



Testing the Threat-Sensitive Hypothesis with Predator Familiarity and Dietary Specificity

Brian A. Crawford*†, Caleb R. Hickman*‡ & Thomas M. Luhring*§

* Savannah River Ecology Laboratory, Aiken, SC, USA

† Warnell School of Forestry and Natural Resources, University of Georgia, Athens, GA, USA

‡ Department of Zoology, University of Wisconsin, Madison, WI, USA

§ Division of Biological Sciences, University of Missouri, Columbia, MO, USA

Correspondence

Brian A. Crawford, Savannah River Ecology Laboratory, Drawer E, Aiken, SC 29802, USA and Warnell School of Forestry and Natural Resources, University of Georgia, 180 E Green Street, Athens, GA 30602, USA.
E-mail: bcrawfor@uga.edu

Received: January 8, 2011

Initial acceptance: March 31, 2011

Final acceptance: October 4, 2011

(D. Zeh)

doi: 10.1111/j.1439-0310.2011.01983.x

Abstract

In a system with multiple predators, the threat-sensitive predator avoidance hypothesis predicts that prey respond differently to predators relative to the risks each poses (e.g., degree of dietary specialization). Aquatic animals often rely heavily on detecting predators via chemical cues (kairomones) and respond with a suite of behaviors including detection and avoidance. However, little is known about how animals respond to kairomones of specialist versus generalist predators. In laboratory experiments, we compared behavioral responses of a poorly studied aquatic salamander, the greater siren (*Siren lacertina*), to cues from specialist and generalist predator snakes to evaluate threat-sensitive responses. Sirens exhibited a novel behavior (gill-flushing) most often in the presence of specialist predator cues. Avoidance behavior (reversing direction following cue detection) was higher in response to specialist predator and novel animal control cues and lowest in response to generalist predator cues. An intermediate response to the animal control, an unfamiliar amphibian predator, indicated that sirens respond cautiously to a novel cue. The gradient of observed responses to different snake cues indicates that sirens may be evaluating predation potential of animals based on their foraging specificity and familiarity.

Introduction

Predation risk can be abated by an animal's ability to detect and avoid potential predators (Endler 1986; Lima & Dill 1990; Brodie et al. 1991). Prey animals experience chronic interactions with potential predators that can vary in their relative predation pressures (Sih 1987; reviewed by Sih et al. 1998). While avoidance of a threat ensures safety for the animal, this response comes with potential costs, as reducing activity or seeking refugia may impair fitness-gaining activities such as foraging or reproduction (Petranka 1989; Kats & Dill 1998). Thus, selection should induce plastic responses in prey animals that discriminate between different threat levels of predators

(Chivers & Mirza 2001). The threat-sensitive predator avoidance hypothesis predicts that the cost of predation avoidance can be ameliorated by a measured response that is proportional to the relative threat that a potential predator represents (Helfman 1989).

Different potential predators signify varied levels of threat according to their identity, size, and diet specialization (reviewed by Lima & Dill 1990; Madison et al. 1999). In comparing predators of different diet specializations, specialists are predicted to be more efficient and successful at obtaining their particular prey items (reviewed in Via 2001; Ferry-Graham et al. 2002), while generalists seem to be less efficient hunters with fewer behavioral and

morphological specializations (e.g., Drummond 1983; Mori & Vincent 2008). For example, Drummond (1983) directly showed that a piscivorous specialist garter snake (*Thamnophis couchi*) caught fish more effectively than closely related generalists (*T. sirtalis* and *T. elegans*). From findings like these, specialists may represent greater predation pressures than generalists on shared prey and should be consistently avoided (Sullivan et al. 2005). Although many studies have found correlations between prey response and predation risk (reviewed by Sih 1987; Chivers & Mirza 2001; Relyea 2001b), few studies have examined whether animals will respond differently to predators of different foraging specificities (Ferrari et al. 2007a). Prey can provide a clue to the relative importance of predators to ecological interactions via behavioral responses (Snyder & Wise 2000).

