Nature of Predation Risk Cues in Container Systems: Mosquito Responses to Solid Residues From Predation

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ABSTRACT   In aquatic systems, prey animals associate predation risk with cues that originate either from the predator or from injured conspecifics. Sources and benefits of these cues have received considerable attention in river, lake, and pond ecosystems but are less well understood in small container ecosystems that can hold less than a liter of water. Mosquitoes *Aedes triseriatus* (Say) and *Aedes albopictus* (Skuse) encounter predatory *Corethrella appendiculata* (Grabham) and *Toxorhynchites rutilus* (Coquillett) in small containers and show antipredatory behavioral responses. We investigated the sources of the predation cues to which these prey larvae respond. We tested whether *Ae. albopictus* larvae show behavioral responses to cues emanating from the predator or from damage to prey caused by the act of predation. We also tested whether *Ae. triseriatus* respond to cues present in fluid or solid residues from predator activity. *Ae. albopictus* showed behavioral modifications only in response to waterborne cues from a feeding predator and not to cues from a starving predator, indicating that *Ae. albopictus* respond to cues created by the act of predation, which could include substances derived from damaged prey or substances in predator feces. *Ae. triseriatus* showed behavioral responses to solid residues from predation but not to fluid without those solids, indicating that the cues to which they respond originate in predator feces or uneaten prey body parts. Our results suggest that cues in this system may be primarily chemicals that are detected upon contact with solid residues that are products of the feeding processes of these predators.

KEY WORDS   predation risk, container community, antipredatory behavior

In aquatic systems, prey from many taxa modify behavior in response to cues to predation risk and these modifications can reduce an individual's risk from predation (Sih and Moore 1993, Hughes et al. 1994, Chivers and Smith 1998, Wisenden 2000, Hamilton and Heithaus 2001, Laurila et al. 2004). The cues by which aquatic prey perceive the risk of predation are often chemical and can originate with the predator itself (Chivers and Smith 1998, Wisenden 2000, Tollrian and Heibl 2004, Gyssels and Stoks 2006, Ferris and Rudolf 2007) or can be created by the act of predation, including alarm cues that arise when prey are damaged (Chivers and Smith 1998, Relyea 2001), and dietary cues that originate only after consumed prey are digested (Chivers and Smith 1998). Understanding the source and nature of the cues to predation is central for understanding the proximate mechanisms of adaptive behavior, and may help in understanding specificity of predator detection mechanisms, the potential costs of antipredatory behavior, and ultimately the evolutionary origin of prey responses to predators. Apart from direct predation, costs due to nonlethal effects can be important in structuring communities and hence studies focusing on nonlethal effects have

increased (Ripple and Beschta 2004, Trussell et al. 2004). As one practical example of the value of understanding the origins of cues to predation risk, knowing sources of cues can be useful when designing experiments in which investigators simulate predator cues to induce behavioral response as a means of testing hypotheses about the costs (e.g., reduced foraging) of prey's responses to the predator (Werner et al. 1983, Werner 1991, Van Buskirk and Arioli 2002, Van Buskirk et al. 2002, Schoeppner and Relyea 2005) and the nonlethal effects of predators (Preisser et al. 2005).

Several studies have reported prey antipredatory behavior in response to alarm cues. Mayfly nymphs (Huryn and Chivers 1999), caddisfly larvae (Gall and Brodie 2009), and amphipods (Wisenden et al. 1999) reduce their activity in the presence of cues from injured conspecifics. The intertidal snail *Littorina scutulata* (Gould) shows a graded response with increase in predation risk perceived from predator alone, injured conspecifics alone, or a combination of both cues (Keppel and Scrosati 2004). Newts, *Notopthalamus viridiscens* (Rafinesque), increase their frequency of low-risk behaviors in the presence of macerated conspecifics (Rohr and Madison 2001). Mechanisms of detection of predation risk are best

