

**Use of Sucrose to Stimulate the Production
of Daphnia Pulex in Aquaria**

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USE OF SUCROSE TO STIMULATE THE PRODUCTION
OF DAPHNIA PULEX IN AQUARIA ¹✓

By Merle G. Galbraith Jr.

Abstract

Twenty aquaria were filled with lake water and inoculated with Daphnia pulex at the rate of 10 adults per liter. Sucrose was added to 15 aquaria and 5 were held as controls. Four experiments were run in sequence and sucrose was applied to the aquaria at the rate of 1.5, 2.0, 4.0, 6.0, 8.0, and 16.0 ppm. Periodic sampling of the daphnid population showed that with few exceptions, sucrose was not successful in stimulating the production of large populations of daphnids.

¹✓ Contribution from Dingell-Johnson Project F-35-R, Michigan.

Introduction

Good growth and survival of many game fish are often dependent on the abundance of zooplankton as food at one time or another in their life. Two of the most important factors governing the abundance of zooplankton in lakes are the degree of predation by planktivores and the availability of the right kind of food. Daphnia is probably one of the most important zooplankters affecting the growth of those fish which at one time or another become planktivorous.

The success of artificially producing walleye fry in Michigan ponds has been found to be dependent on the abundance of daphnids right after the yolk sac has been absorbed. A method for increasing the production of these zooplankters over that produced by current methods would be highly desirable. Any increase in the survival and production of walleyes would help further in meeting the demands of the fish managers for stocking walleyes in Michigan lakes.

Continued good growth and survival of rainbow trout in Michigan lakes appear to be associated with an abundance of large daphnids. Rainbow trout 4-12 inches in size consume large quantities of daphnids in late spring and fall. Based on studies of lakes treated with rotenone, it takes upward of a year or more before the daphnids can reestablish themselves at a density sufficient to provide an adequate source of food for trout. When these lakes are restocked before recovery of daphnids is possible, fish predation appears to exceed the production of daphnids and fishing quality is poor. If the food (bacteria, yeasts, etc.) upon which invertebrates are dependent was increased more rapidly after treatment then perhaps the recovery of the aquatic invertebrates could be accelerated and trout could be planted earlier. Thus an earlier buildup of the food chain in a newly reclaimed lake might increase the time span that a lake can provide high quality fishing. The addition of sucrose to these lakes and ponds might very well significantly increase fish production.

Knowledge of the use of sucrose or other forms of sugar to stimulate production of invertebrates in lakes or ponds is meager. To my knowledge only one study has been published--"Trout production in an experimental stream enriched with sucrose" (Warren et al. 1964). This study, however, cannot be considered directly applicable because of the difference in nature of running water vs. standing water. The study did lead the investigators to believe that the production of bacterium Sphaerotilus natans in the enriched areas was responsible for a two-fold increase in food consumption and a seven-fold increase in the production of trout. They attributed this increase to an increased abundance of aquatic benthic invertebrates, especially chironomid larvae. Algae production was lower in the enriched section than in the unenriched. These authors did not study the effect of sucrose on encrustaceans or on bacteria.

In New Mexico, sucrose was applied weekly to ponds at 1.5 and 2.0 ppm. D. B. Jester Jr. (personal communication) reported that the sucrose stimulated the production of bacteria and chrysophytes, and that zooplankton dominance shifted from copepods to daphnids. It is known that sugars enhance the production of yeasts as well as certain bacteria and that bacteria can be an important source of food for daphnids (Peterson and Hobbie 1978). The objective of the study was to measure in aquaria changes in numbers of daphnids produced by the addition of sucrose at varying concentrations and frequencies of application.

Methods

For the first experiment, aquaria were filled with water comprised of one-half deionized water and one-half lake water. Lake water alone was used in subsequent experiments. Water was collected from Maceday Lake, Oakland County, and was filtered through a nylon net of 76 micron mesh. This water contained 136 mg per liter total alkalinity and 0.029 mg per liter and 0.01 mg per liter of phosphates and nitrates, respectively. Fluorescent lighting, controlled by an electrical timer, was used; a light day ran from 6 AM to 10 PM. The aquaria were aerated and agitated continuously by

pumping air through filter stones. Water temperature and oxygen content were determined at each feeding. Water temperatures fluctuated very slowly and for each experiment variation was small. During the course of the entire study temperatures ranged between 18.9 C and 25.8 C. The oxygen content, which ranged between 6.0 and 9.0 ppm during the study, was determined with a Delta Scientific Model 85 oxygen meter.

