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DIVISION OF FISH AND WILDLIFE  
SECTION OF FISHERIES

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A BIOLOGICAL TRICKLING FILTER SYSTEM  
FOR WATER REUSE IN TROUT REARING

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A BIOLOGICAL TRICKLING FILTER SYSTEM  
FOR WATER REUSE IN TROUT REARING

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ABSTRACT

A biological trickling filter was constructed and tested in a 95% water reuse trout rearing system. The filter was capable of removing ammonia from fish raceway effluent at a maximum rate of  $6.4 \times 10^{-5}$  pounds of  $\text{NH}_3\text{-N}$  per square foot of media surface, which was the equivalent of removing the ammonia output from one pound of high protein trout food by 475 square feet of filter media.

At 95% water reuse levels, all fish rearing water quality parameters including temperature, oxygen and ammonia were maintained at satisfactory levels.

Waste water from the 95% water reuse system was generally within water quality standards for hatchery effluent.

Water temperatures in the system could be regulated by the amount of make up water used to vary growth rates and feeding rates of trout to meet specific requirements.

The trickling filter provides an economical and satisfactory method of extending existing water supplies and improving water quality.

## INTRODUCTION

Water quality and quantity are factors which often limit the scope of cold-water fish hatchery operations. The ideal supply is an abundant flow of spring water at a temperature between 50° and 55°F, with no treatment needed except, perhaps, aeration. This water is used most economically in a single pass through the hatchery by gravity flow and with fish loads kept low enough to maintain good water quality.

If high quality spring water is not available, or if hatchery production is expanded beyond the spring water supply, additional water is usually available from wells or from lakes or streams, but at the added cost of pumping and filtration. This cost is minimized by using the water flow to its maximum potential in a single pass system, and by a limited amount of reuse in a serial rearing system by aerating the water between raceways. This type of use is limited by the build up of fish waste metabolites, the most toxic of which is ammonia (Burrows 1964).

If toxic metabolites can be economically removed from the water, the available water supplies can be reused many times and hatchery production can be expanded proportionately. Recirculation can also conserve on costs if the original water supply needs heating, filtration or ultra-violet sterilization.

Biological nitrifying systems were determined to be the superior method of removing ammonia from water (Kramer, Chin and Mayo, 1972). Nitrifying Nitrosomonas bacteria can convert ammonia to nitrite (also toxic), and Nitrobacter bacteria change nitrite to non-toxic nitrate. A flow-through biological filter consists of a large amount of substrate media upon which the nitrifying bacteria can colonize, and the rate of ammonia removal is proportional to the amount of surface provided by the substrate. The rate of ammonia removal is directly affected by the retention time of the water within the filter, and

may be reduced by the interfering action of other organisms (heterotrophs) within the filter.

In addition to ammonia removal, a water reconditioning system must also have some provision for aerating the water to bring the dissolved oxygen up to acceptable levels, usually eighty percent (or more) of saturation. It may also be necessary to provide heating or cooling if the water is out of the acceptable range of temperatures. The intensive reuse of water may promote rapid acceleration of disease and parasite problems, requiring special methods of treatment.

Various aquaria systems have long utilized the principle of biological nitrification of ammonia to condition the water for reuse (Spotte, 1970). These systems are suitable only when fish loads are relatively light<sup>1/</sup> and are not applicable to intensive fish culture. To meet the needs of large scale fish production, modified and greatly expanded versions of biological filters have been developed for optimum efficiency at low cost.

#### BIOLOGICAL FILTER SYSTEMS

The basic filter system designs are the horizontal-flow, down-flow and up-flow submerged filters, and down-flow trickling filters.

A horizontal-flow filter is simple to install in many existing raceways by placing a coarse media, such as crushed rock, in a screened section of the raceway and allowing the water to pass through the media towards the tail of the raceway. Water is then pumped from the tail and piped to the head of the raceway for reuse. Preliminary tests of this design at the St. Paul Hatchery showed good rates of ammonia removal until accumulations of solid wastes within the filter began to clog the interstices and form anaerobic zones of decom-

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<sup>1/</sup>Larmoyeux, Jack D. Closed circuit water systems for fish hatcheries. Presented at Salmon Workshop, Boston Mass. April 25, 1972.

position. Cleaning the wastes from this filter was very difficult, a problem which is more easily dealt with in the other filter designs.

Burrows and Combs (1968) developed a submerged down-flow filter containing a four-foot bed of crushed rock overlain with one foot of oyster shells. The oyster shells mechanically trap solid wastes, serve as a buffer to keep the pH of the water within a desirable range, and may be a source of necessary trace elements for the growth of nitrifying bacteria. The filter must be periodically backflushed with water and compressed air to remove accumulations of solid wastes. The treated water must also be aerated by a separate aspirator before it can be returned to the fish.

The submerged up-flow filter is similar in principle to the down-flow filter, requiring aeration and periodic flushing, but the modifications in flow design make it simpler to operate (Kramer, et al, 1972). Each time a submerged filter is flushed, its discharge must be diverted as waste and an alternate water source used to maintain the fish. The substrate, freshly scoured from the flushing is usually less effective for ammonia and nitrite removal until the bacteria colonies are fully restored. This requires additional time during which supplemental water must be supplied to the fish. For this reason, submerged filters must be constructed as two or more modules to provide a continuous supply of water to the fish while one unit is being flushed and restored to full activity. The use of a substrate with large interstitial spaces will minimize clogging and reduce the frequency of flushing, but may also require the construction of a larger filter to provide the equivalent media surface for bacteria to colonize.

The down-flow trickling filter is an adaptation of a system much used in sewage treatment plants. Clarified effluent from the fish tank is sprayed on

the surface of a column of porous filter media and allowed to flow by gravity to another clarifier at the bottom (Kramer, et al, 1972). The voids in the column are large enough to permit air to flow through the filter, providing oxygen for biological consumption within the filter and for recharging the water before returning it to the fish. The large voids also let the solid wastes slough from the filter media and wash down to the clarifier without the necessity for periodic flushing and interruption of service. The trickling filter thus has the advantages of steady state operation and simultaneous aeration, while requiring only one filter module in a single water reuse system. Because of the simplicity of design, however, it can easily be separated into several modules for independent water supplies to various rearing facilities without increasing the maintenance effort.

