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Responses of Kit Foxes (*Vulpes macrotis*) to Antipredator Blood-Squirting and Blood of Texas Horned Lizards (*Phrynosoma cornutum*)

WADE C. SHERBROOKE AND GEORGE A. MIDDENDORF III

Six related studies were conducted with four captive juvenile Kit Foxes (*Vulpes macrotis*) to test the hypothesis that blood-squirting from eye-socket tissues by Texas Horned Lizards (*Phrynosoma cornutum*) is a canid antipredator defense. In 16 trials, naive “hungry” foxes killed and ate adult Yarrow’s Spiny Lizards (*Sceloporus jarrovi*; eight of eight trials) slightly more frequently than adult *P. cornutum* (six of eight trials). Adverse responses by foxes (head shaking) were seen in five of six trials in which *Phrynosoma* squirted blood. Later these experienced foxes, fed ad libitum, killed and ate mice (eight of eight trials) while largely ignoring *P. cornutum* (one killed and eaten in eight trials), suggesting a learned aversion to horned lizards as prey. During attacks on mice smeared with horned-lizard blood, foxes displayed behaviors typical of predatory encounters with horned lizard prey (head shaking and prey tearing). These prey-handling behaviors were in striking contrast to those elicited by untreated mice and by mice treated with mouse blood, demonstrating that horned-lizard blood (and its chemical constituents) altered normal behaviors toward mouse prey. Prey-handling times for mice treated with horned lizard blood were significantly longer than mouse-only treatments. Responses of foxes to mice coated with horned lizard Harderian- and lacrimal-gland tissues coupled with responses to mice coated with systemic horned-lizard blood, mouse blood, and untreated mice suggest that (1) no defensive chemicals are added to the blood by orbital glands before blood ejection, and (2) active antipredator chemicals are carried in the circulating blood as well as in squirted blood. In four trials, foxes attacked “de-horned” horned lizards; a role for cranial horns in facilitating predator hesitancy prior to blood squirting is proposed. Evidence is presented that horned lizards visually identify and categorize foxes as appropriate predators for a blood-squirting defense. We conclude that, in many predator-prey encounters with wild canids, blood-squirting by Texas Horned Lizards is an effective chemical defense. We propose a scenario for the evolution of this unique defense and suggest that the defensive compounds found in the blood may be sequestered from the seed-harvester ant prey of horned lizards.

USE of chemical defenses for repulsion of attacks by predators is well known in invertebrates and vertebrates (Berenbaum, 1995). The advantage of delivery of such compounds by prey to attacking predators prior to significant injury has been noted in geckos and salamanders (Rosenberg and Russell, 1980; Brodie and Smatresk, 1990). Some lizards of the genus *Phrynosoma* expel a stream of blood from blood sinuses around their eyes (Bruner, 1907; Burleson, 1942; Heath, 1966). Numerous and diverse hypotheses have been advanced to explain this unique behavior, including the possible use of this sprayed blood as an antipredator defense (Middendorf and Sherbrooke, 1992; Sherbrooke and Middendorf, 2001).

We have experimentally demonstrated a variety of variables in the blood-squirting response of *P. cornutum*, including frequency, repeatability, eyes involved, sex and age, and quantity of blood expelled related to body mass (Sher-

brooke and Middendorf, 2001) and have compared various chemical characteristics of expelled and systemic blood (Middendorf et al., 2001). We proposed that blood-squirting behavior involves the delivery of antipredator chemicals contained in the blood of some horned lizards to a specific category of prey, canids, and possibly other carnivores (Middendorf and Sherbrooke, 1992; Sherbrooke and Middendorf, 2001). Although current evidence supports the canid antipredator hypothesis, no wild canids have been reported to elicit blood squirting nor have any of their responses to horned-lizard blood, systemic or squirted, been recorded.

