Ecol Res (2009) 24: 1223–1231
DOI 10.1007/s11284-009-0607-4

ORIGINAL ARTICLE

Daniel Pincheira-Donoso · David J. Hodgson
Jan Stipala · Tom Tregenza

A phylogenetic analysis of sex-specific evolution of ecological morphology in *Liolaemus* lizards

Received: 22 December 2008 / Accepted: 6 March 2009 / Published online: 25 April 2009

Abstract Adaptive radiation theory predicts that phenotypic traits involved in ecological performance evolve in different directions in populations subjected to divergent natural selection, resulting in the evolution of ecological diversity. This idea has largely been supported through comparative studies exploring relationships between ecological preferences and quantitative traits among different species. However, intersexual perspectives are often ignored. Indeed, although it is well established that intersexual competition and sex-specific parental and reproductive roles may often subject sex-linked phenotypes to antagonistic selection effects, most ecomorphological research has explored adaptive evolution on a single sex, or on means obtained from both sexes together. The few studies taking sexual differences into account reveal the occurrence of sex-specific ecomorphs in some clades of lizards, and conclude that the independent contribution of the sexes to the morphological diversity produced by adaptive radiation can be substantial. Here, we investigate whether microhabitat use results in the evolution of sex-specific ecomorphs across 44 *Liolaemus* lizard species. We found that microhabitat structure does not predict variation in body size and shape in either of the sexes. Yet, we found that males and females tend to occupy significantly different positions in multivariate morphological spaces, indicating that treating males and females as ecologically and phenotypically equivalent units may lead to incomplete or mistaken estimations of the diversity produced by adaptive evolution.

Keywords Natural selection · Adaptive radiation · Adaptation · Convergence · Ecomorphology · Sexual dimorphism · Lizards · *Liolaemus*

D. Pincheira-Donoso · D. J. Hodgson · J. Stipala ·
T. Tregenza (✉)
Centre for Ecology and Conservation,
School of Biosciences, University of Exeter,
Cornwall Campus, Penryn, Cornwall TR10 9EZ, UK
E-mail: T.Tregenza@exeter.ac.uk

Introduction

The quantitative study of phenotypes provides a valuable source of information for understanding the response of organisms to selection (Lynch and Walsh 1998; Pigliucci and Preston 2004). According to adaptive radiation theory, phenotypic evolution occurs when divergent natural selection pulls means of two or more conspecific populations toward different fitness peaks (Gavrilets 2004; Schluter 2000; Simpson 1953), resulting in ecological diversification (Coyne and Orr 2004; Dieckmann et al. 2004). Therefore, a clade is considered to have adaptively radiated when its members exhibit substantial ecological diversity (Givnish 1997; Losos and Miles 2002; Schluter 1996). However, since available niches are finite, two or more non-closely related species (e.g., non-sister species within the same genus or family) may independently converge to exploit similar ecological niches. Under this scenario, the evolutionary outcome will be convergent adaptive radiations (Jackman et al. 1997; Losos et al. 1998; Schluter 2000).

A number of niche components have been recognized as major factors involved in the origin and maintenance of predictable morphological variation between species (Grant 1986; Losos 1994; Schluter 2000). Occupation of different structural microhabitats has proven to be an important one. For example, the adaptive radiation of Caribbean *Anolis* lizards has independently produced similar sets of ecomorph species specialized in the exploitation of similar sets of microhabitats across different islands (Losos et al. 1998; Williams 1983). However, in many organisms, natural and sexual selection also promote the evolution of alternative phenotypes within species (Andersson 1994; Butler et al. 2007). Since conspecific males and females are often subjected to antagonistic selection resulting from competition and intrinsic differences in reproductive, sexual, and parental roles, the evolutionary trajectories followed by sex-specific phenotypes are often expected to differ (Bolnick and Doebeli 2003; Losos et al. 2003; Olsson et al. 2000; Shine

1989). For example, pregnancy is associated with reduction in sprinting performance, which in turn may increase female vulnerability to predation, in contrast to males (Miles et al. 2000; Plummer 1997; Shine 1980). Therefore, a major outcome of disruptive selection acting within species is the evolution of sexual dimorphism (Bolnick and Doebeli 2003; Fairbairn et al. 2007; Shine 1989; Slatkin 1984). This means that a significant proportion of the morphological diversity observed within clades might be explained by the different fitness peaks favored by disruptive selection at different sex-specific phenotypic optima within the same species (Fairbairn et al. 2007; Schulte et al. 2004; Shine 1989; Slatkin 1984). Surprisingly, although these ideas have been around for decades, they have only rarely been considered in relation to studies of adaptive radiation (Butler et al. 2007; Stuart-Fox and Moussalli 2007). Recently, Butler et al. (2007) showed that the independent ecological contribution of males and females to the morphological diversity produced by adaptive radiations may be substantial. These authors observed that the evolutionary history of Caribbean *Anolis* lizards has produced a high diversity of sex-specific morphological plans associated with the occupation of specific structural microhabitats (see also Butler et al. 2000; Losos et al. 2003). Butler et al. (2007) concluded that ignoring the contribution of substantial sexual dimorphism in ecologically relevant traits may result in significant underestimates of the adaptive component of evolutionary radiation. Therefore, ecomorphological studies based exclusively on one of the two sexes, or on a single value obtained from both males and females together, may provide partial or misleading estimates when exploring the outcome of adaptive radiation. In spite of this, only a few explicit attempts to investigate the independent response of both males and females to the process of adaptive radiation have been published (Butler et al. 2007; Stuart-Fox and Moussalli 2007).

