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## CHEMICAL ALARM SIGNALS: PREDATOR DETERRENTS OR PREDATOR ATTRACTANTS?

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*Abstract.*—Although fright responses to alarm signals provide obvious benefits to receivers, benefits to senders of alarm signals are less clear. Ostariophysan fishes produce a chemical alarm signal ("Schreckstoff") that is released only following mechanical damage to the skin, such as that which occurs following capture by a predator. Two mutually exclusive hypotheses for the evolution of chemical alarm signals in senders are predator deterrence and predator attraction. According to the predator-attraction hypothesis, the alarm pheromone functions to attract additional predators that interfere with the predation event, which allows an opportunity for the prey to escape. We used laboratory and field experiments to demonstrate that the alarm substance of fathead minnows (Cyprinidae) is attractive to two different predators, northern pike (Esocidae) and predaceous diving beetles (Dytiscidae). We suggest that damage-released alarm pheromones, such as the alarm substance of ostariophysan fishes, are analogous to the distress calls of some birds and mammals in that they are produced only after the sender has been captured and may function to attract other predators. Alarm signals that are released only following capture by predators may therefore be functionally distinct from other alarm signals and may have evolved through direct benefits to the sender.

Understanding the evolution of alarm signals is one of the more elusive issues in behavioral ecology (Williams 1964, 1992; Taylor et al. 1990). The puzzle is especially perplexing for chemical alarm signals that are released only following mechanical damage to the skin. If such signals evolved as a mechanism to warn conspecifics (e.g., through kin selection; Smith 1977), then it follows that a voluntary release mechanism should allow for a much more efficient warning system. Moreover, because mechanical damage typically occurs only after the prey individual has been captured by a predator, it is difficult to see any direct benefit to the sender.

Damage-induced release of chemical alarm pheromones has been described for a wide variety of taxa, including invertebrates such as anthozoans (Howe and Sheikh 1975), gastropods (Sleeper et al. 1980; Rittschof et al. 1992), collembolans (Purrington et al. 1991), and echinoderms (Snyder and Snyder 1970; Lawrence 1991) and vertebrates such as fishes (e.g., darters, gobies, sculpins, minnows, sticklebacks; reviews in Pfeiffer 1977 and Smith 1992; see also Mathis and Smith 1993a), anuran amphibians (Pfeiffer 1974; Hews 1988), and, possibly, caudate

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amphibians (G. A. Marvin and V. H. Hutchison, unpublished data). The chemical alarm pheromone, or "Schreckstoff," of fishes in the superorder Ostariophysi, which includes over 5,000 species (more than 70% of all freshwater fish species), probably has been the best studied. However, even for ostariophysans, hypotheses concerning the evolution of chemical alarm signals remain speculative as few empirical tests have been done.

Many small fishes in natural populations exhibit scars from failed predator attempts (Smith and Lemly 1986; Reimchen 1988); therefore, senders may survive predatory attacks and thus potentially derive direct benefits from the release of Schreckstoff. Recently, the alarm pheromone of ostariophysan fishes has been suggested to be analogous to the distress calls made by some birds following capture by predators (Smith 1992). In birds, these calls appear to function to attract additional predators that disrupt the predation event, which allows the prey an opportunity for escape (Perrone 1980; Högstedt 1983; Koenig et al. 1991). It has been proposed (Smith 1992) that the alarm pheromone of fishes may similarly provide direct benefits to the sender by attracting additional predators, which would result in an increase in the prey's probability of escape. If this hypothesis is true, then predators should be attracted to areas where the alarm pheromone is present.

In contrast to the predator-attraction hypothesis, the alarm pheromone may possibly function to repel predators (Williams 1964, 1992). If this hypothesis is true, then predators should avoid areas marked by the alarm pheromone. The repellent function may be manifested in several ways. For example, the chemical that is produced may be distasteful, may startle the predator, or may communicate other information to the predator. In some species of birds and mammals, individuals give alarm calls that appear to be directed toward the predator, possibly to inform the predator that it has been sighted and that the prey is alert (see, e.g., Woodland et al. 1980; Sherman 1985; Hasson 1991). Such calls may result in reduced probability of capture for the signaling prey (Sherman 1985).

We distinguished between the predator-deterrence and predator-attraction hypotheses by quantifying responses of two different predators, northern pike (*Esox lucius*) and predaceous diving beetles, to the alarm pheromone of fathead minnows (*Pimephales promelas*). Other hypotheses, including an antipathogen function that would aid in the healing of damaged tissue, were not addressed by our study (see Smith 1977, 1992 for reviews of other hypotheses).

