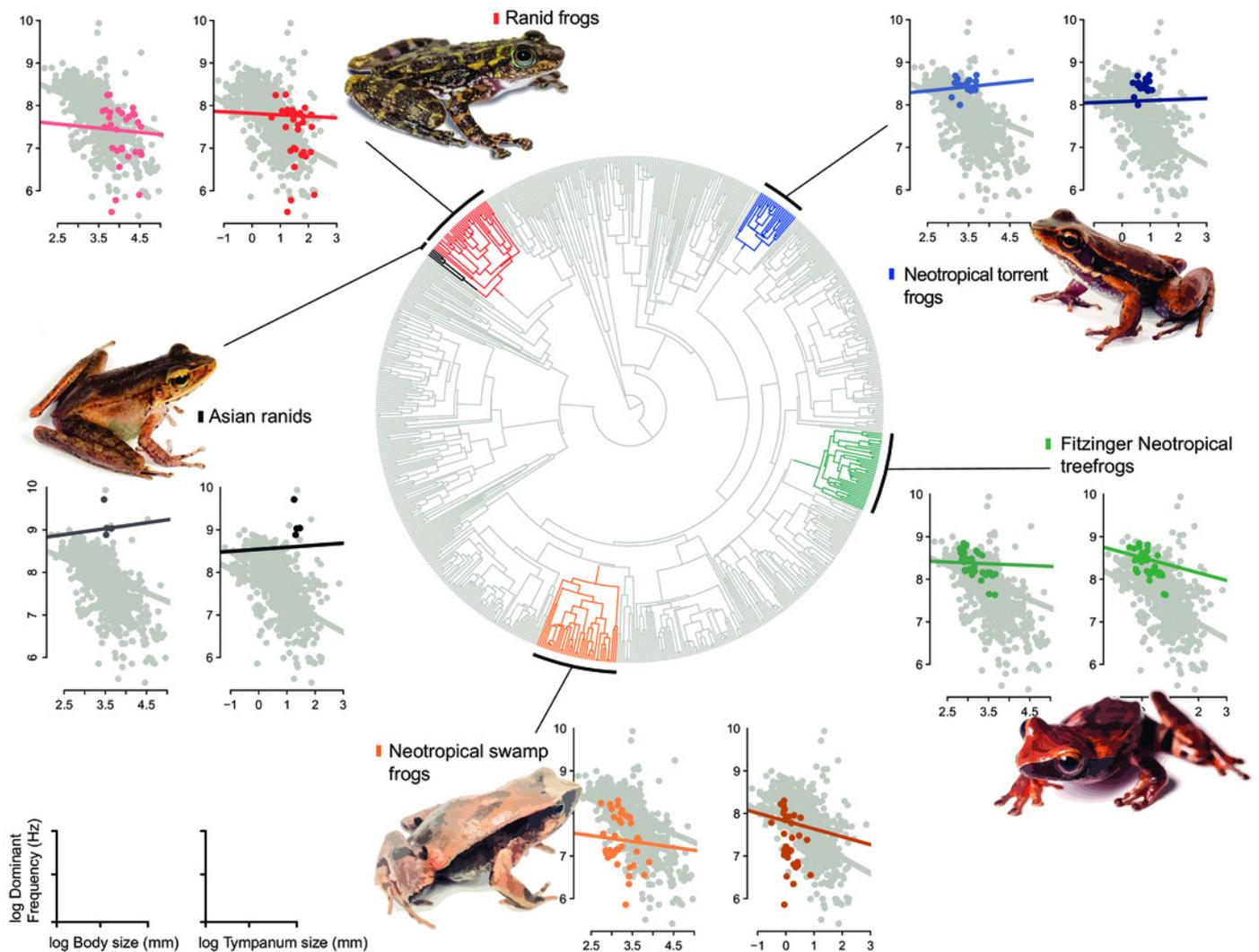
Figure 5

Estimates of intercept and slopes of model

Density plots showing the uncertainty in model parameter estimates of intercept ($\theta_{\text{Dominant frequency}}$) and slopes ($\beta_{\text{Snout-vent length}}$ and $\beta_{\text{Tympanum size}}$) for each escaped lineage: Hylodidae (blue), *Dendropsophus* (green), Leiuperinae (orange), other ranids (red) and *Huia + Meristogenys* (black).