CHAPTER 4. RESULTS & DISCUSSION

The development of the experimental protocols for this research presented a number of challenges. The overall objective was to construct and operate a continuous flow sediment-water microcosm system that could be used to investigate denitrification kinetics in selected reaches of the Occoquan Reservoir during periods of bottom water anoxia. In order to achieve the overall goal, however, a number of practical problems had to be solved. As a result, some early experiments were unsuccessful from the standpoint of the ultimate goal, but nevertheless, they provided information that supported necessary changes in experimental protocols, and also enabled the author to develop the operational proficiency that was necessary for the long-term studies contemplated.

As suggested above, the first three experiments were not successful with respect to observation of denitrification kinetics. A range of problems was encountered, including the measurement of nitrate concentrations and oxidation-reduction potentials; and also in maintaining anoxic conditions by excluding oxygen from the reactor(s). Complete resolution of the operating problems was not achieved until Experiment 6.

The author had previously discovered air leaks around the DO probe insertion point and the mixing motor shaft and bearing assembly. These problems were eliminated by designing and constructing new reactor(s), which were first employed for Experiment 5. Although difficulties continued with the measurement of DO and ORP, some of the results after the third experiment were determined to be useful, and are included in the discussion. In order to provide a complete record of research, however, time series plots of all parameters from all experiments not presented in this section are included in Appendix F.

Denitrification and Chemical Reduction of Nitrate

Denitrification

Although the UOSA discharge sometimes increases the oxidized nitrogen concentrations (NO_2^-+NO_3^-) in Bull Run to more than 20 mg/L, concentrations observed at the Occoquan Reservoir dam have remained relatively low. This has previously been attributed

to denitrification in the reservoir during the period of summer stratification (OWML, 1998). However, because the concentrations at the dam have been observed to remain low throughout the year (OWML, 1998), there may be an additional, as yet unidentified, process depleting the oxidized nitrogen.

The main objective of this study was to quantify denitrification in the Occoquan Reservoir during the summer stratification. In examining the historical oxidized nitrogen record in the Occoquan Reservoir, the author concluded that denitrification seemed to be particularly significant in the upper reaches of the reservoir downstream to a point located between Bull Run Marina (RE30) and Ryan's Dam (RE15). Figure 4-1 shows that the oxidized nitrogen concentrations during the summer of 2000 decreased slightly on passing downstream from Yates Ford (ST40) to RE30, and then decreased more rapidly in the remaining reach before Station RE15. Downstream of that point, no similarly dramatic decrease could be observed.

A similar pattern has been observed every year from 1982 to 2000, as shown in Figure 4-2. The observed pattern could result from a combination of high oxygen concentrations and low detention time in the upper reach of the reservoir from ST40 to RE30, and an increase in effective depth in the lower reservoir as shown, respectively, in Figures 4-3 and 4-4. Oxygen concentrations above 0.2 mg/L in the upper reaches would be expected to result in an absence of denitrification (Cavari and Phelps, 1977; and Seitzinger, 1988). In addition, deep-water systems (such as exist in the lower reservoir) are less affected by sediment processes (William, 1986; and Cole, 1994). After considering the physical conditions of the upper Occoquan Reservoir, it was decided to focus the experimental work on the reservoir reach between RE30 and RE15.

Experiment Four

As noted earlier, data from the first three experiments will not be presented in this discussion. Experiment 4 was the first successful attempt to simulate denitrification in the Occoquan Reservoir. The experimental apparatus consisted of a 3-CSTRs-in-series system. The first reactor represented the reservoir from RE30 to the confluence of Bull Run and Occoquan Creek. The second reactor simulated the reservoir to a point just downstream of RE20, and the last reactor represented the reach from that point to a point just upstream of

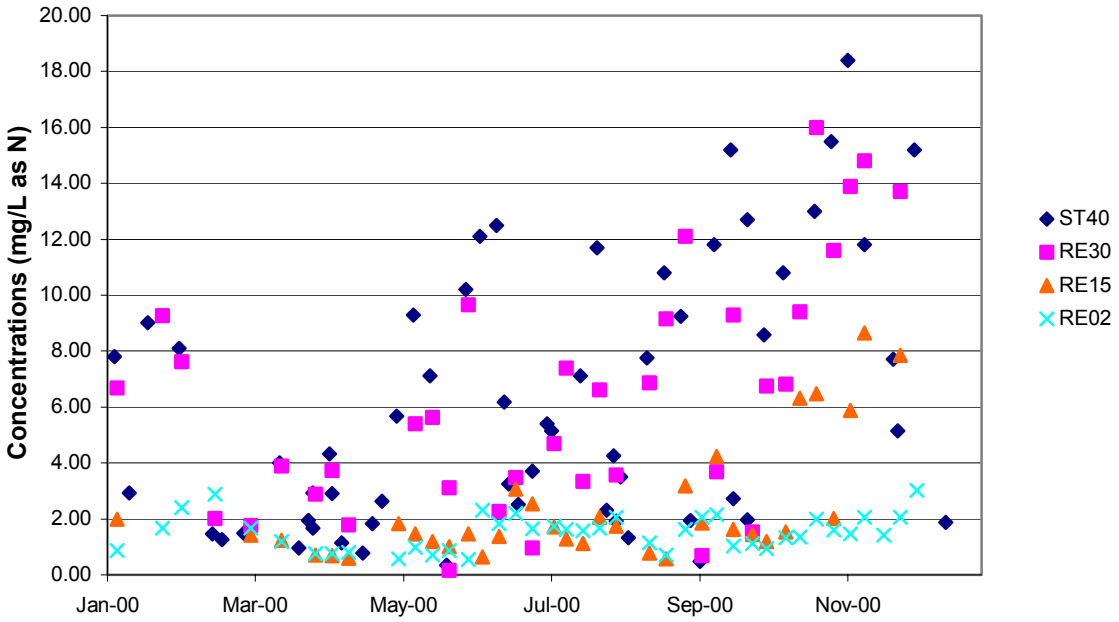

Figure 4-1. Oxidized nitrogen record in the bottom water (10 feet and below) of Occoquan Reservoir at ST40, RE30, RE20, and RE02 in 2000. Source: OWML (2000b).

