

Horned Lizards (*Phrynosoma*) Incapacitate Dangerous Ant Prey With Mucus

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ABSTRACT Horned lizards (Iguanidae, Phrynosomatinae, *Phrynosoma*) are morphologically specialized reptiles characterized by squat, tank-like bodies, short limbs, blunt snouts, spines and cranial horns, among other traits. They are unusual among lizards in the degree to which they specialize on a diet of ants, but exceptional in the number of pugnacious, highly venomous, stinging ants they consume, especially harvester ants (genus *Pogonomyrmex*). Like other iguanian lizards, they capture insect prey on the tongue, but unlike other lizards, they neither bite nor chew dangerous prey before swallowing. Instead, they employ a unique kinematic pattern in which prey capture, transport and swallowing are combined into a single feeding stage, apparently leaving the mouth, pharynx, esophagus and stomach vulnerable to bites and stings. Nevertheless, horned lizards consume dozens of harvester ants without harm. We show that their derived feeding kinematics are associated with unique, mucus-secreting pharyngeal papillae that apparently serve to immobilize and incapacitate dangerous ants as they are swallowed by compacting them and binding them in mucus strands. Radially branched esophageal folds provide additional mucus-secreting surfaces the ants pass through as they are swallowed. Ants extracted from fresh-killed horned lizard stomachs are curled ventrally into balls and bound in mucus. We conclude that the pharyngeal papillae, in association with a unique form of hyolingual prey transport and swallowing, are horned lizard adaptations related to a diet of dangerous prey. Harvester ant defensive weapons, along with horned lizard adaptations against such weapons, suggest a long-term, predator–prey, co-evolutionary arms race between *Phrynosoma* and *Pogonomyrmex*. *J. Exp. Zool.* 309A:447–459, 2008. © 2008 Wiley-Liss, Inc.

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Horned lizards, genus *Phrynosoma* (Squamata, Iguanidae), are small, bizarrely derived reptiles (Fig. 1) ranging across the American west from southern Mexico to southern Canada. They are particularly abundant in the deserts of the southwestern United States and northern Mexico (Sherbrooke, 2003). They belong to a diverse clade of iguanid lizards known as “phrynosomatines” (Schulte et al., 2003; traditionally called “sceloporines,” Etheridge and de Queiroz, ’88), but often accorded familial status as the Phrynosomatidae (Frost and Etheridge, ’89). This clade includes several generalized, basal genera as well as a more derived, monophyletic group known as the “sand lizards” (*Uma*, *Callisaurus*, *Cophosaurus* and *Holbrookia*). *Phrynosoma* is regarded as the sister group of the sand lizard clade (Wiens,

’93; Reeder and Wiens, ’96; Wilgenbusch and de Queiroz, 2000; Leaché and McGuire, 2006; Schulte and de Queiroz, 2008) (Fig. 2).

Most lizards eat a variety of prey types, especially small invertebrates. Dietary specialization, in the sense of stenophagy, is relatively uncommon among lizards and phenotypic specia-

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lization of the feeding apparatus reflecting a specialized diet is even less common (Greene, '82; Schwenk, 2000b; Pianka and Vitt, 2003; Schwenk and Rubega, 2005). Thus, horned lizards



Fig. 1. A Texas horned lizard, *Phrynosoma cornutum*, showing the tank-like body, short limbs, cranial horns and other morphological features typical of the genus and often erroneously attributed to myrmecophagy (see text).

are unusual in the degree to which most species specialize on a diet of ants. It is, therefore, not surprising that their highly derived and unusual morphology is widely believed to reflect their myrmecophagous habits. In this study we examine the functional morphology of ant consumption in horned lizards and consider the question of diet-specific adaptation.

Although most species of *Phrynosoma* eat other prey types, sometimes in significant quantities, ants are nearly always the most important type numerically and many species consume ants almost exclusively (Norris, '49; Pianka, '66; Pianka and Parker, '75; Whitford and Bryant, '79; Rissing, '81; Turner and Medica, '82; Suarez et al., 2000; Lahti and Beck, 2008). In a classic study of *Phrynosoma* ecology, Pianka and Parker ('75:156) concluded that horned lizards encompass a "unique constellation of anatomical, behavioral, physiological and ecological adaptations that facilitate efficient exploitation of ants as a food source