#### EXPERIMENT IA: ATTRACTION OF PIKE TO ALARM SUBSTANCE PREPARED FROM SKIN EXTRACT

##### *Methods*

*Subjects.*—Fathead minnows are widely distributed throughout the central and northern portions of the United States and throughout most of the Canadian provinces. They may be locally abundant in ponds, lakes, or streams and generally are important forage fishes (Scott and Crossman 1973). For fathead minnows and other ostariophysans, the alarm pheromone ("alarm substance," or AS) is

produced by epidermal club cells and is released only by mechanical damage to the skin (Smith 1986). It has been calculated (Lawrence and Smith 1989) that a single AS cell contains enough pheromone to create an active space of about 80 L; a square centimeter of skin would contain enough pheromone to fill a cube that was 3.9 m on each side (Smith 1992). Conspecifics, and sometimes heterospecifics (Mathis and Smith 1993a), that detect the alarm substance perform fright responses such as decreasing activity (Lawrence and Smith 1989), increasing shelter use (Mathis and Smith 1993b), or increasing shoaling (Mathis and Smith 1993b). Alarm substance cells develop within the first few months of life (Waldman 1982) and are present in both females and nonbreeding male fathead minnows (Smith and Murphy 1974). Breeding males develop a dorsal mucus pad and tubercles and lose their alarm substance cells (Smith 1973; Smith and Murphy 1974).

Northern pike are common predators in freshwater systems in both North America and Eurasia. Pike are generally solitary predators (Eklöv 1992), and their diet consists primarily of other fish (Lawler 1965). Both pike (Savino and Stein 1989) and fathead minnows (Sullivan and Atchison 1978) commonly occupy vegetative microhabitats and are often in close proximity. Large pike probably ingest minnows quickly with little mechanical damage to the minnows' skin. However, because of the pike's juvenile-biased population structure (see, e.g., Treasurer et al. 1992), most encounters are probably between minnows and juvenile pike. Pike become fish predators at about 35 mm in length (Treasurer et al. 1992) and can consume minnows of approximately the same size as themselves (A. Mathis, D. P. Chivers, and R. J. F. Smith, personal observation). Smaller pike generally take a considerable amount of time to manipulate the minnow into a headfirst position for swallowing (in our laboratory observations, a handling time of several minutes is not unusual). During manipulation of the prey, there is a great deal of damage to the minnows' skin (A. Mathis, D. P. Chivers, and R. J. F. Smith, personal observation). In addition to pike, a number of other animals also act as predators of fathead minnows, including other fishes (Savino and Stein 1989), birds (McIntyre 1988), and aquatic invertebrates, such as diving beetles (see experiment 2).

*Collection and maintenance.*—Northern pike were captured from Pike Lake in central Saskatchewan. Pike were maintained in artificial stream tanks (300 L) at approximately 15°C on a 14L:10D cycle and were fed once every 5 d with one to two fathead minnows. We tested 18 pike (mean fork length  $\pm$  1 SD = 17.2  $\pm$  4.44 cm).

*Alarm substance (skin extract) preparation.*—Predators might be attracted to skin extract from fathead minnows regardless of the presence of the alarm pheromone. Therefore, we used skin extract from breeding male fathead minnows as a control stimulus; males lose their alarm substance cells when they are in breeding condition (Smith 1973). The experimental stimulus was an extract from the skin of nonbreeding minnows (with AS). Separate skin extract solutions were made from 15 donor individuals each for nonbreeding fathead minnows (with AS) (12 males, three females; mean fork length  $\pm$  1 SD = 5.07  $\pm$  0.501 cm) and breeding male fathead minnows (without AS) (mean fork length  $\pm$  1 SD = 5.67  $\pm$  0.294 cm). Donor fishes were killed by a blow to the head, and most of the skin was

removed from both sides of each fish<sup>1</sup>. The skin samples were immediately placed in 50 mL of chilled glass-distilled water (approximately 5°C) and were homogenized with a Polytron homogenizer. The homogenates were filtered through glass wool to remove scales and other solid particles, and each solution was further diluted by the addition of 350 mL of glass-distilled water (total volume for each stimulus solution, 400 mL). The homogenates were stored in a freezer at approximately -20°C until used. Frozen skin extracts from nonbreeding fathead minnows retain the capacity to elicit fright responses from conspecifics (see, e.g., Lawrence and Smith 1989).

