SHORT COMMUNICATION

The effect of parity on morphological evolution among phrynosomatid lizards

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Abstract

The shift from egg laying to live-bearing is one of the most well-studied transitions in evolutionary biology. Few studies, however, have assessed the effect of this transition on morphological evolution. Here, we evaluated the effect of reproductive mode on the morphological evolution of 10 traits, among 108 species of phrynosomatid lizards. We assess whether the requirement for passing shelled eggs through the pelvic girdle has led to morphological constraints in oviparous species and whether long gestation times in viviparous species have led to constraints in locomotor morphology. We fit models to the data that vary both in their tempo (strength and rate of selection) and mode of evolution (Brownian or Ornstein-Uhlenbeck) and estimates of trait optima. We found that most traits are best fit by a generalized multipeak OU model, suggesting differing trait optima for viviparous vs. oviparous species. Additionally, rates (σ^2) of both pelvic girdle and forelimb trait evolution varied with parity; viviparous species had higher rates. Hindlimb traits, however, exhibited no difference in σ^2 between parity modes. In a functional context, our results suggest that the passage of shelled eggs constrains the morphology of the pelvic girdle, but we found no evidence of morphological constraint of the locomotor apparatus in viviparous species. Our results are consistent with recent lineage diversification analyses, leading to the conclusion that transitions to viviparity increase both lineage and morphological diversification.

Introduction

Transition in reproductive mode or parity, from egg-laying (oviparous) animals to live-bearing (viviparous) animals, is one of the most frequently observed evolutionary shifts among vertebrate taxa and has been observed in numerous lineages (Blackburn, 2000, 2006). Among squamates alone, it is estimated this transition has occurred more than 100 times independently (Blackburn, 2000; Sites et al., 2011); although recent research has suggested reversals back to oviparity from viviparity may exist (Pyron & Burbrink, 2013).

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The frequency with which squamate lineages shift among parity types makes them an ideal group for comparative studies that examine the effects of transitioning to live birth. Years of research have demonstrated the effect of live-bearing on reproductive anatomy and physiology (Andrews, 2002; Thompson & Speake, 2006), why viviparity evolves (Schulte et al., 2000; Hodges, 2004; Lynch, 2009; Lambert & Wiens, 2013) and the effect of viviparity on lineage diversification (Lynch, 2009; Lambert & Wiens, 2013; Pyron & Burbrink, 2013). However, few studies have examined the effect of reproductive mode on overall morphological diversity, despite the potential conflicting demands of reproduction and locomotion on morphology.

Morphological diversity is often estimated by examining rates of morphological diversification, which can be affected by numerous factors (Kazancıoğlu et al., 2009;

Price *et al.*, 2011). While the transition to live birth increases lineage diversification (Lynch, 2009; Lambert & Wiens, 2013; Pyron & Burbrink, 2013), it may also affect rates of morphological evolution and trait optima through its potential to influence functional systems related to maximizing reproductive output and/or locomotor morphology. Oviparous species produce a shelled egg that must pass through the pelvic girdle during oviposition. It has been suggested that there may be morphological constraints due to the production of an optimal-sized egg (Congdon & Gibbons, 1987). If an egg too large to pass through the pelvic girdle is produced, it may rupture upon oviposition, decreasing the survival of the offspring. Conversely, eggs that are too small limit the amount of nutrients provisioned to embryos, result in smaller embryos and may decrease overall offspring survival (Smith & Fretwell, 1974). Therefore, the passage of eggs in oviparous species may constrain the evolution of pelvic morphology to ensure the passage of optimal-sized eggs (Congdon & Gibbons, 1987). Without this constraint, that is when species transition to live birth (viviparity), they may exhibit increased rates of morphological evolution of the pelvic girdle. One benefit of oviparity is the short retention time of the embryos within the body, which may be advantageous in warmer environments, whereas viviparous species retain eggs for longer periods of time until the live birth of offspring, which may be beneficial in colder environments (Lambert & Wiens, 2013). The longer retention time of the embryos in viviparous species, however, may constrain the evolution of the locomotor apparatus (Shaffer *et al.*, 1996; Qualls & Shine, 1998; Shine, 2003). Therefore, the transition to live birth may coincide with decreased rates of morphological evolution of locomotor traits. These two hypotheses, pelvic constraints in oviparous species and locomotor constraints in viviparous species, can be tested in relation to the shift to live birth using modern phylogenetic comparative methods.

In this paper, we test how parity effects tempo and mode of morphological evolution in 108 phrynosomatid lizards (Fig. 1) using phylogenetic comparative methods aimed at examining rates of evolution to detect constraints. Using the OUwie package for R (Beaulieu *et al.*, 2012), first, we test whether oviparous species have decreased rates of diversification in the pelvic girdle, consistent with the pelvic constraint hypothesis to ensure successful passage of the eggs. Second, we test whether the transition to viviparity coincides with decreased diversification rates of the limbs, as may be expected by the longer retention time of eggs constraining the locomotor apparatus. In total, we examined the fit of seven evolutionary models that vary in parameter estimation and model of evolution, Brownian or Ornstein-Uhlenbeck (Table 1), for 10 morphological traits among 108 species of Phrynosomatidae (Fig. 1), which exhibit six independent transitions to viviparity, and a

recent well-supported phylogenetic hypothesis (Lambert & Wiens, 2013) to test for morphological constraints associated with reproductive mode.