Criteria for accepting blood-squirting behavior as an antipredator defense against canids include that (1) it is elicited by the presence of, or attack by, a canid, and not by other potential predators, and (2) the discharge reduces predation. The second criterion remains untested

(Middendorf and Sherbrooke, 1992). Earlier and unpublished studies support the first criterion, in that aggressive actions of a domesticated dog (*Canis familiaris*) elicited blood squirting (70–100% of encounters) by *P. cornutum* (Middendorf and Sherbrooke, 1992), whereas attacks by other potential and known predators did not, such as Greater Roadrunners (*Geococcyx californianus*; Sherbrooke, 1990), Southern Grasshopper Mice (*Onychomys torridus*; Sherbrooke, 1991), Western Diamondback Rattlesnakes (*Crotalus atrox*), whipsnakes (*Masticophis* spp.), and Long-Nosed Leopard Lizards (*Gambelia wislizenii*; WCS unpubl.).

Here we evaluate responses of naive Kit Foxes (*Vulpes macrotis*), that opportunistically feed on reptiles (McGrew, 1979), to being defensively squirted by the blood of Texas Horned Lizards and responses of the same foxes to these lizards after becoming experienced. In addition, we examine the hypothesis that a chemical (or chemicals) serving as an agent of antipredator defense is added to the blood (prior to its expulsion) from glands surrounding the eyes and evaluate the potential of the lizard's horns to confound predators' responses or to facilitate targeted delivery of squirted blood. We also note responses of horned lizards to fox approach, prior to tactile contact and afterward, and evaluate the sensory modality used by lizards for predator categorization.

MATERIALS AND METHODS

Texas Horned Lizards were collected between 11 May and 2 July 1992 from Cochise County, Arizona, and Hidalgo County, New Mexico (Sherbrooke, 2002). They were maintained at the Southwestern Research Station, Portal, Arizona, in an aviary-wire screened (two separated layers) outdoor cage (3.8 × 7.0 × 2.4 m high) in which the lower 60 cm of the walls was covered with sheet metal. Lizards were fed live ants of the genus *Pogonomyrmex* spp. and commercially raised crickets (*Acheta domesticus*) and were sprinkler watered for "rain-harvest" drinking (Sherbrooke, 2003).

Seven Kit Foxes were live-trapped from a den in the Playas Valley, Hidalgo County, New Mexico, between 11 and 18 May 1992. Foxes were maintained at the Southwestern Research Station in the Chiricahua Mountains, 1645 m elevation. After stool samples revealed alimentary-tract parasites, foxes were treated with Panacur[®] (fenbendazole). Three of the foxes were adults, too timid for trials. They were subsequently released at site of capture. Four juveniles acclimated well to captivity and were used

in trials with Texas Horned Lizards, Yarrow's Spiny Lizards (*Sceloporus jarrovi*), and laboratory mice (*Mus musculus*). On 20–21 July, the four foxes were placed in adjacent individual cages (1.7 × 4.3 × 2.3 m high), where they were continuously housed, including during prey-encounter trials. They were fed chicken parts dusted with vitamin-mineral powder and recently sacrificed dead mice. In addition, live crickets and live mice were fed to ensure development of hunting skills. Lizards were only presented as potential food items during trials, thus maintaining the "naive" status of the foxes to these prey. Although the juvenile foxes became acclimated to the presence of observers, this presence may have inhibited some behaviors. During the trials of the six studies, observations were made from outside the trial enclosures (unless otherwise noted), either at the door or from a blind at the edge of the cages, or observers departed, returning later to determine the outcome of predator-prey encounters. As a result, prey-capture/feeding times are not complete for all trials (study 1).

