

**THE CAPACITY OF NATIVE FISH AND A FRESHWATER  
MUSSEL SPECIES TO CONTROL SUSPENDED SOLIDS  
IN WASTEWATER STABILIZATION PONDS**

by

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(ABSTRACT)

Stocking herbivorous aquatic organisms in wastewater treatment ponds specifically to control phytoplankton biomass and reduce suspended solids can provide small, rural communities with inexpensive, secondary wastewater treatment. The capability of several native fish species and the freshwater mussel, *Elliptio complanata*, to reduce suspended solids and phytoplankton was compared in laboratory and field enclosure experiments. Fathead minnows (*Pimephales promelas*), gizzard shad (*Dorosoma cepedianum*), and the common carp (*Cyprinus carpio*) had either no effect, or significantly increased suspended solids levels. None of the fish species consistently reduced concentrations of typical wastewater algal taxa. The ineffectiveness of fish was attributed to numerous algal characteristics such as cell size, shape, relative abundance, resistance to digestion, and palatability. In contrast, *E. complanata* consistently and substantially reduced both suspended solids and algae concentrations in wastewater. Small-sized algae and suspended particles were either directly assimilated, or removed from suspension as pseudofeces, further promoting clarification. The proportion of suspended solids and algae concentrations removed per individual mussel declined with increasing densities of mussels. Declines in suspended solids and algae attributable to mussel filtration were best described by semilogarithmic regression equations. Mean filtration rate of *E.*

*complanata* ranged from 53 ml/h/mussel for colonial blue-green algae, to 134 ml/h/mussel for smaller green algae. The results of this study suggest that freshwater mussels can effectively control suspended solids and algae in eutrophic environments.

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# Introduction

The use of stabilization ponds as a method of wastewater treatment is becoming increasingly popular. As relatively easy and inexpensive systems to operate and maintain, they represent a cost-effective means of wastewater treatment, particularly for small (population less than 10,000), rural communities (Middlebrooks et al., 1979; Tucker et al., 1979).

In a typical waste treatment system, wastewater flows serially through a primary, secondary, and polishing pond or cell. Solid wastes settle out in the primary cell, or are removed by a separate primary clarifier, such as an Imhoff tank. In the primary and secondary cells, bacteria and fungi oxidize organic matter into elemental compounds such as nitrate, sulfate, phosphate, carbon dioxide, ammonia, and methane. Algae utilize nutrients, carbon dioxide, and sunlight near the surface and produce free oxygen required by aerobic bacteria. Finally, biological oxygen demand and suspended solids are reduced to acceptable levels in the polishing cell, the effluent of which is frequently discharged into a receiving stream or river.

A major disadvantage of stabilization ponds is the production of large quantities of algae (Middlebrooks et al., 1979; Dinges 1982). Algal cells can contribute significantly to effluent suspended solids, imposing an organic matter burden on receiving waters (Zickefoose and Hayes 1977; Tucker et al., 1979). As a major stream pollutant, suspended solids reduce oxygen and light transmittance, and smother the stream benthos with a blanket of solids (Austin and Engelbrecht 1968).

Secondary waste treatment, as defined by the EPA, requires a 30-day mean value of 30 mg/L or less of suspended solids. Suspended solids, primarily in the form of algae, can exceed 100 mg/L in some ponds, although such a high level is generally limited to a few months of the year (Middlebrooks et al., 1979). Seasonal changes in suspended solids typically mirror algal dynamics, with highest levels occurring during summer and at spring and fall overturn.

Many algal control techniques have been proposed, but few have been effective (Tucker et al., 1979). Methods such as filtration and alum coagulation are costly and labor-intensive. Chlorination kills algae, but the organics released by dead cells increase the biological oxygen demand of the effluent (Zickefoose and Hayes 1977). Copper sulfate is frequently used to destroy algae during summer months, but this practice may simply substitute one pollutant for another.

Filter-feeding, herbivorous fish may provide an effective, inexpensive approach to suspended solids control, although few conclusive studies have been conducted. Henderson and Wert (1976) concluded that for small municipalities, waste stabilization ponds utilizing herbivorous fish are the most cost-effective strategy to meet current secondary treatment standards.

In Asia, filter-feeding finfish are cultured in algae-rich wastewater to produce an inexpensive, protein-rich food source (Edwards 1980). While aquaculture has focused on maximizing fish production, the culture of selected animal species for the sole purpose of improving water quality is a recent and largely unexplored concept (Dinges 1982). Successful use of herbivorous fish as biological filters for wastewater treatment depends upon food availability in terms of size and quality. Additionally, fish must tolerate unstable physicochemical conditions characteristic of small treatment plant ponds and lagoons.

Henderson (1983) found that silver carp (*Hypophthalmichthys molitrix*) and bighead carp (*Aristichthys nobilis*) stocked in an Arkansas sewage lagoon system significantly reduced suspended solids. In some environments, silver carp feed primarily on phytoplankton, whereas bighead carp consume larger-sized algae, zooplankton, and detritus (Cremer and Smitherman 1980). Both fish species are hardy and tolerate low levels of dissolved oxygen.

In contrast, Smith (1985) found that silver carp significantly enhanced phytoplankton growth through predation on zooplankton grazers. However, a mesh refuge virtually eliminated fish predation on zooplankton in a portion of the water column, and facilitated a significant reduction in algal biomass. Feeding niches of silver carp and zooplakton spanned a broad and complementary size range of phytoplankton, providing an integrated or multi-species approach to biological wastewater clarification.

Filter-feeding blue tilapia (*Sarotherodon aurea*) suppress populations of large phytoplankton, but enhance growth of algal taxa too small to be effectively consumed (Drenner et al., 1984). Gizzard shad (*Dorosoma cepedianum*), a North American filter-feeder, has also been shown to stimulate algal growth (Drenner et al., 1986). Many other planktivorous fish species indirectly enhance phytoplankton by consuming filter-feeding invertebrates (Andersson et al., 1978; Lynch and Shapiro 1981).

The complex relationships between grazing and phytoplankton community structure make it difficult to predict herbivorous fish effects on a sewage lagoon plankton community. Grazing efficiency greatly depends upon phytoplankton size. Because fish act as size-selective predators on the plankton community, a complementary group of species may be necessary to control a wide size range of plankton. In addition to algal size, other factors can influence the effectiveness of grazing. Some algae avoid grazing by being toxic or undigestible (Porter 1977). Fish recycle nutrients that can enhance phytoplankton growth (Meyer et al., 1983). Despite these complexities, planktivorous

fish consume phytoplankton under certain conditions, and might therefore reduce or control suspended solid levels in stabilization ponds.

Exotic, herbivorous, filter-feeding cyprinids and cichlids appear best suited to in-pond removal of suspended solids (Dinges 1982). However, in many regions there are legal constraints on exotic species introduction. Genetic research has produced sterile hybrids and polyploids of some species, but they are expensive and their food habits may differ from the diploid, parental strains (Young et al., 1983).

No fish species native to North America exhibit the same feeding habits as the Asian silver carp or African tilapia species. Fathead minnows (*Pimephales promelas*) and gizzard shad consume a variety of prey items including algae (Becker 1983). Fathead minnows have been cultured in wastewater, although success depends upon the presence of adequate dissolved oxygen and maintenance of sublethal levels of ammonia (Hall and Shelton 1983). Following gill raker development, young gizzard shad feed mainly on phytoplankton as pump filter-feeders, but their diet shifts to zooplankton and detritus as they grow (Mummert and Drenner 1986).

Fathead minnows and young gizzard shad could be useful in controlling medium to large (greater than 100  $\mu\text{m}$ ) algae in a hypereutrophic stabilization pond. However, native fish species may not effectively control smaller phytoplankton that frequently inhabit wastewater ponds and lagoons (e.g., *Scenedesmus*, *Chlamydomonas*, and *Chlorella* spp.). Filter-feeding invertebrates are adapted to remove very small (down to 1  $\mu\text{m}$ ) particles from suspension (Jorgenson 1975), and are the primary grazers of phytoplankton in natural ecosystems (Porter 1977). Culturing these organisms for the purpose of controlling suspended solids has received little attention.

Cladoceran zooplankton have relatively high filtering rates for particles from 1 to 50  $\mu\text{m}$  (Allan 1976). Suspension-feeding zooplankton (e.g., *Daphnia* spp.) are often present in stabilization ponds (Dinges 1982); however, they are susceptible to predation

by planktivorous fish (Smith 1985). In addition to zooplankton, filter-feeding midge larvae of the family Chironomidae are not only abundant in many sewage lagoons, but may consume significant amounts of algae and remove energy as emerged adults (Kimerle and Anderson 1971).

Among filter-feeding mollusc species, only the Asiatic clam, *Corbicula fluminea*, has been investigated as a potential biological filter in catfish rearing ponds (Buttner 1986a; 1986b), and in wastewater treatment (Haines 1977). However, *Corbicula fluminea* is an introduced species, and can be an ecological and economic threat (McMahon 1983). Other native, freshwater bivalve species need to be evaluated for their capability to reduce suspended solids and phytoplankton in wastewater. The eastern elliptio (*Elliptio complanata*) is a large unionid species that occurs in both lotic and lentic habitats (Matteson 1948). It is locally abundant in the James River drainage of Virginia. Various aspects of its biology have been studied, including filtration rate (Paterson 1984), annual production (Strayer et al., 1981), depth distribution in a Canadian lake (Ghent et al., 1978), shell organic content (Cameron et al., 1979), and life history (Matteson 1948). By introducing a species complex composed of both large particle-grazers (e.g., native cyprinids or gizzard shad) and small particle-grazers (e.g., a unionid mussel species) into a waste treatment pond, effluent suspended solids might be suppressed.

The primary objective of this study was to evaluate the capability of fathead minnows, gizzard shad, and the eastern elliptio mussel to reduce suspended solids under controlled laboratory conditions. Grazing effects on different-sized cells and algal taxa were evaluated. This screening process identified the species with the greatest potential to control suspended solids. The most effective species were selected for evaluation under field conditions at the new sewage treatment plant in New Castle, Virginia. My specific objectives were as follows.

1. To compare the capability of fathead minnows, gizzard shad, and eastern elliptio mussels to reduce suspended solids and different taxa and size classes of phytoplankton in suspensions of wastewater under laboratory conditions.
2. To evaluate the most effective candidate species for their capability to reduce suspended solids and algae in wastewater suspensions at the New Castle Sewage Treatment Plant under controlled, field conditions.
3. To compare the effects of different densities of grazing organisms on reductions of suspended solids and algae.
4. To measure the time course of suspended solids and algae reductions by mussels, and to estimate filtration rate of eastern elliptio mussels.

# Methods

## Laboratory Grazing Trials

Eastern elliptio mussels, fathead minnows, and gizzard shad were evaluated for their capability to reduce suspended solids and phytoplankton in grazing trials conducted from July 9 to October 31, 1987. Fish and mussels used in these trials were obtained as follows. Fathead minnows were purchased in May and June, 1987 from Zett's Tri-State Fish Hatchery, Inwood, West Virginia, and from the Fish Wagon, Incorporated, Christiansburg, Virginia. Gizzard shad were collected in September, 1987 by electroshocking in a backwater of the Kanahwa River near Winfield, West Virginia. Gizzard shad were transported to Blacksburg, Virginia in oxygen-injected hauling tanks to which salt (2 g/L), quinaldine (2.5 mg/L), and furacin (10 mg/L) were added. Eastern elliptio mussels were collected in August, 1987 from the James River near Scottsville, Virginia. Fish and mussels were held in recirculating aquaculture systems at the Center Woods facility of the Virginia Tech Department of Fisheries and Wildlife Science for at least 2 weeks prior to any experiments. The animals were fed ground trout chow *ad libitum* prior to and between subsequent grazing trials.

Grazing trials were conducted in a laboratory in Cheatham Hall, Virginia Tech, Blacksburg, Virginia. Light was provided by fluorescent lamps on a 14 h light, 10 h dark photoperiod. In each trial, 4 to 8 aquaria (39.5 L) were filled with municipal wastewater and aged, aerated tap water to a volume of 7 to 10 L. The wastewater was obtained

from a primary sewage treatment lagoon serving Massie's Mobile Home Park, Montgomery County, Virginia. Water temperature was 21 to 22° C. An airstone (30.5 cm length) was placed in the bottom of each aquarium to continuously mix and aerate the water. Two aquaria were randomly selected as controls to monitor algal settling, growth, or senescence. The remaining 2 to 6 aquaria were stocked with one of the candidate species. Trial dates, number of replicates, and the number and size of animals stocked in each trial are shown in Table 1. Trials were initiated when the animals were stocked, and completed after 60 hours. Mortalities were replaced with new animals to maintain a constant stocking density.

At the beginning and end of each trial, a 300 ml water sample was collected from each aquarium for suspended solids analysis and cell enumeration. The sample was a composite from random areas within each aquarium in the event that the suspension was not evenly mixed and circulated by the airstone. Suspended solid and cell concentrations were each measured in duplicate and averaged for each replicate. The percent change was calculated from the pre and post levels (Appendix A). A t-test was used to compare the mean percent change between stocked and control aquaria in each trial. The summary data were calculated by subtracting the mean percent change in control aquaria from the percent change in each stocked replicate. This net change in stocked aquaria was interpreted as the percent change attributable to grazing or other activities of the fish or mussel species.

Suspended solids (mg/L) were measured by standard methods (A.P.H.A. 1981). Phytoplankton concentrations (cells/ml) were determined as follows. A 0.05 ml subsample was placed on a microscope slide under a cover slip. Cells of the predominant algal taxa were counted in one field-strip width across the middle of the cover slip at a magnification of 450 x. The ratio of field-strip width to cover slip width was used to

**Table 1.** Stocking densities and mean lengths and weights of eastern elliptio mussels, fathead minnows, and gizzard shad in 1987 laboratory grazing trials. Standard deviations of mean lengths and weights are in parentheses.

| Species          | Trial | Date<br>(month/day) | Stocked<br>aquaria | Density<br>(number/7 l) | $\bar{X}$ length<br>(mm) | $\bar{X}$ weight<br>(g) |
|------------------|-------|---------------------|--------------------|-------------------------|--------------------------|-------------------------|
| Eastern elliptio | 1     | 8/20                | 2                  | 15 <sup>a</sup>         | 98.1 (7.5)               | 92.4 (18.9)             |
|                  | 2     | 8/10                | 4                  | 4                       |                          |                         |
|                  | 3     | 8/13                | 6                  | 8                       |                          |                         |
|                  | 4     | 8/30                | 4                  | 15                      |                          |                         |
| Fathead minnow   | 1     | 7/9                 | 4                  | 20                      | 50.2 (5.8)               | 1.1 (0.4)               |
|                  | 2     | 7/14                | 4                  | 10                      |                          |                         |
|                  | 3     | 8/20                | 2                  | 80 <sup>a</sup>         |                          |                         |
|                  | 4     | 8/3                 | 2                  | 20                      |                          |                         |
|                  | 5     | 8/3                 | 2                  | 10                      |                          |                         |
| Gizzard shad     | 1     | 9/29                | 4                  | 18                      | 30.0 - 125.0             | 0.6 - 15.3 <sup>b</sup> |
|                  | 2     | 10/2                | 4                  | 18                      |                          |                         |
|                  | 3     | 10/6                | 2                  | 10                      |                          |                         |
|                  | 4     | 10/6                | 2                  | 5                       |                          |                         |
|                  | 5     | 10/9                | 2                  | 10                      |                          |                         |
|                  | 6     | 10/9                | 2                  | 10                      |                          |                         |

<sup>a</sup> number/10 L

<sup>b</sup> estimated using length-weight regression of Bodola (1965)

convert strip counts to the number of cells/0.05 ml. This concentration was multiplied by 20 to convert to cells/ml.