*Testing tanks.*—Pike were tested in aquaria (90 cm × 45 cm × 50 cm) that contained water to a depth of 15 cm. The aquaria were modified to allow for dichotomous choice testing. A widthwise removable screen barrier was placed 28 cm from one end of the aquarium, which formed an acclimation area for the pike. This area also served as a “neutral zone” during testing. An opaque plastic barrier that was impermeable to water divided the remainder of the tank lengthwise into two equally sized compartments. Two artificial plants and one rock were placed against the back wall of each compartment, and an airstone was placed behind the plants. Stimulus solutions were injected into each compartment through Tygon tubing (length, 3.0 m; diameter, 0.5 cm) that was tied to the air line. The end of the tubing was approximately 1 cm from the surface of the airstone so that the inflow of air caused the stimulus water to be rapidly dispersed throughout the compartments.

*Testing protocol.*—Pike were tested separately. Each pike was placed into the acclimation area (behind the removable screen barrier) of a randomly selected testing tank. Testing occurred the day following introduction of the pike into the tank. During each test, an observer sat approximately 1 m in front of the testing chambers, facing the neutral zone. Immediately prior to testing, 60 mL of tank water were drawn through each stimulus-injection tube and discarded to remove any stagnant water that may have collected in the tubing. An additional 60 mL of tank water were drawn through each stimulus injection tube and saved. Five milliliters of the AS skin extract were injected into one stimulus-injection tube at the same time that 5 mL of the “no-AS” skin extract were simultaneously injected into the other stimulus-injection tube. The compartment (right vs. left sides) that received the AS stimulus was determined randomly for each test. Injection of the stimulus solutions was immediately followed by the simultaneous injection of 60 mL of tank water into both stimulus-injection tubes. Trials with vegetable dyes demonstrated that it took approximately 3 min following injection for the dye to reach the pike’s chamber.

Observations began immediately following the injection of the tank water. At 15-s intervals, we recorded (1) whether the pike was located in front of the AS or no-AS compartments (if part of the pike’s body was between the two compartments, we recorded the chamber that contained >50% of the pike’s body) and (2) whether the pike was facing (oriented toward) the AS or no-AS compartment or one of the sides of the neutral compartment. After 10 min of observation, we removed the screen barrier, which allowed the pike access to the AS and no-AS compartments. During the post-barrier-removal period, we also measured the

amount of time that the pike spent in the AS, no-AS, and neutral compartments, in addition to the indexes of behavior described above. Tests were conducted between 0750 and 1530 hours at approximately 22°C.

For each of the recorded categories of behavior, we compared the observed data to a theoretical random distribution using two-tailed Kolmogorov-Smirnov one-sample tests (Siegel 1956). The three indexes used were the percentage of neutral zone locations on the AS side of the tank (including both pre- and post-removal of the screen barrier), percentage of neutral zone orientations that were toward the AS side of the tank (incidences of orientation away from both compartments were deleted), and percentage of time the pike spent in the AS compartment (time in the neutral zone was not considered in the analysis).

### *Results*

The location of pike in the neutral zone (AS vs. no-AS sides) was not significantly different from random (mean percentage time on AS side  $\pm 1$  SD = 50.2%  $\pm$  43.64;  $D = 0.23$ ,  $P > .20$ ). However, while in the neutral zone, pike spent significantly more time orienting toward the AS compartment than expected by chance (fig. 1a;  $D = 0.61$ ,  $P < .01$ ).

Seven of the 18 pike remained in the neutral zone for the entire test. For the 11 pike that entered the stimulus compartments, the distribution of time spent in the two compartments was significantly different from random, with pike spending, on the average, more time in the AS compartment (fig. 1a;  $D = 0.72$ ,  $P < .01$ ). Ten of the 11 pike spent more time in the AS compartment than in the no-AS compartment, with nine of those pike never entering the no-AS compartment.

#### EXPERIMENT 1B: ATTRACTION OF PIKE TO AN ARTIFICIAL ALARM SUBSTANCE (HYPOXANTHINE-3(N)-OXIDE)

### *Methods*

The active component of the skin extract has been tentatively identified as hypoxanthine-3(N)-oxide (Argentini 1976; Pfeiffer et al. 1985). We obtained a commercial preparation of hypoxanthine-3(N)-oxide (artificial alarm substance, or AAS) from Menai Organics Limited (Gwynedd, N. Wales, U.K.). Sixteen milligrams of the AAS were dissolved in 115 mL of glass-distilled water, and the resulting solution was pipetted into polypropylene vials in 5-mL units. The vials were placed into a  $-20^{\circ}\text{C}$  freezer; control vials containing 5 mL of glass-distilled water were also frozen at  $-20^{\circ}\text{C}$ . This experiment followed the same protocol as experiment 1a except that the AAS was used as the AS stimulus, and glass-distilled water was used as the no-AS stimulus. The same pike that were used in experiment 1a were also tested in this experiment; two pike died between the two experiments, so only 16 pike were tested in experiment 1b.

