Teaching wild redbelly dace novel predator cues

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Abstract

Alarm cues in fish have been shown to cause anti-predator responses. Although it has been shown that fish can be taught novel predatory cues in laboratory work, it has yet to be demonstrated in the field. In this study we attempt to teach northern redbelly dace in Deming Lake the alarm cue of the yellow perch, a predatory fish that does not occur in Deming Lake. First, perch-naïve dace were tested for response to perch alarm cue. Then they were exposed to a mixture of perch and dace alarm cue and their response to perch alarm cue was re-evaluated the following day. There were no significant differences in fish response to perch alarm cue from the first to the third day. The study showed that it can be difficult to teach a large population of fish a novel predatory cue.

Introduction

Aquatic habitats are full of behavioral responses that rely heavily on alarm cues (Mirza and Chivers, 2001). Survival and fitness are maintained through an olfactory system of predator risk assessment (Wisenden, 2000). When a species releases an alarm cue, conspecifics and sometimes ecologically similar heterospecifics will react (Pollock et al., 2005). Predator evasion in response to heterospecific alarm cues is innate in closely related species and must be learned in more distant species (Pollock et al, 2005). Studies where conspecific cues were mixed with heterospecific cues have shown increased shoaling and anti-predator behaviors (Brown and Godin, 1997). This has never been demonstrated in a field setting where massive exposures are required for large populations.

The northern redbelly dace (*Phoxinus eos*), a common minnow species in Minnesota, has been shown to use alarm cues. They are one of the main species in Deming Lake, in Lake Itasca State Park, MN. This lake is devoid of all piscivorous fish, such as the yellow perch (*Perca flavescens*). Thus, these dace should not fear the smell of a predatory fish, such as the perch. In a similar manner, stocked game fish often fall prey to unfamiliar predatory fish (Santucci and Wahl, 1993). If the stocked fish were able to recognize the smell of a predator that they had not yet been exposed to, this would allow for better risk assessment by the stocked fish due to predation by others. The walleye (*Sander vitreus*) is a prime example where this type of learning is needed. This fish, being the top game fish in Minnesota, is stocked in many lakes directly from fisheries; the ability to teach these fish to evade predatory fish such as northern pike (*Esox lucius*) and largemouth bass (*Micropterus salmoides*) would benefit the industry.

The hypothesis for this experiment was that the redbelly dace would acquire recognition of perch alarm cue as dangerous and evade such a risk. Our predictions were that, prior to exposure of the perch and redbelly dace alarm cues mixed together, the control and perch scented traps would catch relatively similar amounts of fish. Following the attempted teaching exposure, we presumed that the number of fish caught in perch scented traps would drop significantly to a number more similar to the redbelly dace scented traps. During our experimentation we also observed the risk-assessment of heterospecific fathead minnow (*Pimephales promelas*) and brook stickleback (C*ulaea inconstans*). Successful learned recognition in the redbelly dace would be the first field study of teaching the risk-assessment of a novel allospecific alarm cue to a population of fishes.

Methods

Adult redbelly dace were obtained with minnow traps on Deming Lake in Lake Itasca State Park, Minnesota. They were kept in a live well for a couple days before the experiment commenced. Juvenile yellow perch were collected by seining near the Itasca Biology Station on the shores of Lake Itasca. They were kept in a separate live well until use in the experiment. After the fish were obtained, twelve 3M sponges (152mm x 106mm x 40mm) were cut into 15 roughly equal sized pieces (30.4mm x 35.3mm x 40mm). Then each sponge was fitted with a metal wire approximately 12 cm long, to be used for attaching the sponge for the traps. The sponges were then rinsed twice with tap water, as per the warning on the sponge bag.

