

Are Morphological Specializations of the Hyolingual System in Chameleons and Salamanders Tuned to Demands on Performance?*

Anthony Herrel^{1,†}
 Stephen M. Deban²
 Vicky Schaerlaeken¹
 Jean-Pierre Timmermans³
 D. Adriaens⁴

¹Department of Biology, University of Antwerp, Universiteitsplein 1, B-2610 Antwerp, Belgium; ²Department of Biology, SCA 110, 4202 East Fowler Avenue, University of South Florida, Tampa, Florida 33620; ³Department of Veterinary Sciences, University of Antwerp, Groenenborgerlaan 171, B-2020 Antwerp, Belgium; ⁴Department of Biology, Ghent University, K.L. Ledeganckstraat 35, B-9000 Ghent, Belgium

Accepted 5/8/2008; Electronically Published 11/13/2008

ABSTRACT

Extremely specialized and long tongues used for prey capture have evolved independently in plethodontid salamanders and chameleons. In both systems, the demands on tongue projection are probably similar: to maximize projection velocity and distance. Consequently, many of the design features of the projection system in these two groups have converged to an astonishing degree. Both involve the use of power amplification systems based on helically wound muscle fibers that load internal connective tissue sheets as illustrated in previous studies. Demands imposed on tongue retraction, however, are different to some degree. Although in both groups there is a clear demand for retraction capacity (given the long projection distances), in chameleons there is an added demand for force because they eat large and heavy prey. As indicated by our data, plethodontid salamanders have extremely long tongue retractors with normal striated muscle. Chameleons, on the other hand, evolved long retractors of the supercontracting type. Interestingly, our data show that at least in chameleons, the extreme design of the tongue in function of prey capture appears to have consequences on prey transport, resulting in an increased dependence on the hyoid. In turn, this has led to an increase in transport-

cycle duration and an increase in the number of cycles needed to transport prey in comparison with closely related agamid lizards. Clearly, extreme morphological specializations are tuned to functional and ecological demands and may induce a reduced performance in other functions performed by the same set of integrated structures.

Introduction

Morphological specializations are often assumed to be the result of natural selection on morphology in response to specific functional or ecological demands. For example, the long beaks or snouts of nectar-eating birds and bats are thought to be the result of coevolution between plants and their pollinators (Muchhala 2007a, 2007b). Animals with longer beaks and snouts will have a performance advantage because they are able to sample a wider range of flowers, including those with the deepest corollas (Fleming et al. 2005; Muchhala 2006). Additionally, many bats, birds, and bumblebees have developed long tongues that allow them to reach into the deepest flowers without needing extremely elongated heads (Muchhala 2006).

The vertebrate tongue is an organ that is crucial to a wide variety of functions, including important roles in prey transport and swallowing, drinking, breathing, and even chemoreception and prey capture in some groups, such as lizards (Bels et al. 1994; Schwenk 2000; Herrel et al. 2001c). Thus, specialization of the tongue may be constrained by functional trade-offs imposed by the different functions. For example, specialization of the tongue for chemoreceptive purposes in many lizards and snakes constrains its use in prey capture and transport, and thus alternative capture (i.e., jaw prehension) and transport (e.g., inertial transport) strategies have been developed (Schwenk 2000). In other, animals such as anteaters (Redford 1985), many frogs (Nishikawa 1999, 2000), chameleons (Wainwright et al. 1991; Wainwright and Bennett 1992a, 1992b; Herrel et al. 2000), and plethodontid salamanders (Deban 1997; Deban et al. 1997; Wake and Deban 2000), the tongue has evolved into a highly specialized prey-capture organ.

If the tongue is used for prey capture, the functional demands imposed on it are clearly dependent on the distance of protrusion/projection and the size of prey that are captured. Tongue-extension length is probably relevant to predators because the further the tongue can be extended, the greater the number of prey that come within reach of the tongue without the need of predator movement, a feature especially crucial for cryptic sit-and-wait predators. Generally, two types of tongue extension are possible in vertebrates. Most commonly, the

* This paper was a contribution to the symposium "Functional Consequences of Extreme Adaptations of the Trophic Apparatus in Craniates," organized by Dominique Adriaens and Anthony Herrel, at the Eighth International Congress on Vertebrate Morphology, Paris, France, 2007.

[†]Corresponding author; e-mail: anthony.herrel@ua.ac.be.

tongue functions as a muscular hydrostat. This mechanism depends on the active contraction of intrinsic muscles (Kier and Smith 1985; Smith 1988; Smith and Kier 1989). Although such a system has the advantage of being accurate because it allows for online modulation, it is also slow, and its extension is directly limited by muscle shortening (Nishikawa et al. 1999; Nishikawa 2000; Meyers et al. 2004). Alternatively, the tongue can be projected from the mouth and then lengthen under its own inertia. In ballistic systems, tongue extension is rapid but does not allow for modulation of the trajectory after release from the mouth. Tongue-extension length in such ballistic systems is probably limited by muscle stretch and the presence of connective tissue in the tongue (Peters and Nishikawa 1999; Nishikawa 2000). Additionally, tongue extension will be determined by the “takeoff” velocity of the tongue from the mouth (Hill 1950; Marsh 1994; Aerts 1998). Higher takeoff velocities are, in turn, dependent on the acceleration that can be imparted to the tongue, which is dictated by the ability of the musculoskeletal system to generate mechanical power (Lutz and Rome 1994; Aerts 1998; Askew and Marsh 2002; de Groot and van Leeuwen 2004; Lappin et al. 2006; Deban et al. 2007).

However, if the tongue is projected from the mouth to a great length, this will also impose demands on the tongue-retraction system. Muscle shortening of typical cross-striated vertebrate muscle is limited to 50% of its resting length (Huxley 1965; Gordon et al. 1966). Thus, if ballistic projection involves extreme lengthening of the tongue as observed in chameleons, frogs, and plethodontid salamanders (Wainwright et al. 1991; Nishikawa 2000; Wake and Deban 2000), then the retractor muscles will probably have to be very long. This may, however, limit the functional capacity of the tongue retractor when the tongue is used during prey transport and swallowing (Herrel et al. 2001a, 2002). Near either end of its working range, the tongue retractor is faced with a decrease in its force-generating capacity because the muscle can be expected to be operating on the ascending and/or descending limb of its length-tension curve (i.e., decreased overlap between thick and thin filaments; Burkholder and Lieber 2001; Rome and Lindstedt 1997).

Here, we review the design of the tongue in two groups of vertebrates that have independently evolved extreme tongue-projection and prey-capture behaviors: plethodontid salamanders and chameleons (Deban et al. 1997; Herrel et al. 2000, 2001b, 2002). In doing so we aim to (1) explore whether functional demands imposed by the ecology and lifestyle of these animals have resulted in similar morphologies in these phylogenetically distantly related taxa and (2) investigate whether morphological specialization for extreme functional demands has limited the capacity of the tongue to function in other behaviors. In addition to reviewing the literature, we provide new data on the ultrastructure of the tongue retractors in *Hydromantes* and previously unpublished data on the use of the hyoid during prey transport in an agamid lizard (*Pogona vitticeps*) with a generalized tongue morphology compared with chameleons (*Chamaeleo calypttratus* and *Furcifer oustaleti*).

