Prey-associated head-size variation in an invasive lizard in the Hawaiian Islands

MELISSA J. VAN KLEECK1, LUCIANO M. CHIAVERANO2 and BRENDEN S. HOLLAND2*

1Department of Biology, University of Hawaii at Manoa, Honolulu, HI, USA
2Pacific Biosciences Research Center, University of Hawaii at Manoa, Honolulu, HI, USA

Received 30 April 2015; revised 3 June 2015; accepted for publication 4 June 2015

Biological invasions are recognized as a primary driver of large-scale changes in global ecosystems. This study addresses ecomorphological variation in head size within and among populations of an ecologically destructive invasive predator, and evaluates the potential roles of environmental components in phenotypic differentiation. We used four size-corrected measurements of head morphology in Jackson’s chameleons, Trioceros jacksonii xantholophus (N = 319), collected from multiple Hawaiian Islands to assess phenotypic variation among and within islands. Results of analysis of variance (ANOVA) comparing chameleon head size (PC1) among islands revealed significant differences (mean difference > 5%) associated with variation in both rainfall and diet composition using Mann-Whitney U-tests and chi-squared analyses. These results suggest that morphological differentiation among populations from different islands has occurred over a relatively short ecological timescale, and is likely the result of ecomorphological adaptation to differences in exploited prey hardness. Intra-island allopatric population variation, however, was also detected in this study. Although we might expect that genetic change is the more likely explanation for differences between islands than within, and that plasticity may be more likely an explanation for the within- than the between-island differences, it is also possible that both within- and between-island patterns are the results of genetic change, or of plasticity. © 2015 The Linnean Society of London, Biological Journal of the Linnean Society, 2015, 116, 626–636.


INTRODUCTION

The age-old ‘nature vs. nurture’ argument historically assumed that phenotypic variation resulted from differences in either genotype or environment. As scientists have advanced our understanding regarding the interplay between gene expression and environmental factors, however, a more nuanced perspective has emerged, commonly phrased as gene-environment interaction. Understanding the complex interplay between genetic and environmental drivers over time is a burgeoning field with relevance to a variety of subdisciplines ranging from human disease etiology to ecology of biological invasions. Studies that examine patterns of ecomorphological variation in recently established populations with well-documented invasion histories may help to elucidate the mechanisms underlying the interactions between genetic and environmental factors.

Intraspecific phenotypic variation can occur in sexually reproducing populations of taxa as a result of environmentally induced variation in morphology, known as phenotypic plasticity, evolutionary adaptation over time to local environmental conditions and/or nonadaptive evolutionary change such as genetic drift (Losos, Warheit & Schoener, 1997; Boback, 2006; Measey, Hopkins & Tolley, 2009; Whitman & Argawal, 2009; Gianoli & Valladares, 2012). In reproductively connected populations that differ morphologically in spite of ongoing genetic exchange, phenotypic plasticity in response to different environmental cues is a possible mechanistic explanation (Price, Qvarn-
In situations, however, in which absolute barriers to gene flow are in place, for example when two species or allopatric populations of a single species share phenotypic characters only under certain conditions, phenotypic variance among populations may occur due to adaptation in response to differential selective forces in the local environments and genetic assimilation (Dlugosch & Parker, 2008; Wund et al., 2008; Torres-Dowdall et al., 2012).

Biological invasions are recognized as a primary driver of large-scale changes in global ecosystems and provide opportunities to investigate microevolutionary variation and the relative roles of abiotic and biotic factors in differentiation. Phenotypic variation can play a crucial role in the successful establishment of nonnative species to novel environments (Sakai et al., 2001). For example, following introduction, release from native selective pressures can reduce or remove morphological and physiological constraints, which can result in rapid microevolutionary changes (Baker, Peck & Goldsmith, 1990; Sakai et al., 2001; West-Eberhard, 2005). Likewise, when individuals sharing a single genetically homogeneous lineage are released in different ecological settings, exposure to novel selective pressures can lead to lineage differentiation. Successful invasive lineages also undergo adaptation to local environmental conditions as they encounter novel selective pressures from biotic (predators, pathogens, competitors and prey resources; Elton, 1958) and abiotic factors (temperature, desiccation and habitat structure; Huey, Gilchrist & Hendry, 2005; Urbansky et al., 2012).

