doi: 10.1111/j.1420-9101.2004.00758.x

Omnivory in lacertid lizards: adaptive evolution or constraint?

A. HERREL, B. VANHOOYDONCK & R. VAN DAMME

Department of Biology, University of Antwerp (UIA), Antwerp, Belgium

Keywords:

adaptation;
bite force;
constraint;
digestive tract;
lizard;
omnivory;
teeth;
trade-off.

Abstract

Feeding specializations such as herbivory are an often cited example of convergent and adaptive evolution. However, some groups such as lizards appear constrained in the evolution of morphological specializations associated with specialized diets. Here we examine whether the inclusion of plant matter into the diet of omnivorous lacertid lizards has resulted in morphological specializations and whether these specializations reflect biomechanical compromises as expected if omnivores are constrained by functional trade-offs. We examined external head shape, skull shape, tooth structure, intestinal tract length and bite performance as previous studies have suggested correlations between the inclusion of plants into the diet and these traits. Our data show that omnivorous lacertid lizards possess modifications of these traits that allow them to successfully exploit plant material as a food source. Conversely, few indications of a compromise phenotype could be detected, suggesting that the evolution towards herbivory is only mildly constrained by functional trade-offs.

Introduction

Some of the most compelling cases of adaptation are based on the convergent evolution of similar morphologies in similar ecological contexts (e.g. the independent evolution of wings in bats, birds and insects, or the evolution of grinding mechanisms in herbivorous mammals, dinosaurs and turtles; see e.g. Wainwright & Reilly, 1994; Schluter, 2000). In some cases, however, species that occupy similar environments or exploit similar ecological resources have not converged, or have converged only partially (Vanhooydonck & Van Damme, 1999; Leal *et al.*, 2002). The reasons for this lack of convergence, or 'incomplete' convergence vary, and may include hypotheses of historical contingency or constraint (Harvey & Pagel, 1991; Losos & Miles, 1994). Thus, data that shed light on whether species show morphological convergence, or not, in similar ecological settings, will also provide insights into more general evolutionary processes that underly the origin of phenotypic diversity.

Feeding specializations such as herbivory are an often cited example of convergent and adaptive evolution (e.g. King, 1996; Pérez-Barberia & Gordon, 1999; Reilly *et al.*, 2001). Indeed, many vertebrate groups such as mammals, turtles and archosaurs have independently evolved dental grinding mechanisms and modified intestinal tracts that allow them to efficiently utilize plant material as a food resource (see King, 1996 for an overview). However, when examining correlations between feeding morphology and diet at lower phylogenetic levels, and in an explicit phylogenetic context, many of the correlations are no longer significant (e.g. see Pérez-Barberia & Gordon, 2001). Strikingly, some groups such as lizards appear strongly constrained in the evolution of morphological specializations associated with specialized diets such as herbivory and apparently have not converged on the general morphology observed in other vertebrates (Greene, 1982; Schwenk, 2000). Yet in the past decade it has been demonstrated that the few known strictly herbivorous lizards (i.e. those lizards predominantly or exclusively consuming plants; <6% of all known lizard species) do show specializations that allow them to thrive

Correspondence: Anthony Herrel, Biology Department, University of Antwerp, Universiteitsplein 1, B-2610 Antwerp, Belgium.
Tel.: ++32 3820 2260; fax: ++32 3820 2271;
e-mail: aherrel@uia.ua.ac.be

on plants (see King, 1996; Cooper & Vitt, 2002 for an overview). For example, modifications of the intestinal tract (e.g. partioning of the colon to create fermentive chambers; see Iverson, 1980, 1982; Troyer, 1984; Foley *et al.*, 1992; Espinoza, 1999) and of the teeth (Hotton, 1955; Montanucci, 1968; Espinoza, 1999; but see Schwenk, 2000) have been demonstrated. Quite surprisingly, many species of lizards also occasionally include plant material in their diet (e.g. Greene, 1982; Van Damme, 1999; Schwenk, 2000; Cooper & Vitt, 2002). However, most studies examining these omnivorous lizards conclude that, besides being generally bigger, they are largely unspecialized (Szarski, 1962; Ostrom, 1963; Sokol, 1967; Pough, 1973; Van Damme, 1999; Cooper & Vitt, 2002). Most omnivores are thus considered opportunistic herbivores with a generalist morphology, lacking any obvious 'adaptations' to a partially herbivorous diet (Schwenk, 2000).

Previously, it has been proposed that the generalized morphology observed in these lizards allows them to efficiently exploit a wide diversity of food items, making them 'jacks of all diets masters of all' (Greene, 1982; Schwenk, 2000). If unspecialized omnivores are indeed capable of exploiting even challenging food items such as plants, this would eliminate the need for phenotypic specialization and could thus explain the lack of phenotypically specialized lizards. Alternatively, the transitional stage of omnivory might go hand in hand with functional trade-offs that may result in a decreased 'fitness' when compared with either of the specialized diets (i.e. insectivory vs. herbivory). If this intermediate stage is indeed constrained by functional trade-offs, then a 'compromise phenotype' can be expected with most traits showing a generalized morphology (Huey & Hertz, 1984) converging on neither the insectivorous nor the herbivorous morphologies. As a last possibility, omnivores might be more specialized than previously expected and may show clear functional adaptations to deal with tough and fibrous food items such as plants.

As the feeding system in lizards is a fairly simple biomechanical lever system, distinct predictions can be made regarding the specializations expected for insectivores and herbivores respectively (see below). Moreover, the nature of the system is such that the biomechanical demands for insectivory (fast jaw closing) are in direct conflict with those for herbivory (hard biting). Moreover, in analogy to the adaptations observed in herbivorous mammals and some specialist herbivorous lizards, we might expect differences between omnivores and insectivores in the digestive apparatus, resulting in an increased nutrient extraction and adsorption in omnivores. A previous study of the omnivorous Bonaire island whiptail lizard (Dearing, 1993) indicated that, whereas no specialized gut organs were present, the length of the intestine was significantly longer than that of a closely related insectivorous species, suggesting that increases in intestinal tract length might be one way to increase nutrient adsorption in omnivores. Additionally, large populations of nematodes (often suggested to play an important role in the breakdown of cellulose) were present in the intestine of this omnivorous lizard (Dearing, 1993).

The ability to crop smaller pieces from a plant is likely essential for an omnivore. Not only is it a physical prerequisite (unless the plant or plant part is eaten as a whole), but reducing larger parts of plants into smaller, bite-size pieces may also increase the digestive efficiency by increasing the surface area exposed to the digestive juices in the intestinal tract (Bjorndal *et al.*, 1990; Bjorndal & Bolten, 1992). Given the large forces needed to reduce many types of plant material such as stems and leaves (e.g. Herrel *et al.*, 1999b) and the smaller size of many of these omnivores compared to specialized herbivores (Greene, 1982; King, 1996, but see Van Damme, 1999; Cooper & Vitt, 2002), mechanical adaptations that allow them to reduce plant material are expected. As dental grinding mechanisms are absent in lizards (King, 1996; Herrel *et al.*, 1998a,b; Reilly *et al.*, 2001; but see Throckmorton, 1976 for a possible exception), an increase in bite force is probably the simplest way to improve their ability to mechanically reduce plant material. Moreover, an increase in bite force will not adversely affect the ability of omnivores to reduce insects. However, changes in the jaw system towards improved bite force ability might trade off with jaw closing velocity and thus prey capture efficiency, especially in species using jaw prehension such as lacertid lizards. If this is the case then we would expect changes in the morphology and mechanics of the jaw system to be neither towards improved bite force (short out-levers, large closing in-levers; tall, blunt skulls), nor towards fast jaw closing (long outlevers, long opening inlevers, long pointy skulls).

