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EFFECTS OF DIET ON LOCALIZED DEFECATION BY NORTHERN PIKE, *Esox lucius*

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Abstract—Fathead minnows (*Pimephales promelas*) are able to detect conspecific alarm pheromone in the feces of northern pike (*Esox lucius*) and have been shown to avoid areas labeled with the feces of pike that were fed minnows. The minnows did not avoid areas labeled with the feces of pike that were fed swordtails (*Xiphophorous helleri*), which lack ostariophysan alarm pheromone. In laboratory experiments, pike fed a diet of minnows localized their defecation away from their foraging area. It has been suggested that in doing so, pike may remove chemical cues that label their foraging area as dangerous to prey species. As yet there has been no conclusive evidence to support this hypothesis. In this experiment, we test the effects of different predator diets on localized defecation by pike. Pike were fed minnows, swordtails, or mice (*Mus musculus*). Swordtails and mice lack ostariophysan alarm pheromones. Area use and location of feces were recorded. Pike fed minnows spent significantly more time in the home area (i.e., area of the test tank where they were fed) and defecated significantly more often in the opposite end of the tank. Pike fed swordtails also exhibited a significant preference for the home area in area use, while those fed mice showed no such preference. When fed either swordtails or mice, there was no significant difference between the proportion of time spent and proportion of feces in each area of the test tank. These data suggest that pike localize their defecation only when consuming prey items containing alarm pheromone. The current findings support the hypothesis that pike localize their defecation to remove chemical cues from the foraging area of the home range in order to avoid chemically labeling their foraging area as dangerous to prey.

Key Words—Fathead minnow, *Pimephales promelas*, northern pike, *Esox lucius*, localized defecation, predator labelling, alarm substance.

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INTRODUCTION

Fathead minnows (*Pimephales promelas*) possess epidermal club cells, which, when mechanically damaged, release an alarm pheromone [alarm substance (AS) or Schreckstoff (Smith, 1986, 1992)]. Mechanical damage typically occurs when a minnow is attacked or captured by a predator. When detected by conspecifics, AS elicits a stereotypic fright response, characterized by a variety of antipredator behavior patterns (Heczko and Seghers, 1981; Lawrence and Smith, 1989; Krause, 1993; Mathis and Smith, 1993a). Mathis et al. (1995) have demonstrated that predatory northern pike (*Esox lucius*) are attracted to minnow alarm pheromone.

Mathis and Smith (1993a,b) have recently demonstrated that fathead minnows that had never encountered a pike responded with antipredator behavior to chemical stimuli (water containing urine, feces, mucus that had housed a pike) from pike if the pike had recently consumed minnows, but not swordtails (*Xiphophorus helleri*) or breeding male minnows. Swordtails lack ostariophysan alarm pheromones (Mathis and Smith, 1993a,b), and breeding male fathead minnows lose their alarm substance cells during the spawning season (Smith, 1973, 1976). Brown et al. (1995a,b) have demonstrated that AS is contained in feces of a predatory pike and that minnows that have never encountered a pike will avoid areas labeled with feces of pike that have been fed minnows but do not avoid feces of pike fed swordtails.

Brown et al. (1995a) showed that pike fed fathead minnows localize their defecation away from their foraging area. They suggested that pike may do so in order to remove chemical cues that could label their foraging territory as dangerous to prey fish. As yet, there has been no conclusive evidence to support this hypothesis. In the current study, we examine the effects of different diets on localized defecation by pike. We predict that if pike are localizing their defecation to remove possible chemical cues from their foraging range, then pike fed a diet which lacks AS should not show localized defecation.

METHODS AND MATERIALS

Test Fish. Northern pike were collected with seine nets from Van Patten's Creek in south central Saskatchewan. Pike were transported to the lab and held in 60-liter holding tanks at approximately 16°C, under a 14 : 10 light–dark cycle. Prior to the experiments, pike were fed one or two fathead minnows once every four days.

