UNIVERSITY OF BATH

University of Bath

Research

Parental care and the evolution of terrestriality in frogs

Balázs Vági[1], Zsolt Végvári[2,3], András Liker[4], Robert P. Freckleton[5], Tamás Székely[6]

[1]Department of Evolutionary Zoology and Human Biology, University of Debrecen, Debrecen, Hungary

[2]Department of Conservation Zoology, University of Debrecen, Debrecen, Hungary

[3]Department Hortobágy National Park Directorate, Debrecen, Hungary

[4]Department of Limnology, University of Pannonia, Veszprém, Hungary,

[5]Department of Animal and Plant Sciences, University of Sheffield, Sheffield, United Kingdom

[6]Milner Centre for Evolution, University of Bath, Bath, United Kingdom

Author for correspondence:

Tamás Székely

T.Szekely@bath.ac.uk

Running title: Comparative analyses of frog care

Subject Category

Evolution

Subject Areas

behaviour, evolution

1   Frogs and toads (Anura) exhibit some of the most diverse parental strategies in vertebrates. Identifying

2   the evolutionary origins of parenting is fundamental to understanding the relationships between sexual

3   selection, social evolution and parental care systems of contemporary Anura. Moreover, parenting has

4   been hypothesized to allow the invasion of terrestrial habitats by the ancestors of terrestrial vertebrates.

5   Using comprehensive phylogenetic analyses of frogs and toads based on data from over 1000 species that

6   represent 46 out of 55 Anura families, we test whether parental care is associated with terrestrial

7   reproduction and several life history traits. Here we show that both the duration of care and offspring

8   protection by males and females have co-evolved with terrestrial reproduction. Sexual size dimorphism is

9   also related to care, since large male size relative to female size is associated with increased paternal care.

10   Furthermore, increased egg size and reduced clutch volume are associated with increased care in bivariate

11   but not in multivariate analyses, suggesting that the relationships between care, egg size and clutch volume

12   are mediated by terrestrial reproduction. Taken together, our results suggest that parenting by males and

13   females has co-evolved, and complex parenting traits have evolved several times independently in Anura in

14   response to breeding in terrestrial environments.

1. Introduction

Parental care is a highly diverse social behaviour that has evolved to increase offspring survival, although it tends to be costly to the caregiving parent [1–3]. Frogs and toads (Anura, hereafter frogs) are characterized by a remarkable diversity of care [4,5] that is rivalled among vertebrates only by the older and more speciose bony fishes [6]. Approximately 10–20% of extant frog species exhibit parental behaviour, with the duration of care, the sex of the care provider and the type of care all showing unique diversity and phylogenetic plasticity [5,7,8].

Understanding the evolutionary origin and maintenance of frog reproductive diversity is important for understanding the adaptive significance of parental care both on evolutionary and ecological time scales. Firstly, parental care tends to increase offspring survival especially in hostile environments [9,10], and thus, it may have played a key role in the colonization of terrestrial habitats, i.e. not only in the evolution of recent amphibians, but also in early tetrapods, opening the way to the subsequent radiation into terrestrial niches [11]. Because parenting is one of the traits linked to expansion into non-aquatic niches [12,13], identifying correlates of care in extant taxa will help us to understand major transitions such as the occupation of terrestrial niches by early tetrapods. Secondly, parental care is an ideal system to understand interactions between individuals that has been extensively investigated in experimental and game-theoretic analyses of social interactions [14–16]. Since parenting influences offspring survival and reproduction, parental decisions often impact on reproductive success and population dynamics [14]. Third, phylogenetic comparative analyses are important to uncover ecological and life-history predictors of parenting: they add a time axis to social interactions and link ecological and evolutionary time scales [8,17,18], although these studies rarely cover a whole order of organisms [but see 19,20].

Frog parental care is immensely diverse, and it includes simple types of care such as constructing a foam nest or attending the eggs, as well as more elaborated forms such as internal brooding of offspring [4,5,12], or cooperation between parents to attend and provide food for the growing offspring [18]. Reproductive modes, i.e. the variation in nesting sites and the environment where tadpoles develop, are also linked to care [5,7,21], although it is not clear whether these associations hold for different care types, e.g. male-only, female-only and/or biparentally caring species, or are relevant only at certain stages of reproduction [13].

Terrestrial environments are hostile for anamniotic eggs, given the high risks of desiccation and exposure to diseases, parasites and predators although predation risk tends to be high in aquatic environments as well. Therefore, egg attendance and egg protection, including urination on the eggs to keep them moist, may considerably increase offspring survival in terrestrial environments [10,22]. In

48 addition, several frogs show extensive post-hatch care by carrying the tadpoles (or froglets) on their

49 backs or in specialized brooding organs [21,23]. Terrestrially reproducing frogs may have endotrophic

50 larvae that develop in a protected chamber, or directly developing embryos which skip larval phase and

51 hatch as fully-developed froglets [5,13]. These offspring rely upon parental provisions until they reach

52 the next stage of their development (e.g., metamorphosis, hatching or birth). Consequently, anurans

53 may enhance offspring care by extending the duration of care, by providing more protection for the

54 offspring and/or by increasing nutrient provisioning in nutrient-scarce environments. These behaviours

55 enable the offspring to spend a longer period of their development in a safe place [18,24,25].

56   Here we investigate three hypothesized drivers of parental care. We focus on the evolution of

57 care by scoring aspects of care on a finer scale and, to our knowledge, we present the most detailed

58 phylogenetic analyses of parenting in any taxa. First, we test whether terrestrial vs aquatic reproduction

59 relates to different care types, since caring is expected to provide protection against hostile

60 environments [5,10,13]. Second, we investigate whether life history variables including egg size and

61 clutch size correlate with the duration of care, protection and nourishment provided by any of the

62 parents. Specifically, we hypothesize that large eggs are associated with longer care and more

63 protection than small eggs [1,5,26]. Third, sexual selection has been linked to parental care since

64 Trivers' [27] seminal idea (reviewed by [1,9,28]), therefore we also investigate whether intense sexual

65 selection is associated with reduced care provisioning [29–31]. We use sexual size dimorphism (SSD)

66 as a proxy for the intensity of sexual selection [30,31]. Note that SSD as an indicator of sexual selection

67 has been debated in frogs, since SSD may reflect selections acting on females, e.g. to increase

68 fecundity [32–34]. Nonetheless, large size in males is associated with high reproductive success in

69 several species of frogs (reviewed by [5,35]) due to competition for mates or female choice [36–39], with

70 the latter processes being clearly linked to sexual selection.

71   To address these objectives, we use a comprehensive dataset that represents 46 out of 55 extant

72 anuran families. We analyse three main components of care: duration of care, protection of eggs and

73 young, and nutrient transfer to offspring. We consider these separately, because complex social traits

74 such as caring may have multiple components that evolve independently, or traded off against each

75 other and thus respond to different selection pressures [40–43]. Second, instead of combining male-only

76 care, female-only care and biparental care into a single variable (for instance, presence or absence of

77 care by either parent), we treat care by males and females separately, since ecological and life-history

78 variables may exert stronger effects on one sex than on the other. For instance, reproductive effort such

79 as egg size and clutch volume may be an important constraint of female care, whereas the intensity of

80 sexual competition may be an important constraint of male care [8,42,43]. Our work demonstrates that

81　these distinctions are important, since some of the relationships between care components and

82　ecological and life history variables differ between males and females.

