

Introduction

In a variety of animals, behavioral responses to cues from potential predators are common, and are to be expected if these responses decrease the probability of predation (Mathis and Smith 1993). Differences with regards to responses to predator cues among related species are of particular interest when such differences can alter competitive interactions (Werner and Anholt 1993). These differences can potentially alter the composition of biological communities (Hazlett and Schoolmaster 1998).

The use of chemical signals by aquatic organisms has been documented in a variety of contexts. The ability to gather information from the chemical medium is of great selective advantage to many aquatic organisms from different taxa and functional groups (Atema 1985), particularly when their habits are nocturnal (Brönmark & Hansson 2000), such as crayfish. In aquatic systems, chemical cues are one of the most commonly utilized classes of predator avoidance (Chivers et al. 1996; Kiesecker et al. 1996; Smith 1992). Some species of crayfish respond to predator odors, while others do not respond to these chemical cues (Hazlett and Schoolmaster 1998). When *Orconectes propinquus* or *Orconectes virilis* individuals were exposed to odors from largemouth bass, no changes in behavior were reported (Willman et al. 1994). When individuals of *Orconectes rusticus* were exposed to the same chemical cues, their time spent out of shelters in the presence of the fish predator increased.

In addition to response to chemical cues of predator avoidance, alarm responses shown to be substances released from physically damaged conspecific individuals have been reported in a variety of fish (Smith 1992), amphibians (Pfeiffer 1963), marine gastropods (Stenzler and Atema 1976), echinoderms (Snyder and Snyder 1970), and crayfish (Hazlett 1994). Alarm responses reduce the chances of predation (Mathis and Smith 1993). These alarm responses have been documented many species of crayfish, such as *Orconectes rusticus*, *O. virilis*, and *O. propinquus*, respond to damaged conspecifics, commonly known as alarm signals, which produce chemical signals composed of bodily fluids that can alert the individual to a proximate predation event (Wolf and Moore, 2002). Crayfish also utilize olfactory senses heavily while scavenging for various food sources (Willman et al., 1994). In the presence of a damaged individual, it is hypothesized that an individual will realize the potential danger and alter its foraging behaviors accordingly.

This experiment was conducted to determine whether foraging time is altered in response to alarm cues among *Orconectes virilis* and *Orconectes propinquus* by exposing each species to food along with its own specific alarm signal. We hypothesize that crayfish in the presence of alarm signals as well as food will display more nervous behaviors and spend less time foraging than crayfish that are not exposed to the alarm signals.

Materials and Methods

Individuals of the most common crayfish species (Family Cambaridae) in Michigan were tested in the laboratory to monitor foraging behavior. *Orconectes virilis* were collected from Burt Lake in Cheboygan County, Michigan, and *O. propinquus* were collected from the Maple River in Emmet County Michigan, and were tested at the Lakeside Laboratory of the University of Michigan Biological Station in Pellston, Michigan.

Crayfish were placed in 10-gallon aquaria, with fifteen crayfish in each aquarium. Halves of clay pots were placed at the bottom of each aquarium in order to provide a shelter for each crayfish. These aquaria were isolated from each other and each contained 12.5 liters of lake water, which was continually aerated using air stones, plastic tubing, and an air valve. Crayfish were allowed three days to acclimate to the aquariums, and each crayfish was tested just once. The crayfish were not fed during the three-day acclimation period in order to encourage foraging behaviors during observation periods. Behavioral patterns were noted and the temporal duration of patterns recorded for eight minutes per test.

The test tanks were also 10-gallon tanks filled by 12.5 liters of continually aerated lake water and each tank contained one half of a clay pot, placed at one end of the tank, to act as a shelter. Three different control parameters were run, and were later followed by the experimental tests. These control parameters were as follows: crayfish without food or alarm signal, crayfish with food, and crayfish with alarm signal. Eighteen control trials were performed for each parameter. The food or alarm scent was added to the tank, before the crayfish, to the opposite side of the tank from the shelter, approximately 0.5 meters away. The alarm scent was made by removing two claws from the species of crayfish being studied and crushed in one liter of water. The food was three grams of canned sardines.

The crayfish were added to each and observed for eight minutes, and the behavior was monitored to see how much time was spent in the shelter, how much time was spent outside the shelter, and how much time was spent eating. After each parameter, the tanks were dumped and rinsed out to eliminate any traces of food or alarm scents left over from previous tests. The tanks were also cleaned and refilled between tests for different species to eliminate any confounding variables associated with such tests.