Here, we aim to test the hypothesis that occupation of specific structural microhabitats produces predictable adaptations resulting in convergent evolution of body size and shape among unrelated species exploiting similar environments, using both sexes as independent units of analysis. Hence, we investigate whether similar sex-specific ecomorphological adaptations have independently evolved among ecologically similar species. To address these problems, we used South American *Liolaemus* lizards as a model system. Consisting of 200+ species belonging to at least six main clades (Espinoza et al. 2004; Pincheira-Donoso et al. 2008c; Schulte et al. 2000), the *Liolaemus* genus is an ideal group to study adaptive radiations. These lizards exhibit a great diversity in ecological specializations, morphology, and in the extent of sexual dimorphism, which is widespread. Also, the geographical radiation experienced by *Liolaemus* is among the most extraordinary recorded for reptiles, being adapted to one of the widest environmental ranges observed in lizards (Cei 1993; Donoso-Barros 1966; Pincheira-Donoso et al. 2008a; Pincheira-Donoso and Núñez 2005). Finally, in spite of the exceptional biological diversity of these iguanians, only two comparative studies on their multivariate morphological diversification have been published. The first of these works (Jaksic et al. 1980) was conducted on the basis of single means per species obtained from males, females, and subadults of 12 taxa, when phylogenetic comparative methods were not yet routinely incorporated into evolutionary research. More recently, Schulte et al. (2004) applied a phylogenetic approach to analyze ecomorphological evolution only in adult males of 25 *Liolaemus* species belonging to four of the six known main subclades of this genus. These authors observed that microhabitat structures appear not to predict variation in morphological plans across species. Here, we study the relationship between microhabitat occupation and body size and shape separately for both males and females of 44 *Liolaemus* species belonging to all the six reported subclades, and occurring in all environments recorded for these lizards. Therefore, this study not only covers the entire ecological diversity acquired by *Liolaemus*, but it is also one of the few available studies exploring the evolution of sex-specific adaptations during adaptive radiation.

Materials and methods

Study species and microhabitat classification

We studied a sample of 2,058 adult *Liolaemus* specimens (1,041 males, 1,017 females) belonging to 44 species, and representing all the main clades known for this genus (e.g., Cei 1993; Espinoza et al. 2004; Etheridge 1995; Pincheira-Donoso and Núñez 2005; Fig. 1, Table 1). The 44 studied species occur from the deserts of Atacama and Tarapaca in northern Chile and Peru, to austral Argentinean and Chilean Patagonia. Altitudinally, these species range from sea level to more than 5,000 m of elevation in the Andes. Almost every intermediate environment encompassed between these extreme climates has also been represented (e.g., tropical areas in Brazil, Chilean savanna and austral forests, Argentinean pampa). All the specimens used in this study are housed in the institutions and collections detailed in the Appendix.

Species were assigned to one of seven structural microhabitats recorded for *Liolaemus* (Table 1). The microhabitat type where most members of a species ($\geq$70%) bask and dwell (when active) was considered the preferred perch for that taxon. The survey involved both males and females equally using the same microhabitat. This ecological information was obtained from personal field observations and from published data (Cei 1986, 1993; Donoso-Barros 1966; Etheridge 2000; Jaksic 1998; Jaksic et al. 1980; Pincheira-Donoso and Núñez 2005; Scolaro 2006, Table 1; Schulte et al. 2004). We assigned species to microhabitat types according to the following criteria: (1) tree trunk (species dwelling primarily on the

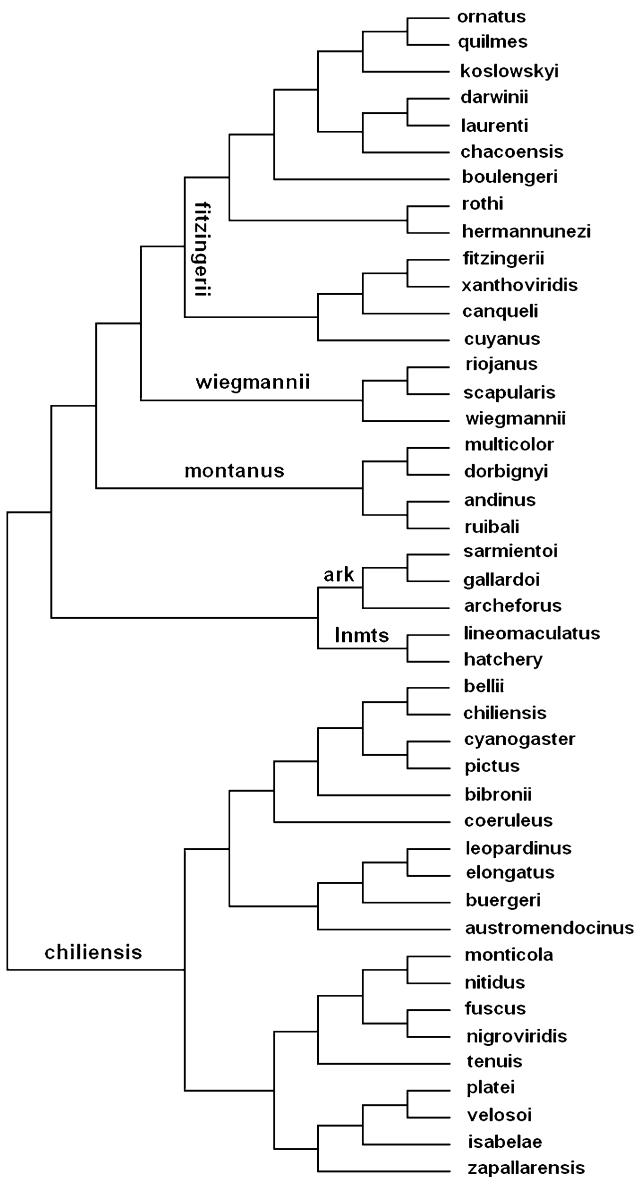

Fig. 1 Phylogeny of *Liolaemus* lizards used in ecomorphological analyses of body size and shape variation. The names of *Liolaemus* subclades are indicated on the branches. Subclades *archeforus-kingii* (*ark*) and *lineomaculatus* (*lnmts*) are abbreviated

trunks of trees, and rarely or never on thin twigs), (2) twigs (species observed on shrubs; none of the species occurring on twigs was observed on trees), (3) rocks, (4) ground, dense vegetation (species dwelling on ground densely covered by shrubs typically separated by less than 100 cm), (5) open ground, shrubs (ground dweller species running between shrubs separated by 101–4,000 cm), (6) open ground (species occurring in open deserts with scarce or almost entirely absent vegetation; when shrubs are present, they are separated by at least four or more meters), and (7) sand (species dwelling on open sandy ground with different degrees of vegetation coverage. In these environments, species tend to exhibit sand-burying behaviors).