In the first study, all four juvenile foxes were inexperienced at encountering live lizard prey. In 16 trials over four mornings (28–31 July; after 0600 h MST), each fox was daily offered a live lizard of one of two phrynosomatid species. The two species offered were *Phrynosoma cornutum* (A) and *Sceloporus jarrovi* (B), with the sequence of species offered being ABBA, foxes numbered 1 and 4, or BAAB, foxes numbered 2 and 3. Predator-prey encounters were observed and partially videotaped, and notes were made of blood squirting by horned lizards, fox reactions, and trial outcome to the potential prey. Following each trial, the foxes were fed a mouse that was immediately eaten, confirming their hunger status.

During the second study, and for four days prior to trials, each fox was provided with dry cat food (Science Diet[®]), ad libitum, a defrosted laboratory rat in the afternoon, and one or two large pieces of chicken (thigh or breast) each evening. The 16 trials (four foxes for four days) were conducted 25–28 August, at 0630–0700 h MST. In previous trials, foxes had been tested with a variety of live prey, such as Texas Horned Lizards ($n = 4$ per fox), mice ($n = 5$ per fox), and Yarrow's Spiny Lizards ($n = 2$ per fox). Horned lizards and mice were presented to the foxes in an ABBA or BAAB sequence. The initial 2 min of encounters were filmed, after which the observers departed from the trial cage area. Two hours later, cages were inspected to determine and record the results; lizards were inspected for evidence of blood squirting.

In the third study, in eight trials (two with each of four foxes; 1–2 August), the head pelage of eight live mice was coated with one of two liquefied tissues. In four gland-tissue tests, heads were coated with entire horned lizard Harderian- and lacrimal-gland tissues (macerated with mortar and pestle in a reptile-saline solution; NaCl 6.5 g/l, KCl 0.14 g/l, CaCl₂ 0.16 g/l, NaHCO₃ 0.2 g/l). In four systemic-blood tests, heads were rubbed into the thoracic cavity of a recently sacrificed Texas Horned Lizard (its arterial and venous supply to the heart had been severed), thus coating the pelage with systemic horned-lizard blood. The gland-tissue tests (A) and systemic-blood tests (B) were run in an AB or BA sequence.

In the fourth study, in eight initial trials, foxes were presented with live mice (4 per day; 13–14 August; 0930–1030 h MST) coated with either systemic horned-lizard blood (A) or uncoated mice (B) in an AB or BA sequence. Then, on 15 August, in four additional trials, all four foxes were presented with live mice whose heads had been smeared with fresh mouse blood. Both lizard and mouse blood were obtained as described above.

In the fifth study, because of the potential deterrent and confounding antipredator role of horns, each of the four foxes was presented (13–14 August; 0630–0900 h MST) with a “dehorned” horned lizard whose occipital and temporal horns had been filed off without causing bleeding. The four trial interactions were observed and videotaped for about 10 min before observers departed, returning an hour later to record the results.

In the sixth study, to investigate whether horned lizards visually classify potential predators, the responses of 28 lizards to the approach of the tamest fox (#1), in its trial/living cage, were examined (3 September; 1610–1730 h MST). The experimenter sat on the dirt floor of the cage and placed a puree of four adult crickets (squashed by mortar and pestle) on the back and head of each lizard while presenting the lizard to the fox. The experimenter was within 20–40 cm of the fox as it approached the lizard and gently removed the food morsels. The fox never attacked or attempted to bite the lizard. Notes and video analysis were made of the following lizard responses: (1) puffiness of eyelids, indicating filling of the ocular bloodsinuses (scored 0–4, increasing intensity from no response to full engorgement; see Middendorff and Sherbrooke, 1992); (2) squirting of blood; (3) tail lifting (scored 0–4, from no response to intense); and (4) raising or tilting of the back toward the fox.