### *Results*

The results of this experiment were similar to those of experiment 1a. The percentage of time that the pike spent on the AAS side of the neutral zone was

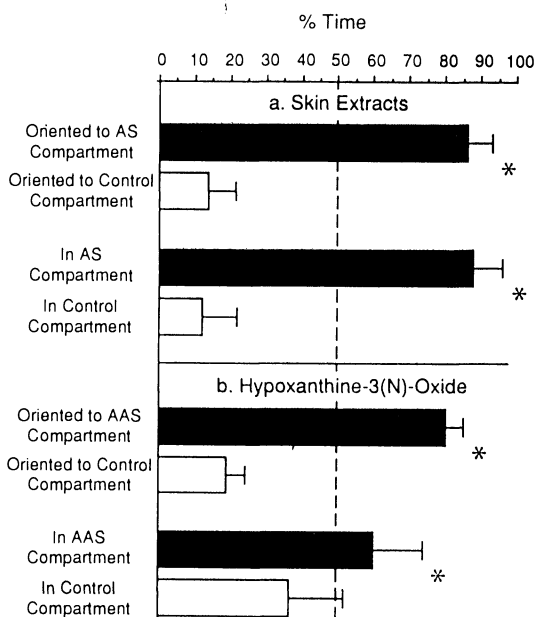


FIG. 1.—Percentage time (mean  $\pm$  1 SE) that pike spent either oriented toward or located in the compartments containing the AS or a control stimulus. *a*, The AS stimulus was skin extract from nonbreeding fathead minnows (with alarm substance cells), and the control stimulus was extract from breeding male fathead minnows (without alarm substance cells). *b*, The AAS stimulus was a solution of hypoxanthine-3(N)-oxide dissolved in distilled water, and the control stimulus was distilled water. Each of the variables was significantly different from random (*dashed line*). An asterisk indicates the results of a nonparametric Kolmogorov-Smirnov test: *a*,  $N = 18$ ; *b*,  $N = 16$ ;  $P < .01$ .

not significantly different from random (mean percentage time on AAS side  $\pm$  1 SD = 53.8%  $\pm$  39.34;  $D = 0.24$ ,  $P > .20$ ). However, while in the neutral zone, pike spent significantly more time orienting toward the AAS compartment than expected by chance (fig. 1*b*;  $D = 0.51$ ,  $P < .01$ ).

Six of the 16 pike that were tested remained in the neutral zone for the entire test. For the 10 pike that left the neutral zone, the distribution was also significantly different from random, with pike spending, on the average, a larger percentage of their time in the AAS compartment (fig. 1*b*;  $D = 0.50$ ,  $P < .01$ ). Qualitatively, this response appears weaker than the other responses. Six of the 10 pike that left the neutral zone spent 100% of their time in the AAS compartment, while the other four pike spent more of their time in the no-AAS compartment.

#### EXPERIMENT 2: ATTRACTION OF PREDACEOUS DIVING BEETLES TO ALARM SUBSTANCE

##### *Methods*

We conducted a field experiment to determine whether predaceous diving beetles (Dytiscidae) were attracted to the fathead minnow alarm substance. To pro-

vide marked areas, we saturated cellulose sponges (1.9 cm × 1.9 cm × 1.9 cm) with the stimulus solutions and placed them into funnel traps. Skin extracts were prepared using the same technique as in experiment 1a, except that the solutions were diluted to a total volume of 200 mL rather than 400 mL. The AS stimulus was prepared from the skin of six nonbreeding male fathead minnows (mean fork length ± 1 SD = 6.45 ± 0.394 cm), and the no-AS stimulus was prepared from the skin of six breeding male fathead minnows (mean fork length ± 1 SD = 5.67 ± 0.266 cm). The saturated sponges were placed in a freezer at approximately -20°C until the day of testing and were kept on ice until the beginning of the experiment.

Traps (Gee's Improved Minnow Traps) consisted of roughly cylindrical wire enclosures (43 cm length × 22 cm diameter) with a funnel located at each end leading into the trap. Funnel entrances were approximately 2.5 cm in diameter. Two sponges from the same treatment (either AS or no AS) were placed into each trap so that one sponge was approximately 2 cm in front of each trap entrance. The two sponges were held in place with wire. Sixteen traps contained sponges saturated with AS skin extracts from nonbreeding male fathead minnows, and 16 traps contained sponges saturated with no-AS extracts from breeding male fathead minnows.