Materials and methods

Following the phylogeny and parity modes of Guillette *et al.* (1980) and Lambert & Wiens (2013), we measured 10 morphological traits in 108 species of alcohol preserved phrynosomatid lizards. Only adult animals were used ($n = 1–6$ individuals). We used digital callipers (Mitutoyo ABSOLUTE Coolant Proof Series, Aurora, IL, USA) to measure the following traits: snout-vent length (SVL), pelvic height (PH), pelvic width (PW), humerus (HU), radius (RA), 4th finger (IVF), femur (FE), tibia (TIB), metatarsal (MET) and 4th toe (IVT) length (Appendix S1). All traits were log_{10}-transformed and phylogenetically size-corrected against SVL (Revell, 2009).

Using the time-calibrated phylogeny with branch lengths in millions of years (Fig. 1) from Lambert & Wiens (2013), we used stochastic character mapping in SIMMAP v1.5 to estimate uncertainty in the timing of the transition to viviparity along the branch lengths (Huelsenbeck *et al.*, 2003; Bollback, 2006). The priors used in the model were similar to previous studies (e.g. Price *et al.*, 2014). We used a branch length prior on the rate parameter, rescaled the tree length so that we did not generate an unbelievably high estimate of the number of transitions, an uninformative prior of $\alpha = 1$, and κ set to the maximum of 101 for all analyses to provide the finest-scale possible description of the distribution. We then generated 10 000 stochastically mapped trees (Fig. 1). This method produced an estimated six independent transitions to viviparity, similar to previous studies (Lambert & Wiens, 2013). It should be noted that although we only have five of the six transitions in our sample (we are missing data for *Sceloporus goldmani*), SIMMAP still estimates six transitions, as there are some instances across the 10 000 sampled trees where a transition to viviparity switches back to oviparity. We did not specify the direction of the transition as recent studies (Pyron & Burbrink, 2013) suggest there may be transitions from viviparity to oviparity. Because of this, there are trees with extant oviparous taxa that have a transition to viviparity and then back to oviparity along a branch, resulting in an estimated six transitions to viviparity.

The 10 000 stochastically mapped trees were then used in the OUwie (v1.34) package for R (2013) to estimate the model and rate of evolution (Beaulieu *et al.*, 2012). OUwie evaluates the fit of different evolutionary models (Table 1; e.g. Brownian vs. Ornstein-Uhlenbeck) as well as their parameter estimates (α = strength of selection or pull towards the optimal trait value, σ^2 = rate of diversification and θ = trait optima). In short, we compare a Brownian motion model with a

Fig. 1 Phylogeny and species used in the study based off of Lambert & Wiens (2013). Colours are a density map (Revell, 2013) for 1000 stochastically mapped trees showing the variation in timing of transition to viviparity. Red = viviparous, blue = oviparous, purple = uncertainty in the timing of the transition. Tip names are spelled out in the Appendix S1.

one-rate σ^2 (BM) and two-rate σ^2 (BMS), a single-peak OU model (OU1) and two-parameter OU models that vary in their parameter estimates: OUM (two-θ model), OUMV (two-θ and -σ^2 model), OUMA (two-θ and -α model) and OUMVA (two-θ, -σ^2, and -α). These seven models were evaluated for each trait independently. If

Table 1 Summary of the models examined using the OUwie package for R. 'None' means that the parameter was not estimated in that model. 1-rate (optimum) means that the model estimates that parameter the same for oviparous and viviparous. 2-rate (optima) means that the model estimates that parameter as different for oviparous and viviparous.

Model	Mode	Strength of selection α	Diversification rate δ^2	Trait optima θ
BM	Brownian	None	1-rate	None
BMS	Brownian	None	2-rate	None
OU1	Ornstein-Uhlenbeck	1-rate	1-rate	1-optimum
OUM	Ornstein-Uhlenbeck	1-rate	1-rate	2-optima
OUMV	Ornstein-Uhlenbeck	1-rate	2-rate	2-optima
OUMA	Ornstein-Uhlenbeck	2-rate	1-rate	2-optima
OUMVA	Ornstein-Uhlenbeck	2-rate	2-rate	2-optima

oviparity constrains the pelvic girdle and viviparity constraints locomotor morphology, we expect either the BMS, OUMV, OUMA or OUMVA models to provide the better fit with the constrained group having lower σ^2 and/or higher α for the traits of interest. Furthermore, examining the optimal trait values allows us to estimate the difference in particular morphological traits between oviparous and viviparous groups and consider the functional implications of these differences. Each of these seven models was evaluated on each of the 10 morphological traits across the 10 000 stochastically mapped trees.

