

Are morphology–performance relationships invariant across different seasons? A test with the green anole lizard (*Anolis carolinensis*)

Duncan J. Irschick, Margarita Ramos, Christine Buckley, Justin Elstrott, Elizabeth Carlisle, Simon P. Lailvaux, Natasha Bloch, Anthony Herrel and Bieke Vanhooydonck

Irschick, D. J., Ramos, M., Buckley, C., Elstrott, J., Carlisle, E., Lailvaux, S. P., Bloch, N., Herrel, A. and Vanhooydonck, B. 2006. Are morphology–performance relationships invariant across different seasons? A test with the green anole lizard (*Anolis carolinensis*). – *Oikos* 114: 49–59.

A key assumption in ecomorphological studies is that morphology–function relationships are invariant due to underlying biomechanical principles. We tested the hypothesis that morphology–performance relationships are invariant across different seasons by examining how a key performance trait, bite force, and two aspects of morphology (head shape and dewlap size) changed seasonally in the field and in the laboratory in the green anole lizard *Anolis carolinensis*. We found that not only did bite force change seasonally (up to 80% within the same individual), but relationships between morphology and bite force are highly plastic. Of the three traits examined (bite force, head shape, and dewlap area), only head shape did not change seasonally. We noted opposing trends for how bite force and dewlap area changed seasonally; whereas dewlap areas were large in the spring, and small in the winter, bite forces were low in the spring and high in the winter. This pattern occurred because of a tradeoff at the individual level: individuals in the spring with large dewlaps and high bite forces diminish their dewlaps (but not bite force), whereas individuals with small dewlaps and low bite forces in the spring increase their bite forces (but not dewlap size). We also show that this trend was apparent both in the field (comparing different individuals) and the laboratory (comparing the same set of individuals under standardized conditions). Finally, seasonal changes were not consistent among individuals for either bite force or dewlap area, as individuals changed seasonally in proportion to their initial state. These findings cast doubt on the widely held view of invariant morphology–performance relationships, and offer a cautionary note for ecomorphological studies.

D. J. Irschick, M. Ramos, C. Buckley, J. Elstrott, E. Carlisle, S. P. Lailvaux, Dept of Ecology and Evolutionary Biology, Tulane Univ., New Orleans, LA 70118, USA (irschick@tulane.edu). – N. Bloch, Dept of Biological Science, Carrera 1 #18A-10, Univ. de Los Andes, Bogota, Colombia. – A. Herrel and B. Vanhooydonck, Laboratory for Functional Morphology, Univ. of Antwerp (UIA), Universiteitsplein 1, BE-2610 Antwerp, Belgium.

Ecological studies with various taxa have established that population structure and social behavior often change dramatically seasonally, particularly in temperate regions (Karr et al. 1990, Johnston et al. 1997, Ghalambor and Martin 2001). Most studies examining seasonal effects have focused on population structure

(e.g. sex ratio), habitat use, behavior (e.g. foraging behavior), or physiology, among others. By contrast, few studies have examined how whole-organism performance capacities change seasonally, or, more interestingly, whether morphology–performance relationships change seasonally. Whole-organism performance capa-

Accepted 15 November 2005
Subject Editor: Tim Benton

Copyright © OIKOS 2006
ISSN 0030-1299

cities (e.g. sprint speed, bite force), are often important for escaping predators, capturing prey, and defending territories (Hertz et al. 1988, Bennett and Huey 1990, Garland and Losos 1994, Herrel et al. 1999, Irschick and Garland 2001, Shine 2003, Lailvaux et al. 2004, Le Galliard et al. 2004, Miles 2004). Previous researchers have postulated that if morphological variation among individuals correlates with variation in performance capacity and fitness, then this would provide evidence for the adaptive nature of morphological variation (Arnold 1983). The assumption that morphology–function relationships are invariant underlies much of the fields of ecomorphology and functional morphology (Karr and James 1975, Miles and Ricklefs 1984, Lauder 1990, Losos 1990, Winemiller 1991, Wainwright and Reilly 1994).

Two potentially independent aspects of morphology can be related to performance. First, simple aspects of morphological shape are expected to be biomechanically related to performance capacity, such as limb dimensions and speed among and within animal species (Bonine and Garland 1999, Irschick and Jayne 1999, Biewener 2003). A second aspect of morphology potentially related to performance capacity is sexual signal size, particularly in male animals (Darwin 1871, Andersson 1994, Badyaev and Qvarnstrom 2002, Whiting et al. 2003). The shape and size of such sexual structures play an important role in male–male interactions, female choice, and hence, reproductive success (Endler 1992, Andersson 1994, Berglund et al. 1996, Backwell and Passmore, 1996, Jennions and Backwell 1996, Backwell et al. 2000, Whiting et al. 2003). However, an on-going debate concerns whether the relative size of such sexual signals acts as indicators of male “quality” (Zahavi 1975, Berglund et al. 1996, Vanhooydonck et al. 2005). Whole-organism performance capacities represent one potential metric of male “quality”, as male performance is correlated with dominance in some species (e.g. lizards, Robson and Miles 2000, Lailvaux et al. 2004).

The assumption that performance capacity should be invariant across different seasons has received little empirical study. However, several considerations suggest that some widely studied performance traits can be expected to change seasonally. For example, seasonal fluctuations in hormone levels (e.g. testosterone) are well-documented in temperate animals (Moore et al. 1984, Moore 1988, Tokarz et al. 1998), and such hormones are also known to affect whole muscle properties. Because many performance capacities (e.g. endurance) are directly affected by hormones (Sinervo et al. 2000), one might also expect performance to change seasonally as well (Garland and Else 1987). On a related note, a large body of work has documented inducible plastic changes in morphology, with consequent effects on performance (e.g. predator-induced morphological changes, Van Buskirk and Saxer 2001,

Wilson et al. 2005). Empirical studies have already demonstrated seasonal fluctuations in morphology across a variety of taxa. Examples include brain morphology in birds (Gulledge and Deviche 1998, Deviche and Gulledge 2000, Tramontin and Brenowitz 2000), cartilage morphology in rodents (Heldmaier and Steinlechner 1981, Ruf et al. 1993), and bone metabolism in Galapagos marine iguanas (Wikelski and Thom 2000).

An important performance trait for many polygynous animals is bite force (Herrel et al. 1999, Lailvaux et al. 2004, Lappin and Husak 2005). For example, bite force is important for both consuming prey and resolving male–male territorial disputes in lizards (Stamps and Krishnan 1997, Herrel et al. 1999, Perry et al. 2004, Lailvaux et al. 2004, Lappin and Husak 2005, Vanhooydonck et al. 2005). Bite force also shows strong biomechanical links with simple aspects of head shape; both theoretical and empirical analyses show that size-adjusted maximum bite force should be positively correlated with size-adjusted head height and head width (Herrel et al. 1999, 2001a,b, Verwaijen et al. 2002). Finally, other studies with lizards have shown that head shape is correlated with both the ability to acquire territories (Hews 1990, Lappin and Husak 2005), and the ability to win male contests (Lailvaux et al. 2004, Perry et al. 2004).