Next the skin solutions were prepared. One solution was made for 30 sponges at a time. A total of 6 solutions were made (one with dace, one with perch, one with water, and three with a mixture of perch and dace). To make the redbelly dace solution, 10 dace were selected and their lengths were measured. They were then euthanized by cervical dislocation with a scissors. Their mass was then obtained. The dace were an average of 4.9 cm long and had average postcranial weight of 0.05g each. Finally their bodies were put in a dish with 100 ml of tap water and pureed with a hand blender. Alarm cues were obtained via this method because it would be the most practical method that a fisheries worker could use. The fish solution was further diluted with 200 ml of water. Then 30 sponges were treated with the solution with 10 ml of solution used per sponge. The perch solution was prepared in a similar manner; however it used 10 perch instead of 10 dace. The perch were an average of 6.3 cm long and had an average postcranial mass of 0.35g. The mixed solution contained 10 perch and 10 dace in 300 ml of water. The water solution only contained 300 ml of tap water. After all sponges were treated with the solution, they were stored in bags of 30 sponges labeled by solution type. Then they were placed in a freezer for storage.

The remainder of the experiment was carried out at Deming Lake over a three day period. On the first and third day, each minnow trap had a treated sponge tied to the inside of it. The traps were laid in a repeating sets based on sponge treatment in a clockwise manner (control, dace, and perch) over the southeastern third of the lake. Traps were laid roughly 5 m apart from each other. Each set was laid roughly every 5 minutes and collected two hours after being laid. After the traps were pulled in, each trap type had one person that would identify each kind of fish caught and another that would record the numbers. On day one, 45 traps were used, while on day three only 39 traps were used. The fish counts were then transferred to a Microsoft Excel spreadsheet for analysis. On day two, 45 pairs of sponges with the mixed solution were attached to the clips on trap ropes. They were laid in repeating clockwise sets as the traps were on days one and three. The sponges were allowed to soak in the lake for 24 hours and collected as the traps were being laid for the third day. Finally, the data was processed using Statistical Package for the Social Sciences (SPSS) 17.0.

Results

Day 1

Three species of fish were caught in our minnow traps on days one and three: redbelly dace, fathead minnow, and brook stickleback. On average the traps had much higher average catches than median catches because some traps caught very large numbers of fish, while others caught none. On day one, the perch traps averaged twice the redbelly dace as the dace or control traps (12.3 dace versus 5.8 and 4.6 respectively). Each type of trap averaged about one brook stickleback per trap, and the dace traps caught the most fatheads (4.8 fish per dace trap, 3.2 fish per perch trap, and 0.8 fish per control trap). The dace traps had a median of only one dace caught per trap. While the perch had a median of two dace, one fathead, and one stickleback per trap, the control had a median of one dace and one stickleback per trap. The amount of fish caught by each trap type was run through a Kruskal-Wallis test. There was no significance for the number of fathead minnows caught by the different trap types ($p=0.061$, $df=2$, $\chi^2=5.607$, **Figure 2**). Furthermore there was no significant difference in the number of redbelly dace caught by the different trap types ($p=0.166$, $df=2$, $\chi^2=3.588$, **Figure 1**). Finally there were also no significant differences between number of sticklebacks caught and trap type ($p=0.476$, $df=2$, $\chi^2=1.484$, **Figure 3**).

Day 3

The same species were caught in our traps as day one. Additionally this day also had a similar amount of discrepancies between mean and medians for the same reason. However this time, the control traps averaged the most redbelly dace (9 fish versus 4.8 fish for the perch traps and 4.6 fish for the dace traps). The control traps also averaged the most sticklebacks (1.2 fish versus 0.5 fish for the perch and dace traps). Although the perch traps averaged about twice as many fatheads as either the dace or control traps (2.2 fish versus 0.8 and 1.3 respectively). The dace traps had a median of two dace caught per trap. The perch traps had a median of three dace and one fathead in each trap. However the control traps had a median of one fathead and twelve dace. Kruskal-Wallis tests were then run to test for significance between numbers of each type of fish caught and trap type. There was no significance in number of dace caught by trap type ($p=0.207$, $df=2$, $\chi^2=3.514$, **Figure 1**). Also, no significance was seen in number of fatheads caught by trap type ($p=0.240$, $df=2$, $\chi^2=2.855$, **Figure 2**). Finally there was no significance seen in the number of sticklebacks caught by trap type ($p=0.484$, $df=2$, $\chi^2=1.450$, **Figure 3**).