The experiment was conducted in Lakeview Pond, an artificial pond located in Saskatoon, Saskatchewan. The pond contained both fathead minnows and dytiscid beetles (A. Mathis, D. P. Chivers, and R. J. F. Smith, personal observation). Pairs of AS and no-AS traps were simultaneously placed into the water along the shoreline so that traps from the two treatment groups spent exactly the same amount of time in the water. Successive traps were separated by approximately 10 m. The linear order of placement of AS and no-AS traps was determined randomly with the proviso that no more than two traps in a row could be of the same treatment condition. Traps were placed into the water at approximately 2130 hours (immediately prior to sunset) and were removed at approximately 0630 hours (immediately after sunrise) the following day. The traps were left in the water overnight because adult diving beetles are most active at night (Holomuzki 1985). Pairs of AS and no-AS traps were removed simultaneously from the water. Dytiscid beetles from each trap were preserved in 40% formalin (later transferred to 70% ethanol) and were identified for species. Minnows caught in the traps were identified, counted, and immediately released at the capture site. Statistical comparisons of the number of dytiscid beetles in AS versus no-AS traps were made using two-tailed Mann-Whitney *U*-tests (Siegel 1956).

### Results

All 58 of the beetles that were captured in the traps belong to the family Dytiscidae, and all are predaceous. Overall, significantly more beetles were captured in the AS traps than in the no-AS traps (table 1; Mann-Whitney *U* = 68, *P* < .05). The beetles belonged to seven different species in four different genera (table 1). Of the seven species that were captured, only *Colymbetes sculpilis* was present in sufficient numbers for a separate statistical analysis. Significantly more *C.*

TABLE 1

PREDACEOUS DIVING BEETLES (DYTISCIDAE) AND FATHEAD MINNOWS (*PIMEPHALES PROMELAS*) CAUGHT IN TRAPS MARKED WITH SKIN EXTRACT FROM EITHER NONBREEDING MALE FATHEAD MINNOWS WITH ALARM SUBSTANCE CELLS (AS) OR FROM BREEDING MALE FATHEAD MINNOWS WITHOUT ALARM SUBSTANCE CELLS (CONTROL)

TAXON	TOTAL NUMBER (RANGE PER TRAP)		P
	AS	Control	
Fish:			
<i>Pimephales promelas</i>	0	54 (0-48)	...
Beetles:			
<i>Acilius semisulcatus</i>	1 (0-1)	1 (0-1)	...
<i>Colymbetes sculptilis</i>	20 (0-3)	7 (0-2)	<.05
<i>Dytiscus alaskanus</i>	9 (0-2)	5 (0-2)	...
<i>Dytiscus circumcinctus</i>	1 (0-1)	1 (0-1)	...
<i>Dytiscus cordieri</i>	0	1 (0-1)	...
<i>Graphoderus occidentalis</i>	5 (0-2)	2 (0-1)	...
<i>Graphoderus perplexus</i>	3 (0-1)	1 (0-1)	...
Total beetles	39 (0-7)	18 (0-5)	<.05

*sculptilis* were present in the AS traps than in the no-AS traps (table 1;  $U = 69.5$ ,  $P < .05$ ).

Fathead minnows were the only species of fishes caught in the traps during the experiment. All of the 54 minnows that were caught were present in two of the no-AS traps. While these data are limited, they are consistent with previous observations (Mathis and Smith 1992) that fathead minnows avoid traps marked with the alarm pheromone.

#### DISCUSSION

Our results clearly do not support the hypothesis that minnow alarm substance deters predators. This inference is consistent with palatability tests demonstrating that rainbow trout (*Oncorhynchus mykiss*) did not distinguish between fathead minnow prey with and without alarm substance cells (Bernstein and Smith 1983). Furthermore, fathead minnows and other ostariophysan fishes are readily consumed by natural predators (e.g., snakes: Herzog and Burghardt 1988; birds: McIntyre 1988; fishes: Smith and Moyle 1945; Savino and Stein 1989) and are often cultured for use as bait (see, e.g., Brown and Gratzek 1980; Martin 1986). It is possible that the alarm pheromone may produce a predator-deterrent effect in conditions different from those in our experiments. For example, the pike (and presumably the diving beetles) in our study were experienced predators; the alarm pheromone may have a deterrent effect only on naive predators. However, we feel that the evidence to date indicates that the predator-deterrence hypothesis is unlikely to account for the evolution of minnow alarm substance.