83

2. Methods

(a) Data collection

86　We compiled the initial dataset from comprehensive phylogenetic comparative publications which

87　contain information on parental care in frogs [8,13,18,26,42,44,45]. Next, we augmented this dataset

88　with data from primary research publications (see Supporting Information), online databases [46,47],

89　and peer-reviewed books [5,48,49]. Our final database holds information from 1044 species; 399 of

90　these species exhibit some form of care. 46 of 55 Anura families are represented in our database that

91　hold approximately 95% of extant species (electronic supplementary material, table S1).

92

(b) Parental care variables

94　We used 4 variables for coding parental care. First, type of care was scored on a five point scale: 0–no

95　care; 1–male-only care; 2–female-only care; 3–biparental care; 4–care either by the male or the female.

96　Because the latter (i.e, uniparental care either by male or female) was reported only from seven

97　species, we excluded these species from the analyses. We considered biparental care if both parents

98　participate in offspring care. In the analysis of the number of care-providing parents, male-only care and

99　female-only care (scores 1 and 2) were combined as uniparental care, whereas score 3 was kept as

100　biparental care.

101　　　　Second, we scored the duration of care based on discrete ontogenetic stages of the offspring

102　(egg, tadpole and juvenile care), and recorded the most advanced stage when a particular caring

103　behaviour has been reported. Care duration was defined as 0–no care; 1–egg care; 2–tadpole care; 3–

104　juvenile care. Care duration was scored separately for males and females.

105　　　　Third, we scored offspring protection as a separate variable on a 6 point scale: 0–no protection;

106　1–offspring protected in a nest but not attended by parent(s); 2–parental attendance; 3–carrying on the

107　back of parent(s); 4–carrying in a closed organ (brooding pouch, dermal invagination, stomach or vocal

108　sack) of parent(s); 5–viviparity. This scoring was based on the logic that protection is more effective

109　when eggs or offspring are enclosed (e.g., in a brooding pouch, stomach, vocal sack, skin invagination)

110　rather than exposed on the back of the parent(s). The highest level of protection appears to be in

111　viviparous species because in these species the offspring only leave the reproductive tract of the mother

112　in a well-developed stage. Protection was scored separately for males and females.

Fourth, nourishment was categorized as follows: 0–exotrophic tadpoles feed mainly on external food sources after depleting their yolk provided in the egg; 1–feeding tadpoles by trophic eggs or skin secretion; 2–endotrophic tadpoles and directly developing species (which complete metamorphosis inside the egg) reach metamorphoses nourishing only upon the egg's yolk. Nourishment was only provided by the female except in two species in which the males provision the offspring (*Ecnomiohyla rabborum, Rhinoderma darwini* [5,50]). Consequently the latter two species were excluded from the analyses of nourishment.

In order to investigate the consistency of our parental care scores with three published datasets that scored parenting as a binary variable (presence/ absence) [13,26,44], we calculated the correlations between these four datasets. The association between our dataset and the three independent datasets were highly significant (electronic supplementary material, table S2).

(c) Life-history variables

Egg size was defined as the diameter of the egg (vitelline) in millimetres, excluding the gelatinous capsule. Clutch size was defined as the number of eggs laid during one egg-laying event. We use clutch volume (calculated as egg volume in cm^3 multiplied by clutch size) instead of clutch size in bivariate analyses, because clutch volume appears to be a more appropriate indicator of female reproductive expenditure than clutch size alone. However, to separate the potential effects of egg size and clutch size in multivariate analyses, we included egg size and clutch size in the models. Snout-vent length (SVL) was calculated separately for males and females, computed as mean values across all available data for a given species. Body size (mean SVL) was calculated as the average of male and female SVLs (in mm) for each species, whereas sexual size dimorphism was $\log_{10} (SVL_{male} / SVL_{female})$. Clutch size, clutch volume and egg size were transformed to logarithmic scale to ensure homoscedasticity. If several data points were available for a given species, we calculated their arithmetic mean.

Terrestrial reproduction and direct development were treated as binary variables (present or absent), following previous classifications [13,21]. Terrestrial reproduction included floating foam nest on water, as in this case the eggs themselves are included in an air-filled chamber, and also viviparity and egg-brooding in different organs (pouches, stomach, vocal sac) provided by terrestrial parents. In contrast, members of the genus *Pipa* which lay eggs in water and brood by aquatic parents were considered aquatic breeders. We established these categories because anuran eggs are adapted primarily to aquatic development and placing them outside water exposing them to hostile conditions, and we considered the strategy for this challenge as an important aspect of parental care.

(d) Phylogeny

We used a comprehensive amphibian phylogenetic tree (the consensus tree from [51]) which included the majority of species in our database. Archaeobatrachians were treated as all anurans outside the Neobatrachia clade, and basal Neobatrachians as all Neobatrachians outside the Hyloidea and Ranoidea clade (figure 1 and electronic supplementary material, figure S1). In figure 1a–c we used Grafen-transformed branch lengths for better visualisation.

Anuran phylogenies tend to hold consistent patterns, at least in the topology of deeper nodes [51,52]. Since most variation in care is between genera and families, our results appear to be robust to different phylogenetic hypotheses. Nonetheless, to check the sensitivity of our results to alternative phylogenies, we re-analysed the major models using an alternative tree: a composite tree based on [53]. We augmented the latter tree [53] with 145 additional species inserted next to their closest species (whenever known), based on recent phylogenetic information. Nodes were collapsed to polytomies when no further information was available on the phylogenetic relationships within a genus. The species we added manually are listed in electronic supplementary material, table S7, along with the references for their phylogenetic relationships. We use the branch lengths of the original trees [51,53]. In composite phylogeny we assumed half branch length for the new species we included using 'phytools' package [54] in R 3.1.0 [55]. Importantly, the results using the alternative phylogeny were highly consistent with those of the main phylogeny (see table 1, electronic supplementary material, tables S3–S6).

(e) Comparative analyses

We tested associations between parental care and life history variables using Phylogenetic Least Squares (PGLS) [56–58]. This approach controls for the non-independence among species by incorporating a variance–covariance matrix that represents their phylogenetic relationships. All analyses incorporated phylogenetic dependence by estimating Pagel's λ [58]. We built separate multipredictor PGLS models for each parental care variable (i.e., care duration by females, care duration by males; protection by females, protection by males, nourishment by females) in which one of the care variables was the dependent variable, and log clutch size, log egg size, average SVL, sexual dimorphism, terrestrial reproduction and direct development were the predictors.

We also included the higher nodes (i.e., superfamily ID, see supporting data s2) as a factor in PGLS models [53,59]. This was to control for the lack of variation in key traits within higher taxa: for traits that do not vary within higher nodes, the effective level of replication and appropriate degrees of freedom can be questioned. Due to the lack of variation within clades, three species-poor lineages ('Crown Hyloidea' that includes Alsodidae, Ceratophryidae, Hylodidae, Odontophrynidae and

7

Rhinodermatidae, 12 species in total; Heleophrynidae, 2 species; and Sooglossoidea, 3 species) were excluded from analyses that included higher node as factor. Higher nodes were not included in analyses on trophic egg feeding (Nourishment excluding species in Nourishment category 2) – in this case, most of the clades showed little variance to the trait.

We tested multicollinearity between predictors using variance inflation factor (VIF) analysis: all predictors had VIF values less than 5 (VIF_{max} = 2.02). In multiple regression models, we included six predictor variables (see table 1) except in models of nourishment we did not include developmental mode since nourishment and developmental mode were correlated by definition. All analyses were carried out using R 3.1.0 [55] with 'caper' package [60].