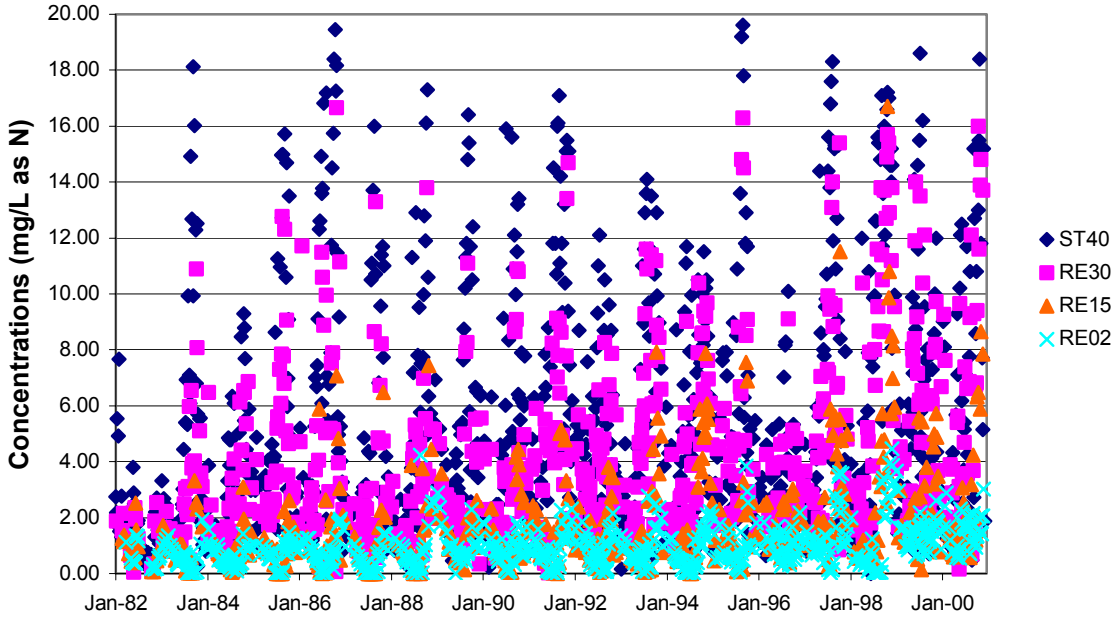

Figure 4-2. Oxidized nitrogen record in the bottom water (10 feet and below) of Occoquan Reservoir at ST40, RE30, RE20, and RE02 from 1982 to 2000. Source: OWML (2000b).

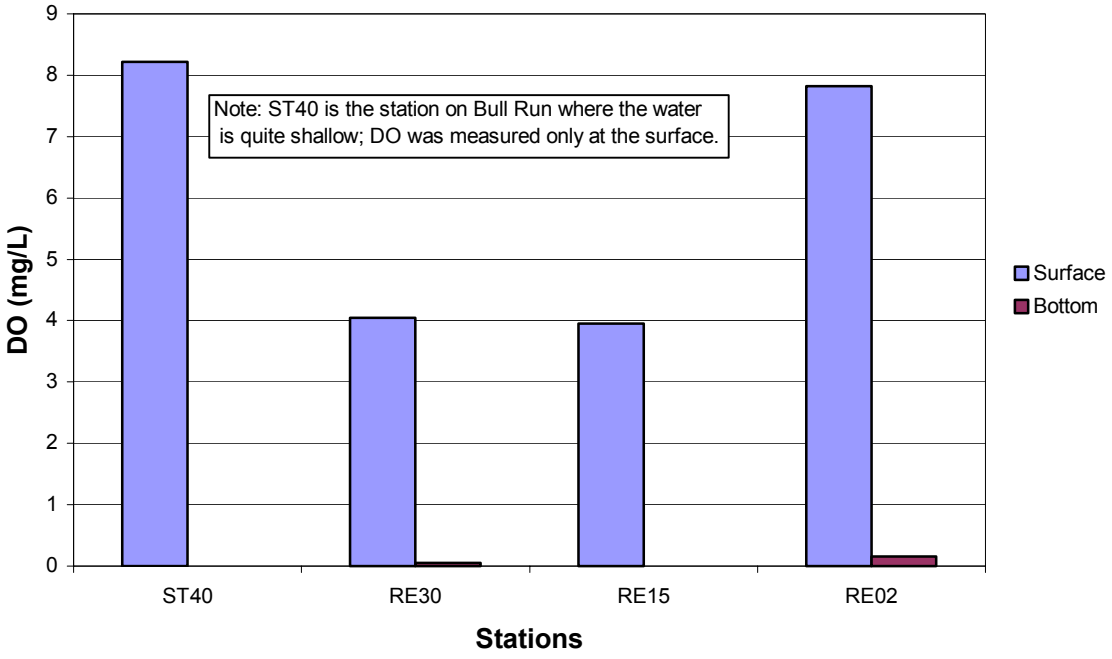

Figure 4-3. Average longitudinal oxygen concentration profile in the summer of 1982 to 2000 along the Occoquan Reservoir. Source: OWML (2000b).

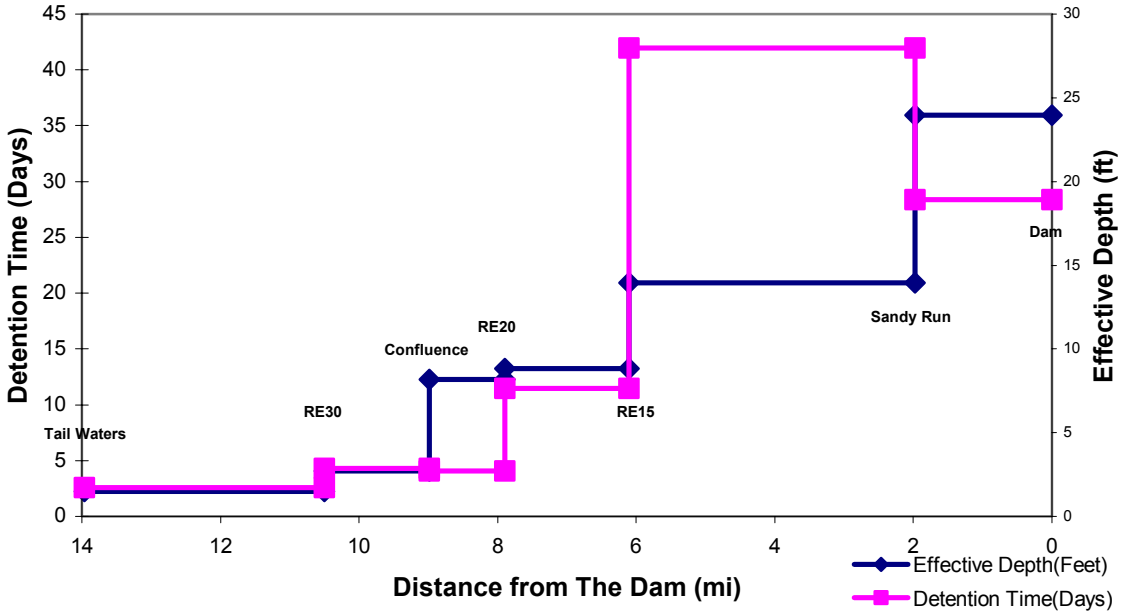

Figure 4-4. Effective depth and detention time profile along the Occoquan Reservoir. The flow rates used to calculate detention times are listed in Appendix D. Source: OWML (2000a).

