Effect of Larval Sea Lamprey Density on Growth

Project Completion Report

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by

William D. Swink

National Biological Service

Great Lakes Science Center

Lake Huron Biological Station

11188 Ray Rd.

Millersburg, Michigan 49759, USA

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Abstract.--Growth of larval sea lampreys Petromyzon marinus was compared at two population densities (~40 and ~400 animals/m^2) to determine if growth was affected by a waterborne inhibitor, in a laboratory study conducted from 1 March to 18 November 1994. Growth over the 8.5 months of the study averaged 0.5 g for animals in the high density tanks and 1.0 g for animals in the low density tanks. Growth differed significantly between population densities only for the September - November period, when the larvae in the low density tanks received significantly more feed per animal. Within a population density, larval growth never differed between the control tanks and the test tanks, which received effluent water from the controls. I detected no evidence of a waterborne growth inhibitor, even among larvae exposed to water from a combined population equivalent to 800 animals/m^2.

Population density has been shown to affect the larval growth of sea lampreys Petromyzon marinus (Purvis 1979; Morman 1987) and Pacific lampreys Lampetra tridentata (Mallatt 1983). The cause of this density effect is unknown, but does not appear to be related to limitations on food. Malmqvist and Brönmark (1981) suggested that only minimal intraspecific competition for food occurred among larval Lampetra planeri in a Swedish stream, even at densities as high as 113 larvae per m^2.

Mallatt (1983) suggested that larval lampreys release a substance into the surrounding substrate that inhibits growth. Such a substance might account for the decline in growth rates that was noted for each succeeding year class of sea lampreys in streams after treatment with lampricides (Purvis 1979). More recent work (P. Sorensen, University of Minnesota, personal communication) showed that spawning-phase sea lampreys could detect effluent water from larvae. Hence, it seems likely that a growth inhibiting substance that can be detected by other larvae might also be carried in the water, rather than just be deposited in the substrate. If a chemical growth inhibitor exists, growth for animals at the same density should progressively decrease when exposed to water from successively denser populations. Any effect on growth should be most pronounced for the animals in a high density population that receives water from another high density population. The purpose of this study was to determine the effect of effluent water from one group of sea lampreys on the growth of sea lampreys in another group.

Methods

The study was conducted at the Lake Huron Biological Station from 1 March to 18 November 1994. Sea lamprey larvae were anesthetized in a 75 ± 25 mg/L solution of tricaine methansulfonate (buffered with sodium bicarbonate to maintain pH within one unit of Lake Huron water; Allen and Harman 1970), measured to the nearest mm, and weighed to the nearest 0.01 g. The sea lamprey larvae used in the study were collected by electrofishing from the Manistee River, Michigan on about 26 October 1993 and were held in the fish culture facilities at the Lake Huron Biological Station until the start of the study. Larvae were fed twice a week during this period with a slurry of bakers' yeast and BioKyowa A-250 fry feed (BioKyowa, Inc., Chesterfield, Missouri; Swink, in review).

Sea lamprey larvae for the study ranged in size from 54 to 110 mm. The larvae were placed in each of 40 37.9-L aquaria (50.0 cm x 25.2 cm x 29.0 cm; 0.126 m^2 bottom area) that contained about 30 L of water and beach sand to a depth of 80 ± 10 mm for substrate. Beach sand was collected in the vicinity of the Lake Huron Biological Station and was sieved to remove larger debris before placement in the tanks. Particle size of a 3,884.5 g sample of beach sand consisted of > 1000 μm, 0.7%; 591 - 1000 μm, 40.9%; 251 - 590 μm, 56.5%; 126 - 250 μm, 1.8%; and ≤ 125 μm, < 0.1%.

Sea lamprey larvae were added to 20 tanks at a rate of 50 per tank (density of 397 animals/m^2) and to the other 20 tanks at

a rate of 5 per tank (density of 39.7 animals/m^2). Twenty of the tanks were situated on the upper level of a rack so the effluent water from a tank drained into the corresponding tank beneath it. Five of the upper tanks that each contained 50 animals drained into five corresponding tanks that each contained 50 animals. Another five of the upper tanks that each contained 50 animals drained into five corresponding tanks that each contained 5 animals. Five upper tanks that each contained 5 animals drained into five corresponding tanks that each contained 5 animals. The other five upper tanks that each contained 5 animals drained into five corresponding tanks that each contained 50 animals.

All tanks were aerated, and flowing Lake Huron water at ambient temperature was supplied to the 20 tanks on the upper level at a rate of 500 ± 75 mL/min. Lake Huron water temperature was continuously recorded on a thermograph.