Material and Methods

Histology

The morphology of the hyobranchial system of *Hydromantes italicus*, *Salamandra salamandra*, *Laudakia stellio*, and *Chamaeleo calypttratus* was studied by serial cross sections. *Laudakia stellio* (an agamid lizard) and *S. salamandra* (a nonplethodontid salamander) were chosen for comparison because they are both nonspecialized but use their tongues for prey capture. Specimens were killed by an overdose of anesthetic (MS222 for the salamanders, Ketamine for the lizards), fixed in an 8% buffered formaldehyde solution, decalcified with Decalc 25% (HistoLab), dehydrated through an alcohol series, and embedded in Technovit 7100 (Kulzer-Heraeus; *C. calypttratus* and *H. italicus*) or paraffin (*L. stellio*, *S. salamandra*). Series of semithin sections (2–5 μm) were cut using a Leica Polycut SM 2500, stained with a nonspecific toluidine blue (*C. calypttratus*) or tissue specific modified trichrome stain (*S. salamandra*, *L. stellio*, and *H. italicus*), and mounted with DPX. Images of the sections were obtained using a digital camera (Colorview 8, Soft Imaging System) mounted on a Polyvar-Reichert light microscope and processed with Analysis Docu (Soft Imaging System, ver. 3.0).

Transmission Electron Microscopy

For transmission electron microscopy (TEM), the tongue retractors of an adult *H. italicus* (snout-vent length [SVL] 50 mm) were removed unilaterally. Tissue samples were removed from both the anterior and posterior parts of the muscle. Several small blocks of tissue taken from the middle part of the muscle were cut from the samples and thoroughly washed three times for 10 min in 0.1 mol L⁻¹ sodium cacodylate buffer containing 7.5% saccharose and 0.05% calcium chloride. Next, the tissue was postfixed for 2 h at 4°C in 1% osmium tetroxide in 0.033 mol L⁻¹ veronalacetate buffer containing 4% saccharose (pH 7.4). After postfixation, samples were rinsed three times for 10 min in 0.05 mol L⁻¹ veronalacetate buffer containing 6% saccharose sodium cacodylate buffer (pH 7.4) and dehydrated through a graded series of ethanols. The samples were then embedded in Durcupan. Suitable longitudinal areas and cross sections of the striated muscle were selected on toluidine blue-stained semithin sections. Subsequently, ultrathin sections were stained with lead citrate and uranyl acetate and examined in a Philips CM10 electron microscope. Photographs were taken of all samples at different locations for several sections within each sample. Pictures were taken of both anteriorly and posteriorly located samples. The sarcomere length, filament lengths (thick and thin), and I-band thickness were measured for 30 sarcomeres using TPSdig software (Rohlf 2004).

Kinematics and Feeding Behavior

Lizards (*Pogona vitticeps*, $N = 2$; *C. calypttratus*, $N = 1$; *Furcifer oustaleti*, $N = 2$) used for feeding trials were obtained from a commercial dealer and transferred to the lab at the University

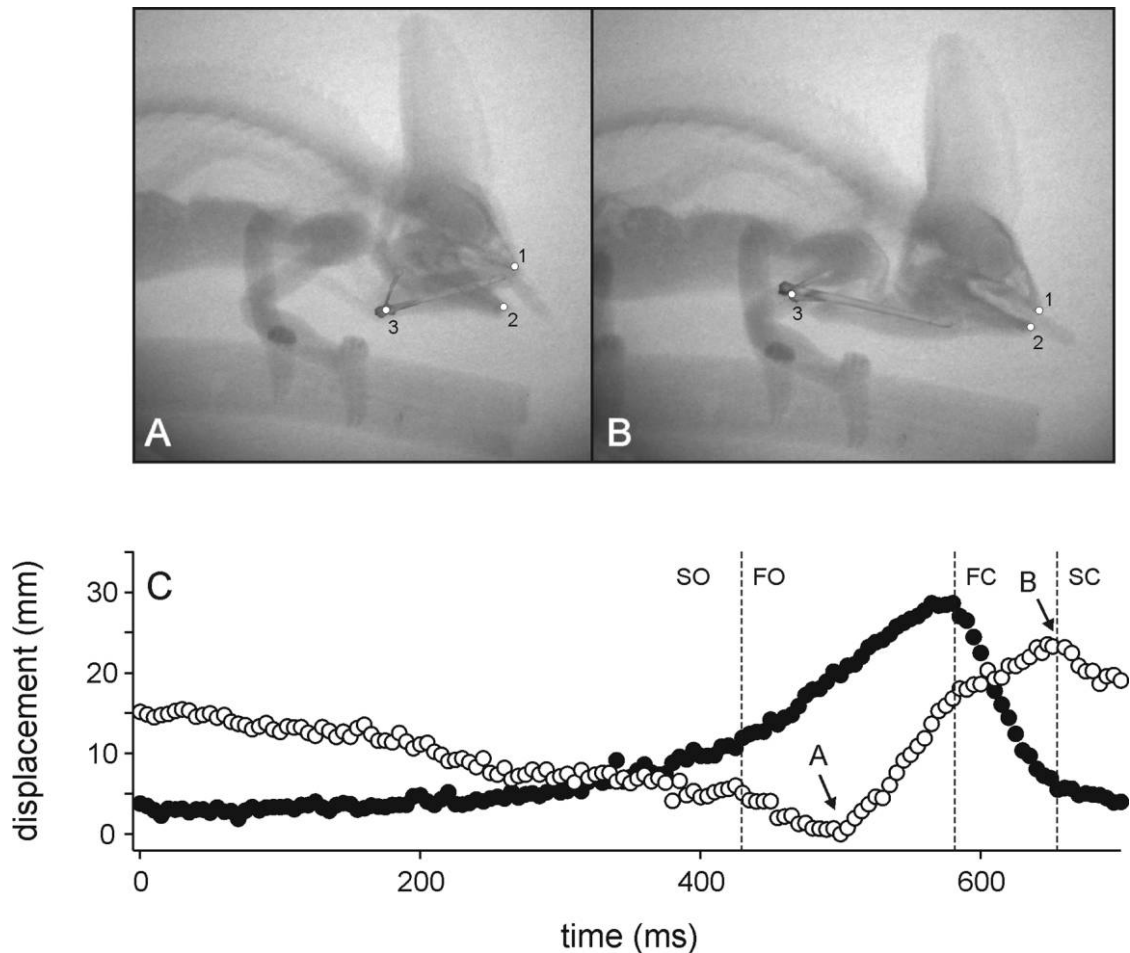


Figure 1. *A*, Picture of the hyoid in *Camaleo calyptratus* in its most protracted position achieved during prey transport superimposed on a frame of an x-ray recording. *B*, The hyoid in its most retracted position. *C*, Displacement profiles of the jaws (filled circles) and the hyoid (open circles) during the transport of a large grasshopper in *C. calyptratus*. Arrows indicate the positions of points where the hyoid is in its most protracted (*A*) and retracted (*B*) position. White circles in *A* and *B* indicate the landmarks digitized on each frame: 1, tip of upper jaw; 2, tip of lower jaw; 3, base of the ceratobranchial. SO, slow opening phase; FO, fast opening phase; FC, fast closing phase; SC, slow closing phase.

of Antwerp. Animals were of similar size, the chameleons having an average lower-jaw length of 36.96 ± 1.25 mm and the *Pogona* having an average lower-jaw length of 34.96 ± 1.37 mm. The animals were kept in a vivarium on a 12L : 12D cycle and were offered crickets, grasshoppers, and waxworms ad lib. The environmental temperature varied from 34°C during day-time to 22°C at night. An incandescent bulb provided the animals with a basking place at higher temperature.

