



Project Summary

Advanced Biological Treatment of Municipal Wastewater Through Aquaculture

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This research project was initiated with the overall objectives of: (a) assessing the potential of aquaculture as a suitable means of treating municipal sewage in a mid-temperate latitude on an annual basis, (b) providing a set of design criteria for implementation of aquaculture as an advanced wastewater treatment method, and (c) achieving an effluent quality amenable to PL 92-500 and the 1977, 1983, and 1985 standards and goals.

Two four-celled raceways constructed in a series adjacent to a primary wastewater stabilization pond were used in the studies. One raceway series functioned as the experimental system, while the other served as a control. The first experimental phase used a source of wastewater from the primary wastewater stabilization pond. The second experimental phase used a source of wastewater from the primary clarifier of an activated sludge treatment plant. The primary clarifier also provided the source of wastewater to the primary wastewater stabilization pond. Under both experimental conditions the experimental raceway was stocked with a native Oklahoma fish, *Pimephales promelas* Raf., at two stocking densities, one for each experimental phase.

An analysis of the wastewater quality data assembled during the two experimental phases revealed moderate reductions in suspended solids during the first experimental phase which could have been attributed to the presence of the fish. No distinguishable reductions in five-day biochemical oxygen demand

(BOD₅) were statistically supported that could be attributed to the fish stock. Analyses of nutrient parameters also indicated no distinguishable reductions due to fish populations. During the second experimental phase, high mortality due primarily to oxygen stress revealed no observable impact on the quality of wastewater that could be attributed to the fish. Retention time of the wastewater within each cell of the raceway appeared to play a strong role in the observed percentage reductions in most of the regulatory and nutrient parameters.

A marked reduction in fecal coliform organisms was attributed to the retention time of the wastewater rather than due to influences of the fish present in the cells.

Biological studies of fish growth and reproductive capabilities revealed moderate successes with respect to reproduction, while the analysis of growth revealed exceptional potentials for production of biomass over a short period of time.

The full project report covers the period of March 1977 through August 1979 and describes the experiments and results in detail.

This Project Summary was developed by EPA's Robert S. Kerr Environmental Research Laboratory, Ada, OK, to announce key findings of the research project that is fully documented in a separate report of the same title (see Project Report ordering information at back).

Introduction

The current demand for advanced wastewater treatment practices, arising from the enactment of PL 92-500, the Federal Water Pollution Control Act of 1972, coupled with a demand for energy conservation and the underlying economic implications, is rapidly creating a dilemma in the field of wastewater treatment. In Oklahoma alone, approximately 80 percent of the communities are currently utilizing wastewater stabilization ponds as a means of treating municipal sewage. Such conventional treatment practices have not adequately produced effluents of a quality amenable to established standards provided by PL 92-500.

The problem that currently exists in finding an equitable solution arises not from a lack of advanced waste treatment technology but from a cost-effective implementation of advanced physico-chemical treatment processes in areas where the serviced population is inadequate to finance and maintain such facilities. Such a problem establishes a strong premise for investigating the potentials of utilizing existing conventional treatment methods and supplementing physico-chemical and biological treatment processes with extended biological activity in the form of aquaculture.

The primary objectives of this study were to: (a) assess the potential of aquaculture as a suitable means of treating municipal sewage in a mid-temperate latitude on an annual basis, (b) provide a set of design criteria for implementation of aquaculture as a wastewater treatment method, and (c) achieve an effluent quality amenable to PL 92-500 and the 1977, 1983, and 1985 standards and goals. Subordinate objectives included a quantitative determination of the degree of sewage treatment during major seasons, and an evaluation of practical uses and economic potentials of biological products of the system.

The scope of this study was somewhat broad in context, yet limited in application due to the limited background information available and the physico-chemical nature of the wastewater environment. Such limitations necessitated the use of certain criteria in selecting a suitable test organism, namely: (a) a species of fish native to Oklahoma waters, (b) a species known to have a high tolerance for unstable conditions which exist at various intervals in wastewater stabilization ponds, primarily dissolved oxygen (D.O.), and (c) a species of fish readily available and easily handled. For these reasons the fathead

minnow (*Pimephales promelas* Raf.), although not the most efficient filter feeder, was selected as the test organism. The fathead minnow has been classified as an opportunistic feeder, utilizing small invertebrate organisms as well as algae, and its long intestinal tract suggests an ability to function efficiently as a herbivore.