Deleterious impacts of anthropogenic biological introductions to native biodiversity have been well documented (e.g. Mack et al., 2000), islands being particularly vulnerable to biological invasion (Li, Wu & Duncan, 2006). Geographic isolation of islands may also help to drive rapid morphological and genetic divergence among populations of newly isolated founding populations of introduced taxa (Baker & Moed, 1979, 1987; Ross & Baker, 1982; Huey et al., 2000). In the presence of environmental heterogeneity, geographically fragmented, patchy distributions of taxa with low vagility, can lead to local phenotypic adaptation among populations in a relatively short period of time (Losos et al., 1997; Grant & Grant, 2002; Welsh et al., 2013). Especially in herpetofauna, however, it has been shown that morphological plasticity can also play an important role in localized variation (Shine et al., 2012; Aguilar-Kirigin & Naya, 2013) despite varying levels of genetic connectivity (Lind et al., 2011). Although ecomorphological variation is observed in sexually reproducing taxa (Bickel & Losos, 2002; Kolbe, Larson & Losos, 2007; Measey et al., 2009), few, if any studies have assessed phenotypic evolution and the associated environmental drivers of variation in sedentary nonnative species with allopatric population structure.

The introduction of Jackson's chameleons (Trioceros jacksonii xantholophus) to the Hawaiian Islands in 1972 is well documented (Fig. 1), although predatory ecology, movement behavior and environmental impacts have only recently begun to receive scientific attention (Hagey, Losos & Harmon, 2010; Holland, Montgomery & Costello, 2010; Goldberg & Kraus, 2011; Kraus & Preston, 2012; Kraus et al., 2012; Chiaverano & Holland, 2014; Chiaverano, Wright & Holland, 2014). In 1998, regulations were adopted prohibiting inter-island transport of chameleons in Hawaii (Carpenter, Rowcliffe & Watkinson, 2004; Kraus et al., 2012), initiating complete geographic isolation of island populations. Preliminary results of a molecular population genetic analysis (B.S. Holland, M.J. Van Kleeck, unpubl. data) indicate extremely low genetic diversity in chameleons in Hawaii, as expected for a single release of a small number of individuals from a single geographic source, providing an opportunity to examine the potential role localized phenotypic variation among genetically homogeneous populations, in an introduced species over a given time interval.

As variation in environmental factors can result in different selective pressures on functionally important morphological features such as head size among populations, the hypothesis we propose is that following several dozen generations of geographic isolation, morphological differentiation will occur among insular populations as a result of differential selection due to environmental heterogeneity of different Hawaiian Islands. In order to test this hypothesis we collected morphological data using four size standardized skull measurements in chameleons from three islands (Hawaii, Maui, and Oahu) and tested for correlation among these features and environmental factors including rainfall and differences in exploited arthropod prey hardness (based on prey type) from gut content data. Our objectives were to: (1) quantify the extent of intraspecific skull size variation within and among multiple populations of T. j. xantholophus from three islands; and (2) conduct and evaluate correlation analyses of morphometric patterns and potential environmental drivers of phenotypic variation in this species.

METHODS

SPECIMEN COLLECTION, MATERIAL EXAMINED, AND ENVIRONMENTAL DATA

We measured 169 preserved Jackson’s chameleon museum specimens (Bernice P. Bishop Museum, Honolulu Hawaii) collected from 1998 to 2010, and 150 chameleons field-collected by hand at night...
between 2011 and 2014, from multiple locations on the islands of Hawaii, Maui and Oahu (Fig. 1). For each individual location (i.e. latitude and longitude) we assigned the correspondent value of annual precipitation using the Rainfall Atlas of Hawaii (Giambelluca et al., 2013).