The trickling filter in this study was tested to determine the efficiency of ammonia removal at various loading rates, and to measure the performance of rainbow trout in the reconditioned water as compared to a single pass system.

#### DESCRIPTION OF TEST FACILITIES

The fish were held in two rectangular concrete raceways, each 10.5 feet long and 2.08 feet wide, in a heated room within the Department of Natural Resources Hatchery in St. Paul. Fresh water was piped to the head of each raceway and flowed straight through to the drain in the tail of the raceway, where the water depth was maintained at 1.17 feet by a standpipe in the drain-hole. The tail of each raceway was screened to prevent fish from entering the drain. One raceway was used as a single pass, flow-through control and the other was used to test the biological filter.

Water for the raceways was pumped from a well and aerated by spraying above the surface of a reservoir. The trace of hydrogen sulfide in the well water was oxidized during the detention in the reservoir. The water entered

the raceways at a constant 52°F, and with 0.1 ppm ammonia and 7 to 8 ppm dissolved oxygen. The total alkalinity was 240 ppm at a pH of 7.8.

The water in the test raceway was recirculated by pumping water from the tail of raceway to the top of a biological trickling filter, from where it flowed by gravity through the filter media to a clarifier basin, and again by gravity, to the head of the raceway (Figure 1). The total water volume in the clarifier and raceway was 27.42 cubic feet, with another 0.4 to 1.0 cubic feet in the filter, depending on the flow rates. The centrifugal pump was rated at 10 gallons per minute, but a by-pass allowed water to be delivered to the filter at any lesser rate. Fresh well water was added to the head of the raceway as needed to keep water temperatures no higher than 60°F.

The trickling filter was a steel cylinder six feet high and 1.8 feet in diameter. It contained five feet of 2-inch diameter limestone chunks with large enough interstices to allow unimpeded passage of water and free circulation of air. A perforated plate at the top spread the flow evenly over the column, and another perforated plate at the bottom retained the limestone media while allowing the water to trickle into the clarifier tank. Sampling tubes and plexiglass inspection ports were installed at one-foot intervals down the side of the filter to obtain water samples and check on water conditions at intermediate points within the column.

The clarifier tank was large enough (1.87 cubic feet of water) to allow solid wastes from the filter to settle out before the water exited to the raceway. A bottom drain in the clarifier allowed the solids to be flushed into the raceway drain or to be collected for analysis. The clarified water was returned to the head of the raceway by way of an open trough which allowed maximum aeration and near-saturation levels of dissolved oxygen upon delivery.

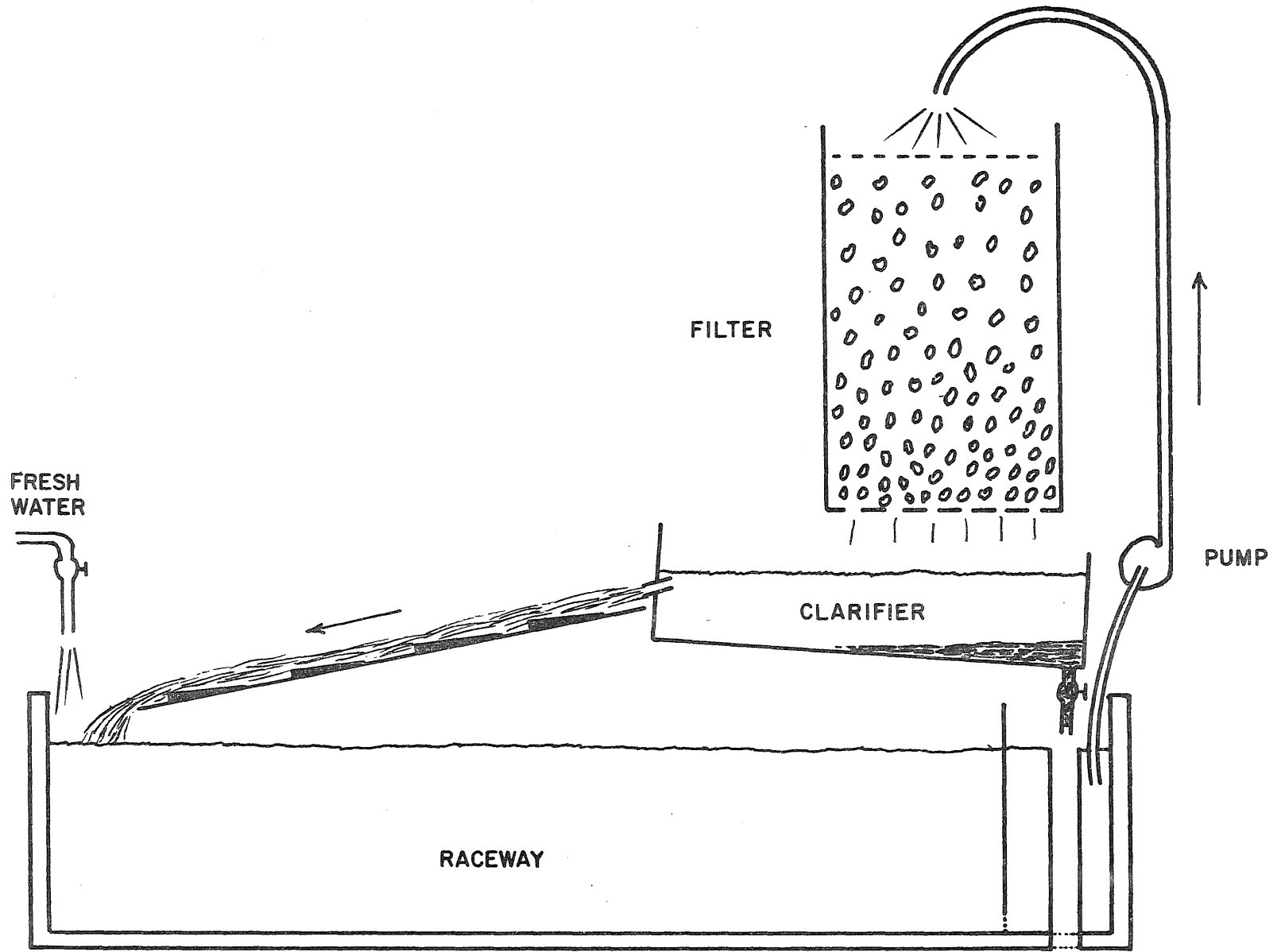


FIGURE I. TRICKLING FILTER FOR CONDITIONING HATCHERY RE-USE WATER.