Video and Cineradiographic Recordings

Pogona vitticeps and *C. calyptratus* were filmed in lateral view at 25 Hz using a handheld digital camera (Sony DCR-HC22E). The animals were offered large grasshoppers (4 cm). Simultaneously, high-speed x-ray videos were recorded using a Philips Optimus x-ray generator coupled to a 14-inch image intensifier with two zoom modes (10 and 6 inch) and a Redlake Motion Pro 2000 camera (1,248 × 1,024 pixels). Recordings of prey transport were made in lateral view using the 6-inch zoom

function at 250 frames per second for *P. vitticeps* and 200 frames per second for *C. calyptratus*. All experiments were approved by the Animal Care and Use Committee at the University of Antwerp.

Analyses

Video recordings were reviewed using Midas Player software (Redlake, San Diego, CA, ver. 2.1.7). Only feeding sequences in which all the transport cycles of one feeding sequence were present were retained for further analysis. Based on these recordings, the number of transport cycles needed to transport a grasshopper and the average duration of a transport cycle were calculated. Cineradiographic recordings were reviewed using Midas Player software (ver. 2.1.7). Only feeding sequences in which all phases of a transport cycle (SO, slow opening; FO, fast opening; FC, fast closing; SC, slow closing; see Fig. 1) were present were used in further analysis. Three landmarks were digitized using Didge (Image Digitizing Software, Alistair Cul-

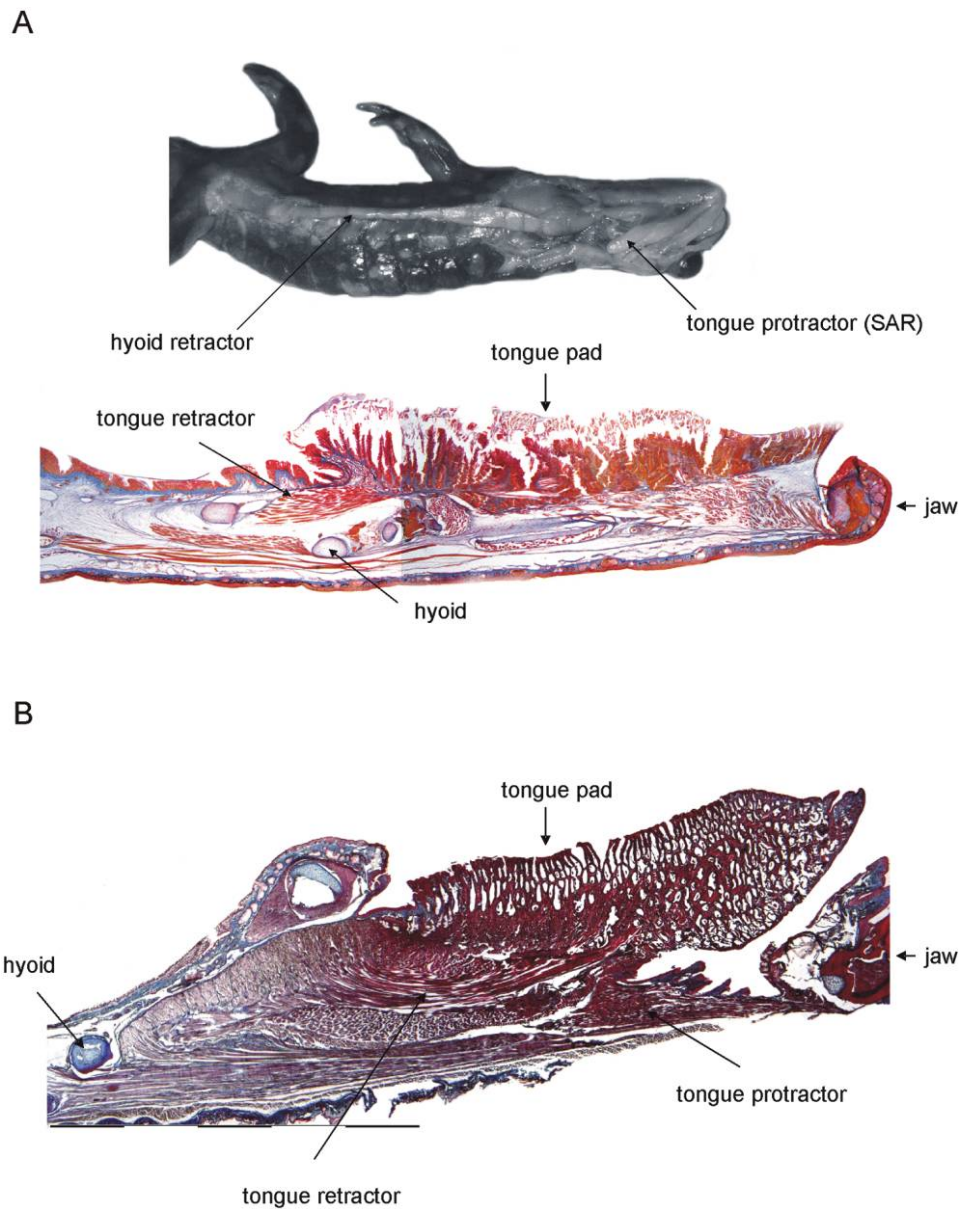


Figure 2. A, Top, ventrolateral view on the hyolingual system in *Salamandra salamandra*; bottom, sagittal section through the tongue of an *S. salamandra*. B, sagittal section through the tongue of *Laudakia stellio* (modified after Herrel et al. 2005). In the stains, muscle and bone are colored red/orange, and connective tissue and cartilage are colored blue.

lum, ver. 2.2.0). These landmarks included the anterior tip of the upper and lower jaw and base of the first ceratobranchial (Fig. 1). Based on x - y coordinates of these markers, gape distance (distance between the jaw tips) and the displacement of the hyoid relative to the jaw during prey transport were calculated. Sixty intraoral transport cycles from seven different feeding sequences were analyzed for *C. calyptatus*, and 27 cycles from three feeding sequences for the first individual and 20 cycles from four feeding sequences for the second individual of *F. oustaleti* were analyzed. A total of 16 intraoral transport cycles from five complete feeding sequences were analyzed for two *P. vitticeps*.

Nested ANOVA with individual nested within group (agamids vs. chameleons) were used to test for differences in prey-transport kinematics between *Pogona* and the chameleons used in this study. Note that cycle-to-cycle and among-sequence variation is incorporated in the total individual variation and was not explicitly introduced as a factor in our analyses.