We tested the hypothesis that morphology–performance relationships are invariant across different seasons by examining how bite force, and two aspects of morphology (head shape and dewlap size) changed seasonally in the field, and in the laboratory in the green anole *Anolis carolinensis*. The dewlap is a sexually dimorphic flap of loose skin that is extended through movements of the hyobranchium (i.e. second ceratobranchial; Bels 1990, Font and Rome 1990, Tokarz et al. 2003, 2005). Male anoles use their dewlaps in various contexts, including non-directed (when the male is alone), or directed (in contact with another male or a female) displays (Jenssen et al. 2000, 2001). Recent work (Vanhooydonck et al. 2005) showed that for several species of Caribbean *Anolis* lizards, the relative size of the dewlap is a significant predictor of maximum bite force, but that study was conducted at a single point in time.

We chose the green anole lizard because it has been a model system for studies of behavioral ecology (Greenberg and Crews 1990, McMann 1993, Lovern et al. 1999, Jenssen et al. 2000, 2001), and performance (Lailvaux et al. 2004, Irschick et al. 2005a). We focused on a well-studied and enclosed southern Louisiana green anole population (Tulane campus). Although seasonal fluctuations in southern Louisiana are not marked compared to more northern localities (Bishop and Echternacht 2004), they do consist of relatively mild spring and fall seasons, a hot summer, and a relatively cool winter season (see www.weather.com for detailed

information on temperature and precipitation in southern Louisiana). Concomitant with such seasonal weather changes are seasonal cycles in reproductive behavior and hormones in temperate *Anolis* lizards (Greenberg and Crews 1990, Tokarz et al. 1998, Jenssen et al. 2000, 2001). Male green anoles compete with one another for access to territories in the spring, based in part on the presence of pre-existing females. Males are more quiescent and less active during the fall and winter (Jenssen et al. 1995). We conducted field studies across three seasons (spring 2003, winter 2003/2004, and spring 2004) in which we measured bite force, dewlap size, and head shape in samples of different male green anoles. We also maintained a group of the same male green anoles in the laboratory across different seasons (spring 2004, fall 2004, winter 2004) to investigate seasonal plastic changes in the above variables.

Material and methods

Field data

Our field site was a 380 m linear transect located on the Tulane University campus in Orleans Parish, LA (Irschick et al. 2005a). The habitat structure for this population consisted of clumps of vegetation separated in space from other clumps by 2–10 m. We only sampled the vegetation along either edge of the Tulane campus transect. Lizards were examined in the field over three time periods (spring 2003, winter 2003/2004, and spring 2004), and for the laboratory for three seasons (spring 2004, fall 2004, and winter 2004). The field sampling periods were: spring; April 1 to May 10; winter; December 1 to January 10). Although we do compare some different times (e.g. spring 2003 in field versus spring 2004 in the laboratory), the actual seasons sampled are similar. We examined the laboratory anoles during both the fall and winter of 2004 to provide additional detail on when seasonal changes occur, and as our data show, these two time periods show similar results, and both are comparable to the winter 2003/2004 field sample.

We focused on adult males because bite force and dewlaps size are most relevant for this age/sex class (Lailvaux et al. 2004, Vanhooydonck et al. 2005). We collected large samples of adult males (>50 per season, see legend of Fig. 1 for exact sample sizes) for each of the three field seasons, although sample sizes differed slightly between the dewlap and the bite force/morphology data sets. Sexually mature males (sex distinguished by the presence of post-anal scales) were greater than or equal to 45 mm SVL, which is when males achieve sexual maturity. After capture, we measured morphology (mass, snout-vent length, head height and head width, dewlap area) and bite performance of each lizard, and released the lizard to its original point of capture within 48 h.

Because each lizard was marked with a permanent tag (Irschick et al. in press), we knew that each lizard was measured once (see Nauwelaerts et al. 2000 for details of the marking procedure).

Laboratory plasticity experiment

We collected 17 adult male *A. carolinensis* during the middle of the spring 2004 sampling period (April 20–27) from the Tulane field site, and measured their mass, SVL, head shape, dewlap area, and bite force. We chose large males for this experiment because we did not want seasonal growth to be a confounding issue. Each lizard was maintained singly in a 10-ga aquarium (lined with cypress mulch) with similar perches (0.5 m × 2 cm) oriented in the same manner towards a uniform light source (75-watt vita-lite bulbs positioned at the same spot over each cage). Each lizard was fed exactly six similarly sized crickets per week, and was provided water ad libitum. Lizards were maintained in the laboratory from the spring until the winter (about 9 months). We maintained lizards under a constant photoperiod mimicking spring hours (12 light: 12 dark), because we wanted to determine if morphological and performance changes would occur under standardized laboratory conditions. Lizards were prevented from viewing one another by dark paper sheets covering each side of each cage. We re-measured mass, SVL, mass, head shape, and bite force for each lizard in the fall (1 October 2004), and winter (1 December 2004).

Measurement of morphology and bite force

For each lizard, we measured the following morphological traits with digital calipers (with the exception of mass): mass (measured to 0.01 g using a Mettler scale), snout vent length (SVL; measured from the tip of the snout to the cloaca), head height (measured at the highest point of the head), and head width (measured at the widest point of the head). We focused on head width and head height because these two measures are expected to be biomechanically linked with bite force (Herrel et al. 1999). All linear dimensions were measured using digital calipers (Mitutoyo CD-15 DC; precision 0.01 mm). To obtain a reliable measure of dewlap size, we positioned the lizard sideways and pulled the base of the second ceratobranchial gently forward with a pair of forceps. Since the dewlap consists of a skin flap attached to the lizard's throat on the one side and to the hyoid bone on the other, the dewlap becomes maximally extended when the ceratobranchial is pulled forward completely. Before taking a digital picture, we positioned the lizard in such that the extended dewlap was parallel to the lens of the camera (Nikon Coolpix 4500). We digitized the outer edge of the dewlap using the program

TPSDIG (v 1.39; J. Rohlf, SUNY Stonybrook) and calculated the total dewlap area for each individual. This method of measuring dewlap size yields repeatable results (Losos and Chu 1998, Vanhooydonck et al. 2005).

We measured biting force by using an isometric Kistler force transducer (type 9023, Kistler Inc. Wintherthur, Switzerland) connected to a Kistler charge amplifier (type 5058a, Kistler Inc.). We induced lizards to bite forcefully on the free ends of the bite force device (Herrel et al. 1999). We measured bite forces five times for each lizard with a 30 min rest period between measures. The largest bite force obtained from each session was taken as the maximal bite force for that individual. All individuals were placed inside an incubator at 32°C for 30 min prior to each bite force measurement.

Statistical analyses

All statistical analyses were conducted in SYSTAT v. 10.0. We first tested whether bite force, head shape, and dewlap area changed within the same individuals (laboratory samples), and for different individuals (field samples) across different seasons. Snout-vent length did not change significantly across seasons for the laboratory samples (ANOVA, $F_{2,48} = 0.75$, $P > 0.50$), or for the field samples (dewlap data set; ANOVA, $F_{2,206} = 0.70$, $P > 0.40$; bite force data set; ANOVA, $F_{2,218} = 2.51$, $P > 0.05$). Hence, we conducted non size-adjusted MANOVAs for both the field (dewlap and bite force/morphology) and the laboratory data using bite force, dewlap area, head width, and head height as the dependent variables, and season (field test, $N = 3$, spring 2003, winter 2003/2004, spring 2004; laboratory test, $N = 3$, spring 2004, fall 2004, winter 2004) as the independent variable. We also tested for homogeneity of variances for all variables using the Levene test (Levene 1960). We addressed whether predicted relationships existed between two sets of variables across different seasons. We tested whether significant relationships existed between relative measures of head shape (head height and head width) and relative bite force for the field data ($N = 6$ tests). We did not conduct similar head shape–bite force analyses for the laboratory sample because of the relatively small sample size ($N = 17$ individuals), which would be unlikely to detect head shape–bite force relationships that tend to exhibit relatively low correlation coefficients.

