



Functional divergence between morphs of a dwarf chameleon: differential locomotor kinematics in relation to habitat structure

TIMOTHY E. HIGHAM^{1*}, G. JOHN MEASEY², ALEKSANDRA V. BIRN-JEFFERY¹, ANTHONY HERREL^{3,4} and KRYSTAL A. TOLLEY^{5,6}

¹Department of Biology, University of California, 900 University Avenue, Riverside, CA, 92521, USA

²Centre for Invasion Biology, Department of Botany & Zoology, Stellenbosch University, Merriman Avenue, Stellenbosch, South Africa

³Département d'Ecologie et de Gestion de la Biodiversité, Centre National de la Recherche Scientifique/Muséum National d'Histoire Naturelle, 55 rue Buffon, 75005, Paris, France

⁴Ghent University, Evolutionary Morphology of Vertebrates, K.L. Ledeganckstraat 35, B-9000, Gent, Belgium

⁵Applied Biodiversity Research Division, South African National Biodiversity Institute, Claremont 7735, Cape Town, South Africa

⁶Department of Botany & Zoology, Stellenbosch University, Merriman Avenue, Stellenbosch, South Africa

Received 23 January 2015; revised 18 March 2015; accepted for publication 18 March 2015

Arboreal lizards are extremely effective at moving in structurally complex habitats, including surfaces of varying diameter and incline. Chameleons exemplify this by exhibiting a number of morphological specializations for moving in these habitats, including the use of prehensile feet and tail to grasp branches. Despite their unique morphology and behaviour, little is known about how locomotor movements vary between species. In addition, some species, such as the Cape Dwarf Chameleon, *Bradypodion pumilum*, consist of two morphs that differ in ecology, morphology, and behaviour. The two morphs can be found in either closed canopy woodland habitat or relatively open fynbos habitat. The morph that occupies the woodland habitat tends to be larger and utilizes larger diameter perches. Although their ecological and morphological divergence is established, whether this translates into differences in three-dimensional kinematics of locomotion is not known. Given the potentially strong selective pressures from structurally different habitats, kinematic differences might reveal the functional basis of incipient speciation. We determined that the two morphs diverge significantly in multidimensional kinematic space, and that this occurs for the forelimb and hindlimb independently. These differences outweigh the effects of substrate within each morph, although the differences between morphs were more pronounced on the vertical treatments. © 2015 The Linnean Society of London, *Biological Journal of the Linnean Society*, 2015, 00: 000–000.

ADDITIONAL KEYWORDS: biomechanics – *Bradypodion* – climbing – hindlimb – locomotion – Western Cape.

INTRODUCTION

An animal's phenotype often reflects, to some degree, the ecological constraints and pressures that it experiences. This relates not only to differences in habitat structure, but also can be impacted by other factors,

such as temperature, competition, and predation (Lavin & McPhail, 1986; Sinervo & Huey, 1990; Van Damme, Aerts & Vanhooydonck, 1998; Herrel, Meyers & Vanhooydonck, 2001; Brinsmead & Fox, 2002; Ghalambor, Walker & Reznick, 2003; Kerfoot & Schaefer, 2006; Herrel *et al.*, 2011; Logan *et al.*, 2012). Interpopulation studies are excellent for teasing apart these pressures and constraints (Garland

*Corresponding author. E-mail: thigham@ucr.edu

& Losos, 1994), especially given that the component of the phenotype resulting from the evolutionary history, which can significantly impact interspecific comparison, is minimal or non-existent. Therefore, this is a critical taxonomic level to examine given that it reflects potential incipient speciation and, consequently, critical evolutionary patterns and processes (Sinervo & Losos, 1991; Macrini & Irschick, 1998; Van Damme *et al.*, 1998; Herrel *et al.*, 2001).

Divergence in habitat use between populations of lizards can lead to disparity in morphology (Malhotra & Thorpe, 1997; Herrel *et al.*, 2001, 2011; Gifford, Herrel & Mahler, 2008; Hopkins & Tolley, 2011; Collins, Russell & Higham, 2015) and performance (Macrini & Irschick, 1998; Gifford *et al.*, 2008; Herrel *et al.*, 2011; Hopkins & Tolley, 2011), although intra-specific studies are relatively rare. Given the incredible range of habitats that lizards occupy, and the numerous morphological specializations associated with habitat type, the link between morphology and habitat is not surprising. Correlations between habitat structure and morphology among populations often reflect varying demands placed on the locomotor system. Although differences in morphology are excellent for predicting functional relationships and trade-offs in response to changing demands (Herrel *et al.*, 2001), biomechanical analyses are needed to link the two. If functional differences can be observed in conjunction with morphological differences, then a mechanistic link can be established.

Quadrupedal vertebrates have more flexibility in executing locomotor movements than bipedal vertebrates given that four limbs, instead of two, can share function. Despite this, most work has focused on the hindlimbs. This is not sufficient for understanding arboreal locomotion given the importance of the forelimb for pulling an animal up when climbing (Cartmill, 1985; Foster & Higham, 2012), as well as assisting with locomotion on downhill surfaces. In addition, the forelimb has the potential to offer sensory feedback for the subsequent hindlimb footfall given that they will contact a new surface prior to the hindlimbs. Regardless, it is difficult to make conclusions regarding locomotor adaptations to a given habitat without examining all of the elements important for locomotion. In addition, it is important to examine these propulsive elements during natural behaviours, which, in the case of arboreal locomotion, often involves moving on varying inclines and perch diameters (Mattingly & Jayne, 2004).

Animals that live in arboreal or saxicolous habitats, which require climbing, often exhibit morphological or behavioural changes to facilitate this type of locomotion (Zaaf *et al.*, 1999, 2001; Clemente, Withers & Thompson, 2013; Birn-Jeffery & Higham,

2014b; Higham *et al.*, 2015). Chameleons exhibit a unique mode of locomotion that is well suited for an arboreal lifestyle (Peterson, 1984; Bickel & Losos, 2002; Higham & Jayne, 2004; Fischer, Krause & Lilje, 2010; Herrel *et al.*, 2013b; Higham & Anderson, 2013; Krause & Fischer, 2013). Their prehensile feet allow them to grip a branch strongly when climbing (Herrel *et al.*, 2013b). This anchor point allows them to pull and push with both their hindlimbs and forelimbs (Higham & Jayne, 2004). Analyses of limb kinematics in chameleons are restricted to *Chamaeleo calyptratus*, despite the fact that other genera of chameleons exhibit extensive diversity in ecology and morphology (Bickel & Losos, 2002). For example, there is considerable variation in species from the southern African genus *Bradypodion*, highlighted by the recent studies of their performance, morphology, and ecology (Butler, 2005; Resinger, Stuart-Fox & Erasmus, 2006; Measey, Hopkins & Tolley, 2009; Stuart-Fox, 2009; Herrel *et al.*, 2011, 2013b; Hopkins & Tolley, 2011; Measey *et al.*, 2011; Carne & Measey, 2013; da Silva & Tolley, 2013; da Silva *et al.*, 2014). It has been noted that populations of the Cape Dwarf Chameleon, *Bradypodion pumilum*, occupy different types of habitat (open fynbos habitat or closed canopy woodland habitats) (Fig. 1) and they exhibit corresponding differences in morphology (Measey *et al.*, 2009; Hopkins & Tolley, 2011), habitat use (Herrel *et al.*, 2011), and locomotor performance (Herrel *et al.*, 2011). Compared to the woodland habitat, the fynbos habitat, a Mediterranean-type heathland, is common in the southern Cape region of Africa (Tolley *et al.*, 2006). The closed canopy morphs (henceforth referred to as the 'woodland morph'), in comparison with the fynbos morphs, are typically larger bodied, brighter in coloration, have higher casques, and have longer tails (Hopkins & Tolley, 2011). In addition, the woodland morph uses larger diameter perches (for sleeping) and sprints faster than the open fynbos morph (Herrel *et al.*, 2011). Despite the differences between the morphs, it is unclear whether the three-dimensional limb movements also differ between the morphs in association with morphological and ecological differences.

We examined the functional divergence between morphs of *B. pumilum* occupying habitats with different structure aiming to test the hypotheses: (1) the three-dimensional movements of the morphs will differ, and this will be related to their respective habitat; (2) the greatest separation between the morphs will occur on the narrow vertical perch, given that it is probably the most demanding treatment; and (3) forelimb and hindlimb motion will be integrated for both morphs, such that hindlimb movements will be predicted by preceding forelimb movements.