Results and Discussion

Salamanders

The tongue skeleton of terrestrial salamanders is composed of a series of articulating, mostly cartilaginous elements (Wake

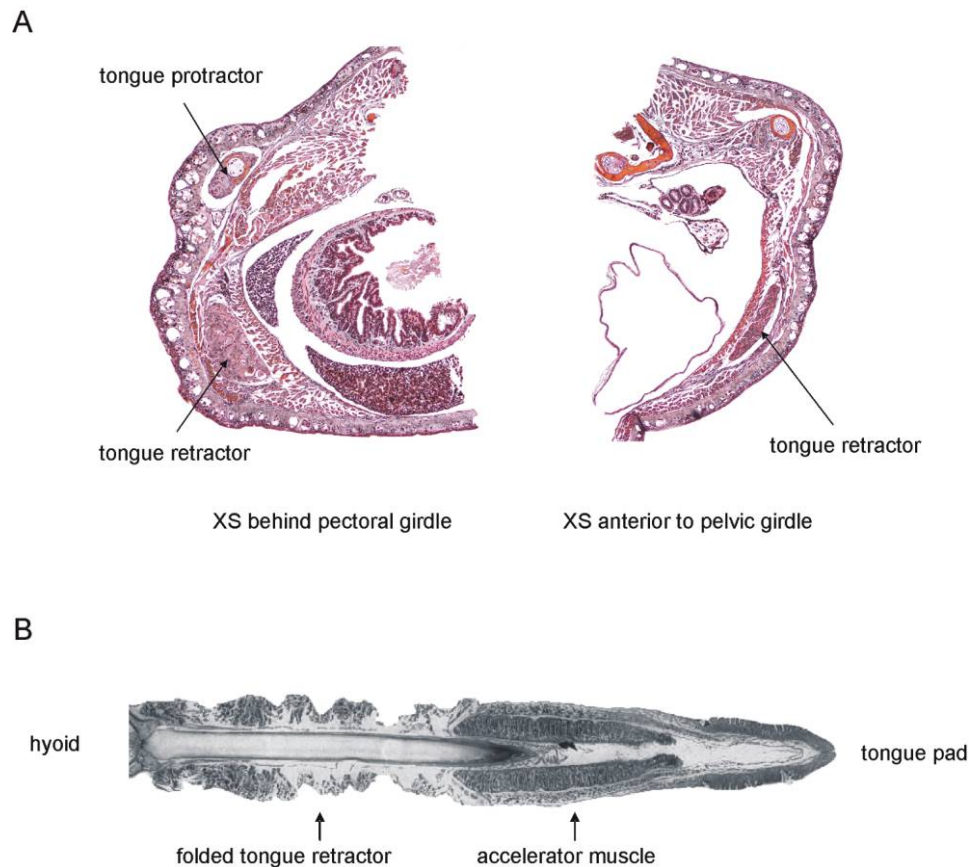


Figure 3. *A*, Left, cross section through the body (one half only) just posterior to the pectoral girdle in *Hydromantes*; right, cross section through the body (one half only) just anterior to the pectoral girdle in *Hydromantes*. Bone is colored orange, and muscle is colored pink. *B*, Frontal section through the hyolingual apparatus of *Chameleo jacksonii* (modified after Herrel et al. 2001a).

and Deban 2000). The basibranchial lies medially and provides support for the tongue pad. On each side, a first and second ceratobranchial articulate with the basibranchial, and an epibranchial articulates with the caudal ends of the first and second ceratobranchials (Lombard and Wake 1977; Wake and Deban 2000; Deban and Dicke 2004). The epibranchial in *Hydromantes* is greatly elongated in comparison with that of *Salamandra* (Lombard and Wake 1977; Wake and Deban 2000). During tongue protrusion/projection, the tongue skeleton folds medially as it is pulled and pushed forward. Whereas in *Hydromantes*, the tongue skeleton is free and can be projected completely from the mouth, in *Salamandra*, the posterior part of the tongue skeleton does not leave the mouth.

The m. subarcualis rectus is the primary tongue-protractor muscle (Figs. 2, 3). It originates broadly along the edge of the ceratohyal and runs posteriorly to the epibranchial. The rostral portion has a fiber orientation that can pull the tongue skeleton forward. The caudal portion forms an elongated sheath around the epibranchial with a helically wound segmented arrangement of short muscle fibers and is active on average 107–117 ms before tongue projection in the plethodontids *Bolitoglossa do-fleini* and *Hydromantes imperialis* (Deban and Dicke 2004; Deban et al. 2007; Fig. 4). In *Salamandra*, the m. subarcualis rectus

does not form a sheath around the epibranchial but rather inserts along the posteroventral third of it. During tongue protrusion in *Salamandra*, the m. subarcualis rectus is active and pulls the epibranchial forward (Dockx and De Vree 1986; F. De Vree, personal communication). In *Hydromantes*, connective tissue sheets are present within the m. subarcualis rectus posterior and between the muscle and the epibranchial, which are presumed to be deformed on contraction of the muscle and may thus have the potential to store elastic strain energy (Deban et al. 2007; Fig. 4). Tongue retraction in salamanders is accomplished by the m. rectus cervicis posterior, which is a long straplike muscle that originates on the pelvis and inserts into the tongue pad (Deban and Dicke 1999; Figs. 2, 3). In *Hydromantes* species, a portion of the m. rectus cervicis posterior lies in a loop in the throat just in front of the heart when the tongue is at rest (Lombard and Wake 1977; Deban et al. 1997). In both *Salamandra* and *Hydromantes*, the muscle is activated before prey contact and remains active throughout tongue retraction (Dockx and De Vree 1986; Deban and Dicke 1999; F. De Vree, personal communication).

TEM data of the m. rectus cervicis collected specifically for this study show that the muscle is composed of typical cross-striated vertebrate muscle (Fig. 5). Neither anterior nor pos-