Conclusions

The analysis of the regulatory parameter suspended solids, under the wastewater and stocking conditions employed in the first experimental phase, revealed an apparent contribution of the fish present within the test cells. Such a contribution was reflected in a comparison between the concentration of suspended solids within the effluent in the final cell of the experimental versus control series raceways. Unlike the suspended solids parameter, another regulatory parameter, BOD₅, showed no statistically significant contrast between experimental and control cells, with respect to concentrations monitored. Such results suggest that the direct reduction in suspended solids was insignificant to indirectly stimulate a reduction in BOD₅. The observed percentage reduction in BOD₅ from influent to effluent of each raceway (control and experimental) was apparently related directly to the retention time of wastewater rather than the presence of the fish within the wastewater.

The analyses of regulatory parameters (BOD₅ and suspended solids), and nutrient parameters, under wastewater and stocking conditions in the second experimental phase revealed no apparent contribution of the fish to a reduction in concentrations.

Considering the nature of the wastewater tested under the second experimental phase (wastewater of primary treatment quality), it is apparent that if cultures of fathead minnows are to be implemented as a cost-effective means of supplemental wastewater treatment, they should be considered as an adjunct to the wastewater stabilization pond and not as a substitute for secondary treatment afforded by the wastewater stabilization pond processes.

Under the wastewater conditions provided in the first experimental phase, wastewater from a primary wastewater stabilization pond, the fathead minnow exhibited the ability to live and reproduce successfully with only limited threat from disease.

Under the wastewater conditions provided in the second experimental phase, wastewater from primary treatment quality, the fathead minnow was able to survive. However, under extreme stresses initiated by wastewater with a high oxygen demand, such an existence was extremely limited and prohibited reproductive rigor and success.

In summary, the wastewater community provides an extremely opportunistic environment which, if managed and monitored closely, may provide a very cost-effective approach to the utilization of nutrients that conventionally are carried away by receiving stream waters and very often end up overloading and upsetting the balance of nutrients provided for in the natural stream environment. Finfish within the wastewater environment provide an opportunity for recycling nutrients, capturing fundamental elements within protein-rich products usable in numerous ways by man. Such an approach can be feasibly implemented as a valuable water reuse technique.

Recommendations

A definite potential exists for the integration of aquaculture techniques and wastewater treatment. If such a goal is to be implemented, further studies are necessary to more clearly define the intricate food web that exists within the waste stabilization pond. Such a definition would provide a stronger basis for selecting the appropriate species of fish to be cultured.

The unstable nature of raw and even primarily treated municipal wastewater limits the potential of introducing a population of finfish due to the high level of competition for oxygen; therefore, if finfish culture in the wastewater environment is to be a successful integration of productivity and wastewater treatment, such operations should focus on supplementing the secondary treatment mode.

Due to the wide range of diurnal fluctuations in oxygen and temperature within the wastewater stabilization pond, finfish culture practices should include: (1) a source of emergency aeration to provide a means of quickly stabilizing septic conditions often experienced during the year-round operation of the lagoon, particularly during late summer and early spring seasons, (2) adequate manpower and equipment for handling fish stock to limit the stresses imposed on the fish during handling, (3) disease control protocol to ensure minimal stock loss, and (4) alternative planning for wastewater flows,

including multiple raceway designs, which allow the operator a means of alternating flow and thus eliminate problems with overloading.

The successful implementation of aquaculture practices with wastewater treatment will rely heavily on interdisciplinary skills of biologists and wastewater treatment specialists particularly in organization and establishment of such an operation. Proper initial organization will ensure a smooth operation which should require only minimal skills to maintain after installation.

Facilities, Operation, and Results

The study was conducted at the Bethany-Warr Acres sewage treatment facility which serves a portion of the northwest Oklahoma City municipal complex. The treatment phases of the facility consisted of: (a) screening, (b) grinding (comminutor), (c) aeration, (d) primary and secondary clarification, and (e) sludge digestion. Following these treatment phases the effluent was discharged to primary and secondary wastewater stabilization ponds, where the wastewater underwent biological oxidation processes before being discharged into the adjacent receiving stream (Bluff Creek, a first-order stream location on the Cottonwood Creek drainage basin).