The hypothesis that predators are attracted by minnow alarm substance is

supported by our results. In addition to the possible relevance of this finding to the evolution of alarm substance cells (see below), these results provide other important insights into the operation of the ostariophysan alarm substance system. First, the significance of the involuntary release mechanism (only following mechanical damage to the skin) has previously been puzzling. If senders benefit either directly or indirectly by warning conspecifics, then the release mechanism should be voluntary, allowing warnings to be broadcast any time a predator is detected rather than only after the sender has been captured. However, if the alarm signal is attractive to predators, the costs of such warnings may be prohibitively high. Second, our results also may explain the loss of alarm substance cells by breeding minnows (Smith 1977). During courtship and mating, fathead minnows defend spawning sites for several days and rub against spawning surfaces, potentially damaging the epithelium and releasing alarm substance (Smith 1973, 1977). Until now, the only hypothesis proposed for the loss of alarm substance cells by breeding minnows has been that inadvertent release of alarm substance may frighten away potential mates (Smith 1977, 1992). Attraction of predators to spawning territories may be an even stronger selective force favoring seasonal loss of alarm substance cells.

Although the focus of this article has been possible benefits for senders of the alarm, the selection pressures on predators are also of interest. Both pike and predaceous beetles found skin extracts with alarm substance more attractive than extracts without alarm substance. From the predator's perspective, attraction to the alarm substance did not evolve because of benefits to the prey but because it may help predators locate their next meal, either by stealing prey (or pieces of the prey) from the first predator or by eating the first predator. Interestingly, if the alarm pheromone attracts predators of the initial predator, then the inclusive fitness of the sender may benefit by the removal of potential predators of close relatives from the population regardless of whether the sender survives the encounter; a similar function has been suggested for the bioluminescence of many marine invertebrates (Gardiner 1972). However, attraction of additional predators to the alarm pheromone might evolve even in the absence of benefits to the receivers.

In addition to predator deterrence and predator attraction, several other hypotheses have been suggested to account for the evolution of alarm substance cells by senders (Smith 1977, 1986, 1992). These hypotheses, including an anti-pathogen function and indirect benefits via kin selection, are not necessarily incompatible with our results and require further testing.

We suggest that damage-released alarm pheromones may have evolved under different selection pressures than alarm pheromones with voluntary release mechanisms. These chemical alarm signals, at least for ostariophysan fishes, appear to function in a fashion analogous to that of distress calls in birds (Perrone 1980; Högstedt 1983; Koenig et al. 1991) by attracting other predators. Furthermore, the relevance of the predator-attraction hypothesis may extend to a wide variety of taxa in which an alarm signal is given following capture by a predator. Distress calls following capture are exhibited by a number of vertebrates (e.g., juvenile alligators: Staton 1978; fishes: Myrberg 1981; mammals: Sherman 1985; frogs:

Kanamadi et al. 1993). Distress calls and damage-released alarm pheromones may therefore be functionally distinct from other alarm signals, at least from the sender's viewpoint.

Although production of a signal by prey to attract predators may seem counter-intuitive, the minnow alarm substance, like a distress call, is only produced when senders are in dire circumstances and have little to lose. In laboratory tanks, we have observed instances in which minnows were captured by pike but escaped following interference by another pike. Interference can be common in laboratory tanks. For example, in a 200-L stream tank containing three pike (mean  $\pm$  1 SD fork length =  $22.3 \pm 1.53$ ), we observed 10 cases of interference when feeding the pike a total of 30 minnows over four different feeding sessions. In two of these 10 cases, the minnows temporarily escaped from the pike. Interference may take several forms. Most commonly in our observations, the second pike attempted to grab the minnow from the first pike, which was manipulating the minnow with its mouth. Other forms of interference occurred when the second pike swam so that it collided with the first pike or bit the pike around the opercula, which caused it to open its mouth and allowed the prey an opportunity to escape. Because pike are notoriously cannibalistic (see, e.g., Lawler 1965), pike in the laboratory must be kept with individuals of similar size. We suspect that as the size difference between the pike increases, interference may be more common. It is easy to imagine a scenario in which a small pike that captures a minnow would immediately release the minnow if a larger pike approached. The frequency of interference may also increase as the hunger level of the pike increases. In addition to its alarm function (Mathis and Smith 1993*b*), the Schreckstoff may also increase the prey's probability of escape by attracting other predators, which thus provides direct benefits to the sender.