We then examined whether the relative size of the dewlap was a significant predictor of relative bite force among individuals within each of the seasons (i.e. for both the laboratory and the field data, $N = 6$ tests). Recent work (Lailvaux et al. 2004) shows that green anoles segregate into two age-specific morphs that differ in relative head shape and bite force. Lightweight males

(<63 mm SVL) have relatively small heads and weak bite forces, whereas heavyweight males (>63 mm SVL) have relatively large heads and strong bite forces. Further, bite force is most relevant for heavyweight males for resolving male territorial disputes (Lailvaux et al. 2004). Thus, we divided each of the field samples into “lightweight” and “heavyweight” correlation tests based on the SVL threshold value of 63 mm. Because the laboratory anoles consisted primarily of large individuals (>63 mm SVL), we did not divide this data set. To test if relationships between morphology and performance varied seasonally, we conducted Pearson correlation tests for each bivariate combination of head shape, bite force, and dewlap area. Because the head shape–performance, and dewlap–performance relationships were among individuals that differed in size (in contrast to the comparisons among seasons in which individuals did not differ in size), we corrected for the effects of size on head shape, dewlap area, and bite force by calculating residuals. We calculated residuals from simple univariate linear regressions of log-transformed values for each of these variables (dependent variables) versus log-transformed values of SVL (independent variable).

Results

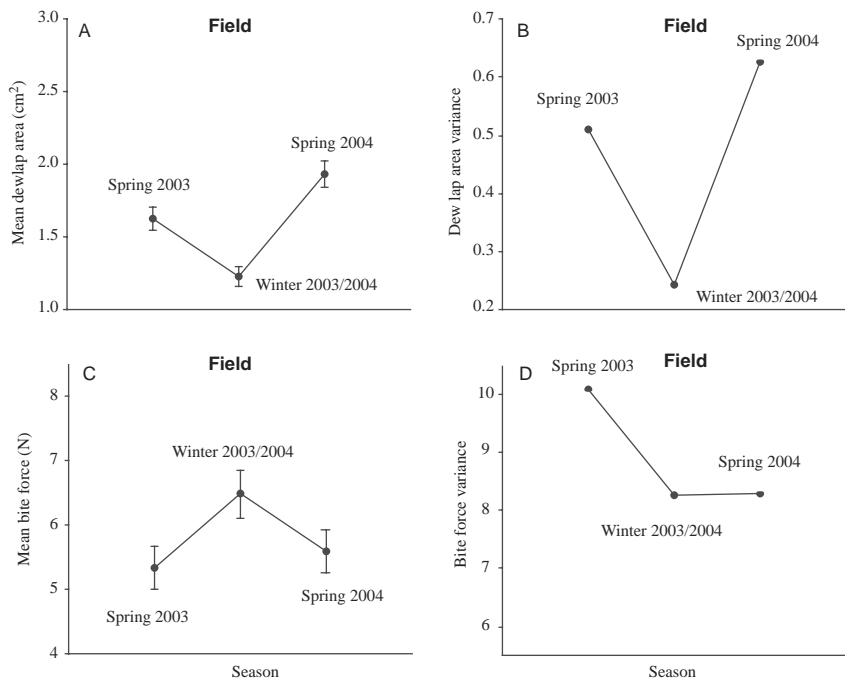
Field data

For the field samples, mean bite force showed a clear seasonal trend in being low during the two spring seasons, and high during the winter (Fig. 1, Table 1). By contrast, the two head shape variables did not differ statistically across different seasons in the field (Table 1). Similarly, mean dewlap areas fluctuated markedly among seasons in the field. In contrast to bite force, average dewlap size was large in the two spring seasons (2003, 2004), and small in the winter (2003/2004) seasons (Fig. 1, Table 1). Similarly, the variance of the field dewlap area samples changed seasonally (Fig. 1, Table 1), being high in the two spring seasons (2003 and 2004), and low in the winter (2003/2004). However, the variances in the two head measurements, and bite force did not change significantly across seasons (Fig. 1, Table 1).

Laboratory data

These above seasonal trends for bite force, head shape, and dewlap area were mirrored in the laboratory. Mean bite forces were significantly lower for the spring of 2004 compared to the fall and winter of 2004 (Fig. 2, Table 1), but the variance of bite force did not change significantly across seasons (Fig. 2, Table 1). Both the mean and the variance of dewlap area were significantly lower for the spring of 2004 compared to the fall and winter of 2004

Fig. 1. Plots of field values of means (A, C), and variances (B, D) for dewlap area and bite force across three seasons (spring 2003, winter 2003/2004, and fall 2004) from the Tulane field site for green anole males. Error bars are ± 1 SE. Note the opposing trends for seasonal changes in mean dewlap area and mean bite force; mean dewlap areas are large in the spring, and small in the winter, whereas mean bite forces are low in the spring, and high in the winter. By comparison, the variances for dewlap area are large in the two spring seasons, and small in the winter. However, the variance for bite force did not change statistically across different seasons. Numbers of individuals sampled were as follows: spring 2003 (dewlap; N = 84, bite force/morphology; N = 91), winter 2003/2004 (dewlap; N = 52, bite force/morphology; N = 61), spring 2004 (dewlap; N = 73, bite force/morphology; N = 70).



(Fig. 2, Table 1). Finally, neither the means or the variances of head shape differed significantly among seasons in the laboratory (Table 1).

The seasonal changes in bite force and dewlap area in the laboratory were not consistent among individuals. For example, large male lizards with high bite forces increased in bite force only slightly between spring and fall/winter (about 5–20%), whereas smaller males with low bite forces increased dramatically in bite force

(the most extreme individual changed about 80%, Fig. 3). Similarly, green anole males with large dewlaps in the spring declined seasonally to the fall and winter more than anoles with small spring dewlaps (Fig. 3). However, one should be cautious when interpreting these scatterplots because the independent and dependent variables in Fig. 3 are not statistically independent. However, these scatterplots provide evidence for a pattern of non-linear seasonal change in bite force and dewlap area among individuals under standardized laboratory settings.

Table 1. Results from MANOVAs (means test) and tests of homogeneity of variances (variance test) for dewlap area, head shape and bite force across different field samples (different sets of individuals sampled across seasons), and laboratory samples (same set of individuals sampled across seasons). Note that mean dewlap area and bite force, but not head shape, both changed significantly across seasons, both in the field and in the laboratory. Only the variance for dewlap area changed significantly across seasons.

