

SHORT COMMUNICATION

Antipredator responses of *Hadrurus arizonensis* (Scorpiones: Caraboctonidae) to chemosensory cue from a mammalian predator

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Abstract. Sensory ecology studies show that reception and utilization of information from the environment is a crucial life process. Scorpions possess a weapon that can be used against predators, but it remains unknown whether scorpions' decision to use it is influenced by chemical cues from predators. We investigated the influence of predators' odors on stinging behavior of *Hadrurus arizonensis* (Ewing, 1928) by stimulating them to sting under two conditions: in the presence of an odor from a potential rodent predator (*Rattus norvegicus*) and in the absence of such an odor. It took fewer probes to elicit a response when predator scent was present, and it resulted in more wet stings than the non-scented treatments. Finally, the smaller scorpions were more reactive than the larger ones. The variances in stinging behavior suggest that the detection of predator odors by *H. arizonensis* elevates its response in potentially threatening circumstances.

Keywords: Arthropod, kairomone, defensive stinging, chemoreception, risk assessment

Antipredator decision-making has received much attention in ecology during the recent decade. Due to the unforgiving nature of predation, preyed organisms are under pressure to readily detect and avoid predators (Lima & Dill 1990; Lima 1998a,b). Many organisms detect and avoid predators from the use of multiple sensory inputs, including visual cues, substrate-borne vibration, and chemical or tactile stimulations.

Predators, in turn, emit a myriad of sensory cues that are detectable by preyed species. Some sensory information is immediate and direct (e.g., visual and auditory), whereas other cues may be indirect and asynchronous (e.g., chemical). The utilization of direct and indirect cues is rather common in arthropods, as the phylum is characterized by tremendous diversity in mechanisms for detection of visual, chemical, and mechanical stimuli (Bate et al. 1978; Root 1990).

Predation is a powerful, threatening force that shapes the behavior and morphology of prey. Many species respond to the odors of their predators and adapt their behaviors according to the source of the odor (Kats & Dill 1998). Predator avoidance is probably the most efficient way to prevent predation. A number of studies have highlighted increased avoidance behavior as a defensive response while under exposure to predator odors. For example, prey often decrease their activity or use different spatial or temporal refuges to decrease their vulnerability to predators (see review by Lima 1998a).

Scorpions possess a large assortment of mechano- and chemoreceptors that provide them with information from their environment (Brownell & Farley 1979a,b,c; Brownell & van Hemmen 2001). For example, trichobothria react to horizontal air streams and possess directional sensitivity (Hoffman 1967). Pectines appear to be involved in chemically-mediated orientation behaviors such as mate recognition and possibly localization of water (Gaffin & Brownell 1992; Gaffin et al. 1992). Fingers of the pedipalps possess a constellation-shaped microscopic array of sensilla that are thought to be involved with chemoreception (Fet et al., 2006a,b; Nisani et al. 2018). However, there is still little information on scorpions using chemosensory cues for predator avoidance. A few studies have suggested that some species may use substrate-borne pheromones to aid in the recognition of female conspecifics (Melville et al. 2003) or to avoid areas exposed to direct cues from conspecific males (Miller & Formanowicz 2010).

Among scorpions, *Parabuthus transvaalicus* Purcell, 1899 has been shown to regulate defensive venom expenditure based on perceived

risk (Nisani & Hayes 2011). However, no study has investigated scorpion stinging behavior in response to predators' chemical cues. The goal of this study was to document whether the odors from a predator might influence the defensive behavior of *Hadrurus arizonensis* (Ewing, 1928). We experimentally tested two hypotheses: (1) that scorpions would become more defensive when exposed to the odors of a potential mammalian predator (*Rattus norvegicus*); and (2) that juvenile scorpions, being more vulnerable to predation, would be more defensive than adult scorpions.

Hadrurus arizonensis were collected during mid-July and August at a single location in Lancaster, California (34°38'36" N, 118°11'38" W), using UV light. They were housed individually in clear plastic containers measuring 31 × 19 × 12 cm (L × W × H), with all containers having a sand substrate and a wet sponge. Scorpions were kept at 21–23°C and 55–60% relative humidity under a 12:12 h light:dark cycle. They were fed an appropriate-sized cricket once a week. In order to avoid possible variation due to sex, only female scorpions were used in this study. Large scorpions weighed 5.66 ± 1.37 g (mean ± S.D.) with a range of 3.23–7.93 g ($n = 17$) and small scorpions weighed 1.15 ± 0.23 g with a range of 0.69–1.49 g ($n = 13$). Scorpions from each weight class were randomly assigned and tested twice, once in absence of odor (control) and once in presence of odor (odor treatment) with minimum intertrial interval of 7 days.

To collect odor cues, a filter paper (20 × 20 cm) was placed in the rat (*Rattus norvegicus*) cage for 4 days. This odor source was selected because rodents have been shown to be predator of scorpions (Polis et al. 1981). The soiled filter paper was then transferred (but separated by a wire mesh to prevent contact) into a Ziploc® bag containing a clean piece of filter paper for another 4 days. The procedure was repeated for each trial with the presence of the predator's odor. For the control trial, a clean piece of filter paper was left in a Ziploc® bag for 4 days and removed prior to testing. For all trials (controlled and uncontrolled), the filter paper was only used once then discarded and the experimental arena was cleaned with alcohol, followed by water and then allowed to dry.