Twenty aquaria were filled each with 34 liters of water. Sucrose was added to 15 aquaria and 5 more aquaria were held as controls. The sucrose-treated aquaria were divided into three lots of five aquaria each and each of these lots received a different concentration of sucrose. At the beginning of each experiment 340 adult-size Daphnia pulex from cultures were introduced into each aquaria. For the first three experiments the aquaria were all treated with the same concentrations of sucrose, that is, 1.5, 2.0, and 4.0 ppm in each of three lots. In between applications sucrose solutions were refrigerated. Aquaria in experiment number 1 were dosed at the rate of twice a week and those in experiments 2 and 3 were dosed everyday except weekends. Aquaria in the 4th (and last) experiment were treated with four times the previous concentrations, that is, 6.0, 8.0, and 16.0 ppm. They were treated everyday except weekends.

Sampling the daphnid population began after visual observations indicated that there was an apparent increase in density in at least some of the aquaria so that the removal of daphnids would not jeopardize the future potential for daphnid reproduction. A plexiglass cylinder 3.5 cm inside diameter was used to sample each aquarium. The cylinder was lowered vertically to the bottom, sealed with a rubber stopper at the bottom, and the column of water removed and preserved. Four samples were collected from each aquarium, one in each quadrant. The water was then replaced with filtered water. The volume of each sample was determined and the daphnids counted later.

The 20 aquaria were arranged in a mixed model array with 16 aquaria arranged in a Latin square. Four more aquaria were arranged randomly in order to determine the sampling error between tanks within rows.

Results

The aquaria were observed daily for increases in the daphnid population. During the first experiment sampling of the population began 3 weeks after the initial introduction. Two months after this experiment began only six aquaria still contained some living daphnids. Results of sampling during this first experiment indicated that the relationship between the density of daphnids vs. time was linear in the control and in the aquaria treated with 1.5 and 2.0 ppm of sucrose, and curvilinear in those treated with 4.0 ppm. Regression analyses showed that there was a significant decrease over time in the daphnid population in the control and in all the other aquaria except in those dosed at 1.5 ppm (Table 1). The daphnids in the aquaria dosed at 1.5 ppm declined similarly to those dosed at the other concentrations until the final sampling. At this time, for some unexplainable reason, the daphnid population in one of the five replicates exploded (33 liter⁻¹) causing too large a variation for the decline to be significant. It is also noteworthy that in the aquaria treated with 4.0 ppm, daphnid production increased during the first 18 days, but subsequently declined.

During the 54 days of the first experiment the average number of daphnids in all the aquaria treated with sucrose was significantly higher (P less than 0.05) than those in the control (Table 2). The highest number of daphnids was produced in aquaria dosed at 4.0 ppm; there was no significant difference between the average number of daphnids produced in the 1.5 and 2.0 ppm concentrations. Even when the individual aquaria are considered, the daphnid population was never very high. The analysis run to determine if outside influences had affected the results due to the position of aquaria in the rows showed no difference ($F = 0.443, 16 \text{ d.f.}$).

Officially this experiment terminated on June 13, 1979, but those aquaria still containing daphnids were treated with the standard dosages of sucrose daily instead of twice a week. For the next 2 weeks of feeding visual observations indicated very little change in the daphnid population. By the third week most daphnids were dead so aquaria were emptied and cleaned.

Experiment number 2 commenced August 1, 1979, but 5 days later few daphnids were alive. I suspect that the hose clamps, used to anchor the air stones, which had been used in an earlier toxicity experiment, remained toxic. The third and final experiment began on August 19, 1979. Sucrose was fed everyday and visual observations indicated good reproduction in some tanks and fair in others. Results of the first sampling 2 weeks later showed increases in the daphnids in the aquaria treated with 1.5 ppm and 4.0 ppm. In the control and in the aquaria treated with 2.0 ppm the daphnid population remained at the same density. During the next 2 weeks the daphnids began declining in all aquaria. A month after the study began two aquaria in each lot of the 2.0 and 4.0 ppm dosages and three control aquaria contained no daphnids. A week later only seven aquaria contained daphnids. There were so few remaining in these aquaria that the experiment was terminated.

After the final sampling in experiment number 3, a four-fold increase of sucrose was applied to the seven aquaria still containing daphnids. No noticeable stimulus occurred during 2 more weeks of feeding.

Regression analysis of data from experiment number 3 showed a significant decline in the daphnids for all dosages of sucrose. There was an early surge in the daphnids during the first 2 weeks just as there had been in experiment number 1, but daphnids in all aquaria declined thereafter. Production was greatest in the aquaria treated with 1.5 and 4.0 ppm sucrose during the first 2 weeks but there was no significant difference by the last day of the experiment. Independent testing for significant differences between rows from outside influences again showed no difference ($F = 0.633, 4 \text{ d.f.}$).