The cell counting procedure assumed that algae cells were randomly distributed on the counting slide, and that the number of cells in one strip count was representative of the total number of cells under the cover slip (e.g. in the entire 0.05 ml sample). This assumption was evaluated using a modification of Schulte's (1975) procedure for measuring variability in cell enumeration. A strip count of *Chlorella* cells was made across the top, middle, and bottom of the cover slip for three 0.05 ml subsamples of wastewater. The mean strip counts for the 3 subsamples were 22.00, 21.33, and 19.33 cells/strip, and the sample standard deviations (sd) were 1.41, 1.70, and 1.25 cells/strip, respectively. The corresponding coefficients of variation (CV) were 6.41, 7.97, and 6.47 % of the mean (mean CV = 6.95 % of the mean). The relatively low CV's indicated that cells were randomly distributed on the counting slides, and one strip count across the middle of the cover slip was representative of the number of cells/0.05 ml. Variability associated with subsampling was evaluated by counting all *Chlorella* cells in three 0.05 ml subsamples. The mean was 342 cells/0.05 ml (sd = 25.57 cells/0.05 ml), and the CV was 7.48 % of the mean.

## New Castle Grazing Experiment

The newly constructed New Castle sewage treatment plant (STP) in Craig County, Virginia began operations in early summer, 1988. The system receives primary treated wastewater which is detained and treated in 3 ponds, serially. Wastewater from the final pond in the series is chlorinated, dechlorinated, and then discharged into Craig Creek.

During this study, water quality in pond 3 was quite good and suspended solid and BOD<sub>5</sub> levels were below the 30 mg/L standard. However, seasonal algal blooms could cause declines in effluent quality in the future. Biological controls for suspended solids are probably best suited to finishing or polishing ponds in the series because environmental conditions generally facilitate survival of herbivorous or filter-feeding organisms. The grazing trial was conducted from September 27 to 29, 1988 at New Castle using wastewater from pond 3.

The candidate species evaluated in this experiment were the eastern elliptio, fathead minnows, and Israeli carp. Gizzard shad were not included in the experimental design because they demonstrated little potential as a biological control species during the 1987 laboratory grazing trials. Israeli carp were readily available and are commonly sold for algae control in farm ponds. The experimental animals were obtained as follows. Fathead minnows were purchased in August, 1988 from Windmill Fish Hatchery, Kernersville, North Carolina. Israeli carp were purchased in August, 1988 from Zett's Tri-State Fish Hatchery. Eastern elliptio mussels were collected in July, 1988 from the James River near Scottsville, Virginia.

Fish and mussels were held in separate floating cages in the upper 0.5 meter of pond 3. Cages consisted of PVC frames around which was attached plastic netting (6 or 13 mm mesh). Fish cages were 55 x 55 x 60 cm. Mussel cages were 120 x 30 x 20 cm.

Pond 3 is approximately 4,460 m<sup>2</sup> in surface area with a mean depth of 3 m. It is 146 m in length and 35.5 m in width. The pond is equipped with 16 submerged aerators which are operated by an electronic timer. Throughout the period that fish and mussels were held in the pond, aeration was supplied daily for at least 15 m/h.

Sixteen plastic buckets (19 L) were filled with 7 L of wastewater from pond 3. To mix and aerate the water, air was supplied continuously from a compressor to an airstone (15.3 cm length) on the bottom of each bucket. The buckets were floated at the

surface on plywood rafts and PVC frames which submerged 50 % of each bucket. During the experiment, diel water temperature in the buckets ranged from 19° C in the early morning to 23 ° C in the late afternoon. The buckets were randomly assigned a stocking of one of the 3 species, or were unstocked to serve as controls. Mean stocking density and mean length and weight for each species were as follows; 8 mussels (101.8 mm shell length, 117.5 g wet weight with shell), 60 fathead minnows (63.1 mm total length, 3.0 g wet weight), and 3 Israeli carp (166.0 mm total length, 79.3 g wet weight). The number of replicates was 4 for each treatment level; control, mussels, fathead minnows, and Israeli carp.

Experimental duration was 60 h. Pre and post suspended solid levels and cell counts (Appendix B) were determined in the same manner described for the 1987 laboratory grazing trials. ANOVA and Duncan's multiple range comparisons were used to test for significant differences in the percent change in suspended solid and algae concentrations among the 4 treatment levels. Summary data are presented as the percent change attributable to the organisms.

## **Density-Specific Experiments**

The effect of 3 mussel densities on suspended solids and algae reduction was evaluated in two experiments. One was conducted in the laboratory on October 29 and 31, 1987 using wastewater from the primary treatment lagoon serving Massie's Mobile Home Park. The other experiment was conducted at New Castle on October 4 and 5, 1988 using wastewater from pond 3. The materials, source of animals, and sampling

procedures in these density-specific experiments were the same as described previously for the laboratory and field grazing trials, except for the differences described below.

In the laboratory experiment, 8 aquaria were filled with wastewater and aged, aerated tap water to a volume of 7 L. The aquaria were randomly assigned one of 4 treatment levels; unstocked control, 4, 8, or 12 mussels (97.4 mm mean shell length, 86.5 g mean wet weight with shell). The experimental duration was 24 h. Water temperature was 21° C. Pre and post suspended solids and algae concentrations were measured as described previously. This experiment was repeated on 3 consecutive days with different mussels and newly acquired wastewater in order to increase the number of replicates to a total of 6 for each treatment level. Analysis of covariance (ANCOVA) was calculated on the pooled data to test for both significant and parallel regression coefficients, using the pre levels of suspended solids and cell counts as covariates. If the regressions were significant and parallel, then an LSMEANS (SAS 1985) procedure was used to test differences in adjusted means among treatment levels. If the regression of percent change on pre levels was not significant, ANOVA and Duncan's multiple range comparisons were used to test significant differences among treatment levels. Summary data were calculated as the percent reduction in suspended solids or algae attributable to mussels.

In the 1988 mussel density experiment conducted in the field at New Castle, 12 buckets were filled with 10.5 L of wastewater. During the experiment, water temperature in the buckets ranged from 19° C in the early morning to 22 ° C in the late afternoon. The buckets were randomly assigned one of 4 treatment levels; control, 4, 8, or 12 mussels (shell length, 102.5 mm; wet weight with shell, 117.3). Experimental duration was 24 h. The total number of replicates was 3 for each treatment level. Suspended solids and cell concentrations are summarized in Appendix D. ANOVA and Duncan's multiple range comparisons were used to test significant differences between treatment levels. Summary data were calculated as the percent reduction attributable to mussels.

## Time-Specific Effects on Mussel Filtration

In the prior experiments and grazing trials with mussels, the change in suspended solid and algae concentrations was measured over a 24 or 60 hour time period. The time-specific experiment was designed to measure reductions in suspended solids and phytoplankton over shorter time intervals within a 24 hour period, and also to measure filtration rate of eastern elliptio mussels on a dense wastewater suspension of algae. The experiment was conducted in a lab at Cheatham Hall, Virginia Tech. The source of the mussels was the 1988 James River collection which were held in pond 3 at the New Castle STP.

Eight aquaria were filled with aged, aerated tap water and wastewater from Massie's Mobile Home Park lagoon to a total volume of 7 L. Four aquaria were randomly assigned as controls, and 4 aquaria each were stocked with 8 mussels (mean shell length 104.6 mm; mean wet weight with shells 123.2 g). The water was aerated and mixed continuously as previously described. The experiment was begun at 8:00 AM on August 19, 1988, and concluded 24 hours later on August 20, 1988. Water temperature was 22° C.

A water sample (300 ml) was collected from each aquarium at intervals of 0, 2, 4, 8, 16, and 24 hours after the mussels were stocked. Suspended solids were measured and algal cells counted as in previous experiments with one exception. Each blue-green algal colony was counted as a single entity, rather than counting the number of cells in each colony. This was found to be necessary because of the short time intervals between sampling, and the need to process samples quickly with a minimum of storage time. Least squares regression analyses were used to analyze the change in suspended solid

and algae concentrations over time. A t-test was used to compare regression coefficients between control and stocked aquaria.

Filtration rate is the volume of water cleared of particles per unit of time. Mean filtration rate of eastern elliptio mussels was calculated from the cell counts during the first 2 hours of the experiment (e.g. at 0 and 2 h). This time interval was used for the following reasons. Filtration rates of bivalves differ as a function of particle concentration (Jorgensen 1975). The continual decline in concentration due to mussel filtration in a closed system represents a confounding factor in the estimation of filtration rate by indirect means, that is by the change in particle concentration over time (Winter 1978). A large change in particle concentration is typically avoided by measuring filtration rate over short time intervals such as 2 hours or less. Mussels produce significant amounts of pseudofeces when particle concentration exceeds some threshold (Winter 1978). When filtration rate is measured indirectly, the resuspension of pseudofeces would result in the underestimation of filtration rate. In previous grazing trials, mussels produced pseudofeces when filtering wastewater. Therefore, the probability of pseudofeces resuspension was minimized by measuring filtration rate over the first 2 hours of the experiment. Finally, variability in phytoplankton concentrations was expected to increase with time due to differential algal growth and senescence between control aquaria and those stocked with mussels. Such confounding effects probably were minimized by calculating filtration rate from the change in cell concentrations during the first 2 hours of the experiment.

Filtration rate was calculated with Quayle's (1948) equation as cited in Coughlan (1969):

---

$$m = \frac{M}{nt} \left[ \left( \log_e \frac{conc_0}{conc_2} \right) - \left( \log_e \frac{conc_0'}{conc_2'} \right) \right]$$

---

where  $m$  is the filtering rate (ml/h/mussel),  $M$  is the total volume (6,700 ml),  $n$  is the number of animals (8),  $conc_0$  and  $conc_2$  are the cell concentrations initially and after 2 hours in the stocked aquaria, and  $conc_0'$  and  $conc_2'$  are those in the control aquaria.

## Growth and Survival of Mussels

Mussels collected in July, 1988 were held in cages in pond 3 at the New Castle STP, except for short periods of time when experiments were conducted. This permitted some observations on the ability of eastern elliptio mussels to survive and grow in a sewage treatment pond. On August 19, 1988, a subsample of 32 individuals was measured and weighed. Shell length was measured to the nearest 0.1 mm with calipers. Mussels were rinsed to remove periphyton and seston attached to the shells, blotted dry, and weighed to the nearest 0.1 g (wet weight with shell). Mussel survival was not quantified between July, 1988 when the animals were collected and October 20, 1988, when all the experiments had been completed. However, mortality was relatively low, and percent survival was estimated to be in excess of 90 %.

Percent survival and weight change were quantified over the interval from October 20, 1988 to June 3, 1989 (approximately 8 months). Ninety-two mussels were measured

and weighed on October 20, 1988. Individual mussels were marked by gluing a specific number of plastic disc tags (6 mm) on the shells in a unique pattern. The mussels were placed in 2 cages (55 x 55 x 60 cm) constructed of wooden frames and plastic netting. The cages were suspended approximately 60 cm above the bottom (depth, 2.4 m) and 50 cm above 2 submerged aerators in pond 3. On June 3, 1989, the cages were raised, the percent of mussels surviving was calculated, and the remaining live mussels were suspended in cages in the upper 20 cm of the water column. On June 28, 1989, the surviving mussels were measured and weighed. Tag loss was low, less than 1 %.

# Results

## Laboratory Assessment of Grazing

Mean percent change in suspended solids varied considerably among species (Table 2). Mean initial suspended solid levels generally ranged from 68 to 114 mg/L, although a higher level (180 mg/L) was present during one experiment. *Elliptio complanata* was the only candidate species that significantly reduced suspended solids. The overall mean reduction was 66.0 % (range: - 57.7 to - 75.8 %) relative to reference aquaria. Suspended solids reductions in aquaria stocked with mussels were visually evident by increased water clarity and an accumulation of discrete bundles of algae (pseudofeces) on the enclosure bottoms. These changes were apparent within the first 24 hours of each experiment. In contrast to mussels, neither fish species reduced suspended solids. Fathead minnows had no significant effect on suspended solids, and the overall mean change was 4.1 % (range: - 1.9 to 11.6 %). Gizzard shad significantly increased suspended solid levels in 3 trials. The overall mean change was 45.7 % (range: 11.0 to 69.8 %).

*Anacystis* (Cyanophyta) was the predominant algal taxon, comprising 48.4 to 92.7 % of the total cell counts during all laboratory experiments. *Anacystis* occurs as colonies of small cells enclosed in a gelatinous sheath, and is characteristic of highly eutrophic environments such as sewage lagoons (Dinges 1982). Colony size ranged from 30 to 250  $\mu\text{m}$ . *Oscillatoria* (Cyanophyta) filaments were next in abundance and occurred

**Table 2.** Initial level and the net percent change in suspended solids (SS) in laboratory grazing trials with eastern elliptio mussels, fathead minnows, and gizzard shad. The p-values correspond to t-tests on the mean percent change in suspended solids between reference and stocked aquaria in each trial.  $\bar{X}$  is overall average of the grazing trial means for each species.

| Species          | Trial     | Stocked aquaria | Initial SS (mg/L)<br>$\bar{X}$ | Net % change<br>$\bar{X}$ (SE) | p-value |
|------------------|-----------|-----------------|--------------------------------|--------------------------------|---------|
| Eastern elliptio | 1         | 2               | 110.0                          | - 64.4 (2.7)                   | 0.010   |
|                  | 2         | 4               | 114.3                          | - 57.7 (10.8)                  | 0.024   |
|                  | 3         | —               | —                              | —                              | —       |
|                  | 4         | 4               | 113.8                          | - 75.8 (6.2)                   | < 0.001 |
|                  | $\bar{X}$ | —               | —                              | - 66.0 (5.3)                   | —       |
| Fathead minnow   | 1         | 4               | 88.1                           | 6.0 (5.2)                      | 0.704   |
|                  | 2         | 4               | 95.5                           | 0.8 (6.7)                      | 0.942   |
|                  | 3         | 2               | 103.7                          | 11.6 (6.8)                     | 0.307   |
|                  | 4         | —               | —                              | —                              | —       |
|                  | 5         | 2               | 180.0                          | - 1.9 (10.5)                   | 0.934   |
|                  | $\bar{X}$ | —               | —                              | 4.1 (3.0)                      | —       |
| Gizzard shad     | 1         | —               | —                              | —                              | —       |
|                  | 2         | 4               | 113.0                          | 72.2 (5.9)                     | 0.001   |
|                  | 3         | 2               | 68.0                           | 13.2 (0.3)                     | 0.025   |
|                  | 4         | 2               | 71.0                           | 29.3 (5.1)                     | 0.033   |
|                  | 5         | 2               | 77.5                           | 83.7 (45.0)                    | 0.159   |
|                  | 6         | 2               | 92.5                           | 30.0 (21.7)                    | 0.301   |
|                  | $\bar{X}$ | —               | —                              | 45.7 (13.7)                    | —       |

in 14 of 15 experiments. It contributed 5.8 to 35.9 % to the initial total cell counts, but generally comprised less than 12 %. Filaments ranged in length from 50 to 300  $\mu\text{m}$ . Green algae (Chlorophyta) comprised the smallest (0.3 to 17.7 %) proportion of algae, generally less than 5 % of the total cell counts. Chlorophyta cells were the smallest (6 to 30  $\mu\text{m}$ ) in the phytoplankton assemblage. *Scenedesmus* was the most abundant green algae taxon, but the group included smaller populations of *Ankistrodesmus*, *Chlorella*, *Closterium*, and *Phacus*.

Mussels reduced *Anacystis* in all 4 experiments (Table 3), with an overall mean reduction of 63.6 % (range: - 31.3 to 131.1 %). Since *Anacystis* colonies comprised the majority (88 to 92 %) of phytoplankton, their effective removal by mussels presumably accounted for the marked reductions in suspended solids. Fathead minnows had little effect on *Anacystis* populations, with an overall mean change of 2.6 % (range: -0.1 to 25.4 %). Gizzard shad increased *Anacystis* populations in most trials, but the changes were not significantly different from those in reference aquaria. The overall mean change attributable to gizzard shad was 9.2 % (range: - 19.9 to 25.6 %).