Morphological measurements

Ten external morphological measurements were taken from both male and female adult specimens by the same person (DPD), to avoid interindividual biases (Lee 1990; Losos et al. 2003). Since lizards typically shrink by about 9% after preservation in ethanol (Roughgarden 1995), we only studied museum samples (see Appendix). These morphometric variables were selected on the basis of their ecological significance and adaptive lability, as reported in several previous lizard studies (Beuttell and Losos 1999; Losos 1990; Schulte et al. 2004; Vanhooydonck and Van Damme 1999; Vitt et al. 1997). We used snout–vent length (SVL) as a proxy for body size. SVL correlates positively with other body variables, such as body mass, and with ecological and life-history traits. We estimated mean values for all studied variables using the largest two-thirds of the total adult sample for each studied species (Losos et al. 2003; Pincheira-Donoso et al. 2008b). In addition, geographical variation in thermal regimes have largely been considered as an important factor promoting predictable variation in body size, and potentially in other traits that might respond to body mass variation. Nevertheless, some recent phylogenetic studies have revealed that body size in *Liolaemus* lizards does not vary predictably with either latitudinal or altitudinal geographical variation (Pincheira-Donoso et al. 2007b, 2008b). Therefore, at least at this level, no significant geographical bias might be expected in *Liolaemus*, which however, requires further studies to explore this assumption more thoroughly.

We measured the following morphological variables: (1) snout–vent length (from the tip of the snout to the anterior edge of the cloacae), (2) head length (lateral measurement from the anterior edge of the ear opening to the tip of the snout), (3) head width (the widest zone of the head immediately anterior to the ear), (4) forelimb length (distance from insertion of the limb into the body wall to the end of the third toe, excluding the claw), (5) hind limb length (distance from the insertion of the limb into the body wall to the end of the fourth toe, excluding the claw), (6) tibia length (from knee to the proximal end of the foot), (7) foot length (from the proximal end of the foot to the end of the fourth toe, excluding the claw), (8) axilla-groin length (from the axilla to the anterior insertion of hind limb on the body wall), (9) pelvic width (width of the body immediately anterior to the insertion of hind limbs in the body wall), and (10) tail length (from the anterior edge of the cloaca to the tip of the tail, excluding individuals with broken, missing or regenerated tails). Except when any damage or break occurred on the right side, all measurements were obtained from the right side of the individuals.

Statistical analyses and phylogenetic control

Statistical analyses were based on the mean value of each variable for each studied species, separately for the

Table 1 Species and microhabitats of the *Liolaemus* lizards used in this study

Clade	Species	N		Microhabitat
		Males	Females	
fitzingerii	*boulengeri*	13	11	Open ground-shrubs
	chacoensis	6	6	Ground-dense vegetation
	canqueli	5	5	Open ground-shrubs
	darwinii	23	22	Open ground-shrubs
	fitzingerii	13	12	Open ground-shrubs
	hermannunezi	12	14	Open ground-shrubs
	koslowskyi	11	12	Open ground-shrubs
	laurenti	7	6	Open ground-shrubs
	ornatus	17	14	Open ground-shrubs
	quilmes	9	8	Open ground
	rothi	14	15	Open ground-shrubs
	xanthoviridis	3	5	Open ground-shrubs
wiegmannii	*riojanus*	6	5	Sands
	scapularis	16	14	Sands
	wiegmannii	26	23	Sands
montanus	*andinus*	27	29	Open ground
	cuyanus	5	4	Sands
	dorbignyi	5	5	Open ground
	multicolor	10	13	Open ground
	ruibali	67	66	Open ground-shrubs
archeforus-kingii	*archeforus*	5	5	Open ground-shrubs
	gallardoi	4	3	Open ground-shrubs
	sarmientoi	8	7	Open ground-shrubs
lineomaculatus	*lineomaculatus*	13	12	Open ground-shrubs
	hatcheri	4	4	Open ground-shrubs
chiliensis	*austromendocinus*	5	6	Open ground-shrubs
	bellii	41	46	Open ground-shrubs
	bibronii	11	13	Ground-dense vegetation
	buergeri	13	19	Rocks
	chiliensis	39	36	Twigs
	coeruleus	11	10	Open ground-shrubs
	cyanogaster	35	31	Twigs
	elongatus	14	12	Rocks
	fuscus	27	23	Rocks
	isabelae[a]	13	11	Rocks
	leopardinus	19	19	Rocks
	monticola	34	38	Rocks
	nigroviridis	97	84	Rocks
	nitidus	32	33	Rocks
	pictus	29	28	Tree trunks
	platei	17	12	Open ground-shrubs
	tenuis	214	230	Tree trunks
	velosoi	23	15	Open ground-shrubs
	zapallarensis	38	31	Open ground-shrubs

Sample sizes of males and females shown represent the largest two-thirds of a total sample of studied adult specimens

[a] See Pincheira-Donoso and Núñez (2007)

sexes. Prior to analyses, all morphometric variables were ln-transformed to reduce skew and make data variances homogeneous (Field 2006; Miles and Ricklefs 1984; Zar 1999). After ln-transformation, all studied variables met the statistical assumptions required for parametric analyses.

We aimed to test whether male and female body size and shape evolve predictably in response to variation in microhabitat characteristics. Therefore, since quantitative differences between species body size may cause allometric bias in body shape variables, we removed the effects of snout–vent length on the remaining traits to compare size-independent shape variables. Least square linear regressions of body size (e.g., SVL) against linear measures of shape have largely been used to produce size-effect-free residual indices (Green 2001). However, recent studies have suggested that a series of relevant assumptions may not hold for residuals, and that the residual index is an ad-hoc sequential procedure with no demonstrated statistical justification, unlike analyses of covariance (Garcia-Berthou 2001; Green 2001; Hayes and Shonkwiler 1996; Smith 1999). Thus, to control for the allometric effect of body size, we applied univariate (ANCOVA) and multivariate (MANCOVA) analyses of covariance, with body size (SVL) as covariate, and the remaining morphological traits as dependent variables (Zelditch et al. 2004) separately for both males and females.