## PROCEDURES

### Fish Rearing

The test fish for each raceway were taken randomly from a single lot of Donaldson-strain rainbow trout. These fish have a growth potential of about one inch of length for every 19 Temperature Units. (Temperature units per month equals average temperature for 30 days, minus 38.6°F Haskell, 1959.) They were fed federal diets SD<sub>4</sub> and PR<sub>6</sub> in amounts calculated by the formula of Buterbaugh and Willoughby (1967), using an expected conversion rate of 1.3.

Acceptable water quality for these fish was considered to be the generally used standard of a minimum of 5 ppm dissolved oxygen and a maximum of 0.5 ppm ammonia, as measured in the raceway tail-water. Fish loads and feeding levels in both raceways were adjusted periodically as the fish grew in order to keep the water quality up to standard. The maximum fish loads for single-pass usage within these parameters was dependent upon the calculated feeding requirements, and was limited to 0.15 pounds of food per day (at three percent ammonia production) for each gallon per minute of well water flow. By using at least seven gpm in a raceway, the fish could be fed up to 1.0 pounds of food per day, enough to keep 22 pounds of two inch fingerlings or as much as 67 pounds of six-inch fish growing at their normal rate of 0.02 inches per day in 52°F water. The fish load must be reduced at higher temperatures, because the same allowable poundage of food will support fewer pounds of fish at accelerated growth rates. In addition, the amount of food would also be reduced if the inflowing water at the head of the raceway contained more than the 0.1 ppm ammonia or less than the 8ppm dissolved oxygen observed in the well water.

The initial load of fingerling trout in each raceway was kept light to allow for the rapid increase in weight and to allow time for a bacterial culture

to develop in the filter. As the fish grew, the water flow in each raceway was increased as necessary to maintain good water quality up to a flow of about seven gpm, after which fish were removed periodically to keep the water flow requirement from going higher. Flow rates in the reuse system were sometimes set higher than necessary to measure the effect of hydraulic loading on the efficiency of the filter, and excessive fish loads were sometimes allowed in order to measure the maximum ammonia loads that could be handled by the filter.

The fish were weighed and counted each month to determine growth rates. Daily records were kept of flow rates, temperatures, amount of food fed, fish mortalities, medications, and systems maintenance.

#### Water Quality Measurements

Daily water samples were taken each noon from the raceway tailwaters, from one foot intervals within the filter, and from the filter effluent, and were tested for dissolved oxygen (Winkler method and Yellow Springs Instrument Model 54<sup>2/</sup> electronic D.O. meter) and for ammonia and nitrite (Dobie 1962). More frequent samples were taken for a one-week period to measure cyclic diurnal loading changes under different feeding rates. Bi-weekly water samples from each system were tested for phosphate, total alkalinity, nitrate, and total Kjeldahl nitrogen, (Dobie, 1962); for B.O.D. according to Standard Methods for the Examination of Water and Wastewater, 1971; and for pH with Corning Model 7 pH meter.<sup>2/</sup>

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<sup>2/</sup> Mention of brand name does not constitute endorsement of product.

### Solid Wastes

Solid wastes from the filter effluent were collected from the clarifier and allowed to settle in 1000 ml graduated cylinders for 24 hours. The settled fraction was recorded as wet weight. The material was then steam dried and weighed again. Samples of solid wastes were also collected from the raceway outlet. The chemical oxygen demand for the material taken from each source was determined to compare the oxygen demands of the fish wastes and filter effluent solids.

### Filter Retention Time

The retention time of water within the filter was determined by shutting off the filter inflow and measuring the volume of water that drained from the filter from that moment. The retention time ( $T_r$ ) in minutes per pass was calculated by dividing that volume ( $V_f$ ) by the flow rate ( $F$ ) in gallons per minute.

$$T_r = \frac{V_f \text{ (gallons)}}{F \text{ (gpm)}}$$

Measurements were made at several flow rates to determine the effect of hydraulic loading on retention time.

The total amount of contact the water has with the filter media is a function of the retention time per pass and the number of passes made by the entire volume of water (210 gallons) in the system. The flow rate divided by the volume of the system ( $V_s$ ) gives the number of passes per minute. The contact time per day ( $T_c$ ) for this system is therefore:

$$\begin{aligned} T_c \text{ (minutes per day)} &= T_r \text{ (minutes)} \times \frac{F \text{ (gpm)}}{V_s \text{ (gallons)}} \times 1440 \text{ minutes/day} \\ &= \frac{V_f}{F} \times \frac{F}{V_s} \times 1440 \\ &= \frac{V_f}{V_s} \times 1440 \\ &= \frac{V_f}{210} \times 1440 = 7 V_f \end{aligned}$$

Because the volume of water in the filter ( $V_f$ ) varies with the flow rate, the total contact time also varies with the flow rate. The effect of total contact time on ammonia removal was compared for several flow rates.

#### Disease Treatment

The filter system was tested for compatibility with treatments for some of the common diseases of trout. Fin rot was treated at various times by dipping the fish in a 1:2000 copper sulfate solution or by applying malachite green to the entire system at 6 ppm for one-half hour, or formalin at 1:4000 for one hour. In a simulated treatment for bacterial diseases, Terramycin (25 percent oxytetracycline) was fed to the trout at the rate of 2.5 grams per day per hundred pounds of fish for ten days (Allison 1971).