MORPHOLOGICAL DATA COLLECTION
Five morphological measurements were recorded from each specimen using a digital caliper (± 0.01 mm): (1) snout-vent length (SVL), (2) head length (HL) as the distance between top of cask to snout, (3) head width (HW) as distance between the midline of each eye, (4) head height (HH) as the distance from bottom of jaw to the top of the cask, and (5) jaw length (JL) as the distance from posterior edge of jaw to the end of the snout (Fig. 2).

GUT CONTENT DATA
To address and compare differences in arthropod prey exploitation among islands, we dissected 21 live collected chameleons from Oahu, and stomach contents were sorted and identified under a dissecting microscope. In addition, we compiled published data from a subset of museum specimens from Maui (N = 34; Kraus et al., 2012) and the island of Hawaii (N = 33; Kraus & Preston, 2012). Prey were identified to order and categorized as hard or soft, determined by average force needed to crush individuals of arthropod orders following the methods of Herrel et al. (2001), Verwaijen, Van Damme & Herrel (2002), and Measey et al. (2011) (Table 1).

STATISTICAL ANALYSIS
We first performed univariate ANCOVAs using log-transformed head size variables as the dependent variable and log SVL as a covariate to test for shared allometries among islands. ANCOVAs revealed differences in the rate of growth (i.e. slopes) in head length among islands (see Supporting Information, Table S1); thereby, morphological features were then size-corrected using the normalization technique by Lleonart & Thorpe (Thorpe, 1975, 1976; Reist, 1985; Lleonart, Salat & Torres, 2000). This transformation standardizes each individual to the same SVL, and
adjusts morphological measurements taking into account differences in allometries among populations, using the formula:

\[ Y^* = Y_i (SVL_m / SVL_i)^b \]

where \( Y^* \) is the size-corrected morphological feature in the individual \( i \), \( Y_i \) is the measured morphological feature in individual \( i \), \( SVL_i \) is the snout-to-vent length of individual \( i \), \( SVL_m \) is the mean (arithmetic) snout-to-vent length of all individuals in the dataset, and \( b \) is the within-island slope of the linear regression between \( \ln(SVL_i) \) and \( \ln(Y_i) \). This size-correction method has been previously used in several taxa, and it has proven to be a robust technique tool for not only removing size effects on morphological variables but also comparing size independent morphology among groups with or without shared allometries (e.g. Rosso, Castellano & Giacoma, 2005; Peig & Green, 2009; Sistrom et al., 2012). The Lleonart–Thorpe method was successful at eliminating size effects on head variables of Jackson's chameleons for this study (see Supporting Information, Fig. S1, Table S2). We also applied the Shearing method, a widely used size-correction technique, to our dataset (Supporting Information, Table S3, Fig. S2). Although this method was also successful at removing size effects on head variables, we chose not to use it because the assumption of homogeneity of slopes among groups is still violated (McCoy et al., 2006) due to \( b \) in our dataset not being constant for all head size variables among islands (see above). All size-corrected features were log-transformed and included in a Principal Component Analyses (PCA, covariance matrix) due to high correlation among them. The first principal component (PC1) was used as a proxy for head size in relation to body size, as all feature loadings scored positively and significantly (\( r > 0.75, P < 0.001; 65\% \) of total variation) on this axis (Table 2). A two-way multivariate ANOVA (MANOVA) was used to test for potential significant effects of location (islands, fixed factor) and gender (fixed factor) on PC1, which were followed by univariate ANOVA's. In addition, collection locations were categorized as low (0–1500 mm) or high (2500–4000 mm) rainfall and Mann–Whitney \( U \)-tests were performed using PC1 to compare head size differences in head size between low and high rainfall locations from the islands of Oahu and Maui. Specimens from Hawaii were excluded from this analysis because specimens were collected from a single location on that island. Variation in Jackson's chameleon diet among islands (i.e. percentage of soft vs. hard prey in diet) was assessed using chi-squared tests, and the potential association of diet composition with relative

![Figure 2. Dotted lines show measurements recorded from the heads of each individual T. j. xantholophus: head length (HL), head height (HH), jaw length (JL) and head width (HW). Male chameleon depicted (three prominent horns shown).](image)

