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# USE OF A WATER RECYCLE SYSTEM FOR CULTURE OF SALMONIDS

by

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## ABSTRACT

A small experimental recycle system was successfully operated for over 320 days with Atlantic salmon *(Salmo salar)* under conditions of increasing biomass and three different, successively lower make-up rates. Changes in make-up rate affected the nitrate levels markedly but had little effect on ammonia or nitrite levels or on filter efficiency.

Filter efficiency was inexplicably low during the first half of the experiment but did not result in significant mortality and did not prevent reasonable fish growth. Efficiency improved during the second half of the experiment to a high level near 60% ammonia removal before declining slightly. The experiment was terminated when efficiency collapsed during an outbreak of chironomid larvae in the system.

During periods of improved efficiency, levels of ammonia and nitrite were comparatively low and there was a marked reduction in ammonia concentration across the filters. Periods of poor efficiency showed relatively high ammonia and nitrite, low or falling nitrate levels and little change in ammonia across the filters.

## INTRODUCTION

Water recycle systems are employed in fish culture to conserve water and energy and hence reduce costs required to heat or cool the water by recirculating and reusing it rather than

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discarding it after a single pass. The water must not only be recycled but also reconditioned by treating it to reduce toxic substances to safe levels (Burrows and Combs 1968; Liao and Mayo 1974; Meade 1974).

Ammonia[2] is the dangerous nitrogenous waste of fish in most hatchery conditions (Burrows 1964). It is commonly maintained at safe levels using biological filters. The process, nitrification, is the biological oxidation of ammonium ($NH_4^+$) to nitrite ($NO_2^-$) and from nitrite to nitrate ($NO_3^-$) by bacterial action (Meade 1974). Unionized ammonia is a gas and is toxic at very low concentrations (Burrows 1964); it is reversibly formed by dissociation from ammonium and is kept at safe levels both by the oxidation of ammonium to nitrite by *Nitrosomonas* and by maintaining pH in the neutral to slightly acid range where the vast majority of ammonia-nitrogen is bound in the ionized form ammonium (Trussell 1972) which is non-toxic. Nitrite, the intermediate, is also highly toxic but is rapidly oxidized by *Nitrobacter* to nitrate, the end product of nitrification. Nitrate is relatively non-toxic and so can be tolerated in high concentrations. It is maintained at safe levels by the continuous addition and removal of a small proportion of fresh water (make-up and waste water, respectively) or by denitrification (see Burrows and Combs 1968; Liao and Mayo 1974; Meade 1974).

We designed and built a small water recycle system to learn whether it would provide a safe, dependable and stable environment for small-scale salmonid rearing. This report describes the first phase of our study using the system to culture Atlantic salmon *(Salmo salar)*.

The purpose of the study was to evaluate the system in respect to the well-being of the salmon under recycle conditions, to demonstrate how to operate and maintain the recycle system and to gather chemical data on filter operation under different biomass levels and make-up rates.

## MATERIALS AND METHODS

Our system is shown schematically in Fig. 1. It is briefly described as follows: a single pump circulates water

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[2]We shall use the following terminology (after Burrows 1964): "ammonium" is a specific term for the ion $NH_4^+$; "unionized ammonia" or "gaseous ammonia" are specific terms for the dissolved gas $NH_3$; "ammonia" is a general term referring to $NH_4^+$ and $NH_3$ combined; "ammonia-nitrogen" or "$NH_3$-N" are general terms referring to the nitrogen bound as $NH_4^+$ and $NH_3$ taken together.

to seven 1-$m^2$ (150-ℓ) Swedish rearing tanks, the fish-rearing unit from which water returns by gravity through the rest of the system: an 1800-ℓ sedimentation tank; two 1800-ℓ submerged, upflow filter tanks in parallel, each containing 1.2 $m^3$ of 9-cm Koch rings (Actifil Biorings, Norton Chemical Products Division, Akron, Ohio, U.S.A.); aeration and temperature regulation tanks and sump. Waste water is removed from the sedimentation tank and/or sump. Ultraviolet-light-sterilized make-up water enters at the aeration tanks. The total volume of the system is approximately 7300 ℓ, of which 1050 ℓ is the rearing unit.

The flow rate was 112 ℓ/min (16 ℓ/min/fish tank). For this experiment, to maintain flow rates into the filters and prevent water loss through sedimentation tank overflow, we resorted to vigorous daily use of a suction plunger. This was a temporary solution to the problem of fungus growth fouling the relatively small connecting pipes (nominal 2-inch, schedule 80 PVC plastic, 4.8 cm inside diameter) and clogging the holes drilled in these pipes inside the filters. (A permanent solution would have required a modification to the plumbing, hence a disruption of the filters and perhaps starting the experiment over again.) Plunger use fell to perhaps weekly later during the 2% make-up period as the fungus problem subsided, probably because increasing numbers of chironomid larvae were eating, or successfully competing with the fungus.