Constraints on bite force generation, however, might be partially circumvented by changes in dentition. Moreover, even when an increase in bite force is present, this would not be likely to improve the ability of an animal to crop plant matter without concomitant changes in tooth shape. Instead of the pointed, conical teeth typical of insectivorous lizards (Dessem, 1985; Mateo & Lopez-Jurado, 1997), medio-laterally flattened blade-like teeth are generally observed in herbivorous lizards (Hotton, 1955; Montanucci, 1968). Medio-laterally flattened teeth with a broad cutting edge may, however, be sub-optimal for killing and holding arthropods (see Mateo & Lopez-Jurado, 1997) and we might expect a generalized morphology if dentition is constrained.

In the present paper we test whether evolutionary changes in diet from insectivory to omnivory in lacertid lizards are accompanied by changes in the digestive tract and/or the jaw apparatus. We further test whether changes in bite force coincide with changes in head size or shape by comparing *in vivo* bite forces and morphometric data for lacertid lizards from different dietary groups (insectivores and omnivores). Finally, we investigate whether these changes in bite force are accompanied by concomitant changes in tooth shape in omnivores that would allow a more efficient cropping of plant material. Lacertid lizards are an ideal group to investigate morphological specializations in relation to the inclusion of plant matter into the diet as omnivores have arisen several times independently within this group (Van Damme, 1999). Moreover, previous studies investigating tooth shape in the partially herbivorous lacertid lizards of the genus *Gallotia* already suggested changes in dentition that were assumed to be associated with the partially herbivorous diet of these lizards (Mateo & Lopez-Jurado, 1992; Valido & Nogales, 2003).

Materials and methods

Animals

Most of the lacertid species used in this study were collected from various locations in Western Europe and the Canary Islands. An additional two species (*Acanthodactylus pardalis*, *Latastia longicaudata*), obtained from the pet trade were also included in the analysis. Where possible bite force measurements were taken at the field site (*Lacerta oxycephala*, *Podarcis melisellensis*, *Podarcis muralis*, *Podarcis tiliguerta*, *Lacerta vivipara*). The other species (*Lacerta bedriagae*, *Podarcis sicula*, *Lacerta viridis*, *Psammodromus algirus*, *Gallotia galloti*, *Podarcis hispanica*, *Podarcis atrata*, *Podarcis lilfordi*) were captured, transported to Belgium and measured in the lab. All animals were housed in groups of three or four and provided with food (insects and fruit dusted with calcium powder) and water *ad libitum*. Environmental temperature varied from 26 °C during daytime to 16 °C at night. An incandescent bulb provided a basking spot at higher temperatures (45 °C).

Head shape

The following measurements were taken from each animal used in the performance trials using digital calipers (±0.01 mm; Mitutoyo, Sakato, Japan): snout–vent length (SVL), head length (HL), head width (HW), and head height (HH). Head length was measured from the tip of the snout to the posterior edge of the parietal bone which can easily be detected by palpation in lacertid lizards. Head width was measured at the widest part of the skull and includes potential bulging of the jaw muscles, and head height was measured at the highest part of the skull just posterior to the orbita, and may again include bulging of the jaw adductors (m. pterygoideus). A summary of the morphometric data is presented in Appendix 1.

Bite forces

In vivo bite forces were measured using an isometric Kistler force transducer (type 9203; Kistler Inc., Wintherthur, Switzerland), mounted on a purpose-built holder and connected to a Kistler charge amplifier (type 5995; Kistler Inc.; see Herrel *et al.*, 1999a, 2001a, b for a more detailed description). Prior to, and inbetween, performance trials, the animals were placed in an incubator set at their preferred temperature (35 °C for most species, 38 °C for *A. pardalis* and *L. longicaudata*; see Bauwens *et al.*, 1995; Castilla *et al.*, 1999). When measured in the field, bite force was measured immediately after capture. Lizards were generally eager to bite when taken out of their bags or container. Where necessary, lizards were induced to bite by gently tapping the sides of their jaws. We tried to elicit defensive bites as these likely best represent a maximal effort (see Wainwright & Reilly, 1994 for the importance of using maximal effort data as measures of performance). The place of application of bite forces was standardized by mounting acrylic stops onto the free end of the holder (see Herrel *et al.*, 1999a, 2001a, b). Gape angle (opening of the jaws) was standardized (10 ± 5°) by moving the bite plates away from each other for larger animals. Measurements were repeated five times for each animal with an inter-trial interval of at least 30 min. The maximal value obtained during such a recording session, was considered to be the maximal bite force for that animal (see Table 1).

Skull and tooth dimensions

The skull dimensions and dentition of the lizards used in this study were examined on preserved specimens. For each species, the skulls of three adult male individuals (where available, see Appendix 2, 3) were cleaned by hand to expose the skull bones and toothrows. Several

Table 1 results of the phylogenetic ANCOVAS performed on the bite force and head shape data testing for differences between omnivores and insectivores.

Variable	d.f.	*F*	*P*	F_{phyl}	P_{phyl}
Males					
SVL	1, 13	1.33	0.27	2.59	0.15
Head length	1, 12	2.91	0.11	3.50	0.07
Head width	1, 12	1.40	0.26	3.04	0.16
Head height	1, 12	0.13	0.73	3.31	0.67
Bite force	1, 12	4.44	0.06	3.27	**0.03**
Females					
SVL	1, 13	0.83	0.380	2.83	0.27
Head length	1, 12	1.74	0.21	3.08	0.12
Head width	1, 12	1.80	0.20	2.83	0.11
Head height	1, 12	0.27	0.62	3.35	0.84
Bite force	1, 12	5.25	**0.04**	3.05	**0.01**

Bold values are significant at $\alpha = 0.05$ after sequential Bonferroni correction.

SVL, snout–vent length.

skull and lower jaw dimensions were measured using digital calipers (±0.01 mm; see Fig. 1): skull length (sl) was measured from the back of the parietal bone to the tip of the premaxilla; skull width (sw, not depicted on Fig. 1) was measured just posterior to the orbits, at the level of the posterior process of the jugal; skull height (sh) was measured at the highest part of the skull, again situated at the back of the orbits; lower jaw length (ljl) was measured from the back of retroarticular process to the tip of the dentary; coronoid height (ch) was measured as the height of the mandible at the level of the coronoid bone; the jaw closing in-lever (ci) was measured as the distance between the anterior aspect of the articular surface to the anterior aspect of the coronoid bone; the jaw opening in-lever (oi) was measured as the distance from the back of the retroarticular process to the anterior aspect of the articular surface; the jaw out-lever (ol) was measured as the distance from the anterior aspect of the articular surface to the anterior tip of the dentary.