Figure 1. Images showing the two morphs and their respective habitats. The woodland morph was collected in Stellenbosch (top) and the fynbos morph was collected in Kogelberg (bottom).

MATERIAL AND METHODS

ANIMALS

Individuals from two morphs of *B. pumilum* were collected in South Africa in January 2012. The fynbos morph chameleons were collected from the Kogelberg Nature Reserve ($N = 5$, snout-vent length = 60.3 ± 2.2 mm, mass = 4.6 ± 0.5 g) and woodland morphs were collected from Stellenbosch ($N = 4$, snout-vent length = 68.1 ± 2.3 mm, mass = 6.6 ± 0.9 g), as described in previous studies (Herrel *et al.*, 2011). These two habitats are non-overlapping, and a specific morph has not been observed in the other morph's habitat type. Lizards were brought back to the laboratory in breathable opaque cotton bags and held for no more than 24 h. During this time, the bags were kept moist to maintain hydration. After the experiments, lizards were released at the place of capture. Animals were not fed during this period of time, although they were hydrated regularly.

HABITAT MEASUREMENTS

Perch diameter availability (fynbos: 1.35 ± 0.93 mm; woodland: 3.42 ± 1.71 mm) and use (during sleep) for both morphs was quantified in a previous study, and shows that the fynbos habitat has significantly smaller diameter perches available (Herrel *et al.*, 2011). In the present study, we also quantify the incline (perch angle) of the available habitat using two 100-m transects in each habitat in a 1-m wide

swath, every 10 m along the transect. Angles between 0° and 90° were recorded, and values of 0° were converted to 1° to facilitate the log transformation. Both transects were combined for each habitat. The fynbos (Kogelberg) is structurally low (approximately 1 m high) and so angle measurements were taken 10 cm from the top of the vegetation because this is the height at which chameleons are usually found when perching at night. For the woodland habitat (Stellenbosch), measurements were taken at 1.5 m above the ground, although chameleons can be found from approximately 1 m to several metres in height. These measurement heights are consistent with perch heights that chameleons utilize and were, therefore, considered representative.

EXPERIMENTS

Chameleons were filmed walking on perches of varying incline (0° , 45° and 90°) and diameter (small, 3 mm; large, 9 mm). The small perch diameter reflects the smallest dowel that was available, and our goal was to avoid any significant deflections of the surface, which would probably have other confounding impacts. Dots of white correction fluid were placed on a number of locations on the chameleons (described below) to visualize the movements on the videos. Chameleons were encouraged to walk across the perch by gently tapping the tail. Each trial was filmed using two high-speed video cameras (Photron

APX-RS and Casio Exilm) operating at 200 Hz. Lateral and dorsal views were obtained simultaneously, and both cameras were synchronized using a flashing light-emitting diode held within the view of both cameras. Multiple trials were obtained from each individual.

DATA ANALYSIS

Three-dimensional coordinates of markers on the dorsal midline (five to seven markers), between the two girdles, as well as limb markers on the shoulder/hip, elbow/knee, wrist/ankle, and digit tips were digitized using DLTDV3 (Hedrick, 2008) in MATLAB, release R2012b (The Mathworks Inc.) (Fig. 2). The x , y , and z -axes represent the fore-aft, vertical, and medio-lateral planes, respectively. After digitization, the coordinates were processed using custom written script in MATLAB to obtain a proxy for centre of mass, joint angles, and spatio-temporal characteristics for both forelimb and hindlimb. The dorsal markers along the spine were averaged to obtain the centre of mass proxy, which was then used to determine speed. Stride length and step length were calculated based on the mid dorsal spine marker.

All three-dimensional joint angles were calculated in accordance with methods described previously (Higham & Jayne, 2004; Fuller, Higham & Clark, 2011; Foster & Higham, 2012). Smaller values for the elbow, wrist, knee, and ankle angles represent greater flexion at those joints. Depression, retraction, and long-axis rotation were calculated for the most proximal limb bone (humerus/femur) to describe the three-dimensional movements (Foster & Higham, 2012). Negative values of retraction indicate greater protraction (anterior to the girdle), whereas positive

values indicate retraction (posterior to the girdle). Positive values of depression indicate greater depression (below the girdle) and negative indicates greater elevation (above the girdle). Clockwise rotations are positive angles, whereas anti-clockwise rotations are negative angles. Finally, pectoral girdle rotations were also calculated where positive values show that the left shoulder is in front of the right shoulder and vice versa for negative values. Joint angles were measured at footfall (FF), mid-stance (MS) and end of stance (ES) events (Fig. 2).

STATISTICAL ANALYSES

All statistics were run in JMP, version 11 (SAS) and/or MATLAB. Perch angle availability was compared using a Mann–Whitney test. Discriminant function analyses (DFAs) were used to test the separation of the morphs in multivariate kinematic space, although only 0° and 90° were used to minimize the number of tests. A total of 18 kinematic variables from each of the hindlimb and forelimb were included in each DFA. Each DFA included all perch diameters for a given combination of incline (level or vertical) and limb (forelimb and hindlimb) and variables with loadings > 0.4 were considered significant for that specific axis. Successful classification as either fynbos or woodland was assessed, and the scores from the complete canonical matrix from the DFAs were then examined using two-way analysis of variance (ANOVA) (morph and diameter as the fixed independent factors) to determine whether the morphs occupied significantly different kinematic space. The dependent variable in these ANOVAs was the scores from the DFA. Key variables that loaded strongly in the DFAs were also used as dependent

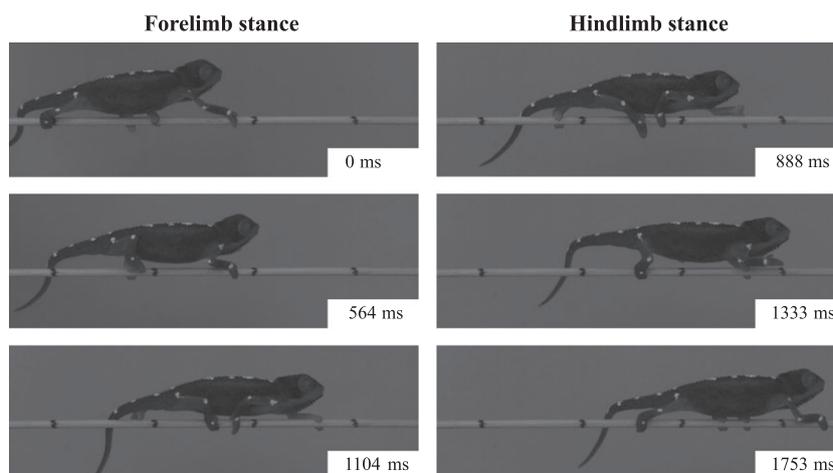


Figure 2. Still images of a fynbos morph moving on a small 0° perch. The left images indicate footfall (0 ms), mid-stance (564 ms), and end-stance (1104 ms) for the forelimb, and the right images reflect the same (888 ms, 1333 ms, and 1753 ms, respectively) for the hindlimb.

variables in univariate analyses. Three-way ANOVAs were used to examine the impact of morph (fixed factor), individual (random factor nested within morph), and treatment (fixed factor). Speed was not included as a factor because it did not significantly impact upon the variables of interest. Because individual was included as a factor, the denominator in the F -test was modified (Zar, 1996). Because of unequal sample sizes, Games-Howell post-hoc tests, rather than Tukey's honestly significant difference post-hoc tests, were used to determine which treatments exhibited significant differences for the morphs.

RESULTS

PERCH INCLINE AVAILABILITY

The mean perch angle in the fynbos habitat 10 cm below the top of the vegetation was 80.8°. The mean incline in the woodland habitat 1.5 m above the ground was 39.6°, which was significantly less vertical than the fynbos ($P < 0.001$).

MULTIVARIATE DIFFERENCES BETWEEN MORPHS

On the 90° treatments, morphs were classified correctly 100% of the time for each DFA. Correct classification was also high on the level for the forelimb and hindlimb, with values of 86% and 91.5%, respectively. From the DFAs and subsequent ANOVAs on the scores, the morphs were significantly different for the hindlimbs and forelimbs on all treatments ($P < 0.01$, ANOVAs) (Fig. 2). These separations in 'kinematic space' were especially evident for the forelimb on the vertical treatment (Fig. 3C). Depending on the DFA, different variables appeared to be more important for driving the separation. For example, the knee angle at ES was greater for the fynbos morph on the 90° conditions (Fig. 3D, Table 1), although the knee angle at FF was greater for the woodland morph on the same treatment (Fig. 3D, Table 1). For the forelimb on the 90° narrow condition, greater values of MS humerus retraction for the woodland morph appear to contribute to the separation along the DF1 axis (Fig. 3C, Table 1). The wrist angle at FF and MS is much greater for the fynbos and woodland morphs on the larger perch diameter than the narrow perch diameter on the 90° condition (Table 1).