RESULTS

Study 1: Predator-prey encounters between naive, “hungry” Kit Foxes and both Texas Horned Lizards and Yarrow’s Spiny Lizards.—All eight *Sceloporus jarrovi* were killed and eaten, whereas six of eight *P. cornutum* were killed and at least partially eaten. Although no significant differences in prey survival were observed between the two lizard species (Fisher’s Exact Test, $P = 0.233$), significantly fewer *Phrynosoma* (3) were consumed completely compared to *Sceloporus* (8) (Fisher’s Exact Test, $P = 0.013$). Both non-blood-squirting horned lizards were killed, whereas of the six that squirted, four were killed and two survived. Thus, both surviving *Phrynosoma* squirted blood. In five of six trials in which blood-squirting occurred, the foxes immediately shook their heads laterally in a vigorous manner, with intermittent jaw opening-closing sequences including tongue protrusions. Three horned lizards were killed and partially eaten, and the remaining three were completely eaten. Prey consumption time differences were due largely to handling; in general, *S. jarrovi* were immediately killed, severed into large pieces and rapidly swallowed (prey-capture/feeding times; mean = 66.3 ± 38.9 sec SD; range = 33, 39, 54, 55, 78, 139 sec), whereas *P. cornutum* were slowly torn apart into small pieces, and ingested piecemeal over longer time intervals (mean = 1288.2 ± 1023.8 sec SD; range = 372, 519, 750, 2400, 2400 sec).

Study 2: Behaviors of experienced and ad libitum-fed Kit Foxes to live Texas Horned Lizards and laboratory mice.—In spite of the availability of abundant and diverse nonliving food prior to and during the trials, in all cases (eight of eight) foxes killed and consumed the live mouse presented. In contrast, in only one of eight trials was a horned lizard killed and consumed. Fox feeding responses to horned lizards and mice differed significantly (Fisher’s Exact Test, $P = 0.013$). In the seven trials in which horned lizards survived, posttrial inspections of cages did not reveal evidence of injuries to lizards nor blood from defensive blood-squirting. In the single trial in which a lizard was killed, only the posterior half was eaten. Blood in the cage and on the lizard suggested it had attempted a blood-squirting defense from both eyes.

Study 3: Feeding responses of Kit Foxes to laboratory mice coated with either macerated Texas Horned Lizard Harderian- and lacrimal-gland tissues or horned-lizard systemic blood.—All four foxes killed and gulped gland-smeared mice in a normal mouse-prey fashion, in which a rapid approach and capture was followed by the quick use of,

mainly, carnassial teeth to kill and sever the mouse into two or three pieces, and then gulping of large pieces of mouse. No head-shaking behavior by the foxes was observed. In contrast, in the trials with systemic horned-lizard blood, foxes treated mice similarly to horned lizards that had squirted blood. Three of four foxes responded to the blood-smear mice with vigorous lateral head-shaking and all four held mice to the ground with a front paw while employing the incisor teeth to nibble at and shred the legs and tail, as well as extracting entrail contents prior to swallowing the stomach and intestines. This feeding process ("tearing") was previously seen with foxes killing and eating horned lizards. In these trials, the four foxes responded with significantly distinct feeding behaviors to mice coated with horned lizard gland tissue and to mice coated with systemic blood (Fisher's Exact Test, $P = 0.014$).

The two fox prey-handling behaviors of mice, gulping and tearing, also differed in the amount of time required to execute killing and consumption. Average and total times of the four gland-tissue trials (gulping) were as follows: mean = 53.8 ± 18.6 sec SD (range = 35, 41, 66, and 73 sec). For the four systemic-blood trials (tearing) times were: mean = 259.0 ± 277.4 sec SD (range = 80, 104, 182, and 670 sec).