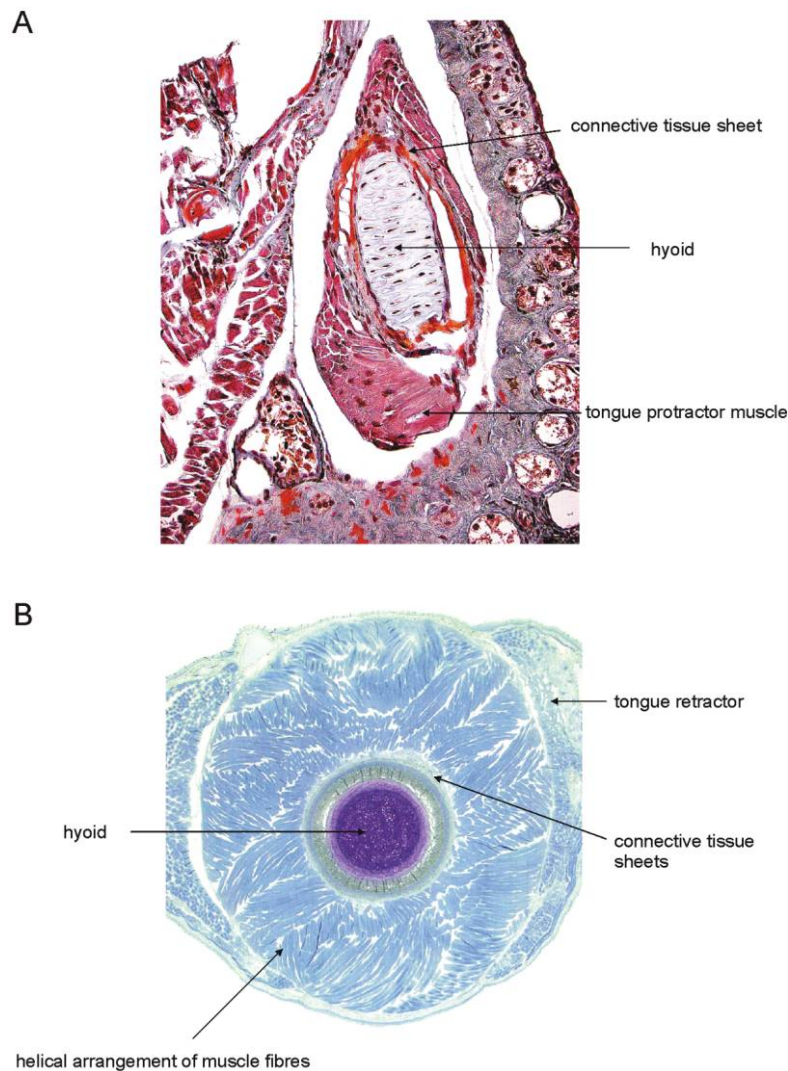


Figure 4. A, Cross section through the hyolingual apparatus in *Hydromantes italicicus* at the level of the pectoral girdle. Note the cartilagenous epibranchial (blue) surrounded by connective tissue (orange) and muscle (pink). B, Cross section through the hyolingual system in *Chameleo calyptratus* at mid tongue. Note the dense centrally positioned cartilage of the entoglossal process surrounded by connective tissue sheets and the helically wound accelerator muscle.

terior samples show supercontracting muscle fibers. Sarcomere lengths are slightly shorter than typical vertebrate sarcomeres ($1.74 \pm 0.06 \mu\text{m}$; Table 1) with fairly typical I bands ($0.33 \pm 0.06 \mu\text{m}$). Filament lengths also do not deviate greatly from the typical vertebrate pattern (thick: $1.41 \pm 0.02 \mu\text{m}$; thin: $0.81 \pm 0.03 \mu\text{m}$). Interestingly, the length of the tongue retractor of *Hydromantes* is slightly longer than the tongue-protrusion length (80% of body length; see Table 1), suggesting that it can indeed operate using a typical vertebrate skeletal-muscle sarcomere structure contracting to approximately 50% of resting length. However, near full extension and retraction, the force output of the muscle is probably reduced.

Lizards

The tongue skeleton of the lizards examined here consists of a centrally positioned entoglossal process and three pairs of ar-

ticulating elements in *Pogona* but only two pairs in *Chameleo*. Chameleons differ from agamid lizards by the presence of an elongated and parallel-sided entoglossal process, the absence of the second pair of ceratobranchials, a reduction of the length of the ceratohyals, and an anteriodorsad rotation of the first pair of ceratobranchials (see Herrel et al. 2001b). During tongue protrusion, the tongue skeleton is protracted, and the ceratobranchials are folded backward by the combined action of the mandibulohyoideus and sternohyoideus muscles (Wainwright and Bennett 1992a, 1992b; Meyers and Nishikawa 2000). In agamid lizards, the hyoid is also protracted, but to a lesser degree (Schwenk 2000). Neither in chameleons nor in agamids does the tongue skeleton leave the mouth entirely on tongue protrusion/projection.

The main tongue-protractor muscles in agamid lizards consist of the paired lateral, medial, and internal mm. genioglossi,

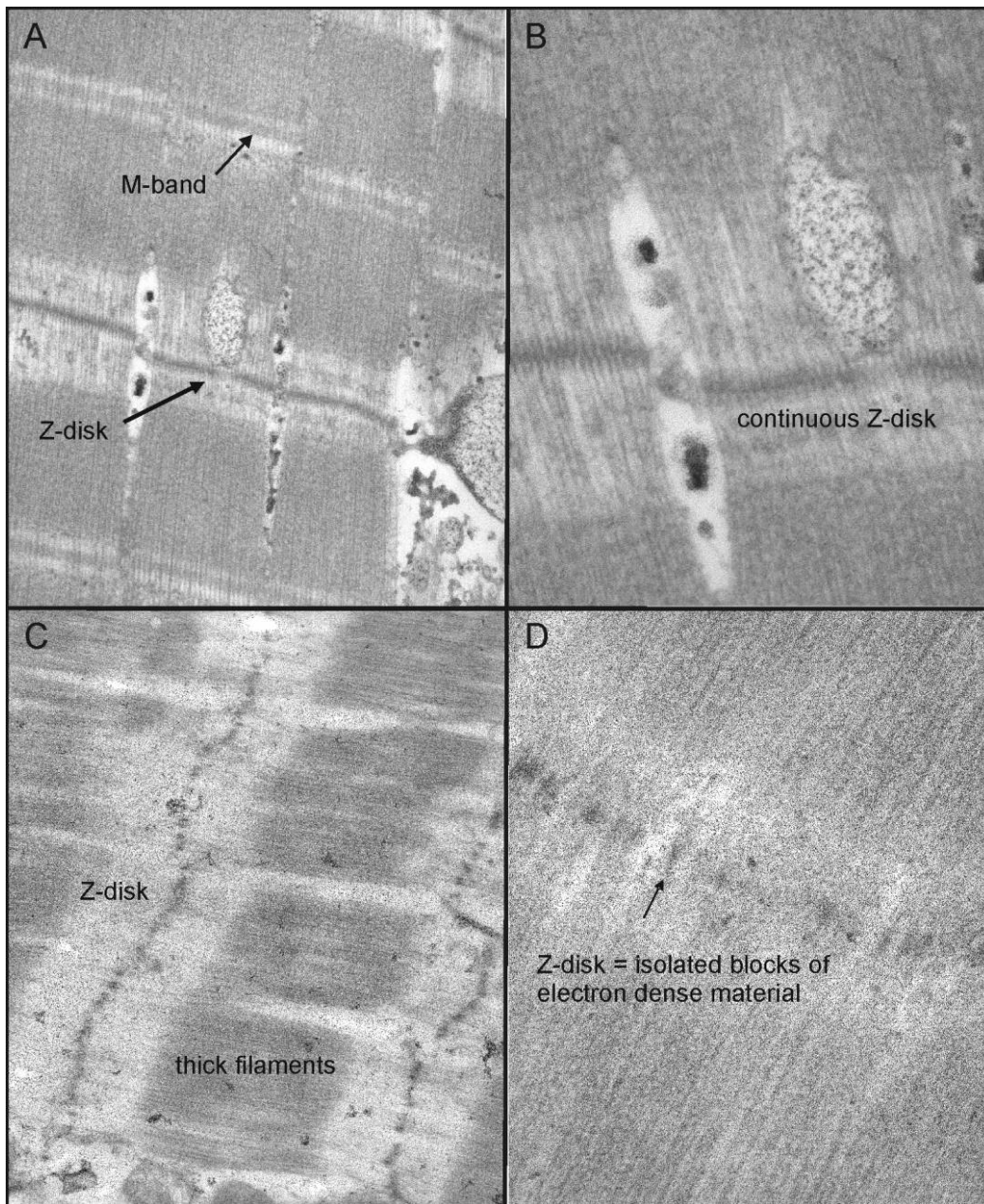


Figure 5. A, Transmission electron micrographs (TEMs) through the tongue-retractor muscle in *Hydromantes italicus*. B, detail of A showing the continuous Z-disk structure. C, TEMs through the tongue-retractor muscle in *Chameleo calyptatus*. D, Detail showing how the Z disk is composed of isolated blocks of electron-dense material.