MULTIVARIATE INTERACTIONS

Interaction terms (morph \times perch diameter) were significant in many cases, with the greatest interaction occurring on the level and vertical for the

hindlimb (Fig. 3B, D). For the level condition, hindlimb values for the morphs significantly separated on DF2 for the small perch, although these were not separated on the large perch condition (Fig. 3B). This suggests that the morphs diverge in kinematics on smaller perches on the level. On the vertical (for DF1), hindlimb values also separated for the morphs on the small (but not large) perch conditions (Fig. 3D). This appears again for the forelimb values on the vertical treatment along DF2, where the morphs differ on the small (but not large) perch condition (Fig. 3C).

UNIVARIATE DIFFERENCES BETWEEN MORPHS

From the univariate analyses, some striking differences were evident between the morphs for variables at FF (Fig. 4, Table 1). The elbow and knee angles at FF were significantly greater for the woodland morph on the small 45° and 90° treatments (Fig. 4A, B, Table 1), although only the elbow angle was greater for the woodland morph for the large 45° treatment (Fig. 4A, Table 1). Interestingly, the pectoral girdle was rotated more in the counterclockwise direction at FF in the fynbos morph for the small 45°, small 90°, large 0°, and large 90° treatments (Fig. 4C, Table 1). By contrast, the pelvic girdle was rotated more in the counterclockwise direction at FF for the woodland morph on the small 0°, small 45°, and large 45° treatments (Fig. 4D, Table 1).

Locomotor speed was relatively invariable among the fynbos individuals, regardless of condition (Table 1). However, the woodland morph exhibited a marked decrease in speed on the vertical conditions relative to the horizontal conditions. For example, the speed on the small 90° was, on average, 52% that of the small level condition, and the large 90° was, on average, 63% that of the large level condition (Table 1).

INCLINE AND PERCH DIAMETER

Changes in incline and perch diameter had significant impacts on limb kinematics for both morphs (Fig. 4, Table 1). For example, although elbow angle exhibited no significant differences between treatments in the woodland morph, the fynbos morph exhibited significantly lower elbow angles at FF on the small 90° treatment compared to the small 0°, large 0°, and large 90° treatments ($P < 0.05$) (Fig. 4A).

The effects of perch diameter were often dependent on the morph and the set of limbs. Proximal joint flexion at MS was not impacted by perch diameter for the hindlimb, although the elbow was more flexed at MS on the small 90° treatment compared to the large 90° ($P < 0.05$) (Table 1). Although the wrist

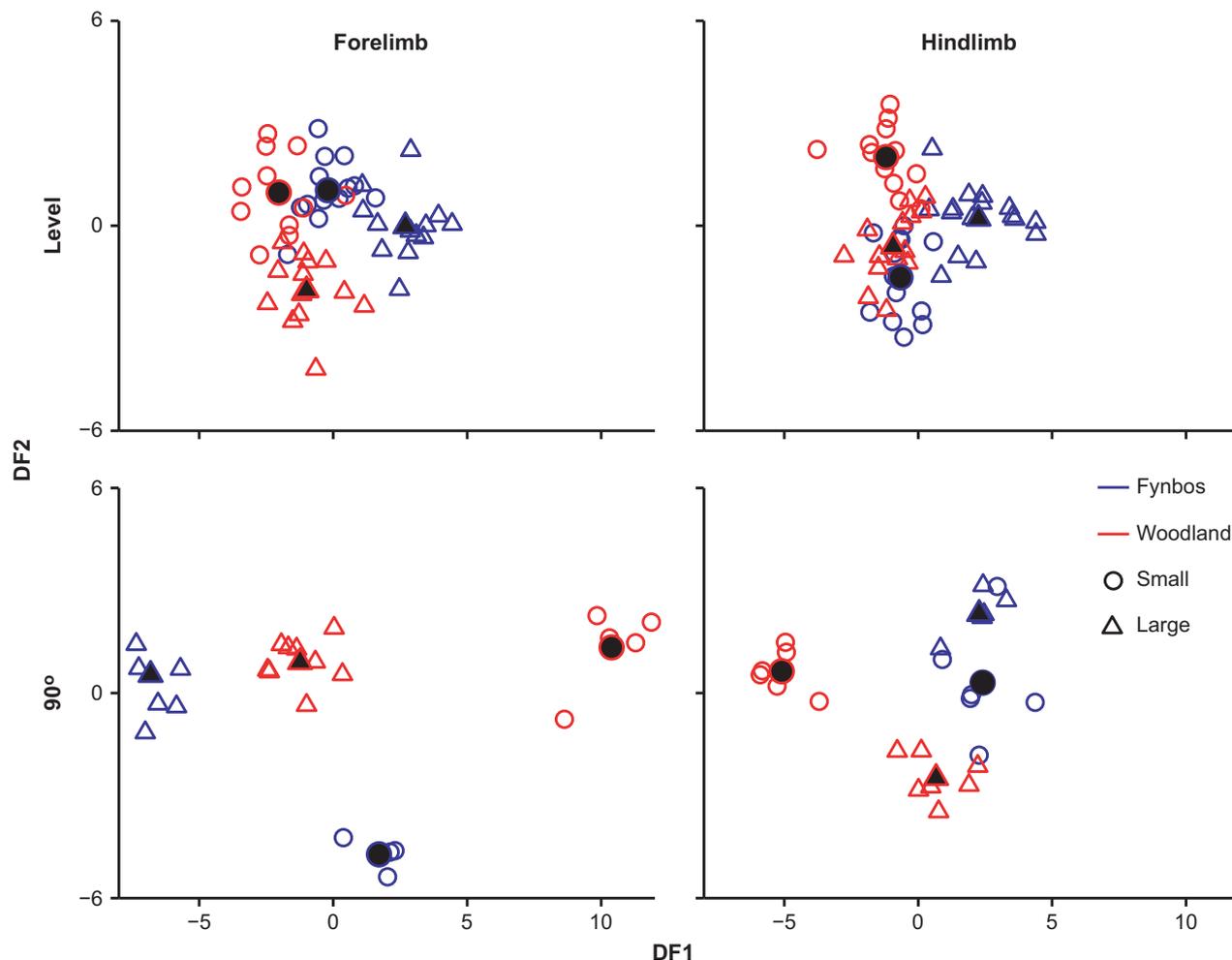


Figure 3. Scores from discriminant function analyses for the forelimb (left column) and hindlimb (right column) on 0° (top row) and 90° (bottom row) conditions. Correct classification reached 100% for both vertical discriminant function analyses (DFAs) and was above 80% for both level DFAs. The woodland morph is in red and the fynbos morph is in blue. In all cases, the morphs significantly separated in multivariate space (analysis of variance: $P < 0.01$).

and ankle were often more flexed on the small 90° treatments compared to the large 90° treatments, this would probably not contribute to a lower centre of mass given that these were at FF and ES. Hip height at MS was greater on the narrow level surface for the fynbos (10.7 ± 0.45 mm compared to 9.3 ± 0.39 mm, $P < 0.05$) and woodland (10.6 ± 0.65 mm compared to 7.8 ± 0.40 mm, $P < 0.05$) morphs compared to the large level surface. This was associated with greater degrees of femur and humerus depression at MS on the narrow level condition (Table 1).

COORDINATION WITHIN AND BETWEEN FORELIMBS AND HINDLIMBS

Although many values were not coordinated between different parts of the limb, the degree of pelvic

rotation was significantly (and positively) correlated with the degree of femur protraction at FF ($r^2 = 0.91$, $P < 0.001$) (Fig. 5A). In addition, changes in the forelimb were correlated with changes in the hindlimb in that the wrist angle at FF was positively correlated with the ankle angle at FF ($r^2 = 0.53$, $P < 0.05$) (Fig. 5B).