Fathead minnows were taken from Pike Lake, an oxbow lake of the South Saskatchewan River near Saskatoon, Saskatchewan. Minnows were held in 300-liter artificial stream channels under a 14 : 10 light–dark cycle. Minnows were

fed daily with commercial fish food. Swordtails (maintained on same diet as minnows) and frozen neonatal mice (*Mus musculus*) were obtained commercially. Swordtails were used as a fish prey because they lack ostariophysan alarm pheromone (Mathis and Smith, 1993a). It is unlikely that swordtails possess any form of alarm pheromone, as two additional species of *Xiphophorus* (*X. maculatus* and *X. variatus*) have been shown to lack alarm pheromones (Pfieffer, 1977). Mice were chosen as a representative terrestrial prey because they are taken opportunistically by inshore pike (Lawler, 1965; Scott and Crossman, 1973) and lack ostariophysan alarm pheromone. Mice possess disturbance chemical alarm signals, which are released in urine and/or feces when disturbed (Rottman and Snowdon, 1972). These cues are likely not present in our trials, since the mice were dead when fed to the pike. In addition, under natural conditions, mice would be swimming near the surface, while feces would be restricted to the substratum. Mice were thawed in water prior to use.

Test Tanks. Three, identical 500-liter glass aquaria (183 cm long × 49 cm wide × 56 cm high) were used as test tanks. The test tanks were not filtered, and there was no appreciable flow. A single airstone was suspended near the back wall of the tank, halfway down the length of the tank. We divided each tank into seven compartments, as in Brown et al. (1995a), by drawing gridlines on the exterior of the tank. The first compartment was 38 cm wide, and each of the remaining six compartments were 24 cm wide. The first compartment was designated as the home area and contained rocks and artificial plants that extended to within 15 cm of the water surface. In addition, opaque plastic covers were placed over the home range to provide shade. The floor of the entire tank was covered with silica sand to a depth of 4 cm. Water temperature in the testing tanks was maintained at approximately 18°C. Test tanks were illuminated with overhead fluorescent lights and maintained on a 14:10 light–dark cycle.

Experimental Protocol. Trials lasted one week, and each trial consisted of five days of acclimation and two days of testing (days 6 and 7). An individual pike was placed in the test tank on the morning of day 1. During days 1–5, we fed each pike one of the following diets: fathead minnow, swordtail, or mouse. For each condition, pike were fed one food item daily (mean ± 1 SD = 1.45 ± 0.21, 1.82 ± 0.32, and 1.71 ± 0.27 ml, minnow, swordtail, and mouse, respectively, measured by volumetric displacement in water). Prior to feeding, minnows and swordtails were killed by a blow to the head. Each food item was injected with approximately 0.5 ml of ultraviolet fluorescent dye (Tracer Glo). Since pike do not readily accept dead prey (Huntingford, 1984), prey items were suspended on the end of a fine-gauge stainless steel wire. The use of the wire allowed us to manipulate the prey item and induce the pike to strike. In the case of minnow or swordtail diets, the wire was placed at the base of the caudal peduncle. In the case of the mouse diet, the wire was placed through a fold of

skin near the hind legs. This allowed us to pull the wire free without disturbing the pike. Prey were always presented in the home area.

At the end of day 5, all feces were removed from the test tank with a siphon. On day 6, we began recording two measures: (1) proportion of time the pike spent in each area of the tank (area use), and (2) the location of feces. We quantified area use by recording the location of each pike once every 10 sec for a 10-min observation period once each hour for eight consecutive hours on days 6 and 7. Observations began at approximately 08:00 hr each day.

The location of feces was quantified after the final area use observation on days 6 and 7. We recorded (and removed with a siphon) the number of fecal pellets in each compartment of the test tank. Fecal pellets within 1 cm of each other were recorded as a single pellet. After recording the location of feces on day 7, we removed the pike and carefully drained the tank so as not to disturb the substrate. A UV light was used to check for any fecal pellets that may have partially dissolved or been covered with sand. We recorded the presence of fluorescent dye as a fecal pellet, using the same criterion as above.

A total of nine pike (mean $\pm$ SD = 18.21 $\pm$ 2.61 cm) were used in the experiment. Each pike was tested once in each diet condition. The order of trials was random, and pike were tested once every three weeks. Individual pike were tested in different tanks for each of the three diet conditions. No qualitative differences were observed in the acceptability of each of the three prey types.

Statistical Analysis. We employed a one-way analysis of variance (ANOVA; Sokal and Rohlf, 1981) to determine if there was a significant difference in the amount of time spent or the number of fecal pellets in each of the seven areas of the test tank. We compared the percent of time versus the percent of total feces in each area of the tank using paired t tests, corrected for increasing alpha using a modified Bonferronni procedure (Keppel 1982).