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understood for ostariophysian fishes (reviewed by Wisenden 2000, Chivers and Smith 1998, Chivers et al. 2007). Alarm substances from damage to epidermis play a major role in this process and these fishes show behavioral responses to lab synthesized hypoxanthine-3-*N*-oxide (Brown et al. 2000). These alarm chemicals are species-specific (Brown et al. 1995a) and can be released from the act of predation (Wisenden 2008) or be present in the feces of the predators that fed on conspecifics (Brown et al. 1995b). A single conditioning treatment with a combination of predator stimuli (visual or odor) from a novel predator and injured conspecifics is sufficient for fathead minnows to recognize the novel predator and their taxonomic siblings as potential predators and to show avoidance behaviors (Ferrari et al. 2008). Studies described above all relate to large, heterogeneous aquatic systems (e.g., rivers, lakes, and ponds), but predation and the nature of cues are less well understood in small confined systems like containers. Rainwater collects in small (often <1 liter) containers such as depressions in trees, cemetery vases, and abandoned tires, and these are inhabited by communities of aquatic insects which are dominated by mosquitoes. Such small aquatic habitats limit prey ability to avoid subhabitats where predation risk is high; hence, we expect prey in such habitats to respond to cues to predation risk by modifying behavior or life history. Although the presence of responses to predation risk cues are known in container systems (Juliano and Gravel 2002; Kesavaraju and Juliano 2004, 2008; Kesavaraju et al. 2007a,b, 2008), the sources and type of those cues are poorly investigated.

Prey Mosquitoes. *Aedes albopictus* (Skuse) is an invasive mosquito, introduced into North America from Asia in the mid-1980s (Hawley et al. 1987), that has become abundant in the southeastern United States (Juliano and Lounibos 2005). Larvae inhabit natural (tree holes) and artificial container habitats (e.g., water filled tires and cemetery vases), co-occurring with the native mosquito *Aedes triseriatus* (Say), the predatory midge *Corethrella appendiculata* (Grabham), and the predatory mosquito *Toxorhynchites rutilus* (Coquillett) (Griswold and Lounibos 2005). *C. appendiculata* and *Tx. rutilus* are sit and wait predators and use mechanoreceptors to capture their prey and typically make most captures at the bottom of containers (Juliano and Reminger 1992, Kesavaraju et al. 2007a). Prey larvae that are more active at the bottom are at a higher risk of capture than are those that are motionless at the surface (Juliano and Reminger 1992, Kesavaraju et al. 2007a).

Responses to *Corethrella appendiculata*. Second instars of both *Ae. albopictus* and *Ae. triseriatus* are highly vulnerable to predation by fourth-instar *C. appendiculata*, and both species reduce activity at the bottom of containers in the presence of waterborne cues from *C. appendiculata* preying on *Aedes* larvae (Kesavaraju et al. 2007a). These changes reduce the risk of predation (Kesavaraju et al. 2007a). For *Ae. albopictus*, the degree of behavioral change is less than that shown by *Ae. triseriatus*, so that *Ae. albopictus*

larvae are more vulnerable to *C. appendiculata* predation (Kesavaraju et al. 2007a). Where *C. appendiculata* abundances are low *Ae. albopictus* dominates, and where *C. appendiculata* abundances are high, these species coexist (Kesavaraju et al. 2008).

The waterborne cues to which *Ae. albopictus* and *Ae. triseriatus* larvae respond could come from the act of predation or from the predator itself, independent of the act of predation (Lima and Dill 1990, Lima 1998). Both species modify behavior in water that had held *C. appendiculata* feeding on *Aedes* larvae for 5 d, but both altered behavior less when a living *C. appendiculata* was added to the container only minutes before the trial (Kesavaraju et al. 2007a). These results suggest that cues either emanate from predation, or emanate from the predator itself, but must accumulate for 5 d to be effective.