RE15. The sediment in each reactor was taken at an upstream location in the reach represented.

After sealing the reactors from the atmosphere, nitrate concentrations in all reactors remained relatively stable until Day 11 as shown in Figure 4-5. It should be noted that the analytical protocol used in the present study, as previously described in the *Materials and Methods* section, does not exclusively measure nitrate; it also includes nitrite in the

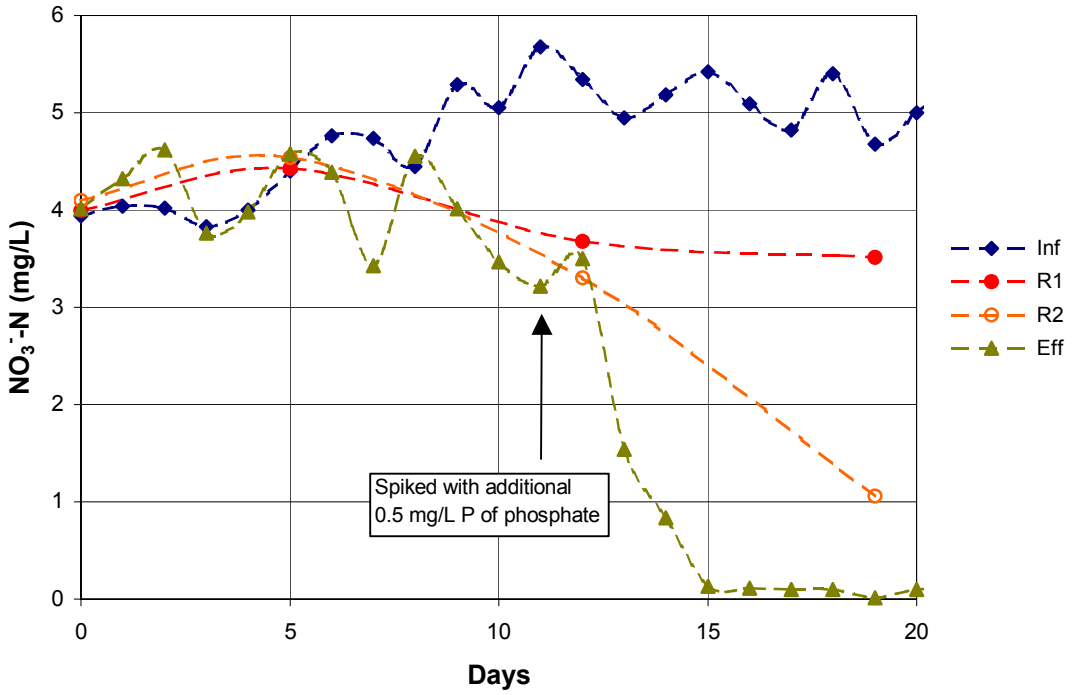

Figure 4-5. Time series of fourth experiment nitrate concentrations in feed water (Inf), Reactor 1 (R1), Reactor 2 (R2), and the effluent (Eff).

result. Nevertheless, because nitrite is a relatively unstable state of oxidized nitrogen and normally exists at low concentrations (Snoeyink and Jenkins, 1980; Stumm and Morgan, 1996), the analytical results for oxidized nitrogen have been taken to be an acceptable surrogate for nitrate, and will be referred to as such herein.

It should be noted also that the sampling strategy produced fewer data points for Reactors 1 and 2 than for Reactor 3. The observed delay in the onset of denitrification prompted the speculation that a phosphorus deficiency might be responsible for the lack of biological activity. This speculation was reinforced by the low observed total phosphorus (TP)

concentrations, which are shown in Figure 4-6, and may be seen to be near the detection limit of 0.01 mg/L P.

In order to determine if phosphorus deficiency was responsible for the lack of activity, the reactors were treated with an additional 0.5 mg/L of inorganic phosphate phosphorus on Day 11. After one day, it was observed that the effluent nitrate concentrations began to decrease, and, as may be seen in Figure 4-7, the denitrification-rate constants dipped into and remained in the negative region, indicating an immediate response to the limiting nutrient addition. The denitrification rates were also found to increase moving down the reactor series. The average denitrification-rate constants in Reactors (R1), Reactor 2 (R2), and Reactor 3 (R3) of Experiment 4 were observed to be, respectively, -0.08 day-1, -0.39 day-1, and –0.97 day-1.

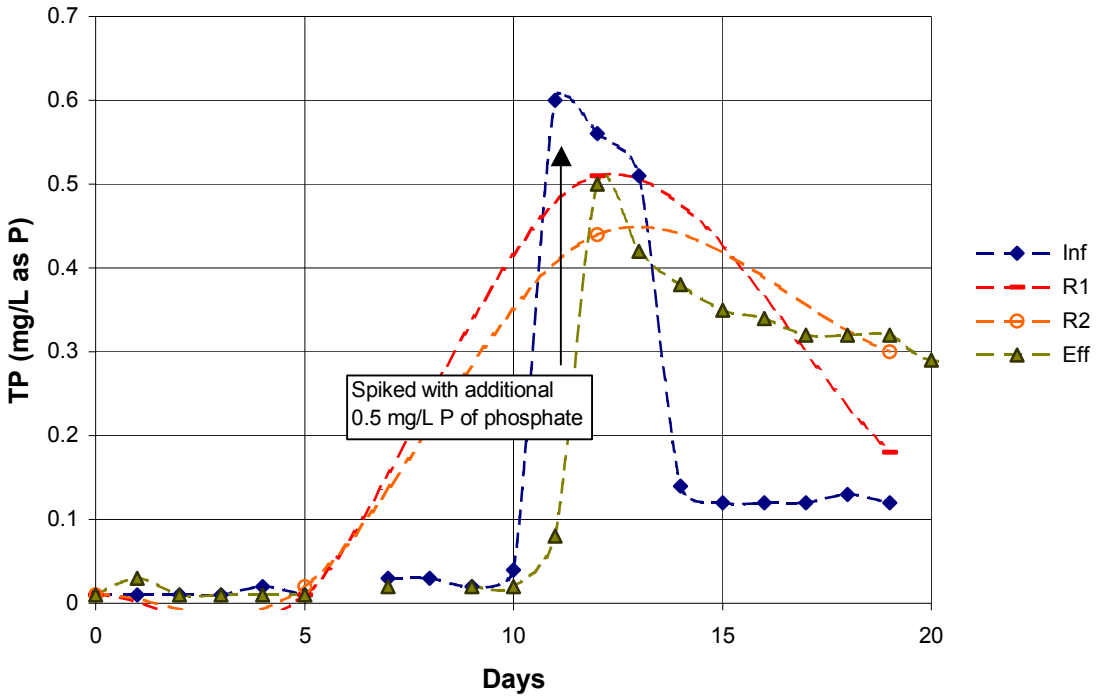

Figure 4-6. Time series of fourth experiment total phosphorus concentrations in feed water (Inf), Reactor 1 (R1), Reactor 2 (R2), and the effluent (Eff).