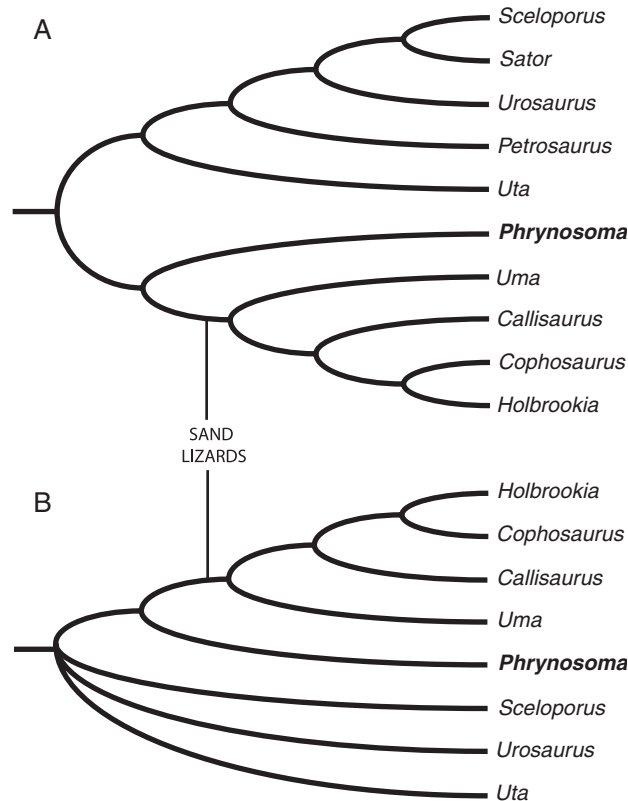


Fig. 2. Phylogenetic relationships among phrynosomatine lizards. Note that in both hypotheses, *Phrynosoma* is the sister taxon to a monophyletic group of genera known as the "sand lizards." Based on mitochondrial and nuclear DNA sequences, Leaché and McGuire (2006) found a similar sister relationship between *Phrynosoma* and the sand lizards with the exception that the positions of *Uma* and *Callisaurus* were reversed. (A) Relationships according to Wiens ('93) and Reeder and Wiens ('96) based on morphology and mitochondrial DNA sequence data. (B) Relationships according to Wilgenbusch and de Queiroz (2000) based on mitochondrial DNA sequence data.

and set [them] apart from most other species of lizards." These features include a squat, tank-like body that is dorsoventrally compressed, scaly spines, short limbs, a short tail, a head with blunted snout and posteriorly directed cranial horns, large stomach volume, relaxed thermoregulation and specialized foraging techniques, among others (Fig. 1). Many of these attributes may be a consequence of the poor nutritional quality of ants (Redford and Dorea, '84), which requires myrmecophages to consume them in large numbers, often at exposed colony sites. According to Pianka and Parker ('75), horned lizards adapted to the challenge of feeding on small, nutritionally poor prey by increasing stomach volume relative to body size so that they could consume the ants in large numbers. This putatively led to a reduction in locomotor speeds that, in conjunction with exposure during foraging in open habitats, led, in turn, to adaptations for increasing levels of predator defense, including cryptic coloration, freezing as a response to alarm, horns, spines, blood-squirting of defensive chemicals, etc. (Pianka and Parker, '75; Sherbrooke and Middendorf, 2001, 2004; Sherbrooke, 2003; Young et al., 2004). As such, horned lizards feed on many small prey items (ants) that are unevenly distributed (i.e., clumped) within open habitats that afford little cover or protection from visually hunting predators. Although the derived horned lizard phenotype is often assumed to reflect dietary specialization (e.g., Hotton, '55; Whitford and Bryant, '79; Schmidt and Schmidt, '89; MacMahon et al., 2000), most of the lizards' unusual morphology and behavior is actually attributable to selection stemming from predation pressure and resource competition only indirectly related to feeding on ants. In other words, most aspects of the derived horned lizard phenotype reflect the circumstances under which the lizards feed rather than the food they feed on. No adaptations of the trophic apparatus related directly to a diet of ants have been demonstrated for *Phrynosoma* thus far.

In fact, many lizards eat ants without any apparent phenotypic specialization (reviewed by Schwenk, 2000b; Pianka and Vitt, 2003; Vitt et al., 2003). However, horned lizards are exceptional—probably unique—in the number of dangerous, highly venomous ants included in their diets, particularly harvester ants of the genus *Pogonomyrmex* (Formicidae) (Pianka and Parker, '75; Whitford and Bryant, '79; Rissing, '81; Turner and Medica, '82; Schmidt and Schmidt, '89; Schmidt

et al., '89; Suarez et al., 2000). Indeed, several horned lizard species eat almost nothing else. In contrast, most lizards select more nutritious prey and avoid dangerous prey as much as possible (Vogel and von Brockhusen-Holzer, '84; Hasegawa and Taniguchi, '93, '96; López and Martín, '94; Vitt et al., 2003). A closely related sand lizard, *Uma inornata*, for example, only resorts to feeding on *Pogonomyrmex* ants in drought years when other insects are unavailable. The lizards fail to thrive on this diet and lose weight (C. W. Barrows, personal communication).

Harvester ants have powerful mandibles for cropping seeds (Hermann and Blum, '81; Schmidt and Schmidt, '89) and these are also used defensively against predators, including horned lizards (Schmidt and Blum, '78; Hermann and Blum, '81; Schmidt and Schmidt, '89; Sherbrooke, personal observation). In addition, *Pogonomyrmex* ants have an autotomizing sting (i.e., one that breaks off in the victim) that injects a potent venom. Stinging is used solely as a defensive weapon (Cole, '68; Hermann and Blum, '81; Schmidt and Schmidt, '89). One species (*P. maricopa*) produces the most toxic venom known for any arthropod (according to LD₅₀ studies with mice [Schmidt and Schmidt, '89]). Sting autotomy is thought to have evolved in ants specifically as a defense against vertebrate predators (Hermann, '71; Schmidt and Blum, '78; Kugler, '79; Hermann and Blum, '81; Schmidt and Schmidt, '89). In humans *Pogonomyrmex* stings cause excruciating pain (Cole, '68; Schmidt and Schmidt, '89). However, horned lizards are less sensitive to *Pogonomyrmex* venom than related lizards because of a blood plasma factor that detoxifies the venom to some extent (Schmidt and Schmidt, '89; Schmidt et al., '89). Nonetheless, they are not immune—the venom is fatal if a lizard is stung too many times (Edwards, 1896; Schmidt et al., '89).

While foraging, horned lizards must balance the need to maximize the number of ants consumed with the need to avoid mobbing (and potentially fatal stinging) by an aroused colony, as well as the need to minimize movements that might attract predators. They have evolved an unusual foraging strategy in which they move through the environment seeking columns of foraging ants, whereupon they remain still (Whitford and Bryant, '79; Rissing, '81; Shaffer and Whitford, '81; Munger, '84), capturing ants with exceptional speed by using precisely targeted tongue protrusion and rapid prey handling (see below; Schwenk, 2000b,

and unpublished data; Ott et al., 2004, and unpublished data; Meyers and Herrel, 2005). Before the ant colony can respond defensively, the lizards cease predation and move on in search of another column (Whitford and Bryant, '79; Rissing, '81).