The filter column was by-passed and left dry for periods up to three weeks to simulate periods in which chemicals that might damage the filter would be applied to the raceway. The filter was then put back into operation and monitored for evidence of any decline in its biological activity caused by the dry period.

## FINDINGS AND DISCUSSION

### Raceway Water Quality Under Fish Load

The total alkalinity in both raceways remained stable at 240 ppm, and the pH varied in a narrow range between 7.6 and 8.0.

The water temperature in the single pass raceway remained 52°F with no measureable rise at the tailwater. The water in the reuse system gained less than 1°F per pass at room temperatures of 65°F to 70°F, and the raceway temperature was held at 60°F by adding 0.3 gpm (5 to 10 percent) make-up water at 52°F. Varying the reuse flow rate between 3 and 6 gpm had little effect on the final temperature because changes in the rate of exposure to ambient temperatures during each pass were compensated for by changes in the calculated percentage of cooling water. A change in the absolute amount of make-up water had a more pronounced effect on water temperature.

Ammonia levels and oxygen consumption increased directly with increased fish loads and feeding levels. The total daily production of ammonia or consumption of oxygen closely matched the calculations made by Speece (1973) to determine the effects of various feeding rates and water flows. During each day, however, the ammonia load and oxygen demand climbed from a low point just before first feeding to a maximum in late afternoon, and tapered off slowly until early the following morning (Figure 2). The most consistent level occurred at the 8:00 A.M. low point just before feeding. The peak and time of maximum load was less predictable, and was affected by the number of feedings per day and the amount of food per feeding. It was necessary to measure the ammonia level several times each day in order to calculate the total daily load. Day-to-day comparisons could be made based on single daily samples only if the samples were taken at the same time each day preferably 8:00 A.M. for minimum load or about 3:00 P.M. for maximum load.

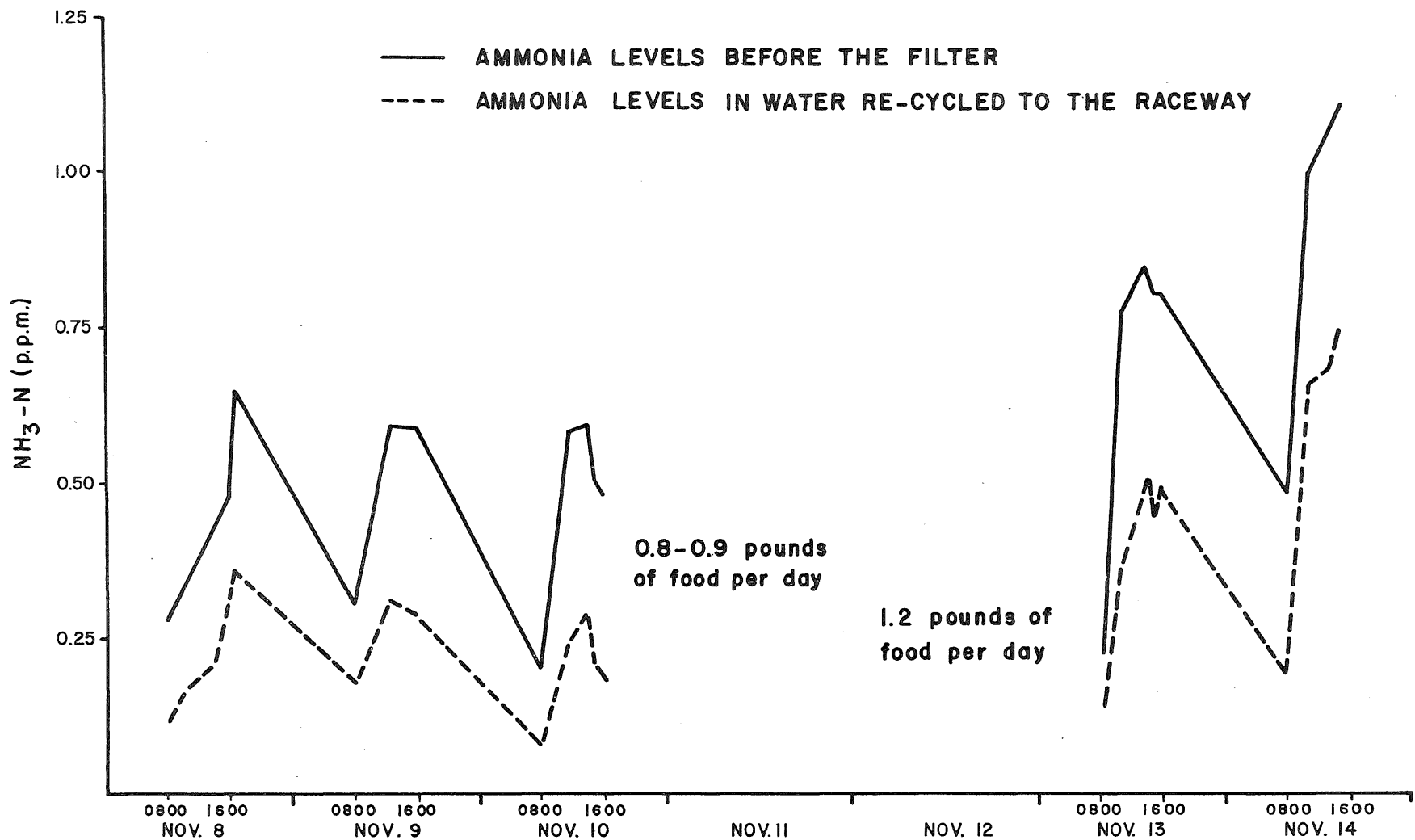


FIGURE 2. AMMONIA LEVELS IN A TRICKLING FILTER RE-USE SYSTEM AT TWO FEEDING RATES.

### Trickling Filter Hydraulics

The maximum test flow that was applied to the filter was 10 gpm, or 3.9 gpm per square foot of filter column upper surface. The water passed readily through the filter at this flow with no ponding, even when the filter was heavily charged with bacterial slimes and solid wastes. Because of the irregular size and shape of the limestone chunks, however, the smaller interstices sometimes became plugged and diverted the flow, causing a small amount of "short-circuiting" and decreasing the effective surface area of the filter media.