Responses to *Tx. rutilus*. Fourth instars of *Ae. triseriatus* reduce their activity at the bottom of the container in the presence of waterborne predation risk cues from *Tx. rutilus* (Kesavaraju and Juliano 2004). *Ae. triseriatus* larvae show the same behavioral response to *Tx. rutilus* feeding on conspecifics or on *Ae. albopictus*, indicating that the cues are not species specific (Kesavaraju and Juliano 2004). In contrast to the small response of *Ae. albopictus* to cues from *C. appendiculata* (Kesavaraju et al. 2007a), they show no significant response to *Tx. rutilus* predation risk cues (Kesavaraju and Juliano 2004). Behavioral responses of *Ae. triseriatus* to *Tx. rutilus* predation risk cues decrease as the concentration of cue-laced water and suspended solids decreases via dilution with distilled water, suggesting a graded, threat sensitive response to the abundance or activity of predators (Kesavaraju et al. 2007b). The source of the cues to which *Ae. triseriatus* respond is not clear. *Ae. triseriatus* showed no significant behavioral response to feeding *Tx. rutilus* isolated in a cage (Hechtel and Juliano 1997), suggesting that solid components (e.g., predator feces) that were retained in the cage may be the source of the cue.

To understand the mechanisms by which these mosquitoes detect their predators, we tested the hypotheses 1) *Ae. albopictus* larvae respond to waterborne cues from either *C. appendiculata* itself or from conspecifics injured during the act of predation and 2) *Ae. triseriatus* larvae either respond to dissolved waterborne cues or to cues in the solid residues of *Tx. rutilus* predation.

Materials and Methods

Origin of Larvae. *Ae. albopictus* were F_1 progeny from a colony collected initially as larvae from tree holes (Indrio Road, Fort Pierce, FL). Both *C. appendiculata* and *Tx. rutilus* are common at this site (S.A.J., personal observations). *Ae. triseriatus* were F_1 progeny of individuals collected initially as larvae from tree holes at Parklands Merwin Reserve near Lexington, IL. *Tx. rutilus* are rare at this site but occur sporadically in tree holes (Juliano 1989). Thus, larvae in both experiments were descendents of field-collected indi-

viduals that probably encountered the test predator in each experiment. Both *Aedes* were propagated via weekly blood feeding. *C. appendiculata* and *Tx. rutilus* larvae were from laboratory colonies maintained at the Florida Medical Entomology Laboratory, Vero Beach, FL. Both predator colonies were established from larvae that were collected in Florida.

Experiment 1. Response of *Ae. albopictus* to Different Types of Cues From *C. appendiculata*. Behavior of *Ae. albopictus* second-instar larvae was recorded in water treated in four ways: control, predator with prey, predator alone, and deionized water (blank). All treatments were prepared for 5 d in 10-ml polystyrene cups with 10 ml of deionized water. Control was prepared by holding 10 second-instar *Ae. albopictus.* Predator with prey was prepared by holding 10 second-instar *Ae. albopictus* with three fourth-instar *C. appendiculata.* Dead, eaten, and pupated larvae were replaced daily. Predator alone was prepared by holding three *C. appendiculata* fourth instars without food. Finally, the blank treatment consisted of 10 ml of water held without addition of larvae. Each treatment was replicated 24 times, for a total of 96 replicated units.

Second-instar *Ae. albopictus* were used as test larvae for recording behavior. The test larvae were hatched and held with 5 ml of water in 15-ml vials and fed with 1 ml of liver powder suspension (LPS), which was prepared by stirring 0.3 g of liver powder in a 1,000-ml beaker with 1,000 ml of water on a stir plate and transferred using an Eppendorf pipette (Juliano and Gravel 2002, Kesavaraju and Juliano 2004). A single feeding was sufficient for *Ae. albopictus* to develop to the second instar.

Test larvae were starved for 24 h in 10-ml cups with 10 ml of water before being transferred to treatment cups for behavior recording. Before test larvae were transferred into the treatment cups, all predator and prey treatment larvae were removed from the treatment cups, leaving behind only any waterborne cues (e.g., uneaten body parts, feces, dissolved chemicals) emanating from the treatments. One second-instar *Ae. albopictus* larva was placed in treatment water in each container and their behavior was recorded on a computer in MPEG2 format by using a Panasonic video camera and zoom lens (WV-D5100 and WV-LZ14/15, respectively) and a Winfast XP 2000 PCI card (Leadtek Research Inc., www.leadtek.com) for 15 min. A video clip contained four cups with all treatments represented in each clip.