DISCUSSION

We found that the two morphs of *B. pumilus* varied dramatically in their limb kinematics, which supports our first hypothesis. The divergence in function is evident in both the forelimb and hindlimb, suggesting that both are critical for locomotion of the animal in its habitat. In support of our second hypothesis, the morphs appear to diverge more on

Table 1. Joint angle and general stride parameters

Variable	Fynbos			Woodland				
	Small 0	Large 0	Small 90	Large 90	Small 0	Large 0		
	Small 90	Large 90	Small 0	Large 0	Small 90	Large 90		
Speed (cm s ⁻¹)	4.1 ± 0.39	4.2 ± 0.5	3.6 ± 0.69	4.2 ± 0.40	4.8 ± 0.60	5.9 ± 0.36	2.5 ± 0.36	3.7 ± 0.60
Duty factor	0.6 ± 0.02	0.62 ± 0.04	0.63 ± 0.02	0.67 ± 0.02	0.68 ± 0.02	0.72 ± 0.02	0.71 ± 0.03	0.71 ± 0.04
Stride frequency (Hz)	0.65 ± 0.04	0.72 ± 0.07	0.64 ± 0.12	0.9 ± 0.07	0.81 ± 0.1	1.15 ± 0.07	0.46 ± 0.07	0.67 ± 0.09
Humerus retraction (FF)	-14.75 ± 8.1	-17.35 ± 8.6	-44.99 ± 30.9	-86.84 ± 23.9	-11.09 ± 6.3	-1.57 ± 3.3	-36.88 ± 8.2	-13.93 ± 10.6
Humerus retraction (MS)	56.3 ± 7.29	48.0 ± 6.4	13.7 ± 13.3	12.0 ± 6.3	58.1 ± 4.3	59.3 ± 5.0	56.1 ± 12.3	45.2 ± 6.0
Humerus retraction (ES)	58.14 ± 5.14	44.88 ± 5.39	23.6 ± 12.04	13.5 ± 4.64	55.57 ± 3.69	49.31 ± 3.7	38.61 ± 15.51	44.23 ± 7.57
Elbow angle (FF)	112.8 ± 5.05	112.57 ± 5.51	97.59 ± 5.92	123.29 ± 5.18	112.93 ± 3.31	117.4 ± 3.56	124.68 ± 9.4	111.11 ± 8.35
Elbow angle (MS)	56.3 ± 2.27	59.6 ± 4.42	64.86 ± 5.6	81.45 ± 2.58	48.89 ± 2.92	55.79 ± 3.28	64.56 ± 3.36	65.05 ± 3.2
Elbow angle (ES)	106.8 ± 3.08	99.96 ± 4.79	99.02 ± 3.99	107.17 ± 5.55	105.65 ± 2.79	107.25 ± 1.61	99.23 ± 9.19	96.26 ± 4.49
Wrist angle (FF)	109.4 ± 4.94	105.29 ± 8.08	94.61 ± 17.03	126.87 ± 7.34	111.58 ± 5.27	118.51 ± 5.99	68.83 ± 14.51	122.43 ± 6.54
Wrist angle (MS)	123.0 ± 4.81	125.5 ± 9.18	113.16 ± 10.59	143.76 ± 10.13	117.08 ± 5.92	130.05 ± 3.82	81.53 ± 11.78	143.89 ± 6.66
Wrist angle (ES)	119.3 ± 3.33	110.5 ± 9.97	117.94 ± 7.24	121.63 ± 4.29	114.5 ± 2.36	117.06 ± 2.13	121.18 ± 3.39	136.1 ± 2.33
Pectoral girdle rot. (FF)	-32.4 ± 8.26	-21.67 ± 7.24	13.33 ± 21.52	26.42 ± 16.36	-41.96 ± 3.85	-41.45 ± 3.25	-33.23 ± 4.84	-35.69 ± 7.72
Pectoral girdle rot. (ES)	18.6 ± 4.97	7.3 ± 7.93	56.59 ± 11.02	64.86 ± 5.92	21.94 ± 3.73	26.38 ± 3.35	40.96 ± 9.52	36.32 ± 8.64
Humerus depression (MS)	28.6 ± 7.29	15.98 ± 7.91	43.74 ± 12.9	54.5 ± 13.75	38.53 ± 5.07	43.04 ± 2.37	50.65 ± 15.48	39.01 ± 5.23
Femur retraction (FF)	-41.5 ± 3.78	-44.51 ± 4.93	-39.48 ± 14.16	-82.19 ± 23.59	-74.21 ± 10.79	-47.81 ± 11.46	-60.7 ± 14.12	-66.7 ± 15.48
Femur retraction (MS)	-21.8 ± 2.97	-16.48 ± 8.84	-39.39 ± 16.42	-15.64 ± 25.93	-49.69 ± 8.85	-46.51 ± 8.13	-37.52 ± 10.23	-52.54 ± 12.39
Femur retraction (ES)	40.2 ± 4.89	13.7 ± 6.9	17.99 ± 11.7	3.79 ± 7	23.53 ± 10.35	7.99 ± 10.04	31.77 ± 11.96	0.83 ± 9.95
Knee angle (FF)	136.2 ± 3.74	135.62 ± 5.12	131.27 ± 4.23	133.34 ± 2.43	131.57 ± 5.9	132.27 ± 2.03	148.3 ± 4.55	139.3 ± 4.37
Knee angle (MS)	89.3 ± 2.28	98.66 ± 3.39	83.08 ± 6.09	79.79 ± 3.25	81.28 ± 2.59	84.3 ± 1.84	76.76 ± 11.81	82.72 ± 3.63
Knee angle (ES)	127.2 ± 3.42	123.85 ± 4.35	114.49 ± 9.47	121.46 ± 4.96	128.36 ± 2.89	115.53 ± 3.44	95.73 ± 5.47	109.21 ± 5.21
Ankle angle (FF)	156.7 ± 3.15	154.05 ± 3.75	138.73 ± 10.1	149.33 ± 6.07	158.28 ± 4.27	164.46 ± 2.45	120.79 ± 9.36	161.95 ± 2.97
Ankle angle (MS)	111.9 ± 3.23	121.61 ± 3.26	97.97 ± 4.59	99.2 ± 5.57	104.35 ± 2.95	114.06 ± 4.58	77.11 ± 13.89	118.95 ± 4.48
Ankle angle (ES)	94.2 ± 1.64	102.24 ± 4.6	81.93 ± 5.05	87.51 ± 4.37	87.96 ± 3.59	89.44 ± 2.52	78.5 ± 11.51	86.19 ± 3.93
Pelvic girdle rot. (FF)	-32.8 ± 5.12	-27.35 ± 5.32	-42.38 ± 14.53	-5.89 ± 19.59	-12.38 ± 9.95	-28.35 ± 11.69	-27.57 ± 17.4	-13.02 ± 15.33
Pelvic girdle rot (ES)	4.7 ± 3.63	-6.26 ± 5.4	28.28 ± 15.09	22.2 ± 9.46	23.59 ± 9.47	22.77 ± 8.98	15.93 ± 10.02	38.85 ± 10.63
Femur depression (MS)	36.8 ± 3.52	22.18 ± 4.84	46.17 ± 9.33	55.37 ± 14.59	40.39 ± 1.93	38.18 ± 5	39.2 ± 9.71	23.89 ± 8.53

For all angles, the values are in degrees. FF, footfall; ES, end of stance; MS, middle of stance; rot, rotation; values are the mean ± SEM.