The retention time of water passing through the filter varied from 1.5 minutes at 2 gpm, to about 1.0 minute at 3 gpm, but then remained between 0.75 and 1.0 minutes at flows up to 7 gpm (Figure 3). The total daily contact time of the 210 gallons of water in the system at 2 to 3 gpm flow was about 20 minutes. This increased to about 30 minutes at 5 gpm and 50 minutes at 7 gpm. At the heavier flows, however, the thicker film of water on the rocks probably resulted in less effective contact with the bacterial slimes, thereby reducing the effect of the greater contact time.

### Aeration

The dissolved oxygen in the raceway dropped from 9 ppm (90 percent of saturation) at the head to 7-8 ppm at the tail when the fish loads were relatively light. The oxygen content rose to 8.5-9.0 ppm as it passed through the filter, and to 9 ppm as it flowed back to the head of the raceway.

At moderate fish loads, the oxygen dropped to 6-7 ppm in the tailwater, rose to 8-8.5 at the end of the filter, and again returned to the head of the raceway at 9 ppm.

At maximum permissible fish loads, the oxygen was only 5-6 ppm in the tailwater, but rose to 7-8 ppm as it passed through the filter. Water samples

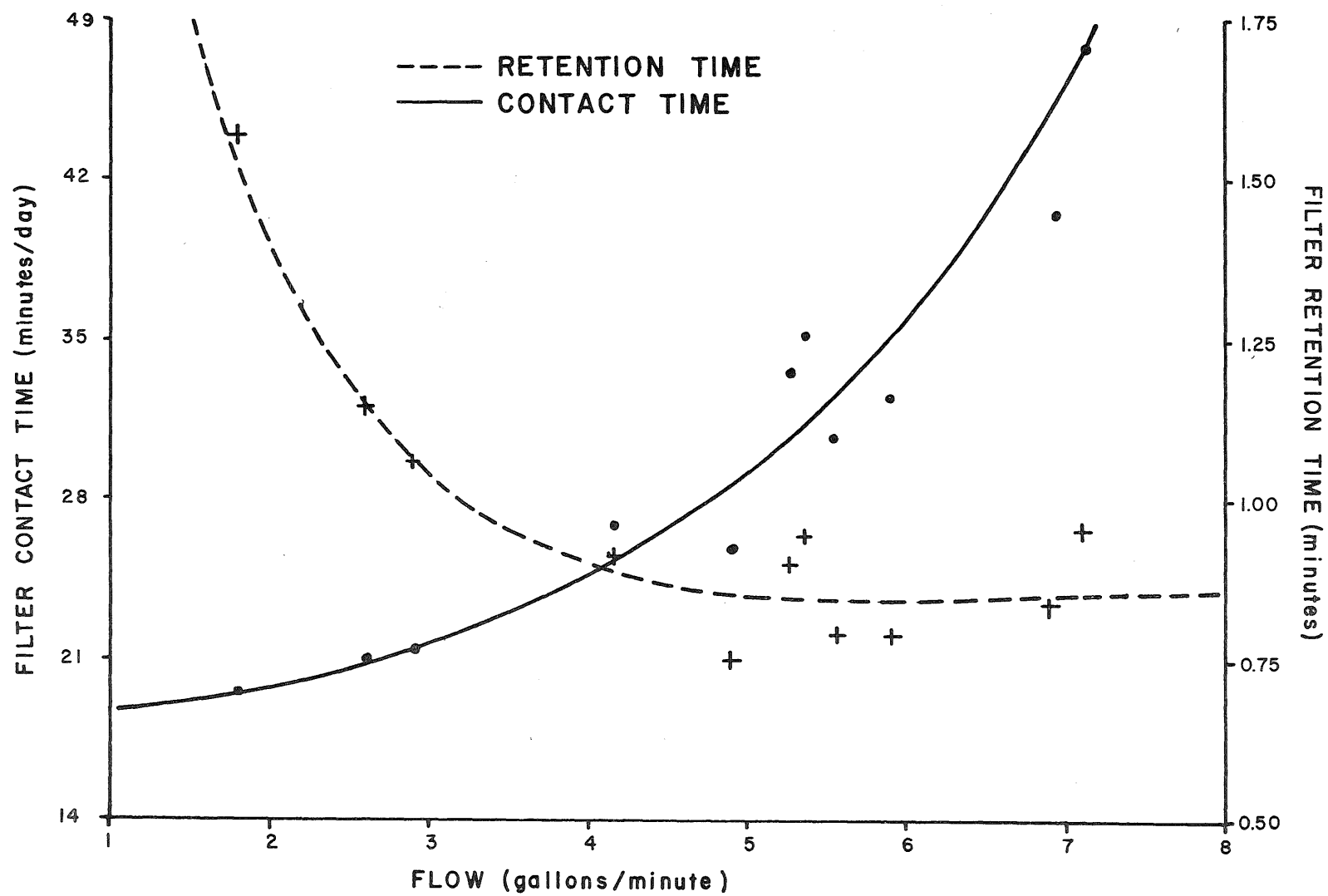


FIGURE 3. FILTER RETENTION AND CONTACT TIME AT VARIOUS HYDRAULIC LOADINGS.



taken at one-foot intervals through the filter sometimes showed a net reduction at some intermediate points because of the high consumption of oxygen by bacteria and heterotrophic organisms which were feeding on the heavy loads of ammonia and organic wastes. In all cases, however, the water left the filter with the dissolved oxygen at 70 to 80 percent of saturation and returned to the head of the head of the raceway at 85 to 90 percent saturation which is entirely satisfactory for trout rearing. Oxygen levels higher than 90 percent saturation are difficult to achieve, and the effort to attain more than 95 percent can seldom be justified (Liao and Mayo, 1972).