The effect of each species on *Oscillatoria* was more variable when compared to *Anacystis* (Table 4). The overall mean reduction of 36.7 % (range: -90.7 to 16.9 %) by the eastern elliptio indicated that *Oscillatoria* was filtered less effectively than *Anacystis*. Nevertheless, the overall mean reduction was greater for mussels than for either fish species. The overall mean change attributable to fathead minnows was - 10.0 % (range: -55.1 to 16.3 %), but the percent changes were not statistically significant. Gizzard shad enhanced *Oscillatoria* populations overall by 20.9 %, but the mean response among experiments was quite variable (range: -145.8 to 168.7 %), and included both significant reductions and increases.

Mussels provided the most effective biocontrol for Chlorophyta cell concentrations (Table 5). Although the proportional reduction was not statistically significant in all

**Table 3.** Initial concentration and the net percent change in *Anacystis* populations in laboratory grazing trials with eastern elliptio mussels, fathead minnows, and gizzard shad. (%) is the percent of initial phytoplankton counts comprised of *Anacystis* cells. The p-values correspond to t-tests on the mean percent change in *Anacystis* between reference and stocked aquaria in each trial.  $\bar{X}$  is the overall average of the grazing trial means for each species.

| Species          | Trial     | Stocked aquaria | $\bar{X}$ initial <i>Anacystis</i> cells/ml x 10 <sup>4</sup> (%) | Net % change $\bar{X}$ (SE) | p-value |
|------------------|-----------|-----------------|---|-----------------------------|---------|
| Eastern elliptio | 1         | 2               | 351.0 (89.5)  | - 39.8 (2.4)                | 0.098   |
|                  | 2         | 4               | 176.4 (87.7)  | - 31.3 (6.8)                | 0.083   |
|                  | 3         | 6               | 123.6 (81.8)  | - 52.2 (8.7)                | 0.017   |
|                  | 4         | 4               | 369.0 (91.9)  | - 131.1 (7.9)               | < 0.001 |
|                  | $\bar{X}$ | —               | —   | - 63.6 (22.9)               | —       |
| Fathead minnow   | 1         | 4               | 43.7 (82.3)   | - 8.7 (8.1)                 | 0.551   |
|                  | 2         | 4               | 82.5 (48.4)   | - 10.5 (4.4)                | 0.691   |
|                  | 3         | 2               | 257.4 (88.8)  | 25.4 (17.0)                 | 0.357   |
|                  | 4         | 2               | 639.9 (92.7)  | 4.0 (2.1)                   | 0.258   |
|                  | 5         | 2               | 858.6 (93.0)  | 2.8 (2.2)                   | 0.392   |
|                  | $\bar{X}$ | —               | —   | 2.6 (6.4)                   | —       |
| Gizzard shad     | 1         | 4               | 100.3 (82.5)  | 6.3 (9.5)                   | 0.682   |
|                  | 2         | 4               | 92.3 (88.0)   | 18.8 (11.5)                 | 0.402   |
|                  | 3         | 2               | 82.8 (83.1)   | - 19.9 (0.9)                | 0.380   |
|                  | 4         | 2               | 77.4 (88.1)   | 25.6 (1.7)                  | 0.288   |
|                  | 5         | 2               | 59.4 (88.5)   | 21.4 (5.4)                  | 0.059   |
|                  | 6         | 2               | 85.5 (87.9)   | 3.0 (4.3)                   | 0.559   |
|                  | $\bar{X}$ | —               | —   | 9.2 (6.8)                   | —       |

**Table 4.** Initial concentration and the net percent change in *Oscillatoria* populations in laboratory grazing trials with eastern elliptio mussels, fathead minnows, and gizzard shad. (%) is the percent of phytoplankton counts comprised of *Oscillatoria* cells. The p-values correspond to t-tests on the mean percent change in *Oscillatoria* between reference and stocked aquaria in each trial.  $\bar{X}$  is the overall average of the grazing trial means for each species.

| Species          | Trial     | Stocked aquaria | $\bar{X}$ initial <i>Oscillatoria</i> cells/ml x 10 <sup>4</sup> (%) | Net % change $\bar{X}$ (SE) | p-value |
|------------------|-----------|-----------------|--|-----------------------------|---------|
| Eastern elliptio | 1         | 2               | 40.1 (10.2)  | - 77.3 (2.0)                | 0.003   |
|                  | 2         | 4               | 20.7 (10.3)  | 4.4 (23.2)                  | 0.905   |
|                  | 3         | 6               | 24.6 (16.3)  | 16.9 (12.5)                 | 0.486   |
|                  | 4         | 4               | 30.8 (7.7)   | - 90.7 (8.4)                | 0.057   |
|                  | $\bar{X}$ | —               | —  | - 36.6 (27.6)               | —       |
| Fathead minnow   | 1         | 4               | —  | —                           | —       |
|                  | 2         | 4               | 61.2 (35.9)  | - 55.1 (15.8)               | 0.102   |
|                  | 3         | 2               | 31.0 (10.7)  | 6.8 (13.3)                  | 0.868   |
|                  | 4         | 2               | 39.9 (5.8)   | 16.3 (6.6)                  | 0.168   |
|                  | 5         | 2               | 53.3 (5.8)   | - 8.1 (4.2)                 | 0.296   |
|                  | $\bar{X}$ | —               | —  | - 10.0 (15.8)               | —       |
| Gizzard shad     | 1         | 4               | 21.3 (17.5)  | - 145.8 (10.4)              | 0.007   |
|                  | 2         | 4               | 12.6 (12.0)  | 55.5 (18.9)                 | 0.187   |
|                  | 3         | 2               | 13.5 (13.5)  | - 78.7 (3.7)                | 0.008   |
|                  | 4         | 2               | 7.2 (8.2)  | 15.9 (25.1)                 | 0.600   |
|                  | 5         | 2               | 4.5 (6.7)  | 168.7 (22.1)                | 0.030   |
|                  | 6         | 2               | 8.6 (8.9)  | 109.8 (11.8)                | 0.043   |
|                  | $\bar{X}$ | —               | —  | 20.9 (47.8)                 | —       |

**Table 5.** Initial concentration and net percent change in Chlorophyta populations in laboratory grazing trials with eastern elliptio mussels, fathead minnows, and gizzard shad. (%) is the percent of total phytoplankton counts comprised of Chlorophyta cells. The p-values correspond to t-tests on the mean percent change in Chlorophyta between reference and stocked enclosures in each trial.  $\bar{X}$  is the overall average of grazing trial means for each species.

| Species          | Trial     | Stocked aquaria | $\bar{X}$ initial Chlorophyta cells/ml x 10 <sup>4</sup> (%) | Net % change $\bar{X}$ (SE) | p-value |
|------------------|-----------|-----------------|--|-----------------------------|---------|
| Eastern elliptio | 1         | 2               | 1.2 (0.3)  | - 89.7 (4.1)                | 0.027   |
|                  | 2         | 4               | 3.9 (2.0)  | - 9.9 (13.3)                | 0.706   |
|                  | 3         | 6               | 2.9 (1.9)  | - 13.6 (9.0)                | 0.443   |
|                  | 4         | 4               | 1.8 (0.4)  | - 110.1 (7.9)               | 0.001   |
|                  | $\bar{X}$ | —               | —  | - 55.8 (25.8)               | —       |
| Fathead minnow   | 1         | 4               | 9.4 (17.7)   | 19.7 (9.4)                  | 0.236   |
|                  | 2         | 4               | 26.7 (15.7)  | - 11.46 (8.71)              | 0.589   |
|                  | 3         | 2               | 1.6 (0.5)  | 17.3 (27.5)                 | 0.631   |
|                  | 4         | 2               | 10.7 (1.5)   | - 6.0 (10.5)                | 0.866   |
|                  | 5         | 2               | 11.4 (1.2)   | - 19.7 (13.0)               | 0.807   |
|                  | $\bar{X}$ | —               | —  | - 0.1 (7.9)                 | —       |
| Gizzard shad     | 1         | 4               | —  | —                           | —       |
|                  | 2         | 4               | —  | —                           | —       |
|                  | 3         | 2               | 3.4 (3.4)  | 17.5 (22.9)                 | 0.584   |
|                  | 4         | 2               | 3.2 (3.7)  | 14.5 (4.9)                  | 0.447   |
|                  | 5         | 2               | 3.2 (4.8)  | 18.4 (0.0)                  | 0.227   |
|                  | 6         | 2               | 3.1 (3.2)  | 0.6 (9.9)                   | 0.973   |
|                  | $\bar{X}$ | —               | —  | 12.7 (4.2)                  | —       |

trials, the overall mean reduction of 55.4 % (range: - 110.0 to - 9.9 %) was nearly as great as that for *Anacystis* (63.6 %). During experiments with fathead minnows, responses ranged from a 19.7 % reduction to a 19.7 % increase, but none of the changes were significantly different from those in reference aquaria. The overall mean decline was 0.04 %. Gizzard shad had no significant effect on green algae levels, with an overall mean increase of 12.7 % (range: 0.5 to 18.4).

In summary, mussels were more effective than both fathead minnows and gizzard shad at reducing suspended solids, *Anacystis*, *Oscillatoria*, and Chlorophyta concentrations (Figure 1). Mussels were least effective at reducing filamentous algae concentrations, but the overall mean reduction still exceeded that of either fish species. In contrast to mussels, fathead minnows and gizzard shad had either no effect or enhanced suspended solid levels. While population levels of each algal taxon were suppressed by fish during some trials, responses were not consistent.

## Field Assessment of Grazing

Mussels significantly reduced suspended solids by 121.0 % relative to reference enclosures in the grazing experiment conducted at the New Castle STP (Table 6). Both fish species significantly increased suspended solids relative to reference enclosures. The net percent change attributable to fathead minnows and Israeli carp was 353.5 and 209.6 %, respectively. The mean changes attributable to the candidate species were significantly different from each other. Mean suspended solids reduction by mussels in the field was 1.6 times greater than the maximum suspended solids reduction of 75.8 %

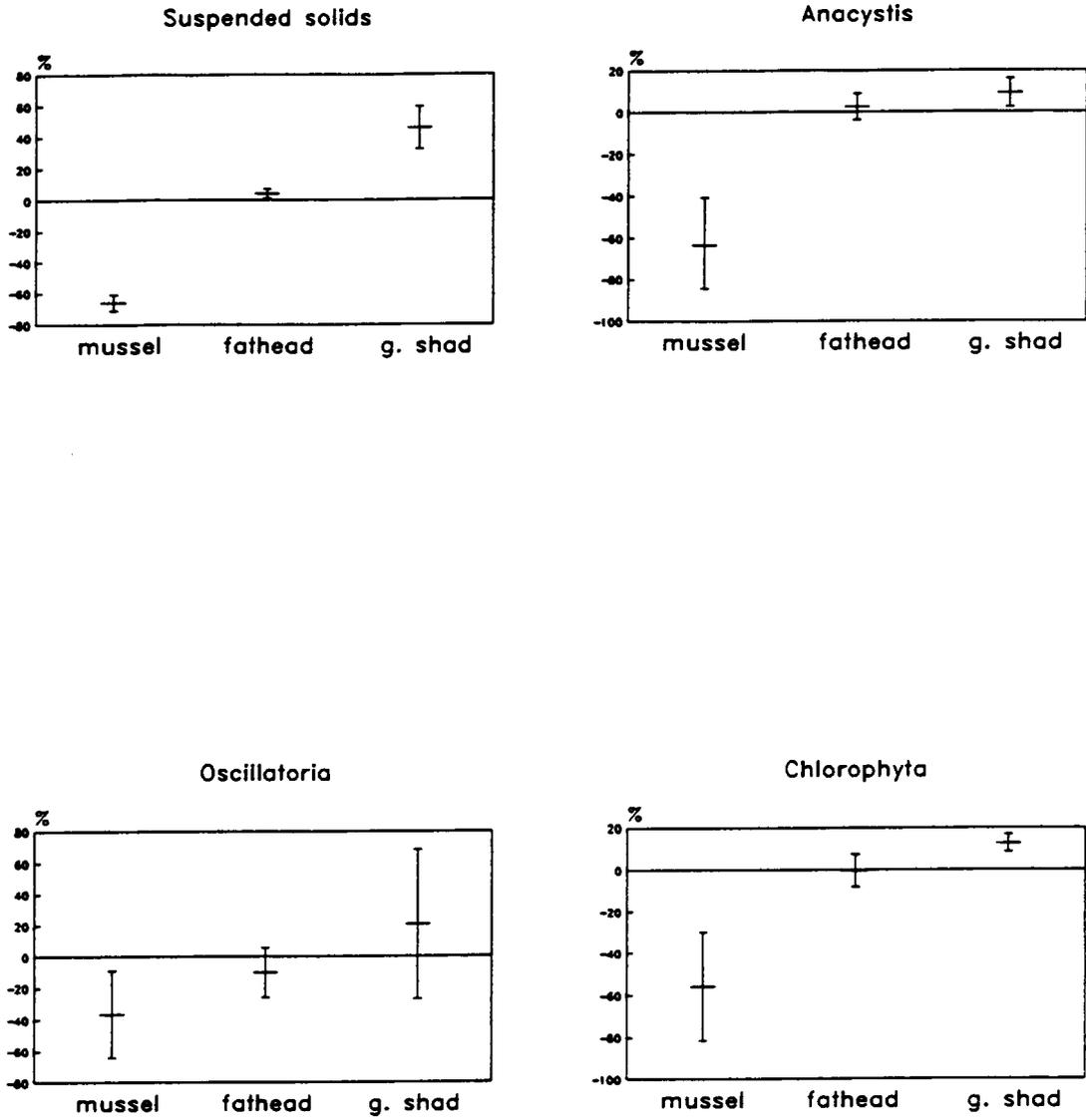


Figure 1. Mean ( $\pm$  1 SE) changes in suspended solids and algae attributable to mussels and fish in laboratory grazing trials.

**Table 6.** Mean change in suspended solids, *Acanycystis*, and Chlorophyta concentrations attributable to fish and mussels under field conditions. The three means for both parameters are significantly different ( $P > 0.05$ ).

| Parameter                                      | $\bar{X}$ initial concentration | $\bar{X}$ net % change (SE) |                |               |
|--|---------------------------------|-----------------------------|----------------|---------------|
|  |                                 | Eastern elliptio            | Fathead minnow | Israeli carp  |
| Suspended solids (mg/L)                        | 15.0                            | - 121.0 (4.8)               | 353.5 (41.7)   | 209.6 (15.7)  |
| <i>Chlorella</i> (cells/ml x 10 <sup>4</sup> ) | 17.8                            | - 307.1 (15.7)              | 19.5 (15.4)    | - 84.1 (36.2) |

achieved during laboratory trials. The 353.5 % increase by fathead minnows in the field experiment exceeded any increases during laboratory trials with that species.

Both mussels and Israeli carp significantly reduced *Chlorella* populations relative to reference enclosures, however the 307.1 % net reduction by mussels was 3.7 times greater than the 84.1 % reduction attributable to carp. The 19.4 % increase attributable to fathead minnows was not significantly different from reference enclosures. The mean net changes for the 3 species were significantly different from each other. The marked reduction of *Chlorella* populations by mussels exceeded maximum reductions of algae by mussels in the laboratory by 2 to 3 times.

The wastewater suspension from Pond 3 at the New Castle STP was very different from those in laboratory experiments. The mean initial suspended solids level of 15.0 mg/L was 5 to 10 times lower than those present in laboratory trials. The algae community was a monoculture of *Chlorella* (Chlorophyta), which are small (6 to 12  $\mu\text{m}$ ), spherical cells representative of stabilization ponds (Dinges 1982). The mean initial *Chlorella* concentration of 178,000 cells/ml was an order of magnitude lower than total algal concentrations present in laboratory grazing trials.

The relatively low suspended solid and algae concentrations in the field were a consequence of light waste loading in the New Castle STP, which was started up only a few months prior to the field experiment. In contrast, wastewater suspensions in laboratory experiments came from a heavily loaded, primary treatment pond that supported dense concentrations of blue-green algae.