Study 4: *Feeding responses of Kit Foxes to mice coated with Texas Horned Lizard blood, uncoated mice, and mice coated with mouse blood.*—All four uncoated mice were killed and consumed by the foxes in rapid fashion, involving use of carnassial teeth to section the prey into pieces and rapidly gulping these down (prey-handling time: mean = 36.3 ± 5.7 sec SD; range = 31, 33, 37, and 44 sec). Foxes killed and ate the mice coated with mouse blood in a fashion identical (gulping) to that used by the foxes to kill and eat uncoated mice (prey-handling time: mean = 36.3 ± 4.9 sec SD; range = 30, 36, 37, and 42 sec). In contrast, mice coated with horned-lizard blood were killed and eaten using the method of paw holding and tearing typically associated with attacks on horned lizards. Prey handling times were much longer (mean = 587.5 ± 123.8 sec SD; range = 428, 552, 673, and 697 sec). Fox prey-handling behavior of mice was significantly dependent on the presence of horned lizard blood, versus no blood or mouse blood ($\chi^2 = 12.0$, $P = 0.002$).

In summary, the eight mice with only mouse chemical information (uncoated mice and mice coated with mouse blood) were gulped. Foxes handling mice coated with horned-lizard blood tore them into small pieces, and, in three of four trials, exhibited head-shaking behavior. Di-

rection of prey ingestion also differed. Foxes swallowed the uncoated and mouse-blood-coated mice head section first, whereas mice coated with horned-lizard blood were eaten tail or thorax section first.

Study 5: *Feeding responses of Kit Foxes to "dehorned" Texas Horned Lizards.*—All four dehorned horned lizards were attacked, and of the three that squirted, two survived, although they were partially eaten. The only nonsquirting lizard was killed and partially eaten. In three trials, fox attacks focused on limbs and tails, and included tearing, behaviors typical of those seen in horned lizard predator-prey interactions with foxes. In the fourth, the fox, despite being squirted, ate head-first and exhibited behaviors more typical of mouse consumption.

Study 6: *Responses of Texas Horned Lizards to visual approach and contact by a Kit Fox.*—Although no eye puffiness or bulging, or other responses were noted when the prepared puree of cricket was placed on the head and back of the lizard, all 28 lizards responded to the approach of the fox, some in more than one way. Prior to contact, four lizards exhibited initial stages of eye puffiness, five exhibited tail-lifting, and two raised or tilted their backs toward the fox. Within five seconds of contact 22 lizard subjects exhibited various degrees of eye puffiness (puffiness response index: mean = 3.2), 20 lifted tails (tail-lift response index: mean = 2.0), and 14 tilted or raised their back toward the fox. Five of the lizards even squirted blood; all were of small amounts, single squirts, and from a single eye.

DISCUSSION

In initial trials, naive Kit Foxes, inexperienced with Texas Horned Lizards and Yarrow's Spiny Lizards, attacked them as prey. Responses of foxes to the two prey species differed in terms of fox prey-handling technique and handling times, and adverse reactions (head-shaking; Study 1). When no-longer naive foxes (fed ad libitum) were offered familiar prey items in the form of mice and horned lizards, they clearly distinguished between the two prey species, with clear survival value to horned lizards (Study 2). These data suggest an inverse relationship between predator hunger levels and survivorship of horned lizards as potential prey of foxes. We are aware of only one account of canid consumption of a blood-squirting species of horned lizard (two Texas horned lizards; see Middendorf and Sherbrooke, 1992), although Duncan et al. (1994) reported possible Kit Fox preda-

tion on a "non-blood-squirting" species (Sherbrooke and Middendorf, 2001).

The head-shaking reaction by foxes to blood squirting (not seen in *Sceloporus* trials nor in nonsquirting horned lizard trials; Study 1) and to horned-lizard blood on mice (Studies 3 and 4) suggests that foxes respond adversely to horned-lizard blood. Also, different prey types (Studies 1 and 2) elicited very different prey subjugation and handling behaviors. Horned-lizard blood, on a mouse, clearly modified and lengthened prey-handling behavior and time by foxes (Studies 3 and 4). Further, fox feeding methods and prey-handling times used with gland-treated mice (Study 3) were identical to those observed with uncoated mice and mice coated with mouse blood (Study 4). These data refute the hypothesis of Burleson (1942), Heath (1966), and Cowles (1977) that Harderian- or lacrimal-gland secretions are added to horned-lizard systemic blood when squirted through orbital tissues and that these compounds serve as a chemical deterrent to predation.