3. Results

Types of care varied across Anura, with each type of care occuring in several clades (figure 1; electronic supplementary material, table S1). Major clades exhibited substantial variations in sex of care provider, protection and nourishment (figure 1): exceptional diversity was exhibited by five clades that include Eleutherodactylidae, Dendrobatidae, Leptodactylidae and Microhylidae, figure S1).

Care duration, protection and nourishment were not different between species with female-only care, male-only care and biparental care (Phylogenetic Generalised Least Squares PGLS, care duration: figure S2, $F_{2,379}$ = 0.716; p = 0.489; protection: $F_{2,375}$ = 0.502; p = 0.610; nourishment: $F_{2,370}$ = 0.502; p = 0.426), nor between uniparental and biparental species (PGLS, care duration: $F_{1,387}$ = 0.415; p = 0.520; protection: $F_{1,382}$ = 0.788; p = 0.375; nourishment: $F_{1,378}$ = 1.694; p = 0.194). Thus, males and females provide similar extents of care in anurans. Interestingly, the extent of parental care by males was associated with the extent of female care both in care duration (PGLS; $F_{1,1006}$ = 8.674; p < 0.0001) and protection ($F_{1,1005}$ = 54.58; p < 0.0001).

Terrestrial reproduction was a key factor associated with parental care (figure 1). All forms of care were more common in terrestrial taxa than in aquatic ones (figure 2) including protection by males (5.5% and 46.5% of aquatic and terrestrial taxa, respectively), protection by females (1% and 39.0%), and nourishment (5.0% and 34.5%). Terrestrial reproduction was associated with increased levels of care by both males and females (figure 2 and electronic supplementary material, table S3). Consequently, the number of caring parents was significantly higher in terrestrial frogs than in aquatic ones (PGLS; $F_{1,591}$ = 80.47; p < 0.0001).

Large eggs and small clutches were associated with extended parenting and protection by both sexes, and provisioning by the female (figure 3 and electronic supplementary material, table S4). However, since egg size and clutch volume often depend on body size, we also investigated the

212 relationship between egg size, clutch volume and care by including body size as an explanatory variable

213 in phylogenetically corrected models (table S5). When body size was statistically controlled for, neither

214 egg size nor clutch volume remained correlated with care with the exception of nourishment, and small

215 clutch volume remained associated with male care (electronic supplementary material, table S5).

216 Sexual size dimorphism was associated with male care but not female care (electronic

217 supplementary material, table S4 and figure S3). However, male care was associated with increased

218 male size relative to female size (table S4). The latter relationship remained significant when absolute

219 body size was controlled for in the analysis (table S5). The latter relationship between size dimorphism

220 and body size suggests that Anura exhibit an allometric relationship between sizes of males and

221 females known as Rensch's rule [44,61] (PGLS; $F_{1,430}$ = 7.39; p = 0.007).

222 Terrestrial reproduction remained the main predictor of both care duration and offspring

223 protection in multipredictor analyses, but not for nourishment (table 1). These results suggest that the

224 relationships between life history and care we uncovered using bivariate analyses (electronic

225 supplementary material, table S4) may be mediated by terrestrial reproduction. Nevertheless, in multi-

226 predictor models male-biased size dimorphism remained associated with male care (table 1), and

227 nourishment remained associated with clutch size and body size.

228 Trophic egg feeding (i.e, exotrophic tadpoles feed on external food sources versus tadpoles fed

229 by trophic eggs or skin secretion) was associated with sexual dimorphism and clutch volume (electronic

230 supplementary material, tables S3–S4), and these relationships remained significant after controlling for

231 body size (table S5, S6).

232

4. Discussion

234 Our comprehensive phylogenetic analyses of the extent of male and female care show that care is

235 extremely variable both within and among major clades of frogs. Not only the presence or absence of

236 care varies – that has been uncovered by previous studies [18,26] – but also the type and duration of

237 care are highly variable. In contrast to reptiles and mammals, in which the females are the main care

238 provider, or to birds in which biparental care is the predominant form of care [9,62], in frogs female-only,

239 male-only and biparental care are all widespread among various lineages, and the involvement of males

240 and females in care is comparable. Because in ~20% of newts and salamanders (urodeles) one of the

241 parents guards the eggs or the offspring [5,9,63,64], and caecilians in which females feed their offspring

242 using an excretion of their skin [65,66], the overall richness of caring is spectacular in amphibians. This

243 suggests that over the course of amniote evolution, the phylogenetically younger tetrapod clades (e.g.,

244 reptiles, birds and mammals) became specialised to a limited set of care patterns [62].

245   Consistently with previous studies [11–13], we found that the transition towards terrestrial

246   reproduction facilitated parental care. Moreover, our work advances the understanding of evolutionary

247   relationships by showing that terrestrial reproduction is related to all forms of both male and female

248   care, except nourishment. Thus, when early tetrapods invaded terrestrial niches, both males and

249   females may have been under the effects of selection forces to improve the survival of their offspring, so

250   that both males and females evolved various forms of care provisioning in response to terrestrial

251   reproduction. Therefore, the subsequent canalization of parental care largely towards females (e.g., in

252   reptiles and mammals) and cooperation by both sexes (in birds) may have been the result of additional

253   selective pressures that the ancestors of these clades faced during their radiation into various ecological

254   niches. This implies that the predominance of maternal care coevolved with internal fertilization [67, but

255   see 68]. In urodeles, where internal fertilization is more frequent, only phylogenetically basal external

256   fertilizers with aquatic reproduction appear to provide paternal care [5,63], although clutch attending by

257   females is widespread especially in those with terrestrial reproduction [63].

258   We also found that egg size and clutch volume are related to parental care, although these

259   associations became non-significant by including terrestriality in the models. On the one hand, terrestrial

260   egg-layers have larger eggs and smaller clutches than aquatically reproducing frogs [8,13,26], which

261   may be predicted by other factors besides parental care, such as selection on offspring size [69] or

262   protection against the hostile environment [11]. However, egg size and clutch size were no longer

263   associated with care duration and protection when body size was statistically controlled. Therefore, the

264   associations between egg size, clutch size and parenting showed by previous studies [8,13,26] may

265   have been mediated by other factors, e.g. body size and/or terrestrial reproduction. On the other hand,

266   increased nutrient transfer to the offspring is associated with reduced clutch size, which seems to be the

267   result of an increased investment to individual offspring [3] traded off against fecundity. Moreover,

268   trophic egg feeding is also associated with reduced egg size [table S6], implying that mothers may

269   reduce the cost egg production using this type of nourishment.

270   Finally, the evolutionary relationship between male care and size dimorphism has been debated

271   [32-34], and our results using fine-scaled care variables, multi-predictor models and more extensive

272   taxonomic coverage than previous studies, confirm that male care is associated with sexual size

273   dimorphism [44]. We suggest two mutually non-exclusive explanations for the increased male size

274   (relative to female size) with the extent of male care. On the one hand, sexual selection may favour

275   larger males in male caring species if females prefer large males and/or large males are more

276   successful in coercive mating [38,39], provided that these males are more successful in nursing the

277   offspring. On the other hand, male care may reduce the fecundity selection pressure on females, so that

female size decreases in those species in which the males provide care [44,61]. To distinguish between these scenarios, further experimental and phylogenetic analyses are warranted [9,17].