## Density-Specific Effects on Mussel Filtration

Initial levels of suspended solids had no significant effect on the percent reduction of suspended solids by mussels under laboratory conditions. Low, medium, and high densities of mussels significantly reduced suspended solids relative to reference aquaria. The percent reductions attributable to mussels in 7 liters of wastewater were 37.4, 51.4, and 57.9 % at densities of 4, 8, and 12 mussels, respectively (Table 7). Reduction at high density was significantly greater than at low density, but was not significantly different from medium density. Reduction of suspended solids at medium mussel density was not significantly different from the low density treatment.

Initial concentrations of *Anacystis* had a significant effect on percent reduction of *Anacystis*, and analysis of covariance was conducted. At low, medium, and high densities, the net reductions of *Anacystis* were 28.0, 39.5, and 53.8 % (least square means adjusted by initial *Anacystis* concentrations). All three mussel densities significantly reduced *Anacystis* concentrations relative to reference aquaria. Mussels at high density significantly reduced *Anacystis* concentrations to a greater extent than both low and medium density treatments.

Initial concentrations of Chlorophyta had no significant effect on reduction of Chlorophyta cells by mussels. Low, medium, and high densities of mussels significantly reduced Chlorophyta concentrations relative to reference aquaria. The percent reductions attributable to low, medium, and high densities of mussels were 28.4, 40.7, and 54.4 %, respectively. Reduction at high density was significantly greater than at low density, but was not significantly different from the medium density treatment. Net reductions at low and medium mussel densities were not significantly different.

**Table 7.** Mean net reductions in suspended solids, *Anacystis*, and Chlorophyta concentrations by 3 densities of mussels under laboratory conditions. Low, medium, and high densities = 4, 8, and 12 mussels/7 L, respectively. SE is one standard error of the mean. N = 6 replicates at each density level. Means underscored by the same line are not significantly different ( $P > 0.05$ ).

| Parameter                                      | $\bar{X}$ initial concentration | $\bar{X}$ net % reduction (SE) |                    |             |
|--|---------------------------------|--------------------------------|--------------------|-------------|
|  |                                 | low                            | medium             | high        |
| Suspended solids (mg/L)                        | 111.6                           | <u>37.4 (5.7)</u>              | <u>51.4 (3.3)</u>  | 57.9 (5.5)  |
| <i>Anacystis</i> (cells/ml x 10 <sup>4</sup> ) | 85.6                            | <u>28.0* (4.4)</u>             | <u>39.5* (4.4)</u> | 53.8* (4.4) |
| Chlorophyta (cells/ml x 10 <sup>4</sup> )      | 22.6                            | <u>28.4 (7.9)</u>              | <u>40.7 (8.5)</u>  | 54.4 (7.3)  |

\* least square means adjusted by the initial concentration of *Anacystis*

Net reductions of suspended solids under field conditions averaged 47.6, 48.8, and 60.5 % at densities of 4, 8, and 12 mussels/10.5 L, respectively (Table 8). Mussels at all three densities significantly reduced suspended solids relative to reference enclosures. However, the three mean net reductions attributable to mussels were not significantly different from each other.

Low, medium, and high mussel densities removed 57.1, 82.5, and 92.0 % of initial *Chlorella* concentrations relative to reference enclosures. Reductions at both medium and high densities were significantly greater than at the low density level, but reductions at medium and high densities were not significantly different. Mussels at all three density levels significantly reduced *Chlorella* concentrations relative to reference enclosures.

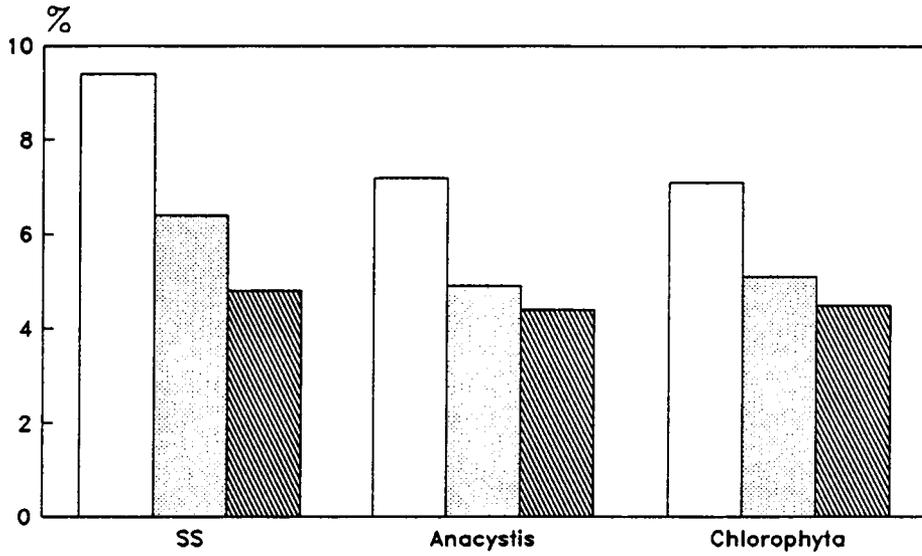
Mussels removed a greater proportion of initial suspended solid and phytoplankton concentrations in the field than under laboratory conditions. Reductions of suspended solids and algae in the field were greater than they appeared relative to reductions in the laboratory because the volume of wastewater in field enclosures (10.5 L) was 50.0 % greater than in laboratory enclosures (7.0 L). In addition to differences in wastewater volumes, initial field concentrations of suspended solids and phytoplankton were 7 to 8 times lower than initial laboratory levels (Tables 7 and 8). The phytoplankton assemblage also differed between laboratory and field conditions. The algae in laboratory suspensions were mainly comprised of relatively large cells (primarily *Anacystis* and *Scenedesmus*), whereas a monoculture of small cells (*Chlorella*) predominated in field suspensions.

In summary, greater numbers of mussels removed greater proportions of suspended solids and algae cells. However, the percent removed per mussel (e.g. mean reductions divided by the number of mussels at each treatment level) declined as mussel density increased under both laboratory and field conditions (Figure 2).

**Table 8.** Mean net reductions in suspended solids and algae concentrations at three densities of mussels under field conditions. Low, medium, and high densities = 4, 8, and 12 mussels/10.5 L, respectively. Means underscored by the same line are not significantly different ( $P > 0.05$ ).

| Parameter                                      | $\bar{X}$ initial concentration | $\bar{X}$ net % reduction (SE) |             |            |
|--|---------------------------------|--------------------------------|-------------|------------|
|  |                                 | low                            | medium      | high       |
| Suspended solids (mg/L)                        | 17.5                            | 47.6 (8.2)                     | 48.8 (11.5) | 60.5 (8.4) |
| <i>Chlorella</i> (cells/ml x 10 <sup>4</sup> ) | 14.4                            | 57.1 (4.4)                     | 82.5 (3.3)  | 92.0 (1.9) |

## Laboratory



## Field

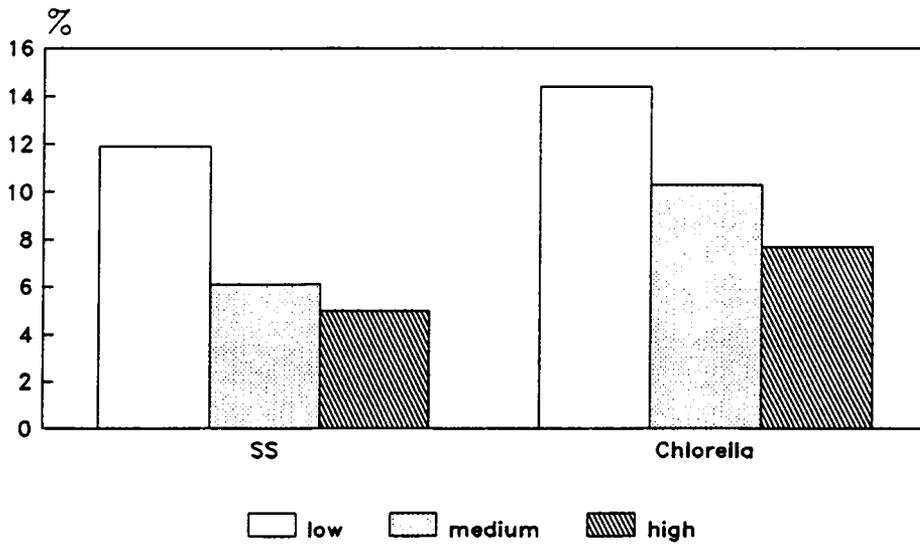


Figure 2. Density-specific reduction (per mussel) of suspended solids and algae under laboratory and field conditions.

## Time-Specific Effects on Mussel Filtration

Mussels reduced suspended solid levels by a total of 36 mg/L in 24 hours, or 66.1 %, compared to a decline of 10.3 mg/L, or 18.2 % in reference aquaria (Figure 3 and Table 9). The net suspended solids reduction (treatment levels minus reference levels) by mussels was 25.7 mg/L, or 47.2 %. More than 50 % of the total reduction occurred during the first 8 hours. The decline in suspended solids due to mussel filtration was significantly greater than the decline in control enclosures. The following regression equation best described the decline in suspended solids over time in laboratory aquaria:

$$\log_e SS = -0.042 T + 3.912 \quad (R^2 = 0.91)$$

where SS is suspended solids (mg/L) and T is time in hours.

Mussels reduced *Anacystis* concentrations by 66,011 colonies/ml, or 67.8 %, relative to a decline of 24,004 colonies/ml or 22.1 % in control aquaria (Table 10 and Figure 3). The net reduction of *Anacystis* by mussels was 42,007 colonies/ml, or 45.7 % in 24 hours. Nearly 50 % of the total *Anacystis* reduction occurred during the first 4 hours. *Anacystis* population reduction by mussels was significantly greater than the decline in reference aquaria. The following equation best described the decline in *Anacystis* concentrations over time in laboratory aquaria:

$$\log_e C = -0.043 T + 11.332 \quad (R^2 = 0.83)$$

where C is concentration of *Anacystis* (colonies/ml) and T is time in hours.

Mussels reduced *Scenedesmus* population levels by a total of 66,363 cells/ml, or 77.4 % (Table 11 and Figure 3). Concentrations in reference aquaria declined by 20,474 cells/ml, or 22.6 %. The reduction of *Scenedesmus* populations attributable to mussels was 42,889 cells/ml, or 54.8 % in 24 hours. Over 50 % of the *Scenedesmus* cell reduction occurred within the first 4 hours of the experiment. Mussel reduction of *Scenedesmus*

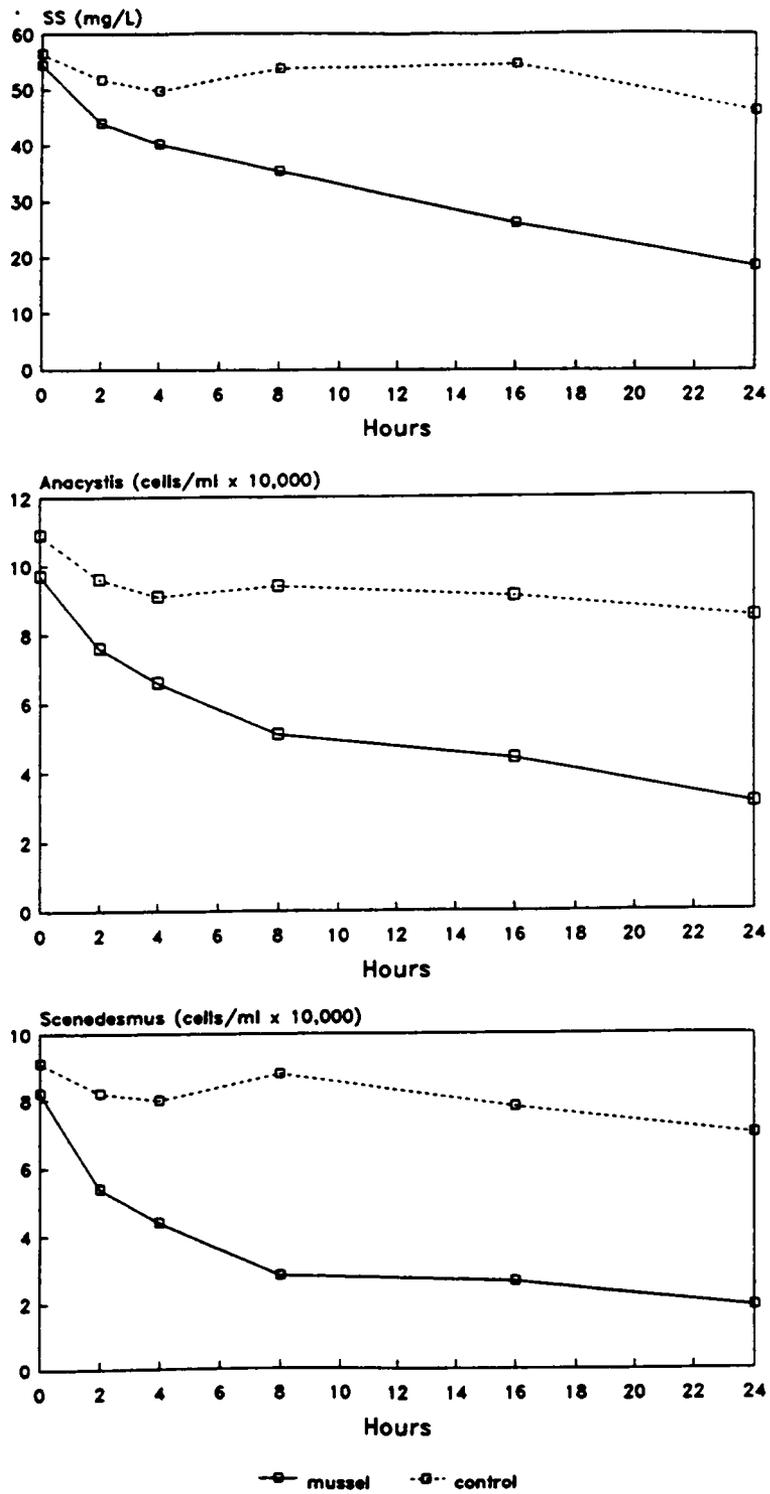


Figure 3. Mean suspended solids and algae concentrations in mussel (solid line) and reference (broken line) aquaria in 24 hours.