Like other members of the iguanian clade (Iguanidae, Agamidae and Chamaeleonidae), horned lizards capture small prey with the tongue (Schwenk and Throckmorton, '89; Schwenk, 2000b; Meyers and Herrel, 2005). They are able to modulate tongue trajectory and distance as they target fast-moving ant prey using rapid head movements in response to visual feedback (Ott et al., 2004, and unpublished data). Our initial observations suggested that, unlike other insectivorous lizards, horned lizards neither bite nor chew captured ants before swallowing (Schwenk, 2000b; Schwenk and Sherbrooke, 2003; Sherbrooke, 2003; Ott et al., unpublished data). Therefore, during prey capture, intraoral transport and swallowing, they potentially expose the vulnerable soft tissues of the mouth and pharynx to bites and stings. In contrast, other lizards that only occasionally eat stinging hymenopterans or other dangerous prey adopt an unusual capture and processing behavior involving rapid biting and chewing to kill the prey before it can sting (Vitt and Cooper, '88; O'Connell and Formanowicz, '98; Lappin and German, 2005; C. W. Barrows, personal communication). However, killing each prey item by biting or chewing incurs a significant cost in handling time, and this may make such a strategy unfeasible for horned lizards (see below).

In this study we address the problem of how horned lizards are able to consume large numbers of pugnacious, biting, stinging, highly venomous ants without being injured or killed in the process.

MATERIALS AND METHODS

We examined feeding behavior in Texas horned lizards (*P. cornutum*, $n = 4$) using high-speed (250 frames per sec) 16 mm cine film and frame-by-frame motion analysis. During filming, lizards were fed small, juvenile crickets comparable in size to *Pogonomyrmex* ants. We also examined the stomach contents of ten fresh, road-killed specimens of *P. cornutum* from southwestern New Mexico with particular attention to prey condition. Finally, we used standard paraffin histology to examine the microscopic anatomy of the mouth and pharynx ($n = 4$). Specimens were decalcified in formic acid A solution (Presnell and Schreiber, '97), sectioned at 8–10 μm and stained with either

Ehrlich hematoxylin and eosin or Weigert iron hematoxylin and picro-ponceau (protocols slightly modified from Presnell and Schreiber, '97 and available on request from K. S.). We compared our results to similar sections made of other *Phrynosoma* species (*P. asio*, *P. coronatum*, *P. hernandesi*, *P. douglasii*, *P. modestum*, *P. mcallii*, *P. orbiculare*, *P. platyrhinos* and *P. solare*), as well as sister and outgroup taxa including the sand lizards (*C. draconoides*, *U. scoparia*, *C. texanus*, *H. maculata*), other phrynosomatines (*Sceloporus* spp., *Urosaurus graciosus*, *Uta stansburiana*), other iguanids, agamids and more distant, noniguanian, taxa (including representatives of all lizard families). Included in our outgroup samples were other ant-eating lizards such as *Anolis bonairensis*, *Liolaemus monticola* (Iguanidae) and the agamids *Draco volans* and *Moloch horridus*. We also examined the morphology of the esophagus in *P. cornutum*, *P. modestum*, *P. platyrhinos*, *P. mcallii*, *P. hernandesi*, *P. coronatum* and *P. solare*, and compared our results with data in the literature for other squamates.

RESULTS

Feeding kinematics

Horned lizards capture prey using a mostly typical iguanian lizard pattern of lingual prehension (Schwenk and Throckmorton, '89; Schwenk, 2000b; Meyers and Herrel, 2005). They differ slightly from other iguanid species in protruding the tongue relatively farther (tongue protrusion distance/jaw length = 0.50 as compared with 0.17–0.31; some agamids have comparable protrusion distances, however; Schwenk and Throckmorton, '89) and in having a derived tongue and hyobranchial morphology (Schwenk, '94, and in preparation). However, they show dramatic departures from other iguanian taxa in two ways. First, the films confirmed that the teeth are not used at all to chew or crush small prey after capture (Fig. 3A–C) (noted also in *P. platyrhinos*; Schwenk, unpublished data; Meyers and Herrel, 2005). In contrast, the usual iguanian pattern is to place the prey item between upper and lower tooth rows for killing and chewing immediately upon capture (Fig. 3D–F) (Schwenk and Throckmorton, '89; Schwenk, 2000b). Second, horned lizards exhibit a unique hyolingual behavior after lingual retraction of prey into the mouth (Fig. 3A–C). Retraction is extremely fast (approx. 30 msec) and in a single motion the prey item is pulled past the teeth postero-ventrally, deep into the pharynx and

directly to the esophagus. This is evident externally as a pronounced bulge in the throat caused by movement of the tongue and hyobranchial apparatus. Swallowing is coincident with this behavior so that when the tongue is once again protracted, the prey item has disappeared from the oral cavity and pharynx. This behavior contrasts sharply with other iguanians in which such extreme bulging is evident neither during lingual prey capture, nor during processing, transport and swallowing cycles (Fig. 3D–F). Furthermore, in other lizards intraoral transport and swallowing are accomplished during separate, kinematically distinct hyolingual cycles that are independent of the ingestion (prehension/capture) stage. As such,

the merging of ingestion, transport and swallowing into a single feeding (ingestion) stage in *Phrynosoma* is exceptional, if not unique (see Schwenk, 2000a,b and Schwenk and Rubega, 2005, for reviews of feeding stages in lizards and other vertebrates).