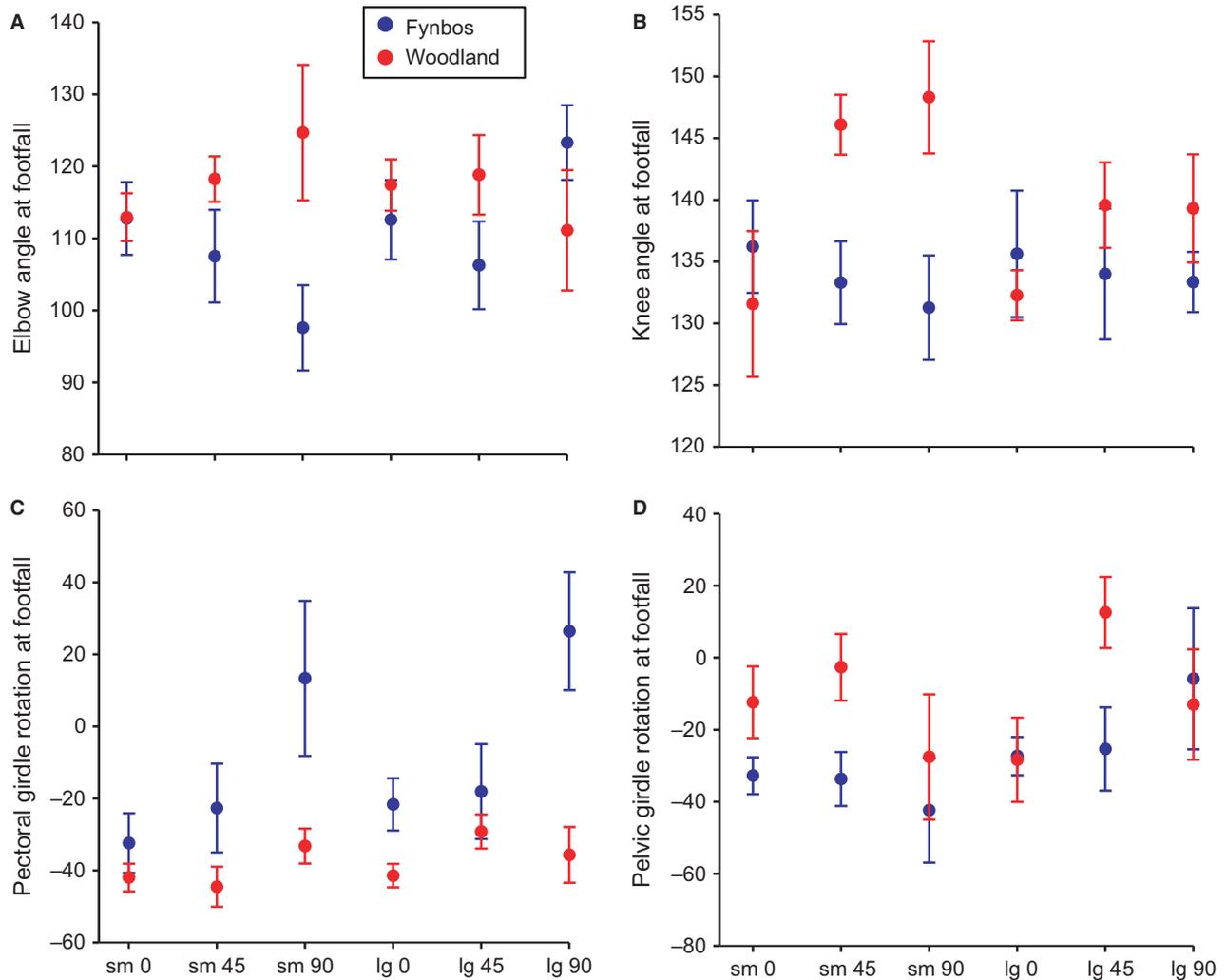


Figure 4. Mean values for elbow angle (A), knee angle (B), pectoral girdle rotation (C), and pelvic girdle rotation (D) at footfall for each treatment (sm, small diameter; lg, large diameter). The woodland morph is in red and the fynbos morph is in blue.

the narrower perches than the larger diameter perches (Fig. 3), possibly indicating differences in the ability to stabilize themselves on narrower perches. However, data regarding perch use during active behaviours are needed to determine whether the perch diameters we used are reflective of perch use. Finally, we found strong correlations within and between limbs, supporting our third hypothesis. Collectively, this supports the idea that functional differences can be driven by differences in habitat use, and probably precipitated a functional shift toward locomotion on narrow perches (Tolley *et al.*, 2006). The functional differences are probably not a result of phenotypic plasticity, given that common garden experiments have not detected morphological plasticity in this genus relating to habitat (Miller & Alexander, 2009). In combination with several other

studies of these morphs, we conclude that this system could provide an excellent resource for understanding how evolution shapes an animal's phenotype.

IMPACTS OF HABITAT STRUCTURE ON KINEMATICS

Although the impacts of incline on kinematics have been examined in a wide range of taxa (Irschick & Jayne, 1998; Spezzano & Jayne, 2004; Lammers, Earls & Biknevicus, 2006; Herrel *et al.*, 2013a; Birn-Jeffery & Higham, 2014a), the impacts of perch diameter are understood far less. From recent work on frogs and lizards, a key alteration that is made when moving on narrow perches is to lower the centre of mass (Cartmill, 1985; Schmitt, 1998; Foster & Higham, 2012, 2014; Herrel *et al.*, 2013a). This is

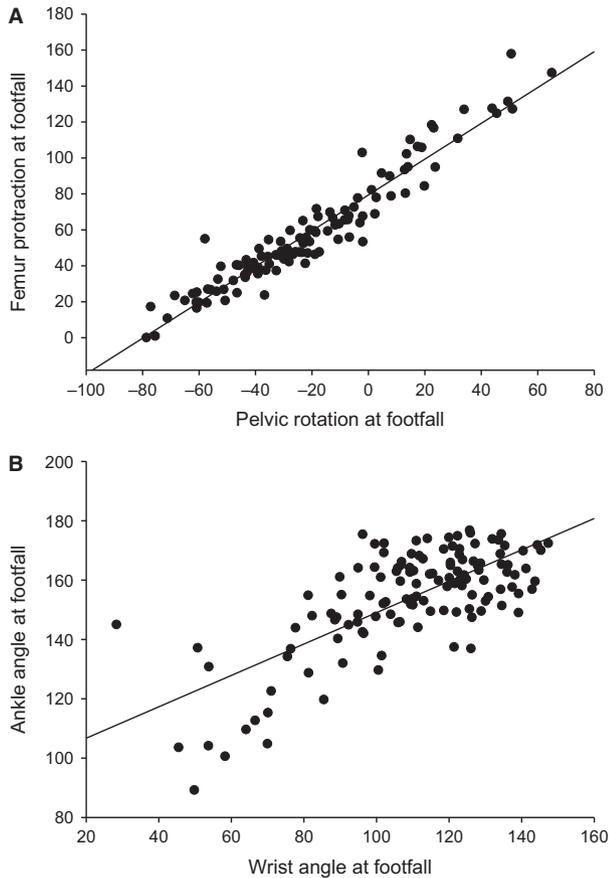


Figure 5. The integration of joint angles between and within limbs. (A) pelvic rotation versus femur protraction at footfall for all trials. The r^2 value was 0.91, and $P < 0.01$. (B) wrist angle versus ankle angle at footfall for all trials. Note that the wrist angle occurred prior to the ankle angle, and thus predicts the subsequent hindlimb angle at footfall. The r^2 value was 0.52, and $P < 0.05$.

demonstrated by increased joint flexion and femur elevation at mid-stance. For another species of chameleon, *C. calypttratus*, reduced hip height on narrow perches (compared to flat) was primarily driven by greater knee flexion at mid-stance (Higham & Jayne, 2004). In the present study, the fynbos morph had a more flexed knee at MS on the narrow level perch compared to the large level perch, although the femur and humerus were depressed to a greater extent (Table 1). This resulted in a greater hip height at MS on the narrow level perch, suggesting that the limbs might be constrained to being more upright on the narrow perch given that the base of support is closer to the animal's midline. Although raising the hip away from the substrate would typically decrease stability (by increasing the toppling moment), the incredible gripping ability of small chameleons (Herrel *et al.*, 2011), which includes the

limbs and tail, might ameliorate this destabilization. Overall, the impacts of habitat structure on stability are probably more prevalent for larger animals.

Chameleons generally move differently on narrow perches compared to broad surfaces (Higham & Jayne, 2004; Herrel *et al.*, 2013b). One of the key variables driving the kinematic differences for *C. calypttratus* when moving on a narrow perch (2.4 cm) compared to a flat surface (stationary treadmill) is the ankle angle at footfall, which is consistently greater on the narrow perch (Higham & Jayne, 2004). By contrast, we found no difference in the ankle angle at footfall between perch diameters within either morph. However, it is important to note that, although the relative lengths of the upper (femur) and lower (tibia) hindlimb segments are almost identical between *C. calypttratus* and *B. pumilium*, *C. calypttratus* is much larger (mean weight = 81 g) than the *B. pumilium* used in the present study (fynbos mean weight = 4.6 g; woodland mean weight = 6.6 g). Therefore, it is difficult to compare the effects of perch diameter without scaling this to body size. Given that chameleons do exhibit an incredible range of body size, this would be a fruitful avenue for future work.