Chemical cues emitted by predators ('kairomones') can be used by prey species to identify and evaluate the risk posed by a potential predator (reviewed by Chivers & Smith 1998; Kats & Dill 1998). Animals that live in low-light or structurally complex environments often rely on kairomones more heavily than visual or tactile cues to detect predators (Stauffer & Semlitsch 1993; reviewed by Kats & Dill 1998; Mathis & Vincent 2000). In aquatic environments, kairomones diffuse through visual barriers (e.g., vegetation) and can be detected earlier than visual or tactile cues (Lima & Dill 1990). In addition to low visual acuity and physical barriers, aquatic amphibians may find chemosensory cues important to detect cryptic predators in clear habitats (Hickman et al. 2004). When predator cues are detected, amphibians in general (e.g., Brodie et al. 1991; Mathis & Vincent 2000; Mathis et al. 2003) – and aquatic amphibians in particular (e.g., Relyea 2001a; Epp & Gabor 2008) – demonstrate various response types such as increased detection behavior, cue avoidance, increased use of refugia, reduced activity, and flight (reviewed by Kats & Dill 1998). Increased ventilation via gill movement has been observed in fish as a detection response to predator and novel animal chemical cues (Gibson & Mathis 2006). Although not previously studied, other gilled vertebrates, such as aquatic salamanders, may also respond to kairomones with increased ventilation.

For our model system, we used a large, obligately aquatic salamander, the greater siren (*Siren lacertina*), that commonly co-occurs with an aquatic salamander specialist, the mudsnake (*Farancia abacura*) and a generalist predator, the banded watersnake (*Nerodia fasciata*). We used a controlled laboratory setting to test for threat-sensitive responses of greater sirens to

kairomones of two predators (specialist mudsnakes, generalist watersnakes) sympatric to each other and siren, a non-sympatric amphibian predator, the eastern hognose snake (*Heterodon platirhinos*), as a novel animal control, and dechlorinated tap water as a blank control. When presented with kairomones from these different sources, we hypothesized that sirens (1) detect predator kairomones and (2) respond to the presence of predator kairomones with intensity relative to predation risk. Specifically, we expected increased detection and avoidance behavior in the presence of specialist predator cues, relative to generalist predator and novel animal control cues.

Materials and Methods

Model Prey and Predators

Greater sirens, an obligately paedomorphic species, are cryptic and nocturnal salamanders that inhabit a variety of sluggish, murky, and heavily vegetated Coastal Plain wetlands of the Southeastern United States (Duellman & Schwartz 1958). Of ten of the currently recognized salamander families, at least seven are known to respond to a chemical cue of some sort (reviewed in Kiemnec 2009), suggesting that chemosensory detection is ancestral to the caudate clade. There has only been one previous study on the chemosensory abilities of sirenids, considered the most basal group of caudates (Zhang & Wake 2009), which found no evidence that lesser sirens (*S. intermedia*) use chemical cues for food detection (Sullivan et al. 2000). However, the authors cautioned that the types and concentrations of prey chemical cues used in the study may have been insufficient to elicit a response from sirens. Thus, it is still unknown whether sirenids can detect chemical cues of any kind, including predator kairomones.

While there are several known and likely predators of sirens (reviewed by Luhring 2008), mudsnakes (*F. abacura*) are often sympatric with sirenids (Jensen et al. 2008) and are known specialists of aquatic salamanders, including sirenids (Gibbons & Dorcas 2004; Jensen et al. 2008). Banded watersnakes (*N. fasciata*) are known to consume a variety of animals, including sirens (TML, pers. obs.). However, the majority of *N. fasciata* diets are comprised of fishes and anurans (Mushinsky et al. 1982). The novel animal control chosen for this experiment, eastern hognose snake (*H. platirhinos*), is terrestrial and specialist predators of toads (*Bufo* spp.) and therefore unlikely to encounter or consume siren. We used hognose kairomones in this study as a

control for chemical cues that may be emitted by snakes in general and compared siren response to two specific predators with this ecologically and phylogenetically related control.

Collection, Husbandry, and Stimulus Preparation

Sirens were collected from three wetlands in Aiken and Barnwell counties, South Carolina, USA. Animals were captured between Jun. 16 and Jul. 2, 2008 using steel and plastic minnow traps, hoop nets, and trashcan traps (Luhring & Jennison 2008) to ensure a broad range of animal size, age, and mass (TML, unpubl. data). Sirens were housed individually at 22°C under a 12 h light:12 h dark cycle. Sirens were fed earthworms (*Lumbricus terrestris*) weekly, and water was changed once a week.

Predators (mudsnakes and watersnakes) were captured in the same localities as sirens, housed for a short time to collect chemical cues, and then promptly returned to their site of capture. Several species respond to chemical cues from predators that have recently eaten a conspecific (reviewed by Chivers & Mirza 2001), and prey may even respond to unfamiliar predators when these diet cues are associated (Chivers & Smith 1998). Because cues from recent meals may detract from the predator-specific cues we seek to address (Madison et al. 1999; Ferrari et al. 2007a), we only used snakes that were devoid of prey items and then housed for 1 wk before collecting the stimulus (to allow any residual diet cues to dissipate). The animal controls (hognose snakes) were long-term captives maintained by the Savannah River Ecology Laboratory and fed a non-amphibian diet of *Mus musculus*, which controlled for possible amphibian diet cues. Stimulus was collected from animal controls 1 wk after their last meal.