The sharp cranial horns of horned lizards have been implicated as structures used during antipredator defense, especially for predators such as snakes that swallow their prey whole (Sherbrooke, 2003). Our earlier results suggest that the horns play a minor role in interactions with canids (Middendorf and Sherbrooke, 1992). Trials with de-horned horned lizards resulted in attack and feeding behaviors by the foxes that were not dissimilar to those seen with "horned" horned lizards, except that in one fatal case the head was atypically consumed before the carcass. Apparently, Texas Horned Lizard cranial horns do not confer significant antipredator defense, by themselves, with Kit Foxes. Nevertheless, horns may play an important role with these predators in facilitating efficient blood-squirting by inducing hesitancy by predators to initiate carnassial cutting of the head, which may result in rapid crushing of the cranium. Such a time delay of potentially lethal prey-subjugation methods provides opportunity for effective employment of a blood-squirting defense.

Like predators, prey exhibit choices during encounters, such as initial recognition (and classification) of a potential predator, decisions as to how to respond, and subsequent reevaluation (Sherbrooke, 2003). Horned lizards responded to the approach of a fox, exhibiting anticipatory antipredator-defensive responses, even prior to contact. Our observations support visual identification and categorization of predators, especially given that two of the behaviors exhibited toward the fox, eye puffiness and tail-

lifting, are not known to be exhibited toward other predator types (Sherbrooke, 2003). The idea of a taxonomically limited role for these antipredator chemicals (Middendorf and Sherbrooke, 1992) is supported by the lack of strong negative responses by humans, a primate, to squirted blood when tasted (Hay, 1892; Cowles, 1977; WCS and GAM, pers. tasting [20+]). Future testing of the responses of various predators to systemic blood samples (now known to potentially carry the antipredator chemicals) of different species of *Phrynosoma* may elucidate the taxonomic range of activity among predators of this antipredator defense, and determine the relative effectiveness of the blood-borne chemicals in the "non-blood-squirting" and blood-squirting species of horned lizards (Sherbrooke and Middendorf, 2001; Sherbrooke and Mendoza-Quijano, in press; Sherbrooke et al., in press; W. C. Sherbrooke and J. R. Mason, unpubl.). The absence of barking or growling during attacks by foxes on horned lizards suggests that auditory cues were not important in predator identification. Apparently, olfactory cues from dog saliva are also unimportant (Middendorf and Sherbrooke, 1992).

Defensive behaviors are likely to increase survivorship if they are tailored to appropriate predator threats. Tail lifting by horned lizards appears to increase the likelihood of tail-biting and tossing by canids. Tail biting by foxes may follow unsuccessful cranial bites on Texas Horned Lizards (pers. obs.). Like eye puffiness and blood squirting, it has only been observed in dog, Kit Fox, and Coyote (*Canis latrans*; W. C. Sherbrooke and J. R. Mason, unpubl.) trial encounters. The likelihood of a lizard being lost by a canid following a tail-biting toss, with little directional control, is increased if the lizard subsequently remains motionless, a common horned lizard strategy (Sherbrooke, 2003).

When a canid bites a horned lizard's head (richly endowed with mechanoreceptors, including along the eyelids; Sherbrooke and Nagle, 1996), the lizard, having already visually identified and categorized the predator, is ready to respond to that tactile stimulation by squirting blood into the predator's mouth (see photographs in Middendorf and Sherbrooke, 1992; Sherbrooke, 2003), causing a negative feeding response by the canid and increasing the lizard's potential for survival. Successful use of blood squirting as an antipredator defense requires proper identification of predators prior to its use and appropriate prey and predator responses. Twenty-two of 28 horned lizards engorged blood sinuses or squirted blood before, or within 5 sec, of contact by a Kit Fox. Seventy-

five percent of challenged horned lizards tested in Studies 1 and 5 (nine of 12) squirted blood at foxes, similar to the percentage that squirted at a dog (Middendorf and Sherbrooke, 1992; Sherbrooke and Middendorf, 2001), and about 25% of these survived.