Locomotor speed is often compared among and within species to determine the impacts of habitat structure on maximum performance (Losos, 1990). Speed was relatively constant for the fynbos morph among treatments, whereas the woodland morph exhibited a significant decrease when on the vertical treatments compared to the level. It is important to note that we did not attempt to induce maximum performance and so these differences must be interpreted with caution. The larger body mass of the woodland morph may have hindered forward progression on the vertical surfaces given the requirement for increasing the potential energy of the body with each stride. Future work should examine the impacts of perch diameter and incline on maximum running performance in chameleons aiming to determine any potential fitness consequences.

There are several possible explanations for the increased extension of the elbow and knee at FF (Fig. 4) on inclined narrow perches. First, this increased 'reach' could lead to a greater reliance on pulling the centre of mass up (as opposed to pushing it up), and this pulling can occur with both the hindlimbs and forelimbs (Higham & Jayne, 2004). Pulling might be beneficial for locomotor efficiency, maximizing muscle force production, preventing pitching of the body (and therefore maintaining stability) or increasing step length. Although future work is needed to determine the benefits and costs associated with pulling as a form of propulsion in these lizards, it is clear that the woodland morph might utilize pulling to a greater extent than the fynbos morph.

This again suggests that incline use differences in their habitat might be driving these changes in kinematics. However, this increase in pulling is evident on the narrow (and not the large) perch diameter, suggesting that the narrower perches are more suited for gripping. Reaching farther on a broader perch might be problematic if they cannot exert a sufficiently large gripping force. Given that grip strength increases with decreasing perch diameter in *Bradypodion* chameleons, narrow perches may be better for pulling (da Silva *et al.*, 2014), supporting our interpretation. Another explanation may be related to the fact that chameleons are extremely effective at bridging large gaps (T. E. Higham & G. J. Measey, pers. obs.), and this might be driving the increased extension in the woodland morph. In other words, they might frequently bridge gaps (more often than the fynbos morph), leading to increased joint extension on inclines. The fynbos morph probably bridges gaps by either moving on the ground between plants or by moving horizontally between relatively vertical perches within the vegetation. Thus, the increased extension in the woodland morph when moving up a vertical surface may also reflect the propensity to bridge vertical gaps, especially given that their available habitat is not as inclined as for the fynbos morph. In other words, morphs that live in a relatively horizontal habitat must bridge vertical gaps to ascend. Conversely, morphs that live in a relatively vertical habitat could simply climb up a perch to ascend.

ECOLOGICAL AND FUNCTIONAL DIVERGENCE BETWEEN MORPHS

Morphological, behavioural, and performance differences between species of lizards are widespread and are often linked to variation in habitat structure. Conversely, species can also converge on a similar morphology when occupying similar habitats. A stunning example of this is the *Anolis* ecomorphs, which exhibit comparable suites of morphological characters (and levels of performance) for a given microhabitat despite lack of a recent shared ancestry (Losos & Sinervo, 1989; Losos, 1990). However, convergence is not always the end result, and advantageous structures among species independently occupying comparable habitats do not always indicate adaptation (Revell *et al.*, 2007). Given this added complexity of evolutionary history, addressing habitat–morphology–biomechanics questions using an intraspecific framework provides many benefits. Indeed, several studies have directly linked habitat structure with intraspecific differences in morphology and performance. In populations of *Urosaurus ornatus*, for example, morphology depends strongly

on the structure of the habitat (Herrel *et al.*, 2001). Cliff populations exhibit a wide and flat body, with relatively short limb segments, whereas the boulder population has relatively long toes on the hindlimb, which enhance propulsion, and short proximal limb segments, and the arboreal population exhibited slender bodies, long tails, and long proximal limb segments. Similar work has been conducted on *B. pumilum*, and shows that morphological features associated with gripping (i.e. hand, foot, and tail size) differ, and these differences are associated with performance (Herrel *et al.*, 2011; 2013a, b). Despite these types of studies, little is known about the kinematic differences between populations of lizards or any group of vertebrate. In the present study, clear kinematic differences are correlated with habitat use and availability. For both the hindlimbs and forelimbs, the morphs of *B. pumilum* separate in multivariate kinematic space for each treatment (Fig. 3). The striking differences that we observed corroborate the results from several other studies that identified divergence in a variety of traits between these two morphs (Measey *et al.*, 2009, 2011; Herrel *et al.*, 2011; Hopkins & Tolley, 2011). When combined, it is clear that this is a potentially fruitful system not only for understanding how speciation occurs, but also the consequences and causes of phenotypic diversification. In addition, this framework can be extended to other morphs and species with the *Bradypodion* genus. For example, if a morph/species climbs vertical perches more often, or if they commonly bridge vertical gaps, more extension of the limbs at footfall should occur to maximize climbing efficiency and performance.

Incipient speciation is probably widespread but not always identifiable. One potential way of examining this is via population-level analyses (Stronen *et al.*, 2014). Although populations may differ simply by chance, phenotypic divergence can be a result of directional selection in response to environmental differences. One of the most demanding challenges for an organism is moving within complex three-dimensional habitats. Arboreal habitats are incredibly challenging, containing an array of perch diameters, inclines, and compliant structures, amongst others (Mattingly & Jayne, 2004). Thus, it is safe to say that selection pressures resulting from the demands of habitat structure are probably strong. The kinematic differences that we found between the two morphs *B. pumilum* provide support for this, given that they occupy and use significantly different habitats (Herrel *et al.*, 2011). A key question that remains is whether the morphs differ most in the treatments that are most demanding and/or those that most reflect a given morphs habitat. The Kogelberg site, where the fynbos morph is found, contains narrower and steeper

perches (mean = 1.35 mm and 80.8°, respectively) than the Stellenbosch sites (mean = 3.42 mm and 39.6°, respectively), where the woodland morph is found (Herrel *et al.*, 2011). Indeed, the narrower (3-mm experimental perch) 90° treatment appeared to pull the morphs further apart in multivariate space, and this was the case for both the forelimb and hindlimb (Fig. 3). Future studies should examine how this available habitat is used.

The differences between the morphs were more pronounced for certain conditions but not divergent for others. This highlights the fact that differences in kinematics (or biomechanics in general) depend on the conditions in which locomotion is compared. Divergence is probably more pronounced in extreme situations, or situations that are experienced in nature by one morph and not by the other. In the present study, the vertical condition appeared to separate the morphs in multivariate kinematic space to a greater degree than the level condition (Fig. 3), suggesting that these morphs may diverge in their use of inclines during locomotion under natural conditions. Given the increased demand associated with moving on inclines, specifically the increased work required to move the animal's centre of mass uphill (Birn-Jeffery & Higham, 2014a), it would be expected that this is where the functional differences would arise, especially if the incline use differs in the habitat. Although we found that the fynbos habitat, compared to the woodland habitat, consists of steeper inclines (80.8° versus 39.6°), it is currently unclear how these chameleons actually use their habitat when moving during the day. Given that the fynbos habitat is low to the ground, and that the plants are separated spatially, these morphs probably utilize the ground more often than the woodland morph. Thus, it might be possible that the fynbos morph occasionally uses shallower inclines relative to the available habitat quantified. However, the fynbos morph probably utilizes steeper inclines when above the ground, where these chameleons are expected to spend most of their time. Future work, however, is clearly needed to address these possibilities.

Perch diameter availability and sleeping perch use have been quantified, and differ between the morphs. The woodland morph sleeps on larger diameter (2.82 mm) perches than the fynbos morph (1.98 mm) (Herrel *et al.*, 2011). However, few data exist for daytime perch use, which is critical given that these chameleons are diurnal. Butler (2005) quantified foraging patterns of the woodland morph, and found that the mean daytime perch diameter used was 9 mm, which is the same diameter as the large perch used in the present study. However, additional data obtained by radio-tracking six

individuals of the same species suggests daytime perch diameters used by the woodland morph may be much smaller, averaging 4.5 mm, which is only slightly larger than the night-time perches used (3.9 mm; K. A. Tolley & E. Katz, unpubl. data). No comparable data exist for the fynbos morph. If the perches utilized in the day are larger than the sleep perches for the woodland morph, sleeping on a narrower perch may position the chameleon in a location that is hard to reach for a predator. The trade-off might be a sub-optimal perch diameter for locomotion. However, the lizard is stationary during the night, reducing the need for dynamic stability. Indeed, in a study of *Anolis* lizards, Singhal, Johnson & Ladner (2007) found that *A. valencienni* uses narrower branches for sleeping perches and larger diameter branches for diurnal use. Future work detailing the perch use (incline and diameter) during the day for these two chameleon morphs would be fruitful for understanding how animals use their habitat.