which run from the mandible to the tongue (Fig. 2; Herrel et al. 1995; Meyers and Nishikawa 2000). Additionally, the so-called ring muscle causes forward sliding of the tongue on the tapered entoglossal process (Smith 1988; Herrel et al. 1995). Hyoid protraction is achieved by the action of the paired mandibulohyoideus muscles (Herrel et al. 1995; Meyers and Nishikawa 2000). Tongue retraction is achieved by activity of the m. hyoglossus, which runs from the first ceratobranchial into the tongue. Simultaneously, the hyoid is retracted by the paired hyoid retractors (m. sternohyoideus superficialis and profundus, m. omohyoideus), which run from the pectoral girdle to the

hyobranchium (Herrel et al. 1995; Meyers and Nishikawa 2000). The major difference in chameleons compared with agamid lizards is the shift in insertion of the mm. genioglossi from the tongue to the floor of the mouth (Herrel et al. 2001b). Thus, tongue protrusion/projection in chameleons is achieved solely by the activity of the m. accelerator (the homologue to the ring muscle in agamids; see Wainwright and Bennett 1992a, 1992b). Protrusion of the hyobranchium in chameleons is achieved by the mm. mandibulohyoidei, as is the case in agamids (Wainwright and Bennett 1992a; Herrel et al. 1995). The m. accelerator consists of helically wound short muscle

Table 1: Summary of morphological and functional attributes of the hyolingual system in a typical plethodontid salamander (*Hydromantes italicus*) and a chameleon (*Chameleo calyptratus*)

	<i>Hydromantes italicus</i>	<i>Chameleo calyptratus</i>
Body mass (g)	5	100
Snout-vent length (mm)	50	150
Tongue length (mm)	3.6	17
Tongue-extension length (mm)	39.2	315
Tongue-extension duration (ms)	14	30
Tongue-retractor length (mm)	34.5	41
Tongue-retractor sarcomere length (μm)	$1.74 \pm .06$	$2.28 \pm .1$
Tongue-retractor Z-disk perforations	No	Yes
Maximal prey size (% body mass)	3 ^a	30 ^b

Note. Table entries are based on one adult individual of each species. Sarcomere lengths were determined for 30 sarcomeres in the salamander and 15 sarcomeres in the chameleon.

^a Based on Vignoli et al. 2006.

^b Personal observation (by A. Herrel) of the maximal prey size the *C. calyptratus* individuals were willing to eat.

fibers surrounding the entoglossal process (Figs. 3, 4; Herrel et al. 2001*b*; de Groot and van Leeuwen 2004). Between the accelerator muscle and the entoglossal process, there are several layers of connective tissue sheets present (de Groot and van Leeuwen 2004). The accelerator muscle is activated 200–300 ms before the onset of tongue projection; during this time, these collagenous sheets are hypothesized to strain and store elastic energy for the tongue launch (de Groot and Van Leeuwen 2004).

A review of the literature shows that the tongue retractor in chameleons is unusual, as it is composed of supercontracting muscle fibers characterized by perforations in the Z disks (Fig. 5; Rice 1973; Herrel et al. 2001*a*, 2002). It is complexly folded in rest (Fig. 3), and its length is about half that of the body (Table 1). However, on projection, the muscle can be stretched up to four times its resting length, given that tongue-projection distances of up to two body lengths are not uncommon in many chameleons (Wainwright et al. 1991; A. Herrel, personal observation).

Convergence in Morphology and Design

The degree of convergence in the tongue-projection system in chameleons and plethodontid salamanders is striking. Independently, both groups have evolved similar tongue-projection systems relying on power amplification. Given similar functional demands but a radically divergent Bauplan, evolution has resulted in a remarkably similar solution in the mechanics (power amplifier) and morphology (helically wound muscle surrounding cartilaginous structure with circular connective tissue sheets that are preloaded and can store elastic energy; see Fig. 4). The difference in the projection system lies in the details. Whereas the tongue-protractor muscle is shot out of the mouth in the chameleons, the muscle remains inside the body in the plethodontids, and the hyobranchium with adhering tongue is propelled from the mouth. Although both

systems are clearly high-performance systems, salamanders appear to be generating substantially higher power outputs during tongue projection (de Groot and van Leeuwen 2004; Deban et al. 2007). However, this may be partially a scaling effect, because smaller animals are expected to produce higher power outputs (Hill 1950). Clearly, an analysis of tongue projection in juvenile chameleons or dwarf chameleons of the genus *Brookesia* or *Rhampholeon* could shed light on this.

Tongue retraction clearly involves different morphologies associated with the different mechanical demands. Whereas in chameleons, there is a demand for both force production (chameleons are known to eat relatively large prey; see Luiselli and Rugiero 1996; Pleguezuelos et al. 1999; Herrel et al. 2000; Keren-Rotem et al. 2006) and retraction capacity (the tongue can be extended up to 200% of body length), plethodontids eat relatively smaller prey (up to 3% of body mass; see Lynch 1985; Larsen et al. 1989; Vignoli et al. 2006) and do not extend their tongues that far (Wake and Deban 2000). Consequently, different solutions are presented to the problems associated with tongue retraction in the two groups. In the plethodontids, the retraction system is characterized by an extremely elongated tongue-retractor muscle that attaches onto the pelvis and, in rest, lies in a loop in front of the heart (Deban et al. 1997). Tongue retraction is relatively rapid compared with chameleons, sometimes taking as little as 30 ms. In chameleons, on the other hand, the muscle is also relatively long but is characterized by the presence of supercontracting muscle fibers that allow cross-bridge cycling and thus force production beyond the limits of typical cross-striated vertebrate muscle (Rice 1973; Herrel et al. 2001*a*, 2002). Tongue retraction, however, is much slower in chameleons (± 300 ms; Wainwright et al. 1991).

Despite the differences mentioned above, there are also a number of similarities in the retraction systems of both groups. Clearly, both plethodontid salamanders and chameleons have evolved long retractor muscles that are folded at rest and have broad length-tension curves. Because the tongue retractors need

Table 2: Summary kinematics of the transport of grasshoppers in *Pogona vitticeps* and *Chameleo calyptratus*

	Average Number of Transport Cycles	Average Transport Cycle Duration (s)	Average Hyoid Displacement (mm)
<i>Pogona vitticeps</i> ($N = 2$)	12.29 \pm 5.02	.46 \pm .09	5.87 \pm 2.41
<i>Chameleo calyptratus</i> ($N = 3$)	20.26 \pm 7.28	1.47 \pm .36	12.79 \pm 3.35

to generate force throughout tongue retraction, in chameleons, they developed supercontracting sarcomeres. Plethodontids, on the other hand, benefited from the design of generalized salamanders, as illustrated by *Salamandra*; these animals also have a long tongue-retractor muscle inserting at the pelvic girdle. Thus, by increasing the length of the tongue retractors to a relatively minor extent, they were able to retract their tongues after projection and sustained only a small cost in retraction speed. Chameleons have strong tongue retractors due to the supercontractile properties of the muscle but are also 10 times slower than salamanders. Clearly, the constraints imposed by Bauplan and ecology define the morphological solutions in light of the mechanical demands imposed.