The tanks were checked every day on weekdays for dead larvae on the surface of the substrate and to ensure that the water flow and air stones were not obstructed. Dead animals were removed from the tank, measured to the nearest mm, and recorded.

Larval sea lampreys were fed twice a week on non-consecutive days with a slurry of 454 g of baker's yeast and 50 g of BioKyowa A-250 fry feed mixed in 8.5 L of Lake Huron water. On days when feeding occurred, the water to the tanks was shut off at about 1600 hours and freshly made slurry added to the tanks. Aeration was maintained in the tanks while the water was shut off. The water remained shut off until about 0800 hours of the next day.

Initially, water flow reestablished after feeding was allowed to drain immediately from tanks on the upper level into tanks on the lower level. After the initial study period (March to May), however, effluent from the tanks on the upper level was routed directly to the drain for about 1 h before being rerouted into the lower level tanks. This allowed any excess food in the upper tanks to be removed and helped prevent a disparity in feeding rate among the upper and lower tanks.

Initially, slurry was supplied to the tanks at a rate of 250 mL for tanks with 50 larvae and 25 mL for tanks with 5 larvae; each larval sea lamprey was given the same ration of about 0.27 g of yeast and 0.03 g of fry feed. At the conclusion of the first growth period (10 May), feed rate in low density tanks was increased to 100 mL of slurry twice a week; the ration per animal was increased to 1.06 g of yeast and 0.12 g of fry feed. Feed rate in the low density tanks was increased again after 12 September to 250 mL of slurry twice a week; the ration per animal increased to 2.67 g of yeast and 0.29 g of fry feed. Feed rate to the high density tanks was maintained throughout the study at 250 mL of slurry twice a week.

About every two months, each tank was drained and the larvae removed by hand from the substrate, anesthetized, measured, and weighed as previously described. Dead or missing larvae were replaced at this time with larvae marked by latex injection (Hanson 1972) to maintain the proper density in the tanks. Marked larvae were not included in the growth estimates. Mean

change in weight was compared for differences among groups of animals at the same population density during the four periods in the study.

The surface of the substrate in the tanks was cleaned with a siphon when excess food and waste built up in a layer more than 5 to 10 mm thick. The sand substrate was removed from the tanks and replaced with fresh sand when the larvae were measured, to prevent the sediment from going anaerobic.

Mean initial sea lamprey length and weight were compared among groups of tanks using the Kruskal-Wallis test (Conover 1971). Mean weight change among larvae held at the same population density and effluent exposure were compared with the mean weight change of larvae held at each of the other combinations of population density and effluent exposure using the Kruskal-Wallis test (Conover 1971).

Results and Discussion

Initial Sea Lamprey Size

The mean initial length of larval sea lampreys in a tank (range, 80.2 - 88.8 mm) did not differ significantly ($P > 0.05$) among the two control and four test groups. Mean initial weight of sea lampreys differed slightly but significantly ($P < 0.05$) between the high (mean, 0.916 g) and low (mean, 0.989 g) density tanks. However, mean weights did not differ significantly among the three groups of tanks (one control and two test groups) within either the high ($P > 0.15$) or low ($P > 0.25$) population

density. Therefore, any differences in weight change among control and test groups at a particular population density should not be caused by a difference in initial weight or feeding level.

Growth

Weight changes of sea lamprey larvae did not vary significantly among population-density groups for any combination of effluent exposures in the March to May study period (Table 1). Effluent from tanks with sea lamprey larvae did not appear to contain any growth suppressant that affected the larvae in the receiving tanks. However, trends in weight change among the low density animals indicated that the feeding rate might be inadequate. The effluent water appeared to be more a source of extra food than a source of growth suppressant. Therefore, the feeding rate for animals in the low density tanks was increased to 100 mL of slurry twice a week for the next two study periods.

The lack of any significant differences in weight changes among groups in the May to July and July to September study periods (Table 1) again indicated that there was no growth suppressant in the effluent. Although feeding levels appeared to provide for adequate growth of larvae at both population densities, I increased the feeding rate in the low density tanks to 250 mL of slurry twice a week for the final study period.

Significantly better growth occurred in the low density tanks than in the high density tanks in the September to November study period (Table 1); growth in the low density tanks was over

three times higher than in the high density tanks. But again, no significant difference in growth occurred among effluent treatments within a group (Table 1). The continued absence of any significant difference in growth that could be attributed to a growth suppressant caused me to end the study after only 8.5 months.