Variable	Means test F-value	Variance test F-value	df
Field			
dewlap area (cm ²)	10.12***	15.40***	2,206
bite force (N)	3.90*	0.03 ^{ns}	2,218
head height (mm)	2.89 ^{ns}	1.95 ^{ns}	2,218
head width (mm)	1.40 ^{ns}	0.66 ^{ns}	2,218
Laboratory			
dewlap area (cm ²)	20.03***	4.32*	2,48
bite force (N)	6.34**	0.12 ^{ns}	2,48
head height (mm)	1.76 ^{ns}	0.15 ^{ns}	2,48
head width (mm)	3.09 ^{ns}	0.73 ^{ns}	2,48

ns = non-significant ($P > 0.05$), * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

Morphology–performance relationships

In the field, the seasonal shifts in bite force resulted in both measures of residual head shape (width and depth) being statistically significant predictors of residual bite force during the two field spring seasons (2003 and 2004), but not during the winter of 2003/2004 (Fig. 4). Similarly, in the field, the simultaneous shifts in dewlap size and bite force resulted in residual dewlap area being a significant predictor of residual bite force during the two spring seasons, but not during the winter season for larger (heavyweight) green anole males (Fig. 5). However, none of the correlations between residual dewlap area and residual bite force were significant for the field samples for smaller (lightweight) males (spring 2003: Pearson $r = -0.12$, $df = 43$, $P > 0.20$; winter 2003/2004: $r = -0.38$, $df = 23$, $P > 0.05$; spring 2004: $r = -0.02$,

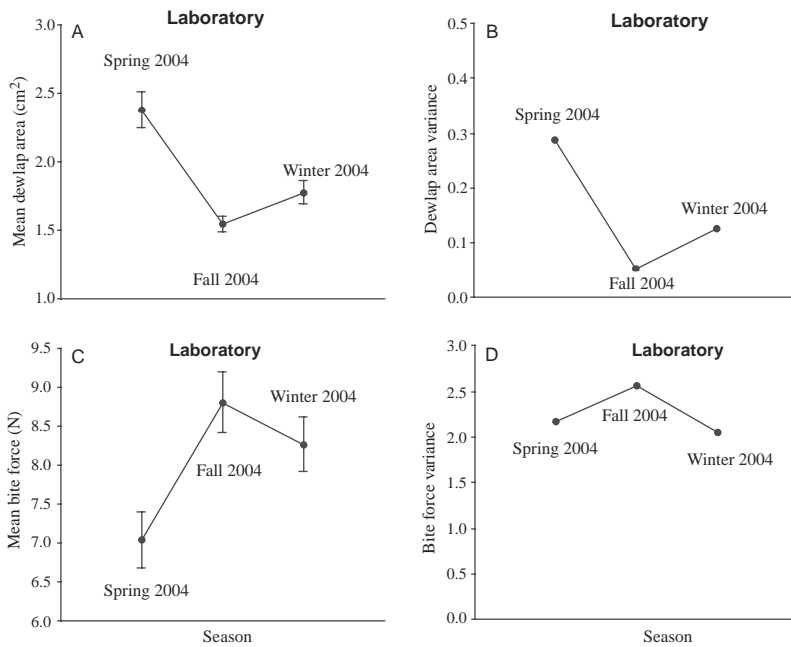


Fig. 2. Plots of laboratory values of means (A, C), and variances (B, D) for dewlap area and bite force across three seasons (spring 2004, fall 2004, and winter 2004) for the same group of 17 green anole males. Error bars are ± 1 SE. Note the opposing trends for seasonal changes in mean dewlap area and mean bite force; mean dewlap areas are large in the spring, and small in the fall and winter, whereas mean bite forces are low in the spring, and high in the fall and winter. By comparison, the variances for dewlap area are large in the spring, and small in the fall and winter. However, the variance for bite force did not change statistically across different seasons.

df = 34, $P > 0.50$). This seasonal plasticity in the dewlap area-bite force relationship also occurred under standardized laboratory settings with the same set of heavy-weight green anole males (Fig. 6). In other words, either across different sets of individuals, or for the same set of individuals, residual dewlap area was significantly related to residual bite force only during the spring for large green anole males.

Discussion

We tested the hypothesis that morphology–performance relationships are invariant across different seasons using a population of green anole lizards as a model system. We rejected the hypothesis of invariance; not only did bite force change seasonally (up to 80% within the same individual across several months), but relationships among morphology and bite force were highly plastic. We noted opposing trends for how bite force and dewlap area changed seasonally; whereas dewlap areas were large in the spring, and small in the winter, bite forces were low in the spring and high in the winter. This pattern occurred because of a tradeoff at the individual level: individuals in the spring with large dewlaps and high bite forces diminish their dewlaps (but not bite force), whereas individuals with small dewlaps and low bite forces in the spring increase their bite forces (but not dewlap size). We also showed that these trends were apparent both in the field (comparing different individuals) and the laboratory (comparing the same set of individuals under standardized conditions).

Recent comparative work with head shape and biting mechanics in fish has shown that the same morphology–function relationship can be acquired by different combinations (Alfaro et al. 2005, Wainwright et al. 2005). Other work has emphasized the importance of behavior as an intermediary between morphology and performance (Garland and Losos 1994, Lauder and Reilly 1996), or potentially, between performance and fitness (Irschick et al. 2005b). Our work builds upon these prior studies by showing that alterations in morphology–performance relationships can occur even in the absence of significant changes in morphological shape. For example, we note that head shape in green anoles remained constant across seasons, both in the field, and in the laboratory, but bite force changed dramatically. Prior work has shown that head shape and bite force plays a key role in dictating dominance in male lizards, which, in turn, can determine access to high-quality territories and females (Hews 1990, Lailvaux et al. 2004, Perry et al. 2004, Lappin and Husak 2005). Thus, at first sight, the diminishment of bite forces during the spring reproductive period seems puzzling, but a closer inspection is informative. We note that for the largest anoles with the highest bite forces, seasonal change in bite force was minimal (Fig. 3). Because bite force is most important for large green anole males (Lailvaux et al. 2004), this indicates little change in their dominance status across seasons. The low bite forces in the spring occurred because smaller anoles that had low bite forces in the spring increased dramatically in the fall and winter. Increases in body size are not responsible for this trend, as the two smallest individuals grew an average of 2 mm in snout-

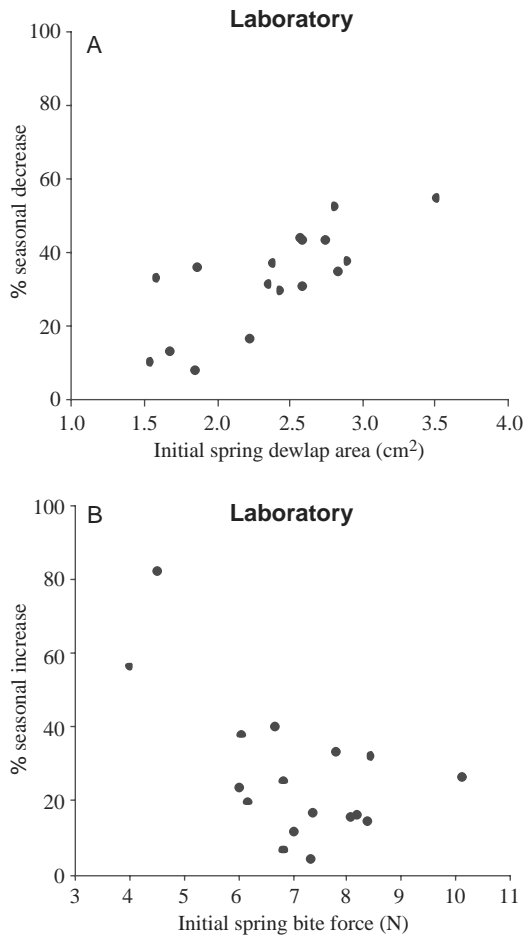


Fig. 3. (A) A scatterplot for laboratory green anoles males showing initial dewlap area during the spring of 2004 (x-axis) versus percent decline in dewlap area between spring 2004 and winter 2004 (y-axis) ($r=0.74$, $N=17$, $P<0.001$). Note that individuals with large spring dewlaps tend to shrink more seasonally compared to individuals with small spring dewlaps. (B) A scatterplot of the bite force in the spring of 2004 (x-axis) versus percent increase in bite force between spring 2004 and winter 2004 (y-axis) for the same laboratory green anoles ($r=0.58$, $N=17$, $P<0.025$). Note that some male lizards increased by as much as 80% in bite force in only four months, despite not changing significantly in body size. These trends remain if one uses laboratory values for fall 2004 for either dewlap area or bite force.

vent length over nine months, but their bite forces nearly doubled (Fig. 3). This finding suggests an age/size dependence on seasonal plastic change, and accordingly, a threshold on seasonal plastic change.