RESULTS

Pike fed minnows and pike fed swordtails spent a significantly greater proportion of time in the home area end of the test tank ($F_{6,56} = 13.22$, $P < 0.0001$ minnow diet, $F_{6,56} = 3.90$, $P < 0.003$ swordtail diet; Figure 1). Pike fed mouse did not localize their time in any section of the test tank ($F_{6,56} = 1.21$, $P = 0.30$; Figure 1). In the minnow diet trials, pike defecated significantly more often in the opposite end of the test tank ($F_{6,56} = 54.30$, $P < 0.0001$; Figure 1). Pike fed swordtails defecated significantly more often in or near the home area ($F_{6,56} = 3.90$, $P < 0.003$; Figure 1). In the trials of pike fed mouse, there was no significant difference in the number of fecal pellets in any of the seven areas of the test tank ($F_{6,56} = 1.21$, $P = 0.32$; Figure 1).

When pike were fed minnows, there was a significantly greater proportion of time spent in the home area and the two adjacent areas compared to the

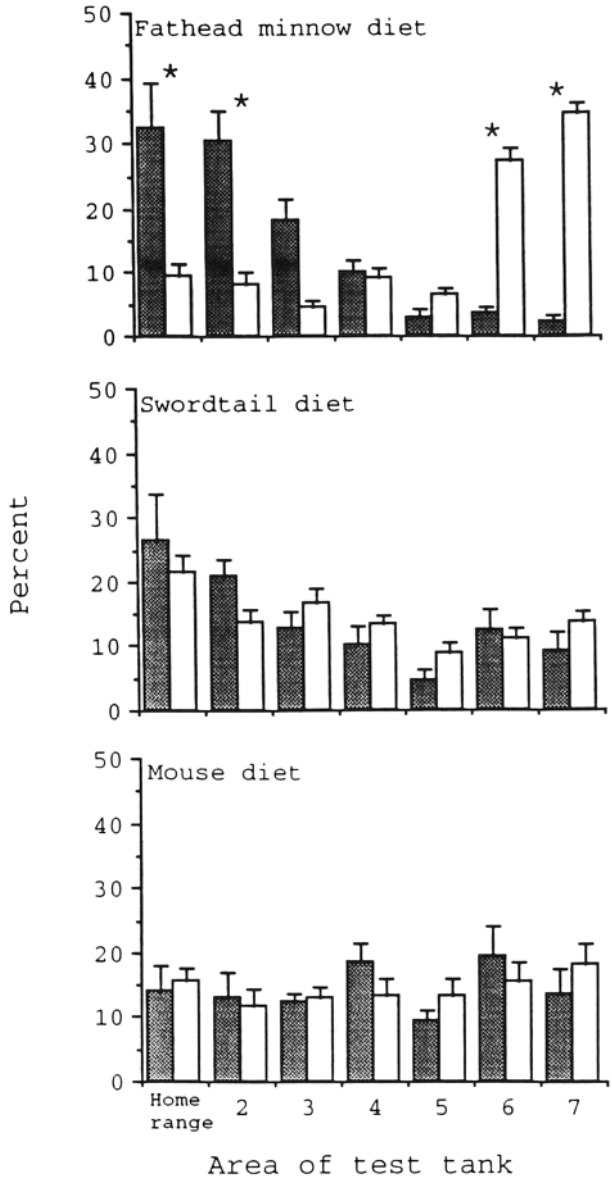

FIG. 1. Mean (±SE) percent time spent (closed bars) and percent total feces (open bars) in each area of the test tank for each of the three diet conditions. An asterisk denotes significant difference ($P < 0.05$) for comparison between percent time and percent feces in each area of the test tank. See text for details.

percent of total feces ($P < 0.05$, Figure 1). Conversely, there was a significantly greater proportion of feces compared to the percent of time in areas 6 and 7 of the test tank (Figure 1) when fed minnows. In both the swordtail and mouse diet conditions, there was no significant difference in the percent time vs. percent feces in any area of the test tank (Figure 1).

DISCUSSION

The results of the current study clearly demonstrate an effect of diet on localized defecation by northern pike. When fed a diet of fathead minnows, pike localized their defecation away from the home or foraging area, confirming the results of Brown et al. (1995a). When fed a diet of either swordtails or mice, which lack alarm pheromones, there was no difference between the percent of time spent and percent of total feces in each area of the test tank, indicating that the pike did not localize their defecation when fed these diets. These results support the hypothesis that pike localize defecation in order to avoid chemically labeling their foraging area as dangerous to potential prey.