Here we treat parental care as an invariable trait for a given species, although this assumption suits some species better than others. For example, *Allobates femoralis* exhibits variation in parenting since females transport tadpoles but this behaviour is only provoked by the absence of the father that is normally the care-providing parent [41]. Therefore, future phylogenetic analyses should pay attention to the flexibility of care provisioning [41,70]. Care provision can be further tuned by variation in the ecological [25,71,72] or social environment [41], and this plasticity not only enables better adaptation to seasonal and unpredictable changes of the environment, but it may also act as the origin of evolutionary changes in the extent of care [41,45] or in parental roles [41,43,45]. Field-based and laboratory-based studies will likely add more examples for this plasticity and would help in identifying environmental factors which provokes shifts.

In summary, parental care is predicted by ecological and life history variables in frogs. Care is a complex social trait and specific aspects of care have different predictors in males and females. Further analyses are needed to investigate the impacts of climate, reproductive modes and mating systems on care strategies. Since new forms of parental care are cropping up [71,72], field-based studies of yet unstudied species are needed to explore breeding systems (including parenting) in frogs that live in remote areas and/or inhabit extreme environments. Taken together, studies of anuran parental care provide important contributions to the understanding of reproduction, evolution and diversification in the most threatened vertebrate class of the Anthropocene.

References

1. Clutton-Brock TH. 1991 *The evolution of parental care.* Princeton: Princeton Univ. Press.

312   2. Székely T, Moore AJ, Komdeur J. 2010 *Social behaviour: genes, ecology and evolution.*
313   Cambridge: Cambridge Univ. Press.

314   3. Smiseth PT, Kölliker M, Royle NJ. 2012 What is parental care? In *The evolution of parental care*
315   (eds Royle NJ, Smiseth PT, Kölliker M), pp. 1 – 17. Oxford: Oxford Univ. Press.

316   4. Crump ML. 1995 Parental care. In: *Amphibian biology, volume 2: Social behavior* (eds
317   Heathvole H, Sullivan BK), pp. 518 – 567. Chipping Norton: Surrey Beatty & Sons.

318   5. Wells KD 2007. Parental care. In *The ecology and behaviour of amphibians* (Wells KD).
319   Chicago and London: Univ. of Chicago Press, pp. 516 – 556.

320   6. Mank JE, Promislow DEL, Avise JC 2005 Phylogenetic perspectives in the evolution of parental
321   care in ray-finned fishes. *Evolution* **59**, 1570 – 1578. (doi: 10.1554/04-734)

322   7. Duellman WE, Trueb L 1986 *Biology of amphibians*. New York: McGraw-Hill.

323   8. Summers K, McKeon CS, Heying H, Patrick W. 2007 Social and environmental influences on
324   egg size evolution in frogs. *J. Zool.* **271**, 225 – 232. (doi: 10.1111/j.1469-7998.2006.00213.x)

325   9. Balshine S. 2012 Patterns of parental care in vertebrates. In *The evolution of parental care* (eds
326   Royle NJ, Smiseth PT, Kölliker M). Oxford: Oxford Univ. Press, pp. 62 – 80.

327   10. Poo S, Bickford DP. 2013 The adaptive significance of egg attendance in a South-East Asian
328   tree frog. *Ethology* **119**, 1 – 9. (doi: 10.1111/eth.12108)

329   11. Martin KL, Carter AL 2013 Brave new propagules: terrestrial embryos in anamniotic eggs.
330   *Integr. Comp. Biol.* **53**, 233 – 247. (doi: 10.1093/icb/ict018)

331   12. Crump ML. 1996 Parental care among the Amphibia. *Adv. Stud. Behav.* **25**, 109 – 144. (doi:
332   10.1016/S0065-3454(08)60331-9)

333   13. Gomez-Mestre I, Pyron RA, Wiens JJ. 2012 Phylogenetic analyses reveal unexpected patterns
334   in the evolution of reproductive modes in frogs. *Evolution* **66**, 3687 – 3700. (doi: 10.1111/j.1558-
335   5646.2012.01715.x)

336   14. McNamara JM, Weissing FJ. 2010 Evolutionary game theory. In: *Social behaviour: genes,*
337   *ecology and evolution* (eds Székely T, Moore AJ, Komdeur J), pp. 88 – 106. Cambridge:
338   Cambridge Univ. Press.

339   15. Houston AI, Székely T, McNamara JM. 2013 The parental investment models of Maynard
340   Smith: a retrospective and prospective view. *Anim. Behav.* **86**, 667 – 674. (doi:
341   10.1016/j.anbehav.2013.08.001)

342   16. Barta Z, Székely T, Liker A, Harrison F. 2014 Social role specialization promotes cooperation
343   between parents. *Am. Nat.* **183**, 747 – 761. (doi: 10.1086/676014)

344   17. Székely T, Reynolds JD 1995 Evolutionary transitions in parental care in shorebirds. *Proc. R.*
345   *Soc. B* **262**, 57 – 64. (doi: 10.1098/rspb.1995.0176)

346   18. Brown JL, Morales V, Summers K. 2010 A key ecological trait drove the evolution of biparental

347   care and monogamy in an amphibian. *Am. Nat.* **175**, 436 – 446. (doi: 10.1086/650727)

348   19. West HER, Capellini I 2016 Male care and life hictory traits in mammals. *Nat. Commun.* **7**,

349   11854. (doi: 10.1038/ncomms11854)

350   20. Remes V, Freckleton R, Tökölyi J, Liker A, Székely T 2015 The evolution of parental

351   cooperation in birds. *PNAS* **112**, 12603 – 13608. (doi: 10.1073/pnas.1512599112)

352   21. Haddad CFB, Prado CPA. 2005 Reproductive modes in frogs and their unexpected diversity in

353   the Atlantic Forest of Brazil. *BioScience* **55**, 208 – 217. (doi: 10.1641/0006-

354   3568(2005)055[0724:RMIFC]2.0.CO;2)

355   22. Bickford DP. 2004 Differential parental care behaviors of arboreal and terrestrial microhylid

356   frogs from Papua New Guinea. *Behav. Ecol. Sociobiol.* **55**, 402 – 409. (doi: 10.1007/s00265-

357   003-0717-x)

358   23. Köhler F, Günther R. 2008 The radiation of microhylid frogs (Amphibia: Anura) on New Guinea:

359   A mitochondrial phylogeny reveals parallel evolution of morphological and life history traits and

360   disproves the current morphology–based classifications. *Mol. Phylogenet. Evol.* **47**, 353 – 365.

361   (doi: 10.1016/j.ympev.2007.11.032)

362   24. Shine R. 1978 Propagule size and parental care: the "safe harbour" hypothesis. *J. Theor. Biol.*

363   **75**, 417 – 424. (doi: 10.1016/0022-5193(78)90353-3)

364   25. McKeon CS, Summers K. 2013 Predator driven reproductive behavior in a tropical frog. *Evol.*

365   *Ecol.* **27,** 725 – 737. (doi: 10.1007/s10682-013-9641-3)

366   26. Summers K, McKeon CS, Heying H. 2006 The evolution of parental care and egg size: a

367   comparative analysis in frogs. *Proc. R. Soc. B* **273**, 687 – 692. (doi: 10.1098/rspb.2005.3368)

368   27. Trivers RL. 1972 Parental investment and sexual selection. In *Sexual selection and the descent*

369   *of man* (ed Campbell B), pp. 136 – 179. Chicago: Aldine-Atherton.

370   28. Klug H, Alonso SH, Bonsall MB. 2012 Theoretical foundations of parental care. In *The evolution*

371   *of parental care* (eds Royle NJ, Smiseth PT, Kölliker M), pp. 21 – 39. Oxford: Oxford Univ.