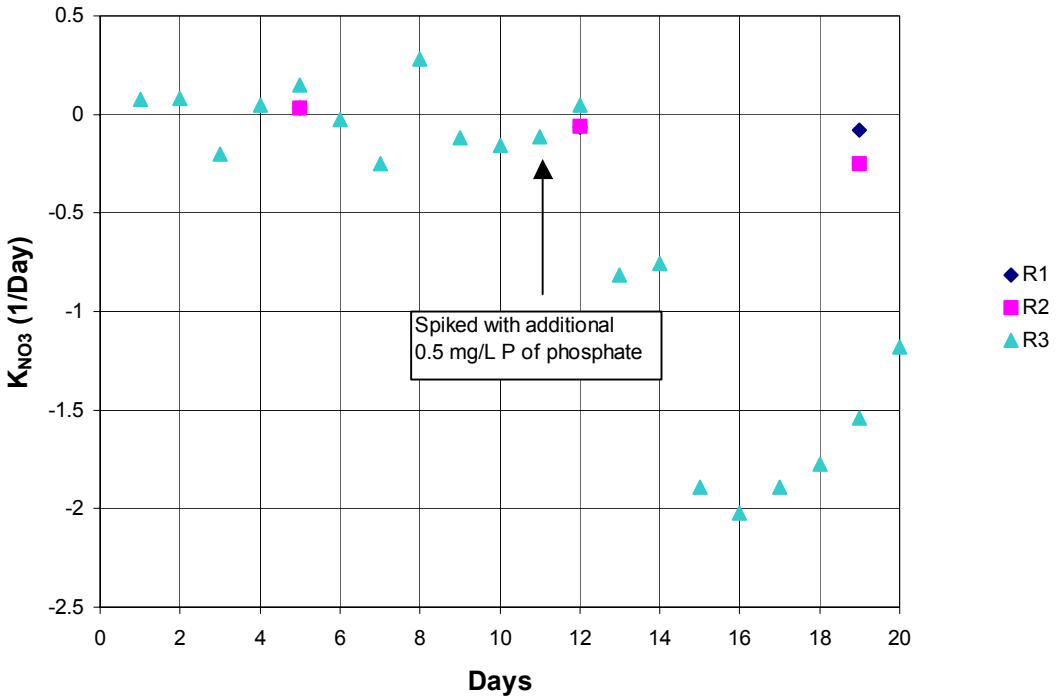

Figure 4-7. Time series of the first-order denitrification-rate constants (K_{NO3}) in Reactor 1, 2, and 3 of Experiment Four.

It was not clear why the denitrification-rate constants of Reactor 3 reached the greatest value on Day 16 and then began to decrease in magnitude. The greatest value of the rate constant also coincided with the disappearance of nitrate from Reactor 3. With regard to the reduced rates of denitrification computed from Day 16 on, it must be remembered that the rates were dependent upon estimates of the influent nitrate concentrations from Reactor 2 interpolated between Days 12 and 19. It is possible that the nature of the CSTR-type interpolation did not accurately represent the activity in Reactor 2. Another possibility is that the denitrification activity after Day 16 had become limited by the very low levels of nitrate (below the detection limit of 0.1 mg/L as N).

Experiment Five

For Experiment 5, only a single reactor of the new design was used in order to reduce the complexity of the experiment and also to test the reactor performance. Except for the design, the reactor was configured similarly to the first reactor of the previous experiment. It was expected that the new design would eliminate previously identified operational problems and would allow observation of denitrification in microcosm systems without phosphate

addition. With the new reactor, it was possible to make DO measurements without introducing air into the reactor headspace. DO had not been measured in Experiment 4 because of this very problem.

In Experiment 5, it was observed that the reactor DO decreased slowly over time as shown in Figure 4-8, and reached 0 mg/L approximately on Day 20. The system was opened on Day 21 to calibrate the DO probe, and atmospheric oxygen entered the water column, as may be seen from the short-term concentration increase to about 1.5 mg/L. Loss of nitrate from the water column began to be observed following the oxygen leak, as shown in Figure 4-9. The computed rate constants are shown in Figure 4-10, and as may be seen, the values fluctuated around zero until Day 25 when they decreased below zero and stayed in the negative region, suggesting that denitrification was taking place. The first-order denitrification rate constant average value from Day 25 to the end of the experiment was -0.13 day^{-1}.

From the results of this experiment, it was concluded that a longer time than anticipated was required to develop anoxic conditions in the reactor. This also prompted some speculation that continuing Experiment 4 for a longer period might have resulted in the onset of denitrification without phosphorus addition. Recognizing that such a long time was required to completely deplete the oxygen in Experiment 5, it was suspected that the upper reach of the reservoir above Station RE30 could play an important role in depleting the oxygen in the waters of the Bull Run Arm of the Reservoir. It is possible that the oxygen in the water column near the bottom may be gradually decreased as the flow approaches RE30, resulting in an observed DO of near zero mg/L at the station. In order to test this hypothesis, it was decided to include a microcosm simulation of the reach of the reservoir from the tail waters on the Bull Run arm to RE30 in Experiment 6.

Experiment Six

Experiment 6 was conducted with a two-CSTR-in-series system simulating the Reservoir from the tail waters on the Bull Run arm to RE15. The first reactor represented the portion from the tail waters to RE20 and the second continued the simulation downstream to RE15. The sediment in Reactor 1 was taken from three sites in the represented reach and sediment of Reactor 2 was taken from the midpoint between RE20 and RE15. The oxygen in