To determine tooth characteristics, digital pictures were taken of the lateral aspect of the posterior teeth using an Olympus digital camera (Olympus Camedia C3030 zoom camera, Olympus Inc., Tokyo, Japan) attached to a Wild M3Z dissecting microscope (Wild Inc., Gais, Switzerland). A ruler was kept in the field of view for scaling purposes. Tooth shape was quantified for the three posterior most teeth of the upper jaw for each individual using Scion image for windows (V 4.02, Scion corporation; freely available on the internet at http://www.scioncorp.com). For each tooth the following variables were determined: the tooth height perpedicular to the jaw (th), tooth width halfway down the tooth (tw), the angle of the tip of the central, most prominent cusp (ta), and the number of cusps on each tooth (see

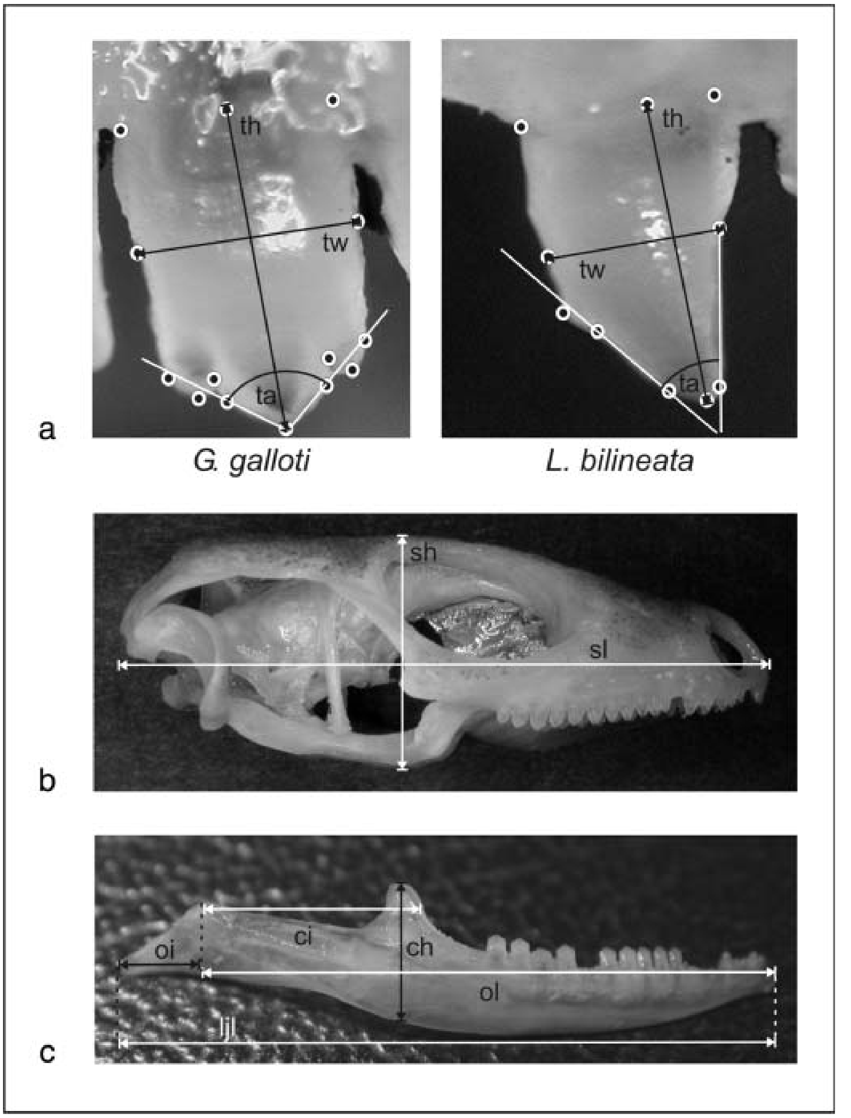

Fig. 1 Quantification of tooth and skull dimensions. (a) The height, width, perimeter, surface area and tip angle were quantified on digital pictures of the teeth in lateral view for three teeth of three individuals per species. On the left a tooth of a typical partial herbivore (*Gallotia galloti*) and on the right a tooth of a typical insectivore (*L. bilineata*) are depicted. (b) Lateral view on the skull of *G. galloti*. Skull length (sl) was determined from the back of the parietal bone to the tip of the premaxilla Skull height (sh) was determined just posterior to the orbits. (c) Lateral view on the mandible of *G. galloti*. Lower jaw length (ljl) was determined form the back of the retroarticular process to the tip of the dentary. Coronoid height (ch) was estimated by measuring the height of the mandible at the level of the coronoid bone. The jaw out-lever length (ol) was measured from the anterior aspect of the articular surface to the tip of the dentary. The jaw closing in-lever was measured form the anterior aspect of the articular surface to the anterior aspect of the coronoid bone. The jaw opening in-lever was measured from the back of the retroarticular process to the anterior aspect of the articular surface.

Fig. 1). Additionally, the tooth perimeter length and tooth surface area were determined by digitising landmarks around the perimeter of the tooth (see Fig. 1). For all traits, measurements were averaged across the three posteriormost teeth and across all individuals to provide a species mean. These measurements reflect tooth shape indicating whether teeth are tall, narrow and pointy as expected for insectivores or rather wide, low and with large surface areas and low cusp angles.

Length of the digestive tract

For five adult male individuals per species (where available, see Appendix 4), the length of the digestive tract and its subdivisions was determined. Preserved specimens were cut open mid-ventrally and the entire digestive tract was removed from the specimen. Connective tissue associated with the digestive tract was removed carefully and the tract was straightened without being stretched, and pinned onto a piece of cardboard. The length of the entire digestive tract was measured from the anteriormost point of the oesophagus to the posteriormost aspect of the rectum. The length of the oesophagus, the stomach, the small intestine, and the large intestine were also measured. No measurements were taken of the recta in the different species, as it was extremely hard to dissect out the terminal part of the intestinal without damaging it. Additionally, we measured the snout–vent length of all specimens. All measurements were taken using digital calipers (±0.01 mm). After measurements were taken, the hindgut of all was cut open mid-ventrally and inspected for the presence of valves or other structures that could function to slow down food passage. Additionally, we noted the presence of potential commensal organisms such as nematodes.

Feeding ecology

We used literature data to asses the diet of the species used in this analysis (see Van Damme, 1999). Species were classified as omnivores if all known populations as listed by Van Damme (1999) showed some degree of herbivory (more than 5% of the diet consisting of plant material). Based on this criterion, *G. galloti*, *P. lilfordi*, *P. atrata* and *L. bedriaga* were classified as omnivores (see also Castilla *et al.*, 1989; Castilla & Bauwens, 1991; Molina Borja, 1991; Pérez-Mellado & Corti, 1993; Valido & Nogales, 1994, 2003). All these species include both softer plant parts such as fruits as well as tough and fibrous elements such as leaves and stems into their diet. All other species were classified as insectivores eating a wide variety of arthropods (see Van Damme, 1999).

Analyses

As sexual head size dimorphism is prominent in lacertid lizards (Olsson & Madsen, 1998) and has been shown to affect bite force (Herrel *et al.*, 1996, 1999a, 2001a, b), analyses of bite force and head shape were performed on separate data sets for male and female lizards.