372   Press.

373   29. Queller DC. 1997 Why do females care more than males? *Proc. R. Soc. Lond. B* **264**, 1555 –

374   1557. (doi: 10.1098/rspb.1997.0216)

375   30. Jennions MD, Kokko H. 2010 Sexual selection. In *Evolutionary behavioral ecology* (eds

376   Westneat DE, Fox CW), pp. 343 – 364. Oxford: Oxford Univ. Press.

377   31. Janicke T, Häderer IK, Lajeunesse MJ, Anthes N. 2016 Darwinian sex roles confirmed across

378   the animal kingdom. *Sci. Adv.* **2**, e1500983. (doi: 10.1126/sciadv.1500983)

379   32. Monnet JM, Cherry MI. 2002 Sexual size dimorphism in anurans. *Proc. R. Soc. Lond. B* **269**,
380   2301 – 2307. (doi: 10.1098/rspb.2002.2170)

381   33. Kupfer A. 2007 Sexual size dimorphism: an overview. In *Sex, size and gender roles:*
382   *evolutionary studies of sexual size dimorphism* (eds Fairbairn DJ, Blanckenhorn WU, Székely
383   T), pp. 50 – 59. Oxford: Oxford Univ. Press.

384   34. Monroe MJ, Alonzo SH. 2014 Sexual size dimorphism is not associated with the evolution of
385   parental care in frogs. *Ecol. Evol.* **4**, 3991 – 3998. (doi: 10.1002/ece3.1263)

386   35. Arak A. 1983 Male-male competition and mate choice in anuran amphibians. In *Mate choice* (ed
387   Bateson P), pp. 181 – 205. Cambridge: Cambridge Univ. Press.

388   36. Davies NB, Halliday TR 1979. Competitive mate searching in male common toads, *Bufo bufo*.
389   *Anim. Behav.* **27**, 1253 – 1267. (doi: 10.1016/0003-3472(79)90070-8)

390   37. Ryan MJ. 1980 Female mate choice in a neotropical frog. *Science* **209**, 523 – 525. (doi:
391   10.1126/science.209.4455.523)

392   38. Rausch AM, Sztatecsny M, Jehle R, Ringler E, Hödl W. 2014 Male body size and parental
393   relatedness, but not nuptial colouration influence paternity success during scramble competition
394   in *Rana arvalis*. *Behaviour* **151**, 1869 – 1884. (doi: 10.1163/1568539X-00003220)

395   39. Vági B, Hettyey A. 2016 Intraspecific and interspecific competition for mates: *Rana temporaria*
396   males are effective satyrs of *Rana dalmatina* females. *Behav. Ecol. Sociobiol.* **70**, 1477 – 1484.
397   (doi: 10.1007/s00265-016-2156-5)

398   40. Székely T, Remeš V, Freckleton RP, Liker A. 2013 Why care? Inferring the evolution of complex
399   social behaviour. *J. Evol. Biol.* **26**, 1381 – 1391. (doi: 10.1111/jeb12148)

400   41. Ringler E, Pašukonis A, Tecumseh Fitch W, Huber L, Hödl W, Ringler M. 2015 Flexible
401   compensation of uniparental care: female poison frogs take over when males disappear. *Behav.*
402   *Ecol.* **26**, 1219 – 1225. (doi: 10.1093/beheco/arv069)

403   42. Zamudio KR, Bell RC, Nali RC, Haddad CFB, Prado CPA. 2016 Polyandry, predation and the
404   evolution of frog reproductive modes. *Am. Nat.* **188**, S41 – S61. (doi: 10.1086/687547)

405   43. Goyes Vallejos J, Grafe TU, Ahmad Sah HH, Wells KD. 2017 Calling behavior of males and
406   females of a Bornean frog with male parental care and possible sex role reversal. *Behav. Ecol.*
407   *Sociobiol.* **71**, 95. (doi: 10.1007/s00265-017-2323-3)

408   44. Han X, Fu J. 2013 Does life history shape sexual size dimorphism in anurans? A comparative
409   analysis. *BMC Evol. Biol.* **13**, 27. (doi: 10.1186/1471-2148-13-27)

410   45. Delia J, Bravo-Valencia L, Warkentin KM. 2017 Patterns of parental care in neotropical
411   glassfrogs: fieldwork alters hypotheses of sex-role evolution. *J. Evol. Biol.* **30**, 898 – 914. (doi:
412   10.1111/jeb.13059)

413   46. Amphibiaweb 2018 [cited 2018 March 18]. Berkeley: University of California. Available from
414   https://www.amphibiaweb.org.

415   47. IUCN. 2018 *The IUCN Red List of Threatened Species.* Version 2018-1 [cited 2018 March 18].
416   Available from https://www.iucnredlist.org.

417   48. Stuart SN, Hoffmann M, Chanson JS, Cox NA, Berridge RJ, Ramani P et al., eds 2008
418   *Threatened amphibians of the world.* Barcelona: Lynx Edition.

419   49. Glaw F, Vences M. 2007 *A field guide to the amphibians and reptiles of Madagascar*. Cologne:
420   Vences & Glaw Verlags GbR.

421   50. Hansen RW. 2012 About our cover: *Ecnomiohyla rabborum*. *Herpetological Review* **42**, 3.

422   51. Jetz W, Pyron RA. 2018 The interplay of past diversification and evolutionary isolation with
423   present imperilment across the amphibian tree of life. *Nat. Ecol. Evol.* (doi: 10.1038/s41559-
424   018-0515-5)

425   52. Feng YJ, Blackburn DC, Liang D, Hillis DM, Wake DB, Cannatella DC et al. 2017
426   Phylogenomics reveal rapid, simultaneous diversification of three major clades of Gondwanan
427   frogs at the Cretaceous–Paleogene boundary. *PNAS* **114**, E5864 – E5870. (doi:
428   10.1073/pnas.1704632114)

429   53. Pyron RA, Wiens JJ. 2011 A large-scale phylogeny of Amphibia including over 2800 species,
430   and a revised classification of extant frogs, salamanders, and caecilians. *Mol. Phylogenet. Evol.*
431   **61**, 543 – 583. (doi: 10.1073/pnas.1704632114)

432   54. Revell LJ 2012 phytools: an R package for phylogenetic comparative biology (and other things).
433   *Methods Ecol. Evol.* **3,** 217 – 223. (doi: 10.1111/j.2041-210X.2011.00169.x)

434   55. R-Core-Team R 2013 Version 3.1.0 [cited 18 March 2018]. *A language and environment for*
435   *statistical computing*. Available from: http://www.r-project.org/.

436   56. Martins EP, Hansen TF. 1997 Phylogenies and the comparative method: A general approach to
437   incorporating phylogenetic information into the analysis of interspecific data. *Am. Nat.* **149**, 646
438   – 667. (doi: 10.1086/303188)

439   57. Pagel M. 1999 Inferring the historical patterns of biological evolution. *Nature* **401**, 877 – 884.
440   (doi: 10.1038/44766)

441   58. Freckleton RP, Harvey PH, Pagel M. 2002 Phylogenetic analysis and comparative data: a test
442   and review of evidence. *Am. Nat.* **160**, 712 – 726. (doi: 10.1086/343873)

443   59. Frost DR, Grant T, Faivovich JN, Bain RH, Haas A, Haddad CFB et al. 2006 The amphibian
444   tree of life. *Bulletin of the American Museum of Natural History* **297,** 1 – 291. (doi:
445   10.1206/0003-0090(2006)297[0001:TATOL]2.0.CO;2)

446 60. Orme D. 2013 *The caper package: comparative analysis of phylogenetics and evolution in R*. R

447   package version, 5 [cited 18 March 2018]. Available from: https://cran.r-

448   project.org/web/packages/caper/

449 61. Fairbairn DJ, Blanckenhorn WU, Székely T. 2007 *Sex, size and gender roles: evolutionary*

450   *studies of sexual size dimorphism.* Oxford: Oxford Univ. Press.