Once the controls were tested, the experiment was performed using the same test tanks and procedures that were used for the controls. Food and alarm scent were added first at approximately the same time, then the crayfish were added and observed. This experimental test was repeated eighteen times.

After the data were collected, a univariate ANOVA was performed to determine whether food, alarm signal, or both had a significant effect on foraging time as well as if there was significant difference between the foraging times of each species.

Results

Significant data was obtained to suggest that the presence of food affects foraging time (Table I, $F=11.030$, $p=.000$). Alarm signals did not have a significant effect on foraging time ($F=2.367$, $p=.127$). Length also had a significant effect on foraging time (Fig. 1, $R^2=.065$, $p=.008$). There was also significant data to suggest a difference in foraging time between *O. propinquus* and *O. virillis* ($F=2.367$, $p=.001$) with *O. propinquus* having a significantly longer foraging time (Fig. 2).

Discussion

There was a significant difference for each control and the experimental conditions between the two species *Orconectes propinquus* and *O. virilis*. This result could stem from a few parameters. The two species are very closely related phylogenetically, but have demonstrated different levels of response to many chemical signals in previous studies (Troy Keller, personal communication). The *O. propinquus* spent much more time foraging, with and without the alarm scent present, than the *O. virilis*. There are a number of factors that could affect this finding. *O. virilis* are nocturnal animals, feeding only at night (Hazlett 1994), while *O. propinquus* eat during all hours of the day. Because the species feed at all times, *O. propinquus* has more of a fitness decline through competition (Hazlett 1994), possibly causing the individuals to feed as much as possible when food was present. Because *O. virilis* only feed at night, they possibly don't have to forage as much because there is less competition at night therefore the species hasn't evolved the necessity to eat as much food is present, for food is abundant during the night hours where less individuals are feeding. The general foraging behavior of each species would also lead to the predictable disparity between results of foraging time for each species. *O. virilis* generally do not feed upon recently killed prey, rather waiting for the prey to be safer and more decayed. *O. propinquus*, on the other hand, will generally feed on anything in the water generally accepted as food. This could mean that instead of recognizing the damage of conspecifics as an alarm signal, the chemical signal was recognized as food (Hazlett 1994).

The results of this study demonstrate the strong effect that the presence of food has on the foraging behaviors of both *O. virilis* and *O. propinquus*. When the crayfish detects the presence of food, their foraging behaviors change. This is a predictable behavior, according to other studies that suggest that crayfish become bolder in the presence of food (Hazlett 1994).

Results from this study do not demonstrate a significant difference in foraging behavior in the presence of an alarm scent for neither *Orconectes virilis* nor *O. propinquus* individuals. For the individuals of *O. virilis*, these results are somewhat surprising. A wide variety of chemical signals, including alarm signals, have been demonstrated to influence the behavior of individuals of *O. virilis* (Hazlett 1994).

For individuals of *O. propinquus*, the observation that individuals did not show a statistically significant alteration of behavior when solutions made from crushed conspecifics were introduced, compared to the control solutions, was still

surprising, but less so. Individuals in the past have been shown not to demonstrate a response to disturbance pheromone (Hazlett 1990), to respond less to sex pheromone (Hazlett 1994), and even to not respond to crushed conspecifics (Hazlett 1994). It has been shown that *O. propinquus* have reduced sophistication of chemical communication compared to that of *O. virilis* (Fitzpatrick 1987), which is surprising considering how closely related the species are phylogenetically and ecologically (Fitzpatrick 1987).

Clearly, much work has to be done to explain the lack of correlation between presence of alarm response and foraging time. There are many factors that could have contributed to the lack of statistically significant results, which would be altered in future studies. The first recommendation for future study would be to do a paired experiment, keeping one individual in a tank, allowing acclimation time, and then testing the same individual for each control, just crayfish, crayfish and food, crayfish and alarm scent, and the experimental test. This would make the results more convincing for this would eliminate possible intraspecific factors that would affect results.

There are a few other courses for future study that would improve the experiment. Because presence of food was the only statistically significant factor affecting foraging time, testing the different effects of different food for the crayfish would be interesting, (freshly dead, long dead, frozen, etc.)

Another factor that would be interesting in future studies could be to study the different alarm responses. Using crushed conspecifics, not just claws, could elicit a different response. Also, the period between conspecific death and use for alarm scent (newly dead, decaying, frozen, etc.) could provide different results in another study.

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