Behaviors were classified into activities and positions (Juliano and Reminger 1992). Activities were as follows: 1) browsing: mouthparts in contact with the container surfaces; 2) filtering: moving in the water column via feeding movements of the mouthparts; 3) thrashing: moving with vigorous lateral flexion of the body; and 4) resting: not exhibiting any previous activities. Positions were as follows: 1) surface: siphon in contact with water surface; 2) wall: within 1 mm of the sides; 3) bottom: within 1 mm of the bottom; and 4) middle: >1 mm from the sides, bottom, and surface.

Activity and position of the test larvae were recorded every 30 s for 15 min (Kesavaraju and Juliano 2008) upon playback of the video clips. Behaviors were then converted to proportions (total number of observations per replicate, 30) for each replicate. The number of behavioral variables was reduced with principal component analysis (PCA). Because past work on these species (Juliano and Gravel 2002; Kesavaraju and Juliano 2004, 2008; Kesavaraju et al. 2007a, 2008) has shown that the primary response of these mosquitoes to predation cues is an increase in the frequency of resting at the surface at the expense of foraging moving below the surface, we focus our analysis on a variable that quantifies this shift in behavior, which in these analyses was always the principal component 1 (PC1) (see Results). Thus, we analyzed PC1 via one-way analysis of variance (ANOVA) (PROC GLM, SAS 9.1, SAS Institute, Cary, NC) (Kesavaraju and Juliano 2004). We used Tukey–Kramer multiple comparisons among treatment least-squares means for pairwise comparisons.

Experiment 2. Nature of *Tx. rutilus* Predation Risk Cues. Behavior of fourth-instar *Ae. triseriatus* were recorded in water treated four ways: control fluid, control solid, predation fluid, and predation solid, all prepared for 5 d in 50-ml disposable cups with 50 ml of deionized water. Predation water was prepared by feeding a fourth-instar *Tx. rutilus* with 10 fourth-instar *Ae. triseriatus* and control water by holding 10 fourth-instar *Ae. triseriatus* larvae. Pupated and dead larvae were replaced daily. After 5 d, both the prey and the predator were removed from the cups leaving behind the only any waterborne cues (e.g., uneaten body parts, feces, and dissolved cues). The control and predation water treatments were then filtered with a Whatman filter paper (grade 2, size 12.5 cm) placed in a funnel on a conical flask that was connected to a vacuum pump. The solid matter retained on the filter paper was washed into a clean 50-ml disposable cup with deionized water and then the volume was brought up to 50-ml with deionized water. The filtrate from the conical flask was transferred to 50-ml disposable cups. Predation and control treatments were replicated 35 times each and because each predation cup and control cup yielded two treatments (predation solid, predation fluid, control solid, control fluid), the total number of replicated units was 140.

Fourth-instar *Ae. triseriatus* were used as test larvae for recording behavior. The test larvae were hatched and held in 5 ml of water in 15-ml vials and fed 1 ml of LPS as described in experiment 1 every 2 d. Test larvae were starved for 24 h in 50-ml cups with 50 ml of water before being transferred for behavior recording. One fourth-instar *Ae. triseriatus* was placed in treatment water in each container and their behavior was recorded using the same Panasonic videocamera and an S-VHS video cassette recorder for 30 min (Kesavaraju and Juliano 2004). A video clip contained six cups and all treatments were represented on each clip. The treatment waters were filtered and the filtrates transferred to the cups 15–30 min before video recording.

Table 1. Rotated factor patterns for the behavioral responses of *A. albopictus* in different types of cues

Variable	PC1 (50%)	PC2 (24%)	PC3 (13%)
Resting	98	−6	5
Browsing	−91	−32	−14
Thrashing	0	98	−12
Filtering	17	7	85
Surface	98	−6	5
Wall	−77	−31	−40
Middle	15	93	23
Bottom	−60	−5	48
Interpretation	Resting, surface vs. browsing, wall, bottom	Thrashing, middle vs. other	Filtering, bottom vs. wall

Percent variation explained by each PC is given within parentheses. Values >40 are in bold.