451 62. Reynolds JD, Goodwin NB, Freckleton RP. 2002 Evolutionary transitions in parental care and

452   live-bearing in Vertebrates. *Phil. Trans. Roy. Soc.* **357,** 269 – 281. (doi:

453   10.1098/rstb.2001.0930)

454 63. Nussbaum RA 1985 *The evolution of parental care in salamanders*. Ann Arbor: The University

455   of Michigan Press.

456 64. Tóth Z, Hoi H, Hettyey A 2011 Intraspecific variation in the egg-wrapping behaviour of female

457   smooth newts, Lissotriton vulgaris. *Amphibia-Reptilia* **32:**77 – 82. (doi:

458   10.1163/017353710X543001)

459 65. Kupfer A, Müller H, Antoniazzi AM, Jared C, Greven H, Nussbaum RA et al. 2006 Parental

460   investment by skin feeding in a caecilian amphibian. *Nature* **440,** 926 – 929. (doi:

461   0.1038/nature04403)

462 66. Kupfer A, Maxwell E, Reinhard S, Kuehnel S 2016 The evolution of parental investment in

463   caecilian amphibians: a comparative approach. *Biol. J. Linn. Soc.* **119,** 4 – 14. (doi:

464   10.1111/bij.12805)

465 67. Gross MR, Shine R 1981 Parental care and the mode of fertilization in ectothermic vertebrates.

466   *Evolution* **35**, 775-793. (doi: 10.1111/j.1558-5646.1981.tb04937.x)

467 68. Beck CW 1998 Mode of fertilization and parental care in anurans. *Anim. Behav.* **55**, 439 – 449.

468   (doi: 10.1006/anbe.1997.0619)

469 69. Nussbaum RA. 1987 Parental care and egg size in salamanders: An examination of the safe

470   harbor hypothesis. *Res. Popul. Ecol.* **29,** 27 (doi: 10.1007/BF02515423)

471 70. Simon MP. 1983 The ecology of parental care in a terrestrial breeding frog from New Guinea.

472   *Behav. Ecol. Sociobiol.* **14**, 61 – 67. (doi: 10.1007/BF00366657)

473 71. Poelman EH, van Wijngaarden RPA, Raaijmakers CE. 2013 Amazon poison frogs (*Ranitomeya*

474   *amazonica*) use different phytothelm characteristics to determine their suitability for egg and

475   tadpole deposition. *Evol. Ecol.* **27**, 661 – 647. (doi: 10.1007/s10682-013-9633-3)

476 72. Schulte LM, Lötters S. 2013 The power of the seasons: rainfall triggers parental care in poison

477   frogs. *Evol. Ecol.* **27**, 711 – 723. (doi: 10.1007/s10682-013-9637-z)

478   73. Gururaja KV, Dinesh KP, Priti H, Ravikath G. 2014 Mud-packing frog: a novel breeding
479   behaviour and parental care in a stream-dwelling new species of *Nyctibatrachus* (Amphibia,
480   Anura, Nyctibatrachidae). *Zootaxa* **3796,** 033 – 061. (doi: 10.11646/zootaxa.3796.1.2)
481   74. Iskandar DT, Evans BJ, McGuire AJ. 2014 A novel reproductive mode in frogs: a new species
482   of fanged frog with internal fertilization and birth of tadpoles. *PLoS ONE* **9**, e115884. (doi:
483   10.1371/journal.pone.0119988)
484

Figure legends

Figure 1. Phylogenetic distribution of parental care and breeding habitat in frogs. (a) Type of care (592 species). (1) Alytidae (*Alytes* sp., male egg transport), (2) Pipidae (*Pipa* sp., eggs embedded in the dorsal skin of female), (3) Hemisotidae (*Hemisus* sp, tadpole guarding by the female), (4) Microhylidae (*Sphenophryne cornuta*, juvenile transport by the male), (5) Rhacophoridae (*Rhacophorus* sp., foam nest made by both parents), (6) Dicroglossidae (*Limnonectes larvaepartus*, viviparity: live birth to larvae), (7) Limnodynastidae (*Limnodynastes peronii*, foam nest made by the female), (8) Myobatrachidae (*Assa darlingtoni*, male carry tadpoles in inguinal pouches), (9) Eleutherodactylidae (*Eleutherodactylus coqui*, direct developing eggs guarded by the male), (10) Hemiphractidae (*Flectonotus* sp., eggs carried in dorsal pouch of the female), (11) Hylidae (*Hypsiboas boans*, male guard eggs in constructed mud pool), (12) Rhinodermatidae (*Rhinoderma darwini*, tadpoles reared in vocal sac of the male), (13) Leptodactylidae (*Leptodactylus podicipinus*, the pair constructs the foam nest, the female guard the tadpoles), (14) Dendrobatidae (*Ranitomeya imitator*, the male transports tadpoles, the female feeds tadpoles in cooperation with the male), (16) Bufonidae (*Nimbaphrynoides* sp., viviparity: live birth to toadlets). (b) Diversity of female care (care duration, protection and nourishment, 594 species). (c) Diversity of male care (care duration and protection, 593 species). Grafen-transformed branch lengths are shown. 0 refers to no care in a particular trait, whereas 3, 5 and 2 refer to the most advanced stage in offspring development in care duration, protection (for males and females separately) and nourishment (for females), respectively.

Figure 2. Care duration, offspring protection and nourishment in relation to aquatic and terrestrial reproduction in frogs. Number of species exhibiting different extent of care duration, offspring protection and nourishment (on the left) and the extent of female and male parental care in aquatic and terrestrial species (mean + SD; on the right). Red shades represent female care, blue shades represent male care.