**Table 9.** Mean levels of suspended solids in stocked (8 mussels/7 L) and reference aquaria over 24 hours. Sample size was 4 stocked and 4 reference aquaria.

| Time<br>(h)   | Suspended Solids      |           |                       |           |
|---------------|-----------------------|-----------|-----------------------|-----------|
|               | Mussel                |           | Reference             |           |
|               | $\bar{X}$ mg/L (1 SE) | % decline | $\bar{X}$ mg/L (1 SE) | % decline |
| 0             | 54.5 (2.2)            | 0.0       | 56.5 (3.2)            | 0.0       |
| 2             | 44.0 (1.8)            | 19.3      | 51.7 (1.0)            | 8.5       |
| 4             | 40.3 (0.8)            | 26.1      | 49.8 (2.1)            | 11.9      |
| 8             | 35.5 (2.7)            | 34.9      | 53.8 (3.4)            | 4.8       |
| 16            | 26.2 (1.7)            | 51.9      | 54.6 (1.9)            | 3.4       |
| 24            | 18.5 (1.8)            | 66.1      | 46.2 (2.1)            | 18.23     |
| Total decline | 36.0                  | 66.1      | 10.3                  | 18.2      |
| Net reduction | 25.7                  | 47.2      | —                     | —         |

**Table 10.** Mean concentrations of *Anacystis* in treatment (8 mussels/7 L) and reference aquaria over 24 hours. Sample size was 4 treatment and 4 reference aquaria.

| Time<br>(h)   | <i>Anacystis</i>             |           |                              |           |
|---------------|------------------------------|-----------|------------------------------|-----------|
|               | Mussel                       |           | Reference                    |           |
|               | $\bar{X}$ colonies/ml (1 SE) | % decline | $\bar{X}$ colonies/ml (1 SE) | % decline |
| 0             | 97,428 (2,446)               | 0.0       | 108,547 (3,752)              | 0.0       |
| 2             | 76,248 (2,868)               | 21.7      | 96,369 (5,172)               | 11.2      |
| 4             | 66,011 (5,450)               | 32.2      | 91,251 (4,290)               | 15.9      |
| 8             | 51,185 (3,894)               | 47.5      | 93,545 (4,096)               | 13.8      |
| 16            | 43,772 (5,000)               | 55.1      | 90,721 (2,342)               | 16.4      |
| 24            | 31,417 (3,708)               | 67.8      | 84,544 (6,998)               | 22.1      |
| Total decline | 66,011                       | 67.8      | 24,004                       | 22.1      |
| Net reduction | 42,007                       | 45.7      | —                            | —         |

**Table 11.** Mean concentrations of *Scenedesmus* in treatment (8 mussels/7 L) and reference aquaria over 24 hours. Sample size was 4 treatment and 4 reference aquaria.

| Time<br>(h)          | <i>Scenedesmus</i>        |             |                           |             |
|----------------------|---------------------------|-------------|---------------------------|-------------|
|                      | Mussel                    |             | Reference                 |             |
|                      | $\bar{X}$ cells/ml (1 SE) | % decline   | $\bar{X}$ cells/ml (1 SE) | % decline   |
| 0                    | 81,896 (2,824)            | 0.0         | 90,544 (11,563)           | 0.0         |
| 2                    | 54,009 (4,589)            | 34.1        | 81,543 (8,501)            | 9.5         |
| 4                    | 43,772 (6,722)            | 46.6        | 80,131 (9,032)            | 11.5        |
| 8                    | 28,240 (4,035)            | 65.5        | 87,897 (11,608)           | 2.9         |
| 16                   | 26,475 (4,328)            | 67.7        | 78,366 (5,469)            | 13.4        |
| 24                   | 18,532 (3,806)            | 77.4        | 70,071 (7,612)            | 22.6        |
| <b>Total decline</b> | <b>63,363</b>             | <b>77.4</b> | <b>20,474</b>             | <b>22.6</b> |
| <b>Net reduction</b> | <b>42,889</b>             | <b>54.8</b> | —                         | —           |

populations was significantly greater than the decline in control enclosures. The decline in *Scenedesmus* concentrations over time was best characterized by the following regression equation.

$$\log_e C = -0.056 T + 10.998 \quad (R^2 = 0.67)$$

where C is concentration of *Scenedesmus* (cells/ml) and T is time in hours.

The mean filtration rate per mussel in the first 2 hours was 134.1 ml/h (s = 39.3 ml/h), based on the decline in *Scenedesmus* concentrations. Mean filtration rate based on the decline in *Anacystis* concentrations was 54.4 ml/h (s = 36.7 ml/h). Filtration rate based on the total cell counts was intermediate (87.7 ml/h) between the rates on *Scenedesmus* and *Anacystis*.

## Survival and Growth of Mussels

From mid-July to October, 1988 (approximately 3 months), survival of mussels at the New Castle STP was relatively high (90 - 95 %). Large individuals (90 to 117 mm) did not appear to lose body weight from late summer to mid-fall, 1988. On August 19, a subsample of 32 individuals with a mean shell length of 103.2 mm (SD = 6.6 mm) had a mean weight of 120.3 g (SD = 21.0 g). On October 20, a subsample of 17 similarly-sized individuals ( $\bar{X}$  = 102.5 mm, SD = 4.6 mm) had a mean body weight of 120.2 g (SD = 19.6 g).

From October 20, 1988 to June 28, 1989, the mean percent survival among 92 mussels was 68.5 %. Mortality did not appear to be related to mussel size. Weight loss occurred in all surviving individuals. Mean individual weight loss (total wet weight) for 60 mussels was - 15.7 % (SD = 3.2 %) and ranged from - 5.2 to -23.9 %.

# Discussion

## Grazing Effectiveness of Fish

When judged solely on the basis of suspended solids removal, the three fish species showed little potential for use in biological wastewater treatment, particularly when compared to eastern elliptio mussels. Complex trophic interactions between fish and phytoplankton, and the variable food habits of each fish species resulted in low and inconsistent grazing effects on suspended solids and phytoplankton in wastewater suspensions.

The fathead minnow has received attention as a biological wastewater clarifier because of its herbivorous food habits (Coyle 1930; Isaak 1961) and relatively high tolerance to unstable environmental conditions (Becker 1983). However, this study and others indicate that fathead minnows have either little effect or enhance suspended solids (Chambers 1978; Hall and Shelton 1983). At an Oklahoma STP, fathead minnows cultured in secondary-treated effluent affected a small (31 %) and statistically insignificant reduction in suspended solids relative to control raceways without fish (Hall and Shelton 1983). During a second experimental phase, fathead minnows increased suspended solids by 64 % when cultured in primary-treated wastewater. The results were attributed to differences in the suitability of suspended solids as food for fathead minnows, depending on the level of treatment the water entering the raceways had received.

Although suspended solids were not reduced by fathead minnows, they consumed some algae as evidenced by changes in the relative abundance of algal taxa and the presence of algae in their digestive tracts. *Anacystis* populations generally were enhanced, whereas *Oscillatoria* and Chlorophyta populations were suppressed during some experiments. However, fathead minnow grazing had little effect on total enumerated algae because filamentous and green algae comprised only a small proportion of the phytoplankton assemblage. Grazing by fathead minnows on green algae was not only minimal, but inconsistent because *Chlorella* concentrations were not reduced in the field experiment.

Isaak (1961) found *Oscillatoria* and green algae cells in the digestive tracts of fathead minnows. Pflieger (1975) considered the fathead minnow a herbivore; however, an omnivorous diet was indicated by the presence of organic detritus, sand, mud, insects, zooplankton, worms, and algae in stomach contents (Coyle 1930; Dobie et al. 1956). Zooplankton were the primary food of fathead minnows in North Dakota prairie pothole lakes (Held and Peterka 1974). Where algae was included in the diet, it was typically grazed from the sediments or the leaves of macrophytes (e.g., settled algae and periphyton), rather than from open water grazing (Isaak 1961). Since animal prey (e.g., zooplankton and insect larvae) were not abundant during enclosure experiments, some algal grazing was consistent with this species' opportunistic feeding habits.

The trophic habits of gizzard shad indicated its potential applicability to wastewater clarification (Pflieger 1975; Becker 1983), although no direct assessments have been conducted. Gizzard shad exceeding a length of 25 to 30 mm possess finely spaced gill rakers that enable them to filter-feed on suspended matter (Cramer and Marzolf 1970; Drenner et al. 1982). However, gizzard shad are not obligate filter-feeders on open water zooplankton, phytoplankton, and suspended detritus. Benthic sediments, molluscs, insect larvae, and organic detritus are obtained by foraging in bottom

sediments, and attached algae and detritus are grazed from submerged objects (Kutkuhn 1958; Bodola 1965; Pierce et al. 1981). Food habits and foraging behavior depend on the distribution and availability of potential prey items (Bodola 1965; Mundahl and Wissing 1988).

Particle retention by filter-feeding gizzard shad is largely a function of particle size (Mummert and Drenner 1986), although ingestion of zooplankton by gizzard shad depends upon both zooplankton size and its ability to escape predation by swimming (Drenner et al. 1978). Minimum particle size retained by gill rakers increases as gizzard shad grow; however, 100 % retention occurs only for particles 100  $\mu\text{m}$  or greater in size (Drenner et al. 1978; Mummert and Drenner 1986). Most wastewater phytoplankton are less than 100  $\mu\text{m}$  (Gloyna et al. 1976), and therefore gizzard shad grazing rates and particle retention efficiency should be relatively low, a prediction supported by this study. Gizzard shad grazing rates were insufficient to reduce suspended solids under laboratory conditions. In fact, gizzard shad enhanced suspended solids to a greater extent than did fathead minnows. Only small amounts of algae were found in the intestinal tracts of gizzard shad following experiments. Nevertheless, *Anacystis* and *Oscillatoria* populations declined in a few experiments. Some *Anacystis* colonies and *Oscillatoria* filaments exceeded 100  $\mu\text{m}$  and therefore were susceptible to grazing by gizzard shad. All Chlorophyta cells were less than 30  $\mu\text{m}$  and their populations were generally enhanced. Gizzard shad were not tested under field conditions in which wastewater suspensions contained a monoculture of very small (6 to 12  $\mu\text{m}$ ) *Chlorella* cells. However, it is unlikely that these small cells would be grazed to any significant extent by gizzard shad.

This study and others indicate that gizzard shad are ineffective grazers when phytoplankton communities are dominated by small algae (Drenner et al. 1984; Drenner et al. 1986; Threlkeld 1987). In large enclosure experiments, gizzard shad enhanced populations of small (less than 100  $\mu\text{m}$ ) algae (Drenner et al. 1984). Under similar con-

ditions, gizzard shad had no consistent effect on filamentous blue-green algae, and enhanced colonial blue-green and Chlorophyta populations (Drenner et al. 1986). Threlkeld (1987) concluded that gizzard shad affected no significant changes in algal abundance.

The unstable nature of many wastewater ponds as well as the need to monitor and manage fish populations for waste treatment make gizzard shad a poor candidate. Gizzard shad are fragile, difficult to transport and handle, and susceptible to stress-induced mortality (Bodola 1965; Reuter and Herdendorf 1974). High mortality, particularly of young fish, has been related to rapid changes in water temperature and prolonged periods of low water temperature, resulting in wide temporal variability in recruitment (Williamson and Nelson 1985; Willis 1987).

As with the fathead minnow and gizzard shad, common carp are omnivores and opportunistic feeders. While Chlorophyta and Chrysophyta cells comprised the majority of the carp's diet in Elephant Butte Lake (Jester 1974), other studies have shown that benthic invertebrates were the predominant prey and plant material generally comprised a small proportion of total stomach contents (Pearse 1919, 1921; Moen 1953). Carp forage in bottom sediments, often uprooting vegetation and increasing turbidity (Becker 1983). These activities have contributed to a perception of carp as a nuisance species (Pflieger 1975; Becker 1983).

The Israeli carp is a genetic variant of the common carp. In this study, Israeli carp significantly enhanced suspended solids by 210 % in field enclosures. The carp's benthic habits in a Texas stabilization pond increased suspended solids from 25 to 90 mg/L over a 2 year period (Orgeron 1976). Carp activity resuspended inorganic sediments and increased total suspended solids even though volatile suspended solids (mainly algae) declined.

Israeli carp reduced *Chlorella* concentrations by 84 % in field enclosures. Jester (1974) showed that carp consumed significant amounts of green algae when preferred prey are scarce. Carp presumably graze algae from the bottom and from submerged objects. As carp consume benthic prey or detritus, they may passively consume some suspended algae as well. The likelihood of passive algae consumption would increase within the relatively small enclosures used in this study, perhaps accounting for the decline of *Chlorella*. Additional experiments are needed to determine whether carp will significantly and consistently graze on wastewater phytoplankton suspensions.

The potential of fish to control suspended solids in sewage lagoons depends on their ability to suppress suspended nanoplankton. While low levels of suspended algae are required to meet effluent suspended solids standards, attached algae may be desirable in a wastewater stabilization pond. Periphyton does not contribute suspended solids. Additionally, attached algae may promote the maintenance of aerobic conditions, serve as a nutrient sink, and actively compete with phytoplankton for nutrients and trace elements. Stocking herbivorous fish that selectively graze on periphyton rather than phytoplankton may be counterproductive to the control of suspended solids in sewage lagoons. Consumption of periphyton has been documented for fathead minnows (Isaak 1961), gizzard shad (Bodola 1965), and carp (Moen 1953).

Some aspects of algal morphology and biology influence their susceptibility to grazing by aquatic herbivores. The characteristically small size of wastewater algae provides a refuge from grazing by most fish species (Drenner et al. 1984). In natural ecosystems, small (less than 30  $\mu\text{m}$ ) phytoplankton are grazed primarily by invertebrate filter-feeders that discriminate among algae on the basis of size, shape, abundance, and palatability (Porter 1977). Fish may select among potential algal prey on the basis of these characteristics as well. For example, carp use olfaction and taste in prey selection (Lagler et al. 1962; Stein et al. 1975). Noxious chemical defenses impart grazing resist-

ance to many colonial and filamentous blue-green algae (Porter 1973a; Reynolds 1984). Toxic effects have been associated with dense concentrations of *Microcystis* (Porter 1977). *Microcystis* is very similar to *Anacystis*, the predominant blue-green taxon in my laboratory grazing trials. It is conceivable that *Anacystis* unpalatability may have contributed to low grazing rates by fish.

Thick cell walls or gelatinous sheaths enable some phytoplankton species to survive predation by invertebrate grazers (Porter 1973b; Reynolds 1984) and planktivorous fish (Velasquez 1939). Algal genera that survive gut passage through gizzard shad include *Microcystis*, *Oscillatoria*, *Scenedesmus*, and *Ankistrodesmus* (Velasquez 1939). These taxa include some of the predominant algae in this study. Fathead minnows were the only fish species that contained significant amounts of algae in their guts. *Anacystis*, *Oscillatoria*, and Chlorophyta cells removed from fathead minnow gastrointestinal tracts produced viable populations when cultured in fertilized medium, indicating incomplete digestion. For some algae, nutrients absorbed during gut passage can stimulate population growth (Porter 1977). These effects undoubtedly accounted for some of the variation in phytoplankton responses to fish.

Interrelationships between fish and phytoplankton are complex. For example, planktivorous fish consume zooplankton as well as phytoplankton. Filter-feeding zooplankton themselves can significantly reduce algal abundance (Lampert et al. 1986; Porter 1977). Selective algae consumption and nutrient release by invertebrate herbivores dramatically affect the species composition of phytoplankton communities (Porter 1973a; Reynolds 1984). The effects of fish predation on zooplankton can cascade to lower trophic levels, resulting in increased nanoplankton abundance (Brooks 1968; Andersson et al. 1978; Lynch and Shapiro 1982; Carpenter et al. 1985; Hurlburt et al. 1986). Fish species that have enhanced phytoplankton by trophic cascading include gizzard shad (Drenner et al. 1984) and silver carp (Smith 1985).

In my study, zooplankton were scarce, and changes in algae concentrations were not attributed to direct grazing by zooplankton or to fish predation on zooplankton. However, cascading effects of fish have important implications for aquacultural approaches to waste treatment. Stocking planktivorous fish in a stabilization pond could increase algae populations and suspended solids if the pond already supports dense populations of grazing zooplankton. Maintaining fish in one section or the upper water column of the pond would provide invertebrate grazers a refuge from fish predation and maximize zooplankton grazing on nanoplankton. Such an approach has been effective in tanks experiments that integrated the total grazing influence of silver carp and zooplankton on algae (Smith 1985).

In addition to the direct effects of predation, fish can affect algae communities indirectly by recycling nutrients. Nutrient-mediated enhancement of phytoplankton can occur through fish egestion and excretion, mortality and decomposition, and through benthic habits that resuspend detritus and nutrients (Threlkeld 1987).

Fish defecation and excretion provide algae with additional nutrients (Meyer et al. 1983; Robison and Bailey 1981). Phytoplankton enhancement has been linked to nutrient regeneration by gizzard shad (Drenner et al. 1986) and carp (Lamara 1975). The nutrient regeneration rate should be directly related to fish biomass. Of the species evaluated in this study, the fathead minnow was tested over the widest range of stocking densities. The greatest enhancement of *Anacystis* and suspended solids occurred when stocking density and presumably excretion rates were high. While it is difficult to independently assess grazing and nutrient-mediated effects on phytoplankton abundance, it is likely that fish defecation and excretion increase phytoplankton and suspended solids in many grazing experiments (Threlkeld 1987).