In order to obtain the necessary controls, with respect to flow and retention of wastewater, a separate small-scaled series of wastewater stabilization ponds were constructed adjacent to the existing primary wastewater stabilization pond. This facility, as shown in Figure 1, consisted of eight earthen cells, arranged in two series with four cells in each series. Each cell had a surface area of approximately 0.1 hectare (0.25 acres). The two series of cells were constructed in parallel, with each individual cell within one series corresponding to the same numbered cell within the adjacent series, with respect to sequence, flow, and retention time. This design allowed for one series to serve as an experimental control for the other series. All cells within a single series were constructed as identically as possible to its corresponding cell of the adjacent series, and all ponds were designed with the same general specifications with respect to depth, area, and distribution receptacles. At the end of the two series of treatment cells, one large cell was constructed to receive flows from both series of cells. The contents of this cell was periodically

used for irrigation purposes or was pumped back into the secondary wastewater stabilization pond of the permanent treatment facility.

The experimental facilities were designed to maintain maximum achievable control on flow of wastewater through use of distribution receptacles equipped with 30° V-notch weirs. The arrangement of these receptacles was designed to allow the investigator an alternate means of distributing flow of wastewater throughout the entire series of experimental cells. Due to the nature of the wastewater received by the experimental system, the serial flow pattern was selected as the most appropriate design for experimentation, as it allowed for optimum retention time which in turn produced a quality of wastewater suitable for fish culture.

During the course of the study, the existing treatment facility was reconstructed and modified. This necessitated the utilization of two experimental phases with two separate sets of conditions. The two sets of conditions differed primarily with respect to wastewater characteristics and stocking density of fish. The physical characteristics of flow and retention were the same for both experimental phases.

During the first experimental phase, from October 19, 1977 through May 17, 1978, primary lagoon effluent was provided as the source of wastewater for experimentation. Each of the four cells within the test series were stocked with *Pimephales promelas* Raf. at a density of approximately 38 kilograms (kg)/0.1 hectare (83 pounds (lb)/0.25 acres).

During the second experimental phase, from August 2, 1978 through May 9, 1979, the wastewater source was supplied directly from the primary clarifier of the existing treatment facility, by means of a submersible pump transmitting wastewater to the centrally located collection point and distributing the wastewater to the two series raceways. Due to the high suspended solids and low D.O. content of the wastewater received from the primary clarifier, it was necessary to retain the wastewater, allowing time for stabilization and production of phytoplankton. This was accomplished by eliminating fish stock from the first cell in the experimental series, which provided a more stable environment for the finfish in the remaining cells of this series. The remaining three cells were stocked at a density of 151 kg/0.1 hectare (334 lb/0.25 acres).

Under both experimental phases, wastewater was received by a centrally located

distribution receptacle (Figure 1) and was distributed to each of the two series raceways at an average flow of 0.95 liters/second (l/sec) (15 gallons per minute [gpm]). The flow rate varied from 0.63 l/sec (10 gpm) to 1.26 l/sec (20 gpm) depending on several uncontrollable factors inherent to gravitational distribution. This variation in flow necessitated daily monitoring and correction by adjustment of the V-notch weirs in the distribution receptacles of the first cell in each series. The flow between each cell within each series varied somewhat from the flow to the first cell in each series, depending on the evaporation rate and the amount of rainfall. Without compensating for input from rainfall or loss from evaporation, the retention time of wastewater in each cell was approximately 15-20 days.

For purposes of identifying sample locations, each of the cells in the two series was assigned numbers sequentially through the series. Along with the numbered location, subscripts were assigned to designate the control series and the test series; A - designating the control series with no fish and B - designating the experimental series which contained fish (Figure 1).

During this study, under both phases, wastewater samples were collected weekly from the discharge point at each cell within the two series and a sample was collected from the initial wastewater supplied to the system. All samples were collected on the same weekday at approximately the same time of day (1100-1200 hour). Along with the wastewater samples, temperature and dissolved oxygen data were collected at each sampling location. Several water quality parameters were analyzed including BOD₅, suspended solids, and nutrients.

The performance of the system under two operational modes was evaluated by determining the mean effluent levels from each cell for each of the selected parameters and by comparing changes in the character of the flow as it moved through the experimental series of cells with the character of the flow at the corresponding points in the control series. In addition, overall performance of each series was determined by comparing the effluent concentration from the last cell in each series with the level of that parameter in the influent to the series.

In order to minimize the variability due to the day-to-day changes in the character of the influent and variability due to seasonal influences on temperature, statistical comparison of each cell in the

experimental series with the corresponding cell in the control series was conducted utilizing a t-test of observations paired according to sample date. This permitted the use of an alpha level of 0.05.