Table 2. Rotated factor patterns for comparing the behavioral responses of fourth-instar *A. triseriatus* between fluid and solid cues

Variable	PC1 (46%)	PC2 (26%)	PC3 (13%)
Resting	−84	−45	−9
Browsing	90	29	−23
Thrashing	−8	78	11
Filtering	−7	−6	95
Surface	−63	−69	−15
Wall	88	−21	−19
Middle	−12	15	94
Bottom	33	83	−12
Interpretation	Resting, surface vs. browsing, wall	Resting, surface vs. thrashing, bottom	Filtering, middle vs. other

Percent variation explained by each PC is given within parentheses. Values higher than 40 are in bold.

The behaviors described in experiment 1 (activity and position) were recorded every minute for 30 min. Behaviors were converted to proportions, variables were reduced with PCA, and our focal variable PC1 was analyzed with by two-way ANOVA (PROC GLM, SAS 9.1) with treatment (control, predation), cues (solid, fluid), and interaction as independent variables. A significant interaction between treatment and cues would indicate that differences in larval behavior were dependent on both treatment and type of cue and provide evidence of a differential response to either dissolved cues in the fluid or contact cues in the solid components. We used Tukey–Kramer multiple comparisons among treatment least-squares means for pairwise comparisons.

Results

Experiment 1. PCA reduced the response variables to three uncorrelated PCs, with eigenvalues >1 accounting for 87% of total variation (Table 1). A greater positive score on PC1 indicated that larvae spent more time resting at the surface, and a negative score indicated more time browsing at the wall. Greater positive scores on PC2 indicated more time spent in thrashing in the middle, and a negative score indicated more time spent in other behaviors. Greater positive score on PC3 indicated more time spent filtering at the bottom, and a negative score indicated more time spent at the wall (Table 1).

Treatments (blank, control, predator alone, predator with prey) differed significantly for PC1 (F = 18.41; df = 3, 90; $P < 0.0001$). Multiple comparisons revealed that there were no significant differences among blank, control, and predator alone treatments, but that all three treatments were significantly different from predator with prey (Fig. 1). *Ae. albopictus* reduced movement and spent more time resting at the surface in water that had held feeding predators compared with the other three treatments (Fig. 1).

Experiment 2. Similar to experiment 1, there were three PCs with eigenvalues >1 (Table 2), accounting for 86% of the variation in behavior. A greater positive score on PC1 indicated that larvae spent more time browsing at the wall, and a negative score indicated more time spent resting at the surface. Signs on PC scores are arbitrary; hence, PC1 from the two experiments describes a similar behavioral axis. A greater positive score on PC2 indicated that larvae spent more time thrashing at the bottom, and a negative score indicated more time spent resting at the surface. A greater positive score on PC3 indicated that the larva spent more time filtering in the middle, and a negative
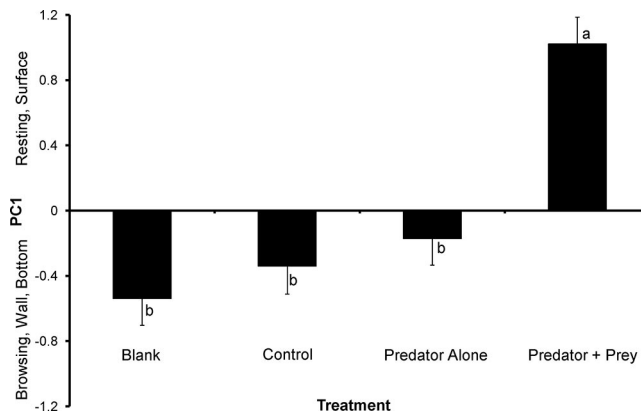