The fit of alternate models was determined using ΔAICc (Table 2). We averaged ΔAICc across the 10 000 stochastically mapped trees. A model was considered to be supported if its average ΔAICc ± SD was < 2 (Burnham & Anderson, 2002). For most traits, more than one model of trait evolution was supported (Table 2); therefore, we used a model-averaging approach to estimate parameter values between the oviparous and viviparous groups (Beaulieu *et al.*, 2012). For most traits, a 2-rate α model was not supported (Table 2); we therefore only evaluated σ^2 and θ between oviparous and viviparous, by averaging each parameter across all models (Table 1), except OUMVA because it is often hard to estimate all three parameters on smaller trees (Beaulieu *et al.*, 2012), weighting the mean by each model's AICc. The parameters were averaged for a given stochastically mapped tree across the 10 000 trees. We then evaluated the histograms for σ^2, if the oviparous and viviparous have similar diversification rates, we would expect overlap in the histograms. If they exhibit different parameter estimates, we expect the histograms to diverge between the two groups.

Results

Most traits were better fit by an OU model than a Brownian motion model (Table 2). Body size, pelvic girdle, two of the three forearm traits and 4th toe were best fit by models that allowed the rate and optima to

vary with reproductive mode. Inspection of the histograms for σ^2 (Fig. 2) for all of the above traits reveals that viviparous species have increased rates of morphological diversification, with clear separation between oviparous and viviparous parameters for all traits. For most hindlimb traits, however, a model with a 1-rate σ^2 is best supported suggesting no difference in the rate for these traits between the two groups. Examining the trait optima (Table 3) shows that oviparous taxa have greater trait optima for body size, pelvic height and hindlimb traits, whereas viviparous taxa have greater optima for pelvic width and forearm traits.

Discussion

Constraints can be defined as anything that reduces potential diversity and have typically been examined in evolutionary biology using cluster analysis, morphospace occupation and phylogenetic analyses (Lauder, 1981; Emerson, 1988; Arnold, 1992; Björklund & Merilä, 1993; Hambright, 1994). Whereas previous methods have taken phylogeny into account by examining how shared ancestry may constrain a species' morphology, modern comparative methods allow for the determination of the variance in morphology over evolutionary time (Garland, 1992; O'Meara *et al.*, 2006; Beaulieu *et al.*, 2012). Although these models have most often been used to determine whether the transition to a different ecology or lifestyle increases rates of morphological evolution (Collar *et al.*, 2011; Lapiedra *et al.*, 2013), they can also be used to examine the converse, a decrease in rates of morphological evolution over time (σ^2), which may indicate the presence of a constraint, as lower rates suggest lower morphological disparity (Garland, 1992; O'Meara *et al.*, 2006; Beaulieu *et al.*, 2012). Further, as several studies have suggested constraints may be due to strong stabilizing selection, we can use these comparative methods to determine whether an Ornstein-Uhlenbeck model (OU, which models traits under stabilizing selection) has a better fit, which would further support the notion of constraints

Table 2 Mean ΔAICc values and AICc weights across 10 000 SIMMAP trees ± SD. Models with a ΔAICc < 2 (in bold for both ΔAICc and AICc weights) are considered equally good fits.