Due to the observed critical periods of low oxygen concentrations encountered during the study, a means of supplementing photosynthetic oxygen production was provided using forced air blowers and perforated distribution lines for diffusers. The supplemental aeration supply was designed to provide aeration to experimental and control cells concurrently to minimize bias to the experimentation. Most of the supplemental aeration was necessary during the winter months when extended periods of ice cover jeopardized the existence of sufficient dissolved oxygen concentrations.

In addition to the monitoring of physico-chemical parameters, biological activity of fish was monitored visually and through analysis of growth patterns. Factors monitored visually included reproductive activity, movement and congregation of fish due to oxygen stress, mortality, disease and periodic inventory of fish stock.

During the first experimental phase, the statistical analysis of BOD₅ data (Table 1) indicated a significantly lower concentration in the first experimental cell than in the first control cell, at both the 0.1 and 0.05 alpha levels. The only other cells that maintained any significant differences were the second cells of the raceway series, which displayed significant differences at the 0.1 level only. Considering the real differences in mean effluent concentrations and the relatively narrow range of percent reduction values, it was assumed that no real differences existed between the control cell effluent BOD₅ levels and the experimental cell BOD₅ levels.

Under the second experimental phase, the results of the BOD₅ analyses indicated no statistically significant differences in effluent concentrations for each of the experimental and control cells of the first three stages in the raceway series. The last experimental cell containing fish, displayed significantly higher effluent BOD₅ than its corresponding control cell. Also, the observed percentage reduction was much higher for the control series than for the experimental series containing fish.

Statistical analysis of the suspended solids data during the first experimental phase revealed a somewhat different trend from that of BOD₅ concentration within the first two experimental cells

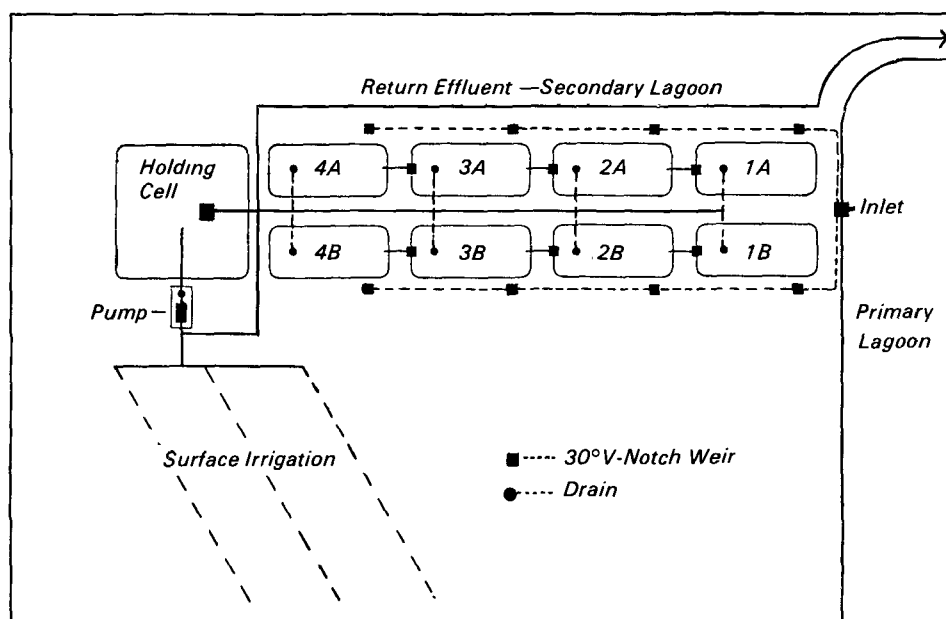


Figure 1. Diagram of Oklahoma State Department of Health, Aquaculture research facilities located at the Bethany-Warr Acres Municipal Wastewater Treatment Plant.