INTER AND INTRA-LIMB COORDINATION

Coordination between joints can occur over both temporal and spatial scales. Given that the forelimb will contact a new substrate before the hindlimb, some feedback is probably present that could modify hindlimb motion depending on what happens at the forelimb. We found striking evidence of this temporal coordination, with forelimb joint angles predicting subsequent hindlimb joint angles (Fig. 5B). This ability to predict a kinematic variable later in time highlights the importance of considering both sets of limbs when attempting to understand how animals adjust their motions in relation to ecology. Whether this coupling is necessarily present in a wide range of taxa is unclear, although we suggest that this is probably common.

A given kinematic motion may arise from the modulation of 'upstream' changes in kinematics, and could therefore be spatially coordinated. Understanding these links is critical for understanding how movements are actuated, as well as how morphology may translate into function. Intralimb coordination has been examined in both mammals and birds (Stoessel & Fischer, 2012) but rarely for lizards. We found that increased pelvic rotation at footfall was positively and tightly correlated with femur protraction at footfall (Fig. 5A). This may indicate that changes in femur protraction are simply a passive consequence of changes in girdle rotation. In any case, this increased femur protraction will result in a greater step length, potentially aiding in stability by providing a greater amount of time with the foot in contact with the substrate.

CONCLUSIONS

We have highlighted the considerable divergence in kinematics between two morphs of a single species of chameleon, and this divergence was especially acute on vertical surfaces; the morphs also exhibit different kinematic patterns on narrow perches within each incline (Fig. 3). Taken together, this suggests that these morphs are moving in very different ways, although additional information regarding mechanics and energetics would help determine the extent to which these movement differences are beneficial. This also highlights the potential for biomechanical studies to inform future ecological work (Aerts *et al.*, 2000). A similar idea was recently proposed by Foster & Higham (2014) regarding *Anolis* lizards. In this case, reduced variation in muscle activation patterns on a certain combination of incline and perch diameter suggested that green anoles (*Anolis carolinensis*) might prefer a specific habitat type. Future work that connects these links between biomechanics and behaviour will add to this integration between disparate fields of biology. The results from the present study can also be extended to other species of *Bradypodion*, given that they occupy a range of habitats in southern Africa (Tolley *et al.*, 2006; Measey, Raselimanana & Herrel, 2013; Tolley & Menegon, 2013). The differences between the morphs of *B. pumilum* can be used as hypotheses for differences in species occupying structurally diverse habitats. For example, some species of *Bradypodion* occur in grassland (Measey *et al.*, 2013), which probably comprises an extreme in terms of habitat structure. The range of habitats among species of *Bradypodion*, coupled with the differentiation into morphs within species, make them a model system for linking ecology and biomechanics.

ACKNOWLEDGEMENTS

We thank the South African National Biodiversity Institute for logistical support. This work was carried out under permits from CapeNature (0056-AAA007-00002 and AAA007-00035-0035) and under ethics clearance certificate (SANBI 003/2011). Our thanks are extended to a team of digitizers, including Kevin Dinh, Stephanie Valiente, Amy Cheu, Cindy Olivas, Angelyn Nepacena, Joseph Soquiati, Shayan Amiri, Amir Azamian, and Sophia Iribarren. Finally, we thank two anonymous reviewers for their very helpful comments. Funding for the present study included the National Research Foundation of South Africa-Key International Science Capacity Fund, start-up funds from the University of California,

Riverside to TEH, and the National Science Foundation (grant number IOS-1147043) to TEH.

REFERENCES

- Aerts P, Van Damme R, Vanhooydonck B, Zaaf A, Herrel A. 2000. Lizard locomotion: how morphology meets ecology. *Netherlands Journal of Zoology* **50**: 261–277.
- Bickel R, Losos JB. 2002. Patterns of morphological variation and correlates of habitat use in chameleons. *Biological Journal of the Linnean Society* **76**: 91–103.
- Birn-Jeffery AV, Higham TE. 2014a. The scaling of uphill and downhill locomotion in legged animals. *Integrative and Comparative Biology* **54**: 1159–1172.
- Birn-Jeffery AV, Higham TE. 2014b. Geckos significantly alter foot orientation to facilitate adhesion during downhill locomotion. *Biology Letters* **10**: 20140456.
- Brinsmead J, Fox MG. 2002. Morphological variation between lake- and stream-dwelling rock bass and pumpkinseed populations. *Journal of Fish Biology* **61**: 1619–1638.
- Butler MA. 2005. Foraging mode of the chameleon, *Bradypodion pumilum*: a challenge to the sit-and-wait versus active forager paradigm? *Biological Journal of the Linnean Society* **84**: 797–808.
- Carne L, Measey GJ. 2013. Chameleons on the cruise: seasonal differences in prey choice of two dwarf chameleons. *Herpetological Journal* **23**: 221–227.
- Cartmill M. 1985. Climbing. In: Hildebrand M, Bramble DM, Liem KF, Wake DB, eds. *Functional vertebrate morphology*. Cambridge, MA: Belknap Press, 73–88.
- Clemente CJ, Withers PC, Thompson G. 2013. Lizard tricks: overcoming conflicting requirements of speed vs climbing ability by altering biomechanics of the lizard stride. *Journal of Experimental Biology* **216**: 3854–3862.
- Collins CE, Russell AP, Higham TE. 2015. Subdigital adhesive pad morphology varies in relation to structural habitat use in the Namib Day Gecko, *Rhoptropus afer*. *Functional Ecology* **29**: 66–77.
- Fischer MS, Krause C, Lilje KE. 2010. Evolution of chameleon locomotion, or how to become arboreal as a reptile. *Zoology* **113**: 67–74.
- Foster KL, Higham TE. 2012. How fore- and hindlimb function changes with incline and perch diameter in the green anole (*Anolis carolinensis*). *Journal of Experimental Biology* **215**: 2288–2300.
- Foster KL, Higham TE. 2014. Context-dependent changes in motor control and kinematics during locomotion: modulation and decoupling. *Proceedings of the Royal Society of London Series B, Biological Sciences* **281**: 20133331.
- Fuller PO, Higham TE, Clark AJ. 2011. Posture, speed, and habitat structure: three-dimensional hindlimb kinematics of two species of padless gecko. *Zoology* **114**: 104–112.
- Garland T Jr, Losos JB. 1994. Ecological morphology of locomotor performance in squamate reptiles. In: Wainwright PC, Reilly SM, eds. *Ecological morphology: integrative organismal biology*. Chicago, IL: University of Chicago Press, 240–302.