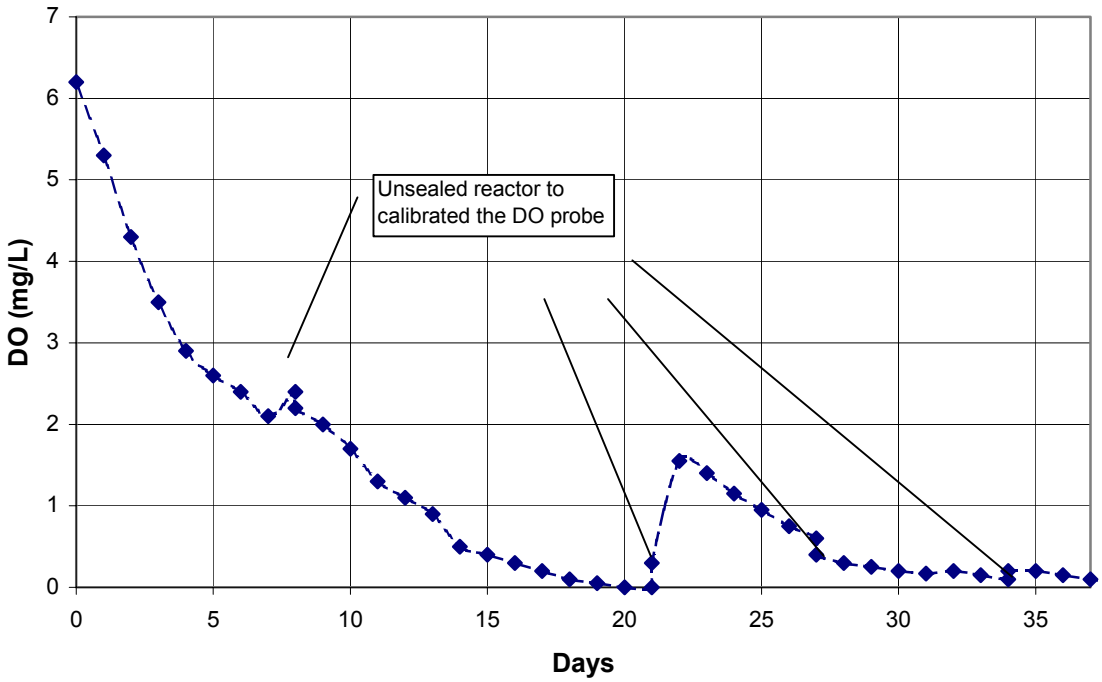

Figure 4-8. Time series of reactor DO in the fifth experiment.

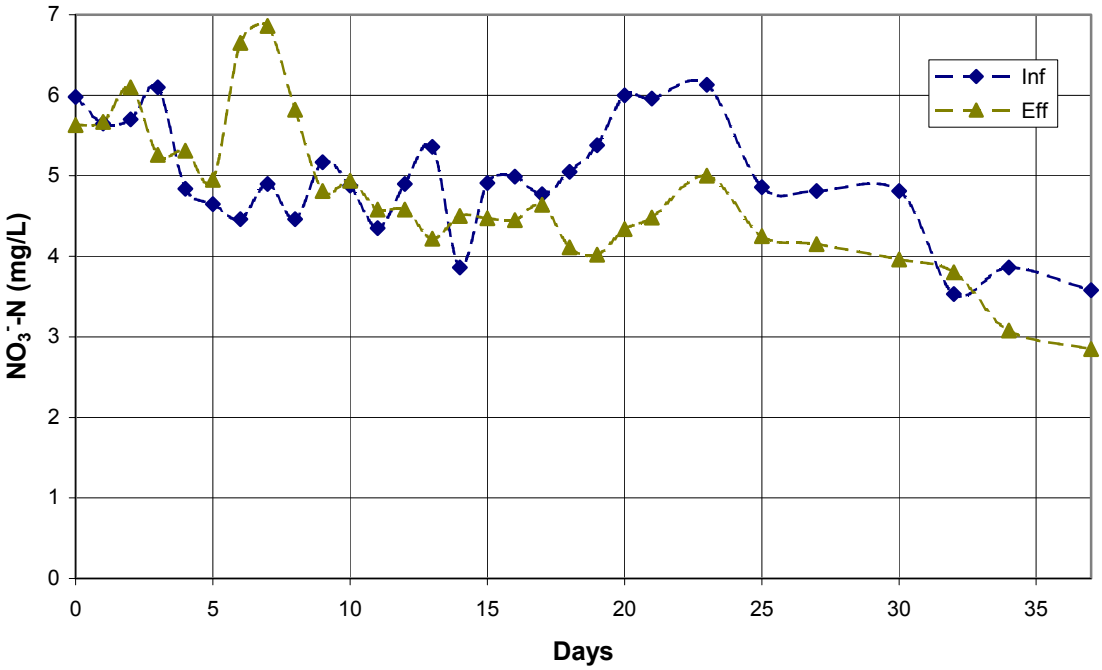

Figure 4-9. Time series of fifth experiment nitrate concentrations in the influent (Inf) and effluent (Eff).

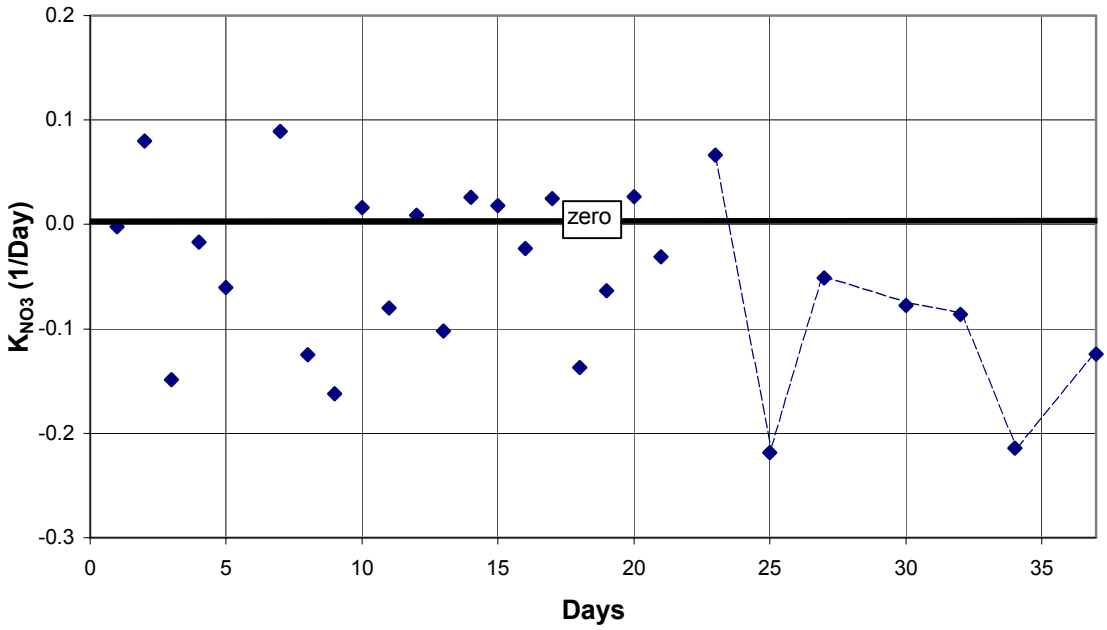

Figure 4-10. Time series of first-order denitrification-rate constants (K_{NO3}) in the fifth experiment.