Horned lizards often exhibit several low-amplitude gape and hyolingual cycles after the initial ingestion cycle (see Schwenk, 2000b, Fig. 8.22), as if clearing the mouth and pharynx of extraneous material. We did not offer our lizards prey in rapid sequence, as would be typical in the field, and it is possible that these extraneous cycles would be eliminated under such circumstances. An entire ingestion cycle, from prehension to swallowing,

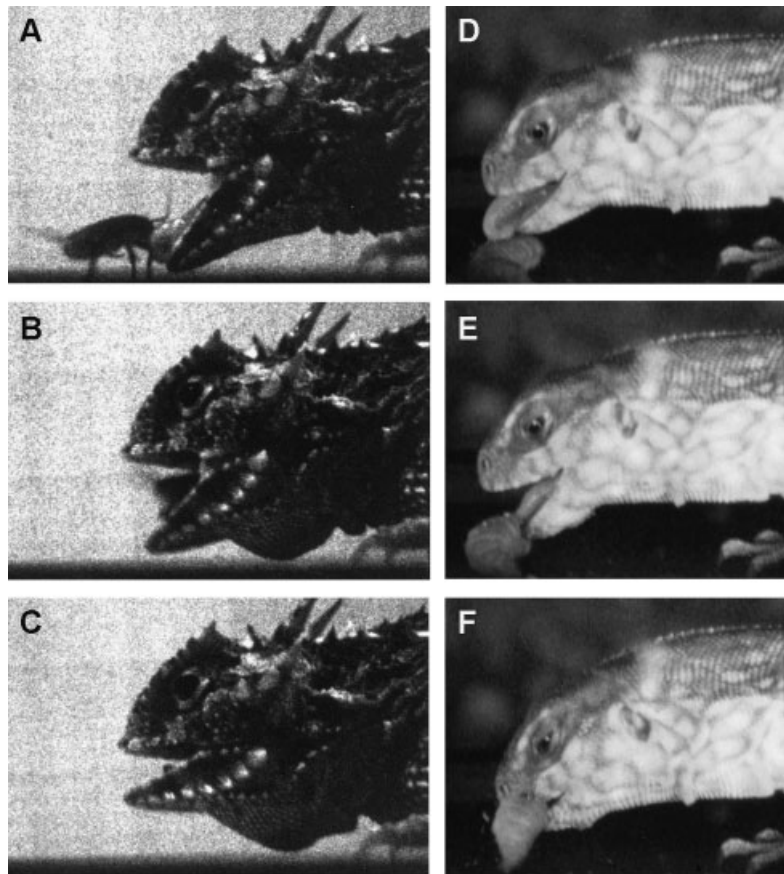


Fig. 3. The kinematics of lingual prehension in a horned lizard (*P. cornutum*) and a generalized iguanid lizard (desert iguana, *Dipsosaurus dorsalis*). (A–C) Three individual frames from a feeding sequence in *P. cornutum* showing retraction of the prey item into the mouth on the tongue's surface. Note the extreme ventral bulge of the gular region as the tongue and hyobranchial apparatus are retracted postero-ventrally. During this rapid movement (approx. 30 msec total), the prey item is forced through the pharyngeal papillae where it is balled, bound in mucus and pushed into the esophagus—an action that combines in one feeding stage/jaw cycle the typically disparate stages of ingestion, processing, transport and swallowing. (D–F) Typical iguanian lizards, such as this desert iguana, capture prey on the tongue and then crush it between the teeth (this is true even for smaller prey than shown). Note that the transit of the hyolingual apparatus is modest and little gular bulging is evident. Separate chewing, transport and swallowing cycles usually follow ingestion and are not accompanied by the kind of extreme gular bulging evident in horned lizards (A–C). Pharyngeal papillae are lacking. Compare also Figs. 4 and 5 in Meyers and Herrel (2005) showing ingestion in *Uma notata* and *P. platyrhinos*.

takes approximately 60 msec, although protrusion time and distance can be modulated so that longer times are often observed (Ott et al., 2004).

Although for our film trials we fed the horned lizards baby crickets, our personal observations, as well as high-speed video recordings (Ott et al., unpublished data), of *Phrynosoma* feeding on *Pogonomyrmex* ants confirm that they use identical feeding kinematics. Horned lizards seem to employ a stereotyped feeding behavior when feeding on a variety of prey types (W. C. S. and K. S., personal observation). As such, there is no reason to believe that our film observations are an artifact of feeding the lizards a non-ant prey type.

Condition of ants in stomach

Stomachs of fresh, road-killed specimens were packed with dozens of *Pogonomyrmex* and other ants. Most ants were intact, although the heads of some had broken off. The ants were almost certainly alive when initially swallowed (e.g., Sherbrooke, 2002; see below). Isolated, individual ants were encased within mucus, usually curled ventrally into a compact ball, with limbs, antennae, mandibles, body segments and stingers bound together by strands of mucus (Fig. 4). This observation suggests circumstantially that during passage through the esophagus, each ant is

immobilized so that stinging and/or biting would be impossible.