As closely related species share a large part of their evolutionary history, they cannot be considered independent data points (Felsenstein, 1985, 1988; Harvey & Pagel, 1991). To take the evolutionary history of the species into account, phylogenetic analyses were used. As these methods require information on the relationships between the species in the analysis, we constructed a current best tree (Fig. 2) based on a combination of morphological (Arnold, 1983, 1989, 1998) and molecular studies (Harris *et al.*, 1998; Harris & Arnold, 1999). The remaining polytomies in the tree were considered 'hard polytomies' as the result of rapid speciation events (Purvis & Garland, 1993; Garland & Diaz-Uriarte, 1999; Fu, 2000; Vanhooydonck & Van Damme, 2001). This approach should be considered conservative as analyses using a tree with resolution within the genus *Lacerta* (see

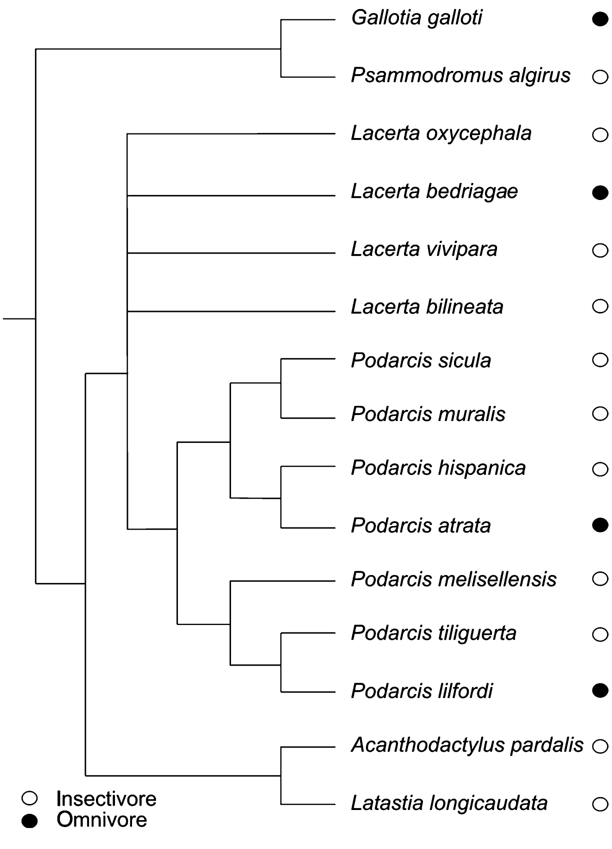

Fig. 2 Putative relationships among the 15 species of lacertid lizards used in this study. The tree is a current best estimate based on morphological and biochemical data (see Materials and methods). Open circles indicate insectivorous species; full circles indicate omnivorous species. Note that the polytomies should be considered 'hard' as they are likely the results of explosive radiation within the group.

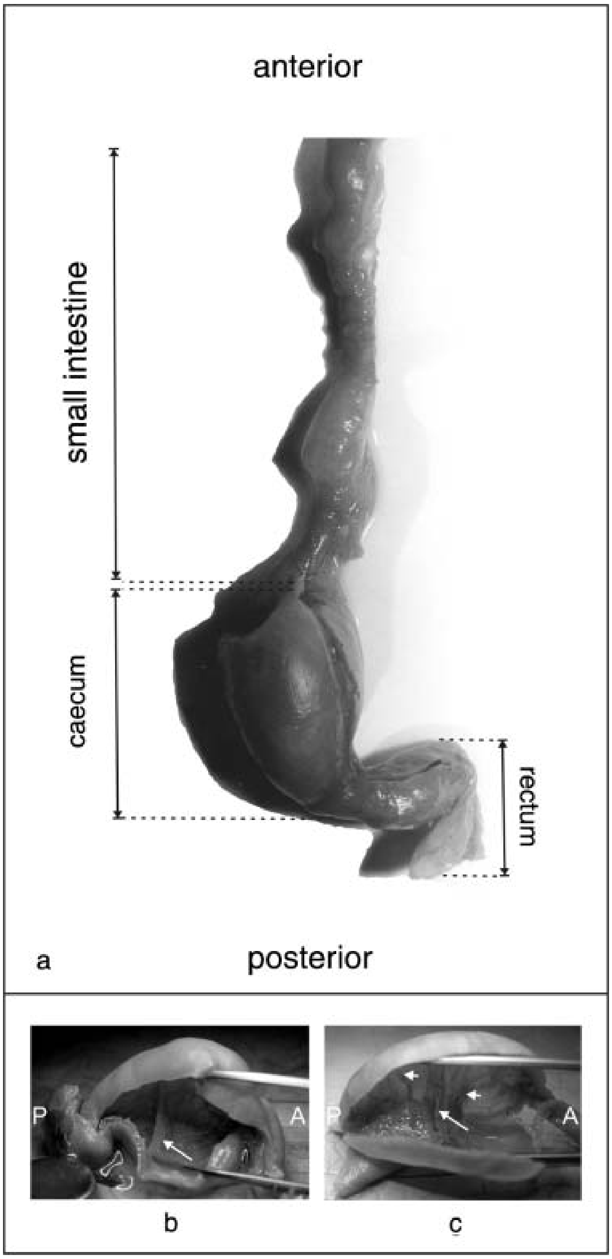

Fig. 3 (a) Picture of the posterior intestinal tract in *Gallotia galloti* showing the enlarged compartment in the large intestine. Distinct ridges and valves as well as large numbers of nematodes were observed inside this caecum-like compartment. (b) Close-up of the caecum-like compartment, cut open to illustrate the presence of the valve (arrow). (c) Close-up of the caecum-like compartment in another individual showing the valve (arrow) as well as several distinct ridges (arrow heads).

Fig. 3 in Harris *et al.*, 1998) resulted in higher phylogenetic *F*-values (see further).

All phylogenetic analyses were performed using the PDTREE or the PDSIMUL and PDANOVA programs (Garland *et al.*, 1999). In the analyses branch lengths were set to unity as no information is available concerning the divergence times of all the species included here (see also Diaz-Uriarte & Garland, 1998; Vanhooydonck & Van Damme, 2001). Analyses using random branch lengths gave the same results indicating that the effects of branch lengths on the outcome of the analyses are minimal. We inspected diagnostics graphs and statistics in the PDTREE program (Garland *et al.*, 1999) to verify that branch lengths were indeed appropriate for all traits (i.e. no correlation between the absolute values of the standardized contrasts and their standard deviations). Where necessary, branch lengths were transformed using the Pagel transformation (Garland *et al.*, 1999). The means of the head size and bite performance traits were calculated per sex and per species, and were Log_{10} transformed before further analysis. For all other traits (skull, intestinal tract lengths and tooth dimensions) species means were calculated and Log_{10} transformed before further analysis.

Simulation analyses were used to asses the differences in head size, bite force, tooth dimensions and intestinal tract length between species differing in feeding ecology (insectivores vs. omnivores). The PDSIMUL program (Garland *et al.*, 1999) was used to simulate trait evolution given the relationships among species as shown in Fig. 2. We ran 1000 unbounded simulations for each trait and used the Brownian motion model as our model for evolutionary change (see also Vanhooydonck & Van Damme, 1999). Next, the PDANOVA program was used to create a distribution of *F*-values based on the simulation data. Differences between groups were considered significant if the *F*-value of our analysis on the original data exceeded the 95% confidence limit extracted from the analysis preformed on the simulated data (F_{phyl}). As body size is known to affect morphological and performance traits (e.g. Valido & Nogales, 2003) analyses of covariance were used with the Log_{10} of snout–vent length as covariate. Regular ANOVA's were used to test for differences in snout–vent length.

To assess the evolutionary relationships between head shape and bite force in lacertid lizards, we used the PDTREE program to calculate independent contrasts of the means per sex and per species for the head size and bite force traits. Subsequently, we regressed the contrasts of all variables against the contrast of snout–vent length (forced through the origin, Garland *et al.*, 1992) and calculated residuals. Multiple regression techniques (backward) were used to examine associations between the residual contrast of bite force (dependent variable) and the residual contrasts of the head shape variables (again forced through the origin, Garland *et al.*, 1992).

Results

In the following paragraphs, only the results from the phylogenetically informed analyses will be discussed. In the tables both the nonphylogenetic (*F*) as well as the phylogenetically 'corrected' *F*-values (F_{phyl}) are given. With the exception of snout–vent length, analyses for all variables are based on analyses of co-variance with snout–vent length as co-variate.