Trait	BM	BMS	OU1	OUM	OUMV	OUMA	OUMVA
ΔAICc							
SVL	9.668 ± 0.114	2.023 ± 0.395	**0.011 ± 0.114**	**1.824 ± 0.145**	**1.282 ± 0.273**	**1.437 ± 0.462**	3.416 ± 1.138
PH	27.764 ± 0.425	27.368 ± 0.557	**0.364 ± 0.425**	**0.022 ± 0.217**	**1.076 ± 0.315**	**1.392 ± 0.373**	14.413 ± 9.868
PW	9.997 ± 1.151	**0.316 ± 0.879**	4.220 ± 1.151	5.487 ± 0.935	**1.371 ± 1.040**	2.022 ± 1.023	3.978 ± 5.592
HU	21.485 ± 0.367	16.055 ± 0.400	**0.351 ± 0.367**	**0.561 ± 0.405**	**0.632 ± 0.641**	**0.300 ± 0.587**	10.785 ± 7.506
RA	21.996 ± 0.692	**0.022 ± 0.156**	10.790 ± 0.692	12.831 ± 0.705	**0.800 ± 0.300**	2.452 ± 1.345	5.991 ± 9.037
IVF	15.110 ± 0.774	15.382 ± 0.625	**0.075 ± 0.774**	2.183 ± 0.777	4.363 ± 0.749	4.307 ± 0.695	8.163 ± 4.062
FE	29.818 ± 0.021	31.938 ± 0.076	**0.000 ± 0.021**	**1.902 ± 0.067**	3.204 ± 0.117	3.297 ± 0.158	23.527 ± 10.793
TIB	8.246 ± 0.179	9.714 ± 0.163	**0.006 ± 0.179**	**1.131 ± 0.456**	3.319 ± 0.457	3.321 ± 0.445	9.910 ± 3.100
MET	9.871 ± 2.094	4.848 ± 2.043	**0.823 ± 2.094**	2.814 ± 2.105	3.103 ± 2.065	2.428 ± 1.809	**1.652 ± 2.582**
IVT	6.282 ± 1.282	**1.145 ± 1.318**	4.631 ± 1.282	6.434 ± 1.297	4.722 ± 1.307	4.190 ± 1.259	**0.608 ± 1.578**
AICc weights							
SVL	0.003 ± 0.0002	0.122 ± 0.019	**0.332 ± 0.030**	**0.134 ± 0.013**	**0.176 ± 0.020**	**0.165 ± 0.030**	0.069 ± 0.037
PH	$3.2\ 10^{-7} \pm 3.9 \times 10^{-8}$	$3.9 \times 10^{-7} \pm 1.8 \times 10^{-7}$	**0.282 ± 0.035**	**0.334 ± 0.034**	**0.197 ± 0.018**	**0.169 ± 0.020**	0.018 ± 0.057
PW	0.003 ± 0.002	**0.366 ± 0.093**	0.056 ± 0.030	0.028 ± 0.010	**0.223 ± 0.074**	0.158 ± 0.051	0.166 ± 0.176
HU	$6.7 \times 10^{-6} \pm 1.8 \times 10^{-6}$	$9.9 \times 10^{-5} \pm 2.0 \times 10^{-5}$	**0.258 ± 0.069**	**0.227 ± 0.024**	**0.222 ± 0.043**	**0.261 ± 0.042**	0.031 ± 0.066
RA	$7.9 \times 10^{-6} \pm 2.5 \times 10^{-6}$	**0.448 ± 0.064**	0.002 ± 0.001	0.001 ± 0.0003	**0.304 ± 0.048**	0.154 ± 0.090	0.090 ± 0.057
IVF	0.0003 ± 0.00004	0.0003 ± 0.0001	**0.597 ± 0.075**	0.208 ± 0.027	0.070 ± 0.009	0.072 ± 0.009	0.052 ± 0.119
FE	$1.9 \times 10^{-7} \pm 4.3 \times 10^{-9}$	$6.5 \times 10^{-8} \pm 6.5 \times 10^{-9}$	**0.559 ± 0.013**	**0.216 ± 0.006**	0.113 ± 0.005	0.108 ± 0.007	0.005 ± 0.014
TIB	0.008 ± 0.001	0.004 ± 0.001	**0.501 ± 0.054**	**0.285 ± 0.032**	0.096 ± 0.011	0.095 ± 0.011	0.011 ± 0.034
MET	0.003 ± 0.001	0.042 ± 0.014	**0.315 ± 0.102**	0.117 ± 0.039	0.100 ± 0.032	0.136 ± 0.039	**0.287 ± 0.216**
IVT	0.024 ± 0.009	**0.318 ± 0.123**	0.055 ± 0.21	0.023 ± 0.009	0.053 ± 0.021	0.069 ± 0.028	**0.457 ± 0.197**

SVL, snout–vent length; PH, pelvic height; PW, pelvic width; HU, humerus; RA, radius; IVF, fourth finger; FE, femur; TIB, tibia; MET, metatarsal; IVT, fourth toe.
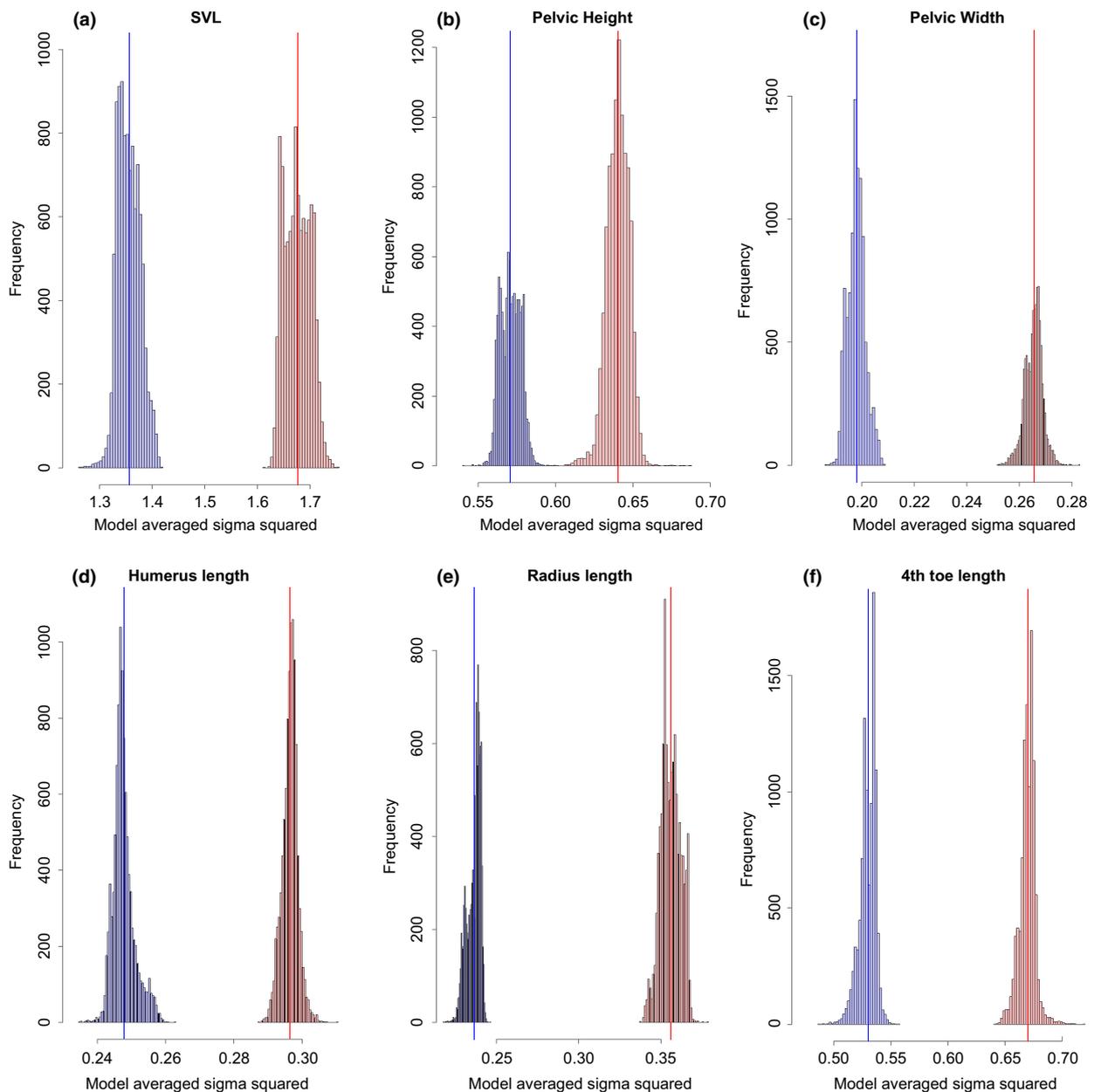
Fig. 2 Histograms of model-averaged σ^2 across 10 000 stochastically mapped trees for traits [A: snout-vent length (SVL), B: pelvic height (PH), C: pelvic width (PW), D: humerus length (HU), E: radius length (RA), F: 4th toe length (IVT)] that included a 2-rate σ^2 model as better fit (Table 2), blue = oviparous, red = viviparous. Lines represent mean σ^2.