The observation that under certain conditions a canid predator will kill and consume a horned lizard (containing 6% blood by weight; Sherbrooke and Middendorf, 2001) does not negate the fact that delivery of blood-containing antipredator compounds directly to oral surfaces (the target area of blood squirting; W. C. Sherbrooke and J. R. Mason, unpubl.; W. C. Sherbrooke and B. A. Kimball, unpubl.) is a different, unpleasant gustatory experience for the predator. Under blood-squirting conditions, the concentration of these compounds in the predator's mouth is much higher, and other flesh-derived compounds that elicit positive-taste responses do not dilute or counteract them.

The fact that circulating horned-lizard systemic blood is used as an effective antipredator agent brings to question the possible source of these chemical components in the blood. They might be derived from their diet, largely ants (Pianka and Parker, 1975). Sequestered dietary-source compounds are known to be employed, with minimal chemical modification, in the defensive armament of monarch butterflies and nudibranch molluscs (Brower, 1984; Edmunds, 1984; Daly, 1995). Among terrestrial vertebrates, dendrobatid and mantelline frogs derive alkaloid defensive compounds from their ant prey (Daly, 1998; Jones et al., 1999; Spande et al., 1999), as may be the case for plumage chemical defenses in some birds (Dumbacher, et al., 2000; Weldon, 2000).

Schmidt et al. (1989) demonstrated that systemic blood plasma of *P. cornutum* can detoxify the venom of a harvester ant (*Pogonomyrmex maricopa*). Harvester ants are important in the diets of horned lizards (Pianka and Parker, 1975; Sherbrooke, 2003). The venom of ants of this genus is highly toxic to vertebrates (Schmidt et al., 1989). The possibility of a link between the antipredator-defensive role of horned-lizard blood squirting and the ability of horned lizard systemic blood to detoxify the venom of *Pogonomyrmex* prey suggests the hypothesis that the two roles of blood chemistry may be linked. If blood-plasma compounds bind with ant venom to neutralize its effects (Schmidt et al., 1989), the resulting compounds could be distasteful to canids. Or, the two blood-chemistry issues might be chemically unrelated, but compounds sequestered from an ant diet

might still enhance the antipredator utility of squirted blood.

Previously, Hay (1892) suggested that horned lizard blood squirting startled or distracted a predator. Although possibly true, we believe, at best, it plays a minor role with foxes. Our study demonstrates that Kit Foxes react negatively to a chemical constituent, or constituents, of the blood of Texas Horned Lizards, even when delivered in the context of mice prey. Our studies strongly support the hypothesis that horned-lizard blood squirting is an antipredator defense against canid predators, increasing the lizard's chances of surviving potentially lethal encounters.

Evolution of this unique (pressurized propulsion of ocular-sinus blood from the eye orbit) antipredator system, limited to species of horned lizards, may have come about as a result of incorporation of chemicals into the lizard's circulating blood, possibly from ant prey, that cause a negative gustatory response in a predator. Subsequently, an evolutionary transition was facilitated by minor modification of a preexisting mechanism, used by lizards to increase cranial and ocular-sinus blood pressure, to allow controlled, external squirting of blood (Bruner, 1907; Heath, 1966). Oozing of blood from orbital membranes during stress has been noted in related lizards, illustrating the existence of an intermediate mechanism for delivery of blood chemicals to a lizard's exterior prior to serious injury (Sherbrooke, 2000). This system was improved by natural selection to enable timely and forceful delivery of blood-borne antipredator compounds directly to oral surfaces of selected predators immediately prior to damaging biting attacks, thus enhancing lizard survival.

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