Head shape and bite force

Simulation analyses showed that, in our sample, male and female omnivorous lacertid lizards are not bigger than insectivorous species (Table 1). Our data do show that omnivores bite harder than insectivores for a given body size (see Table 1, Appendix 1). Omnivores and insectivores, however, did not differ in external head shape (Table 1, Appendix 1). However, our regression analyses indicated that residual head dimensions were strongly intercorrelated for both male and female lizards (all $P < 0.05$). A multiple regression performed on the residual contrast data of head shape and bite force for male lizards retained a model with the residual contrast of head length and the residual contrast in head height ($F_{2,12} = 18.52$, $P < 0.001$; all other variables: $P > 0.05$). For females, a model with only the residual contrast of head width ($F_{1,13} = 6.47$, $P < 0.05$) was retained.

Skull and mandible dimensions

Of the skull and mandible dimensions examined only skull height was significantly different between the two dietary groups (Table 2, Appendix 2). Of all the other variables, only coronoid height approached significance ($P = 0.062$).

Tooth dimensions

Omnivorous lacertid lizards differed significantly in tooth shape from their insectivorous counterparts. Omnivores have wider teeth with a larger number of cusps associated (Table 3, Appendix 3). As a result, omnivores had a larger tooth perimeter and surface area when compared with insectivores (Table 3). Additionally, the sharpness of the main cusp on the teeth (as indicated by the tip angle) was significantly less in omnivores when compared with insectivores (Table 3).

Table 2 Results of the phylogenetic ANCOVAS performed on the skull shape data testing for differences between omnivores and insectivores.

Variable	d.f.	*F*	*P*	F_{phyl}	P_{phyl}
Skull length	1,12	0.32	0.58	3.25	0.35
Skull width	1,12	1.38	0.36	3.11	0.46
Skull height	1,12	9.52	**0.01**	3.01	**<0.01**
Lower jaw length	1,12	0.01	0.94	3.11	0.25
Coronoid height	1,12	2.87	0.12	3.21	0.062
Close in-lever	1,12	0.11	0.75	3.30	0.33
Open-inlever	1,12	0.10	0.75	3.12	0.33
Jaw out-lever	1,12	0.07	0.80	2.93	0.31

Bold values are significant at $\alpha = 0.05$ after sequential Bonferroni correction.

Table 3 Results of the phylogenetic ANCOVAS performed on the tooth data testing for differences between omnivores and insectivores.

Variable	d.f.	F	*P*	F_{phyl}	P_{phyl}
Tooth width	1, 12	5.85	**0.03**	3.18	**<0.01**
Tooth height	1, 12	2.52	0.14	3.49	0.081
Tooth perimeter	1, 12	4.46	0.06	2.98	**0.01**
Tooth surface area	1, 12	4.58	**0.05**	3.41	**0.02**
No. of cusps	1, 12	9.99	**<0.01**	3.76	**<0.01**
Main cusp angle	1, 12	12.34	**<0.01**	3.12	**<0.01**

Bold values are significant at $\alpha = 0.05$ after sequential Bonferroni correction.

Digestive tract length and morphology

Omnivores differed significantly from the other species examined in the overall length of the intestinal tract (Table 4, Appendix 4). Measurements of the different functional components of the intestinal tract showed that this is the result of differences in the length of the small intestine. Of the other functional compartments, only stomach length approached significance ($P = 0.08$). Dissections showed that the anterior part of the large intestine was distinctly enlarged in *G. galloti* (see Fig. 3, Table 5).

Table 4 Results of the phylogenetic ANCOVAS performed on the digestive tract length data testing for differences between omnivores and insectivores.

Variable	d.f.	F	*P*	F_{phyl}	P_{phyl}
Oesophagus	1,11	0.05	0.84	3.92	0.32
Stomach	1,11	2.97	0.11	3.89	0.08
Small intestine	1,11	15.28	**<0.01**	3.74	**<0.01**
Large intestine	1,11	2.18	0.17	3.58	0.11
Small + large intestine	1,11	14.14	**<0.01**	3.57	**<0.01**
Total gut length	1,11	8.12	**0.02**	3.92	**0.01**

Bold values are significant at $\alpha = 0.05$ after sequential Bonferroni correction.

Table 5 Morphometric characterization of the caecum in *Gallotia galloti*.

Individual	Caecum length	Caecum width	Caecum height	Total gut length
1	22.2 (11.1)	11.1 (5.5)	9.8 (4.9)	200.3
2	57.7 (15.0)	18.2 (4.7)	10.4 (2.7)	383.9
3	23.4 (10.6)	11.8 (5.4)	10.0 (4.5)	220.5
4	10.0 (10.3)	5.9 (6.1)	3.3 (3.4)	97.1
5	19.5 (10.1)	12.5 (6.5)	9.9 (5.2)	192.4
Average ± SD	26.6 ± 18.2	11.9 ± 4.4	8.67 ± 3.0	218 ± 103.85

Values are expressed in mm (%).

The caecum-like compartment showed distinct ridges and valves (at least one, see Fig. 3) that resulted in an enlarged surface area, and could potentially function to slow down food passage. All five *G. galloti* specimens examined also had large numbers of nematodes in this compartment.

Discussion

Previous reviews of lizard feeding behaviour have asserted that phenotype and diet generally show no predictable association among lizards (e.g. Greene, 1982; Schwenk, 2000). Whereas such a lack of functional specialisation could be interpreted as the result of conflicting demands imposed by different diets on the morphology and function of the feeding system, morphological specializations resulting in an improved ability to deal with plant matter as a food resource would suggest that functional trade-offs have not constrained the evolution of omnivory in lacertid lizards. Our analyses indicate significant variation in morphology and performance associated with an omnivorous diet in lacertid lizards. As demonstrated here, omnivorous lacertid lizards have converged on a similar morphology that accords well with our *a priori* biomechanical predictions for omnivory. Omnivorous lacertids had higher skulls, tended to have taller coronoid processes (albeit not significant), together likely affecting bite force, which was also significantly higher in omnivores. Additionally, whereas omnivores had significantly wider teeth with more cusps, insectivores had sharper, narrower teeth. Also the overall length of the digestive tract differed between omnivores and insectivores when taking body size into account. However, our data indicate that is purely the result of an elongated small intestine in omnivores.

An examination of the traits included in our analyses suggests that the most 'herbivorous' species included in our analyses is also functionally the most specialized one. Not only does *G. galloti* have the highest residual bite forces of all omnivores examined, it also has a distinct caecum-like compartment in the intestinal tract. As the inclusion of this specialist might potentially bias the overall results of our analyses, we re-analysed all our data in an explicit phylogenetic context exclusing the data for *G. galloti*. The results of these analyses show that the other omnivores still differ significantly from the insectivores for all traits (all $P < 0.05$), with the exception from the *in vivo* bite forces. Bite forces became marginally nonsignificant after excluding data for *G. galloti* for both male ($F = 2.35$; $F_{\mathrm{phyl}} = 3.80$; $P = 0.094$) and female ($F = 2.57$; $F_{\mathrm{phyl}} = 3.47$; $P = 0.085$) lizards. These data suggest that although *G. galloti* is indeed highly specialized, the majority of the results still hold. Thus omnivorous lacertids are generally indeed quite different from their insectivorous counterparts.