As the fish loads and feeding increased, the oxygen in the tailwater dropped to near the 5 ppm level, but better oxygen conditions were restored by increasing the flow rate above the minimum requirements. The trickling filter system served as an effective aerator even for fish loads that were reared beyond the point where ammonia levels became unacceptable.

#### Filter Activation

The freshly crushed limestone in the filter required six weeks of slowly increased fish loading before it acquired a bacterial culture that was fully responsive to maximum loads. The first response of the filter was the development of a Nitrosomonas culture that converted increasing amounts of ammonia to nitrite. The nitrite concentration in the system grew from insignificant levels to about the same concentration as the ammonia for at least a week until the Nitrobacter culture caught up to the load. Toxic levels of nitrite (Kramer, et al 1972) were avoided by keeping the fish loads low and monitoring the nitrite levels frequently. When the filter became fully activated, the nitrite levels dropped to below the ammonia levels.

Once the filter developed a satisfactory culture, it could be left dry for at least three weeks without losing its ability to respond quickly to

ammonia loads when put back in operation. On the other hand, when well water containing 0.1 ppm ammonia was applied to the idle filter in an attempt to retain activity, the filter was completely deactivated within a few weeks and required a full six weeks to regain full capacity. The bacteria apparently needed more than just the ammonia to maintain a healthy culture.

#### Ammonia Removal

The percentage rate of ammonia removal within the activated filter was a function of the ammonia concentration and of the water flow rate. At low-to-moderate ammonia levels and low flow rates, the ammonia concentration was reduced by a net 50 to 60 percent as the water passed through the five feet of filter media. At moderate-to-high water flow rates, the percentage removal of ammonia dropped to 40 to 50 percent on each passage through the filter as a result of the decreasing retention time within the filter.

At higher ammonia concentrations, the percentage removal was generally between 40 and 50 percent, not so much a result of the higher concentration as it was the result of the simultaneous need for higher water flows to meet aeration requirements, thereby lowering the retention time in the filter by as much as a third.

The amount of ammonia removed is the product of the percentage removal and the ammonia load applied to the filter. The ammonia load depends upon the ammonia concentration and flow rate, and is expressed as pounds of ammonia-nitrogen per square foot of filter media surface per day, either as a total daily amount, or as an instantaneous rate that would total that amount if applied to the filter for a full day:

$$\text{lb. (NH}_4\text{-N)/ft}^2\text{/day} = \frac{(\text{NH}_4\text{-N) ppm} \times \text{gpm} \times 8 \text{ pounds/gallon} \times 1440 \text{ minutes/day}}{475 \text{ sq ft. of filter media surface}}$$

Low concentrations of ammonia applied to the filter at low flow rates resulted in ammonia loads of up to  $7 \times 10^{-5}$  pounds of  $\text{NH}_4\text{-N}/\text{ft}^2/\text{day}$  and filter efficiencies higher than 50 percent. As the instantaneous ammonia load to the filter was increased, either by increasing the flow rate or the ammonia concentration, the percentage removal fell below 50 percent, but the amount of ammonia removed continued to increase until the ammonia load approached  $19 \times 10^{-5}$  lb.  $\text{NH}_4\text{-N}/\text{ft}^2/\text{day}$ , after which there was little or no additional ammonia removal (Figure 4). This corresponds closely to the figure of  $20 \times 10^{-5}$  arrived at by Kramer et al (1972) as the maximum capacity per square foot of filter media.

The maximum rate of ammonia removal was approximately  $6.4 \times 10^{-5}$  lb/ $\text{ft}^2/\text{day}$ , or .0304 pounds of ammonia for all 475 square feet of surface on the filter media. This is equivalent to the expected three percent ammonia production from a daily feeding of one pound of trout pellets (Kramer, et al 1972). The trickling filter did, in fact, handle feeding rates up to one pound per day, but higher feeding rates resulted in ammonia levels that rose quickly to dangerous levels. In order to stay within the acceptable level of 0.5 ppm ammonia in the tailwaters at all times, the load to the filter would have to be only  $12 \times 10^{-5}$  lb  $\text{NH}_4\text{-N}/\text{ft}^2/\text{day}$  at 10 gpm, with a total ammonia removal of  $5 \times 10^{-5}$  lb  $\text{NH}_4\text{-N}/\text{ft}^2/\text{day}$ , the equivalent of 0.78 pounds of food per day. There was no evidence, however, that daily peaks of just over 0.5 ppm ammonia were harmful to fish if the ammonia level was lower during most of the day.

Ammonia removal was not always a straight-line function of filter depth. Ammonia levels sometimes actually increased in the upper part of the filter, probably as the result of the breakdown of complex organic compounds by heterotrophic organisms (Spotte 1970). This can be minimized by settling out