Reactors 1 and 2 was depleted within approximately 20 days, as shown in Figure 4-11. The DO in Reactor 2 was not measured during the early stages of the experiment because it was suspected that the measurement could introduce significant amounts of oxygen to the reactor, as had been observed in Experiment 5. Nevertheless, after obtaining some experience with excluding atmospheric oxygen when measuring DO in Reactor 1, measurements were resumed in Reactor 2 on Day 18.

Figure 4-12 shows a time series of the nitrate data for the duration of the experiment. As described earlier in the *Materials and Method*s section, near the end of the experiment (on Day 56), the feed water was changed from a mixture of the UOSA discharge and Bull Run water to Bull Run water only. The purpose was to study sediment-water interactions in the absence of nitrate. Using nitrate data collected from the beginning of the experiment until the feed water change, the first order denitrification-rate constants were computed and are shown in Figure 4-13. In examining the figure, it may be seen that the Reactor 1 constants were always in the negative region, meaning that depletion of nitrate was observed to occur even under aerobic conditions. Aerobic depletion of nitrate will be discussed in the next subsection as a possible chemical reaction with reduced manganese.

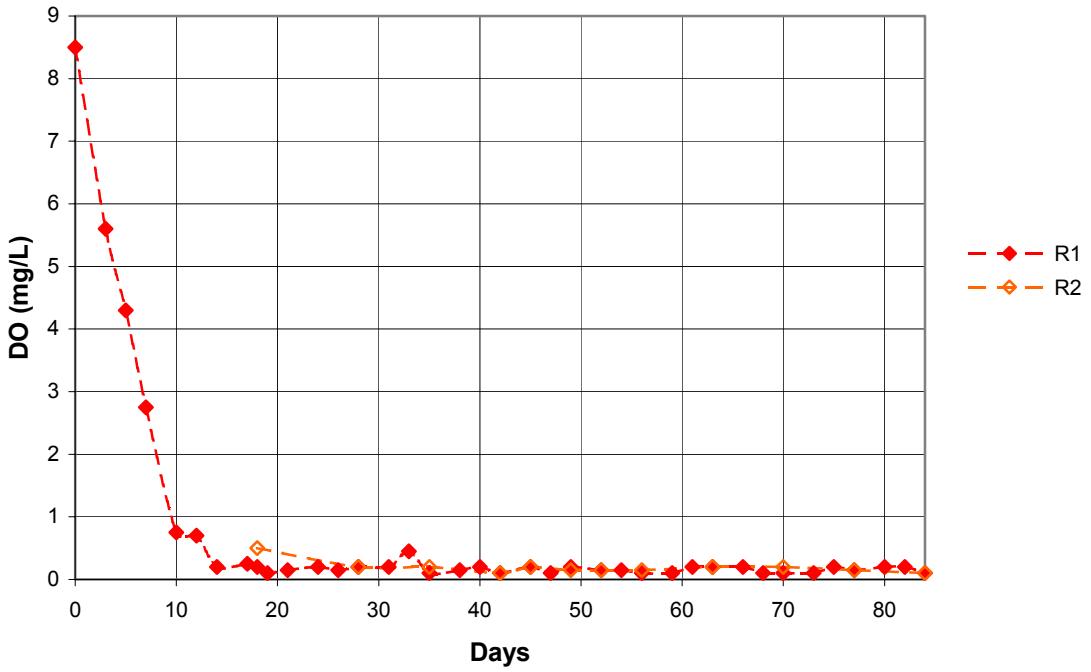

Figure 4-11. Time series of sixth experiment dissolved oxygen concentrations in the solution of the first (R1) and second (R2) reactors.

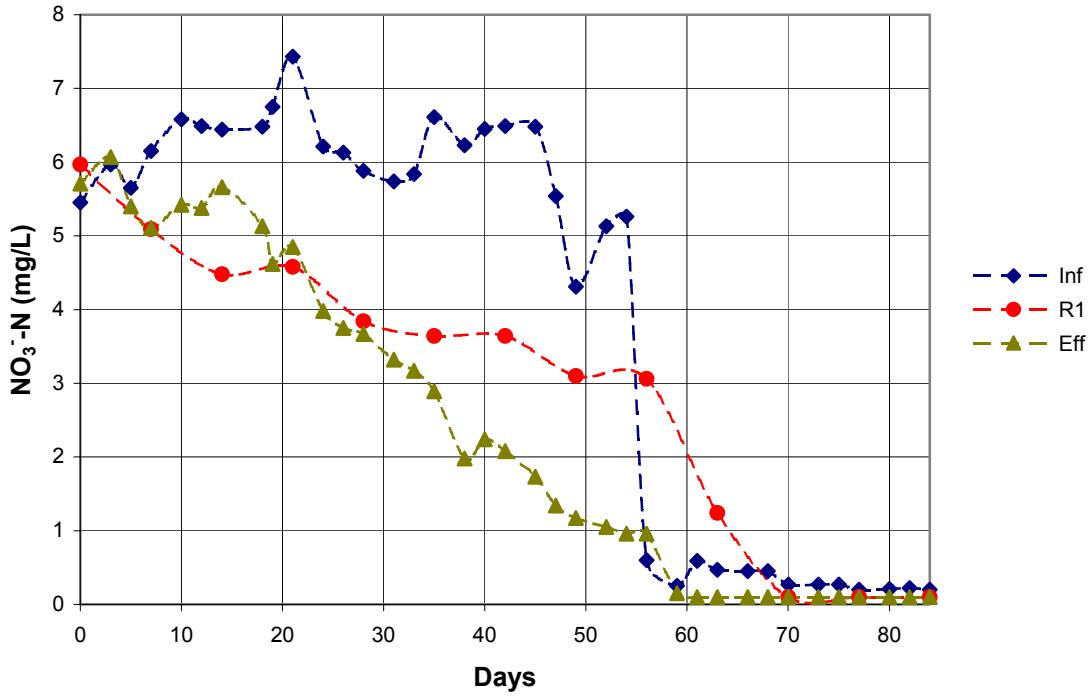

Figure 4-12. Time series of sixth experiment nitrate concentrations in the influent (Inf), first reactor (R1) and effluent (Eff).