Nutrient release from dead fish can enhance phytoplankton (Drenner et al. 1986; Durbin et al. 1979; Threlkeld 1987). Its importance is clearly related to fish decompos-

ition rates and experimental duration. Gizzard shad mortality enhanced both nutrients and phytoplankton concentrations within 30 days (Threlkeld 1987). In my study, fathead minnow and Israeli carp mortality was low (0 to 10 %), although gizzard shad mortality was generally higher (0 to 70 %). However, experimental duration was less than 3 days and fish carcasses were removed within 12 hours of death. Therefore, the influence of fish mortality on phytoplankton was probably small relative to the nutrient-mediated impacts of live fish.

Finally, fish can enhance phytoplankton by swimming and foraging in the sediments and transferring nutrients into the water column (Lamara 1975). Gizzard shad are facultative planktivores that forage in benthic regions (Bodola 1965). The relative importance of swimming to nutrient recycling in this study is uncertain due to the short experimental duration. Additionally, particle settling was minimized by constant aeration. However, it is conceivable that fish grazed some settled or attached algae from the sides and bottom of enclosures. Resuspension of this material following defecation could have increased suspended solids, altered nutrient availability, and facilitated shifts in the relative abundance of algal taxa.

In summary, fathead minnows, gizzard shad, and carp increased suspended solids, but had variable effects on algal populations in wastewater suspensions. Published literature indicates that the effects of herbivorous fish on suspended solids and algae are variable (Table 12). The Asiatic silver carp has shown the most promise in secondary waste treatment applications (Dinges 1982). However, populations of small algae (e.g., *Chlorella*) were increased by silver carp predation on zooplankton (Smith 1985; Burke et al. 1986; Leventer 1987). In addition to its consumption of zooplankton, silver carp contributed to eutrophication by cycling nutrients (Leventer 1987). Reduction of suspended solids and algae typically occurred when silver carp were stocked as part of a polyculture with other cyprinid or *Tilapia* species (Spataru et al. 1982; Henderson 1983;

**Table 12. Effect of fathead minnows, gizzard shad, Israeli carp, and silver carp on suspended solid and phytoplankton concentrations in various systems. Effects are increased levels (+), reduced levels (-), or no effect (0).**

| Species        | Suspended Solids | Phytoplankton   | Experimental System | Citation  |
|----------------|------------------|-----------------|---------------------|---|
| Fathead minnow | 0/+              | 0               | 39 L aquaria        | This study<br>Chambers 1978<br>Hall and Shelton 1983  |
|                | +                |                 | 1,000 L tank        |   |
|                | +/-              |                 | wastewater raceways |   |
| Gizzard shad   | 0/+              | +/-             | 39 L aquaria        | This study<br>Drenner et al. 1984<br>Drenner et al. 1986<br>Threlkeld 1987                                      |
|                |                  | +/-             | pond                |   |
|                |                  | +               | 7,000 L tank        |   |
|                |                  | 0               | 7,000 L tank        |   |
| Israeli carp   | +                | -               | 19 L buckets        | This study  |
| Silver carp    | -                | -               | wastewater lagoons  | Henderson 1983<br>Spataru et al. 1983<br>Wilson et al. 1984<br>Smith 1985<br>Burke et al. 1986<br>Leventer 1987 |
|                |                  | -               | fertilized pond     |   |
|                |                  | -               | eutrophic pond      |   |
|                |                  | +/-             | 1,000 L tank        |   |
|                | +/-              | +               | catfish ponds       |   |
|                | +/-              | fertilized pond |                     |   |

Leventer 1987). Only Wilson et al. (1984) reported significant algae reduction with a silver carp monoculture. Silver carp were not available from commercial sources during this study. Future studies regarding biological control of suspended solids should evaluate the grazing effectiveness of exotic and native fish species simultaneously in a variety of wastewater algae suspensions.

Direct herbivory by fish, trophic-cascade effects, and nutrient-mediated factors can influence phytoplankton abundance and species composition. When fish grazing rates on phytoplankton are low because of small algal size, unpalatability, or the availability of more preferred prey, indirect effects are likely to predominate, enhancing algae and suspended solids. Planktivorous fish alone may not provide a reliable control of suspended solids in wastewater stabilization ponds since phytoplankton are typically small. However, they may contribute to waste stabilization by providing a path for energy flow from low to higher trophic levels, particularly when stocked as part of an integrated biological approach to waste treatment that includes filter-feeding mussels and zooplankton.

## **Grazing Effectiveness of Mussels**

Freshwater mussels have received very little attention as wastewater clarifiers, despite the fact that they possess many appropriate characteristics. Mussels filter a wide size range of particles from 1 to 200  $\mu\text{m}$  (Jorgensen 1975). Food items are either consumed, digested, and expelled as feces, or rejected and expelled as pseudofeces. Both processes remove detritus (organic and inorganic), bacteria, and phytoplankton from suspension with resultant biodeposition in the sediments (Tenore and Dunston 1973),

an objective of wastewater treatment. Mussels are sedentary and more easily handled and transported than fish. They tolerate relatively wide variations in water chemistry, particularly pH and dissolved oxygen (Fuller 1974). Therefore, filter-feeding mussels appear amenable to wastewater treatment applications (Henderson and Wert 1976).

Surprisingly, only the introduced Asiatic clam, *Corbicula fluminea* has been investigated as a biological filter in rivers (Cohen et al. 1984; Lauritsen 1986b), in catfish rearing ponds (Buttner 1986a, 1986b), and in raceways or stabilization basins fed by wastewater effluent (Haines 1977; Dinges 1982). Other bivalve species may not have been considered because of their relatively low filtration rates and lower natural abundance (Lauritsen 1986). Nevertheless, *Corbicula* is generally considered a nuisance species, an economic liability, and a potential threat to native mussel species (Fuller 1974; McMahon 1983).

My results indicate that at least one freshwater mussel species, *Elliptio complanata*, showed a significant capacity to reduce both suspended solids and algae concentrations in wastewater. Proportional reduction of suspended solids relative to controls ranged from 58 to 76 % in laboratory grazing trials and reached 121 % in the field experiment.

Greater suspended solids reduction in the field experiment can be attributed to lower suspended solid and algae concentrations, and smaller algal size. Mean initial suspended solids in laboratory wastewater suspensions were 8 times greater than in field suspensions. Filtration rate declines at high particle concentrations for the eastern elliptio (Paterson 1984), and for many other bivalve species (Morton 1983; Winter 1978). The predominant algae in laboratory suspensions were large (50 to 250  $\mu\text{m}$ ) *Anacystis* colonies, whereas small, single-celled *Chlorella* predominated in field suspensions. Maximum filtration rate for the eastern elliptio occurs for particles 3 to 4  $\mu\text{m}$  in diameter and steadily declines for larger particles (Paterson 1986). Therefore, all algae taxa pres-

ent during both laboratory and field experiments in this were filtered at less than maximum rates. Pseudofeces production was high, particularly in laboratory experiments, reflecting a combination of high particle concentrations beyond usable limits, and large particle sizes. Additionally, dense concentrations of suboptimal food can interfere with mussels' particle-sorting mechanism and further depress filtration rate (Kirby-Smith 1972; Morton 1983). An impaired ability to sort different sized particles was indicated in this study by proportionally low reductions in green algae relative to *Anacystis* during laboratory experiments. Based on size alone, green algae should have been grazed at a higher rate than *Anacystis*.

Other factors may have contributed to differences in the effectiveness of *E. complanata* between laboratory and field conditions. Toxic metabolites produced by some algae (e.g. Cyanophyta species) can inhibit invertebrate filtration (Porter 1973a). The freshwater bivalve *Dreissena polymorpha* may select food particles on the basis of their chemical nature (Ten-Winkel and Davids 1982). The predominant algae during laboratory experiments (*Anacystis* and *Oscillatoria*) may have been unpalatable to *E. complanata*, thereby reducing filtration. Additionally, acclimation to food types and levels can influence invertebrate filtration (Mayzaud and Poulet 1978). Acclimation of *E. complanata* to algal taxa and concentrations in the New Castle wastewater may have enhanced mussel effectiveness during field experiments.

It is apparent that the wastewater suspensions used in laboratory and field experiments did not provide mussels with an optimal food resource (e.g. a homogeneous suspension of 3 to 4  $\mu\text{m}$  particles). Consequently, mussel filtration was probably less than maximum. Nevertheless, suspended solids and total algae concentrations were markedly reduced in all experiments and under a variety of conditions. In addition to direct consumption, pseudofeces production and biodeposition by mussels contributed to reductions in suspended solids. Pseudofeces formation was also observed for *Corbicula*

filtering wastewater (Dinges 1982). Clearly, mussel filtration, consumption, and pseudofeces production rates are affected by the quantity and quality of available food. Additionally, my study has shown that reductions in suspended solids and algae due to mussel filtration were affected by mussel density and the length of time over which suspended solids or phytoplankton removal was measured.

High densities of mussels facilitated greater particle removal under both laboratory and field conditions, although removal expressed on a "per mussel" basis declined with increasing numbers of mussels. This suggests that individual mussel filtration may be inhibited by increased mussel densities, beyond some optimal number. Alternatively, density-dependent filtration may confer some physiological advantage to members of a group. Paterson (1983) observed an 80 % decline in individual oxygen consumption rates of *E. complanata* as density in a respirometer increased from 10.4 to 124.7 mussels/m<sup>2</sup>. Paterson (1983) speculated that in addition to facilitating respiration and feeding, water pumping serves a sensory function by which mussels monitor their environment. Thus, individual metabolic costs may be lowered by the filtration activity of adjacent mussels in an aggregated distribution. Natural density of *E. complanata* has ranged from 0.03 adults/m<sup>2</sup> in Mirror Lake, New Hampshire (Strayer et al. 1981) to 60/m<sup>2</sup> in Lake Pocotopaug, Connecticut (Fisher and Tevesz 1984). In this study, density of *E. complanata* in laboratory enclosures was 31.4, 62.7, and 93.1 mussels/m<sup>2</sup> at low, medium, and high treatment levels, respectively. In field enclosures, low, medium, and high densities corresponded to 75.3, 150.6, and 226.0 mussels/m<sup>2</sup>. Therefore the range of densities used in this study encompassed densities over which individual rate processes (e.g., respiration) are negatively correlated with density.

Suspended solids, and algae concentrations declined logarithmically during 24 hours of mussel filtration in laboratory aquaria. Absolute reductions were greatest during the first 8 hours and subsequently declined, presumably due to lower particle

concentration. Mussel filtration is typically manifested as a logarithmic decline in particle concentration over time (Coughlan 1969). At a constant filtering rate, the absolute number of particles removed from suspension diminishes with declining particle concentration. In my study, the net percentage of algae and suspended solids removed in 24 hours were similar or greater in magnitude to the percentages removed in 60 hours. Therefore, absolute reductions of suspended solids and algae over lengthy time intervals can underestimate actual removal over shorter time periods. To illustrate this point, time-specific suspended solids removal rates are shown for three time intervals in Table 13. Removal rate per hour in the 0 to 2 hour interval (2.7 mg/L) was nearly 3 times greater than that based on the total suspended solids removed over 24 hours (1.1 mg/L). Therefore, the percent reductions attributed to mussels in the 60 hour laboratory and field grazing trials underestimate the filtration capacity of mussels over shorter time periods.

In heterogeneous suspensions, filtration rates represent minimal pumping rates because particle retention by the gills depends upon particle size and concentration, and retention efficiency is usually less than 100 % (Ward and Aiello 1973). In my study, filtration rate of *E. complanata* was 88 ml/h/mussel based upon the change in total cell counts (*Anacystis* and *Scenedesmus*). The rate of 134 ml/h on *Scenedesmus* cells was 2.5 times greater than the rate of 53 ml/h on *Anacystis*, presumably reflecting different retention efficiencies of mussels for the two algal taxa. A higher filtration rate on *Scenedesmus* was probably attributable to their small size, uniform shape, lack of a gelatinous sheath, or greater palatability relative to *Anacystis*. It is conceivable that retention efficiency would be even greater for smaller-sized algal cells (e.g., *Chlorella*) in a more homogeneous suspension, like those in Pond 3 at the New Castle STP. A greater retention efficiency coupled with lower initial cell concentrations in my field experiments probably would have yielded a higher filtration rate for *E. complanata*.

**Table 13.** Time-specific (per hour and per 24 hours) suspended solids removal rates by *E. complanata* (8 mussels/7 L) after 2, 8, and 24 hours of filtration. Total reductions (mg/L) by mussels are adjusted for changes in suspended solids in reference enclosures (e.g., net reductions calculated from Table 9). Removal rates per hour are total reductions divided by the corresponding elapsed time. Removal rates per 24 hours are extrapolated from the corresponding removal rates per hour.

| Elapsed Time<br>(h) | Total Reduction<br>(mg/L) | Suspended Solids Removal Rate |           |
|---------------------|---------------------------|-------------------------------|-----------|
|                     |                           | mg/L/h                        | mg/L/24 h |
| 2                   | 5.9                       | 2.9                           | 69.6      |
| 8                   | 16.4                      | 2.0                           | 48.0      |
| 24                  | 25.7                      | 1.1                           | 25.7      |

Filtration rate measurements are highly variable due to differences in experimental conditions and methods, and by expression of rates in various units (Tenore and Dunston 1973). High variability in filtration rate is often evident within a single study. For example, filtration rate of *Corbicula fluminea* (90 to 100 mg dry flesh weight) ranged from 100 to 1,200 ml/h (Lauritsen 1986). Similarly, Ward and Aiello (1973) reported that individual pumping rates of individual *Mytilus edulus* differed fivefold on consecutive days. Discontinuous filtration by freshwater bivalves undoubtedly contributes to variability in filtration rate measurements. Valves alternately open and close through a 24 hour period (Walz 1978; Benedens and Hinz 1980). However, this behavior may be a response to unnatural laboratory conditions, and active periods may be longer in nature (Jorgensen 1975). If this is true, mussel filtration in my study and others would underestimate the filtration capacity of undisturbed mussels in aquatic habitats.

Despite certain limitations, filtration rate is the most useful measure available to predict the impact of mussels in a wastewater pond. The filtration rate of *E. complanata* in my study can be evaluated by comparison with published literature values of freshwater mussel filtration rates (Table 14). Paterson (1986) found that filtration rate for this species was highly dependent on particle size, with a maximum filtration rate of 410 ml/h/g (dry tissue weight) for particles 3 to 4  $\mu\text{m}$  in diameter. Filtration rate for particles greater than 10  $\mu\text{m}$  was 12/g at particle concentrations greater than 13,000/ml, and 70/g at a particle concentration of 9,000/ml (Paterson 1984). In my study, algae were greater than 10  $\mu\text{m}$  and algal concentrations were 80,000 to 100,000 cells/ml. Filtration rates for *Scenedesmus* and *Anacystis* expressed on a weight-specific basis were approximately 53 and 21/g, respectively, which are well within the range of 12 to 70/g reported by Paterson (1984).

