

RESPONSES OF BLACKNOSE DACE (*Rhinichthys atratulus*) AND BROOK CHAR (*Salvelinus fontinalis*) TO ACIDIFIED WATER IN A LABORATORY STREAM

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Abstract. As part of the Shenandoah National Park: Fish in Sensitive Habitats (SNP: FISH) project, movements of blacknose dace (*Rhinichthys atratulus*) and brook char (*Salvelinus fontinalis*) were examined during exposure to artificial acidification in paired channels of a laboratory stream. The objective of this study was to determine the ability of the fish to avoid depressions in the ambient pH and recognize and use a neutral-pH microhabitat refuge during acute reductions. Fish preference for a particular channel was statistically predictable based on the delivery of food. We tested fish avoidance behavior by manipulating food and the delivery of a pulse of acidified water. Both blacknose dace and brook char avoided the acid pulse (ambient pH reduced from 7.2 to 5.1) by sheltering in the pH-neutral refuge. Extensive field sampling in refuge microhabitats before and during episodic acidification is needed to determine changes in the distributional patterns of these species associated with acid precipitation events.

Key words: laboratory stream, acidified pulses, brook char, blacknose dace

1. Introduction

Water quality in many acid-sensitive streams is highly variable because of inputs of alkaline water from spring seeps and tributaries (Sharpe and DeWalle 1990). This spatial variability creates the potential for alkaline microhabitats to act as refugia for fishes exposed to periodic acidic episodes. Downstream migration and the congregation of fishes at alkaline water inputs have been reported by a number of researchers (Leivestad and Muniz 1976, Hall et al. 1980, Muniz and Leivestad 1981, Watt et al. 1983, Gagen et al. 1989). Fish in laboratory studies have demonstrated behavioral avoidance of a variety of environmental variables including low pH and high Al concentrations (Whitmore et al. 1960, Hill et al. 1981, Jones et al. 1985, Gunn and Noakes 1986). Although increased tolerance of brook char (*Salvelinus fontinalis*) to low pH through acclimation has been documented (Guthrie 1981), most researchers have not accounted for the effect of variables such as food availability, or sensory and physiological acclimation to extreme conditions (Gunn 1986). In addition, few researchers have studied the responses to acidification exhibited by nongame fishes (Charles 1991).

In this study, the effect of water acidification on movement of blacknose dace (*Rhinichthys atratulus*), and brook char indigenous to an acid-sensitive stream (Paine Run) of Shenandoah National Park (SNP), Virginia was tested in the paired channels of a laboratory stream. The objective of this study was to determine the ability of fish to avoid depressions

in ambient pH, and recognize and use a pH-neutral microhabitat refuge during acute pH episodes.

2. Materials and methods

Ninety young-of-year brook char, 52-81 mm total length, and one-hundred thirty adult blacknose dace, 52-78 mm total length, were collected from Paine Run by electrofishing and maintained in aerated aquaria. Experiments were conducted from August 31 through December 31, 1994 in the paired channels of a laboratory stream that was enclosed in a black plastic shroud equipped with viewing ports (Figure 1). Marks were placed along the inside walls to divide the channels into six zones of equal surface area (0.43 m²). Although inputs of food and acid alternated between channels, zone 1 was always closest and zone 6 furthest from the input. The bottom of the stream was covered by a single layer of washed stream gravel.

Water from head tanks was gravity-fed into separate areas at the head of each channel. As determined from 10 trials using a dye tracer, water moved uniformly down each channel and water did not mix between channels. Current velocity was 0.30 ± 0.05 m/s. An acid solution (100 ml H₂SO₄ in 400 ml H₂O) was used to reduce the ambient pH of a selected channel from 7.2 to 5.1 (± 0.1 unit) during an experimental trial. The acid solution was mixed with the gravity-fed water in the selected channel via a peristaltic pump (Masterflex Tubing Pump Drive Fixed Speed Model 7543-20, Cole-Parmer Instrument Company).

Fifteen fish were randomly selected for each trial. Fish were introduced into the downstream end of the laboratory stream and allowed 5 min to adjust before beginning each experiment. Fish were allowed to move freely between channels and among zones of the laboratory stream. Fish in each zone were counted every 30 sec for 30 min. Fifteen preliminary trials (no food or acid additions, 900 observations) were run with each species to assure that fish were not using either channel preferentially. Thirty trials (1800 observations) were run with each species, first with food additions to one of the paired channels (i.e., a brine shrimp suspension delivered via another peristaltic pump), and then with food and acid additions together in a channel for a total of sixty trials per species. Trials with food alone were used to determine if food affected fish distribution. Trials with food and acid combined were used to determine if fish actively avoided acidic conditions in an otherwise favorable environment.