The target make-up rate was initially 10% of the circulating flow rate (11.2 ℓ/min) and was changed to 5% (5.6 ℓ/min) on Day 131 and 2% (2.2 ℓ/min) on Day 262. Waste water rates equalled the make-up rate by using an overflow facility. Temperature was maintained at 14 ± 1°C for the 328 days of the experiment. The filters were pre-activated prior to the addition of fish (Meade 1974).

Our water supply is extremely soft and has very little buffering capacity. To maintain pH in the target 6.8 to 7.0 range, we continuously added small amounts of a solution containing 25.2-75.7 g/ℓ sodium bicarbonate.

$NH_3$-N, $NO_2$-N and $NO_3$-N were assayed spectrophotometrically using the reagents of Kaplan (1969), Bendschneider and Robinson (1952) and Hartley and Asai (1963), respectively. $NH_3$ levels were calculated according to Trussell (1972). Acceptable upper limits for $NH_3$, $NH_3$-N, $NO_2$-N and $NO_3$-N were taken to be .005, 1.0, 0.2 (Liao and Mayo 1972) and 100 (Meade 1974) mg/ℓ, respectively.

The salmon were 3-year-old post-smolts, averaging 173 g, that had been held in the laboratory for a year. Initial stocking of the fish was gradual and completed by Day 11. Pelleted Ewos food was fed automatically; the feeding level was increased from somewhat lower to 180 g/tank/day on Day 41 and to 200 g/tank/day from Day 94 onward.

## RESULTS

Survival and growth of the salmon in our recycle system were good. Only a small proportion (8%) died of unknown causes; 20% of the fish were eventually removed from the system because they were simply too big for the tanks. The largest of these fish exceeded 1 kg (2.2 lb) and most had ripe or ripening gonads. The total weight increased from 39 kg (85 lb) on Day 11 to 168 kg (371 lb) on Day 307. This was more than double our estimate of maximum desirable load for these tanks.

The levels of $NH_3$-N, $NO_2$-N and $NO_3$-N (Fig. 2A, 3A) observed were almost always below allowable upper limits of 1.0, 0.2 and 100 mg/ℓ, respectively. Only $NH_3$-N on Days 150, 167, 173 and 328 and $NO_2$-N on Days 321-328 exceeded these limits. Even on the 4 days when $NH_3$-N exceeded its limit, calculated $NH_3$ concentrations were well below the .005 mg/ℓ target (.0018 to .0027).

The nitrate level was very responsive to the make-up rate and levels in the three make-up periods did not overlap (Fig. 2A); the only apparent exception is the extremely low, spurious value on Day 178. This was a day of heavy system cleaning and a replacement of much of the system's water. The nitrate level rose abruptly following both make-up rate transitions (10%/5% and 5%/2%) and the spurious low level on Day 178.

The $NH_3$-N and $NO_2$-N levels observed on any given day are so much lower than $NO_3$-N that they must be plotted on an expanded scale if the trends are to be evident (Fig. 2B). The pattern seen for them is quite different from that of nitrate. $NH_3$-N levels were comparatively high during the last half of the 10% and first half of 5% make-up phases, and at the last four days at 2% while levels were low during the early part of 10%, the last half of 5%, and until near the end of 2%. There are fewer data for $NO_2$-N but it follows a similar pattern, levels generally rising in the second half of 10%, falling in the second half of 5%, rising slightly at the 5%/2% transition and dramatically higher during the last four days of the experiment.

These fluctuations in $NH_3$-N and $NO_2$-N are not tied to the make-up rate, but are indications of the efficiency of the filters as reflected in the changes in ammonium concentration before and after filtration (Fig. 3A). There are few data for the initial part of the experiment, but during the second half of the 10% make-up period and the early part of 5% there was little or no difference in the $NH_3$-N levels observed before and after the nitrifying filters. Beginning during the 5% period and continuing until the last four days of 2% there was always a marked decrease in $NH_3$-N concentration across the filters.