We collected stimulus from specialist predators (*F. abacura*, $n = 3$, \bar{x} mass \pm SD: 515.33 ± 335.01 g), generalist predators (*N. fasciata*, $n = 4$, \bar{x} mass \pm SD: 69.74 ± 45.09 g), and unfamiliar animal controls (*H. platirhinos*, $n = 2$, \bar{x} mass \pm SD: 415 ± 12.73 g). All snakes were individually placed in separate 18.9-l aquaria with an amount of dechlorinated tap water equaling 2 ml/g of animal mass. We standardized the amount of water per gram mass of snake as an effort to standardize cue concentration because species differ in size, and cue concentration is known to influence prey behavior of predators in aquatic environments (Ferrari et al. 2008). All stimulus aquaria were covered and allowed to sit for 72 h. After this period, snakes were removed, and the stimulus water was filtered through cheesecloth to

exclude coarse material and stored at -12°C in labeled Whirlpak bags (Nasco, Fort Atkinson, WI, USA). Freezing stimulus does not affect cue detection (see Hickman et al. 2004). To control for cue variation in individuals, this process was performed with multiple snakes for each species and mixed throughout the trials. To obtain blank control stimulus, we set up an aquarium identical to stimulus aquaria with 2 l of dechlorinated tap water and allowed it to sit for the same 72-h period.

Experimental Design

We performed trials in two sessions with a different observer in each because of time and material constraints. For each session, conditions and procedures were identical, and observers calibrated before trials began to standardize the scoring of behaviors (observations were not significantly different between sessions – see Results section). The first session compared responses of sirens ($n = 46$) to cues from a blank control, a novel animal control (hognose snake), and a specialist predator (mudsnake). The second session compared the responses of sirens ($n = 27$) to the same blank and animal (hognose snake) control cues with cues from a generalist predator (watersnake). The snout-vent length (SVL) of sirens used in trials averaged 205 mm (range 92–430 mm). For each trial, we randomly selected a siren, a kairomone treatment, and the side of the tank the stimulus was introduced, and each siren was used once for the entire study, receiving one treatment in one experiment. Trials were conducted between Jul. 7 and Aug. 5, 2008 in the same room used for housing test animals. Individuals were tested in rectangular aquaria (120 \times 30 cm) that contained 6 l of well water. To aid in observation, a 20-W fluorescent bulb was suspended over aquaria. Previous to test trials, laboratory and test conditions did not appear to influence normal feeding behaviors (CRH, pers. obs.); therefore, conditions were assumed to be appropriate for testing behavior.

Prior to testing, sirens were gently placed into individual aquaria and allowed to acclimate for ten minutes. During water changes and feeding events, sirens generally resumed exploratory behavior within five to ten minutes of being placed in a new container. Exploratory behavior was similar to foraging movement but much different than the faster and erratic movements that sirens made when physically disturbed (CRH, pers. obs.). To accurately position the stimulus in the aquaria, we prepared four stimulus injection apparatuses (SIA's), each

composed of a 30-ml syringe connected to polyethylene tubing and then mounted on a bamboo splint 0.5 m in length (Mathis et al. 2003; Hickman et al. 2004). Stimulus treatments and SIA's were coded for individual stimulus types and kept blind to the observer. Syringes were filled with 30 ml of stimulus and allowed to reach room temperature. Three drops of food coloring were then added to the SIA so that the observer could visualize the stimulus and determine when the animal came into contact with it. Stimulus was injected into randomly assigned ends of the rectangular tank at a rate of approx. 1 ml/s, and behavior events were recorded for 5 min. Stimulus was slowly thawed and kept on ice for each test to limit degradation (see Hickman et al. 2004). Aquaria were drained and thoroughly cleaned between trials with animal-safe soap (Alconox[®], Alconox Inc., White Plains, NY, USA) and hot water.

A laptop computer was set up facing the aquaria so that the observer could score the animal's behavior using the JWatcher program (Blumstein et al. 2006). Two main behavioral events were recorded: gill-flushing and reversing. Gill-flushing occurred when sirens raised their heads, extended, and pulsed their gills as the mouth was opened and closed to push water through the gill openings. We recorded reversing behavior as an avoidance response to chemical cues, during which the siren's head is initially turned away from the stimulus and tail curled forward to pull the anterior of the body backwards in an S-shape. Ward & Azizi (2004) described a similar escape response, called head retraction, in *Siren intermedia* and other elongated aquatic species. During our trials, head retraction always preceded movement away from the stimulus. Because of the limited size of the test chamber, active sirens often contacted the stimulus multiple times and responded with gill-flushing and/or reversing behaviors multiple times within the 5-min observation period.