Discussion

The results of our experiment lead us to reject the hypothesis that if fish are exposed to a substantial amount of allospecific alarm cue mixed with their own alarm cue, then they should acquire a learned recognition to respond to the allospecific alarm cue isolated. With insignificant data, we have concluded that this learned recognition was not evident. This goes against studies of alarm cue learning in fish. Fish have been documented to learn to respond to alarm cues exposed in the same matter as our experiments in the laboratory setting (Heczko and Seghers, 1981). Possible explanations for this quandary include human error in methodology, insufficient exposure of mixed alarm cues, or the methodology itself.

By blending the fish whole as opposed to using the epithelial tissue alone, error might have been introduced. It is a possibility that the fish were receiving mixed signals: detraction from alarm cues in skin extract and attraction to organ tissue mistaken as food. Although this was a more simple procedure that matched the real-world application most accurately, it could lead to inconsistent and unreliable data.

The amount of exposure might not have been sufficient to teach the entire population also. Repeating this experiment, we would increase the amount of the mixed alarm cue saturation. With our experiment covering under half of the lake's parameter, there comes into consideration fish migration. It is plausible to explore the possibility that fish might have been in areas of the lake during day two that were not experiencing the mixed alarm cues. Fish with this deposition would be found attracted to perch traps as was found in day one. Studies have supported the synergistic interaction between shoaling and alarm cues (Wisenden et al., 2003). If a few fish were to be attracted to traps with alarm cues, this might cancel the effect for the rest of the fish, leading to an even higher source of error.

There are other sources of error that were considered as well. The weather was significantly different on day one and day three. Day one included warm sunny weather, and day three included overcast with rain. This could very well affect the shoaling of fish and their placement in the lake (Keenleyside, 1955). Another source of error is that the behavior of the fish might be affected by all of the experimentation performed on the lake. Multiple experiments are conducted on Lake Deming annually, which could distort the fish behavior to less than natural. We also lacked two traps for day three. With less data, the accuracy is diminished. Repeating the experiment, we would make sure to set at least fifteen sets of traps each trial.

Although we are not able to determine the exact source of error for this experimentation, we are able to evaluate the methodology and how it would change for a repeated study. It is known that fish have highly developed receptors to react to alarm cues of their own species (Wisenden, 2000), and it is also known that fish will sometimes increase anti-predator behavior when exposed to heterospecific alarm cues of fish with which they co-exist (Pollock et al., 2005). There have been some studies that support the idea that the skin extract is in fact not an alarm cue in certain fish (Magurran et al., 1996). Our data coincides with the findings of these studies, though studies that have supported the skin extract as an alarm pheromone (Wisenden, 2000) lead us to conclude our data skew as a result from experiment error. Taking into consideration these sources of error, we feel strongly the need to continue experimentation of learned recognition of alarm cues in a field setting.

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Appendix:

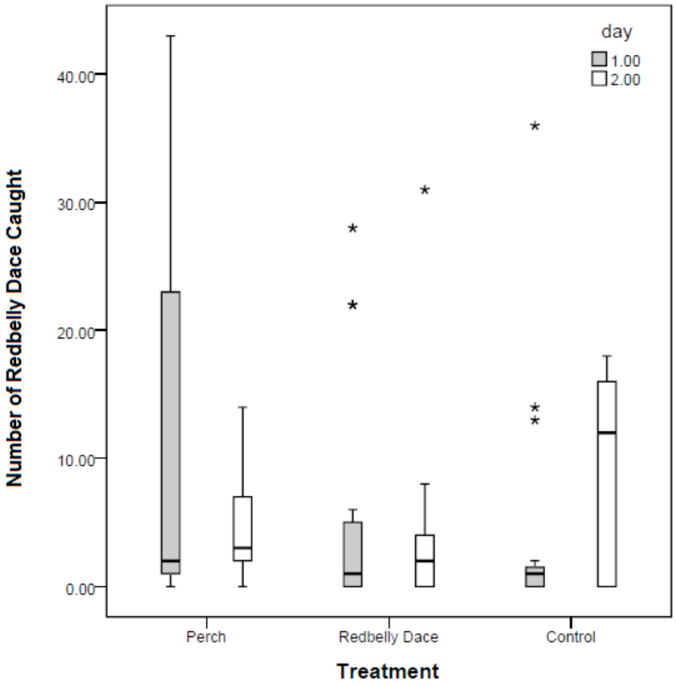

Figure 1: Box plots of the number of redbelly dace caught in each trap treatment on both days.

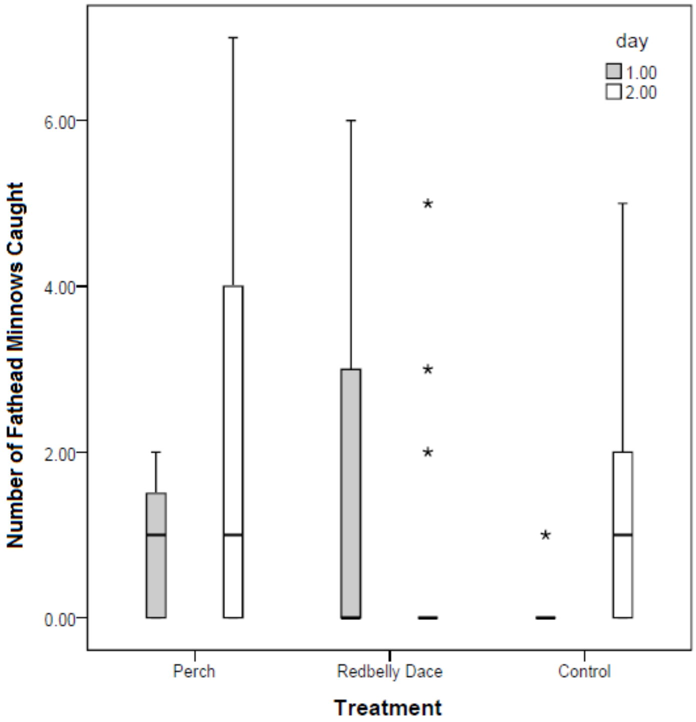

Figure 2: Boxplots of the number of fathead minnows caught by each trap type on both days.

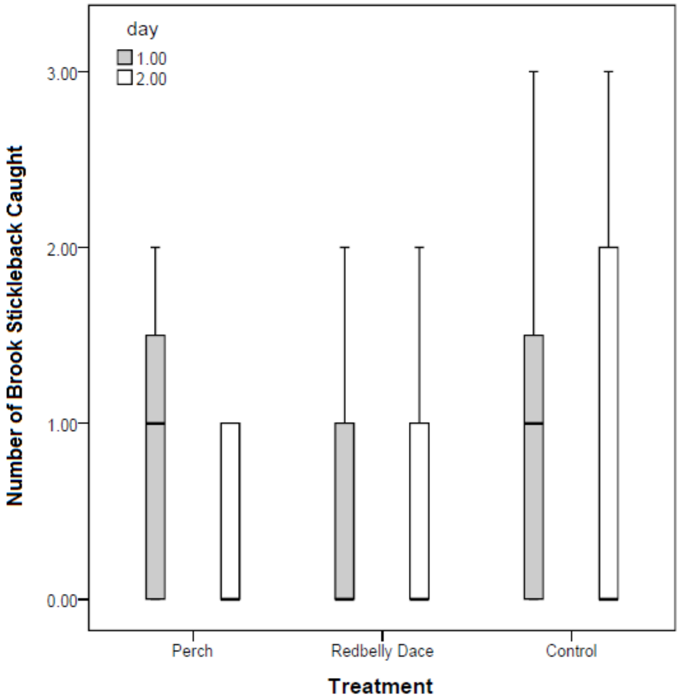

Figure 3: Boxplots of the number of brook stickleback caught by each trap type on both days.