The differences in intestinal tract length observed for omnivorous lacertids accord well with what was observed for the lizard *Cnemidophorus murinus* (Dearing, 1993). In the latter species, the intestine was significantly longer than that of a closely related insectivorous species. Also, in omnivorous frogs an increase in absolute (but not relative) length of the digestive tract was observed (Das, 1995). Our data indicate that the longer intestinal tract is purely the result of an elongation of the small intestine. None of the other compartments were longer in omnivores when compared with insectivores. Interestingly, the stomach was the only other functional component showing a tendency towards being longer in omnivores. These data indicate that an increase of the surface area of the physiologically active tissues may be a way to improve the digestion of plant matter by omnivores. A comparison of our data with the data presented in Valido & Nogales (2003) indicates that whereas omnivores appear to differ in intestinal tract length from insectivores (present study), populations of lizards that are more herbivorous than others do not differ in the relative length of the intestinal tract. Although Valido & Nogales (2003) do not report the length of the small intestine specifically (allowing for cryptic differences in the length of the small intestine not detected by their analyses of overall length), this suggests that it might be the initial inclusion of plant matter into the diet that drives the evolution of the increase in intestinal tract length. Additionally, our data on the structure of the intestinal tract in *G. galloti*, the most herbivorous of the four omnivores studied here, indicated a distinct widening of the posterior part of the large intestine (Table 5). This compartment closely resembles the 'caeca' observed in strictly herbivorous lizards (e.g. Iverson, 1980, 1982) and contained distinct ridges and valve-like structures (see Fig. 3). Additionally, large numbers of nematodes were observed in the caecum-like compartment of this species. This suggests that the higher degree of herbivory in this species (up to 80% in some populations; see Valido & Nogales, 1994, 2003) has lead to the independent evolution of a caecum-like structure in this group.

The analyses of the *in vivo* bite force data, showed that omnivorous lacertid lizards bite harder than insectivorous species. Given the large forces needed to crop plant material such as leaves (much larger than those needed to crush arthropods of similar size; see Herrel *et al.*, 1999b), these results show that bite force is an important performance variable which, most likely, allows these lizards to exploit a novel food resource. As herbivorous lizards extensively 'chew' their food before swallowing (see Herrel & De Vree, 1999; Herrel *et al.*, 1999b) large bite forces will be important in increasing the surface area available to digestive juices. Rather unexpectedly, no differences in head shape or size were observed between omnivores and insectivores. This is even more unexpected given the strong correlations between head dimensions and bite force in lacertid lizards (see results, Fig. 4). Yet, in contrast to the external head dimensions, distinct differences in skull shape were observed. Not only did omnivores have relatively higher skulls, they

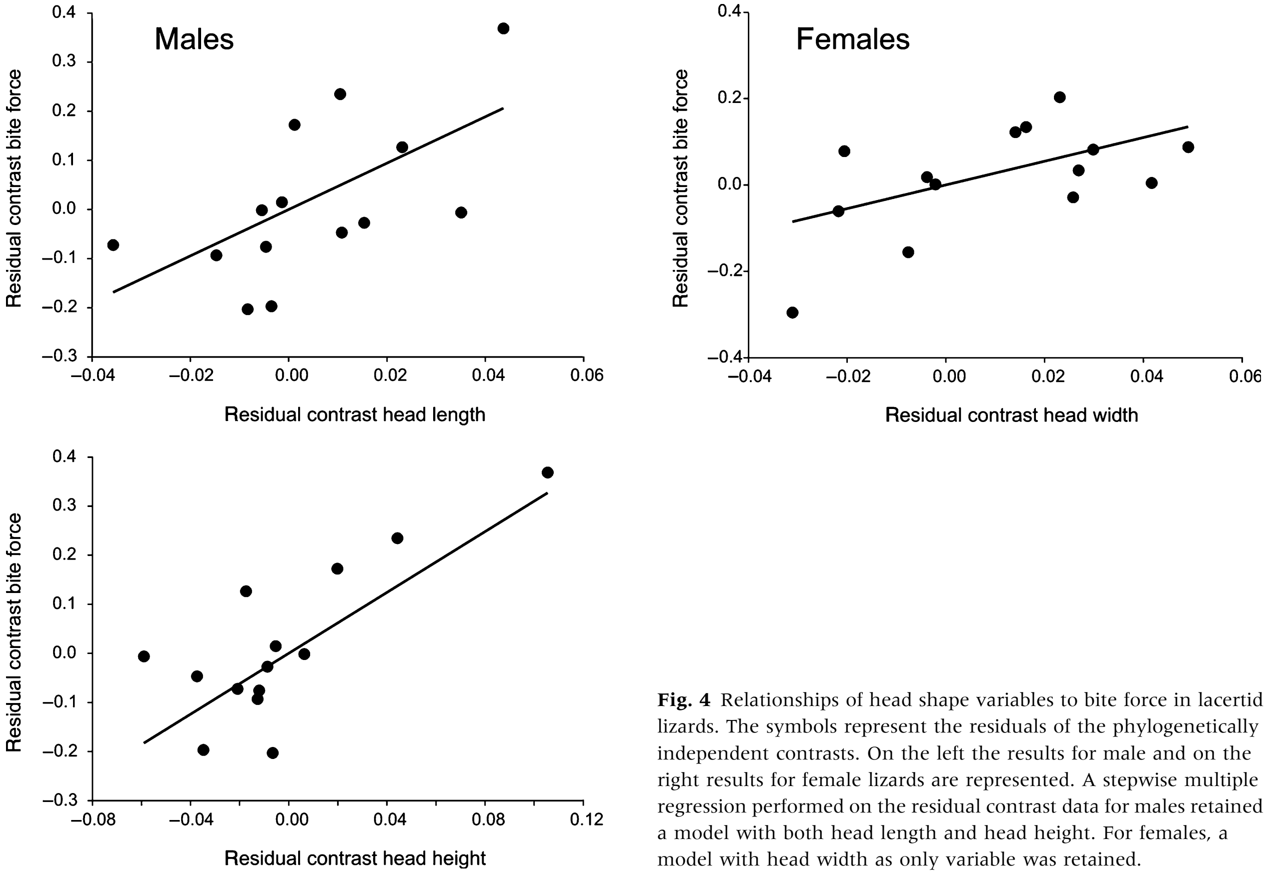

Fig. 4 Relationships of head shape variables to bite force in lacertid lizards. The symbols represent the residuals of the phylogenetically independent contrasts. On the left the results for male and on the right results for female lizards are represented. A stepwise multiple regression performed on the residual contrast data for males retained a model with both head length and head height. For females, a model with head width as only variable was retained.

also tended to have a higher coronoid processus on the lower jaw. Both of these features can be functionally linked to bite force. Higher skulls allow for a more perpendicular orientation of the jaw adductors, which results in the generation of higher torques for a given muscle mass. The higher coronoid, on the contrary provides a larger area of attachment for the jaw adductors. The absence of differences in the lever arms of the jaw system (Table 2) suggests little intrinsic changes in the biomechanics of jaw closing in omnivores compared with insectivores. Although the lack of differences in the lever system may reflect a constraint on jaw closing speed needed to grab elusive prey such as insects, this remains to be investigated further. Besides the observed changes in skull shape, changes in muscle mass or architecture (e.g. muscle orientation, degree of pennation, fibre length) or in the muscle recruitment patterns may also have occurred in the evolution towards omnivory in lacertid lizards. Whereas such changes have been documented for some of the omnivores included in this study (Herrel *et al.*, 1996, 1999a), detailed morphological and biomechanical analyses of the jaw system in a wider range of species are needed to investigate this further.