Comparative analyses of interspecific data may require phylogenetic control as closely related species share parts of their evolutionary history. Therefore, they cannot be considered independent data points for statistical analyses (Felsenstein 1985; Harvey and Pagel 1991). To explore the effect of microhabitat occupation on body shape, the creation of empirical null distributions of *F*-statistics using phylogenetic analyses of covariance (PANCOVA, as implemented in PDAP) offers a powerful approach to circumvent the problem of phylogenetic dependence (Garland et al. 1993). This approach relies on the fact that the use of conventional (non-phylogenetic) ANCOVAs increases the risk of inflating degrees of freedom when analyzing interspecific data (due to non-independence of data points), leading ultimately to potentially spurious significance levels. We introduced into the model the phylogenetic hypothesis derived from previous studies shown in Fig. 1 (Espinoza et al. 2004; Pincheira-Donoso et al. 2007a; Schulte et al. 2000, 2004). Since this phylogeny is based on both a molecular and morphological dataset, we performed analyses under a speciational Brownian motion model of evolutionary change, assuming branch lengths equal to 1.0 (e.g., Garland et al. 1993; Rohlf et al. 1990). To conduct the phylogenetic ANCOVAs, we first conducted a conventional non-phylogenetic ANCOVA separately on each dependent body shape variable (SVL as covariate), and independently for both sexes in the 44 species included in the phylogeny (Fig. 1; Appendix), to create standard tabular *F*-statistics for each trait. We then simulated character evolution along the phylogeny for each dependent variable using PDSIMUL and then performed the PANCOVA in PDANOVA (Garland et al. 1993). To explore variation in body size, we applied the same procedure, but used phylogenetic analyses of variance (PANOVA). Simulations were repeated 1,000 times for each trait. PDANOVA generates empirical null distributions of *F*-statistics for each trait. We considered that morphological traits differ significantly between microhabitat types if the conventional critical values (i.e., *F*-values) obtained from non-phylogenetic ANCOVAs (for each body shape trait) and ANOVAs (for body size) are larger than the 95th percentile of the empirical null distribution created by PDANOVA (Garland et al. 1993, 2005). For all the phylogenetic analyses (in PDSIMUL), the means and variances of the simulations were set to the means and variances of the original data (Garland et al. 1993; Schulte et al. 2004).

We then quantified the extent of multivariate morphological differences between the sexes using a multivariate analysis of covariance (MANCOVA), with SVL as a covariate to control for the allometric effect of body size on body shape components (see above). To graphically represent the positions occupied by males and females in morphological space (Fig. 2), we calculated Anderson–Rubin scores from principal component analysis (PCA) on size-adjusted body shape variables (for analytical criteria, see Jolliffe 1972, 1986). Body shape variables were obtained using an approach similar to that suggested by Mosimann (1970) to remove the effect of SVL (since a large number of species had broken or regenerated tails, this variable was excluded from this analysis). Size effects were controlled by taking the difference of each ln-dependent variable with lnSVL, with the following formula: size-controlled dependent variable = ln(dependent variable) − ln(SVL) (see also Butler and Losos 2002). We recognize that the use of ratios (ln-ratios in this case) may have unwanted statistical effects if the variables are substantially skewed or non-normally distributed (Butler and Losos 2002; Garcia-Berthou 2001; Green 2001). However, we tested these parameters and found that the application of this size-control procedure does not affect our analysis.

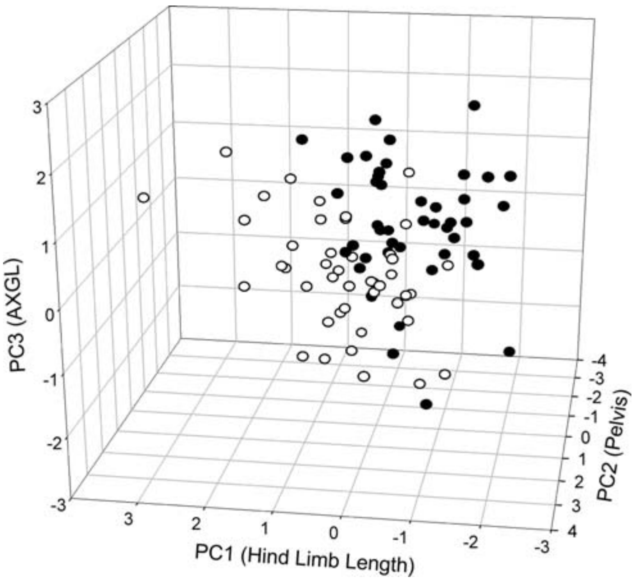

Fig. 2 The distribution of male (*black circles*) and female (*open circles*) *Liolaemus* species in a tri-dimensional morphospace based on scores from the first three morphology principal component axes (*PC1* hind limb length, *PC2* pelvis width, *PC3* axilla-groin length) using size-adjusted data

Results

Conventional and phylogenetic analyses of variance (ANOVA and PANOVA, for body size) and covariance (ANCOVA and PANCOVA, for body shape components) revealed that occupation of different microhabitat structures does not predict morphological variation in either of the sexes among *Liolaemus* species (Table 2). Significant ecomorphological relationships were only observed in male foot length ($F_{6,37} = 2.89$, $P = 0.017$) and tail length ($F_{6,37} = 4.14$, $P = 0.012$), and in female tail length ($F_{6,37} = 4.4$, $P = 0.009$) when conducting non-phylogenetic ANCOVAs. However, any signal of predictable variation vanished when incorporating phylogenetic relationships into the model (PANCOVAs, $P > 0.05$ for all traits; Table 2).

In contrast to interspecific analyses, the intersexual comparison using multivariate analysis of covariance (MANCOVA, SVL as covariate) revealed significant differences in body shape between males and females

Table 2 Results of phylogenetic analyses of variance (PANOVA, for SVL) and covariance (PANCOVA, for the remaining body shape variables) conducted among microhabitat types exploited by males and females of the 44 studied *Liolaemus* species

Trait	Males					Females				
	Non-phylogenetic			Phylogenetic		Non-phylogenetic			Phylogenetic	
	F	*df*	*P*	*F*-crit	*P*	*F*	*df*	*P*	*F*-crit	*P*
SVL	0.99	6,37	0.449	5.22	> 0.05	1.07	6,37	0.404	5.28	> 0.05
HL	2.09	6,37	0.069	4.86	> 0.05	1.96	6,37	0.089	4.79	> 0.05
HW	2.13	6,37	0.065	5.06	> 0.05	2.28	6,37	0.05	4.87	> 0.05
FLL	1.66	6,37	0.150	5.03	> 0.05	1.24	6,37	0.308	5.27	> 0.05
HLL	0.91	6,37	0.506	5.32	> 0.05	0.66	6,37	0.703	4.81	> 0.05
AXGL	1.08	6,37	0.394	5.17	> 0.05	0.85	6,37	0.552	4.84	> 0.05
TBL	0.30	6,37	0.948	5.19	> 0.05	0.76	6,37	0.620	5.13	> 0.05
FTL	2.89	6,37	**0.017**	4.47	> 0.05	1.24	6,37	0.310	4.60	> 0.05
PL	1.19	6,37	0.329	4.57	> 0.05	1.56	6,37	0.181	4.82	> 0.05
TLL	4.14	6,37	**0.012**	4.41	> 0.05	4.40	6,37	**0.009**	4.52	> 0.05