Histology of the pharynx and esophagus

In all *Phrynosoma* species examined, gross examination and histological sections revealed that the ventral surface of the pharynx is covered by a deep pile of long papillae, densely covered with columnar mucocytes (Figs. 5, 6). Thiomine staining confirmed that the mucocytes are secretory cells rich in mucopolysaccharides. The long, mucous papillae typically extend from the posterior portions of the tongue, over the larynx and the trachea, and throughout the ventral portion of the pharynx. The pharyngeal papillae are oriented posteriorly, slightly overlapping those behind (Figs. 5A, 6B, D, E). Note that the lingual morphology of *Phrynosoma* is highly derived relative to other iguanians (Schwenk, '94; in preparation). One derived characteristic is the separation of the genioglossus lateralis muscles from the flanks of the tongue body so that they insert much farther posteriorly compared with other species, forming prominent ridges alongside the tongue proper (Fig. 5; compare to *Sceloporus*). The papillae that cover these muscles and the back end of the tongue are unusually long and robust; they are unlike the lingual papillae typically found

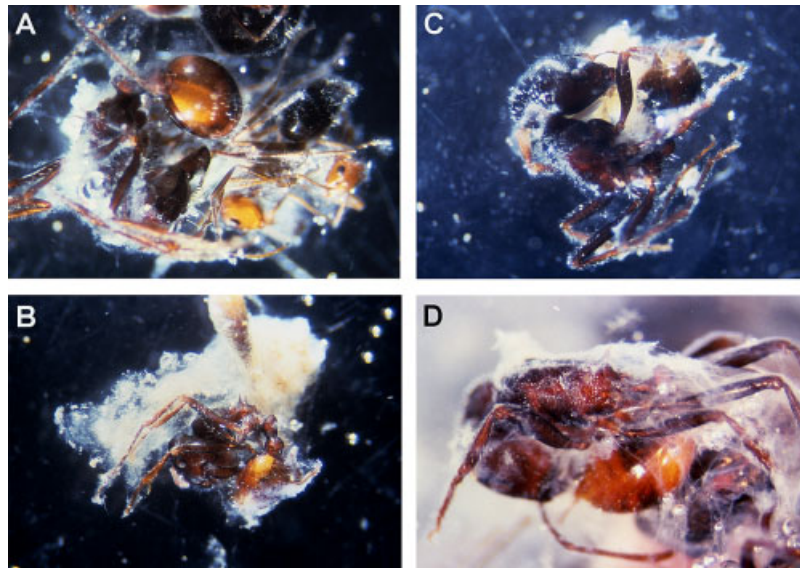


Fig. 4. Photomicrographs of ants extracted from the stomachs of fresh, road-killed specimens of *P. cornutum* in New Mexico. (A) A small group of ants balled and bound in mucus; (B–D) individual harvester ants (*Pogonomyrmex* sp.) tightly bound in mucus. Note that each ant is curled ventrally into a ball, bound by strands of mucus and presumably incapacitated, though it may remain alive in the gut for some time. The stomach may be further protected by mucus that forms a discrete sac lining the stomach (Lahti et al., in preparation).

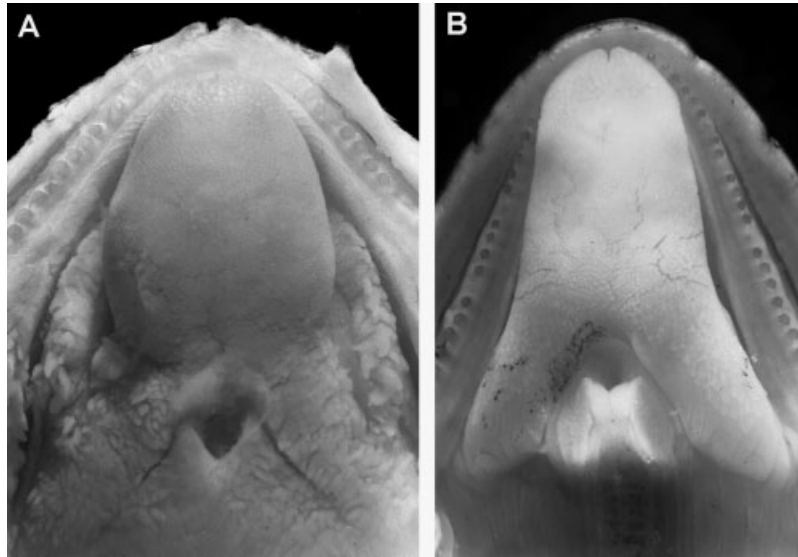


Fig. 5. Photomicrographs illustrating the gross form of the lower jaw, tongue, larynx and floor of the pharynx in two phrynosomatine lizards. (A) A horned lizard, *P. douglasii*, showing the unusual tongue morphology and the thick carpet of robust, mucous papillae covering the posterior limbs of the tongue, the larynx (dark central hole is the glottis) and the floor of the pharynx posterior to the tongue. The papilla-covered bands of tissue curving on either side of the tongue are the genioglossus lateralis muscles that in other lizards form the lateral walls of the tongue, but in horned lizards are discrete (Schwenk, '94 and in preparation). The tongue, itself, is covered with much finer papillae used during prey prehension on the tongue's anterior surface (see Schwenk, 2000b). (B) Tongue and pharyngeal form in a generalized, basal phrynosomatine, *Sceloporus* sp. Note the lack of papillae on the larynx and the floor of the pharynx. The pharynx is covered, instead, with thin striations and a typical mucous epithelium (see Fig. 6G). The form of the pharynx in this species is typical of most lizards.