This is shown more clearly in Fig. 3B where the percent of $NH_3$-N removed by nitrification is compared with dilution by the make-up water. During the late 10% and early 5% make-up periods (periods of high $NH_3$-N levels), dilution by make-up water resulted in a greater reduction in $NH_3$-N level than that from the nitrifying filters: the filters were simply not working well. Beginning around Day 160 the percentage removal of $NH_3$-N increased; this coincided with the fall in $NH_3$-N concentrations to low levels throughout the system for most of the rest of the experiment (Fig. 2, 3A). The percent removal increased steadily throughout the 5% make-up period, reaching nearly 60% for the first observation at 2%; during the 2% make-up phase, the percent removal tended downward somewhat to 50%, then 40%, and fell abruptly on the last four days to 10% or less.

The amounts of $NH_3$-N removed by nitrification and dilution are compared with the total amounts removed in Fig. 3C. Here we see that the *proportion* of removal by nitrification actually fell during the 10% make-up phase, rose during 5% and held steady at over 90% during 2% until the abrupt change during the last four days.

The nitrite data (Fig. 2B) show that the levels of $NO_2$-N fell as ammonium removal efficiency increased (during 5%) and the levels rose again as the efficiency dropped (during 2%). This means both of the steps in nitrification and hence both species of bacteria were responding similarly.

An unforeseen problem forced us to terminate the experiment on Day 328. The collapse of filter efficiency occurring then was associated with a very heavy outbreak of chironomid larvae which seemed to be consuming the nitrifying bacteria.

DISCUSSION

We are satisfied with the first phase of our study of this system. The fish not only survived, but also grew and matured. The system itself worked well from at least that point of view and although demanding of time, was not especially difficult to maintain. We can only be encouraged by this since we had no prior experience with the operation of such systems. However, the changes in filter efficiency were entirely unanticipated.

We had expected that a steady state would be established for each make-up phase but this did not develop. The most stable periods were those of comparatively good filter efficiency during late 5% and most of 2%, but even here there were fluctuations in absolute levels and changes in removal rates.

We know the filters were always working to some extent because nitrate was always present as the dominant nitrogen form and would not otherwise have been detectable. Even during the late 10% and early 5% make-up periods, when nitrification, as indicated by the drop in ammonium levels across the filters, was the worst observed in the experiment, the levels of ammonium and nitrite were nonetheless near or below allowable upper limits. Make-up rates of 5% and 10% in such a system alone would therefore seem to be adequate to keep ammonium levels from becoming disastrous in situations where the filters required emergency service. This is a significant observation and has practical application. One of the worries about recycle systems is what happens when and if the filters fail. A make-up rate that was high but still substantially below the rate for single-pass, flow-through conditions, could prevent mortality even if the filters were out of use for an extended period (days or weeks for reactivation, for example). This would be particularly possible if coupled with reduced feeding rates to reduce ammonia excretion and reduced pH and temperature to reduce the proportion that was unionized ammonia. In our case a favorable pH almost certainly prevented our high ammonia levels from causing mortality.

We offer no explanation for the observation that nitrification was ineffective in late 10% and early 5%. Although there were few data collected during the first half of the 10% make-up period, we do not believe the filters then were as ineffective as subsequently. The data collected early in 10%, though incomplete, show comparatively low levels of $NH_3$-N which is an indication of good nitrification. Throughout that latter part of 10% the ammonium levels rose[3] (Fig. 2B, 3A), and the total proportion of ammonium removed by dilution increased (Fig. 3C), both of which indicate worsening, not merely low, nitrification. Our unpublished data show that the filters were certainly pre-activated at the beginning of 10%.

The increasing efficiency and declining ammonia and nitrite levels during the 5% make-up period were gradual and probably represented reactivation of the filters. That is to say, the system appeared to begin recovering from previously existing adverse conditions.

From these results and others from our analysis of filter pre-activation and start-up and the dynamics of the system through 24-hour periods, we feel quite encouraged that successful application of water recycling technology is appropriate for use in small-scale salmonid production.

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[3]The single low value recorded for $NH_3$-N concentration on Day 124 is spurious in the same sense as the nitrate value on Day 178. Day 123 was a day on which the system was undoubtedly thoroughly cleaned and flushed with an excess of clean water.