Non-phylogenetic *F*-values, degrees of freedom (*df*) and *P*-values were obtained from conventional statistical tests. Critical *F*-values (*F*-crit) for phylogenetic tests were obtained from PDANOVA (see "Materials and methods" section for details). Significant associations between microhabitat and phenotype are indicated in *bold*. Traits abbreviated as detailed in the main text

(Wilks's $\lambda = 0.42$, $F_{6,37} = 13.58$, $P < 0.001$; homogeneity of covariance matrices was confirmed with a Box's test, Box's $M = 28.49$, $P = 0.89$). The first three axes of a principal component analysis (PCA) extracted from the eight size-adjusted body shape variables (tail length excluded, see above) account for 84.1% of the body shape variation, where hind limb length (axis 1, 54.9% of the variance), pelvis width (axis 2, 17.5% of the variance) and axilla-groin length (axis 3, 11.7% of the variance) are the major explanatory axes. These factors plotted separately for males and females show the differential occupation of morphospace between the sexes revealed by the multivariate analysis of covariance (Fig. 2).

Discussion

Our results suggest that in *Liolaemus* lizards the regimes of natural selection imposed by structural microhabitats alone may not have exerted a significant impact on body size and shape variation in either of the sexes, in strong contrast with other highly diverse lizard lineages (Butler et al. 2007; Losos 1994; Losos et al. 1998; Stuart-Fox and Moussalli 2007). In *Liolaemus*, the exploitation of similar microhabitat types by different unrelated species has not promoted events of convergent evolution in the phenotypic characteristics explored in this study (i.e., no ecomorphs can be identified). Yet remarkably, our results show that the morphological trajectories followed by males and females often differ significantly in these lizards (Fig. 2). This finding supports Butler et al.'s (2007) claim that the common a priori assumption that males and females are ecologically or phenotypically equivalent (which often leads to analyze individuals of both sexes together, or to restrict analyses to only one of the two sexes) may provide an incomplete or even mistaken picture of the process of morphological diversification that takes place during adaptive radiations (see also Bolnick et al. 2003).

Lack of convergent evolution

The convergent evolution of ecologically relevant morphological traits (i.e., those targeted by natural selection) between phylogenetically unrelated species exploiting similar environments has often been regarded as evidence of adaptation (Grant 1986; Harvey and Pagel 1991; Herrel et al. 2002; Leal et al. 2002; Losos et al. 1998). However, some phylogenetic comparative analyses reveal that associations between ecology and morphology are often complex and difficult to detect. Hence, lack of convergence does not mean a lack of adaptive evolution, but may instead reflect selective mechanisms that conceal adaptation to any single pressure.

Some authors have argued that an explanation for the lack of relationships between ecological preferences and morphology observed in some clades might be that adaptations may have not yet been achieved (e.g., Vanhooydonck and Van Damme 1999). However, since functional adaptations are of critical importance to face major ecological challenges, such as predator avoidance or exploitation of different resources (Fox et al. 2001; Reilly et al. 2007; Schluter 2000; Wainwright and Reilly 1994), it is hard to accept that groups of species within a clade can exist in the absence of adaptations to their environments.

In a recent study, Schulte et al. (2004) suggested a series of factors that might explain why convergent evolution in morphology is not invariably observed in species exploiting similar niches. For example, different reproductive modes (e.g., viviparity and oviparity) might cause a differential effect of natural selection acting on ecologically similar species, resulting in the evolution of alternative morphological plans to exploit the same environment. Also, Schulte et al. (2004) suggested that alternative behavioral adaptations may play an important role in the adaptive directions followed by morphology. These authors argued that, for example, species that rely on crypsis to avoid predators might not evolve longer legs to increase running speed, unlike runner species. Since primarily cryptic and primarily runner species are often found occupying similar microhabitats (e.g., in *Liolaemus*), signals of morphological convergence might not be detected on the basis of microhabitat type alone (Schulte et al. 2004). However, even after taking these factors into account, convergences might still be hard to predict, for a number of reasons: (1) Natural selection regimes may vary substantially across different ecosystems (e.g., deserts vs. forests) containing similar microhabitat structures (e.g., rocks, grass patches). Hence, even if morphological convergence occurred in species with the same reproductive mode (e.g., viviparous) and microhabitat preference (e.g., rock-dwellers) living in similar ecosystems (e.g., all in forests), this convergence might be obscured if rock-dweller and viviparous species living in totally different ecosystems (e.g., deserts) were included in the same analysis. For example, predator foraging strategies and prey antipredatory responses may interact differently in ecosystems where dense vegetation (e.g., in forests) makes predator–prey visual contacts difficult, compared to entirely open ecosystems, such as deserts. Likewise, exposure of ecologically similar species to different thermal environments may result in the acquisition of alternative adaptations to exploit similar structural microhabitats, affecting the predictability of convergences in morphology. Testing this possibility would require intercontinental studies based on the comparative analyses of different clades living in similar ecosystems around the world (e.g., Melville et al. 2006). (2) Evolution is a multivariate process driven simultaneously by natural and sexual selection (Bell 2008; Coyne and Orr 2004; Shine et al. 1998). For example, in low predation environments, the expression of secondary sexual traits (i.e., sexual dimorphism and dichromatism) may be significantly released from natural selection, favoring the evolution by sexual selection of conspicuous traits involved in mating success (Andersson 1994; Endler 1986; Stuart-Fox and Ord 2004). Hence, if the environment experienced by species occupying similar microhabitat structures differs in natural selection pressures and in the opportunities for sexual selection, evolution of their morphologies might follow different adaptive trajectories (see Schulte et al. 2004). (3) Finally, direct selection on certain traits often affects the simultaneous expression of other traits not directly selected when the same genes (pleiotropy) or pairs of genes (linkage disequilibrium) determine the expression of two or more traits (Conner and Hartl 2004; Lynch and Walsh 1998). However, estimating the extent to which genetic correlations affect the expression of functional adaptations is difficult. At least one of these possibilities might explain the lack of convergent evolution observed here in *Liolaemus* lizards.