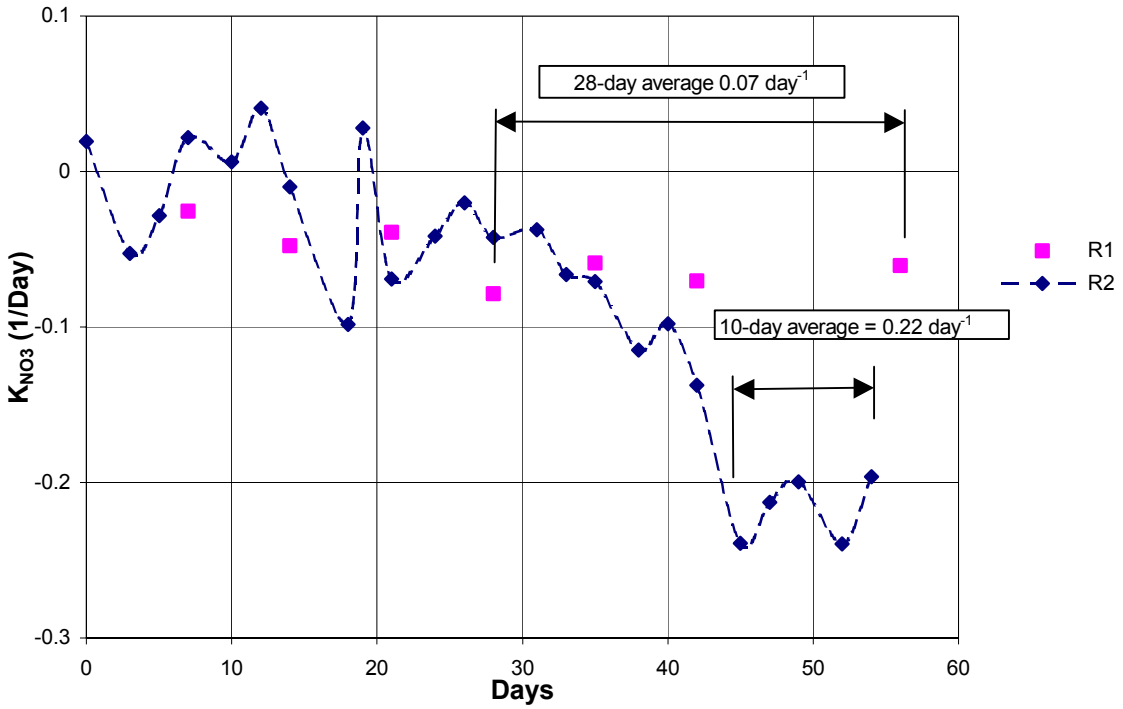

Figure 4-13. Time series of sixth experiment denitrification-rate constants (K_{NO3}) in the first (R1) and second (R2) reactors.

In contrast to the denitrification-rate constants observed in Reactor 1, the constants in Reactor 2 displayed more variability under aerobic conditions, leaving some uncertainty about the fate of the nitrate prior to the onset of anoxia. After the oxygen had disappeared from both reactors, however, the denitrification-rate constants in both reactors remained negative, suggesting that nitrate was being removed from the water column in each. In Reactor 1, the values decreased to a slightly higher negative number, and then remained stable. The increase may be attributed to biological denitrification, in addition to the possible chemical reduction of nitrate, which will be discussed later. For Reactor 1, the average denitrification-rate constant from Day 28 until the feed water change was -0.07 day^{-1}. The constants of Reactor 2 gradually decreased to a greater negative value than found in Reactor 1, and then became more stable after Day 45. The average denitrification-rate constant from Day 45 until the feed water change was -0.22 day^{-1}. Again, the magnitude of the constants increased going down the reactor series.

The increase in denitrification-rate constants in reactors further down the series in Experiments 4 and 6 could be attributed to the effect of oxygen in the feed water. Most of the heterotrophic electron-acceptor requirement in Reactor 1 may have been satisfied by the oxygen in the feed water, resulting in apparently lower denitrification rates than in Reactor 2. In addition, it is possible that due to short-circuiting, some of the oxygen in the feed water may have passed on to Reactor 2. This oxygen would reduce the electron-acceptor demand in Reactor 2, and would explain why, in Experiment 4, the denitrification-rate constant in Reactor 2 was of a greater magnitude than that of Reactor 3.

In order to use the microcosm systems to measure denitrification rates that are likely to be encountered *in situ*, it must be considered that the reservoir behaves more like a plug-flow reactor, or an infinite series of CSTRs. During the summer stratification, once hypolimnetic oxygen is depleted at any point in the reservoir, the sections downstream will not receive any additional oxygen supply in the liquid flow. Hence, the reservoir location where the oxygen concentration is observed to be zero should behave more like the second or third reactors in the experiments conducted for this study. Furthermore, the sixth experiment was operated in a manner closest to the actual condition of the reservoir, unlike the fourth experiment, in which an unrealistic amount of phosphorus was added to the system (0.5 mg/L as P). As a result, the second reactor of the sixth experiment may be expected to be the best simulation of the hypolimnion in the upper Bull Run arm of the Occoquan Reservoir, and in the opinion of the author, the observed rate in this reactor should be closest to the actual rate.

As noted earlier in this section, the 2nd reactor denitrification rate constant in Experiment 6 was observed to be 0.22 day^{-1}. This value is almost 50 percent higher than the highest of the recommended values (0.15 day^{-1}) for the CE-QUAL-W2 model (USACE WES, 2000). However, the value is within the range of values listed in *Rates, Constants and Kinetic Formulations in Surface Water Quality Modeling* (ERLORD, 1985): 0.0 – 1.0 day^{-1}. The discrepancies between the rate observed in this work and the values cited from other sources may be attributed, as suggested by Messer and Brezonik (1984), to the site-specificity of the denitrification phenomenon in reservoirs.

Chemical Reduction of Nitrate

During the course of this study, nitrate concentrations in some reactors were also observed to decrease under aerobic conditions. Because light was excluded from the reactors, photosynthetic uptake was assumed to be negligible. When oxygen was present in a reactor, it was also assumed that the microbially mediated denitrification rate was close to zero. Therefore the possible causes of the observed aerobic decreases were limited to either chemical transformation or some other microbial uptake. The aerobic decrease was observed in Experiment 6 and again in Experiment 7, which was a batch experiment that was conducted to confirm the earlier finding.

Experiment 6

The time series of rates of change of Experiment 6 nitrogen species in Figure 4-14 shows that a significant rate of nitrate concentration decrease occurred in the first reactor of Experiment 6 between Days 7 and 14. There was no significant observed rate of TKN concentration change over the same time span, indicating that the nitrate was not being incorporated into water column biomass *via* assimilatory nitrate reduction. Therefore, it is more likely that the loss of nitrate was due to a chemically-mediated reduction, or by some unknown microbial process in the sediment. The disappearance of nitrate in this manner was not observed in the earlier experiments or even in the second reactor of Experiment 6. In fact, the rate of change of nitrate concentration in the second reactor was observed to be positive from day 7 - 12, presumably due to nitrification, as may be seen in Figure 4-15. Measurements of DO in the second reactor were not begun until Day 18, and oxygen was not depleted until approximately Day 20. Because the positive rate of nitrate concentration change was not accompanied by a comparable rate of loss of water column ammonium or TKN, it is likely that the nitrification might have occurred near the sediment surface utilizing reduced nitrogen forms diffusing from the sediment.