Statistical Analyses

We used a generalized linear mixed effect model (GLMM) with a negative binomial distribution and log link function for counts of each behavioral response (gill-flushing and reversing) followed by a Tukey–Kramer adjustment for multiple comparisons (PROC GLIMMIX in SAS, v9.2; SAS Institute Inc., Cary, NC, USA). We modeled cue and time as fixed effects and time between experiments as a random effect which can account for both type I and II errors (Schielzeth & Forstmeier 2009). If time was not significant as a fixed effect, it was removed as a covari-

ate and the model was run with cue as a fixed effect alone (Bolker et al. 2009). These models fit the data well with reasonable under dispersion for gill-flushing and overdispersion for reversing (deviance/df = 0.96 and 1.19, respectively).

Results

For the gill-flushing behavioral response, main effects for time ($F_{1,47} = 0.07$, $p = 0.80$) and the interaction between cue and time ($F_{1,47} = 0.14$, $p = 0.71$) were not significant, supporting that pre-experiment calibration eliminated observer bias. Therefore, we tested cue as a fixed effect and time as a random effect for gill-flushing behavior. There was a significant main effect of cue on the number of gill-flushes exhibited by sirens ($F_{3,49} = 13.09$, $p < 0.001$; Fig. 1a). Pairwise comparisons revealed that sirens gill-flushed significantly more when presented with

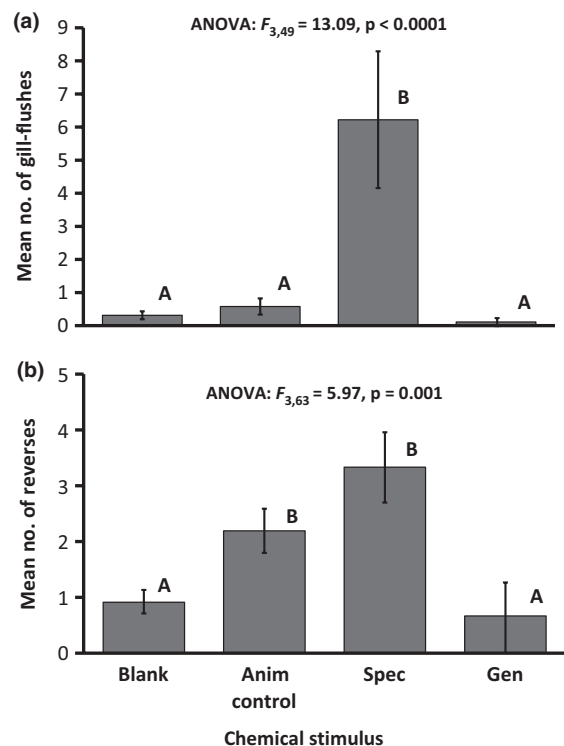


Fig. 1: (a) Mean (± 1 SE) number of gill-flushes by siren in response to chemical stimulus from a blank control ($n = 16$), animal control hog-nose snake ($n = 19$), predatory specialist mudsnake (*Farancia abacura*) ($n = 9$), and predatory generalist watersnake (*Nerodia fasciata*) ($n = 9$). (b) Mean (± 1 SE) number of reverses by siren in response to chemical stimulus from a blank control ($n = 23$), animal control hog-nose ($n = 26$), predatory specialist mudsnake (*Farancia abacura*; $n = 15$), and predatory generalist watersnake (*Nerodia fasciata*; $n = 9$). Letters indicate significant differences among groups using a Tukey/Kramer *post hoc* comparison ($p < 0.05$).

specialist cues compared to any other cue source. Prevalence of gill-flushing did not differ between other treatment combinations (all $p > 0.14$).

Following the gill-flushing behavior, sirens often exhibited a reversing response. Time did not contribute to any variation in our data where main effects of time ($F_{1,64} = 0.001$, $p = 0.95$) and cue by time interaction ($F_{1,64} = 1.95$, $p = 0.31$) were not significant. Time was treated as a random effect, and cue as the only fixed effect in the final model. The main effect of cue on number of reverses was significant ($F_{3,63} = 5.97$, $p = 0.001$; Fig. 1b). Pairwise comparisons revealed that sirens reversed significantly more when exposed to specialist predator cues compared to blank control and generalist predator cues. Response to specialist predator and animal control cues was not significantly different. Sirens reversed more in response to animal control cues compared to blank control and generalist predator cues. There was no difference in reverses to blank control and generalist predator cues.