Sex-specific natural selection and sexual dimorphism

Frequency-dependent intraspecific competition for ecological resources and different parental, sexual, and reproductive roles between the sexes may favor the evolution of alternative phenotypic plans for conspecific males and females (Andersson 1994; Bolnick and Doebeli 2003; Darwin 1871; Fairbairn et al. 2007; Shine 1989). Such a pervasive component of evolutionary diversification has recently been illustrated by studies exploring the independent evolution of the sexes during adaptive radiation in a few groups of lizards, i.e., studies that do not assume a priori that both sexes are equivalent biological units (Butler 2007; Butler et al. 2007; Stuart-Fox and Moussalli 2007). For example, a significant proportion of the morphological diversity observed in *Anolis* is explained by sexual differences alone (Butler et al. 2007). Our analysis in *Liolaemus* reveals a similar situation. While microhabitat use appears not to be an important source of variation within the *Liolaemus* genus (in contrast to *Anolis* and chameleons), intersexual differentiation explains a significant proportion of the morphological variation produced by the evolutionary radiation of this taxon. These morphological differences are expressed not only in body mass (as intuitively expected in species with sexual dimorphism), but also in body shape (Fig. 2). These findings may therefore illustrate the role of sex-specific natural selection on the divergent adaptive trajectories followed by males and females during adaptive radiation.

Our study reinforces the view that since selection may often favor alternative phenotypic optima between the sexes, studies exploring the impact of selection on phenotypic evolution should not assume males and females as equivalent units (e.g., Bolnick et al. 2003; Butler et al. 2007). Finally, it is worth considering (as discussed earlier in relation to Schulte et al.'s 2004 observations) the idea that ecological challenges faced by conspecific males and females might result in behavioral adaptations that may reduce or replace intersexual adaptations at the morphological dimension (e.g., Kerr and Bull 2006). Therefore, integrative studies taking both ecological morphology and behavior into account might offer broader views of the adaptive response of sex-specific phenotypes to the impact of natural selection.

Acknowledgments We thank J. Cheverud, T. Garland, E. Martins, E. Rezende and B.I. Tieleman for valuable advice on phylogenetic comparative analyses. I.P.F. Owens provided useful suggestions. We also thank H. Núñez (MNHN), J. Artigas, M. Contreras and E. Solar (MZUC), J. Navarro (DBCGUCH), E. Pereyra (IBA-UNC), F. Videla (IADIZA), C. McCarthy (NHML), J.M. Cei (JMC-DC) and J.A. Scolaro (JASDC) for permission to study collections under their direction or for providing data from these samples. T. Garland supplied the programme PDAP for phylogenetic analyses. F.M. Jaksic, H. Núñez, and J.A. Scolaro sent essential literature. Two anonymous referees provided valuable criticism and interesting suggestions. DP-D thanks the financial support provided by Universities UK through an Overseas Research Student Award, the University of Exeter for Scholarships, Phrynosaura Chile and Oxford University Press. TT is funded by a Royal Society Research Fellowship. This work was supported by the European Social Fund.

Appendix

The studied material is housed in the following institutions. Collections identified with an asterisk (*) indicate the existence of specimens with collection data, but without official collection number at the moment of the study. Museo Nacional de Historia Natural, Chile (MNHN*), Zoological Museum, Universidad de Concepcion, Chile (MZUC*), Museo de Historia Natural de Concepcion, Chile (MHNC*), Department of Cell Biology and Genetics, Universidad de Chile (DBCGUCH*), Instituto de Biologia Animal, Universidad Nacional de Cuyo, Argentina (IBAUNC*), Instituto Argentino de Investigaciones de las Zonas Aridas, Argentina (IADIZA), Natural History Museum of London (NHML), Muséum National d'Histoire Naturelle, Paris (MNHNP), J. M. Cei Diagnostic Collection (JMC-DC), J. A. Scolaro Collection (JAS-DC), and in the Herpetological Collection of the senior author, D. Pincheira-Donoso (CHDPD*).

References

Andersson M (1994) Sexual selection. Princeton University Press, Princeton

Bell G (2008) Selection. The mechanism of evolution. Oxford University Press, Oxford

Beuttell K, Losos JB (1999) Ecological morphology of Caribbean anoles. Herpetol Monogr 13:1–28. doi:10.2307/1467059

Bolnick DI, Doebeli M (2003) Sexual dimorphism and adaptive speciation: two sides of the same ecological coin. Evolution 57:2433–2449

Bolnick DI, Svanbäck R, Fordyce JA, Yang LH, Davis JM, Hulsey CD, Forister ML (2003) The ecology of individuals: incidence and implications of individual specialization. Am Nat 161:1–28. doi:10.1086/343878

Butler MA (2007) Vive la difference! Sexual dimorphism and adaptive patterns in lizards of the genus *Anolis*. Integr Comp Biol 47:272–284. doi:10.1093/icb/icm061

Butler MA, Losos JB (2002) Multivariate sexual dimorphism, sexual selection, and adaptation in Greater Antillean *Anolis* lizards. Ecol Monogr 72:541–559

Butler MA, Schoener TW, Losos JB (2000) The relationship between sexual size dimorphism and habitat use in Greater Antillean *Anolis* lizards. Evolution 54:259–272

Butler MA, Sawyer SA, Losos JB (2007) Sexual dimorphism and adaptive radiation in *Anolis* lizards. Nature 447:202–205. doi:10.1038/nature05774

Cei JM (1986) Reptiles del centro, centro-oeste y sur de la Argentina. Herpetofauna de las zonas áridas y semiáridas. Museo Regionale di Scienze Naturali di Torino, Torino

Cei JM (1993) Reptiles del noroeste, nordeste y este de la Argentina. Herpetofauna de las selvas subtropicales, puna y pampas. Museo Regionale di Scienze Naturali di Torino, Torino

Conner JK, Hartl DL (2004) A primer of ecological genetics. Sinauer Associates, Massachusetts