**Table 1.** Relative abundance of hard and soft prey items, by order, found in the stomachs of dissected chameleons from each island. Oahu (\( N = 21 \)), Hawaii (\( N = 33 \)), Maui (\( N = 34 \))

<table>
<thead>
<tr>
<th>Order</th>
<th>Relative abundance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Oahu</td>
</tr>
<tr>
<td>Hard</td>
<td></td>
</tr>
<tr>
<td>Coleoptera</td>
<td>16.3</td>
</tr>
<tr>
<td>Gastropoda</td>
<td>2.1</td>
</tr>
<tr>
<td>Squamata</td>
<td>1.4</td>
</tr>
<tr>
<td>Diplopoda</td>
<td>0</td>
</tr>
<tr>
<td>Hemiptera</td>
<td>19.9</td>
</tr>
<tr>
<td>Dermaptera</td>
<td>1.4</td>
</tr>
<tr>
<td>Isopoda</td>
<td>0.7</td>
</tr>
<tr>
<td>Soft</td>
<td></td>
</tr>
<tr>
<td>Hymenoptera</td>
<td>9.9</td>
</tr>
<tr>
<td>Orthoptera</td>
<td>1.4</td>
</tr>
<tr>
<td>Collembola</td>
<td>0.07</td>
</tr>
<tr>
<td>Neroptera</td>
<td>0</td>
</tr>
<tr>
<td>Diptera</td>
<td>22.7</td>
</tr>
<tr>
<td>Lepidoptera</td>
<td>10.6</td>
</tr>
<tr>
<td>Araneae</td>
<td>7.1</td>
</tr>
<tr>
<td>Blattodea</td>
<td>2.1</td>
</tr>
<tr>
<td>Psocodea</td>
<td>1.4</td>
</tr>
<tr>
<td>Total hard</td>
<td>41.8</td>
</tr>
<tr>
<td>Total soft</td>
<td>55.27</td>
</tr>
</tbody>
</table>

© 2015 The Linnean Society of London, Biological Journal of the Linnean Society, 2015, 116, 626–636
head size (PC1) among islands was evaluated via Pearson Correlation Coefficient. All the statistical analyses were performed in Statistica v.6 (Statsoft Inc.).

RESULTS

In total, 319 individuals were examined in this study from 33 locations spanning three main islands (Fig. 1): Hawaii (males = 31, females = 12), Maui (males = 40, females = 29), and Oahu (males = 126, females = 81). Gender biased sampling was due to the natural patterns evident in the field.

HEAD MORPHOLOGY COMPARISON AMONG ISLANDS

There was neither a net increase nor a net decrease in relative head size (PC1) of chameleons over time, from 1998 to 2014 \( F(1,314) = 1.1, r = 0.06, P < 0.5 \). Data from all years were pooled. MANOVA detected a significant island effect \( [\text{Wilk's } \lambda = 0.96; F(4,622) = 3.01, P < 0.03] \) but no significant gender \( [\text{Wilk's } \lambda = 0.96; F(2,311) = 1.69, P < 0.2] \) and interaction \( [\text{Wilk's } \lambda = 0.99; F(2,311) = 0.73, P < 0.6] \) effects on relative head size (PC-Head) and SVL. ANOVA detected no significant differences in SVL among islands \( [\text{ANOVA}: F(2,312) = 0.46, P < 0.65] \), but indicated significant differences in PC1 among islands \( [\text{ANOVA}: F(2,312) = 5.7, P < 0.03] \). Chameleons from Oahu had significantly smaller heads than those of their counterparts from Hawaii (Tukey’s, \( P < 0.001 \); Fig. 3), while no significant differences in head size were detected between Maui and other islands (Tukey’s, \( P > 0.05 \); Fig. 3).