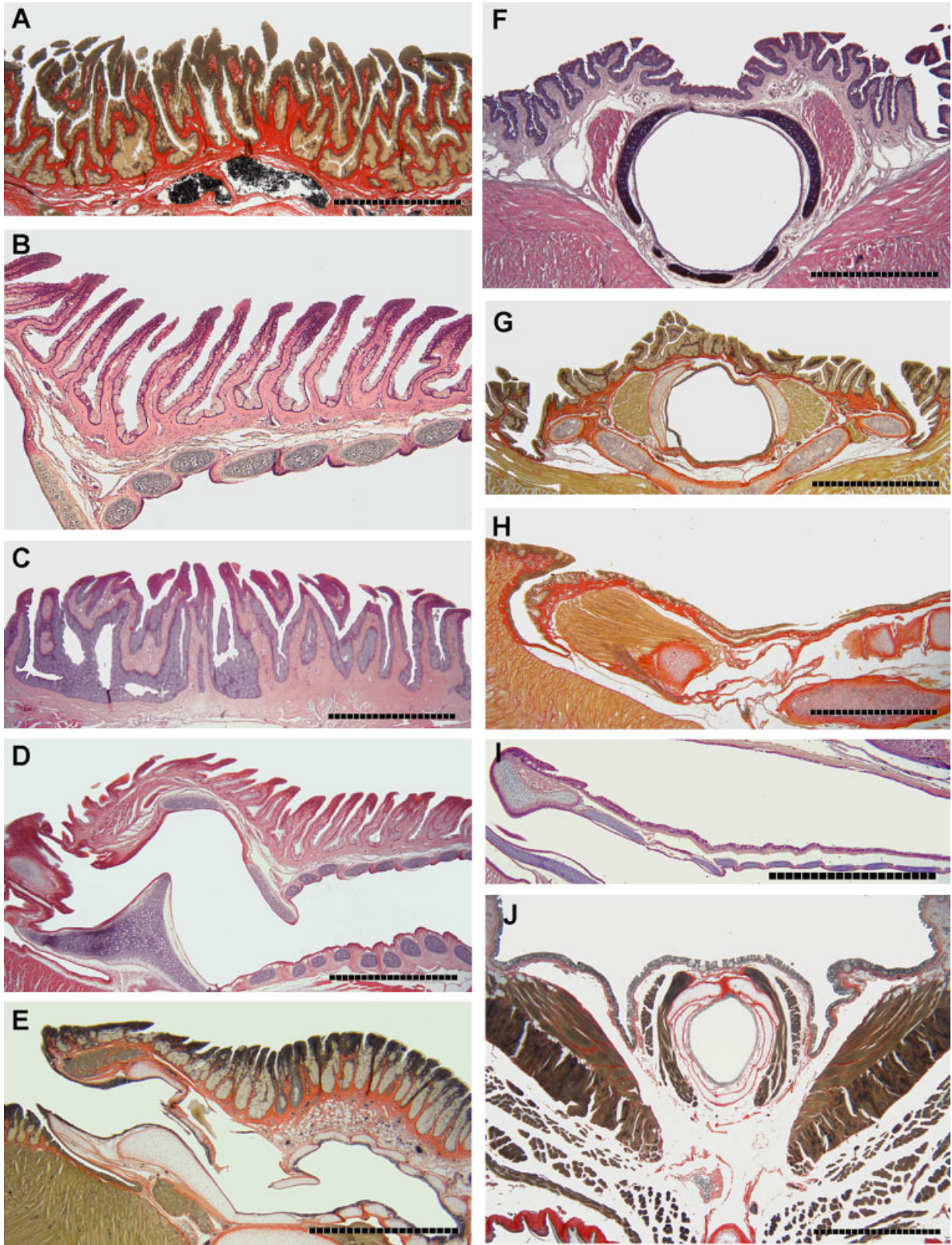
on this part of the tongue (e.g., Schwenk '88) and histologically appear to be an anterior extension of the "pharyngeal" papillae described here.

Pharyngeal papillae end abruptly at the beginning of the esophagus where they are replaced by longitudinal folds running the length of the esophagus to the stomach. The folds comprise major and minor parts; each major fold represents a set of several radially branching minor folds or pleats, visible as longitudinal striations on the surface of each major fold (Fig. 7). Each pleat is covered with a dense layer of goblet cells that release their mucus into the longitudinal grooves between pleats (Fig. 7C). Note that in Figures 7A and B the esophagus has been slit longitudinally and flattened to visualize the luminal surface. In life, however, the folds project well into the esophageal lumen, virtually occluding it (Fig. 7C). Thus, engulfment of ants in mucus-secreting tissues would be undiminished as they travel throughout the pharynx and esophagus to the stomach.

In all sister and outgroup species examined, pharyngeal papillae are entirely absent (e.g., Fig. 6H, I, J), or are only weakly developed (e.g., Fig. 6F, G). Within the phrynosomatines, basal taxa (*Sceloporus*, *Urosaurus* and *Uta*), as

well as two sand lizard taxa (*Callisaurus* and *Uma*) lack pharyngeal papillae completely, whereas *Cophosaurus* and *Holbrookia* exhibit very weakly developed papillae (which become even less developed posterior to the sections illustrated in Fig. 6). Pharyngeal papillae are absent in all more distant outgroup taxa, as well. As an outgroup control, we included in our study several iguanian taxa that are putative ant specialists (*A. bonairensis*, *L. monticola*, *D. volans*, *M. horridus*; see Schwenk, 2000b, Table 8.6, for references). All of these taxa completely lack pharyngeal papillae (e.g., Fig. 6J). These results strongly suggest that the well-developed, mucus-secreting pharyngeal papillae present in all *Phrynosoma* species examined are uniquely derived within the genus.