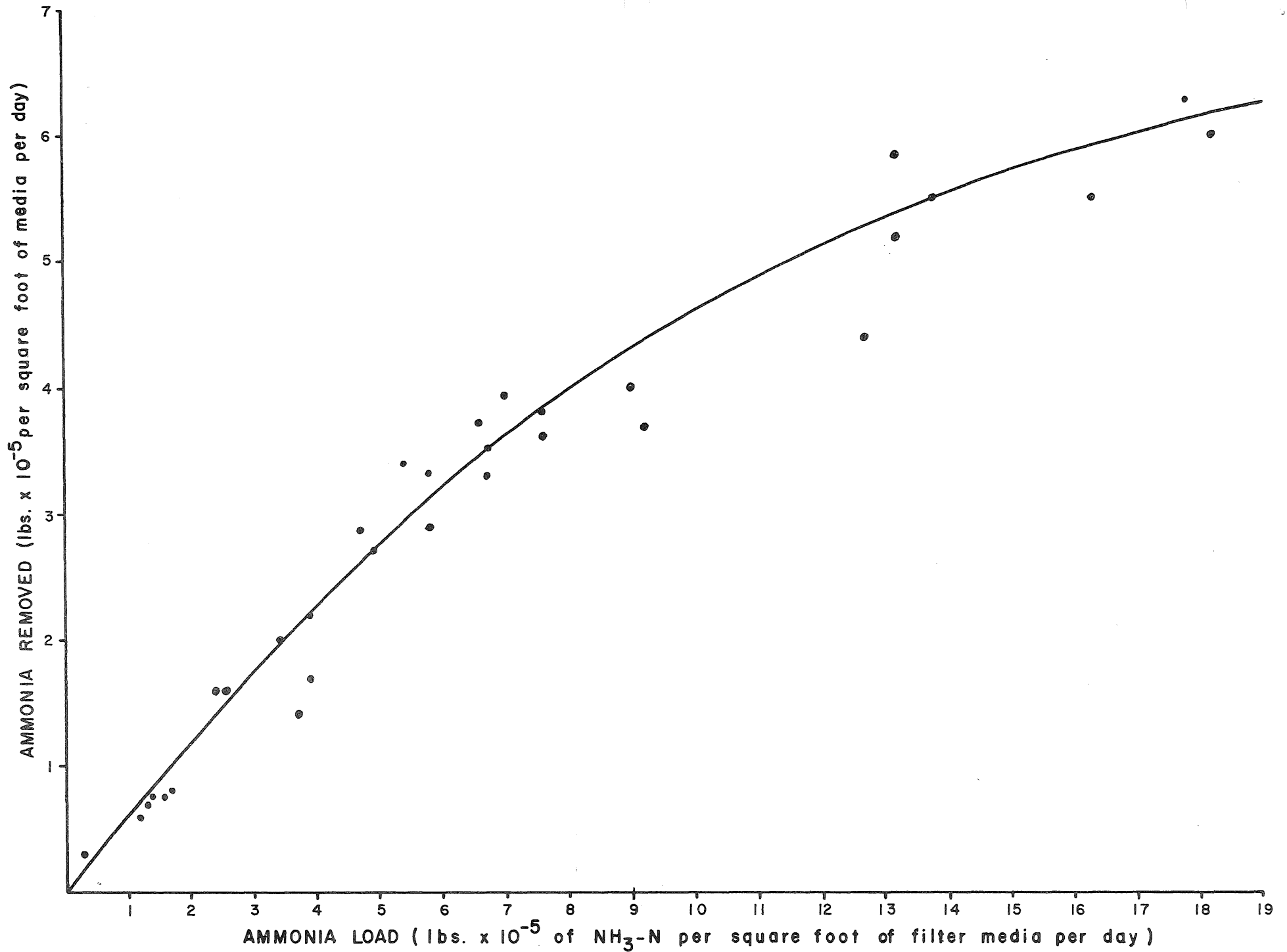


FIGURE 4. AMMONIA REMOVAL IN A TRICKLING FILTER AS A FUNCTION OF AMMONIA LOADING.

as much of the fish raceway solid waste as possible before it gets to the filter. This is especially critical at near-maximum ammonia loads, which can easily become overloads if the filter fluctuates in efficiency. At lower, less critical ammonia loading, the breakdown of organic chemicals can be handled within the filter capacity and is beneficial to the general water quality.

If a constant daily feeding rate were to cause a constant rate of ammonia production in the raceway, the ammonia in the system would stabilize at a constant ammonia load to the filter that would result in ammonia removal exactly equaling the ammonia production, less the diluting effect of make up water. The daily schedule of feeding, however, resulted in cyclic ammonia production and varying loads to the filter. An increase in ammonia production in the raceway meant a higher ammonia concentration in the tailwater and a higher ammonia load to the filter. Because only about half of the extra load could be removed by the filter, the remainder was returned to the head of the raceway, resulting in a higher ammonia level than at the beginning of the previous pass. The additive effect of increased ammonia production and higher initial raceway ammonia, resulted in successively higher ammonia concentrations on each pass until the tailwater ammonia load rose high enough to cause the ammonia removal rate to equal the production rate. As an alternative to letting the ammonia level rise, an increased hydraulic load would also have provided the additional ammonia load to the filter to cause an adequate level of ammonia removal.

The opposite trend took place as the ammonia production diminished later in the day. The high levels of ammonia remaining in the system caused more ammonia to be removed in the filter than was produced in the raceway, resulting in successively lower ammonia levels on each pass until the beginning of the next feeding period. As long as the total daily ammonia load did not exceed

the rated capacity of the filter media, the highs and lows of the cycle remained stable or shifted gradually with changes in the feeding rate (Figure 2). If the daily capacity of the filter was exceeded, the excess ammonia production was raised to a higher level, and the filter was unable to reduce the concentration to the previous day's low, resulting in a higher base level from which to add to for an even higher peak the next day. A continued overload resulted in cumulative gains in daily average ammonia levels and successively higher peaks and lows.

#### Nitrite Removal

The major source of nitrite was from the oxidation of ammonia by Nitrosomonas bacteria. Nitrite is oxidized to nitrate by Nitrobacter bacteria, which lagged behind Nitrosomonas in initial development of a culture, resulting in a temporary build up of nitrite for a week or two. When fully established, the Nitrobacter bacteria kept the nitrite concentrations below the toxic level at all times while the filter was operating within its capacity for ammonia removal. The concentration of nitrite tended to rise in the re-cycled water as the level of ammonia increased, and it neared the minimum anoxia level of 0.2 ppm when ammonia loads approached the capacity of the filter. Nitrite levels tended to cycle with the ammonia cycle, but with a slight lag in the timing of the peaks and lows.

Within the trickling filter, ammonia oxidation in the upper part led to a build up of nitrite toward mid-filter and then a recovery to normal levels as the Nitrobacter culture developed to handle the nitrite load. At steady state, the concentration of nitrite was equal in filter infow and outflow.

### Nitrate Accumulation

Nitrate, as the end product of the oxidation of ammonia and nitrite, tended to accumulate in the re-cycled water to concentrations approaching 3 ppm. In totally closed reuse systems, it has been reported that nitrate has built up to 200 ppm with no apparent harm to fish<sup>3/</sup>. At 90 to 95 percent reuse, the nitrate could only build up to 10 to 20 times the nitrate production per pass, after which the amount of nitrate in the waste water would equal the nitrate production.