Interestingly, however, on complete retraction of the tongue into the mouth, the tongue-retractor muscle in chameleons is no longer able to generate much force (Herrel et al. 2001a, 2002). Although the overall prey-transport kinematics are similar in chameleons and agamid lizards (So et al. 1992; Herrel et al. 1996), the hyoid rather than the tongue is recruited as the principal element ensuring prey transport and repositioning in chameleons as suggested by our data. Indeed, movements of the hyoid in chameleons are more extensive than in *Pogona vitticeps* ($F_{1,3,61} = 9.955$; $P = 0.040$) while transporting identical prey (Table 2). Moreover, chameleons tend to use more transport cycles on average ($F_{1,3,45} = 5.79$; $P = 0.08$), and each individual transport cycle takes significantly longer compared with *Pogona* ($F_{1,5,13} = 72.25$; $P < 0.001$). Interestingly, on complete retraction, the hyoid nearly touches the sternum (Fig. 1), and consequently the sternohyoideus muscle that runs from the sternum to the back of the basihyal must be contracting to more than 50% of its resting length. Although this suggests that this muscle may also be of the supercontracting type, an analysis of the ultrastructure of the m. sternohyoideus is needed to test this hypothesis. Plethodontid salamanders are probably faced with a similar problem because in rest, the tongue-retractor muscle is slack and lies in a loop in front of the heart. Preliminary electromyographic data do suggest that the muscle is active during intraoral transport (S. Deban, personal observation), but unfortunately, little is known about the role of the tongue during prey transport in these animals. Future experiments involving sonomicrometry or cineradiography may help elucidate this further.

An extreme specialization in function of the tongue in prey capture in chameleons and salamanders may obviously have consequences for other functions performed by the same system. Although these negative effects could potentially constrain the evolution of these extreme morphologies and behaviors,

plethodontid salamanders have partly overcome these constraints by decoupling at least one function (respiration) from the tongue. Whether the specialization of the tongue affects the efficiency of prey transport, however, remains unknown. In the chameleons, the role of the tongue during prey transport has been shifted to the hyoid apparatus. However, this shift comes at a cost as evidenced by both the larger number of transport cycles needed to transport a prey and the greater transport-cycle duration in comparison with a closely related lizard with a nonspecialized tongue, as suggested by our data. However, given the lifestyle of chameleons, characterized by cryptic colors and an extreme sit-and-wait foraging strategy, the incurred fitness cost of an increased prey-transport duration is probably minimal.

In summary, our review and data suggest extensive convergence in two distantly related taxa faced with similar mechanical demands on the use of the tongue during prey capture. When functional demands differ as a result of differences in ecology (as illustrated in the case of tongue retraction), the design of the system differs as well. Thus, our data suggest that the morphology of the prey-capture system in chameleons and plethodontid salamanders is the result of an extensive coevolution of ecology, function, and behavior.

Acknowledgments

We thank B. De Kegel and J. Christiaens for their help with sectioning chameleons and salamanders and Francis Terloo for help with the transmission electron microscopy. The research of Vicky Schaerlaeken is funded by a PhD grant of the Institute for the Promotion of Innovation through Science and Technology in Flanders.

Literature Cited

- Aerts P. 1998. Vertical jumping in *Galago senegalensis*: the quest for a hidden power amplifier. *Philos Trans R Soc Lond B* 353:1607–1620.
- Askew G.N. and R.L. Marsh. 2002. Muscle designed for maximum short-term power output: quail flight muscle. *J Exp Biol* 205:2153–2160.
- Bels V.L., M. Chardon, and K.V. Kardong. 1994. Biomechanics of the hyolingual system in Squamata. Pp. 197–240 in V.L. Bels, M. Chardon, and P. Vandewalle, eds. *Advances in Com-*