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#### LITERATURE CITED

- Argentini, M. 1976. Isolierung des Schreckstoffes aus der Haut der Elritze *Phoxinus phoxinus* (L.). Ph.D. diss. University of Zürich.
- Bernstein, J. W., and R. J. F. Smith. 1983. Alarm substance cells of fathead minnows do not affect the feeding preference of rainbow trout. *Environmental Biology of Fishes* 9:307–311.
- Brown, E. E., and J. B. Gratzek. 1980. *Fish farming handbook*. Avi, Westport, Conn.
- Eklöv, P. 1992. Group foraging versus solitary foraging efficiency in piscivorous predators: the perch, *Perca fluviatilis*, and pike, *Esox lucius*, patterns. *Animal Behaviour* 44:313–326.
- Gardiner, M. S. 1972. *The biology of invertebrates*. McGraw-Hill, New York.
- Hasson, O. 1991. Pursuit-deterrent signals: communication between prey and predator. *Trends in Ecology & Evolution* 6:325–329.

- Herzog, H. A., and G. M. Burghardt. 1988. Development of antipredator responses in snakes. III. Stability of individual and litter differences over the first year of life. *Ethology* 77:250–258.
- Hews, D. K. 1988. Alarm response in larval western toads, *Bufo boreas*: release of larval chemicals by a natural predator and its effect on predator capture efficiency. *Animal Behaviour* 36:125–133.
- Högstedt, G. 1983. Adaptation unto death: function of fear screams. *American Naturalist* 121:562–570.
- Holomuzki, J. R. 1985. Life history aspects of the predaceous diving beetle *Dytiscus dauricus* (Gebler) in Arizona. *Southwestern Naturalist* 30:485–490.
- Howe, N. R., and Y. M. Sheikh. 1975. Anthopleurine: a sea anemone alarm pheromone. *Science* (Washington, D.C.) 189:386–388.
- Kanamadi, R. D., H. Schneider, C. R. Hiremath, and C. S. Jirankali. 1993. Vocalization of the tree frog *Polypedates maculatus* (Rhacophoridae). *Journal of Biosciences* 18:239–245.
- Koenig, W. D., M. T. Stanback, and P. N. Hooge. 1991. Distress calls in the acorn woodpecker. *Condor* 93:637–643.
- Lawler, G. H. 1965. The food of the pike, *Esox lucius*, in Heming Lake, Manitoba. *Journal of the Fisheries Research Board of Canada* 22:1357–1377.
- Lawrence, B. J., and R. J. F. Smith. 1989. The behavioral response of solitary fathead minnows, *Pimephales promelas*, to alarm substance. *Journal of Chemical Ecology* 15:209–219.
- Lawrence, J. M. 1991. A chemical alarm response in *Pycnopodia helianthoides* (Echinodermata: Asteroidea). *Marine Behavior and Physiology* 19:39–44.
- Martin, M. 1986. The fathead minnow: an overview on propagation, culturing and uses. *Aquaculture Magazine* 12:48–50.
- Mathis, A., and R. J. F. Smith. 1992. Avoidance of areas marked with a chemical alarm substance by fathead minnows (*Pimephales promelas*) in a natural habitat. *Canadian Journal of Zoology* 70:1473–1476.
- . 1993a. Intraspecific and cross-superorder responses to chemical alarm signals by brook stickleback. *Ecology* 74:2395–2404.
- . 1993b. Chemical alarm signals increase the survival time of fathead minnows (*Pimephales promelas*) during encounters with northern pike (*Esox lucius*). *Behavioral Ecology* 4:260–265.
- McIntyre, J. W. 1988. *The common loon, spirit of northern lakes*. University of Minnesota Press, Minneapolis.
- Myrberg, A. A. 1981. Sound communication and interception in fishes. Pages 395–452 in W. Tavolga, A. N. Popper, and R. R. Fay, eds. *Hearing and sound communication in fishes*. Springer, New York.
- Perrone, M., Jr. 1980. Factors affecting the incidence of distress calls in passerines. *Wilson Bulletin* 92:404–408.
- Pfeiffer, W. 1974. Pheromones in fish and Amphibia. Pages 269–296 in M. C. Birch, ed. *Pheromones: frontiers of biology*. Vol. 32. North-Holland, Amsterdam.
- . 1977. The distribution of fright reaction and alarm substance cells in fishes. *Copeia* 1977:653–665.
- Pfeiffer, W., G. Riegelbauer, G. Meir, and B. Scheibler. 1985. Effect of hypoxanthine-3(N)-oxide and hypoxanthine-1(N)-oxide on central nervous excitation of the black tetra *Gymnocorymbus ternetzi* (Characidae, Ostariophysi, Pisces) indicated by dorsal light response. *Journal of Chemical Ecology* 11:507–524.
- Purrlington, F. F., P. A. Kendall, J. E. Bater, and B. R. Stinner. 1991. Alarm pheromone in a gregarious poduromorph collembolan (Collembola: Hypogastruridae). *Great Lakes Entomologist* 24:75–78.
- Reimchen, T. E. 1988. Inefficient predators and prey injuries in a population of giant stickleback. *Canadian Journal of Zoology* 66:2036–2044.
- Rittschof, D., D. W. Tsai, P. G. Massey, L. Blanco, G. L. Kueber, Jr., and R. J. Haas, Jr. 1992. Chemical mediation of behavior in hermit crabs: alarm and aggregation cues. *Journal of Chemical Ecology* 18:959–984.
- Savino, J. F., and R. A. Stein. 1989. Behavioural interactions between fish predators and their prey: effects of plant density. *Animal Behaviour* 37:311–321.