shaping morphological evolution. Additionally, OU models require an additional parameter to estimate the strength of the pull towards an adaptive peak (Arnold, 1992). Therefore, if constraints exist, we would expect that pull to be stronger in our constrained group (higher alpha). In OU models, we can compare the trait optima (θ) to consider functional implications between our two groups. If there are constraints due to a particular character, such as oviparity constraining pelvic

girdle, then we expect a two-rate σ^2 model to provide a better fit (BM or OU), with our 'constrained' group having lower values for σ^2 (i.e. lower rates of morphological diversification). Further support for constraints would be provided by two-rate α OU models having a better fit; with the constrained group having a higher α; indicative of a stronger pull towards the adaptive peak, keeping those species' morphology closer to the optimum. Lastly, if there are constraints associated with

Table 3 Model-averaged mean optimal trait values ± SD across the 10 000 stochastically mapped trees. SVL represents $\log_{10}$ snout to vent length, all other traits are phylogenetically size-corrected $\log_{10}$-transformed traits.

Trait	Oviparous θ	Viviparous θ
SVL	1.817 ± 0.0003	1.780 ± 0.0040
PH	0.0040 ± 0.0002	−0.0113 ± 0.0009
PW	−0.0013 ± 0.0005	0.0160 ± 0.0043
HU	−0.0060 ± 0.0002	0.0093 ± 0.0016
RA	−0.0060 ± 0.0005	−0.0012 ± 0.0019
IVF	0.0097 ± 0.0003	0.0077 ± 0.0048
FE	−0.0092 ± 0.00004	−0.0132 ± 0.0006
TIB	−0.0053 ± 0.00008	−0.0253 ± 0.0042
MET	0.0063 ± 0.0003	−0.0017 ± 0.0034
IVT	0.0034 ± 0.0005	−0.0148 ± 0.0042

SVL, snout-vent length; PH, pelvic height; PW, pelvic width; HU, humerus; RA, radius; IVF, fourth finger; FE, femur; TIB, tibia; MET, metatarsal; IVT, fourth toe.

a particular group, we might also expect OU models with two-rate θ to be better fit, suggesting different phylogenetic optima for the two groups, which can then be interpreted based on functional considerations. Using the OUwie package for R (Beaulieu et al., 2012), we tested the fit of seven different evolutionary models that vary in their model of evolution (Brownian vs. OU) as well as their parameter estimations (Table 1) to determine whether there are constraints associated with reproductive mode and found constraints in the pelvic girdle and forelimb morphology among species that produce a shelled egg (oviparous).

Our study examined the fit of two evolutionary models (OU and Brownian) on morphology in phrynosomatid lizards. Other evolutionary models, such as a speciational model, may provide a better fit (see Laurin et al., 2011). However, a speciational model was not tested because there is a priori, no reason to think it might provide a better fit than the OU or Brownian motion models. Furthermore, the current methods used to analyse rates of morphological evolution, such as OUwie, do not include a speciational model of evolution to test. Therefore, we would not be able to directly compare parameter estimates for trait optima (θ) and diversification (σ^2) between gradual and speciational models.