- Ghalambor CK, Walker JA, Reznick DN. 2003.** Multi-trait selection, adaptation, and constraints on the evolution of burst swimming performance. *Integrative and Comparative Biology* **43**: 431–438.
- Gifford ME, Herrel A, Mahler DL. 2008.** The evolution of locomotor morphology, performance, and anti-predator behaviour among populations of *Leiocephalus* lizards from the Dominican Republic. *Biological Journal of the Linnean Society* **93**: 445–456.
- Hedrick TL. 2008.** Software techniques for two- and three-dimensional kinematic measurements of biological and biomimetic systems. *Bioinspiration & Biomimetics* **3**: 034001.
- Herrel A, Meyers JJ, Vanhooydonck B. 2001.** Correlations between habitat use and body shape in a phrynosomatid lizard (*Urosaurus ornatus*): a population-level analysis. *Biological Journal of the Linnean Society* **74**: 305–314.
- Herrel A, Measey GJ, Vanhooydonck B, Tolley KA. 2011.** Functional consequence of morphological differentiation between populations of the Cape Dwarf chameleon (*Bradypodion pumilum*). *Biological Journal of the Linnean Society* **104**: 692–700.
- Herrel A, Perrenoud M, Decamps T, Abdala V, Manzano A, Pouydebat E. 2013a.** The effect of substrate diameter and incline on locomotion in an arboreal frog. *Journal of Experimental Biology* **216**: 3599–3605.
- Herrel A, Tolley KA, Measey GJ, da Silva JM, Potgieter DF, Boller E, Boistel R, Vanhooydonck B. 2013b.** Slow but tenacious: an analysis of running and gripping performance in chameleons. *Journal of Experimental Biology* **216**: 1025–1030.
- Higham TE, Anderson CV. 2013.** Function and Adaptation in chameleons. In: Tolley KA, Herrel A, eds. *The biology of chameleons*. Berkeley, CA: University of California Press, 63–83.
- Higham TE, Jayne BC. 2004.** Locomotion of lizards on inclines and perches: hindlimb kinematics of an arboreal specialist and a terrestrial generalist. *Journal of Experimental Biology* **207**: 233–248.
- Higham TE, Birn-Jeffery AV, Collins CE, Hulseay CD, Russell AP. 2015.** Adaptive simplification and the evolution of gecko locomotion: morphological and biomechanical consequences of losing adhesion. *Proceedings of the National Academy of Sciences of the United States of America* **112**: 809–814.
- Hopkins KP, Tolley KA. 2011.** Morphological variation in the Cape Dwarf Chameleon (*Bradypodion pumilum*) as a consequence of spatially explicit habitat structure differences. *Biological Journal of the Linnean Society* **102**: 878–888.
- Irschick DJ, Jayne BC. 1998.** Effects of incline on speed, acceleration, body posture and hindlimb kinematics in two species of lizard *Callisaurus draconoides* and *Uma scoparia*. *Journal of Experimental Biology* **201**: 273–287.
- Kerfoot JRJ, Schaefer JF. 2006.** Ecomorphology and habitat utilization of *Cottus* species. *Environmental Biology of Fishes* **76**: 1–13.
- Krause C, Fischer MS. 2013.** Biodynamics of climbing: effects of substrate orientation on the locomotion of a highly arboreal lizard (*Chamaeleo calyptrotus*). *Journal of Experimental Biology* **216**: 1448–1457.
- Lammers AR, Earls KD, Biknevicus AR. 2006.** Locomotor kinetics and kinematics on inclines and declines in the gray short-tailed opossum *Monodelphis domestica*. *Journal of Experimental Biology* **209**: 4154–4166.
- Lavin PA, McPhail JD. 1986.** Adaptive divergence of trophic phenotype among freshwater populations of the three-spine stickleback (*Gasterosteus aculeatus*). *Canadian Journal of Fisheries and Aquatic Science* **43**: 2455–2463.
- Logan ML, Montgomery CE, Boback SM, Reed RN, Campbell JA. 2012.** Divergence in morphology, but not habitat use, despite low genetic differentiation among insular populations of the lizard *Anolis lemurinus* in Honduras. *Journal of Tropical Ecology* **28**: 215–222.
- Losos JB. 1990.** Ecomorphology, performance capability, and scaling of West Indian *Anolis* lizards: an evolutionary analysis. *Ecological Monographs* **60**: 369–388.
- Losos JB, Sinervo B. 1989.** The effects of morphology and perch diameter on sprint performance of *Anolis* lizards. *Journal of Experimental Biology* **145**: 23–30.
- Macrini TE, Irschick DJ. 1998.** An intraspecific analysis of trade-offs in sprinting performance in a West Indian lizard species (*Anolis lineatopus*). *Biological Journal of the Linnean Society* **63**: 579–591.
- Malhotra A, Thorpe RS. 1997.** Size and shape variation in a Lesser Antillean anole, *Anolis oculatus* (Sauria: Iguanidae) in relation to habitat. *Biological Journal of the Linnean Society* **60**: 53–72.
- Mattingly WB, Jayne BC. 2004.** Resource use in arboreal habitats: structure affects locomotion of four ecomorphs of *Anolis* lizards. *Ecology* **85**: 1111–1124.
- Measey GJ, Hopkins KP, Tolley KA. 2009.** Morphology, ornaments and performance in two chameleon ecomorphs: is the casque bigger than the bite? *Zoology* **112**: 217–226.
- Measey GJ, Rebelo AD, Herrel A, Vanhooydonck B, Tolley KA. 2011.** Diet, morphology and performance in two chameleon morphs: do harder bites equate with harder prey? *Journal of Zoology* **285**: 247–255.
- Measey GJ, Raselimanana AP, Herrel A. 2013.** Ecology and Life History of Chameleons. In: Tolley KA, Herrel A, eds. *The biology of chameleons*. Berkeley, CA: University of California Press, 85–113.
- Miller AK, Alexander GJ. 2009.** Do dwarf chameleons (*Bradypodion*) show developmental plasticity? *African Zoology* **44**: 45–54.
- Peterson JA. 1984.** The locomotion of *Chamaeleo* (Reptilia: Sauria) with particular reference to the forelimb. *Journal of Zoology* **202**: 1–42.
- Resinger WJ, Stuart-Fox DM, Erasmus BF. 2006.** Habitat associations and conservation status of an endemic forest dwarf chameleon (*Bradypodion* sp.) from South Africa. *Oryx* **40**: 183–188.
- Revell LJ, Johnson MA, Schulte JA, Kolbe JJ, Losos JB. 2007.** A phylogenetic test for adaptive convergence in rock-dwelling lizards. *Evolution* **61**, 2898–2912.
- Schmitt D. 1998.** Forelimb mechanics during arboreal and terrestrial quadrupedalism in Old World monkeys. In:

- Strasser E, Fleagle J, Rosenberger A, McHenry H, eds. *Primate locomotion: recent advances*. New York, NY: Plenum Press, 175–200.
- da Silva JM, Tolley KA. 2013.** Ecomorphological variation and sexual dimorphism in a recent radiation of dwarf chameleons (*Bradypodion*). *Biological Journal of the Linnean Society* **109**: 113–130.
- da Silva JM, Herrel A, Measey GJ, Vanhooydonck B, Tolley KA. 2014.** Linking microhabitat structure, morphology and locomotor performance traits in a recent radiation of dwarf chameleons. *Functional Ecology* **28**: 702–713.
- Sinervo B, Huey RB. 1990.** Allometric engineering: an experimental test of the causes of interpopulational differences in performance. *Science* **248**: 1106–1109.
- Sinervo B, Losos JB. 1991.** Walking the tight rope: arboreal sprint performance among *Sceloporus occidentalis* lizard populations. *Ecology* **72**: 1225–1233.
- Singhal S, Johnson MA, Ladner JT. 2007.** The behavioral ecology of sleep: natural sleeping site choice in three *Anolis* lizard species. *Behaviour* **144**: 1033–1052.
- Spezzano LC Jr, Jayne BC. 2004.** The effects of surface diameter and incline on the hindlimb kinematics of an arboreal lizard (*Anolis sagrei*). *Journal of Experimental Biology* **207**: 2115–2131.
- Stoessel A, Fischer MS. 2012.** Comparative intralimb coordination in avian bipedal locomotion. *Journal of Experimental Biology* **215**: 4055–4069.
- Stronon AV, Navid EL, Quinn MS, Paquet PC, Bryan HM, Darimont CT. 2014.** Population genetic structure of gray wolves (*Canis lupus*) in a marine archipelago suggests island-mainland differentiation consistent with dietary niche. *BMC Evolutionary Biology* **14**: 11.
- Stuart-Fox D. 2009.** A test of Rensch's Rule in dwarf chameleons (*Bradypodion* sp.), a group with female-biased sexual size dimorphism. *Evolutionary Ecology* **23**: 425–433.
- Tolley KA, Menegon M. 2013.** Evolution and biogeography of chameleons. In: Tolley KA, Herrel A, eds. *The biology of chameleons*. Berkeley, CA: University of California Press, 131–150.
- Tolley KA, Burger M, Turner AA, Matthee CA. 2006.** Biogeographic patterns and phylogeography of dwarf chameleons (*Bradypodion*) in an African biodiversity hotspot. *Molecular Ecology* **15**: 781–793.
- Van Damme R, Aerts P, Vanhooydonck B. 1998.** Variation in morphology, gait characteristics and speed of locomotion in two populations of lizards. *Biological Journal of the Linnean Society* **63**: 409–427.
- Zaaf A, Herrel A, Aerts P, De Vree F. 1999.** Morphology and morphometrics of the appendicular musculature in geckoes with different locomotor habits (Lepidosauria). *Zoology* **119**: 9–22.
- Zaaf A, Van Damme R, Herrel A, Aerts P. 2001.** Spatio-temporal gait characteristics of level and vertical locomotion in a ground-dwelling and a climbing gecko. *Journal of Experimental Biology* **204**: 1233–1246.
- Zar JH. 1996.** *Biostatistical analysis*. Upper Saddle River, New Jersey: Prentice Hall.