We used a G-test to evaluate differences in the distribution of fishes (Sokal and Rohlf 1995). For preliminary trials without food or acid additions and for food-only trials, we hypothesized that the fifteen fish in a trial would use all six zones of the laboratory stream uniformly (i.e., on average 2.5 fish observed per zone over the course of a trial). In trials

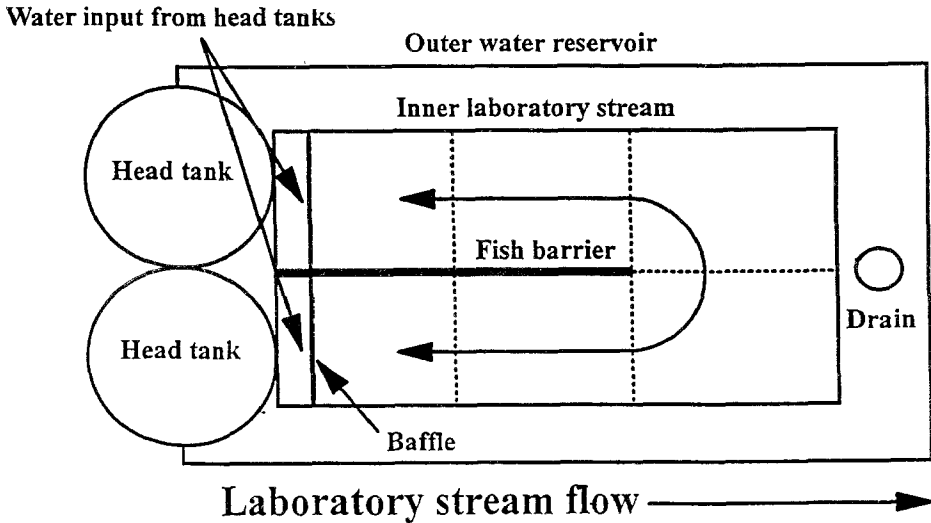


Fig. 1. Schematic diagram of laboratory stream.

where food and acid were introduced together, expected frequencies were based on the results of the food-only trials. Each test had 5 degrees of freedom.

3. Results

The initial experiments showed no preferential use of any zone in either channel by brook char ($G = 5.4, P > 0.50$) or blacknose dace ($G = 5.9, P > 0.40$). Both species used the zones of the laboratory stream uniformly (i.e., on average 2.5 fish per zone) when no food or acid was added to either channel.

3.1 BROOK CHAR RESPONSES

In the second set of experiments, brook char showed a marked preference for the channel receiving food ($G = 11.8, P < 0.05$) and nearly 42% of the fish were observed in zone 1, more than in any other zone. Therefore, the frequencies of zone use by brook char observed in these trials were used as the expected values (i.e., the control) in subsequent experiments that combined food and acid inputs.

In the third set of experiments on char, food and acid were alternately introduced into the upstream ends of the paired channels. Char clearly avoided the acidified channel ($G = 77.5,$

$P < 0.001$) (Figure 2a). Brook char use of zone 6 increased by more than 47% over the expected frequency (Table I).

3.2 BLACKNOSE DACE RESPONSES

After food was added, the distribution of blacknose dace shifted from uniform across all zones to zones in the channel that received food ($G = 17.9$, $P < 0.005$). Over 46% of the blacknose dace were observed in zone 2, therefore, the frequencies of zone use by blacknose dace observed in these trials were used as the expected values in subsequent experiments that combined food and acid.

In the third set of experiments with dace, food and acid were alternately introduced into the upstream ends of the paired channels. Dace clearly avoided the acidified channel ($G = 41.4$, $P < 0.001$) (Figure 2b). Blacknose dace generally decreased their use of the acidified channel, increased use of the channel receiving only gravity fed water, and increased use of zone 4 by 52.1% over the expected frequency (Table I).