We found that most traits were fit better by OU models than a Brownian motion model; two-rate σ^2 models were better supported than two-rate α models; and lower diversification rates (σ^2) of several traits in oviparous species. Our results suggest that egg laying may constrain the pelvic girdle and forelimb morphology, but not hindlimb morphology. How morphology changes in conjunction with parity is poorly understood despite extensive studies into shifts in reproductive mode (Guillette et al., 1980; Blackburn, 2000, 2006; Thompson & Speake, 2006). The use of modern phylo-

genetic comparative methods may be useful in detecting constraints in relation to shifts in ecology or lifestyle over evolutionary time.

We found support for the hypothesis that oviparity constrains the pelvic girdle, with reduced diversification rates of both pelvic height and width over time. Morphological constraints in relation to egg laying have been suggested for squamates and turtles and have mostly been examined within species by inspection of the regression between female size, pelvic size and egg size (Congdon & Gibbons, 1987; Clark et al., 2001; Hofmeyr, 2005; Oufiero et al., 2007). Similar slopes for the relationship between body size and pelvic opening in three species of turtles were interpreted as constraints on the pelvic girdle (Congdon & Gibbons, 1987). However, no studies have examined potential morphological constraints through evolutionary time as species transition to other modes of reproduction (i.e. viviparity). Our results suggest decreased diversity of pelvic height and width through evolutionary time, suggesting there may be less flexibility in the variation in these traits for species that produce a shelled egg. Inspections of trait optima show an increased pelvic height in oviparous compared with viviparous species (Table 2), which would allow females to produce larger eggs that may not rupture upon oviposition. However, the optimum for pelvic width is narrower for egg laying species, opposite of predictions that oviparous species are constrained to have an overall larger pelvic girdle. Because pelvic width may also be important for locomotion, there may be competing selection pressures and functional demands on the morphology associated with egg laying and that associated with movement (Snyder, 1954; Congdon & Gibbons, 1987; Irschick & Jayne, 1999). As Congdon and Gibbons note in reference to the turtle species they examined (1987): '[...] the architecture of the pelvic girdle is under strong counter-selection pressure associated with the requirements of locomotion, limb retraction and body support that prevents expansion of the aperture to accommodate greater parental investment in individual offspring'. Although the turtle bauplan is unique, it nevertheless is subject to many of the same functional considerations as other, less specialized groups. Thus, it is likely that many of the same constraints and functional trade-offs are occurring in oviparous lizards. Therefore, a higher pelvis may ensure large enough eggs can pass through, but a narrow pelvis may interact with locomotor abilities in oviparous species. However, to fully understand these relationships, studies are needed that examine the kinematics of gravid oviparous and viviparous species during differing locomotor modes to determine the functional role of pelvic width and height during locomotion and its relationship to offspring size.

We did not find support for locomotor constraints on morphology in the viviparous species – although

no direct measures of locomotor performance were measured. We found no difference in the diversification rates of hindlimbs, and rates were actually higher in the forelimbs of viviparous species. Most forelimb and hindlimb optima in viviparous species were larger and smaller, respectively, than for oviparous species. The length of the limbs and their ratio are important for locomotion in lizards (Huey *et al.*, 1990; Bonine & Garland, 1999; Aerts *et al.*, 2000; Vanhooydonck & Van Damme, 2001). For example, a low ratio of forelimbs to hindlimbs is expected to be beneficial for ground-dwelling species, whereas similar length fore- and hindlimbs may be better for climbing; furthermore, several limb characteristics have been correlated with various locomotor performance measures (Vanhooydonck & Van Damme, 2001). In fact, among 27 species of phrynosomatid lizards, hindlimb length was significantly correlated to sprint speed (Bonine & Garland, 1999). Therefore, the different optima for the limbs in oviparous and viviparous species may be related to locomotor ability and/or habitat usage. However, the only study that has examined the effect of parity on locomotion found no difference in escape speed in *Lerista bougainvillii* (Qualls & Shine, 1998), and they did not quantify kinematics or limb morphology between the different parity modes. Whereas comparisons within oviparous species have found that gravidity decreases locomotor performance in squamates (Seigel *et al.*, 1987; Sinervo *et al.*, 1991; Miles *et al.*, 2000; Olsson *et al.*, 2000; Shine, 2003), more studies are warranted to determine the interaction of limb morphology and locomotion between oviparous and viviparous species. Furthermore, because limb morphology has been shown to vary across habitats, studies are warranted that examine the effect of habitat on morphological diversification in these traits.

Many studies have examined the effect of viviparity on various aspects of the organism (Blackburn, 2000); our study is the first to examine its effect on macroevolutionary patterns of morphological diversification. Our results demonstrate that patterns of morphological evolution are consistent with lineage diversification; viviparity leads to increased diversification (Lynch, 2009; Lambert & Wiens, 2013; Pyron & Burbrink, 2013). Our study also suggests that differences exist in trait optima for the pelvic girdle and limbs between oviparous and viviparous taxa, which warrants further investigation into the functional role of these traits in egg laying and live-bearing species during locomotion. Further, our study demonstrates that the transition to viviparity is another mechanism that can increase morphological diversification, a fact that may become more important in the future, as viviparous species may be more susceptible to the effects of climate change (Pincheira-Donoso *et al.*, 2013); thus, we may lose an important source of morphological diversification.