Figure 3. Parental care in relation to life histories in frogs. Egg size and clutch volume are plotted against offspring care, protection and nourishment in females (red) and males (blue, *see statistics in electronic supplementary material, table S4–S5*). The variables were scored as follows. Care duration: 0–no care; 1–egg care; 2–tadpole care; 3–juvenile care; Protection: 0–no protection; 1–nest building; 2–attending; 3–carrying on back; 4–carrying in a closed organ; 5–viviparity; Nourishment: 0–exotrophic tadpoles; 1–trophic egg feeding; 2–endotrophic tadpoles, direct development or viviparity.
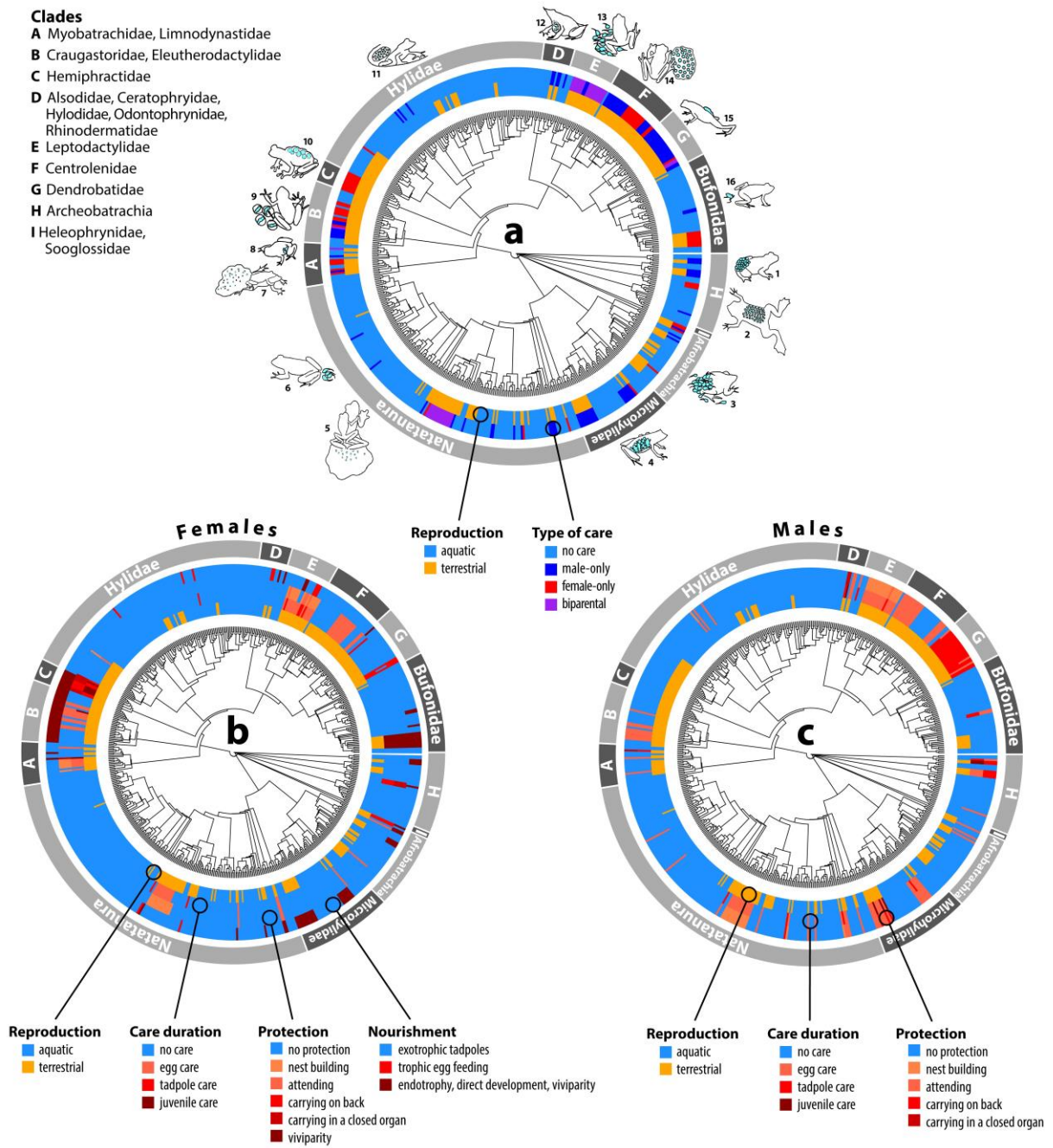
518
519 **Figure 1**

Clades
A Myobatrachidae, Limnodynastidae
B Craugastoridae, Eleutherodactylidae
C Hemiphractidae
D Alsodidae, Ceratophryidae, Hylodidae, Odontophrynidae, Rhinodermatidae
E Leptodactylidae
F Centrolenidae
G Dendrobatidae
H Archeobatrachia
I Heleophrynidae, Sooglossidae

Females

Males

Reproduction
aquatic
terrestrial

Type of care
no care
male-only
female-only
biparental

Reproduction
aquatic
terrestrial

Care duration
no care
egg care
tadpole care
juvenile care

Protection
no protection
nest building
attending
carrying on back
carrying in a closed organ
viviparity

Nourishment
exotrophic tadpoles
trophic egg feeding
endotrophy, direct development, viviparity

Reproduction
aquatic
terrestrial

Care duration
no care
egg care
tadpole care
juvenile care

Protection
no protection
nest building
attending
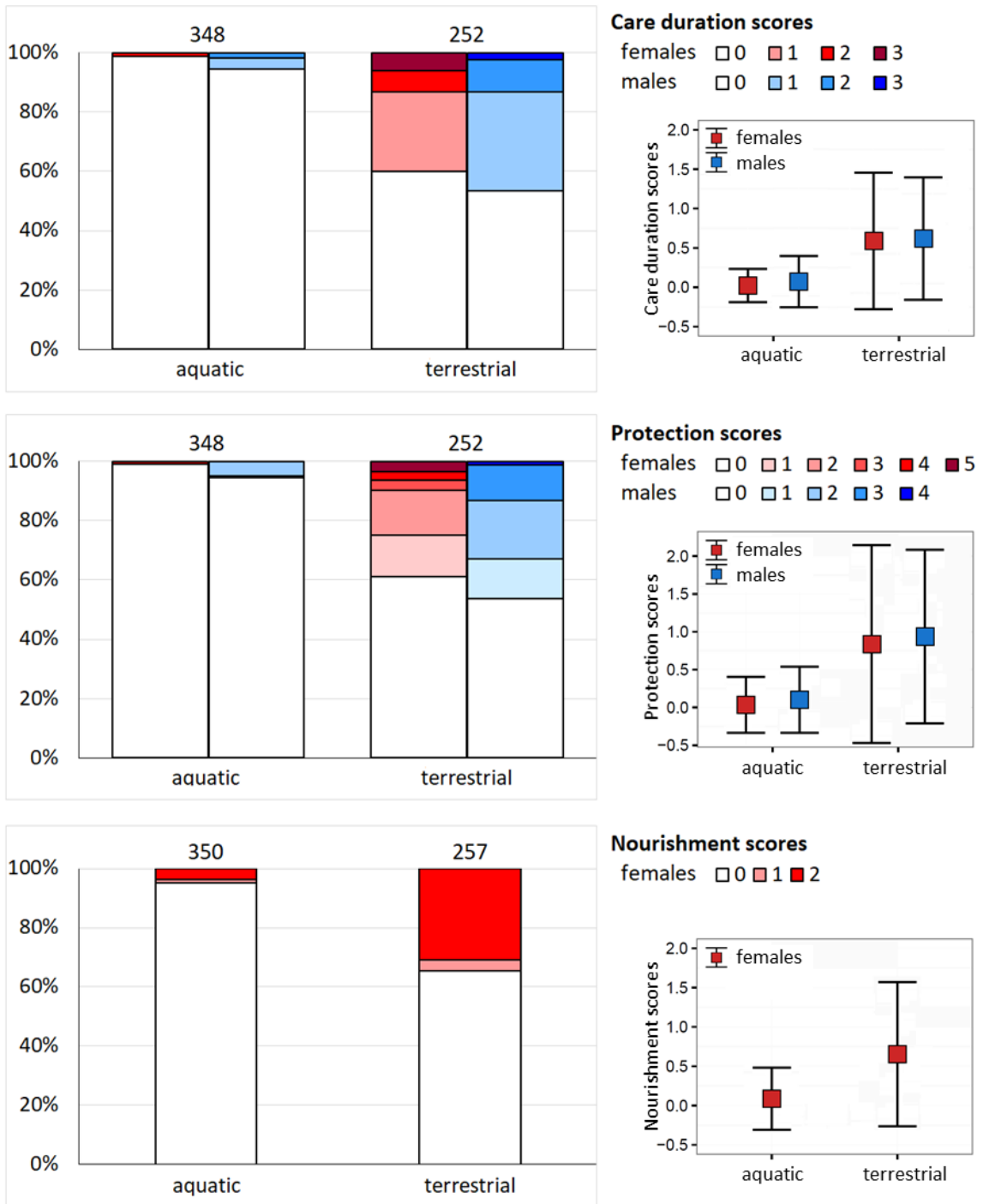carrying on back
carrying in a closed organ