### Solid Waste Removal

Most of the solid fish wastes in the raceway settled out in the tailwaters and were flushed down the drain with the waste water. The water pumped to the filter was relatively free of solids, but contained quantities of ammonia and dissolved organic compounds, part of which was converted to solid material in the filter by being incorporated within the culture organisms. Solids that sloughed off the filter media flushed continuously into the clarifier, where they settled into a flocculent layer on the bottom.

The steam-dried weight of the settled material was about half of the wet weight. When the fish were fed at the maximum sustained rate of 1.0 pound of food per day for this system, the filter solids accumulated at an average rate of 0.18 pounds of steam-dried material per day. Larger amounts were sometimes flushed from the filter during sudden increases in flow rates and during fish disease treatments with formalin or malachite green.

The settled, wet solids from the filter had a chemical oxygen demand of 13,186 mg/l, compared to 45,613 mg/l for fish waste solids collected from the raceway tailwaters.

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<sup>3/</sup> Peronel Communication, Oxman Pet Supply, 2450 Louisiana Avenue North, Minneapolis, Minnesota

### Biological Oxygen Demand

At the maximum feeding rate of one pound of food per day, and with reuse flow rate of 5.7 gpm, the B.O.D. reached a maximum of 18.8 ppm in raceway tail-water and 12.9 ppm in the filter effluent, a B.O.D. reduction of 5.9 ppm or almost one-third per pass. By adding 0.3 gpm fresh make-up water to the 5.7 gpm recirculated water, the 12.9 ppm B.O.D. filter effluent was diluted to 12.2 ppm at 6.0 gpm at the head of the raceway. At steady state, the difference between 12.2 ppm at the head and 18.8 ppm at the tail represents a net B.O.D. production of 6.4 ppm for one pass through the raceway at 6.0 gpm and maximum fish loading. Under equivalent feeding rates and single-pass water usage, the raceway effluent would be an outflow of 6 gpm at 6.4 ppm B.O.D. (net gain per pass) for a total B.O.D. of  $3.07 \times 10^{-4}$  lb/minute in the waste water. At the 95 percent reuse rate, the waste water contained a maximum B.O.D. of 18.8 ppm, but was drained off at only 0.3 gpm, for a total B.O.D. of  $4.51 \times 10^{-5}$  lb/minute, a reduction of at least 85.3 percent from the single pass load. Even though the smaller flow from the trickling filter produces a more concentrated effluent, it represents a B.O.D. load reduction comparable to the best secondary sewage treatment plants.

### System Maintenance

The entire system functioned as a steady state operation at less than maximum loads, requiring no backflushing, additional aeration, or attention other than draining off the settled solids from the post-filter clarifier every few days. Even these could be disposed of by wasting a small percentage of the flow continuously through the bottom of the clarifier. In a system such as this, provision must be made to have pipe diameters large enough to avoid clogging from the layer of slime that develops on all surfaces.



### Fish Performance

Because of higher water temperatures and more rapid fish development in the reuse system, the initial mortality of swim-up fry occurred sooner than in the single pass system and was more intense for a short period. However, the overall mortality was nearly the same. Fry in the reuse system had 5.3 percent mortality compared to 7.0 percent for the control, not a significant difference under test conditions.

The growth rate of fish in the reuse system was more rapid than in the control, primarily because the higher water temperatures were closer to the physiological optimum for rainbow trout. The fish in the reuse system also assimilated food at a more efficient conversion rate and required fewer temperature units per inch of growth under normal conditions. However, the onset of disease or parasite infestation was also accelerated in the warmer water and had more serious effects on growth rates and food conversion than in the control.

Manipulation of water temperatures by varying the amount of make-up water in the reuse system allowed a great deal of flexibility in adjusting growth rates and feeding rates. The water could be warmed to accelerate the growth rate, or the water could be cooled to slow down the growth rate and feeding rate if the system became overloaded before the fish load could be thinned out.

Part of the reason for the efficient conversion rates observed in the reuse system may have been an unknown amount of extra food that washed into the raceway from the filter. Sewage fly larvae were very abundant within the filter, and large number of copepods were observed in samples of solid wastes taken from the post-filter clarifier. No measurements were made of how many of these organisms reached the raceway or were eaten by the fish.

Fish Parasites, Diseases, and Treatment

An outbreak of fin rot occurred in both raceways, but was much more serious in the reuse than in the single pass system. Attempts to control the fin rot by dipping the fish in a 1:2000 copper sulfate solution were ineffective. A one-hour treatment with formalin at 1:4000 in each raceway reduced the fin rot in the single-pass system but eliminated all symptoms and mortalities in the reuse system. The warmer water may have caused a greater outbreak of fin rot, but it also made the formalin treatment more effective. The formalin in the reuse system caused a temporary increase in the flushing of solids from the filter, but did not affect the filter's ability to remove ammonia and nitrite.

One group of test fish from an outdoor earthen pond apparently carried with them a very light infestation of Ichthyophthirius and Tricodina parasites. When this group was split and placed in the test raceways, the control group developed no observable symptoms, but the fish in the reuse system rapidly became so badly infested that therapeutic treatments only increased the mortalities. The fish were eliminated from the reuse raceway, the entire system was flushed, and the raceway was restocked with fish taken from the control raceway. The parasites developed rapidly again in the reuse system and treatments again were ineffective. Water temperatures of 52°F or below in the control raceway and outdoor pond apparently inhibited the development of these parasites, but the 60°F water in the reuse system caused rapid and uncontrolled development of the parasites with heavy mortality to the fish. The fish in both raceways were eliminated, and both systems were flushed without chemical treatment. Fish that had been taken as spawn and hatched within the strictly isolated St. Paul rearing facility were then placed in both raceways with no further trouble

from either parasite.

A therapeutic dose of oxytetracycline was given with the feed to healthy fish in the reuse system to determine the effect of this commonly used antibiotic on the performance of the trickling filter. There was no decrease in biological activity of the filter during or after the treatment, nor was there evidence of any other effect on the reuse system.

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