A key tenant of eco-morphology is that morphology and function should be tightly linked (Wainwright and Reilly 1994), but the seasonal changes in bite force occurred in the absence of changes in head shape. We tested the possibility that seasonal changes in head muscles were responsible by dissecting the head muscles of a sample of male green anoles from the spring of 2005 ($N=7$) and comparing them to different individuals

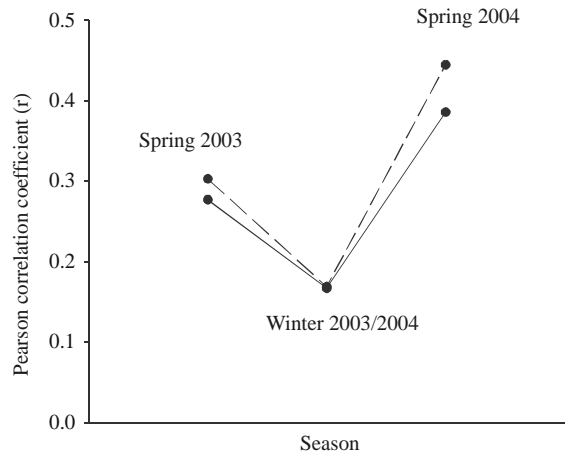


Fig. 4. Plots of Pearson correlation coefficients from two predicted biomechanical relationships (residual head height vs residual bite force [solid line], and residual head width vs residual bite force [dashed line]) across different sets of male green anoles across different seasons in the field ($N=6$ tests total, two per season). Note that green anoles exhibit the predicted significant positive relationships between residual head dimensions and bite force during the two spring seasons (2003 and 2004), but not during the winter season (2003/2004). Exact r -values are: spring 2003 ($N=91$ individuals, head height: $r=0.277$, $P<0.01$; head width: $r=0.303$, $P<0.01$), winter 2003/2004 ($M=61$ individuals, head height: $r=0.167$, $P>0.10$; head width: $r=0.169$, $P>0.10$), spring 2004 ($N=70$ individuals, head height: $r=0.386$, $P<0.001$; head width: $r=0.445$, $P<0.001$).

($N=7$) from the fall of 2004. We detected no significant differences in the cumulative relative masses of head muscles between spring and fall ($P>0.05$), and can offer no mechanistic explanation for why bite force changes seasonally. Further studies that examine more detailed properties of head muscles (e.g. fiber type, etc.) across a greater number of green anole populations would be useful for examining the mechanistic basis of seasonal changes in bite force.

Seasonal changes in sexually dimorphic structures, such as the anole dewlap, are well-documented (Andersson 1994). The anole dewlap has been cited as a key factor in driving the extensive anole radiation (Losos and Chu 1998, Tokarz et al. 2003). The anole dewlap is a flap of skin that is extended via the second ceratobranchial, which is composed primarily of cartilage (Bels 1990, Font and Rome 1990, Tokarz et al. 2003, 2005). In contrast to bite force, green anoles with large dewlaps changed the most across seasons, whereas green anoles with small dewlaps remained largely constant. Prior studies have shown costs (e.g. energetic, predation) associated with unusually large sexual signals (Ryan 1985, Endler 1992, Andersson 1994, McCoy et al. 2003), but the fact that the dewlap can be folded (and hence hidden from view) makes this hypothesis unlikely. Nevertheless, our results are consistent with the seasonal reproductive patterns of green anole lizards in showing

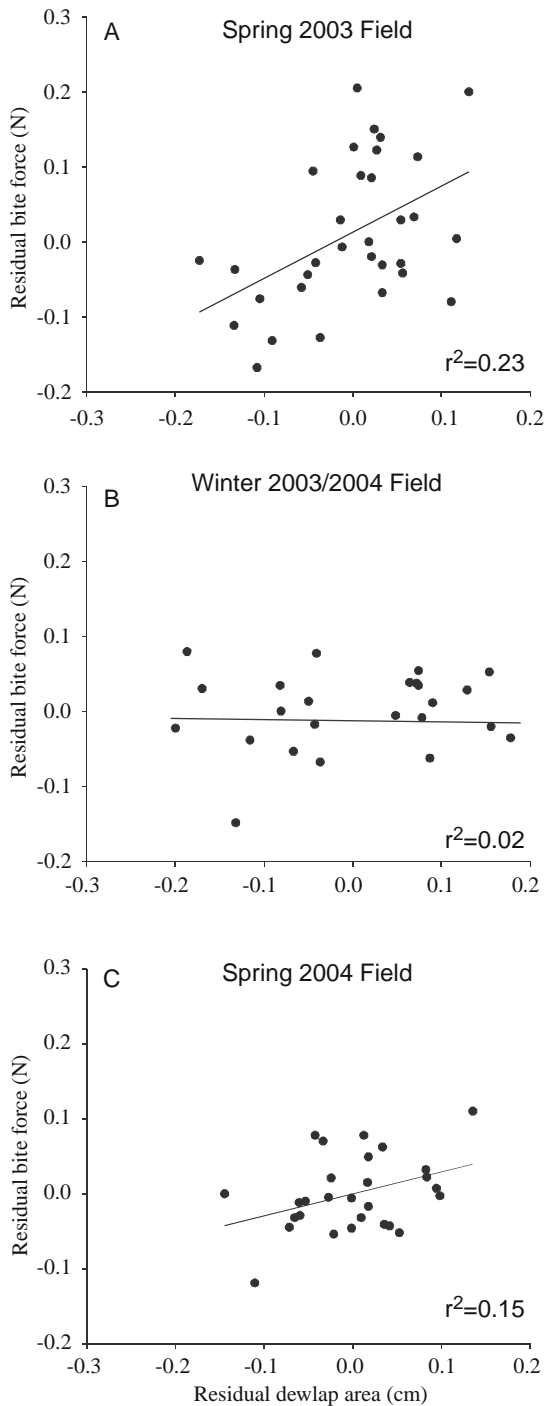


Fig. 5. Scatterplots of residual dewlap areas (x-axis) versus residual bite forces (y-axis) for each of three field samples of heavyweight (≥ 63 mm SVL) male green anoles. Note that for the two spring seasons (2003 and 2004, panels A and C), residual bite force and residual dewlap area are significantly and positively related, whereas for the winter sample (2003/2004, panel B), these variables are not significantly related. Relevant statistics are: spring 2003, $df = 31$, r -value = 0.40, $P < 0.025$), winter 2003/2004, $df = 22$, r -value = 0.13, $P > 0.50$, spring 2004, $df = 26$, r -value = 0.38, $P < 0.05$).