Large bite forces may, however, be inadequate to reduce or crop plant material without a specialized tooth structure. Whereas the typical lacertid dentition consists of simple, conical pointed teeth ideal for puncturing the chitinous exoskeleton of insects (see Barahona, 1996; Fig. 1a – right panel), to crop plants a knife-edged tooth row is required (see also Lucas & Luke, 1984). In specialist herbivores this is associated with the presence of multiple cusps (Valido & Nogales, 2003), and a medio-lateral flattening of the teeth (Hotton, 1955; Montanucci, 1968). Our analyses indicated that the teeth in omnivorous lacertid lizards are wider, have more cusps, a larger surface area and tooth perimeter (see also Mateo & Lopez-Jurado, 1992). Interestingly, our data also show that the angle of the central cusp is significantly smaller (increasing the sharpness of the tooth) in insectivores. Tooth shape thus seems to be a good indicator for omnivory and herbivory in lizards as tooth morphology apparently evolves predictably, even in response to only a partial use of plants as a food resource. At least in lacertids, dentition seems to change in response to the physical demands of the breakdown of different food items (see also Mateo & Lopez-Jurado, 1997).

Despite their suspected unspecialized nature, omnivorous lacertid lizards show distinct adaptations that allow them to successfully exploit plant material as a food source. Conversely, no indications of a compromise phenotype (as would be suggested by differences among groups not in the direction of our *a priori* predictions) could be detected, suggesting that the evolution towards herbivory is apparently not constrained by functional trade-offs. The lack of changes in the jaw lever system, on the contrary, suggests that omnivores have retained the ancestral morphology associated with specialized insectivores. Whereas this would be expected to be suboptimal for the generation of high bite forces, omnivores have apparently circumvented this constraint by having higher skulls and presumably by having an absolutely larger jaw adductor mass. However, this remains speculative at this point and needs to be tested by quantitative analyses of jaw adductor mass in these species. In conclusion, increases in bite force, in skull shape, a modified dentition and an elongated intestine all seem to be crucial elements in the evolution towards omnivory in these species. Whether our findings can be generalized to other lizard families remains to be tested.

Acknowledgments

The authors would like to thank D. Bauwens for allowing us to measure bite performance of the lizards in his care; A. Castilla for providing us with preserved specimens of *P. atrata*; M. Baez for providing us with preserved specimens of *G. galloti* and Irena Grbac and Svjetlana Lupret-Obradovic for valuable field assistance in Croatia. AH and BV are postdoctoral fellows of the Fund for Scientific Research – Flanders, Belgium (FWO-Vl).

Supplementary material

The following material is available from http://www.blackwellpublishing.com/products/journals/suppmat/jeb/jeb758/jeb758sm.htm

Appendix A1 Species, morphological and performance measures for each sex of the 15 species used in this study.

Appendix A2 Skull dimensions of the 15 species of lacertid lizards used in this study.

Appendix A3 Tooth shape in the 15 species of lacertid lizards used in this study.

Appendix A4 Length of the intestinal tract and its components in the 15 species of lacertid lizards used in this study.

References

Arnold, E.N. 1983. Osteology, genitalia and the relationships of *Acanthodactylus* (Reptilia: Lacertidae). *Bull. Brit. Mus. Nat. Hist. (Zool.)* **44**: 291–339.

Arnold, E.N. 1989. Towards a phylogeny and biogeography of the Lacertidae: relationships within an old-world family of lizards derived from morphology. *Bull. Brit. Mus. Nat. Hist. (Zool.)* **44**: 291–339.

Arnold, E.N. 1998. Structural niche, limb morphology and locomotion in lacertid lizards (Squamata, Lacertidae); a preliminary survey. *Brit. Mus. Nat. Hist. (Zool.)* **55**: 209–257.

Barahona, F.F. 1996. *Osteologia craneal de lacértidos de la peninsula ibérica e islas canarias: analisis sistematico filogenético*, Unpublished PhD thesis. Universidad Autonoma de Madrid, Madrid.

Bauwens, D., Garland, T. Jr., Castilla, A.M. & Van Damme R.1995. Evolution of sprint speed in lacertid lizards: morphological, physiological and behavioral covariation. *Evolution* **49**:848–863.

Bjorndal, K.A. & Bolten, A.B. 1992. Body size and digestive efficiency in a herbivorous freshwater turtle: advantages of a small bite size. *Physiol. Zool.* **65**: 1028–1039.

Bjorndal, K.A., Bolten, A.B. & Moore, J.E. 1990. Digestive fermentation in herbivores: effect of food particle size. *Physiol. Zool.* **63**: 710–721.

Castilla, A.M. & Bauwens, D. 1991. Observations on the natural history, present status, and conservation of the insular lizard *Podarcis hispanica atrata* on the Columbretes Archipelago, Spain. *Biological Conservation* **58**: 69–84.

Castilla, A.M., Bauwens, D., Van Damme, R. & Verheyen, R.F. 1989. Notes on the biology of the high altitude lizard *Lacerta bedriagae*. *Herpetological Journal* **1**: 400–403.

Castilla, A.M., Van Damme, R. & Bauwens, D. 1999. Field body temperatures, mechanisms of thermoregulation and evolution of thermal characteristics in lacertid lizards. *Natura Croatica* **8**: 253–274.

Cooper, W.E. Jr. & Vitt, L.J. 2002. Distribution, extent, and evolution of plant consumption by lizards. *J. Zool., Lond.* **257**: 487–517.

Das, I. 1995. Comparative morphology of the gastrointestinal tract in relation to diet in frogs from a locality in South India. *Amphibia Reptilia* **16**: 289–293.

Dearing, M.D. 1993. An alimentary specialisation for herbivory in the tropical whiptail lizard *Cnemidophorus murinus*. *J. Herpetol.* **27**: 111–114.

Dessem, D. 1985. Ontogenetic changes in the dentition and diet of *Tupinambis* (Lacertilia: Teiidae). *Copeia* **1985**: 245–247.

Diaz-Uriarte, R. & Garland, T. Jr. 1998. Effects of branch length errors on the performance of phylogenetically independent contrasts. *Syst. Biol.* **47**: 654–672.

Espinoza, R.E. 1999. Is evolution progressive? Insights from herbivorous lizards. *Am. Zool.* **39**: 11A.

Felsenstein, J. 1985. Phylogenies and the comparative method. *Am. Nat.* **125**: 1–15.

Felsenstein, J. 1988. Phylogenies and quantitative characters. *Ann. Rev. Ecol. Syst.* **19**: 445–472.

Foley, W.J., Bousila, A., Shkolnik, A. & Chosniak, I. 1992. Microbial digestion in the herbivorous lizard *Uromastyx aegyptius* (Agamidae). *J. Zool., Lond.* **226**: 387–398.

Fu, J. 2000. Toward the phylogeny of the family Lacertidae – why 4708 base pairs of mtDNA sequences cannot draw the picture. *Biol. J. Linn. Soc.* **71**: 203–217.

Garland, T. Jr. & Diaz-Uriarte, R. 1999. Polytomies and phylogenetically independent contrasts: examination of the bounded degrees of freedom approach. *Syst. Biol.* **48**: 547–558.

Garland, T. Jr., Harvey, P.J. & Ives, A.R. 1992. Procedures for the analysis of comparative data using phylogenetically independent contrasts. *Syst. Biol.* **41**: 18–32.

Garland, T. Jr., Midford, P.E. & Ives, A.R. 1999. An introduction to phylogenetically based statistical methods, with a new method for confidence intervals on ancestral states. *Am. Zool.* **39**: 374–388.

Greene, H.W. 1982. Dietary and phenotypic diversity in lizards: why are some organisms specialised? In: *Environmental Adap-*

tation and Evolution (D. Mossakowski & G. Roth, eds), pp. 107–128. Fischer Verlag, Stuttgart.