Coyne JA, Orr HA (2004) Speciation. Sinauer Associates, Massachusetts

Darwin C (1871) The descent of man and selection in relation to sex. John Murray, London

Dieckmann U, Doebeli M, Metz JAJ, Tautz D (2004) Adaptive speciation. Cambridge University Press, Cambridge

Donoso-Barros R (1966) Reptiles de Chile. Ediciones Universidad de Chile, Santiago

Endler JA (1986) Natural selection in the wild. Princeton University Press, New Jersey

Espinoza RE, Wiens JJ, Tracy CR (2004) Recurrent evolution of herbivory in small, cold-climate lizards: breaking the ecophysiological rules or reptilian herbivory. Proc Natl Acad Sci USA 101:16819–16824. doi:10.1073/pnas.0401226101

Etheridge R (1995) Redescription of *Ctenoblepharys adspersa* Tschudi, 1845, and the taxonomy of Liolaeminae (Reptilia: Squamata: Tropiduridae). Am Mus Novit 3142:1–34

Etheridge R (2000) A review of lizards of the *Liolaemus wiegmannii* group (Squamata, Iguania, Tropiduridae), and a history of morphological change in the sand-dwelling species. Herpetol Monogr 14:293–352. doi:10.2307/1467049

Fairbairn DJ, Blanckenhorn WU, Szekely T (2007) Sex, size & gender roles. Evolutionary studies of sexual size dimorphism. Oxford University Press, Oxford

Felsenstein J (1985) Phylogenies and the comparative method. Am Nat 125:1–15. doi:10.1086/284325

Field A (2006) Discovering statistics using SPSS, 2nd edn. Sage, London

Fox CW, Roff DA, Fairbairn DJ (2001) Evolutionary ecology. Concepts and case studies. Oxford University Press, Oxford

Garcia-Berthou E (2001) On the misuse of residuals in ecology: testing regression residuals vs. the analysis of covariance. J Anim Ecol 70:708–711. doi:10.1046/j.1365-2656.2001.00524.x

Garland T, Dickerman AW, Janis CM, Jones JA (1993) Phylogenetic analysis of covariance by computer simulation. Syst Biol 42:265–292. doi:10.2307/2992464

Garland T, Bennett AF, Rezende EL (2005) Phylogenetic approaches in comparative physiology. J Exp Biol 208:3015–3035. doi:10.1242/jeb.01745

Gavrilets S (2004) Fitness landscapes and the origin of species. Princeton University Press, Princeton

Givnish TJ (1997) Adaptive radiation and molecular systematics: issues and approaches. In: Givnish TJ, Sytsma KJ (eds) Molecular evolution and adaptive radiation. Cambridge University Press, Cambridge, pp 1–54

Grant PR (1986) Ecology and evolution of Darwin's finches. Princeton University Press, New Jersey

Green AJ (2001) Mass/length residuals: measures of body condition or generators of spurious results? Ecology 82:1473–1483

Harvey PH, Pagel MD (1991) The comparative method in evolutionary biology. Oxford University Press, Oxford

Hayes JP, Shonkwiler JS (1996) Analyzing mass-independent data. Physiol Zool 69:974–980

Herrel A, Meyers JJ, Vanhooydonck B (2002) Relations between microhabitat use and limb shape in phrynosomatid lizards. Biol J Linn Soc Lond 77:149–163. doi:10.1046/j.1095-8312.2002.00101.x

Jackman T, Losos JB, Larson A, de Queiroz K (1997) Phylogenetic studies of convergent adaptive radiations in Caribbean *Anolis* lizards. In: Givnish TJ, Sytsma KJ (eds) Molecular evolution and adaptive radiation. Cambridge University Press, Cambridge, pp 535–557

Jaksic FM (1998) Ecología de los vertebrados de Chile. Ediciones de la Universidad Católica de Chile, Santiago

Jaksic FM, Núñez H, Ojeda FP (1980) Body proportions, microhabitat selection, and adaptive radiation of *Liolaemus* lizards in central Chile. Oecologia 45:178–181. doi:10.1007/BF00346457

Jolliffe IT (1972) Discarding variables in a principal component analysis, I: artificial data. Appl Stat 21:160–173. doi:10.2307/2346488

Jolliffe IT (1986) Principal component analysis. Springer, Berlin Heidelberg New York

Kerr GD, Bull CM (2006) Movement patterns in the monogamous sleepy lizard (*Tiliqua rugosa*): effects of gender, drought, time of year and time of day. J Zool 269:137–147

Leal M, Knox AK, Losos JB (2002) Lack of convergence in aquatic *Anolis* lizards. Evolution 56:785–791

Lee JC (1990) Sources of extraneous variation in the study of meristic characters: the effect of size and of inter-observer variability. Syst Zool 39:31–39. doi:10.2307/2992206

Losos JB (1990) Ecomorphology, performance capability, and scaling of West Indian *Anolis* lizards: an evolutionary analysis. Ecol Monogr 60:369–388. doi:10.2307/1943062

Losos JB (1994) Integrative approaches to evolutionary ecology: *Anolis* lizards as model systems. Annu Rev Ecol Syst 25:467–493. doi:10.1146/annurev.es.25.110194.002343

Losos JB, Miles DB (2002) Testing the hypothesis that a clade has adaptively radiated: Iguanid lizard clades as a case study. Am Nat 160:147–157. doi:10.1086/341557

Losos JB, Jackman TR, Larson A, De Queiroz A, Rodriguez-Schettino L (1998) Contingency and determinism in replicated adaptive radiations of island lizards. Science 279:2115–2118. doi:10.1126/science.279.5359.2115

Losos JB, Butler M, Schoener TW (2003) Sexual dimorphism in body size and shape in relation to habitat use among species of Caribbean *Anolis* lizards. In: Fox SF, McCoy JK, Baird TA (eds) Lizard social behaviour. John Hopkins University Press, Baltimore, pp 356–380

Lynch M, Walsh B (1998) Genetics and analysis of quantitative traits. Sinauer Associates, Massachusetts

Melville J, Harmon LJ, Losos JB (2006) Intercontinental community convergence of ecology and morphology in desert lizards. Proc Biol Sci 273:557–563

Miles DB, Ricklefs RE (1984) The correlation between ecology and morphology in deciduous forest passerine birds. Ecology 65:1629–1640. doi:10.2307/1939141