that the relative size of the anole dewlap acts as an honest signal of male fighting ability (Zahavi 1975, Leal 1999, Backwell et al. 2000, Vanhooydonck et al. 2005) during the spring reproductive season, but not during the non-reproductive fall or winter periods. Because green anoles can damage one another during territorial fights (McMann 1993, Lailvaux et al. 2004), honest signaling during the intense spring period may prevent injury in some cases. The change in the dewlap area–bite force relationships (Figs. 5 and 6) is due to non-linear seasonal change in both bite force and dewlap area (Fig. 3), resulting in the dewlap being a “dishonest” signal during the fall and winter. However, Tokarz et al. (2003, 2005) failed to show a key role for either possession of a dewlap, or use of a dewlap, for the ability of adult male *A. sagrei* to defend territories and acquire matings. Two scenarios for the physical changes in dewlap area are seasonal elongation/shrinking of the ceratobranchial, and seasonal elongation/shrinking of the extendable dewlap skin. We dissected and measured the lengths of the second ceratobranchial for a sample of anoles ($N = 7$ per season) from the spring (2005) and fall (2004) seasons and detected significantly longer ceratobranchial lengths for spring anoles ($P < 0.05$). These data provide some support for seasonal elongation of the ceratobranchial, but do not exclude seasonal elongation/shrinking of the skin. We also note that previous studies have documented seasonal fluctuations in cartilage in rodents (Heldmaier and Steinlechner 1981, Ruf et al. 1993).

A common mechanism that may explain the seasonal changes in bite force and dewlap area are endogenous seasonal rhythms in hormone levels, such as testosterone and corticosterone, which are well-documented in temperature lizards (Moore et al. 1984, Moore 1988, Greenberg and Crews 1990, Tokarz et al. 1998). For example, testosterone levels are known to be high in North American anoles during the active spring reproductive season, and decline dramatically between spring and fall (Greenberg and Crews 1990, Tokarz et al. 1998), a pattern closely matching the seasonal changes in bite force and dewlap area. Indeed, differences among individuals in the relative allocation of hormones could explain the apparent tradeoff between seasonal changes in dewlap size and bite force if individuals differ in both initial (e.g. spring) hormone state, as well as how these hormones decline seasonally. Manipulative studies with hormone implants would be useful for testing this hypothesis.

Eco-morphological studies have provided a foundation for testing ideas about adaptation (Wainwright and Reilly 1994). However, these studies also have a mixed record, with some detecting predicted ecology–morphology relationships, whereas others have not (Karr and James 1975, Rotenberry and Wiens 1980, Wainwright and Reilly 1994, Vanhooydonck and Van Damme 1999).

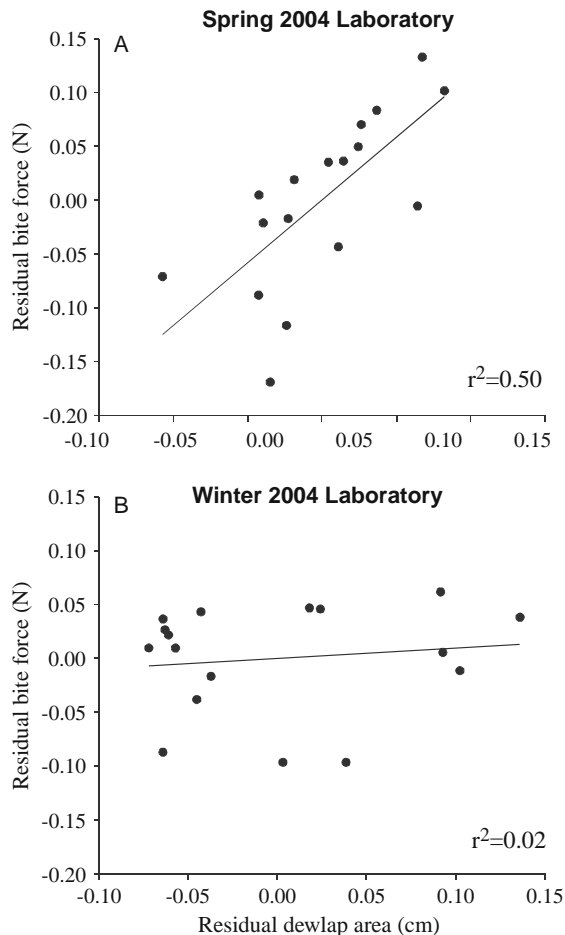


Fig. 6. Scatterplots of residual dewlap areas (x-axis) versus residual bite forces (y-axis) for the same set of 17 green anole males from the laboratory across two seasons (spring 2004 and winter 2004, panels A and B). Note that for the spring season, residual bite force and residual dewlap area are significantly and positively related, whereas for the winter sample these variables are not significantly related. Relevant statistics are: spring 2004: $df=15$, $r\text{-value}=0.71$, $P<0.0025$, winter 2004: $df=15$, $r\text{-value}=0.13$, $P>0.50$. The results for fall 2004 are similar to winter 2004, and hence are not shown.

Based on our findings, we suggest that one confounding factor that may add “noise” to any eco-morphological comparison is seasonal plasticity in morphology and/or performance capacity. We note that during the spring, green anoles showed all the predicted relationships between morphology and performance, whereas these relationships disappeared within only a span of months. Additionally, at least in the laboratory, the changes in both bite force and dewlap area were non-linear, in that individuals changed seasonally in proportion to their initial state. This form of seasonal plastic change is problematic for eco-morphological studies, as seasonal change will differ for every individual. We do not intend the above statements as a critique of any of the above studies, as the primary author (DJI) has also published

eco-morphological studies in which seasonal plasticity in performance and morphology were not considered (Irschick et al. 1997). Nevertheless, we argue that researchers should consider repeating eco-morphological studies across different seasons to verify whether seasonal plasticity could be a confounding factor.

Acknowledgements – We thank T. Jenssen, R. Tokarz and T. Sherry for advice and criticism. This work was supported by an NSF grant to D. Irschick (IOB 0421917). AH and BVH are postdoctoral researchers of the Fund for Scientific Research, Flanders (FWO-VI). The work in this paper was done in accordance with an approved animal use protocol (IACUC 0189-2-16-0301).