Discussion

Predator kairomones can indicate the level of threat that each predator represents (reviewed by [Kats & Dill 1998](#)) and can be used as a signal by animals that reduce opportunity costs of avoidance by altering their responses accordingly ([Sih 1992](#); [Anholt et al. 2000](#)). Sirens detected predator kairomones and varied their responses among predator types. Specialist kairomones elicited increased detection (gill-flushing) and avoidance (reversing) behaviors when compared to kairomones from generalist predators. Furthermore, sirens displayed a higher level of detection but not avoidance behaviors for specialist kairomones when compared to animal control kairomones. These differential responses to cues from potential threats suggest that sirens can respond to perceived threats with both detection and avoidance behaviors, as would be predicted by the threat-sensitive predator avoidance hypothesis ([Helfman 1989](#)).

Assuming that the magnitude of a prey's avoidance response correlates with its perception of risk, our study indicates that the specialist predator *F. abacura* poses a greater threat to sirens than the generalist predator *N. fasciata*. Additionally, sirens may treat novel species cues (i.e., our animal control) with a level of caution that is intermediate between those of known specialists and generalists. Behavioral responses to generalist cues did not differ from responses to the blank control; however, we

recorded two types of behaviors, and animals may deploy a suite of predator-specific responses (e.g., [Relyea 2001a,b](#)). It is possible that sirens perceive the generalist predator as a threat but respond with another behavior that was not recorded in this experiment. Alternatively, we believe the lack of response sirens exhibited toward kairomones of the generalist predator could indicate a familiarity with cues for a weak threat of predation. Generalists are less efficient hunters with fewer behavioral and morphological specializations and a wider diet breadth (e.g., [Drummond 1983](#); [Mori & Vincent 2008](#)), which may result in a reduced predation threat compared to specialist predators ([Orrock et al. 2008](#)). The Predator Continuum Hypothesis ([Ferrari et al. 2007b](#)) predicts that prey should exhibit the strongest responses to predators that are most likely to attack (i.e., specialists). Alternatively, prey should benefit from plastic responses to variably threatening predators (e.g., seasonal diet switches).

Few studies have directly compared prey response to cues from specialists and generalists, and none to our knowledge have examined response to predator cues alone without including diet or alarm signals from prey conspecifics. [Ferrari et al. \(2007a\)](#) found that minnows increased refugia usage more intensely in response to diet cues of specialist than to generalist fish predators, but this study did not examine prey response to predator cues alone. Furthermore, injured conspecifics and predator diet of conspecifics, both of which compose alarm substances, have been implicated in prey ability to learn to recognize novel predators (reviewed in [Mandrillon & Saglio 2005, 2007](#)). By excluding diet cues during stimulus acquisition in our study, the stronger prey response toward specialist predator cues are solely attributable to the predator-specific kairomones, not predator diet. However, there remains an intermediate response to the novel cue without relation to alarm substances – which is consistent with a more general response to related taxa (see [Webb et al. 2010](#)).

Although predator-specific responses can be elicited from many animals (e.g., [Relyea 2001a,b](#)), some may exhibit a more generalized response when encountering cues of predators they have not experienced before and adjust the degree of response through their experience with predator-specific cues ([Kats & Dill 1998](#)). *Eurycea nana*, another aquatic salamander, exhibits experience-specific reactions to a predator scent (*Lepomis auritus*), with predator-naïve animals demonstrating a stronger response than predator-experienced animals ([Epp & Gabor 2008](#)).

In our study, the novel cue from *H. platirhinos* elicited an intermediate response compared to the two known predator treatments, represented by significantly less gill-flushing than specialist, but not generalist, cues and more reversing than generalist, but not specialist, cues. Although we consider *H. platirhinos* a good 'novel' animal control for siren behavior, they may only be so in ecological time. Phylogenetic similarities between the animal control and specialist snakes and their diets (both are amphibian predators) could account for the intermediate responses we observed (e.g., *Heterodon* and *Farancia* are in the same subfamily, Xenodontinae, apart from *Nerodia*, in Natricinae). [Ferrari et al. \(2008\)](#) found that prey animals can exhibit similar antipredator responses to unfamiliar close relatives of a familiar high-risk, but not low-risk, predator. Sirens likely perceive *F. abacura* as an imminent threat and, therefore, respond appropriately in this study. The unfamiliar *H. platirhinos* in this study is more closely related to the high-risk specialist *F. abacura* than to the familiar low-risk *N. fasciata* predator, which coincides with reversing responses we observed. Therefore, our results indicate that sirens respond along a gradient that reflects the presumed danger represented.