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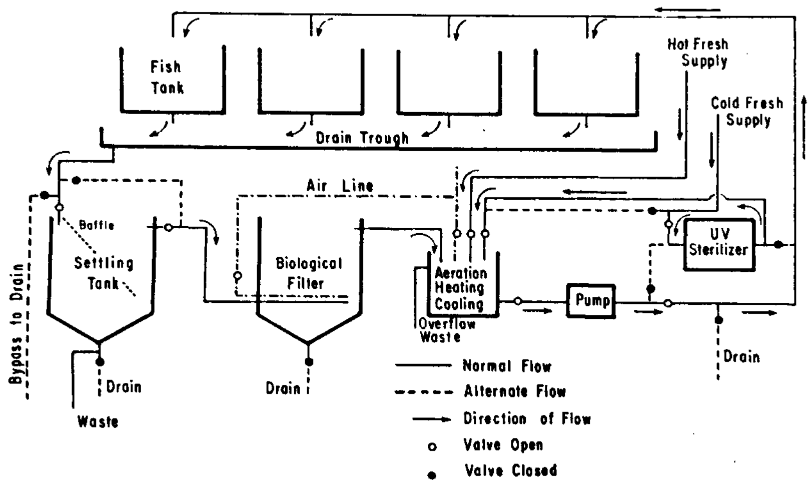


Fig. 1. Diagramatic representation of water recycling system.

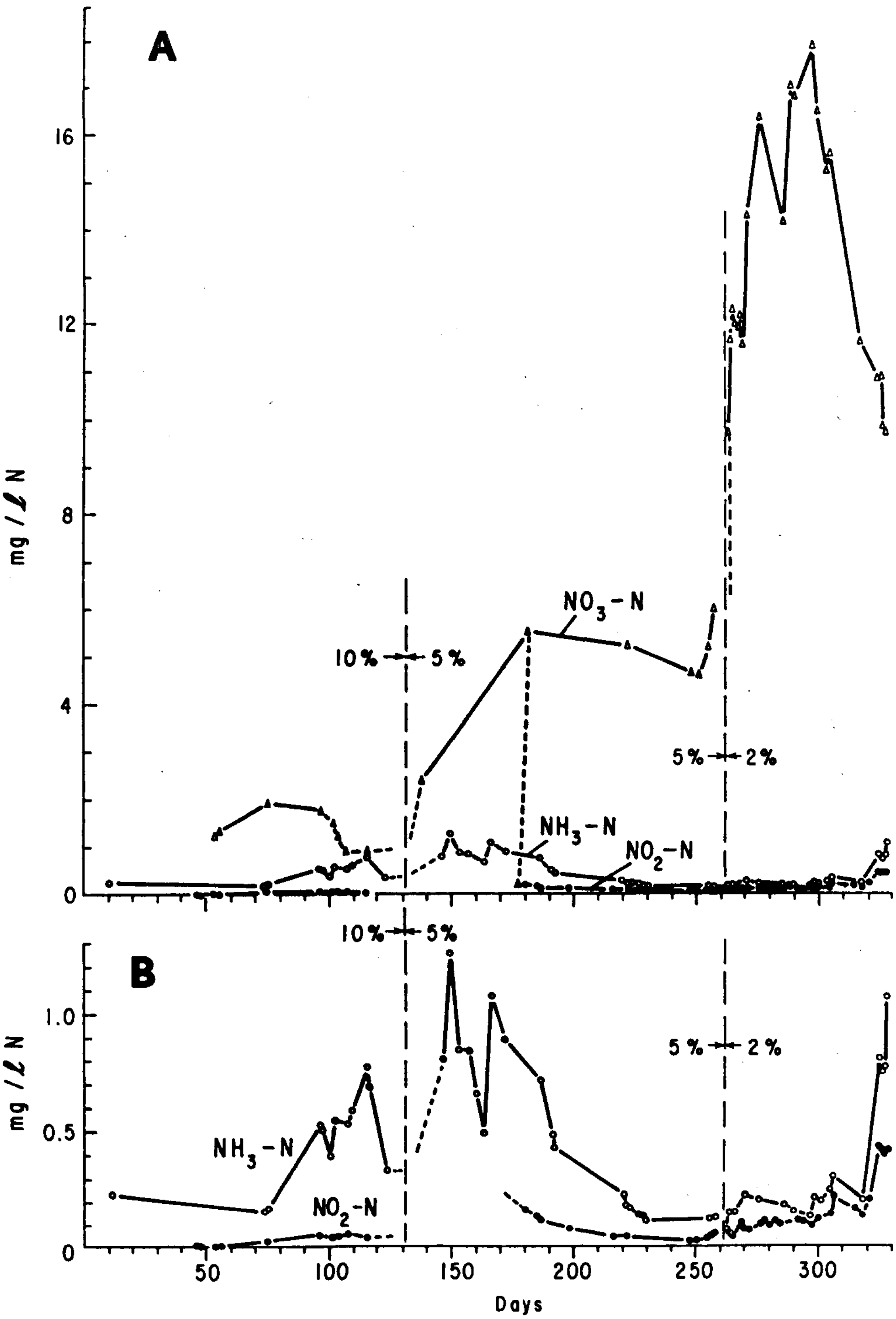


Fig. 2. A - Levels of ammonia-, nitrite- and nitrate-nitrogen in filter effluents; B - Levels of ammonia- and nitrate-nitrogen on an expanded scale.