- Scott, W. B., and E. J. Crossman. 1973. Freshwater fishes of Canada. Bulletin of the Fisheries Research Board of Canada 184:480-484.
- Sherman, P. W. 1985. Alarm calls of Belding's ground squirrels to aerial predators: nepotism or self-preservation? Behavioral Ecology and Sociobiology 17:313-323.
- Siegel, S. 1956. Nonparametric statistics for the behavioral sciences. McGraw-Hill, New York.
- Sleeper, H. L., V. J. Paul, and W. Fenical. 1980. Alarm pheromones from the marine opisthobranch *Navanax inermis*. Journal of Chemical Ecology 6:57-70.
- Smith, L. L., and J. B. Moyle. 1945. Factors influencing production of yellow pike-perch, *Stizostedion vitreum citreum*, in Minnesota rearing ponds. Transactions of the American Fisheries Society 73:243-261.
- Smith, R. J. F. 1973. Testosterone eliminates alarm substance in male fathead minnows. Canadian Journal of Zoology 41:875-876.
- . 1977. Chemical communication as adaptation: alarm substance of fish. Pages 303-320 in D. Müller-Schwarze and M. M. Mozell, eds. Chemical signals in vertebrates. Plenum, New York.
- . 1986. The evolution of chemical alarm signals in fishes. Pages 99-115 in D. Duvall, D. Müller-Schwarze, and R. M. Silverstein. Chemical signals in vertebrates. Vol. 4. Plenum, New York.
- . 1992. Alarm signals in fishes. Reviews in Fish Biology and Fisheries 2:33-63.
- Smith, R. J. F., and A. D. Lemly. 1986. Survival of fathead minnows after injury by predators and its possible role in the evolution of alarm signals. Environmental Biology of Fishes 15:147-149.
- Smith, R. J. F., and B. D. Murphy. 1974. Functional morphology of the dorsal pad in fathead minnows (*Pimephales promelas* Rafinesque). Transactions of the American Fisheries Society 103:65-72.
- Snyder, N. F. R., and H. A. Snyder. 1970. Alarm response of *Diadema antillarum*. Science (Washington, D.C.) 168:276-278.
- Staton, M. A. 1978. "Distress calls" of crocodylians—whom do they benefit? American Naturalist 112:327-332.
- Sullivan, J. F., and G. J. Atchison. 1978. Predator-prey behavior of fathead minnows, *Pimephales promelas*, and largemouth bass, *Micropterus salmoides*, in a model ecosystem. Journal of Fish Biology 13:249-253.
- Taylor, R. J., D. F. Balph, and M. H. Balph. 1990. The evolution of alarm calling: a cost-benefit analysis. Animal Behaviour 39:860-868.
- Treasurer, J. W., R. Owen, and E. Bowers. 1992. The population dynamics of pike, *Esox lucius*, and perch, *Perca fluviatilis*, in a simple predator-prey system. Environmental Biology of Fishes 34:65-78.
- Waldman, B. 1982. Quantitative and developmental analysis of the alarm reaction in the zebra danio, *Brachydanio rerio*. Copeia 1982:1-9.
- Williams, G. C. 1964. Measurement of consociation among fishes and comments on the evolution of schooling. Publications of the Museum, Michigan State University Biological Series 2:349-384.
- . 1992. Natural selection: domains, levels, and challenges. Oxford University Press, Oxford.
- Woodland, D. J., Z. Jaafar, and M.-L. Knight. 1980. The "pursuit deterrent" function of alarm signals. American Naturalist 115:748-753.

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