In our system, prey more often responded with reversing and gill-flushing when the threat of predation was of utmost concern. Gill-flushing was a slow, stereotyped action and, to our knowledge, has not been reported before in salamanders. However, this behavior may be analogous to increased opercular movement observed in fishes in response to chemical cues of predators, novel species, and injured conspecifics ([Gibson & Mathis 2006](#)). Two viable explanations for this behavior have been suggested for fish, and applying these to gill-flushing behavior in salamanders seems reasonable and supports a general response in gill-breathing vertebrates. First, increased opercular movement increases oxygen consumption ([Dalla Valle et al. 2003](#)), which may be precursory to energy-expensive flight responses ([Rottmann et al. 1992](#)). Interestingly, gill-flushing preceded avoidance responses in 14 of the 19 trials in which it was observed. The avoidance response was likely associated with refugia seeking, a behavior typically observed in the field to reduce encounters with predators (reviewed by [Lima & Dill 1990](#)). Second, increased opercular movement could increase the flow of water, containing chemical cues, across olfactory sensory receptors, potentially enhancing the animal's chemosensory ability ([Wisenden & Chivers 2006](#)). While the exact benefit

of this behavior is still unknown, increased gill-flushing in sirens exposed to specialist predator kairomones provides strong evidence for its use in risk assessment. However, the unfamiliar and perhaps unknown risk of *H. platirhinos* exhibited no difference in gill-flushing to control and generalist cues. Future research should focus on the function and mechanics of this type of detection behavior in sirens and other aquatic salamanders.

Chemosensory ability in Sirenidae, the out group to all salamanders ([Zhang & Wake 2009](#)), supports chemosensory ability as a primitive caudate characteristic for detecting predators. The relative strength of behavioral responses to predator kairomones may indicate the relative selective pressures that these predators have on sirenids in the wild. Future research into community-level interactions of sirenids and other animals may provide additional insight into the relative importance of this locally dense (some of the highest densities of known freshwater vertebrates) and widely distributed group of enigmatic amphibians.

Acknowledgements

The authors thank J.W. Gibbons, J. Greene, S. Poppy, R.D. Semlitsch, and A. Tucker for their support and assistance in this opportunity. Furthermore, we thank C. Schalk for assistance in field collections and trial preparations. Sirens were collected under South Carolina Department of Natural Resources Scientific Collection permit G-08-07, and procedures used in the study were approved by the University of Georgia (AUP approval # 2006-10069). This work was supported by the National Science Foundation (grant number DBI-0453493) given to REU students. Some data used in this study were collected as part of an ongoing research project funded by the National Science Foundation (DBI-0453493), the American Museum of Natural History's Theodore Roosevelt Memorial Fund (awarded to TML), and the Savannah River Ecology Laboratory under Financial Assistance Award DE-FC09-96SR18-546 between the University of Georgia and the U.S. Department of Energy. We also thank D.W. Zeh and two anonymous reviewers for their helpful comments that improved this manuscript.

Literature Cited

Anholt, B. R., Werner, E. & Skelly, D. K. 2000: Effect of food and predators on the activity of four larval ranid frogs. *Ecology* **81**, 3509–3521.