Evolutionary inferences about esophageal morphology in *Phrynosoma* are more tentative because we did not undertake a systematic study of the esophagus for this study and published data are inadequate for comparative purposes. Longitudinal folds and striations are typical of the esophagus in most squamate reptiles, as are mucus-secreting goblet cells (Luppa, '76; Parsons and Cameron, '76; Imai and Shibata, '92; Loubourdis, 2003). Indeed, longitudinal folds are typical of most vertebrates owing to the need for



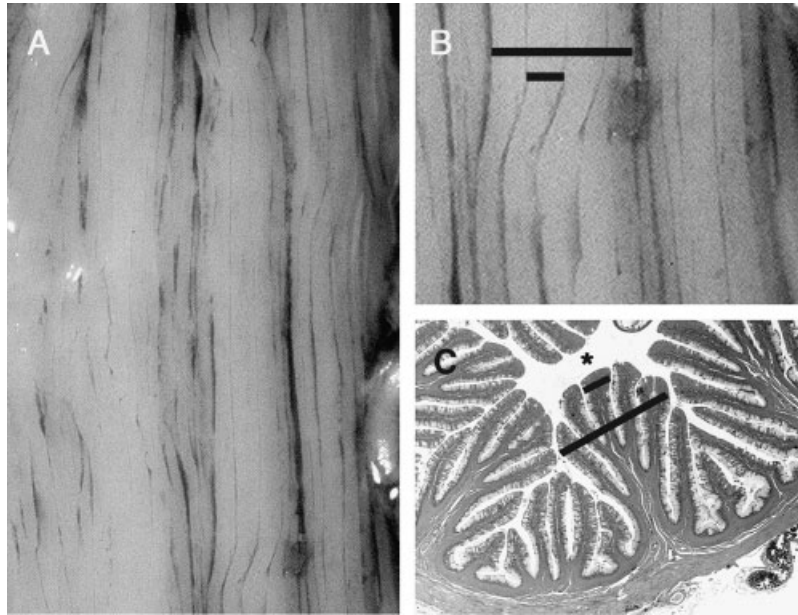


Fig. 7. Surface morphology and histology of the esophagus in *Phrynosoma*. (A) Gross morphology of the luminal surface in *P. cornutum*. Anterior is at the top. Note the major folds and their longitudinal striations indicating the minor folds, or pleats. (B) A digital enlargement of (A) delimiting the major folds and pleats with long and short lines, respectively. (C) Transverse section of the esophagus in *P. solare* (H & E). Note the extreme development of the major folds (long line) and the radial branching of pleats (short line) within each major fold. In situ the folds virtually occlude the lumen (*) Each pleat is covered by a dense mucous epithelium such that the mucus drains into the clefts between pleats. No scale bars available.

the esophagus to expand during swallowing of food. However, in only a few squamate species examined (e.g., *Agama [Stellio] stellio* and *Natrix* sp.) is there evidence of three-dimensional relief significant enough to cause luminal occlusion approaching what we observe in *Phrynosoma* (Luppa, '76; Loumbourdis, 2003). We can find no evidence that any other taxa exhibit the radial branching of the major folds into pleats (a morphology that greatly enhances the surface

area of the mucus-secreting epithelium). Thus, it is possible that the esophagus in *Phrynosoma* is derived both quantitatively and qualitatively as compared with other squamates (in the relative development of the longitudinal folds and in the subdivision of the folds into pleats that increase mucosal surface area, respectively). Any such conclusion must be verified by additional, comparative data, however, particularly for other phrynosomatine species. Regardless, it is clear

Fig. 6. Histology of the pharynx in phrynosomatine lizards. In all cases the scale bar = 1 mm. (A) *P. cornutum*, transverse section, showing pharyngeal papillae covering the larynx. Red-staining tissue is the collagenous, connective tissue core of the papillae; the yellowish-tan tissue is the mucous epithelium covering the sides of the papillae. In the transverse plane the papillae are complex and often form anastomoses with mucous crypts. Weigert hematoxylin and picro-ponceau stain (H & PP). (B) *P. cornutum*, mid-sagittal section, of the larynx. Anterior is to the left. The space at the bottom of the picture is the lumen of the trachea; note the purple-staining laryngeal cartilages below. Long pharyngeal papillae extend dorsally and slightly posteriorly. Mucocytes covering the papillae are stained pale pinkish-lavender. The epithelium at the papillary apices is nonmucous and stains more darkly. This photograph was taken without reference to a scale, so no scale bar is present, but the scale is similar to the other photos. Ehrlich hematoxylin and eosin stain (H & E). (C–D) *P. coronatum*, transverse and mid-sagittal sections, respectively (H & E). (E) *P. orbiculare*, mid-sagittal section, showing larynx and the opening of the glottis at the back of the tongue to the left (H & PP). (F) *Uma scoparia*, a sand lizard, transverse section through the larynx showing weakly developed pharyngeal papillae mostly lacking mucous cells (H & E). (G) *Holbrookia maculata*, another sand lizard, transverse section through the larynx showing weakly developed pharyngeal papillae with a mucous epithelium. In sand lizards, the minimal pharyngeal papillae do not extend very far posteriorly (H & PP). (H) *Sceloporus occidentalis*, a basal phrynosomatine, parasagittal section through larynx with posterior end of the tongue at left. The yellow-staining tissue is primarily lingual and intrinsic laryngeal muscle. Note smooth surface of the pharyngeal epithelium dorsally. Only the occasional goblet cell is evident. This type of featureless, nonpapillose epithelium is characteristic of nearly all lizards examined (H & PP). (I) *Uta stansburiana*, another basal phrynosomatine, mid-sagittal section through the larynx (H & E). (J) *Anolis bonairensis*, an ant-eating iguanid unrelated to horned lizards and other phrynosomatines, showing a complete absence of pharyngeal papillae. Transverse section (H & PP).

that the structure of the horned lizard esophagus would further promote mucus binding of ants as they move to the stomach.