Fig. 1. Experiment 1: *Ae. albopictus* behavioral response on PC1 (mean ± SE) to different types of cues. Means associated with the same letters are not significantly different from each other.
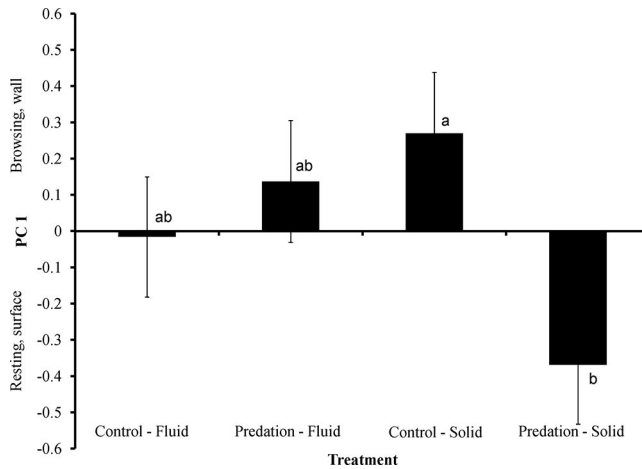
Fig. 2. Experiment 2: differences in behavioral PC1 (mean ± SE) of *Ae. triseriatus* between fluid and solid cues of predation risk from *Tx. rutilus*.

score indicated more time spent in other behaviors (Table 2).

The interaction between cues (fluid and solid) and treatment (control and predation) was significant for PC1 ($F = 5.65$; df = 1, 135; $P = 0.0188$). Multiple comparisons revealed that control solid and predation solid were significantly different from each other ($P = 0.0364$), but there were no differences between any other treatment pairs, indicating that *Ae. triseriatus* responded primarily to cues present in the solid material (Fig. 2).

Discussion

Ae. albopictus reduced movement and increased resting at the surface of the containers when predators were feeding on other *Ae. albopictus*, and their behavior differed significantly from all treatments without feeding predators (Fig. 1). These results suggest that *Ae. albopictus* larvae respond only to the cues that are created by the act of predation. Several arthropods respond to damaged conspecifics (Huryn and Chivers 1999, Wisenden et al. 1999, Gall and Brodie 2009); our data are consistent with a similar pattern of response in *Ae. albopictus.* Alternatively, *Ae. albopictus* may respond to the excrement of *C. appendiculata* because the larvae significantly altered their behavior in response to feeding *C. appendiculata* but not in response to nonfeeding *C. appendiculata.* Because *C. appendiculata* larvae were in the predator alone treatments for 5 d, it is likely that they excreted in the cups, albeit in low levels compared with those *C. appendiculata* that were actively feeding. Although not significantly different from the controls, the mean for PC1 for *Ae. albopictus* larvae in the predator alone treatment indicated slightly greater frequency of resting at the surface (Fig. 1), which may have been a result of the limited amount of feces produced by the nonfeeding *C. appendiculata.* Our results indicate that *Ae. albopictus* larvae show no significant response to

waterborne predation risk cues emanating from nonfeeding *C. appendiculata* and thus indicate that the principal cue to predation risk is derived in some way from the act of predation.

Previous experiments show that both *Ae. albopictus* and *Ae. triseriatus* reduce their activity at the bottom of the containers in response to the physical presence of *C. appendiculata* (Kesavaraju et al. 2007a). The present experiment did not test for effects of the physical presence, but together with these previous results, suggests that these *Aedes* may use multiple cues (chemical, visual, tactile) to evaluate predation risk.

Ae. triseriatus increased low risk behaviors (resting in the surface) in water containing filtered solids from predation compared with either solid residues or filtered residues from living *Ae. triseriatus.* Thus, it seems that solid residues from *Tx. rutilus* predation are the main source of cues to predation perceived by *Ae. triseriatus* (Fig. 2). Although most of the literature on cues to predation in freshwater systems emphasizes dissolved chemical cues (Chivers and Smith 1998, Wisenden 2000), there are other examples of prey responses to solid residues. Fathead minnows showed antipredatory behavior in the presence of feces from northern pike that had fed on conspecifics (Brown et al. 1995a,b, 1996). Brown et al. (1995a) prepared the stimulus by collecting, filtering, and freezing the feces of the predator and argued that the chemical alarm pheromone in the feces gets released slowly when they are resuspended in water. Brown et al. (1996) showed that behavior of their predator was attuned to the perception by prey of chemical alarm pheromones in the feces, because northern pike defecated away from their foraging area.