- Blumstein, D. T., Evans, C. S. & Daniels, J. C. 2006: JWatcher 1.0. Available at: <http://www.jwatcher.ucla.edu/>.
- Bolker, B. M., Brooks, M. E., Clark, C. J., Geange, S. W., Poulsen, J. R., Stevens, M. H. & White, J. S. 2009: [Generalized linear mixed models: a practical guide for ecology and evolution](#). *Trends Ecol. Evol.* **24**, 127—135.
- Brodie, E. D. Jr, Formanowicz, D. R. Jr & Brodie, E. D. III 1991: Predator avoidance and antipredator mechanisms: distinct pathways to survival. *Ethol. Ecol. Evol.* **3**, 73—77.
- Chivers, D. P. & Mirza, R. S. 2001: [Predator diet cues and the assessment of predation risk by aquatic vertebrates: a review and prospectus](#). In: *Chemical Signals in Vertebrates* (Marchlewska-Koj, A., Lepri, J. & Müller-Schwarze, D., eds). Kluwer Academic/Plenum, New York, pp. 277—284.
- Chivers, D. P. & Smith, R. J. F. 1998: [Chemical alarm signaling in aquatic predator-prey systems: a review and prospectus](#). *Ecoscience* **5**, 338—352.
- Dalla Valle, A. Z., Rivas-Diaz, R. & Claireaux, G. 2003: [Opercular differential pressure as a predictor of metabolic oxygen demand in the starry flounder](#). *J. Fish Biol.* **63**, 1578—1588.
- Drummond, H. 1983: [Aquatic foraging in garter snakes: a comparison of specialists and generalists](#). *Behaviour* **86**, 1—30.
- Duellman, W. E. & Schwartz, A. 1958: [Amphibians and reptiles of southern Florida](#). *Bull. Fl. St. Mus., Gainesville* **3**, 181—324.
- Endler, J. A. 1986: [Defense against predators](#). In: *Predator-Prey Relationships* (Feder, M. E. & Lauder, G. V., eds). Univ. of Chicago Press, Chicago, IL, pp. 109—134.
- Epp, K. J. & Gabor, C. R. 2008: [Innate and learned predator recognition mediated by chemical signals in *Eurycea nana*](#). *Ethology* **114**, 607—615.
- Ferrari, M. C. O., Brown, M. R., Pollock, M. S. & Chivers, D. P. 2007a: [The paradox of risk assessment: comparing responses of fathead minnows to capture-released and diet-released alarm cues from two different predators](#). *Chemoecology* **17**, 157—161.
- Ferrari, M. C. O., Gonzalo, A., Messier, F. & Chivers, D. P. 2007b: [Generalization of learned predator recognition: an experimental test and framework for future studies](#). *Proc. R. Soc. Lond. Ser. B-Biol. Sci.* **274**, 1853—1859.
- Ferrari, M. C. O., Messier, F., Chivers, D. P. & Messier, O. 2008: [Can prey exhibit threat-sensitive generalization of predator recognition? Extending the predator recognition continuum hypothesis](#). *Proc. R. Soc. Lond. Ser. B-Biol. Sci.* **275**, 1811—1816.
- Ferry-Graham, L. A., Bolnick, D. I. & Wainwright, P. C. 2002: [Using functional morphology to examine the ecology and evolution of specialization](#). *Integr. Comp. Biol.* **42**, 265—277.
- Gibbons, J. W. & Dorcas, M. E. 2004: *North American Watersnakes: A Natural History*. Univ. Oklahoma Press, Norman, OK.
- Gibson, A. K. & Mathis, A. 2006: [Opercular beat rate for rainbow darters *Etheostoma caeruleum* exposed to chemical stimuli from conspecific and heterospecific fishes](#). *J. Fish Biol.* **69**, 224—232.
- Helfman, G. S. 1989: [Threat-sensitive predator avoidance in damselfish-trumpetfish interactions](#). *Behav. Ecol. Sociobiol.* **24**, 47—58.
- Hickman, C. R., Stone, M. D. & Mathis, A. 2004: [Priority use of chemical over visual cues for detection of predators by graybelly salamanders, *Eurycea multiplicata* griseogaster](#). *Herpetologica* **60**, 203—210.
- Jensen, J. B., Camp, C. D., Gibbons, J. W. & Elliot, M. J. 2008: *Amphibians and Reptiles of Georgia*. The Univ. of Georgia Press, Athens, GA.
- Kats, L. B. & Dill, L. M. 1998: [The scent of death: chemosensory assessment of predation risk by prey animals](#). *Ecoscience* **5**, 361—394.
- Kiemiec, K. M. 2009: *Chemical Cues and the Molecular Basis of Olfactory Chemoreception in Caudate Amphibians*. PhD Thesis, Oregon State Univ., Corvallis, OR.
- Lima, S. L. & Dill, L. M. 1990: [Behavioral decisions made under the risk of predation: a review and prospectus](#). *Can. J. Zool.-Rev.* **68**, 619—640.
- Luhring, T. M. 2008: *Population Ecology of Greater Siren, *Siren lacertina**. MS Thesis, Univ. of Georgia, Athens, GA.
- Luhring, T. M. & Jennison, C. A. 2008: [A new stratified aquatic sampling technique for aquatic vertebrates](#). *J. Fresh. Ecol.* **23**, 445—450.
- Madison, D. M., Maerz, J. C. & McDarby, J. H. 1999: [Optimization of predator avoidance by salamanders using chemical cues: diet and diel effects](#). *Ethology* **105**, 1073—1086.
- Mandrillon, A. L. & Saglio, P. 2005: [Prior exposure to conspecific chemical cues affects predator recognition in larval common toad \(*Bufo bufo*\)](#). *Arch. Hydrobiol.* **124**, 1—12.
- Mandrillon, A. L. & Saglio, P. 2007: [Herbicide exposure affects the chemical recognition of a non native predator in common toad tadpoles \(*Bufo bufo*\)](#). *Chemoecology* **17**, 31—36.
- Mathis, A. & Vincent, F. 2000: [Differential use of visual and chemical cues in predator recognition and threat-sensitive predator-avoidance responses by larval newts \(*Notophthalmus viridescens*\)](#). *Can. J. Zool.-Rev.* **78**, 1646—1652.
- Mathis, A., Murray, K. L. & Hickman, C. R. 2003: [Do experience and body size play a role in responses of larval ringed salamanders, *Ambystoma annulatum*, to](#)