Harris, D.J. & Arnold, E.N. 1999. Relationships of wall lizards, *Podarcis* (Reptilia: Lacertidae) based on mitochondrial DNA sequences. *Copeia* **1999**: 749–754.

Harris, D.J., Arnold, E.N. & Thomas, R.H. 1998. Rapid speciation, morphological evolution, and adaptation to extreme environments in South African sand lizards (*Meroles*) as revealed by mitochondrial gene sequences. *Mol. Phylogen. Evol.* **10**: 37–48.

Harvey, P.H. & Pagel, M.D. 1991. *The Comparative Method in Evolutionary Biology*. Oxford University Press, New York.

Herrel, A. & De Vree, F. 1999. Kinematics of intraoral transport and swallowing in the herbivorous lizard *Uromastix acanthinurus*. *J. Exp. Biol.* **202**: 1127–1137.

Herrel, A., Van Damme, R. & De Vree, F. 1996. Testing the niche divergence hypothesis by bite force analysis. *Neth. J. Zool.* **46**: 253–262.

Herrel, A., Aerts, P. & De Vree, F. 1998a. Static biting in lizards with a primitive skull: functional morphology of the temporal ligaments. *J. Zool., Lond.* **244**: 135–143.

Herrel, A., Aerts, P. & De Vree, F. 1998b. Ecomorphology of the lizard feeding apparatus: a modelling approach. *Neth. J. Zool.* **45**: 1–25.

Herrel, A., Spithoven, L., Van Damme, R. & De Vree, F. 1999a. Sexual dimorphism of head size in *Gallotia galloti*; testing the niche divergence hypothesis by functional analyses. *Funct. Ecol.* **13**: 289–297.

Herrel, A., Verstappen, M. & De Vree, F. 1999b. Modulatory complexity of the feeding repertoire in scincid lizards. *J. Comp. Physiol. A.* **184**: 501–518.

Herrel, A., Van Damme, R., Vanhooydonck, B. & De Vree, F. 2001a. The implications of bite performance to diet in two species of lacertid lizards. *Can. J. Zool.* **79**: 662–670.

Herrel, A., De Grauw, E. & Lemos-Espinal, J.A. 2001b. Head Shape and bite performance in xenosaurid lizards. *J. Exp. Zool.* **290**: 101–107.

Hotton, N. 1955. A survey of adaptive relationships of dentition to diet in North American Iguanidae. *Am. Midl. Nat.* **55**: 88–114.

Huey, R.B. & Hertz, P.E. 1984. Is a jack of all temperatures master of none? *Evolution* **38**: 441–444.

Iverson, J.B. 1980. Colic modifications in iguanine lizards. *J. Morphol.* **163**: 79–93.

Iverson, J.B. 1982. Adaptations to herbivory in iguanine lizards. In: *Iguanas of the World* (G. M. Burghardt & A. S. Rand, eds), pp. 60–76. Noyes, NJ, USA.

King, G. 1996. *Reptiles and Herbivory*. Chapman and Hall, London.

Leal, M., Knox, A.K. & Losos, J.B. 2002. Lack of convergence in aquatic *Anolis* lizards. *Evolution* **56**: 785–791.

Losos, J.B. & Miles, D.B. 1994. Adaptation, constraint and the comparative method: phylogenetic issues and methods. In: *Ecological Morphology* (P. C. Wainwright & S. M. Reilly, eds), pp. 60–98. University of Chicago Press, Chicago.

Lucas, P.W. & Luke, D.A. 1984. Basic principles of food breakdown. In: *Food Aquisition and Processing in Primates* (D. A. Chivers, B. A. Woods & A. P. Møller, eds), pp. 283–301. Plenum Press, New York.

Mateo, J.A. & Lopez-Jurado, L.F. 1992. Study of dentition in lizards from Gran Canaria island (Canary Islands) and its ecological and evolutionary significance. *Biol. J. Linn. Soc.* **46**: 39–48.

Mateo, J.A. & Lopez-Jurado, L.F. 1997. Dental ontogeny in *Lacerta lepida* (Sauria, Lacertidae) and its relationship to diet. *Copeia* **1997**: 461–463.

Molina Borja, M. 1991. Notes on the alimentary habits and spatio-temporal distribution of eating patterns in a natural population of lizards (*Gallotia galloti*). *Vieraea* **20**: 1–9.

Montanucci, R.R. 1968. Comparative dentition in four iguanid lizards. *Herpetologica* **24**: 305–315.

Olsson, M. & Madsen, T. 1998. Sexual selection and sperm competition in reptiles. In: *Sperm Competition and Sexual Selection* (T. R. Birkhead & A. P. Møller, eds), pp. 503–610. Academic Press, London.

Ostrom, J.H. 1963. Further comments on herbivorous lizards. *Evolution* **17**: 368–369.

Pérez-Barberia, F.J. & Gordon, I.J. 1999. The functional relationship between feeding type and jaw and cranial morphology in ungulates. *Oecologia* **118**: 157–165.

Pérez-Barberia, F.J. & Gordon, I.J. 2001. Relationships between oral morphology and feeding style in the Ungulata: a phylogenetically controlled evaluation. *Proc. R Soc. Lond. B.* **268**: 1023–1032.

Pérez-Mellado, V. & Corti, C. 1993. Dietary adaptations and herbivory in lacertid lizards of the genus *Podarcis* from western Mediterranean Islands (Reptilia: Sauria). *Bonn. Zool. Beitr.* **44**: 193–220.

Pough, F.H. 1973. Lizard energetics and diet. *Ecology* **54**: 838–844.

Purvis, A. & Garland, T. Jr. 1993. Polytomies in comparative analyses of continuous characters. *Syst. Biol.* **42**: 569–575.

Reilly, S.M., McBrayer, L.D. & White, T.D. 2001. Prey processing in amniotes: biomechanical and behavioral patterns of food reduction. *Comp. Biochem. Physiol. A.* **128**: 397–415.

Schluter, D. 2000. *The Ecology of Adaptive Radiation*. Oxford University Press, Oxford.

Schwenk, K. (ed.) 2000. Feeding in lepidosaurs. In: *Feeding: Form, Function, and Evolution in Tetrapod Vertebrates*, pp. 175–291. Academic Press, San Diego.

Sokol, O.M. 1967. Herbivory in lizards. *Evolution* **21**: 192–194.

Szarski, H. 1962. Some remarks on herbivorous lizards. *Evolution* **16**: 529.

Throckmorton, G.S. 1976. Oral food processing in two herbivorous lizards, *Iguana iguana* (Iguanidae) and *Uromastix aegyptius* (Agamidae). *J. Morphol.* **148**: 363–390.

Troyer, K. 1984. Structure and function of the digestive tract of a herbivorous lizard *Iguana iguana*. *Physiol. Zool.* **57**: 1–8.

Valido, A. & Nogales, M. 1994. Frugivory and seed dispersal by the lizard *Gallotia galloti* (Lacertidae) in a xeric habitat of the Canary Islands. *Oikos* **70**: 403–411.

Valido, A. & Nogales, M. 2003. Digestive ecology of two omnivorous Canarian lizard species (*Gallotia*, Lacertidae). *Amphibia-Reptilia* **24**: 331–344.

Van Damme, R. 1999. Evolution of herbivory in lacertid lizards: effects of insularity and body size. *J. Herpetol.* **33**: 663–674.

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

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.

Wainwright, P.C. & Reilly, S.M. 1994. *Ecological Morphology*. University of Chicago Press, Chicago.

Received 26 March 2004; accepted 4 April 2004