Previous studies of *Anodonta* and *Unio* species indicated a range of filtration rates from 150 ml/h (Gussman 1978) to 630 ml/h (Cameron and Paterson 1985). Filtration

Table 14. Filtration rates of freshwater mussels. Rates (ml/h) are expressed per mussel, except where indicated as per gram (dry weight).

| Superfamily  | Species                     | Filtration rate (ml/h)               | Particle concentration   | Shell length (mm)          | Citation                                  |            |
|--------------|-----------------------------|--------------------------------------|--|----------------------------|---|------------|
| Unionacea    | <i>Anodonta cataracta</i>   | 150 - 580<br>357 - 632               | 6 - 165/ml x 10 <sup>5</sup><br>11.2 - 12.5 mg C/L             | 93 - 118<br>70 - 80        | Gussman 1978<br>Paterson and Cameron 1985 |            |
|              | <i>A. piscinalis</i>        | 300                                  | not reported   | 62                         | Lewandowski and Stanczykowska 1975        |            |
|              | <i>Eliptio complanata</i>   | 12 - 70/g <sup>a</sup><br>75 - 410/g | 9,000 - 18,000/ml x 10 <sup>3</sup><br>275,700/ml <sup>b</sup> | 60 - 70<br>60 - 70         | Paterson 1984<br>Paterson 1986            |            |
|              | <i>Unio tunidus</i>         |                                      | 53   | 81,900/ml <sup>c</sup>     | 103                                       | This study |
|              |                             |                                      | 134  | 90,500/ml <sup>c</sup>     | 103                                       | This study |
|              |                             | 300                                  | not reported   | 50                         | Lewandowski and Stanczykowska 1975        |            |
| Dreissenacea | <i>Dreissena polymorpha</i> | 77                                   | 1,800,000/ml   | 25                         | Walz 1978                                 |            |
| Corbiculacea | <i>Corbicula fluminea</i>   | 347                                  | 3,400 - 17,700/ml  | 0.8 - 7.4 g <sup>e</sup>   | Buttner and Heidinger 1981                |            |
|              |                             | 11                                   | 20 - 130 mg/L/   | not reported               | Haines 1977                               |            |
|              |                             | 587 - 770<br>278 - 782               | not reported<br>0.33 - 2.67 mm <sup>3</sup> /L                 | 20.0 - 23.3<br>21.2 - 24.1 | Lauritsen 1986a<br>Lauritsen 1986b        |            |
|              | <i>Sphaerium striatinum</i> | 0.6 - 8.4                            | 2 - 64 mg/L <sup>f</sup>                                       | 1 g AFDW <sup>h</sup>      | Hornbach et al. 1984                      |            |

<sup>a</sup> filtration on particles  $\geq 10 \mu\text{m}$

<sup>b</sup> estimated from concentrations of different sized particles (Table 2 in Paterson 1986)

<sup>c</sup> *Anacystis* colonies/ml

<sup>d</sup> *Scenedesmus* cells/ml

<sup>e</sup> wet weight with shell

<sup>f</sup> measured as turbidity

<sup>g</sup> measured as concentration of polyvinyltoluene (PVT) beads

<sup>h</sup> AFDW is ash free dry weight

of *E. complanata* in my study was lower than this range. The unionid mussels represented in Table 14 are lake-dwelling species, including *E. complanata* studied by Paterson (1984; 1986). Experimental animals used in my study were adapted to riverine conditions, and the lack of flow during my experiments may have contributed to their comparatively low filtration rates. Additionally, mussel filtration declines with increasing particle size and concentration beyond usable levels (Winter 1978). In my study, mussels filtered a primary wastewater pond suspension with a high (54.5 mg/L) suspended solids load and a total cell concentration of 200,000 cells/ml. Since only live algae cells were enumerated, total particle concentration (algae, bacteria, organic and inorganic detritus) was presumably far greater. It is likely that natural lake water typically contains far less suspended particles than does wastewater from a primary sewage lagoon. Therefore, filtration rate as measured in my study was probably representative of unionid mussels filtering wastewater suspensions containing dense concentrations of relatively large algae. However, my rate estimates for *E. complanata* are lower than what could be expected if suspended solids were less than 55 mg/L and the wastewater suspension was comprised of small, readily consumable algae (e.g., *Chlorella*).

Filtration rate of *C. fluminea* generally ranged from about 280 to 780 ml/h (Lauritsen 1986b). These rates are comparable to those for unionid species, which are much larger in size. The relatively low filtration rate of 11 ml/h reported by Haines (1977) was attributed to low oxygen levels, high water temperatures, and poorly utilized food in the form of filamentous blue-green algae (Buttner and Heidinger 1981). Additionally, clams were maintained in wastewater, and the high turbidities probably reflected high particle concentrations and suspended solids levels. These are some of the same factors that presumably accounted for relatively low filtration rates of *E. complanata* in my study.

The deposit-feeding *Sphaerium striatinum* relies on suspended food particles to a far lesser extent than the other freshwater mussel species, which is reflected in the relatively low filtration rate for this species (Hornbach et al. 1981).

## Management Implications

Results of experiments with *E. complanata* indicate that freshwater mussels can control algal biomass and reduce suspended solids in secondary-treated wastewater. The grazing ability and physiological ecology of freshwater mussel species has received far less attention than that of marine bivalves (Jorgensen 1975). Before mussel culture can be successfully applied to wastewater treatment, our understanding of the performance of different species, sizes, and densities of mussels under differing wastewater system conditions must be enhanced. Some ecological characteristics that could affect different species' performance include habitat requirements (lentic vs. lotic, substratum requirements), physicochemical water parameter requirements (temperature, dissolved oxygen, pH), susceptibility to disease and parasites, life expectancy, reproductive requirements, and filtration capacity. Candidate species should also be relatively abundant over a wide distributional area so that adequate collections can be made without threatening existing wild populations. It may be necessary to initiate culture programs to provide an abundance of mussel seed stocks for waste treatment sites, and to maintain adequate stock numbers to achieve optimum wastewater clarification.

*E. complanata* inhabits both lotic and lentic environments with a relatively firm and coarse substratum (Matteson 1948; Ayers 1984). Many freshwater mussel species avoid fine mud and shifting sand bottoms (Fuller 1974), although some species (e.g., *Anodonta*

*grandis*) are morphologically adapted to inhabit environments with soft substratum (Ghent et al. 1978). Specimens of *E. complanata* used in this study were from a riverine population. It is not known to what extent this origin influenced its filtration and survival in my study; however, other unionid species (e.g., *Anodonta* species) may be better adapted to filter and survive in wastewater lagoons. In my experiments, mussels rested on one valve on the bottom of enclosures without substratum, and were not in their natural life position (Fisher and Tevesz 1976). One can speculate that filtration might be enhanced by placing mussels in a natural position in their preferred substratum, although no literature exists to support this speculation. Conversely, orientation and substratum preference may not be significant if an adequate food supply is present and environmental conditions are suitable.

Tolerance of freshwater mussels to fluctuating water temperatures differs among species (Fuller 1974). *E. complanata* in a Michigan lake move to cooler waters when water temperature exceeds 24 °C (Matteson 1948). However, *E. complanata* are abundant in the Pamunkey River, Virginia, where water temperatures range from 3.5 °C to 28° C (Ayers 1984). Observations of *E. complanata* at the New Castle STP indicated that mussels survived short-term (less than 48 h) exposure to daily high temperatures of 30 to 34° C. A low (10.8 %) to moderate (31 %) mortality rate over 4 months for *Corbicula fluminea* maintained in secondary-treated wastewater was partially attributed to water temperatures of 25 to 32° C, (Haines 1977). Seasonal temperatures affect filtration and respiration rates of freshwater mussels (Walz 1978; Burky 1983; Hornbach et al. 1984). In general, filtration increases with temperature up to some optimum or range, beyond which it declines (Winter 1978). Filtration by *E. complanata* was readily observed at water temperatures recorded during my laboratory and field experiments (19 to 23° C); however, mussels did not filter when water temperatures exceeded 30° C. Lewis (1984) reported that *A. grandis* respired at a greater rate than did *E. complanata* between 5 and

10° C., suggesting that *A grandis* may filter more actively in cool temperatures. This physiological adaptation would extend the water quality benefits of mussel filtration and biodeposition in a sewage lagoon over a broader time period (e.g. early spring and late fall). A few freshwater mussel species have been shown to undergo seasonal adjustments or acclimation in lethal temperature limits and physiological rate responses to temperature (Burky 1983).

Many mussel species exhibit some degree of resistance adaptation to anoxic conditions in aquatic environments (Fuller 1974). Among unionid mussels, survival time under anoxic conditions ranged from 3 days for *Anodonta cygnaea* (Lukacsovics 1966), to 30 days for *Anodonta hallenbeckii* (Culbreth 1941). Oxygen concentrations affect respiration rates of mussels. *E. complanata* and *A. grandis* maintained a constant rate of oxygen consumption over oxygen concentrations of 9 to 1 mg/L, below which respiration declined (Lewis 1984). In contrast, oxygen consumption of *Parreysia corrugata* steadily declined as oxygen concentration dropped from 5.3 to 1.0 mg/L (Lomte and Nagabhushanam 1971). Because anoxic conditions typically prevail throughout much of the water column in anaerobic stabilization ponds, and near the bottom in facultative wastewater ponds, these systems are not appropriate for maintaining mussels (or fish). Mussels may thrive in the aerobic portion of facultative ponds, but episodic or seasonal mixing of the entire water column could expose them to anoxic conditions. Aerobic stabilization ponds would provide a less hazardous environment for mussels. While proper system design and operation minimizes the risk of oxygen depletion, many wastewater lagoons can undergo dramatic shifts in physicochemical conditions. Therefore, supplemental aeration is highly recommended to maintain dissolved oxygen levels of at least 4 to 5 mg/L.

Mussels generally require a pH of 6.5 to 9.0, as do many other aquatic organisms (Spehar et al. 1982). Respiration rate of *Parreysia corrugata* was depressed at a pH of

less than 6.0, and greater than 8.3 (Lomte and Nagabhushanam 1971). Additionally, high pH and temperature increase the concentration of toxic unionized ammonia which exists in an equilibrium with the much less toxic ammonium ion (Emerson et al. 1975). Concentrations of unionized ammonia that are lethal to freshwater bivalves ranged from 0.237 mg/L (Goudreau 1988) to 1.29 mg/L (West 1985), which are equal or greater than the limiting concentrations of 0.2 to 0.4 mg/L for common and silver carp (Buras et al. 1987). Ammonia-nitrogen concentrations were not measured during this study, and the sublethal effects of ammonia on mussel filtration have not been studied. However, pH levels as high as 9.0 were recorded in mussel enclosures, and it is possible that unionized ammonia concentrations may have inhibited mussel filtration to some extent. The total ammonia-nitrogen concentration in a sewage lagoon may be relatively high and pose a hazard to mussels, particularly when pH and temperatures are high.

Mussels can harbor a variety of parasites including bacteria, protozoans, and trematodes (Fuller 1974). The maintenance of suitable environmental conditions is the best insurance against excessive parasitism and disease. This may be particularly important in an organically enriched, stabilization pond environment. Fish reared in treated domestic wastewater accumulate high concentrations of bacteria and viruses in the gut, organs, and muscles (Buras et al. 1987).

Unionacean species (e.g., *Elliptio* and *Anodonta* spp.) are generally larger and longer-lived (e.g., 10 to 20 years) than corbiculacean clams (Ghent et al. 1978; Ayers 1984; Paterson and Cameron 1985). Their longevity makes them good candidates for wastewater treatment applications by reducing the need to replenish stocks in a pond. However, efforts should be directed toward propagation of the captive stock to avoid exploitation of natural populations. Knowledge of unionid mussel species' reproductive habits and requirements must be considered when selecting candidate species. In general, mature larvae (glochidia) are shed from a female mussel's gills, after which the

larvae parasitize a host fish (Burky 1983). After a time, the glochidia drop from the fish, settle on the bottom, and begin an independent life. The parasitic process is readily accomplished in the laboratory, provided gravid females contain mature larvae, and the appropriate host fish species are available. Some unionid mussel species may rely upon a single host species to complete their life cycle, whereas others can successfully parasitize a variety of fish species (Fuller 1974). Hosts for *E. complanata* glochidia in the Pamunkey River, Virginia, include the American eel (*Anguilla rostrata*), blueback herring (*Alosa aestivalis*), American shad (*Alosa sapidissima*), white perch (*Marone americana*), and yellow perch (*Perca flavescens*) (Ayers 1984). Declines in mussel abundance following a variety of aquatic habitat alterations are often attributable to the loss of appropriate host fish species, rather than direct physiological effects on mussels (Fuller 1974).

In addition to the effects of lagoon water quality, it is possible that long-term exposure to unnaturally high food concentrations present in wastewater may have deleterious effects on mussels. Jorgensen (1975) argued that filter-feeding invertebrates are morphologically and physiologically adapted to feed continuously on relatively low concentrations of food typically present in unaltered aquatic and marine environments. Furthermore, discontinuous feeding reflects the sensitivity of experimental animals to unnatural laboratory conditions, and particle concentration regulates filtration only when food is unusually abundant, as would be the case in eutrophic environments. From this perspective, the low filtration rate of *E. complanata* relative to other unionid mussels (Table 14) may have been typical for a filter-feeder in an algae-rich wastewater suspension. While short-term (60 h or less) reductions in suspended solids were apparent, longer-term exposure to a constant, high food level (e.g., higher than mussels are typically exposed to in their natural environment) may have contributed to mortality and weight-loss. Ingestion rate is often reduced when food levels are high enough to stimu-

late the production of pseudofeces (Winter 1978). A priority of future research regarding mussels and wastewater treatment should be an evaluation of mussel growth, production, and condition, particularly the integrity of the gills, during long-term exposure to high food concentrations. In addition, there is a need for longer-term measurements of mussel filtration over continuously changing conditions (Burky 1983). Such research could shed light on the ecological significance of particle size, shape, quality, and quantity in regulating mussel filtration in nature, as well as facilitate an understanding of the potential impacts of atypical food types and quantities in eutrophic environments.

Despite many sources of variability, individual filtration rate may provide a representative estimate of the mussel density needed to achieve a particular treatment level. For example, consider a 7.5 million liter pond with a retention time of 20 days (e.g., Pond 3 at the New Castle STP), resulting in a constant flow rate of 378,500 L/day, (15,771 L/h). If the phytoplankton were mainly comprised of colonial blue-green algae, complete suspended solids removal could be accomplished with a stock density of about 300,000 *E. complanata*, assuming a filtration rate of 53 ml/h. If the phytoplankton were dominated by green algae such as *Scenedesmus*, 120,000 mussels would provide complete treatment, assuming a filtration rate of 134 ml/h/mussel. Suspended solids levels in Pond 3 have remained below 20 mg/L, and *Chlorella* have comprised the majority of the phytoplankton. Under these conditions, a filtration rate of 300 ml/h might be expected, based on published rates for unionid mussel species (Table 14). In this case, 53,000 mussels would be required for complete treatment.

Complete particle removal may not be needed or desired. For example, if the incoming wastewater contained 40 mg/L suspended solids and the desired treatment was to meet the legal effluent standard of 30 mg/L, fewer mussels would be required. Assuming the same range of individual filtration rates (e.g., 53, 134, and 300 ml/h), a stock density of 75,000, 30,000, or 13,000 mussels could reduce suspended solids by 25 %.

Technical aspects of applying mussels to waste treatment clearly are undeveloped at present. This study represents the first known investigation of the potential of a unionid mussel species to control suspended solids and algae in wastewater. In contrast to the native fish species studied, mussels affected significant reductions in suspended solids and appear better suited to wastewater treatment due to their trophic status as filter-feeders on very small particles, adaptability to captive conditions, and tolerance to environmental fluctuations. It is hoped that the results, discussion, and suggestions presented here will stimulate additional investigations so that freshwater mussels can be evaluated and compared with conventional algal removal technologies.

# Conclusions

1. The native fish species (fathead minnow, gizzard shad and Israeli carp) tested in laboratory and field experiments in this study generally were ineffective at reducing suspended solids and algal cell densities from wastewater suspensions.
2. Fathead minnows had no effect on suspended solid levels and little impact on algal cell abundance in field and laboratory studies. Their opportunistic food habits were too general and unpredictable to recommend them as a reliable biological control species.
3. Gizzard shad consumed minimal amounts of certain algae and increased, rather than depressed, suspended solid levels. Mortality of gizzard shad was high. This species generally was intolerant of handling, transport, and the environmental conditions in waste stabilization systems.
4. Grazing by Israeli carp accounted for a modest reduction in certain algae (*Chlorella*), but its potential to consistently suppress significant levels of phytoplankton and suspended solids in wastewater systems remains questionable. Resuspension of periphyton and nutrient regeneration by carp was considered likely and undesirable in stabilization ponds.
5. Exotic species, such as triploid silver carp, may graze more effectively on some phytoplankton taxa than the native species examined. Silver carp were proposed for

this study and although we were unable to obtain triploid stocks, this species may prove to be an effective control agent and should be tested in future pilot-scale studies.