- predator kairomones? Laboratory and field assays. *Ethology* **109**, 159–170.
- [Mori, A. & Vincent, S. E. 2008: An integrative approach to specialization: relationships among feeding morphology, mechanics, behaviour, performance and diet in two syntopic snakes. *J. Zool.* **275**, 47–56.](#)
- [Mushinsky, H. R., Hebrard, J. J. & Vodopich, D. S. 1982: Ontogeny of water snake foraging ecology. *Ecology* **63**, 1624–1629.](#)
- [Orrock, J. L., Grabowski, J. H., Pantel, J. H., Peacor, S. D., Peckarsky, B. L., Sih, A. & Werner, E. E. 2008: Consumptive and nonconsumptive effects of predators on metacommunities of competing prey. *Ecology* **89**, 2426–2435.](#)
- [Petranka, J. W. 1989: Response of toad tadpoles to conflicting chemical stimuli: predator avoidance versus optimal foraging. *Herpetologica* **45**, 283–292.](#)
- [Relyea, R. A. 2001a: Morphological and behavioral plasticity of larval anurans in response to different predators. *Ecology* **82**, 523–540.](#)
- [Relyea, R. A. 2001b: The relationship between predation risk and antipredator responses in larval anurans. *Ecology* **82**, 541–554.](#)
- [Rottmann, R. W., Francis-Floyd, R. & Durborrow, R. 1992: The Role of Stress in Fish Disease. Southern Regional Aquaculture Center Publication No. 474. Texas A. & M. Univ., College Station, TX.](#)
- [Schielzeth, H. & Forstmeier, W. 2009: Conclusions beyond support: overconfident estimates in mixed models. *Behav. Ecol.* **20**, 416–420.](#)
- [Sih, A. 1987: Predator and prey lifestyles: an evolutionary and ecological overview. In: *Predation: Direct and Indirect Impacts on Aquatic Communities* \(Kerfoot, W. C. & Sih, A., eds\). Univ. Press of New England, Hanover, NH, pp. 263–278.](#)
- [Sih, A. 1992: Prey uncertainty and the balancing of anti-predator and feeding needs. *Am. Nat.* **139**, 1052–1069.](#)
- [Sih, A., Englund, G. & Wooster, D. 1998: Emergent impacts of multiple predators on prey. *Trends Ecol. Evol.* **13**, 350–355.](#)
- [Snyder, W. E. & Wise, D. H. 2000: Antipredator behavior of spotted cucumber beetles \(Coleoptera: Chrysomelidae\) in response to predators that pose varying risks. *Environ. Entomol.* **29**, 35–42.](#)
- [Stauffer, H. P. & Semlitsch, R. D. 1993: Effects of visual, chemical and tactile cues of fish on the behavioural responses of tadpoles. *Anim. Behav.* **46**, 355–364.](#)
- [Sullivan, A. M., Frese, P. W. & Mathis, A. 2000: Does the aquatic salamander, *Siren intermedia*, respond to chemical cues from prey? *J. Herpetol.* **34**, 607–611.](#)
- [Sullivan, A. M., Picard, A. L. & Madison, D. M. 2005: To avoid or not to avoid? Factors influencing the discrimination of predator diet cues by a terrestrial salamander. *Anim. Behav.* **69**, 1425–1433.](#)
- [Via, S. 2001: Sympatric speciation in animals: the ugly duckling grows up. *Trends Ecol. Evol.* **16**, 381–390.](#)
- [Ward, A. B. & Azizi, E. 2004: Convergent evolution of the head retraction escape response in elongate fishes and amphibians. *Zoology* **107**, 205–217.](#)
- [Webb, J. K., Pike, D. A. & Shine, R. 2010: Olfactory recognition of predators by nocturnal lizards: safety outweighs thermal benefits. *Behav. Ecol.* **21**, 72–77.](#)
- [Wisenden, B. D. & Chivers, D. P. 2006: The role of public chemical information in antipredator behaviour. In: *Communication in Fishes, Vol. 1.* \(Kapoor, B. G., Ladich, F., Collin, S. P. & Moller, P., eds\). Oxford & IBH Publishing House Co, Janpath, New Delhi.](#)
- [Zhang, P. & Wake, D. B. 2009: Higher-level salamander relationships and divergence dates inferred from complete mitochondrial genomes. *Molec. Phylog. Evol.* **53**, 492–508.](#)