The lack of any difference in growth among treatments does not preclude the existence of a waterborne growth suppressant. However, given the same rate of feeding, a growth suppressant should have inhibited growth significantly less among larvae in high density control tanks than among larvae in the high density test tanks that received effluent water from the high density controls. Water in the control tanks should carry only half the amount of growth suppressant (from a population at ~400/m^2) that would exist in water in the test tanks (from a combined population equivalent to ~800 animals/m^2). However, a difference in growth was never observed, even though the population density and the resultant concentration of any growth suppressant were both higher than normally found in the wild. In addition, the failure of the effluent water to suppress growth in any group of my lampreys corresponds with recent findings of B. Zielinski (University of Windsor, personal communication) that the olfactory receptors of larval sea lampreys responded only minimally when exposed to wash water from larval lampreys. Logically, a chemical factor that actively suppresses growth should elicit an olfactory response.

Mortality

Mortality of sea lamprey larvae was very low; only 29 of 1,100 larvae died during the almost 9 months of the study. However, 10 of those animals died in two low density tanks when the air stones were accidentally removed during a feeding period. Only 19 larvae (1.7%) died from handling or other natural causes.

Implications for Future Studies of Growth

Lack of evidence for a waterborne growth suppressant indicates that observed decreases in growth at higher larval densities (Purvis 1979; Morman 1987) are caused by other factors. The most likely causes of growth suppression among larval lampreys would seem to be an inhibiting substance released into the substrate, as suggested by Mallatt (1983), a response to tactile stimulation from overcrowding, or a limitation on food.

Although a species- or family-specific growth inhibitor might be released into the substrate, growth might simply be inhibited by the accumulation of normal waste products (e.g., ammonia). My current laboratory study did not test this possibility because the sand in the tanks was changed every 2 months to avoid the buildup of waste. Past experience in culturing larval sea lampreys shows that holding animals in anaerobic sediments under laboratory conditions can result in substantial disease outbreaks and high mortality. However, the effect of low-oxygen sediments on larval growth has never been quantified. Any effect of accumulated waste products on larval

growth would probably be less in streams than in the laboratory, because higher current velocities would tend to retard the buildup of waste products.

Larvae living in dense population groups would also more likely be affected by physical encounters with other burrowed larvae. Competition for space might cause more frequent movement and reburrowing, which increases energy use and could reduce growth. However, crowding by itself does not appear to affect larval growth. Larvae in my high density tanks were held at a higher population density (397 animals/m^2) and initial biomass (mean, 363 g/m^2) than are normally found in the wild. Yet individual lampreys gained an average of 0.5 g during the 8.5 months of the study. Average growth of larvae in my low density tanks was even higher (1.0 g in 8.5 months; density, 40 animals/m^2). In contrast, the greatest increases in mean weight for caged animals of similar size (mean, 66 - 108 mm) in the wild were calculated at 1.21 g for the Jordon River, Michigan (density, 27 animals/m^2) and 0.46 g for the Manistee River, Michigan (density, 44 animals/m^2), over a 12 month period (Morman 1987). Morman (1987) suggested that the cage environment favored growth despite maintaining animals at a higher density than is usually found in the wild. In all these cases, perhaps the lampreys' inability to escape confinement and disperse limited movement and reburrowing activity that normally occurs in streams, and reduced the level of energy loss.

Alternatively, larval growth might simply be limited by the

available food supply. Malmqvist and Brönmark (1981) concluded that larval lampreys suffered only minimal intraspecific competition for food, based on the observation that larval lampreys remove little of the total suspended organic material from a stream. However, Mallatt (1982) noted that, whereas most filter feeders rapidly process dilute suspensions, larval lampreys slowly process concentrated suspensions. Hence, the feeding mechanism of larval lampreys would tend to limit their effect on the supply of suspended food in a stream, which is usually in dilute suspension. However, the fact that lampreys do not affect the total supply of suspended food in a stream does not guarantee that larval growth is not food limited.

The effect of increased concentrations of suspended food on larval growth was readily noted in my low density tanks (Table 1). Although direct comparison may be somewhat misleading because of differences in water temperature between the two periods, increasing the food supply per feeding from 25 mL of slurry in the March to May period to 250 mL of slurry in the September to November period (a 10 fold increase) increased average weight change per animal from -0.052 to 0.716 g per study period. Increases in growth might also have occurred in the high density tanks, had I increased the amount of slurry per feeding. Animals in the high density tanks could probably have ingested more food at each feeding, based on the observed lower turbidity in the high density test tanks relative to the low density tanks at the end of a 16-h feeding period.