Miles DB, Sinervo B, Anthony FW (2000) Reproductive burden, locomotor performance, and the cost of reproduction in free ranging lizards. Evolution 54:1386–1395

Mosimann J (1970) Size allometry: size and shape variables with characterizations of the lognormal and generalized gamma distributions. J Am Stat Assoc 65:930–945. doi:10.2307/2284599

Olsson MM, Shine R, Bak-Olsson E (2000) Locomotor impairment of gravid lizards: is the burden physical or physiological? J Evol Biol 13:263–268. doi:10.1046/j.1420-9101.2000.00162.x

Pigliucci M, Preston K (2004) Phenotypic integration. Studying the ecology and evolution of complex phenotypes. Oxford University Press, Oxford

Pincheira-Donoso D, Núñez H (2005) Las especies chilenas del género Liolaemus. Taxonomía, sistemática y evolución. National Museum of Natural History Press, Santiago

Pincheira-Donoso D, Núñez H (2007) Conspecificity of *Liolaemus isabelae* Navarro & Núñez, 1993 and *Liolaemus nigroventrolateralis* Ortiz, 1994 (Iguania: Tropiduridae: Liolaeminae) from Northern Chile. Herpetol J 17:65–67

Pincheira-Donoso D, Scolaro JA, Schulte JAII (2007a) The limits of polymorphism in *Liolaemus rothi*: molecular and phenotypic evidence for a new species of the *Liolaemus boulengeri* clade (Iguanidae, Liolaemini) from boreal Patagonia of Chile. Zootaxa 1452:25–42

Pincheira-Donoso D, Tregenza T, Hodgson DJ (2007b) Body size evolution in South American *Liolaemus* lizards of the *boulengeri* clade: a contrasting reassessment. J Evol Biol 20:2067–2071. doi:10.1111/j.1420-9101.2007.01394.x

Pincheira-Donoso D, Hodgson DJ, Tregenza T (2008a) Comparative evidence for strong phylogenetic inertia in precloacal signalling glands in a species-rich lizard clade. Evol Ecol Res 10:11–28

Pincheira-Donoso D, Hodgson DJ, Tregenza T (2008b) The evolution of body size under environmental gradients in ectotherms: why should Bergmann's rule apply to lizards? BMC Evol Biol 8:68. doi:10.1186/1471-2148-8-68

Pincheira-Donoso D, Scolaro JA, Sura P (2008c) A monographic catalogue on the systematics and phylogeny of the South American iguanian lizard family Liolaemidae (Squamata, Iguania). Zootaxa 1800:1–85

Plummer MV (1997) Speed and endurance of gravid and nongravid green snakes, *Opheodrys aestivus*. Copeia 1997:191–194. doi:10.2307/1447857

Reilly SM, McBrayer LB, Miles DB (2007) Lizard ecology. Cambridge University Press, Cambridge

Rohlf FJ, Chang WS, Sokal RR, Kim J (1990) Accuracy of estimated phylogenies: effects of tree topology and evolutionary model. Evolution 44:1671–1684. doi:10.2307/2409346

Roughgarden J (1995) Anolis lizards of the Caribbean. Ecology, evolution, and plate tectonics. Oxford University Press, Oxford

Schluter D (1996) Ecological causes of adaptive radiation. Am Nat 148:S40–S64. doi:10.1086/285901

Schluter D (2000) The ecology of adaptive radiation. Oxford University Press, Oxford

Schulte JA, Macey JR, Espinoza RE, 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 Lond 69:75–102. doi:10.1006/bijl.1999.0346

Schulte JA, Losos JB, Cruz FB, Núñez H (2004) The relationship between morphology, escape behaviour and microhabitat occupation in the lizard clade *Liolaemus* (Iguanidae: Tropidurinae: Liolaemini). J Evol Biol 17:408–420. doi:10.1046/j.1420-9101.2003.00659.x

Scolaro JA (2006) Reptiles patagónicos norte. Una guía de campo. Editorial Universidad Nacional de la Patagonia, Comodoro Rivadavia

Shine R (1980) "Costs" of reproduction in reptiles. Oecologia 46:92–100. doi:10.1007/BF00346972

Shine R (1989) Ecological causes for the evolution of sexual dimorphism: a review of the evidence. Q Rev Biol 64:419–464. doi:10.1086/416458

Shine R, Keogh S, Doughty P, Giragossyan H (1998) Costs of reproduction and the evolution of sexual dimorphism in a 'flying lizard' *Draco melanopogon* (Agamidae). J Zool 246:203–213

Simpson GG (1953) The major features of evolution. Columbia University Press, New York

Slatkin M (1984) Ecological causes of sexual dimorphism. Evolution 38:622–630. doi:10.2307/2408711

Smith RJ (1999) Statistics of sexual size dimorphism. J Hum Evol 36:423–459. doi:10.1006/jhev.1998.0281

Stuart-Fox D, Moussalli A (2007) Sex-specific ecomorphological variation and the evolution of sexual dimorphism in dwarf chameleons (*Bradypodion* spp.). J Evol Biol 20:1073–1081. doi:10.1111/j.1420-9101.2007.01295.x

Stuart-Fox DM, Ord TJ (2004) Sexual selection, natural selection and the evolution of dimorphic coloration and ornamentation in agamid lizards. Proc Biol Sci 271:2249–2255

Vanhooydonck B, Van Damme R (1999) Evolutionary relationships between body shape and habitat use in lacertid lizards. Evol Ecol Res 1:785–805

Vitt LJ, Caldwell JP, Zani PA, Titus TA (1997) The role of habitat shift in the evolution of lizard morphology: evidence from tropical *Tropidurus*. Proc Natl Acad Sci USA 94:3828–3832. doi:10.1073/pnas.94.8.3828

Wainwright PC, Reilly SM (1994) Ecological morphology: integrative organismal biology. University of Chicago Press, Chicago

Williams EE (1983) Ecomorphs, faunas, island size, and diverse end points in island radiations of *Anolis*. In: Huey RB, Pianka ER, Schoener TW (eds) Lizard ecology: studies of a model organism. Harvard University Press, Massachusetts, pp 327–370

Zar JH (1999) Biostatistical analysis. Prentice-Hall, New Jersey

Zelditch ML, Swiderski DL, Sheets HD, Fink WL (2004) Geometric morphometrics for biologists. Elsevier, California