References

- Alfaro, M. E., Bolnick, D. I. and Wainwright, P. C. 2005. Evolutionary consequences of many-to-one mapping of jaw morphology to mechanics in labrid fishes. – *Am. Nat.* 165: 140–154.
- Andersson, M. 1994. Sexual selection. – Princeton Univ. Press.
- Arnold, S. J. 1983. Morphology, performance and fitness. – *Am. Zool.* 23: 347–361.
- Backwell, P. R. Y. and Passmore, N. I. 1996. Time constraints and multiple choice criteria in the sampling behaviour and mate choice of the fiddler crab, *Uca annulipes*. – *Behav. Ecol. Soc.* 38: 407–416.
- Backwell, P. R. Y., Christy, J. H., Telford, S. R. et al. 2000. Dishonest signaling in a fiddler crab. – *Proc. R. Soc.* 267: 719–724.
- Badyaev, A. V. and Qvarnström, A. 2002. Putting sexual traits into the context of an organism: life-history perspective in studies of sexual selection. – *Auk* 119: 301–310.
- Bels, V. L. 1990. The mechanism of dewlap extension in *Anolis carolinensis* (Reptilia, Iguanidae) with histological analysis of the hyoid apparatus. – *J. Morphol.* 206: 225–244.
- Bennett, A. F. and Huey, R. B. 1990. Studying the evolution of physiological performance. – *Oxford Surv. Evol. Biol.* 7: 251–284.
- Berglund, A., Bisazza, A. and Pilastro, A. 1996. Armaments and ornaments: an evolutionary explanation of traits of dual utility. – *Biol. J. Linn. Soc.* 58: 385–399.
- Bishop, D. C. and Echternacht, A. C. 2004. Emergence behavior and movements of winter-aggregated green anoles (*Anolis carolinensis*) and the thermal characteristics of their crevices in Tennessee. – *Herpetologica* 60: 168–177.
- Biewener, A. A. 2003. Animal locomotion. – Oxford Univ. Press.
- Bonine, K. T. and Garland, T. Jr. 1999. Sprint performance of phrynosomatid lizards, measured on a high-speed treadmill, correlates with hindlimb length. – *J. Zool.* 248: 255–265.
- Darwin, C. 1871. The descent of man and selection in relation to sex. – Murray, London.
- Deviche, P. and Gulledge, C. C. 2000. Vocal control region sizes of an adult female songbird change seasonally in the absence of detectable circulating testosterone concentrations. – *J. Neurobiol.* 42: 202–211.
- Endler, J. A. 1992. Signals, signal conditions, and the direction of evolution. – *Am. Nat.* 139: 5125–5153.
- Font, E. and Rome, L. C. 1990. Functional morphology of dewlap extension in the lizard *Anolis equestris* (Iguanidae). – *J. Morphol.* 206: 245–258.
- Garland, T. Jr. and Else, P. L. 1987. Seasonal, sexual, and individual variation in endurance and activity metabolism in lizards. – *Am. J. Physiol.* 252: R439–R449.
- Garland, T.Jr. and Losos, J. B. 1994. Ecological morphology of locomotor performance in squamate reptiles. – *Ecological*

- morphological integrative organismal biology. – Univ. Chicago Press, pp. 240–302.
- Ghalambor, C. K. and Martin, T. E. 2001. Fecundity survival tradeoffs and parental risk-taking in birds. – *Science* 292: 494–497.
- Greenberg, N. and Crews, D. 1990. Endocrine and behavioral responses to aggression and social dominance in the green anole lizard, *Anolis carolinensis*. – *Gen. Comp. Endocrinol.* 77: 246–255.
- Gulledge, C. C. and Deviche, P. 1998. Photoperiod and testosterone independently affect vocal control region volumes in adolescent male songbirds. – *J. Neurobiol.* 36: 550–558.
- Heldmaier, G. and Steinlechner, S. 1981. Seasonal control of energy requirements for thermoregulation in the Djungarian hamster (*Phodopus sungorus*), living in natural photoperiod. – *J. Comp. Phys.* 142: 429–437.
- Herrel, A., Spithoven, L., Van Damme, R. et al. 1999. Sexual dimorphism of head size in *Gallotia galloti*: testing the niche divergence hypothesis by functional analyses. – *Funct. Ecol.* 13: 289–297.
- Herrel, A., De Grauw, E. and Limos-Espinol, J. A. 2001a. Head shape and bite performance in xenosaurid lizards. – *J. Exp. Zool.* 290: 101–107.
- Herrel, A., Van Damme, R., Vanhooydonck, B. et al. 2001b. The implications of bite performance for diet in two species of lacertid lizards. – *Can. J. Zool.* 79: 662–670.
- Hertz, P. F., Huey, R. B. and Garland, T. Jr. 1988. Time budgets, thermoregulation, and maximal locomotor performance: are reptiles olympians or boy scouts? – *Am. Zool.* 28: 927–938.
- Hews, D. 1990. Examining hypotheses generated by field measures of sexual selection on male lizards, *Uta palmeri*. – *Evolution* 44: 1956–1966.
- Irschick, D. J. and Jayne, B. C. 1999. Comparative three-dimensional kinematics of the hindlimb for high-speed bipedal and quadrupedal locomotion of lizards. – *J. Exp. Biol.* 202: 1047–1065.
- Irschick, D. J. and Garland, T. Jr. 2001. Integrating function and ecology in studies of adaptation: studies of locomotor capacity as a model system. – *Annu. Rev. Ecol. Syst.* 32: 367–396.
- Irschick, D. J., Vitt, L. J., Zani, P. et al. 1997. A comparison of evolutionary radiations in Mainland and West Island *Anolis* lizards. – *Ecology* 78: 2191–2203.
- Irschick, D. J., Carlisle, E., Elstrott, J. et al. 2005a. A comparison of habitat use, morphology, clinging performance, and escape behavior among two divergent green anole lizard (*Anolis carolinensis*) populations. – *Biol. J. Linn. Soc.* 85: 223–234.
- Irschick, D. J., Herrel, A., Vanhooydonck, B. et al. 2005b. Locomotor compensation creates a mismatch between laboratory and field estimates of escape speed in lizards: a cautionary tale for performance to fitness studies. – *Evolution* 59: 1579–1587.
- Irschick, D. J., Gentry, G., Herrel, A. et al. In press. Effects of Sarcophagid fly infestations on green anole lizards (*Anolis carolinensis*): an analysis across seasons and age/sex classes. – *J. Herpetol.*
- Jennions, M. D. and Backwell, P. R. Y. 1996. Residency and size affect fight duration and outcome in the fiddler crab *Uca annulipes*. – *Biol. J. Linn. Soc.* 57: 293–306.
- Jenssen, T. A., Greenberg, N. and Hovde, K. A. 1995. Behavioral profile of free-ranging male lizards, *Anolis carolinensis*, across breeding and post-breeding seasons. – *Herpetol. Monogr.* 9: 41–62.
- Jenssen, T. A., Orrell, K. S. and Lovern, M. B. 2000. Sexual dimorphism in aggressive signal structure and use by a plogynous lizard, *Anolis carolinensis*. – *Copeia* 2000: 140–149.
- Jenssen, T. A., Lovern, M. B. and Congdon, J. D. 2001. Field-testing the polyandry-based mating system for the lizard, *Anolis carolinensis*: does the model organism have the right model? – *Behav. Ecol. Soc.* 50: 162–172.
- Johnston, J. P., Peach, W. J., Gregory, R. D. et al. 1997. Survival rates of tropical and temperate passerines: a Trinidadian perspective. – *Am. Nat.* 150: 771–789.
- Karr, J. R. and James, F. C. 1975. Ecomorphological configurations and convergent evolution in species and communities. Ecology and evolution of communities. – Belknap Press, pp. 258–291.
- Karr, J. R., Nichols, J. D., Klimkiewicz, M. K. et al. 1990. Survival rates of birds of tropical and temperate forests – will the dogma survive? – *Am. Nat.* 136: 277–291.
- Le Galliard, J. F., Clobert, J. and Ferriere, R. 2004. Physical performance and darwinian fitness in lizards. – *Nature* 432: 502–505.
- Lailvaux, S., Herrel, A., VanHooydonck, B. et al. 2004. Performance capacity, fighting tactics, and the evolution of life-stage male morphs in the green anole lizard (*Anolis carolinensis*). – *Proc. R. Soc.* 271: 2501–2508.
- Lappin, K. A. and Husak, J. F. 2005. Weapon performance, not size, determines mating success and potential reproductive output in the collared lizard. – *Am. Nat.* 166: 426–436.
- Lauder, G. V. 1990. Functional morphology and systematics: studying functional patterns in a historical context. – *Annu. Rev. Ecol. Syst.* 21: 317–340.
- Lauder, G. V. and Reilly, S. M. 1996. The mechanistic basis of behavioral evolution: comparative analysis of muscosk eletal function. Phylogenies and the comparative method in animal behavior. – Oxford Univ. Press, pp. 105–137.
- Leal, M. 1999. Honest signalling during prey-predator interactions in the lizard *Anolis cristatellus*. – *Anim. Behav.* 58: 521–526.
- Levene, H. 1960. Contributions to probability and statistics. Essays in honor of Harold Hotelling. – Stanford Univ. Press, pp. 278–292.
- Losos, J. B. 1990. Ecomorphology, performance capability and scaling of West Indian *Anolis* lizards: an evolutionary analysis. – *Ecol. Monogr.* 60: 369–388.
- Losos, J. B. and Chu, L. 1998. Examination of factors potentially affecting dewlap size in Caribbean anoles. – *Copeia* 1998: 430–438.
- Lovern, M. B., Jenssen, T. A., Orrell, K. S. et al. 1999. Comparison of temporal display structure across contexts and populations in male *Anolis carolinensis*: signal stability or liability? – *Herpetologica* 55: 222–234.
- McCoy, K. J., Baird, T. A. and Fox, S. F. 2003. Sexual selection, social behavior, and the environmental potential for polygyny. – In: Fox, S. F., McCoy, J. K. and Baird, T. A. (eds), Lizard social behavior. Johns-Hopkins Press, pp. 149–171.
- McMann, S. 1993. Contextual signaling and the structure of dyadic encounters in *Anolis carolinensis*. – *Anim. Behav.* 46: 657–668.
- Miles, D. B. 2004. The race goes to the swift: fitness consequences of variation in sprint performance in juvenile lizards. – *Evol. Ecol. Res.* 6: 63–75.
- Miles, D. B. and Ricklefs, R. E. 1984. The correlation between ecology and morphology in deciduous forest passerine birds. – *Ecology* 65: 1629–1640.
- Moore, M. C. 1988. Testosterone control of territorial behavior – tonic release implants fully restore seasonal and short-term aggressive responses in free-living castrated lizards. – *Gen. Comp. Endocrinol.* 70: 450–459.
- Moore, M. C., Whittier, J. M. and Crews, D. 1984. Environmental control of seasonal reproduction in a parthenogenetic lizard *Cnemidophorus uniparens*. – *Physiol. Zool.* 57: 544–549.
- Nauwelaerts, S., Coeck, J. and Aerts, P. 2000. Visible implant elastomers as a method for marking adult anurans. – *Herpetol. Rev.* 31: 154–155.
- Perry, G., Levering, K., Girard, I. et al. 2004. Locomotor performance and dominance in male *Anolis cristatellus*. – *Anim. Behav.* 67: 37–47.
- Robson, M. A. and Miles, D. B. 2000. Locomotor performance and dominance in tree lizards, *Urosaurus ornatus*. – *Funct. Ecol.* 14: 338–344.