ENVIRONMENTAL EFFECTS

Chameleon diet composition in terms of prey hardness varied significant among islands \( (\chi^2 = 7.69, \text{d.f.} = 2, P < 0.03) \). Individuals from Hawaii showed an exploitation pattern with the highest percentage of hard prey items (62.5%), while in the diet of chameleons from Oahu, the lowest percentage of hard prey items was observed (37.3%; Fig. 4). Intermediate percentage values (52.5%) were found in individuals from Maui (Fig. 4). Significant differences in PC1were observed between low and high rainfall sites on both Maui \( (Z = -2.89, \text{d.f.} = 61, P < 0.005) \) and Oahu \( (Z = -2.08, \text{d.f.} = 182, P < 0.05) \; \text{Fig. 5}) \).

DISCUSSION

In this study we present evidence of both within and between-island variation in head size in an introduced predatory lizard over a relatively short ecological time frame (~43 generations). Larger heads in Jackson’s chameleons coincided with a higher proportion of hard prey consumed: individuals collected on the island of Hawaii had the largest heads and the highest percentage of hard prey in their guts among the three islands. Chameleon counterparts from Oahu exhibited the smallest heads and lowest percentage of exploited hard prey. Head size negatively correlated with rainfall on the islands of Oahu and Maui. This pattern likely represents an indirect effect on head size via an effect of precipitation levels on abundance of arthropods categorized as hard, as these two factors are known to be inversely related because hard chitinous exoskeletons tend to inhibit desiccation by water loss. Differences in head size in disparate environments may have resulted from the

Table 2. Character loadings and percentage of total variance explained for the first four principle components among four head morphometric characters of *T. j. xantholophus* \( (N = 319) \). Due to high % variance explained, PC1 was used in subsequent analyses

<table>
<thead>
<tr>
<th>Character</th>
<th>PC1</th>
<th>PC2</th>
<th>PC3</th>
</tr>
</thead>
<tbody>
<tr>
<td>HL</td>
<td>0.81</td>
<td>0.27</td>
<td>-0.21</td>
</tr>
<tr>
<td>HW</td>
<td>0.82</td>
<td>-0.54</td>
<td>0.17</td>
</tr>
<tr>
<td>HH</td>
<td>0.87</td>
<td>0.08</td>
<td>-0.33</td>
</tr>
<tr>
<td>JL</td>
<td>0.70</td>
<td>0.46</td>
<td>0.53</td>
</tr>
<tr>
<td>% Variance</td>
<td>65</td>
<td>15</td>
<td>10</td>
</tr>
</tbody>
</table>

action of divergent selective pressures resulting in local adaptation among islands in the absence of genetic connectivity, with a possible role for phenotypic plasticity as well.

As an organism’s morphology is tightly linked to its ecology (Miles, 1994), differences in features such as head size, observed in this study, have a number of potential environmental drivers. Variation in head size in reptiles has been attributed to a number of different ecological factors that include, sexual size dimorphism and diet composition (Herrel, Van Damme & De Vree, 1996; Herrel, Spithoven & Van Damme, 1999; Boback, 2006). In this study there was no evidence for sexual dimorphism in size-corrected head dimensions, although this species shows dimorphic head ornament characteristics (Fig. 1), so it is unlikely that variation in head morphology was driven by sexually associated behaviours as seen in a number of other chameleon species (Stuart-Fox & Whiting, 2005; Measey et al., 2009).

In various reptile taxa, however, prey hardness has been suggested as an important factor driving head size variation (Herrel et al., 1999, 2001), including chameleons (Measey et al., 2011). In this study, larger heads of T. j. xantholophus were associated with a significantly higher proportion of hard prey consumed based on gut content analysis (Fig. 4B), suggesting that inter-island geographic variation in head size could be due to local adaptation to differences in composition of prey exploited (i.e. hard vs. soft prey items). Geographically isolated populations are likely to diverge morphologically from one another given time as they approach local fitness optima, especially in the presence of distinct environmental and prey assemblage differences (Losos et al., 1997; Boback, 2006). In cases in which gene flow is
absent, divergent selection has been shown to drive local adaptation over short ecological time scales (O’Steen, Cullum & Bennet, 2002; Torres-Dowdall et al., 2012).