References

Aerts, P., Van Damme, R., Vanhooydonck, B., Zaaf, A. & Herrel, A. 2000. Lizard locomotion: how morphology meets ecology. *Netherlands J. Zool.* **50**: 261–277.

Andrews, R.M. 2002. Low oxygen: a constraint on the evolution of viviparity in reptiles. *Physiol. Biochem. Zool.* **75**: 145–154.

Arnold, S. 1992. Constraints on phenotypic evolution. *Am. Nat.* **14**: S85–S107.

Beaulieu, J.M., Jhwueng, D., Boettiger, C. & O'Meara, B.C. 2012. Modeling stabilizing selection: expanding the Ornstein-Uhlenbeck model of adaptive evolution. *Evolution* **66**: 2369–2383.

Björklund, M. & Merilä, J. 1993. Morphological differentiation in *Carduelis* finches: adaptive vs. constraint models. *J. Evol. Biol.* **373**: 359–373.

Blackburn, D.G. 2000. Reptilian viviparity: past research, future directions, and appropriate models. *Comp. Biochem. Physiol., Part A Mol. Integr. Physiol.* **127**: 391–409.

Blackburn, D.G. 2006. Squamate reptiles as model organisms for the evolution of viviparity. *Herpetol. Monogr.* **20**: 131–146.

Bollback, J.P. 2006. SIMMAP: stochastic character mapping of discrete traits on phylogenies. *BMC Bioinformatics* **7**: 88.

Bonine, K.E. & Garland, T. Jr 1999. Sprint performance of phrynosomatid lizards, measured on a high-speed treadmill, correlates with hindlimb length. *J. Zool.* **248**: 255–265.

Burnham, K.P. & Anderson, D.R. 2002. *Model Selection and Multimodel Inference: A Practical Information-theoretic Approach*. Springer Verlag, New York, NY.

Clark, P.J., Ewert, M.A. & Nelson, C.E. 2001. Physical apertures as constraints on egg size and shape in the common musk turtle, *Sternotherus odoratus*. *Funct. Ecol.* **15**: 70–77.

Collar, D.C., Schulte, I.I., James, A. & Losos, J.B. 2011. Evolution of extreme body size disparity in monitor lizards (*Varanus*). *Evolution* **65**: 2664–2680.

Congdon, J.D. & Gibbons, J.W. 1987. Morphological constraint on egg size: a challenge to optimal egg size theory? *Proc. Natl. Acad. Sci. USA* **84**: 4145–4147.

Emerson, S.B. 1988. Testing for historical patterns of change; a case study with frog pectoral girdles. *Paleobiology* **14**: 174–186.

Garland, T. Jr 1992. Rate tests for phenotypic evolution using phylogenetically independent contrasts. *Am. Nat.* **140**: 509–519.

Guillette, L.J. Jr, Jones, R.E., Fitzgerald, K.T. & Smith, H.M. 1980. Evolution of viviparity in the lizard genus *Sceloporus*. *Herpetologica* **36**: 201–215.

Hambright, K.D. 1994. Morphological constraints in the pisci-vore-planktivore interaction: implications for the trophic cascade hypothesis. *Limnol. Oceanogr.* **39**: 897–912.

Hodges, W.L. 2004. Evolution of viviparity in horned lizards (*Phrynosoma*): testing the cold-climate hypothesis. *J. Evol. Biol.* **17**: 1230–1237.

Hofmeyr, M. 2005. Overcoming environmental and morphological constraints: egg size and pelvic kinesis in the smallest tortoise, *Homopus signatus. Can. J. Zool.* **1352**: 1343–1352.

Huelsenbeck, J.P., Nielsen, R. & Bollback, J.P. 2003. Stochastic mapping of morphological characters. *Syst. Biol.* **52**: 131–158.

Huey, R.B., Dunham, A.E., Overall, K.L. & Newman, R.A. 1990. Variation in locomotor performance in demographically known populations of the lizard *Sceloporus merriami. Physiol. Zool.* **63**: 845–872.

Irschick, D.J. & Jayne, B.C. 1999. Comparative three-dimensional kinematics of the hindlimb for high-speed bipedal and quadrupedal locomotion of lizards. *J. Exp. Biol.* **202**: 1047–1065.

Kazancıoğlu, E., Near, T.J., Hanel, R. & Wainwright, P.C. 2009. Influence of sexual selection and feeding functional morphology on diversification rate of parrotfishes (Scaridae). *Proc. Biol. Sci.* **276**: 3439–3446.

Lambert, S.M. & Wiens, J.J. 2013. Evolution of viviparity: a phylogenetic test of the cold-dlimate hypothesis. *Evolution* **67**: 2614–2630.

Lapiedra, O., Sol, D., Carranza, S. & Beaulieu, J.M. 2013. Behavioural changes and the adaptive diversification of pigeons and doves. *Proc. Biol. Sci.* **280**: 20122893.