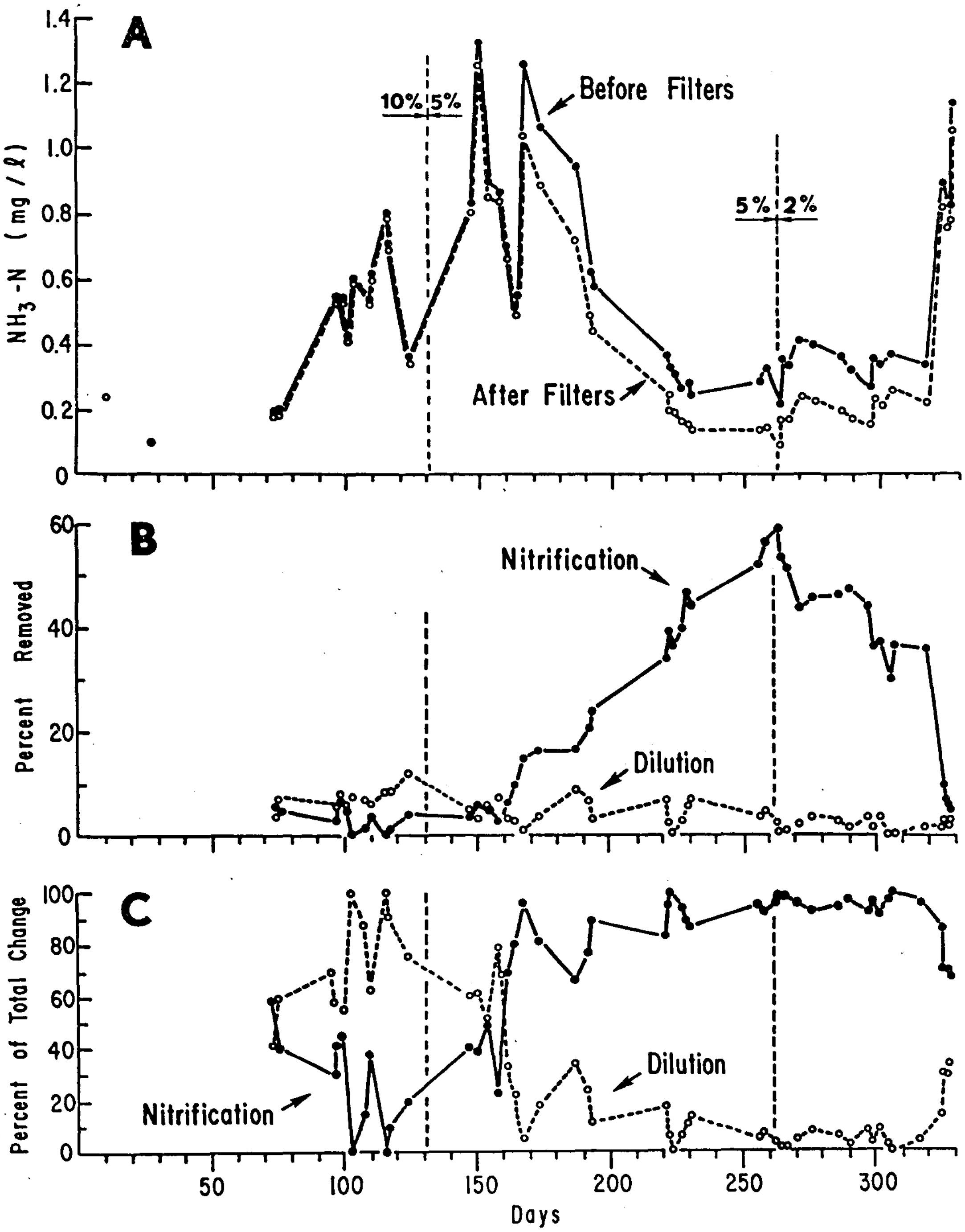


Fig. 3. Changes in ammonia-nitrogen across the filters. A - Levels of ammonia-nitrogen observed before and after nitrification; B - Reduction in ammonia-nitrogen level from nitrification and dilution as percentages of the value observed before nitrification; C - Reduction of ammonia-nitrogen resulting from nitrification and dilution as percentages of the combined total reduction.