- Rotenberry, J. T. and Wiens, J. A. 1980. Habitat structure, patchiness, and avian communities in North American steppe vegetation – a multivariate analysis. – *Ecology* 61: 1228–1250.
- Ruf, T., Stieglitz, A., Steinlechner, S. et al. 1993. Cold exposure and food restriction facilitate physiological responses to short photoperiod in Djungarian hamsters (*Phodopus sungorus*). – *J. Exp. Zool.* 267: 104–112.
- Ryan, M. J. 1985. The Tungara frog: a study in sexual selection and communication. – Univ. Chicago Press.
- Shine, R. 2003. Locomotor speeds of gravid lizards: placing “costs of reproduction” within an ecological context. – *Funct. Ecol.* 17: 526–533.
- Sinervo, B., Miles, D., Frankino, A. W. et al. 2000. Testosterone, endurance and Darwinian fitness: natural and sexual selection on the physiological bases of alternative male behaviors in side-blotched lizards. – *Horm. Behav.* 38: 222–233.
- Stamps, J. A. and Krishnan, V. V. 1997. Functions of fights in territory establishment. – *Am. Nat.* 150: 393–405.
- Tokarz, R., McMann, R. S., Seitz, L. et al. 1998. Plasma corticosterone and testosterone levels during the annual reproductive cycle of male brown anoles (*Anolis sagrei*). – *Physiol. Zool.* 71: 139–146.
- Tokarz, R. R., Paterson, A. V. and McMann, S. 2003. Laboratory and field test of the functional significance of the male’s dewlap in the lizard *Anolis sagrei*. – *Copeia* 2003: 502–511.
- Tokarz, R. R., Paterson, A. V. and McMann, S. 2005. Importance of dewlap display in male mating success in free-ranging brown anoles (*Anolis sagrei*). – *J. Herpetol.* 39: 174–177.
- Tramontin, A. D. and Brenowitz, E. A. 2000. Seasonal plasticity in adult brains. – *Trends Neuro.* 23: 251–258.
- Van Buskirk, J. and Saxer, G. 2001. Delayed costs of an induced defense in tadpoles? Morphology, hopping, and development rate at metamorphosis. – *Evolution* 55: 821–829.
- Vanhooydonck, B. and Van Damme, R. 1999. Evolutionary relationships between body shape and habitat use in lacertid lizards. – *Evol. Ecol. Res.* 1: 785–805.
- Vanhooydonck, B., Herrel, A., Van Damme, R. et al. 2005. Does dewlap size predict male bite performance in Jamaican *Anolis* lizards? – *Funct. Ecol.* 19: 38–42.
- Verwajen, D., Van Damme, R. and Herrel, A. 2002. Relationships between head size, bite force, prey handling efficiency and diet in two sympatric lacertid lizards. – *Funct. Ecol.* 16: 842–850.
- Wainwright, P. and Reilly, S. M. 1994. Ecological morphology: integrative organismal biology. – Univ. of Chicago Press.
- Wainwright, P. C., Alfaro, M. E., Bolnick, D. I. et al. 2005. Many-to-one mapping of form to function: a general principle in organismal design? – *Integr. Comp. Biol.* 45: 256–262.
- Whiting, M. J., Nagy, K. A. and Bateman, P. W. 2003. Evolution and maintenance of status-signalling badges: experimental manipulations in lizards. Lizard social behavior. – Johns-Hopkins Press, pp. 47–82.
- Wilson, R. S., Kraft, P. G. and Van Damme, R. 2005. Predator-specific changes in the morphology and swimming performance of larval *Rana lessonae*. – *Funct. Ecol.* 19: 238–244.
- Wikelski, M. and Thom, C. 2000. Marine iguanas shrink to survive El Nino: changes in bone metabolism enable these lizards to reversibly alter their length. – *Nature* 403: 37–38.
- Winemiller, K. O. 1991. Ecomorphological diversification in lowland freshwater fish assemblages from five biotic regions. – *Ecol. Monogr.* 61: 343–365.
- Zahavi, A. 1975. Mate selection – a selection for a handicap. – *J. Theor. Biol.* 53: 205–214.