Lauder, G.V. 1981. Form and function: structural analysis in evolutionary morphology. *Paleobiology* **4**: 430–442.

Laurin, M., Gussekloo, S.W.S., Marjanović, D., Legendre, L. & Cubo, J. 2011. printed 2012. Testing gradual and speciational models of evolution in extant taxa: the example of ratites. *J. Evol. Biol.* **25**: 293–303.

Lynch, V.J. 2009. Live-birth in vipers (Viperidae) is a key innovation and adaptation to global cooling during the Cenozoic. *Evolution* **63**: 2457–2465.

Miles, D.B., Sinervo, B. & Frankino, W.A. 2000. Reproductive burden, locomotor performance, and the cost of reproduction in free ranging lizards. *Evolution* **54**: 1386–1395.

Olsson, M., Shine, R. & Bak-Olsson, E. 2000. Locomotor impairment of gravid lizards: is the burden physical or physiological? *J. Evol. Biol.* **13**: 263–268.

O'Meara, B., Ané, C., Sanderson, M. & Wainwright, P. 2006. Testing for different rates of continuous trait evolution using likelihood. *Evolution* **60**: 922–933.

Oufiero, C.E., Smith, A.J. & Angilletta, M.J. Jr 2007. The importance of energetic versus pelvic constraints on reproductive allocation by the eastern fence lizard (*Sceloporus undulatus*). *Biol. J. Linn. Soc.* **91**: 513–521.

Pincheira-Donoso, D., Tregenza, T., Witt, M.J. & Hodgson, D.J. 2013. The evolution of viviparity opens opportunities for lizard radiation but drives it into a climatic cul-de-sac. *Glob. Ecol. Biogeogr.* **22**: 857–867.

Price, S.A., Holzman, R., Near, T.J. & Wainwright, P.C. 2011. Coral reefs promote the evolution of morphological diversity and ecological novelty in labrid fishes. *Ecol. Lett.* **14**: 462–469.

Price, S.A., Schmitz, L., Oufiero, C.E., Eytan, R.I., Dornburg, A., Smith, W.L. *et al.* 2014. Two waves of niche-filling straddling the K-Pg boundary formed the modern reef fish fauna. *Proc. Biol. Sci.* **281**: 20140321.

Pyron, R.A. & Burbrink, F.T. 2013, printed in 2014. Early origin of viviparity and multiple reversions to oviparity in squamate reptiles. *Ecol. Lett.* **17**: 13–21.

Qualls, C.P. & Shine, R. 1998. Costs of reproduction in conspecific oviparous and viviparous lizards, *Lerista bougainvillii. Oikos* **82**: 539–551.

Revell, L.J. 2009. Size-correction and principal components for interspecific comparative studies. *Evolution* **63**: 3258–3268.

Revell, L.J. 2013. Two new graphical methods for mapping trait evolution on phylogenies. *Methods Ecol. Evol.* **4**: 754–759.

Schulte, J.A. II, Macey, J.R., Espinoza, R.E. & Larson, A. 2000. Phylogenetic relationships in the iguanid lizard genus *Liolaemus*: multiple origins of viviparous reproduction and evidence for recurring Andean vicariance and dispersal. *Biol. J. Linn. Soc.* **69**: 75–102.

Seigel, R.A., Huggins, M.M. & Ford, N.B. 1987. Reduction in locomotor ability as a cost of reproduction in gravid snakes. *Oecologia* **73**: 481–485.

Shaffer, L.R., Formanowicz, J.R. & Daniel, R. 1996. A cost of viviparity and parental care in scorpions: reduced sprint speed and behavioural compensation. *Anim. Behav.* **51**: 1017–1024.

Shine, R. 2003. Effects of pregnancy on locomotor performance: an experimental study on lizards. *Oecologia* **136**: 450–456.

Sinervo, B., Hedges, R. & Adolph, S.C. 1991. Decreased sprint speed as a cost of reproduction in the lizard *Sceloporus occidentalis*: variation among populations. *J. Exp. Biol.* **155**: 323–336.

Sites, J.W. Jr, Reeder, T.W. & Wiens, J.J. 2011. Phylogenetic insights on evolutionary novelties in lizards and snakes: sex, birth, bodies, niches, and venom. *Annu. Rev. Ecol. Evol. Syst.* **42**: 227–244.

Smith, C. & Fretwell, S. 1974. The optimal balance between size and number of offspring. *Am. Nat.* **108**: 499–506.

Snyder, R.C. 1954. The anatomy and function of the pelvic girdle and hindlimb in lizard locomotion. *Am. J. Anat.* **95**: 1–45.

Thompson, M.B. & Speake, B.K. 2006. A review of the evolution of viviparity in lizards: structure, function and physiology of the placenta. *J. Comp. Physiol. B.* **176**: 179–189.

Vanhooydonck, B. & Van Damme, R. 2001. Evolutionary trade-offs in locomotor capacities in lacertid lizards: are splendid sprinters clumsy climbers? *J. Evol. Biol.* **14**: 46–54.

Supporting information

Additional Supporting Information may be found in the online version of this article:

Appendix S1 Raw data file of species means for the ten morphological traits and reproductive mode.