The fourth and final experiment commenced on November 20, 1979. Four times the amount of the initial dosages was applied to the aquaria on four subsequent days before the introduction of daphnids in an attempt to build up the bacteria. Eight days later daphnids were introduced into the tanks and the increased dosages of sucrose were applied daily except on weekends. Before the end of the first week most populations in the aquaria had declined drastically. In order not to jeopardize the few remaining

daphnids no more samples were collected. A month after this experiment began visual observations indicated that 13 aquaria contained no daphnids, 4 aquaria had one and 3 aquaria had a few. Four days later (December 30, 1979) no living daphnids remained. At this time I terminated this study because at no time did sucrose seem to have the potential to produce large populations of daphnids.

Summary

Daphnids in the control aquaria which contained nothing but a natural biota did poorly. In all experiments, as one might expect, the average number of daphnids declined; there probably was not enough food. But the addition of sucrose, which maintained a 4.0 ppm concentration, twice a week in experiment number 1, apparently was high enough to initially cause a short spurt in the population. Evidence that this spurt was more than due to chance alone is presented in experiment 3. In experiment 3 aquaria were treated with the same dosages of sucrose but on a daily basis rather than twice a week. Before their eventual decline daphnids in both the lowest and the highest sucrose concentrations increased during the first 14 days, and the daphnids in the 2.0 ppm concentration remained at their initial level.

The aquaria in the last experiment of this study were treated with a four-fold increase in sucrose, but by the first week of the experiment the daphnids had declined drastically.

Apparently sucrose alone is not sufficient to stimulate and sustain a large population of daphnids. The highest number of daphnids counted in any tank in the first experiment was 33 individuals per liter and in the third experiment, 44 per liter. By comparison with densities in natural ponds this is very low. Ward (1940) found peak densities of 1,000 to 2,000 per liter in small ponds, and Pennack (1946) gives a figure of 435 per liter in a larger body of water. Frank (1952) consistently maintained populations

greater than 500 per liter in his laboratory studies. Beyerle (1979) found densities of daphnids in his walleye holding ponds, fed manure and yeast, of 205 per liter and this with continuous predation on the daphnids by walleye fry.

Further evidence that sucrose by itself is not the answer for producing large quantities of daphnids is presented by Beyerle (1979). He compared population densities of daphnids in his sucrose-fed ponds versus manure-yeast fed ponds. Sucrose at 2.0 ppm was applied once a week in one pond and twice a week in another. His results indicated that twice-a-week applications of sucrose produced no more daphnids than weekly applications. And, his manure-yeast ponds produced much greater quantities of daphnids and faster fish growth. Sucrose may still be a cheap source of organic carbon for bacteria feeding and have practical application for fish management, but the role of other important factors, such as the nutrient input into the system, must be understood before sucrose can be used.

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Table 1. --Analyses of the decline of D. pulex fed different concentrations of sucrose.

Dosage	Number of observations	Regression for daphnids per liter	R ²
<u>Experiment 1</u>			
Control	25	8.72-0.18(D) ^a ✓	0.8867** ^b ✓
1.5 ppm	25	9.46-0.06(D)	0.1286
2.0 ppm	25	11.4-0.21(D)	0.8419**
4.0 ppm	25	6.9+3.3(D)-0.19(D)+0.0023(D)	0.9900**
<u>Experiment 3</u>			
Control	15	10.18-0.18(D)-0.002(D)	0.6414**
1.5 ppm	15	8.9+1.17(D)-0.034(D)	0.3944**
2.0 ppm	15	9.9+0.62(D)-0.0068(D)	0.5638**
4.0 ppm	15	8.2+1.81(D)-0.050(D)	0.5088**

^a✓ D = day of experiment.

^b✓ ** = significant at the 95% level.

Table 2.--Average number of daphnids in the control and in aquaria fed 1.5, 2.0, and 4.0 ppm of sucrose, summarized by day of sampling.

Day of experiment	Date (1979)	Average number of daphnids per liter by sucrose concentration			
		Control	1.5 ppm	2.0 ppm	4.0 ppm
<u>Experiment 1</u>					
1	April 19	10.0	10.0	10.0	10.0
18	May 7	5.2	12.8	10.6	20.4
21	May 10	3.8	5.8	7.4	15.0
25	May 14	3.0	7.0	5.2	9.0
28	May 18	4.0	4.0	4.6	5.0
54	June 13	0.0	7.6	0.0	6.6
	Average ^a ✓	3.2 ±1.4	7.4 ±1.4	5.6 ±1.4	11.2 ±1.4
<u>Experiment 3</u>					
1	August 16	10.0	10.0	10.0	10.0
14	August 30	7.2	18.6	9.5	23.8
39	September 24	0.0	3.0	2.0	3.2
	Average ^a ✓	3.6 ±2.5	10.8 ±2.5	5.8 ±2.5	13.5 ±2.5

^a✓ Does not include the first day; estimates with 95% confidence limits.

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