520
521
522

19

523
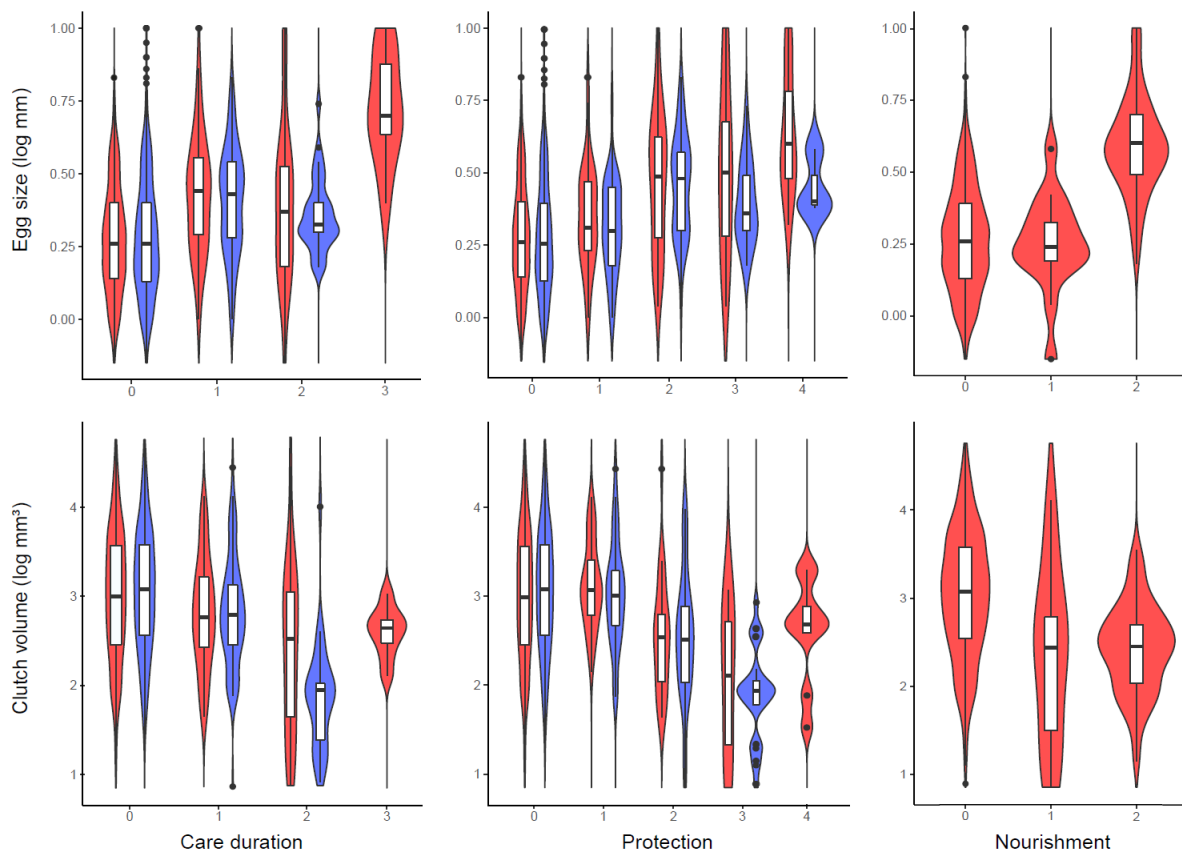524 **Figure 2**

525
526

527

Figure 3

529

530

531

532

533   **Table 1.** Parental care in relation to ecology, life-history and sexual dimorphism in Anura using

534   phylogenetically corrected generalized linear squares (PGLS) models. Multipredictor PGLS models for

535   each care variable are provided separately for males and females; note that only females provide

536   nourishment. Higher node was included in the models except for nourishment (see Methods). Italics

537   indicate significant predictors. Egg size is provided as diameter in mm. Clutch volume is calculated as

538   egg volume × clutch size and provided as mm^3. Clutch volume and egg size were log-transformed prior

539   to the analyses. Body size refers to the average snout-vent length (SVL) in mm. Sexual size dimorphism

540   was calculated as $\log_{10}$ (SVL$_{male}$ / SVL$_{female}$). We provide parameter estimates with standard error ($\beta \pm$

541   *SE),* the corresponding *t* and *P* values, and the adjusted R^2 for the model including *F*(df$_{effect}$, df$_{error}$) and

542   *P* values, respectively.

Care duration	by females			by males		
	β ± SE	*T*	*P*	*β ± SE*	*t*	*P*
Terrestrial reproduction	*0.227 ± 0.103*	*2.209*	*0.028*	*0.278 ± 0.093*	*3.000*	*0.003*
Direct development	-0.386 ± 0.224	1.721	0.087	-0.015 ± 0.197	0.077	0.938
Clutch size	0.007 ± 0.056	0.130	0.897	-0.006 ± 0.053	0.110	0.913
Egg size	0.011 ± 0.177	0.061	0.951	0.009 ± 0.166	0.052	0.959
Body size	-0.001 ± 0.001	0.407	0.685	0.002 ± 0.001	1.421	0.157
Sexual dimorphism	-0.110 ± 0.388	0.282	0.778	*1.070 ± 0.376*	*2.842*	*0.005*
Model	0.155	2.961 (18, 175)	0.0001	0.175	3.254 (18, 174)	< 0.0001

Protection	by females			by males		
	β ± SE	*T*	*P*	*β ± SE*	*t*	*P*
Terrestrial reproduction	*0.426 ± 0.137*	*3.113*	*0.002*	*0.414 ± 0.158*	*2.626*	*0.009*
Direct development	-0.452 ± 0.295	1.532	0.127	0.086 ± 0.332	0.261	0.795
Clutch size	0.045 ± 0.087	0.524	0.601	-0.016 ± 0.097	0.168	0.867
Egg size	-0.059 ± 0.285	0.209	0.835	0.084 ± 0.310	0.272	0.786
Body size	0.000 ± 0.001	0.038	0.969	0.001 ± 0.002	0.656	0.513
Sexual dimorphism	-0.208 ± 0.640	0.325	0.746	*2.156 ± 0.701*	*3.075*	*0.002*
Model	0.282	5.231 (18, 176)	< 0.0001	0.125	2.539 (18, 176)	< 0.001

Nourishment	by females			by females excluding species with endotrophic tadpoles, direct development and viviparity		
	β ± SE	*T*	*P*	*β ± SE*	*t*	*P*
Terrestrial reproduction	0.018 ± 0.098	0.186	0.853	0.014 ± 0.055	0.265	0.792
Clutch size	*-0.180 ± 0.053*	*3.389*	*< 0.001*	*-0.066 ± 0.030*	*2.162*	*0.032*
Egg size	0.119 ± 0.169	0.706	0.481	*-0.195 ± 0.097*	*2.010*	*0.046*

Body size	*0.003 ± 0.001*	*2.043*	*0.042*	*0.003 ± 0.001*	*3.513*	*0.001*
Sexual dimorphism	-0.148 ± 0.373	0.398	0.691	0.162 ± 0.347	0.208	0.437
Model	0.194	3.781 (17, 179)	< 0.0001	0.060	3.283 (5, 174)	0.007