Relative head size of Jackson’s chameleons was significantly smaller at high rainfall locations on Oahu and Maui (Fig. 5). Although rainfall may not directly impinge on morphological variation (James, 1970), ecological factors that are correlated with rainfall, such as resource availability and quality, may drive variation in functional features such as head size, among others (Nevo, 1981; Grant & Dunham, 1990; Tracy, 1999). For example, prey type and availability have been shown to vary with precipitation levels (e.g. Janzen & Schoener, 1968; Field, 1975; Strong & Sherry, 2000), and food abundance has shown to be higher with higher rainfall in a seasonal tropical ecosystem (Griffiths & Christian, 1996). Arthropods are the main food source for Jackson’s chameleons in the Hawaiian Islands (Kraus & Preston, 2012; Kraus et al., 2012), and these taxa tend to have thickened cuticles in arid environments to prevent desiccation (Hadley, 1972; Cloudsley-Thompson, 1975), while in wetter environments arthropods had thinner cuticles, and softer body types (Hadley, 2008). Higher rainfall has been shown to translate to increased availability of softer prey, requiring relatively lower bite force and smaller predator head size (Herrel et al., 1996, 2001). In fact Cape dwarf chameleons exhibit a preference for smaller, softer arthropod prey when available (Measey et al., 2011; Carne & Measey, 2013). The inverse relationship observed between chameleon head size and precipitation, therefore, is likely an indirect effect based on the link between damp environments and softer prey, suggesting that head size variation within populations may be a result of phenotypic plasticity, but further investigation will be required to confirm this notion. Intra-specific character gradients occur across multiple taxa (James, 1970; Grant & Dunham, 1990; Lind et al., 2011) and have been attributed to plastic responses to environmental variation as long as there is genetic connectivity among populations (Crispo, 2008). Although chameleon populations in Hawaii are patchily distributed, gene flow among populations has not been addressed within islands, therefore phenotypic plasticity and/or local adaptation in head size are both potential factors in within-island variation.

Results of this study suggest that local adaptation to environmental conditions, i.e. exploitation of different dietary resources, whether due to availability or selective preference, may be driving evolutionary divergence in head size of Jackson’s chameleon populations on different islands. Although we might expect that adaptation is the more plausible explanation for differences among islands than within, and that plasticity may be the more likely explanation for intra- than the inter-island differences, it is also possible that both patterns are the results of genetic change, or of plasticity. As we currently lack data regarding prey availability, molecular variance, fitness and ecomorphology from the native range in East Africa to support this assertion, alternative explanations should be also considered. As chameleons are opportunistic cruise forage predators, exploited diet composition may be affected by prey type availability, preference, and/or intra-specific competition for resources (Measey et al., 2011; Kraus & Preston, 2012; Kraus et al., 2012; Carne & Measey, 2013). To elucidate whether prey preference is a factor influencing head size variation, however, data regarding prey availability are needed. It is possible that the observed head size differences among islands are a result of neutral variation due, for example, to multiple founder events, in which small numbers of chameleons likely established each sampled population, and sampling bias has resulted in the phenotypes exhibited by those small populations. In previous studies, phenotypic plasticity has also been invoked as an important factor in facilitating invasive lineage establishment, particularly during early phases of range expansion, and could be a contributing factor in the patterns seen among islands (Yeh & Price, 2004; Richards et al., 2006).