- parative and Environmental Physiology. Vol. 18. Springer, Berlin.
- Burkholder T.J. and R.L. Lieber. 2001. Sarcomere length operating range of vertebrate muscles during movement. *J Exp Biol* 204:1529–1536.
- Deban S.M. 1997. Modulation of prey-capture behavior in the plethodontid salamander *Ensatina eschscholtzii*. *J Exp Biol* 200:1951–1964.
- Deban S.M. and U. Dicke. 1999. Motor control of tongue movement during prey capture in plethodontid salamanders. *J Exp Biol* 202:3699–3714.
- . 2004. Activation patterns of the tongue-projector muscle during feeding in the imperial cave salamander, *Hydromantes imperialis*. *J Exp Biol* 207:2071–2081.
- Deban S.M., J.C. O'Reilly, U. Dicke, and J.L. van Leeuwen. 2007. Extremely high-power tongue projection in plethodontid salamanders. *J Exp Biol* 210:655–667.
- Deban S.M., D.B. Wake, and G. Roth. 1997. Salamander with a ballistic tongue. *Nature* 389:27–28.
- de Groot J.H. and J.L. van Leeuwen. 2004. Evidence for an elastic projection mechanism in the chameleon tongue. *Proc R Soc B* 271:761–770.
- Dockx P. and F. De Vree. 1986. Prey capture and intra-oral food transport in terrestrial salamanders. *Stud Herpetol* 1986:521–524.
- Fleming T.H., N. Muchhala, and P. Ornelas. 2005. New world nectar-feeding vertebrates: community patterns and processes. Pp. 161–182 in V. Sanchez-Cordero and R.A. Medellín, eds. *Contribuciones Mastozoológicas en Homenaje a Bernardo Villa-R*. Instituto de Biología e Instituto de Ecología, UNAM, Mexico City.
- Gordon A.M., A.F. Huxley, and F.J. Julian. 1966. The variation of isometric tension with sarcomere length in vertebrate muscle fibres. *J Physiol* 184:170–192.
- Herrel A., M. Canbek, Ü. Özelmas, M. Uyanoglu, and M. Karakaya. 2005. Comparative functional analysis of the hyolingual anatomy in lacertid lizards. *Anat Rec* 284:561–573.
- Herrel A., J. Cleuren, and F. De Vree. 1995. Prey capture in the lizard *Agama stellio*. *J Morphol* 224:313–329.
- . 1996. Kinematics of feeding in the lizard *Agama stellio*. *J Exp Biol* 199:1727–1742.
- Herrel A., J.J. Meyers, P. Aerts, and K.C. Nishikawa. 2001a. Functional implications of supercontracting muscle in the chameleon tongue retractors. *J Exp Biol* 204:3621–3627.
- Herrel A., J.J. Meyers, K.C. Nishikawa, and P. Aerts. 2000. The mechanics of prey prehension in chameleons. *J Exp Biol* 203:3255–3263.
- Herrel A., J.J. Meyers, K.C. Nishikawa, and F. De Vree. 2001b. Morphology and histochemistry of the hyolingual apparatus in chameleons. *J Morphol* 249:154–170.
- . 2001c. The evolution of feeding motor patterns in lizards: modulatory complexity and constraints. *Am Zool* 41:1311–1320.
- Herrel A., J.J. Meyers, J.-P. Timmermans, and K.C. Nishikawa. 2002. Supercontracting muscle: producing tension over extreme muscle lengths. *J Exp Biol* 205:2167–2173.
- Hill A.V. 1950. The dimensions of animals and muscular dynamics. *Sci Prog* 38:209–230.
- Huxley H.E. 1965. The mechanism of muscular contraction. *Sci Am* 213:18–27.
- Keren-Rotem T., A. Bouskila, and E. Geffen. 2006. Ontogenetic habitat shift and risk of cannibalism in the common chameleon (*Chamaeleo chamaeleon*). *Behav Ecol Sociobiol* 59:723–731.
- Kier W.M. and K.K. Smith. 1985. Tongues, tentacles and trunks: the biomechanics and movement of muscular hydrostats. *Zool J Linn Soc* 83:207–324.
- Lappin A.K., J.A. Monroy, J.Q. Pilarski, E.D. Zepnewski, D.J. Pierotti, and K.C. Nishikawa. 2006. Storage and recovery of elastic potential energy powers ballistic prey capture in toads. *J Exp Biol* 209:2535–2553.
- Larsen J.H., Jr., J.T. Beneski Jr., and D.B. Wake. 1989. Hyolingual feeding systems of the Plethodontidae: comparative kinematics of prey capture by salamanders with free and attached tongues. *J Exp Zool* 252:25–33.
- Lombard R.E. and D.B. Wake. 1977. Tongue evolution in the lungless salamanders, family Plethodontidae. II. Function and evolutionary diversity. *J Morphol* 153:39–80.
- Luiselli L. and L. Rugiero. 1996. *Chamaeleo chamaeleon* (common chameleon) diet. *Herpetol Rev* 27:78–79.
- Lutz G.J. and Rome L.C. 1994. Built for jumping: the design of the frog muscular system. *Science* 263:370–372.
- Lynch J.F. 1985. The feeding ecology of *Aneides flavipunctatus* and sympatric plethodontid salamanders in northwestern California. *J Herpetol* 19:328–352.
- Marsh R.L. 1994. Jumping ability of anuran amphibians. Pp. 51–111 in J.H. Jones, ed. *Advances in Veterinary Science and Comparative Medicine: Comparative Vertebrate Exercise Physiology*. Academic Press, New York.
- Meyers J.J. and K.C. Nishikawa. 2000. Comparative study of tongue protrusion in three iguanian lizards: *Sceloporus undulatus*, *Pseudotrapelus sinaitus* and *Chamaeleo jacksonii*. *J Exp Biol* 203:2833–2849.
- Meyers J.J., J.C. O'Reilly, J.M. Monroy, and K.C. Nishikawa. 2004. Mechanism of tongue protrusion in microhylid frogs. *J Exp Biol* 207:21–31.
- Muchhala N. 2006. Nectar bat stows huge tongue in rib cage. *Nature* 444:701–702.
- . 2007a. Adaptive trade-off in corolla shape mediates specialization for flowers pollinated by bats and hummingbirds. *Am Nat* 169:494–504.
- . 2007b. Character displacement among bat-pollinated flowers of the genus *Burmeistera*: analysis of mechanism, process, and pattern. *Proc R Soc B* 274:2731–2737.
- Nishikawa K.C. 1999. Neuromuscular control of prey capture in frogs. *Philos Trans R Soc B* 354:941–954.
- . 2000. Feeding in frogs. Pp. 117–144 in K. Schwenk, ed. *Feeding: Form, Function and Evolution in Tetrapod Vertebrates*. Academic Press, San Diego, CA.
- Nishikawa K.C., W.M. Kier, and K.K. Smith. 1999. Morphology and mechanics of tongue movement in the African pig-nosed

- frog (*Hemismus marmoratum*): a muscular hydrostatic model. *J Exp Biol* 202:771–780.
- Peters S.E. and K.C. Nishikawa. 1999. Comparison of isometric contractile properties of the tongue muscles in three species of frogs, *Litoria caerulea*, *Dyscophus guinetti* and *Bufo marinus*. *J Morphol* 242:107–124.
- Pleguezuelos J.M., J.C. Poveda, R. Monterrubio, and D. Ontiveros. 1999. Feeding habits of the common chameleon, *Chamaeleo chamaeleon* in the southeastern Iberian Peninsula. *Isr J Zool* 45:267–276.
- Redford K.H. 1985. Feeding and food preference in captive and wild giant anteaters (*Myrmecophaga tridactyla*). *J Zool (Lond)* 205:559–572.
- Rice M.J. 1973. Supercontracting muscle in a vertebrate. *Nature* 243:238–240.
- Rohlf F.J. 2004. tpsDIG32. Version 1.40. <http://life.bio.sunysb.edu/morph/index.html>.
- Rome L.C. and S.L. Lindstedt. 1997. Mechanical and metabolic design of the muscular system in vertebrates. Pp. 1587–1651 in W.H. Dantzler, ed. *Handbook of Physiology*. Section 13, vol. 2. Oxford University Press, New York.
- Schwenk K. 2000. Feeding in lepidosaurs. Pp. 175–291 in K. Schwenk, ed. *Feeding: Form, Function and Evolution in Tetrapod Vertebrates*. Academic Press, San Diego, CA.
- Smith K.K. 1988. Form and function of the tongue in agamid lizards with comments on its phylogenetic significance. *J Morphol* 196:157–171.
- Smith K.K. and W.M. Kier. 1989. Trunks, tongues, and tentacles: moving with skeletons of muscle. *Am Sci* 77:28–35.
- So K., P.C. Wainwright, and A.F. Bennett. 1992. Kinematics of prey processing in *Chamaeleo jacksonii*: conservation of function with morphological specialisation. *J Zool (Lond)* 226:47–64.
- Vignoli L., F. Caldera, and M.A. Bologna. 2006. Trophic niche of cave populations of *Speleomantes italicus*. *J Nat Hist* 40:1841–1850.
- Wainwright P.C. and A.F. Bennett. 1992a. The mechanism of tongue projection in chameleons. I. Electromyographic tests of functional hypotheses. *J Exp Biol* 168:1–21.
- . 1992b. The mechanism of tongue projection in chameleons. II. Role of shape change in a muscular hydrostat. *J Exp Biol* 168:23–40.
- Wainwright P.C., D.M. Kraklau, and A.F. Bennett. 1991. Kinematics of the tongue projection in *Chamaeleo oustaleti*. *J Exp Biol* 159:109–133.
- Wake D.B. and S.M. Deban. 2000. Terrestrial feeding in salamanders. Pp. 95–116 in K. Schwenk, ed. *Feeding: Form, Function and Evolution in Tetrapod Vertebrates*. Academic Press, San Diego, CA.