Larval growth may be limited less by the quantity than by the quality of food. The addition of commercial fry feed to the yeast in the laboratory diet significantly increased larval growth and survival (Swink, in review). In the wild, productivity varies among streams and nutritional value of lamprey food probably differs from the laboratory diet. Examination of gut contents from larval sea lampreys and northern brook lampreys Ichthyomyzon fossor from throughout the Great Lakes basin shows that about 97% of the diet consists of detritus (Sutton 1993). Assimilation of detritus is accomplished by passing small amounts slowly through the gut (Sutton 1993). More abundant or higher quality detritus may interact with lamprey density to affect growth between streams or between years within the same stream.

For all three factors, the distribution of larvae within a stream would play a role in suppressing growth. Larval habitat is not uniform, which results in the patchy distribution of larvae in a stream. Localized population densities were found to vary from 61 to 332 animals/m^2 in Salem Creek, Ontario (J. G. Weise, Canadian Department of Fisheries Oceans, personal communication). Larvae in a higher density population group (e.g., 332 animals/m^2; biomass, 147 g/m^2) would live in closer proximity to each other and be more affected by growth inhibitors in the substrate or tactile stimulation than larvae in a lower density (e.g., 61 animals/m^2; 31 g/m^2), more dispersed population group in the same stream. Even if food supply were the limiting

factor in growth, larvae in higher density groups should have consistently less food available in their local area than larvae in lower density groups, given a uniform supply of suspended food throughout a stream. Hence, growth of larvae in a particular year class could vary within a stream based on the variation in localized population density. However, this variation would only be detectable if no significant migration occurred among low- and high-density population groups within a stream.

Future studies might better be conducted in the field rather than the laboratory. A field study could be conducted that measures the quantity and quality of suspended food in various parts of a stream and compares the growth of larval lampreys relative to local population densities and local food availability. Such a study might require caging the larvae to prevent movement, but this could affect the distribution and deposition of food, and interfere with competition from other organisms.

Finally, the low mortality and generally high level of growth in my laboratory study confirms the benefits of supplemental feeding with a commercial fry feed (Swink, in review). Further studies could be conducted to optimize the growth of cultured lamprey larvae. However, increased feed levels would probably require more frequent cleaning of tanks and increase the risk of developing anaerobic sediments.

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Table 1. Mean weight change (g; ± 2 SE, ranges in parentheses) of larval sea lampreys held at two densities and subjected to water from Lake Huron (control) or effluent water from larval populations at two densities (5 or 50 animals per tank) from 1 March to 18 November 1994. Mean weight changes in a row with different letters were significantly different at the $P < 0.001$ level.

	5 animals per tank (~40 /m²) with effluent from			50 animals per tank (~400 /m²) with effluent from		
Study period	Control	Low density	High density	Control	Low density	High density
Mar - May a	-0.015 ± 0.020	0.000 ± 0.020	0.020 ± 0.026	0.144 ± 0.038	0.129 ± 0.012	0.131 ± 0.010
	(-0.068 to 0.038)	(-0.022 to 0.030)	(-0.020 to 0.046)	(0.100 - 0.168)	(0.118 - 0.154)	(0.118 - 0.152)
May - Jul b	0.067 ± 0.048	0.193 ± 0.156	0.126 ± 0.080	0.051 ± 0.012	0.083 ± 0.034	0.076 ± 0.038
	(-0.022 to 0.208)	(0.062 - 0.472)	(-0.028 to 0.204)	(0.008 - 0.090)	(0.044 - 0.139)	(0.004 - 0.119)
Jul - Sep b	0.115 ± 0.074	0.134 ± 0.108	0.162 ± 0.078	0.123 ± 0.022	0.080 ± 0.046	0.140 ± 0.018
	(-0.054 to 0.324)	(-0.017 to 0.308)	(0.072 - 0.268)	(0.068 - 0.176)	(0.020 - 0.143)	(0.082 - 0.121)
Sep - Nov c	0.751 ± 0.084y	0.744 ± 0.148y	0.633 ± 0.080y	0.229 ± 0.018z	0.194 ± 0.042z	0.211 ± 0.034z
	(0.572 - 0.908)	(0.622 - 1.032)	(0.528 - 0.724)	(0.163 - 0.264)	(0.152 - 0.267)	(0.143 - 0.240)

a Larvae held at 5 animals per tank were given 25 mL of yeast and fry feed slurry per feeding.

b Larvae held at 5 animals per tank were given 100 mL of yeast and fry feed slurry per feeding.

c Larvae held at 5 animals per tank were given 250 mL of yeast and fry feed slurry per feeding.