6. Grazing by herbivorous fish was found to be dependent on many environmental conditions and a function of numerous physiological and morphological characteristics of the algae represented such as cell size, cell metabolites, shape, abundance, cell wall thickness, and palatability. These and other attributes strongly influence the ability of fish to effectively reduce suspended solids in the form of algae.
7. Three major problems are apparent in the use of herbivorous or omnivorous fish for wastewater clarification. Overall these fish may promote, rather than suppress, algae and suspended solid levels by: (1) acting as nutrient pumps, recycling dissolved nutrients and viable algae cells through their digestive systems, (2) resuspending bottom sediments and periphyton materials through grazing and swimming activities, and (3) preying on beneficial, zooplankton grazers.
8. *Elliptio complanata*, a riverine mussel species, survived relatively well (> 65 %) in waste systems and provided effective suspended solids control in enclosure studies. It demonstrated a significant capacity to filter suspended solids and phytoplankton from wastewater in both laboratory and field trials. Percent reductions were well in excess of 50 %. Algae and suspended particles, especially small-sized particles that are difficult to remove (1 to 7  $\mu\text{m}$ ), were either directly assimilated or repackaged and sedimented out of suspension as pseudofeces, further promoting clarification.
9. The reductions in suspended solids and algae attributable to mussels may underestimate actual removal since vigorous aeration undoubtedly resuspended some settled

inorganic particles and pseudofeces. Moreover, nutrient recycling by filtering mussels and grazing fish may have stimulated greater production of bacterial biomass in the stocked than the unstocked aquaria. Percent reductions were higher when the algal community was dominated by relatively low densities of small, uniformly-shaped green algae and water quality conditions were acceptable and consistent.

10. Reduction of suspended solids and phytoplankton abundance was correlated with, but not directly proportional to mussel density. Increasingly greater mussel densities improved bioclarification, but not uniformly. The percent reduction per individual declined with increasing densities of mussels. Crowding may have inhibited filtration, but since mussels naturally aggregate at high densities, this response may have been an artifact of the experimental conditions. Understanding density-dependent intraspecific relationships that may inhibit or promote filtering effectiveness requires subsequent study. Determining the optimum stocking rates for biological control is dependent on prevailing physicochemical and biotic conditions and the characteristics of the mussel species selected.
11. Other mussel species (not screened in this study), such as large, pond or lake species (e.g., *Anodonta* species) may be better suited to the standing or slowly flowing environments and water quality conditions characteristic of waste treatment lagoons than *E. complanata*. Further studies are necessary to identify the ideal species or assemblage of mussels to render effective control over the range of sizes and taxa encountered in waste ponds.
12. Time-specific filtration rates by mussels in static systems were characterized by a negative exponential relationship; a precipitous decline in suspended particle con-

centrations occurred through time. For *E. complanata* (shell length 10 cm), mean filtration rate depended on the size, shape, or palatability of different algal taxa. Filtration rate for *Scenedesmus* was 134 ml/h, whereas filtration rate for *Anacystis* was 53 ml/h. filtration rate was 87 ml/h/individual. The greater filtration rates reported in the literature for this species and others suggest that even greater biocontrol than was measured in my study could be achieved.

13. Conceivably, a treatment combination of selected species of mussels, herbivorous fish (e.g., silver carp), zooplankton, and other potential grazers (e.g., insect larvae) may represent the most effective biocontrol system for suppression of suspended solids and phytoplankton in waste stabilization pond systems. Management strategies to avoid lethal and sub-lethal levels of dissolved oxygen, ammonia, nitrite, pH, carbon dioxide, hydrogen sulfide, and water temperature are essential to the survival and efficiency of suspension feeders in sewage lagoons.

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**Appendix A.** Mean initial and final (60 h) concentrations of suspended solids (mg/L) and algae (cells/ml x 10<sup>4</sup>) in treatment and reference aquaria in laboratory grazing trials. Categories are suspended solids (SS), *Anacystis* (ANA), *Oscillatoria* (OSC), and green algae (GRN). Species are eastern elliptio mussels (EE), fathead minnows (FM), and gizzard shad (GS).

| Species | Trial | Category | Treatment $\bar{X}$ (s) |               | Reference $\bar{X}$ (s) |              |             |
|---------|-------|----------|-------------------------|---------------|-------------------------|--------------|-------------|
|         |       |          | initial                 | final         | initial                 | final        |             |
| EE      | 1     | SS       | 110.0 (14.1)            | 52.6 (12.7)   | 101.3 (1.8)             | 113.4 (9.4)  |             |
|         |       | ANA      | 351.0 (33.1)            | 72.0 (10.2)   | 304.2 (84.0)            | 176.4 (5.1)  |             |
|         |       | OSC      | 40.0 (6.2)              | 2.0 (1.3)     | 41.9 (2.5)              | 34.6 (0.5)   |             |
|         |       | GRN      | 1.2 (0.7)               | 0.1 (0.0)     | 1.0 (0.1)               | 1.1 (0.2)    |             |
|         | 2     | SS       | 124.6 (17.3)            | 90.6 (17.0)   | 76.6 (14.1)             | 101.2 (15.9) |             |
|         |       | ANA      | 176.4 (35.8)            | 130.5 (18.2)  | 135.0 (2.5)             | 144.0 (25.5) |             |
|         |       | OSC      | 20.7 (2.2)              | 25.1 (10.1)   | 21.7 (4.5)              | 25.1 (5.2)   |             |
|         |       | GRN      | 3.9 (0.5)               | 3.0 (0.8)     | 4.4 (0.1)               | 3.9 (1.4)    |             |
|         | 3     | ANA      | 123.6 (25.5)            | 65.4 (17.8)   | 144.0 (25.5)            | 154.8 (30.5) |             |
|         |       | OSC      | 24.6 (7.9)              | 8.2 (7.6)     | 25.1 (5.2)              | 4.1 (1.3)    |             |
|         |       | GRN      | 3.0 (0.7)               | 1.6 (0.6)     | 3.9 (1.4)               | 2.7 (0.8)    |             |
|         | 4     | SS       | 113.7 (11.1)            | 44.2 (10.0)   | 125.0 (7.1)             | 143.3 (4.7)  |             |
|         |       | ANA      | 369.0 (37.7)            | 180.0 (23.9)  | 385.2 (45.8)            | 694.8 (61.1) |             |
|         |       | OSC      | 30.8 (7.0)              | 4.7 (4.3)     | 24.1 (6.1)              | 23.2 (12.0)  |             |
|         |       | GRN      | 1.9 (0.6)               | 0.3 (0.1)     | 1.6 (0.1)               | 2.1 (0.4)    |             |
|         | FM    | 1        | SS                      | 88.1 (9.5)    | 95.6 (9.2)              | 83.7 (1.7)   | 86.5 (26.1) |
| ANA     |       |          | 36.6 (8.3)              | 15.2 (2.5)    | 31.3 (7.5)              | 11.4 (6.6)   |             |
| GRN     |       |          | 9.4 (1.3)               | 7.2 (0.8)     | 9.6 (1.5)               | 5.6 (0.7)    |             |
| 2       |       | SS       | 95.5 (4.1)              | 139.2 (11.35) | 98.0 (8.5)              | 142.5 (14.1) |             |
|         |       | ANA      | 82.5 (10.8)             | 91.5 (12.5)   | 76.7 (34.3)             | 83.9 (0.0)   |             |
|         |       | OSC      | 61.2 (4.9)              | 91.6 (12.7)   | 59.9 (6.0)              | 123.1 (2.3)  |             |
|         |       | GRN      | 26.7 (1.6)              | 32.4 (3.1)    | 22.6 (4.1)              | 29.4 (2.1)   |             |
| 3       |       | SS       | 103.8 (5.3)             | 128.3 (16.5)  | 101.3 (1.8)             | 113.3 (9.4)  |             |
|         |       | ANA      | 257.4 (12.7)            | 219.6 (50.9)  | 304.2 (84.0)            | 176.4 (5.1)  |             |
|         |       | OSC      | 31.0 (6.1)              | 26.1 (10.4)   | 41.9 (2.5)              | 34.6 (5.0)   |             |
| 4       |       | GRN      | 1.6 (0.2)               | 1.9 (0.3)     | 1.0 (0.1)               | 1.1 (0.2)    |             |
|         |       | ANA      | 639.9 (26.7)            | 327.6 (5.1)   | 677.3 (13.5)            | 320.4 (20.4) |             |
|         |       | OSC      | 39.9 (4.5)              | 40.0 (0.8)    | 47.5 (3.1)              | 40.2 (5.2)   |             |
| 5       |       | GRN      | 10.7 (1.0)              | 10.4 (0.6)    | 9.6 (1.1)               | 9.8 (3.0)    |             |
|         |       | SS       | 190.0 (7.1)             | 282.5 (17.7)  | 185.0 (7.1)             | 280.0 (56.6) |             |
|         |       | ANA      | 858.6 (99.3)            | 432.0 (76.4)  | 677.3 (13.5)            | 320.4 (20.4) |             |
|         |       | OSC      | 53.3 (18.8)             | 40.1 (11.2)   | 47.5 (3.1)              | 40.2 (5.2)   |             |
|         |       | GRN      | 11.8 (2.1)              | 9.8 (0.4)     | 9.6 (1.1)               | 9.8 (3.0)    |             |
| GS      |       | 1        | ANA                     | 100.3 (10.2)  | 92.3 (11.8)             | 95.4 (2.5)   | 82.8 (0.0)  |
|         |       |          | OSC                     | 21.3 (2.1)    | 12.6 (4.6)              | 15.1 (0.0)   | 31.0 (8.1)  |
|         | 2     | SS       | 113.0 (10.9)            | 149.4 (17.6)  | 106.0 (5.7)             | 63.8 (1.8)   |             |
|         |       | ANA      | 92.2 (11.8)             | 63.2 (18.7)   | 82.8 (0.0)              | 41.8 (19.9)  |             |
|         |       | OSC      | 12.6 (4.6)              | 13.5 (7.4)    | 31.0 (8.1)              | 13.9 (10.4)  |             |

Appendix A. Continued.

| Species | Trial | Category | Treatment $\bar{X}$ (s) |              | Reference $\bar{X}$ (s) |              |
|---------|-------|----------|-------------------------|--------------|-------------------------|--------------|
|         |       |          | initial                 | final        | initial                 | final        |
|         | 3     | SS       | 68.0 (2.8)              | 77.5 (3.5)   | 73.0 (4.2)              | 73.5 (2.1)   |
|         |       | ANA      | 82.8 (5.1)              | 59.4 (2.5)   | 89.1 (8.9)              | 82.8 (30.5)  |
|         |       | OSC      | 13.5 (4.3)              | 4.5 (0.8)    | 14.6 (1.8)              | 16.4 (0.8)   |
|         |       | GRN      | 3.4 (1.1)               | 3.2 (0.0)    | 3.2 (0.6)               | 2.6 (0.2)    |
|         | 4     | SS       | 71.0 (4.2)              | 92.5 (10.6)  | 73.0 (4.2)              | 73.5 (2.1)   |
|         |       | ANA      | 77.4 (5.1)              | 90.7 (4.1)   | 89.1 (8.9)              | 82.8 (30.5)  |
|         |       | OSC      | 7.2 (3.6)               | 8.6 (2.0)    | 14.6 (1.8)              | 16.4 (7.6)   |
|         |       | GRN      | 3.2 (0.1)               | 3.1 (0.1)    | 3.2 (0.6)               | 2.6 (0.2)    |
|         | 5     | SS       | 77.5 (3.5)              | 133.3 (35.4) | 73.5 (2.1)              | 65.7 (2.6)   |
|         |       | ANA      | 63.5 (3.3)              | 93.2 (9.7)   | 92.7 (16.5)             | 116.1 (21.6) |
|         |       | OSC      | 4.5 (0.8)               | 11.2 (0.5)   | 16.4 (0.8)              | 13.3 (4.1)   |
|         |       | GRN      | 3.2 (0.0)               | 2.9 (0.0)    | 2.6 (0.2)               | 1.8 (0.3)    |
|         | 6     | SS       | 92.5 (10.6)             | 108.9 (15.8) | 73.5 (2.1)              | 65.7 (2.6)   |
|         |       | ANA      | 90.7 (4.1)              | 116.1 (0.3)  | 92.7 (16.5)             | 116.1 (21.6) |
|         |       | OSC      | 8.6 (2.0)               | 16.7 (5.3)   | 16.4 (0.8)              | 13.3 (4.1)   |
|         |       | GRN      | 3.1 (0.1)               | 2.2 (0.5)    | 2.6 (0.2)               | 1.8 (0.3)    |

Appendix B. Mean initial and final (60 h) concentrations of suspended solids (mg/L) and algae (cells/ml x 10<sup>4</sup>) under field conditions. Categories are suspended solids (SS) and *Chlorella* (CLO). Treatments are reference (REF), eastern elliptio mussels (EE), fathead minnows (FM), and Israeli carp (IC).

| Category | Treatment | Initial $\bar{X}$ (s) | Final $\bar{X}$ (s) |
|----------|-----------|-----------------------|---------------------|
| SS       | REF       | 15.1 (1.7)            | 32.4 (3.9)          |
|          | EE        | 14.9 (0.5)            | 14.0 (1.3)          |
|          | FM        | 15.5 (1.2)            | 92.1 (3.2)          |
|          | IC        | 14.7 (1.1)            | 62.4 (8.6)          |
| CLO      | REF       | 18.7 (1.4)            | 94.4 (11.2)         |
|          | EE        | 17.7 (1.7)            | 35.5 (8.4)          |
|          | FM        | 18.0 (1.5)            | 94.2 (8.9)          |
|          | IC        | 17.7 (1.0)            | 75.2 (17.3)         |

Appendix C. Mean initial and final (24 h) concentrations of suspended solids (mg/L) and algae (cells/ml  $\times 10^4$ ) under laboratory conditions. Categories are suspended solids (SS), *Anacystis* (ANA), and green algae (GRN). Treatments are reference (REF), and low, medium, and high densities of mussels.

| Category | Treatment | Initial $\bar{X}$ (s) | Final $\bar{X}$ (s) |
|----------|-----------|-----------------------|---------------------|
| SS       | REF       | 108.4 (8.2)           | 94.5 (8.6)          |
|          | low       | 108.3 (11.9)          | 53.1 (1.7)          |
|          | medium    | 113.9 (8.9)           | 41.0 (6.8)          |
|          | high      | 112.5 (9.4)           | 33.6 (11.6)         |
| ANA      | REF       | 82.8 (8.2)            | 78.0 (11.5)         |
|          | low       | 86.4 (8.2)            | 55.8 (14.5)         |
|          | medium    | 85.2 (9.6)            | 46.2 (10.0)         |
|          | high      | 85.2 (17.3)           | 33.0 (8.9)          |
| GRN      | REF       | 21.8 (2.0)            | 20.2 (2.9)          |
|          | low       | 22.4 (2.0)            | 14.4 (4.2)          |
|          | medium    | 22.3 (2.5)            | 11.3 (2.9)          |
|          | high      | 23.1 (2.4)            | 8.5 (2.4)           |

**Appendix D.** Mean initial and final (24 h) concentrations of suspended solids (mg/L) and algae (cells/ml  $\times 10^4$ ) under field conditions. Categories are suspended solids (SS) and *Chlorella* (CLO). Treatments are reference (REF), and low, medium, and high densities of mussels.

| Category | Treatment | Initial $\bar{X}$ (s) | Final $\bar{X}$ (s) |
|----------|-----------|-----------------------|---------------------|
| SS       | REF       | 17.2 (0.5)            | 18.2 (1.8)          |
|          | low       | 17.5 (0.8)            | 10.3 (2.9)          |
|          | medium    | 17.7 (0.9)            | 10.0 (3.0)          |
|          | high      | 17.4 (0.4)            | 7.9 (2.6)           |
| CLO      | REF       | 14.1 (1.5)            | 17.2 (1.6)          |
|          | low       | 14.2 (0.7)            | 8.0 (0.7)           |
|          | medium    | 14.5 (1.1)            | 4.5 (0.7)           |
|          | high      | 14.5 (0.9)            | 3.1 (0.6)           |

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