Each scorpion was transferred individually to a 150 ml glass beaker and moved to the plastic experimental arena measuring 50 × 24 × 5 cm with appropriate filter paper (control or odor treated) placed in the center of the arena and illuminated with a fiber optic illuminator. Scorpions were guided into the beaker without any physical contact to their bodies. After the scorpion had acclimated in the arena and

Table 1.—The frequency of defensive responses of *Hadrurus arizonensis* scorpions in relation to presence or absence of potential mammalian predator odor (*Rattus norvegicus* urine). See text for description of responses.

	Wet sting	Dry sting	Jab	No sting
Large Scorpion				
Control	2	3	5	7
Odor treatment	11	2	4	0
Small Scorpion				
Control	0	12	1	0
Odor treatment	10	2	1	0

was on top of the filter paper for 5 min, we provoked it to sting by gently touching its dorsum (1 probe per second) with the edge of a round, parafilm-covered plastic cup (2 cm high \times 4.5 cm diameter). The cup was presented using a pair of forceps, 29 cm long. We recorded the number of probes necessary to provoke a response and the type of each response (i.e., no sting, jab, dry sting, wet sting, or retreat). A response would qualify as a “jab” if the top of the scorpion’s telson made contact with the parafilm without the aculeus actually puncturing the parafilm. A “sting” was recorded when the aculeus pierced the parafilm and, if venom was secreted, it was recorded as a “wet sting”. If the scorpion ran off the filter paper, it was characterized as a “retreat”.

All scorpions in the trial eventually retreated after either jabbing or stinging the parafilm. It took the larger scorpions on average (\pm S.D.) 3.59 ± 3.04 (control) compared to 1.82 ± 1.07 (odor treated) probes to elicit a response. This difference in response was statistically significant, Mann–Whitney $U = 85$, $P < 0.05$ two-tailed. The incidence of wet stings was 65% higher in the presence of a predator odor, while a majority of larger scorpions (41%) did not sting during control treatment (Table 1). It took the smaller scorpions an average of 3.15 ± 5.13 probes compared to 1.08 ± 0.28 probes to induce a response in the control versus the odor treated case, respectively. This difference was significant (Mann–Whitney $U = 38$, $P < 0.05$ two-tailed). Smaller scorpions stung in both treatments with the majority of stings (77%) in the odor treatment being wet while all stings to control treatment were dry (Table 1). Comparatively, it took smaller scorpions fewer probes to induce a response than in larger scorpions (Mann–Whitney $U = 50$, $P < 0.05$ two-tailed) in the odor treated trials, while there was no statistical difference between large and small scorpions in the control trials (Mann–Whitney $U = 75$, $P < 0.144$ two-tailed).

Overall, scorpions reacted after fewer probes and delivered more wet stings in the presence of predator odor compared to the control condition. The smaller scorpions, presumably more vulnerable to predators, tended to be more reactive, as they stung (dry in control, or wet in odor treatment) more often compared to larger scorpions. This study seems to support the assertion that scorpions are more defensive when exposed to predator odor and will sting more often when stimulated.

Scorpion risk assessment studies have shown that *P. transvaalicus* are more defensive and inject more venom under higher threatening climate compared to a low-threat situation (Nisani & Hayes 2011). Chemoreception is well developed in scorpions (Gaffin & Brownell 1992; Melville et al. 2003; Steinmetz et al. 2004; Taylor et al. 2012), but much remains to be learned about the structures, contexts, and behaviors involved. Kats & Dill (1998) reviewed the responses of prey species to predator cues, with increased defensive responses in arthropods, reptiles and mammals being among them. For example, a New Zealand freshwater crayfish (*Paraneohpops zealandicus*) exhibits a significantly greater number of defensive chela displays when exposed to chemical cues from the skin mucus of eels (Shave et al. 1994). Other studies have shown that crotaline snakes will increase

biting in response to snake predators (Cowles 1938; Meade 1940). Finally, kairomones from another parasitoid increased the incidence of probing and stinging by the *Eupelmus vuilleti* parasitoid during kleptoparasitism, however this study was not performed in a defensive context (Jaloux & Monge 2006). To date, this is the first study to document variability in stinging behavior of a scorpion in relationship to predators’ odor.

Many antipredator strategies have costs associated with them; thus, one would expect that organisms would have evolved an accurate adaptation for that risk. This is especially important when it comes to venom expenditure, as it has been shown to be metabolically expensive (McCue 2006; Nisani et al 2007). It remains to be investigated whether scorpions respond specifically to the chosen predator’s odor or if they react to scent in general, and if they modulate the quantity of venom delivered in accordance to the level of perceived threat due to the scent. Despite these questions, we believe that this study sheds further light on this fascinating aspect of the biology of scorpions and hope this will encourage further investigations.

ACKNOWLEDGMENTS

This research was conducted as part of Antelope Valley College Undergraduate Research Initiative (AVC-URI). This project was performed by Raul Curiel as part of the AVC-URI, and was supported by United States Department of Education grant P031C110009-13.

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Manuscript received 30 August 2018, revised 7 August 2019.