There exist alternate hypotheses that may explain the observed localized defecation in pike. In particular, pike may localize defecation for sanitary reasons. By defecating away from the home or foraging range, pike may decrease the likelihood of pathogen or parasite infection. Such a system has been shown for a variety of avian and mammalian species (Poole, 1985). While the current data cannot refute the antipathogen/parasite hypothesis, we observed localized defecation only in response to the minnow diet. Our data do support the hypothesis that pike localized defecation to remove chemical cues from their foraging territory. In addition, these hypotheses are not mutually exclusive and could both serve as selection pressures for localized defecation behavior in pike.

These data demonstrate an effect of diet on the foraging patterns of pike. In both the minnow and swordtail diet conditions, pike spent a significantly greater proportion of time in the home end of the test tank, while in the mouse diet condition, pike did not exhibit such a preference. Since pike are primarily ambush predators (Hobson 1979; Chapman and MacKay, 1984; Savino and Stein, 1989), it would benefit a pike to remain under the cover of vegetation and wait for suitable prey to come within its attack range. Conversely, terrestrial prey items, such as mice, would be swimming near the surface. Hence, when fed a mouse diet, it would benefit the pike to remain in open water. These differences in foraging strategy would account for the lack of localized area use observed in the mouse diet condition, even though the pike were fed in the same manner regardless of diet condition.

These results also indicate a more general phenomenon, whereby the foraging strategy of predators is affected by diet. By altering the foraging strategy

to match the most abundant prey type, the pike would maximize its probability of future foraging success. As a result, if pike have experienced terrestrial prey in open water and are subsequently fed terrestrial prey, then we would predict that they should not show a preference for area of the test tank containing vegetation.

There are a variety of mechanisms by which the presence of alarm pheromone in the home or foraging area could decrease a pike's foraging efficiency. Initially, Brown et al. (1995a) demonstrated that the alarm pheromone is present in the pike's feces. The presence of feces containing minnow alarm pheromone has been shown to decrease area use by minnows (Brown et al., 1995b). This would serve to decrease the pike's foraging efficiency by deterring prey fish from entering the pike's foraging area.

Chivers and Smith (1995a) have shown that fathead minnows are able to learn the identity of risky habitats when the chemical cues of a particular microhabitat are paired with AS. If AS in pike feces were present in the pike's foraging range, then the minnows may learn to avoid the risky area. Alternatively, the minnows may be more vigilant when in the area and therefore less likely to be captured (Mathis and Smith, 1993c; Chivers and Smith, 1995a). Given that minnows can culturally transmit recognition of dangerous habitats (Chivers and Smith, 1995b), the effects may extend to other minnows.

Cross-species reactions to alarm pheromones have been identified in a variety of species (e.g., Smith, 1982; Smith et al., 1991; Mathis and Smith, 1993d; Chivers and Smith, 1994; Wisenden et al., 1994, 1995; Chivers et al., 1995a,b). Prey species typically avoid areas containing heterospecific alarm pheromones of members of their prey guild (i.e., those species with which they share predators). The presence of minnow alarm pheromone in the pike's foraging range would likely also decrease area use by other species, such as brook sticklebacks (Mathis and Smith, 1993d), that recognize and respond to minnow AS. By defecating away from their foraging range, pike can increase the likelihood of future predation events in that location not only on minnows, but on other species of the prey guild as well.

Minnow alarm pheromone has been shown to attract pike (Mathis et al., 1995). A pike that is attracted to AS (i.e., a secondary predator) could interfere with a primary predator that has captured a minnow. The secondary pike could compete for the prey item directly, or it could attack the primary predator as a prey item. Avoidance of secondary predator interference could account for the observed localized defecation. By removing minnow alarm pheromone from their foraging range, pike could decrease the possibility of attracting secondary predators.

In summary, the data presented in the current study demonstrate an effect of diet on the localized defecation of northern pike. Localized defecation was observed when pike had consumed minnows, but not when pike had been fed

swordtails or mice. These data thus support the hypothesis that pike localized their defecation in order to avoid chemically labeling their foraging area with the alarm pheromone of their prey.

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