Modern theoretical predictions regarding the role of plasticity in morphological variation over geographic space, originally termed the Baldwin effect (Baldwin, 1896; also see Robinson & Dukas, 1999), posit that colonization of a novel environment leads to transient initial increases in plasticity, which accelerate evolution of a new phenotypic optimum, followed by slow genetic assimilation of this phenotype and secondary reduction of plasticity (Pigliucci, Murren & Schlichting, 2006; Lande, 2015). Jackson’s chameleons have been established in the Hawaiian Islands in excess of 43 generations, and have likely reached numbers in the tens of thousands on each of the three islands sampled in this study. Strict enforcement of regulations prohibiting inter-island transport has effectively eliminated the possibility of inter-island gene flow (Kraus et al., 2012). Assuming that the larger head ecomorph and its concomitant enhanced bite force is an optimal phenotype under the lower precipitation/harder prey habitat scenario observed on the island of Hawaii, this phenotype likely evolved on a relatively short, ecological time scale. These chameleon populations, therefore, may have entered the phase of reduced plasticity, rather than initial invasion, and if this is indeed the case, we may have documented insipient adaptive divergence of Oahu and Hawaii populations. Although dif-
ferentiating the relative roles of plasticity and adaptation in generation of biodiversity remains challenging (Westley, 2011), both theoretical predictions and case studies support this conclusion (Nosil, 2012; Westley, Ward & Fleming, 2013; While et al., 2015). Successful establishment of anthropogenically released nonnative taxa is one factor that is dramatically altering our global ecosystems, but it can also provide opportunities to investigate the pace and process of microevolutionary change. In such instances when lineages with reduced genetic diversity are placed into varied, novel environments, the primary drivers, whether plasticity or adaptation, can be addressed and potentially elucidated. The role of ecomorphological plasticity in phenotypic differences in T. j. jacksonii within islands with potential for gene flow should also be considered, especially in 'new', ongoing range expansions (West-Eberhard, 2005; Lind et al., 2011). In further studies, integrated genomic, ecomorphological and behavioural approaches will be useful in elucidating relative genetic vs. environmental contributions to evolutionary change in established nonnative populations. Such studies that take place during contemporary invasion and range expansion events can detect biological change as it plays out, on ecological time scales, and therefore hold potential to provide real-time insights into the ability of species to adapt in the face of our rapidly changing global ecosystems.

ACKNOWLEDGEMENTS

First, we thank the Hawaii Department of Land and Natural Resources, and Oahu Army Natural Resources Program for providing permits, specimens, access, and funding. For field assistance (despite ‘buckets of rain’) we thank Vincent Costello, Jamie Tanino, Stephanie Joe, Bonnie Dietrich, Thomas Arthur Hooper Smith, Cierra Howard, Marat Khramov, Rose Rico, Zachary Williams, Bob Thompson, Amber Wright, Kyle Edwards, and Levi Gray. For access to museum collections we thank Molly Hagemann and the staff of the Bernice Pauahi Bishop Museum. We are grateful to Jonathan Losos for helpful discussions during the preparation of this manuscript, and to three anonymous reviewers for their valuable insights and helpful comments.

REFERENCES


SUPPORTING INFORMATION

Additional Supporting Information may be found in the online version of this article at the publisher’s website:

Figure S1. Relationships among raw head dimension variables and SVL in mm (A–D), size-corrected variables and log SVL using Lleomart–Thorpe method (E–H), and size-corrected variables and log SVL using residuals (I–L) for all islands (Hawaii: triangles, Maui: filled circles, and Oahu: open circles). Head size variables are (A, E and I) head length; (B, F and J) head width; (C, G and K) head height; (D, H and L) jaw length. (A, E) head length; (B, F) head width; (C, G) head height; (D, H).
**Figure S2.** Relative chameleon head size among islands calculated from PCA of residuals of regression between log-corrected head size variables and log SVL. Increasing PC scores indicate an increase in head morphometric variables. Lowercase letters represent significant differences among sites using ANOVA \(F_{(2,317)} = 6.42; \ P < 0.003\).

**Table S1.** Results of univariate ANCOVAs comparing each head morphometric (log-transformed) among islands using SVL (log-transformed) as covariate. Critical assumption of homogeneity of slopes is violated for head length, indicating different allometries.

**Table S2.** Linear regressions between raw and size-corrected head measurements and SVL. The Lleonart–Thorpe size-correction method used here does eliminate the effect of body size for all variables. HL, head length; HH, head height; HW, head width; JL, jaw length.

**Table S3.** Results of the principal component analysis (PCA) including head features that were size-corrected using the Shearing method. This method uses the residuals of the linear regressions between each head feature (log-transformed) and SVL (log-transformed). Feature loadings and percentage of the total variance explained by each principal component (PC) are shown.