DISCUSSION

Our data provide strong circumstantial evidence that horned lizards are able to consume dangerous ants by immobilizing them with mucus within the pharynx immediately after capture. Mucus binding presumably occurs when the ants are pushed deep into the papillae by the tongue, simultaneously expressing mucus from the papillae, compacting the ant and incapacitating its weapons (mandibles and stinger). This phase of the process is evident externally as pronounced gular bulging, reflecting extreme postero-ventral retraction of the hyolingual apparatus as the insect is transported through the pharyngeal papillae and into the esophagus. This scenario is also supported by high-speed video images of feeding *P. cornutum* that reveal thick strands of mucus running across the pharynx when the mouth is opened (Ott et al., unpublished data). Most iguanian lizards swallow by pushing food into the esophagus with posterior lobes of the tongue in a cyclical behavior known as "pharyngeal packing" (Smith, '84; Schwenk, 2000b). Horned lizards, however, are exceptionally short-necked and the pharynx is correspondingly abbreviated so that the ant is pushed directly into the esophagus after traversing the pharyngeal papillae. Horned lizards, therefore, combine the three stages of ingestion, transport and swallowing typically seen in lizards into a single feeding stage. Prey processing with the jaws and teeth, also evident in most lizards, is eliminated and replaced with a unique form of pharyngeal prey processing.

The precise manner in which ants are transported through the pharyngeal papillae to the esophagus and bound with mucus remains unclear. Preliminary data suggest that horned lizards deform the posterior end of the tongue in an unusual way during this transport process and that the tongue exhibits several derived attributes that might be related to this behavior (Schwenk, unpublished data).

Crushing ants with the teeth might be a simpler way to render them harmless, but this may not be feasible in horned lizards because the time required to position prey between the teeth for chewing could allow an ant to bite or sting vulnerable oral tissues. By immobilizing ants almost immediately after prey capture, horned

lizards disable the ant's weapons before they can be used defensively. Furthermore, by substituting single-cycle pharyngeal processing for multiple cycles of chewing, transport and swallowing, horned lizards are able to reduce dramatically the handling time of individual prey items. Handling time is especially important for horned lizards because they feed rapidly on multiple prey items during a single feeding bout as they pick-off individual ants from foraging columns. They consume as many ants as possible in a short time so that they can move on to another colony before being detected and mobbed by aggressive ant workers, or attacked by predators. Indeed, Meyers and Herrel (2005) showed that the specialized ant eaters, *P. platyrhinos* and *M. horridus* (Agamidae), differed from other lizards in the rapidity of their feeding events owing to the elimination of an oral processing stage.

Small movements of ants in the pharynx and esophagus of horned lizards have been visualized by X-ray videography, demonstrating that ants remain alive as they are swallowed (J. Meyers, personal communication). Presumably, insects not crushed by chewing remain alive in a lizard's gut for some time before chemical digestion. By incapacitating the ants' weapons at the time of capture, horned lizards not only avoid bites and stings within the mouth and throat, but also in the stomach. The reality of this danger is indicated by the deaths of several round-tail horned lizards (*P. modestum*) kept in outdoor enclosures (Sherbrooke, 2002). Postmortem examination revealed that the lizards had died after consuming large beetles that had bored through their stomachs. *P. modestum*'s mucus-binding defense was probably ineffective because of the beetles' size, form and strength or possibly because this small horned lizard species has secondarily reduced its dietary dependence on *Pogonomyrmex* ants (Whitford and Bryant, '79; Shaffer and Whitford, '81; Munger, '84; Schwenk and Sherbrooke, in preparation).

To our knowledge the mucus prey-binding system we describe is unique among vertebrates. Mammalian myrmecophages, especially edentates and pangolins, have exceptionally large salivary glands and sticky mucus that coats the tongue for prey adhesion (e.g., Doran and Allbrook, '73; Reiss, 2000), and this might passively incapacitate some dangerous ants. However, mammals in general avoid exposure to dangerous ant species (Swart et al., '99) and feed preferentially on harmless and more nutritious termites. Mammalian myrmecophages do share some interesting

attributes of the feeding apparatus with horned lizards, notably lingual prehension of prey, copious salivary mucus, reduced bite forces and lack of oral processing (Reiss, 2000; Meyers et al., 2006).

Some marine turtles have heavily keratinized pharyngeal and esophageal papillae, but these only protect the pharyngeal surfaces against abrasion and stings (respectively) from the siliceous sponges and medusae they consume, as well as directing the gelatinous prey toward the esophagus (e.g., Schumacher, '73; Parsons and Cameron, '76; Meylan, '88; Winokur, '88); they do not disarm the prey item, itself. Some freshwater, pelomedusid (side-necked) turtles have similar, but unkeratinized papillae in the esophagus (Parsons and Cameron, '76; Vogt et al., '98), but these are believed to serve as filters to retain small food items in the gut during water expulsion after feeding (Belkin and Gans, '68; Vogt et al., '98).

We have shown that horned lizards possess several uniquely derived traits—a single, combined ingestion/transport/swallowing stage, elimination of chewing cycles, pronounced postero-ventral hyolingual retraction, and mucus-secreting pharyngeal papillae—that we infer to be a character complex functionally related to incapacitating the weapons of the dangerous ant prey they consume in large numbers. It is reasonable to conclude that these traits represent adaptations to their specialized diet. Nonetheless, it is important to recognize that such adaptations are not a response to myrmecophagy, per se, but rather reflect the need to cope with highly defended, dangerous prey. Thus, many lizards are able to consume large quantities of ants without apparent phenotypic specialization (Schwenk, 2000b). Other than their low nutritional value, most ants do not seem to pose a particular challenge to the generalized lizard feeding system (Schwenk, 2000b; Vitt et al., 2003). This finding highlights the fact that it is a food item's mechanical attributes, rather than its taxonomic affiliation, that mediates the evolutionary link between dietary and phenotypic specialization (see discussion in Schwenk, 2000b). The presence of effective antipredator, defensive weapons in harvester ants and unique adaptations (behavioral, morphological and physiological) to cope with these weapons in horned lizards suggests the possibility of a long-standing predator-prey, co-evolutionary arms race in this system, a hypothesis we are presently exploring (Schwenk and Sherbrooke, in preparation).

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