Table 1. Analytical Results for Regulatory Parameter, BOD₅, Sampled During First and Second Experimental Phases

Stat. Para.	Infl.	Cell	First Phase				% Red.
			Effluent				
			1	2	3	4	
\bar{x} mg/l	57.8	Contr.	24.6	27.0	27.5	22.4	61
S	34.0		12.6	17.2	15.9	13.2	
N			25.0	25.0	24.0	24.0	
\bar{x} mg/l		Exp.	19.9	21.5	24.6	21.5	63
S			11.6	14.1	14.7	11.1	
N			24.0	25.0	24.0	22.0	
t (paired observations)			a,b	b			
Second Phase							
\bar{x} mg/l	60.9	Contr.	42.8	32.5	30.3	26.7	41 ^c
S	41.7		23.8	17.8	20.4	14.6	
N	31		33.0	33.0	33.0	33.0	
\bar{x} mg/l		Exp.	47.2	31.7	28.4	30.1	33 ^c
S			28.3	21.3	15.7	14.0	
N			33.0	32.0	32.0	33.0	
t (paired observations)						a,b	

^aIndicates statistical significance at $X = 0.05$.

^bIndicates statistical significance at $X = 0.10$.

^cComposite of cell 1 ($\bar{x} = 15.0$, $S = 26.1$, $N = 66$) was used to calculate % reduction.

(Table 2). No significant difference exists for effluent suspended solids concentration between test cells and corresponding control cells. The third experimental cell showed a significantly lower concentration of suspended solids at an alpha level of 0.1 than its corresponding control and the fourth experimental cell showed a significantly lower concentration at the 0.1 and the .05 alpha levels. Comparison of the overall 20 percent reduction in suspended solids from the point of introduction of influent into the raceway to the effluent from the number four experimental cell, and the 11 percent increase in suspended solids observed across the control cells indicates that a real difference exists. This difference can be attributed to the high finfish population present in the third and fourth experimental cells and their role in reducing suspended solids. Also, the absence of significant reductions in the first two cells of the series likely resulted from the low finfish population levels.

Concentrations of suspended solids during the second experimental phase tend to reflect the same pattern. Experimental cells three and four were found to contain significantly higher concentrations of suspended solids than the corresponding control cells. In fact, a 94 percent increase in suspended solids was observed from the influent into the raceway series to the effluent of the fourth experimental cell, and a four percent increase was observed over the control series. Such results support the view that a large proportion of the suspended solids present in the primary clarifier waste stream were not suitable for consumption by the finfish and their population levels contributed to the suspended solids load.

Experimental results for nutrient parameters revealed a somewhat contradictory pattern. Observed differences in concentrations between experimental cells and corresponding controls failed to reveal any significance on which to base a sound conclusion about a reduction in nutrients due to fish living within the cells.

During the course of study, visual inspection of the general condition of the fish within the wastewater environment revealed a healthy and reproductively viable population with only a moderate amount of disease, apparently initiated by handling during seining operations. Although spawning was observed during the study, the apparent success of such reproductive activity was low as reflected by standing crop harvested during the second experimental phase. Low oxygen

Table 2. Analytical Results for Regulatory Parameter, Suspended Solids, Sampled During First and Second Experimental Phases.

Stat. Para.	Infl.	Cell	First Phase				% Red.
			Effluent				
			1	2	3	4	
\bar{x} mg/l	32.8	Contr.	24.2	34.3	34.1	36.5	-11
S	19.2		21.7	20.8	20.5	23.4	
N	25		24.0	26.0	25.0	26.0	
\bar{x} mg/l		Exp.	22.5	30.3	26.3	26.4	20
S			17.0	23.8	11.7	8.5	
N			26.0	25.0	25.0	25.0	
<i>t</i> (paired observations)					b	a,b	
Second Phase							
\bar{x} mg/l	45.1	Contr.	30.1	26.5	29.6	28.6	4 ^c
S	31.3		21.2	15.5	23.1	13.0	
N	32		32.0	34.0	34.0	34.0	
\bar{x} mg/l		Exp.	20.3	28.6	37.8	57.5	-94
S			14.2	19.2	20.8	28.4	
N			34.0	33.0	34.0	34.0	
<i>t</i> (paired observations)					a,b	a,b	

^aIndicates statistical significance at $X = 0.05$.

^bIndicates statistical significance at $X = 0.10$.

^cComposite of cell 1 ($\bar{x} = 29.7$, $S = 17.9$, $N = 66$) was used to calculate % reduction.

concentrations experienced throughout the course of the study probably contributed the single most detrimental impact on the ability of the young minnows to survive the embryonic stages of development.

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The complete report, entitled "Advanced Biological Treatment of Municipal Wastewater Through Aquaculture," (Order No. PB 83-159 319; Cost: \$11.50, subject to change) will be available only from:

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