Although some cues may leach from solid residues into solution, it is unlikely that the observed response to filtered solids resulted from small amounts of soluble chemical cues that might have leached from solids during the filtering process. Kesavaraju

et al. (2007b) prepared *Tx. rutilus* predation water by feeding *Tx. rutilus* with *Ae. triseriatus* in treatments similar to those in the present experiment and tested the behavior of *Ae. triseriatus* at different dilutions from the original concentration. Behavior of *Ae. triseriatus* in response to low levels of predation risk cues was not significantly different from no-predation control. The strong responses we observed in the present experiment to solid material suggest that contact with solid residues (e.g., while *Aedes* larvae are foraging) provides the cues to the presence of a predation threat. If these behavioral responses were induced by leached, dissolved chemicals from the feces, we also would have expected the predator fluid treatment to have yielded a significant change in behavior of *Ae. triseriatus;* it did not (Fig. 2). Kesavaraju and Juliano (2004) showed that fourth-instar *Ae. triseriatus* increased their low-risk behaviors in both predation water from *Tx. rutilus* feeding on conspecifics or on *Ae. albopictus*, indicating that solid residues from a variety of victims of predation can serve as predation risk cues.

Hechtel and Juliano (1997) held feeding fourth-instar *Tx. rutilus* in a small cage inside cups and recorded no increase in low-risk behavior of *Ae. triseriatus* outside of the cage. Our current study indicates that lack of behavioral response from *Ae. triseriatus* observed by Hechtel and Juliano (1997) probably arose because the solid cues were retained within the cage with *Tx. rutilus.* Although in experiment 1 the response of *Ae. albopictus* was tested in the presence of a different predator (*C. appendiculata*), the lack of response in predator only water is also consistent with the hypothesis that *Ae. albopictus*, like *Ae. triseriatus*, perceives risk of predation via solid residues created by the act of predation. We cannot at this point determine whether those cues arise from damage to the victim at the time of attack (e.g., bits of uneaten victim), or from the products of digestion of the victim that are in predator feces, or from some combination of those sources (Wisenden 2000, Schoppner and Relyea 2005).

Antipredator responses of prey have been typically studied by recording the behavior of prey in water that had caged feeding predators (e.g., Relyea 2000) or in water that had held a feeding predator (e.g., Kesavaraju and Juliano 2004), or by combining visual cues with water that had held a nonfeeding predator (e.g., Chivers et al. 2001). Many of these studies indicate that these predation risk cues are waterborne chemical and visual cues (e.g., Chivers et al. 2001). The results described in this study are, to our knowledge, among the first to show that the filtered solid material from a feeding predator also can serve as predation risk cues. Some aquatic prey such as mosquitoes show antipredator responses only in the presence of cues that are created by actual predation on conspecifics and not to the cues from a nonfeeding predator (Ferrari et al. 2007b, this study). Antipredatory responses are costly because reduced activity also means reduced

foraging opportunities that favor threat sensitive responses to predation risk (Ferrari et al. 2007b, Kesavaraju et al. 2007b). Because these *Aedes* occur in small aquatic habitats, dissolved cues from the predator itself may be pervasive, and responses to such cues too costly. Cues derived from the act of predation provide a more specific indication of an immediate threat, and thus may be more efficient cues, particularly in such small water volumes. Solid residues of predation also may provide more specific information than would dissolved chemical cues, because solid residues encountered on the bottom by a forager may provide information about the spatial distribution of predation risk.

Predation risk cues in aquatic systems degrade if not replenished by additional predation, and prey may alter their responses depending on the degradation level (Ferrari et al. 2007a). Furthermore, pupation of predators removes the source of new predation cues. Modulating the degree of behavioral response to correspond with cue amount, as it changes with natural degradation of solid cues created by predation, would help these *Aedes* limit costly antipredatory behaviors to times when those responses are most advantageous.

Acknowledgments

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