4. Conclusion

Both brook char and blacknose dace actively avoided water having a pH of 5.10, which is

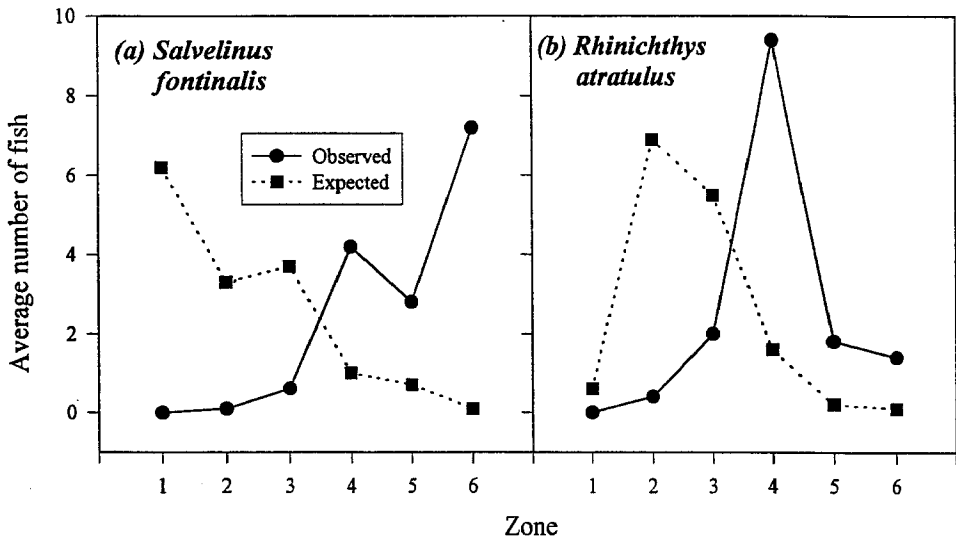


Fig. 2. Average number of brook char (a) and blacknose dace (b) observed over 30 trials in the zones of a laboratory stream after the introduction of food (expected) and food combined with acid (observed). Input was closest to zone 1 and farthest from zone 6.

TABLE I

Percent use of experimental zones by brook char and blacknose dace during an acid pulse, and percent deviation from control.

		Experimental zones					
		1	2	3	4	5	6
Brook char	Number observed	0.03	0.14	0.57	4.22	2.81	7.23
	Percent use	0.2	0.9	3.8	28.1	18.8	48.2
	Percent deviation	-41.3	-21	-20.9	21.7	14	47.5
Blacknose dace	Number observed	0.01	0.35	2.03	9.43	1.8	1.38
	Percent use	0.1	2.3	13.5	62.9	12	9.2
	Percent deviation	-3.9	-44	-23.1	52.1	10.4	8.5

lower than that reported by Charles (1991) to elicit an adverse effect on these species. The lowest level of pH experienced by fish in the laboratory stream was higher, however, than pH observed in Paine Run, where pH has fallen from 5.80 to 4.86 during a natural event (Art Bulger, University of Virginia, personal communication). The lower pH limit for brook char survival is 4.50 (Power 1980), and waters below pH 5.50 are considered borderline (Schofield 1976). Waters ranging in pH from 5.00 to 5.90 have been reported to cause mortality in different life stages of blacknose dace (Johnson et al. 1987, Schofield and Driscoll 1987, Kretser et al. 1989, and Haliwell 1989). These results suggest that Paine Run fish did not use sensory or physiological acclimation to withstand acute acidic conditions. Availability of other resources (i.e., food) also did not appear to deter these fish from leaving an experimentally acidified channel. Typically, both species vacated the acidified channel within a few minutes of acid introduction, and after a brief period of feeding forays into the acidic water permanently resided in the non-acidified channel for the remainder of a trial. Consequently, we concluded that both species of fish from Paine Run actively avoided acute reductions in ambient pH and recognized and used a pH-neutral microhabitat refuge in this laboratory stream.

As in with any laboratory study, there is danger in extending inferences made from behavior observed in a controlled environment to behavior in a natural setting. Based on laboratory data, Neville (1985) hypothesized that juvenile salmonids in natural streams could escape death caused by acidic episodes by seeking refugia. Other researchers have documented congregations of stream salmonids near alkaline water inputs during acidifying conditions (Leivestad and Muniz 1976, Hall et al. 1980, Gagen et al. 1989). More recent research, however, suggests that movements of fish during an acidic episode may be the result of passive drift rather than an active search for refugia (Gagen 1991).

Future research should clarify the inconsistencies between field observations and laboratory data. Both species clearly exhibited an adaptive response to depressed pH under the sublethal conditions presented to them in this study. These experiments represent an important step in understanding the relationship between behavioral modifications, acidic episodes, and fish population resilience in acid-sensitive streams.

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