Another unusual observation in the first reactor of Experiment 6, which may be linked to the chemical transformation of nitrate, is that manganese was released from the sediment on Day 20, shortly after the disappearance of DO from the water column on approximately Day 14, and while nitrate still persisted. The manganese release might actually have followed more

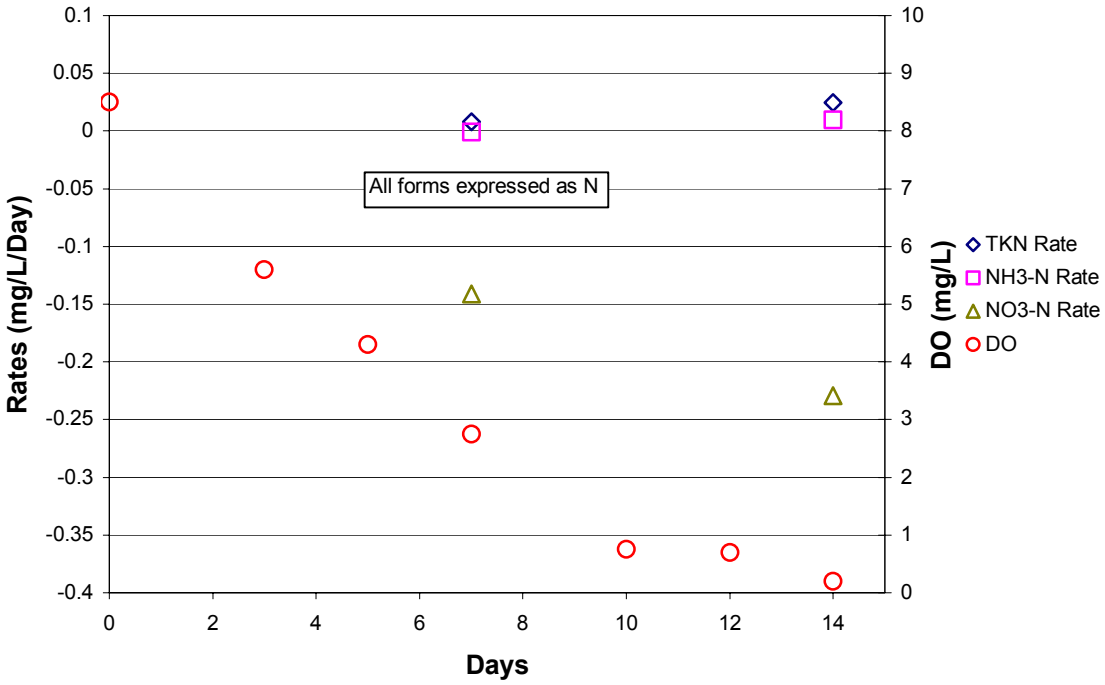

Figure 4-14. Time series of the rates of nitrogen transformations and dissolved oxygen depletion in the first reactor of Experiment 6.

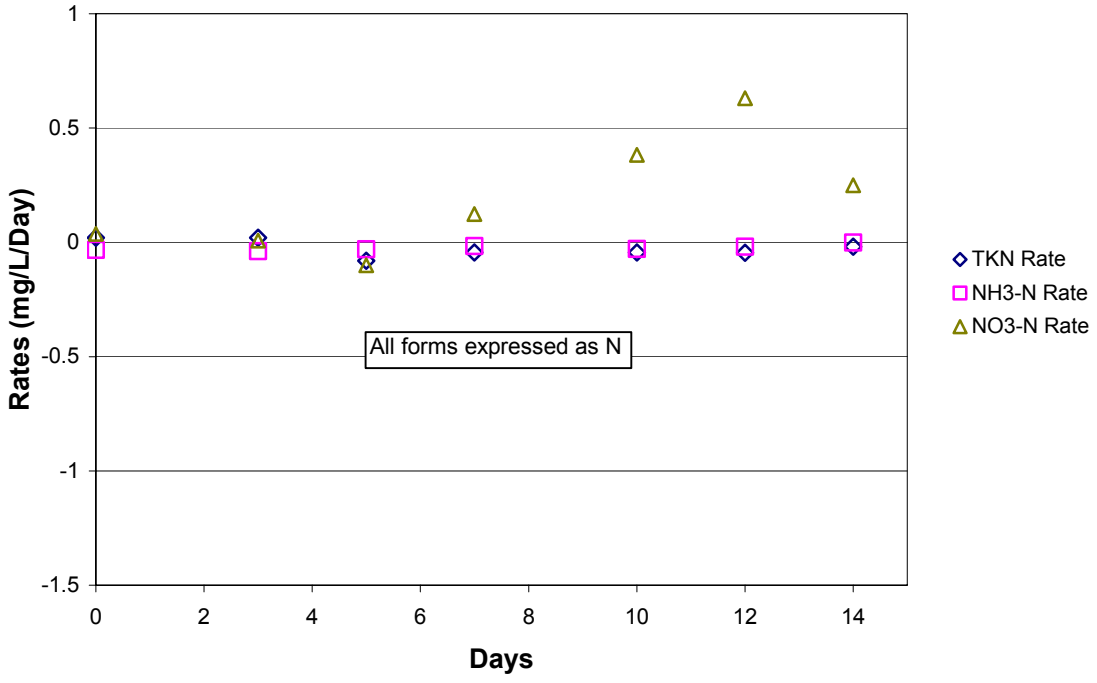

Figure 4-15. Time series of the rates of nitrogen transformations in the second reactor of Experiment 6.

closely the depletion of oxygen, however, the concentration was not measured until Day 20, thereby making it difficult to identify the exact point when the release began. The time series is illustrated in Figure 4-16. Normally, the release of sediment manganese has been reported as following the completion of nitrate reduction in the water column (Sherman, 1983; and Song and Muller, 1999), which is consistent with the fact that microbial oxidations with manganese as the terminal electron acceptor (TEA) yield less energy than with nitrate as the TEA (Stumm and Morgan, 1996).

Luther *et al.* (1997) found that reactive (non-structural) manganese on surface sediments could chemically oxidize organic-N and ammonia to nitrogen gas and that biological mediation could enhance this reaction. The reactive manganese on a sediment surface is a result of upward migration of dissolved manganese, and re-precipitation of particulate manganese at the sediment surface (Calvert and Price, 1977). Luther *et al.* also postulated that manganese oxides act as a catalyst in the oxidation of organic matter by oxygen. Sequentially, manganese oxides oxidize organic matter, and then the reduced manganese is re-oxidized by

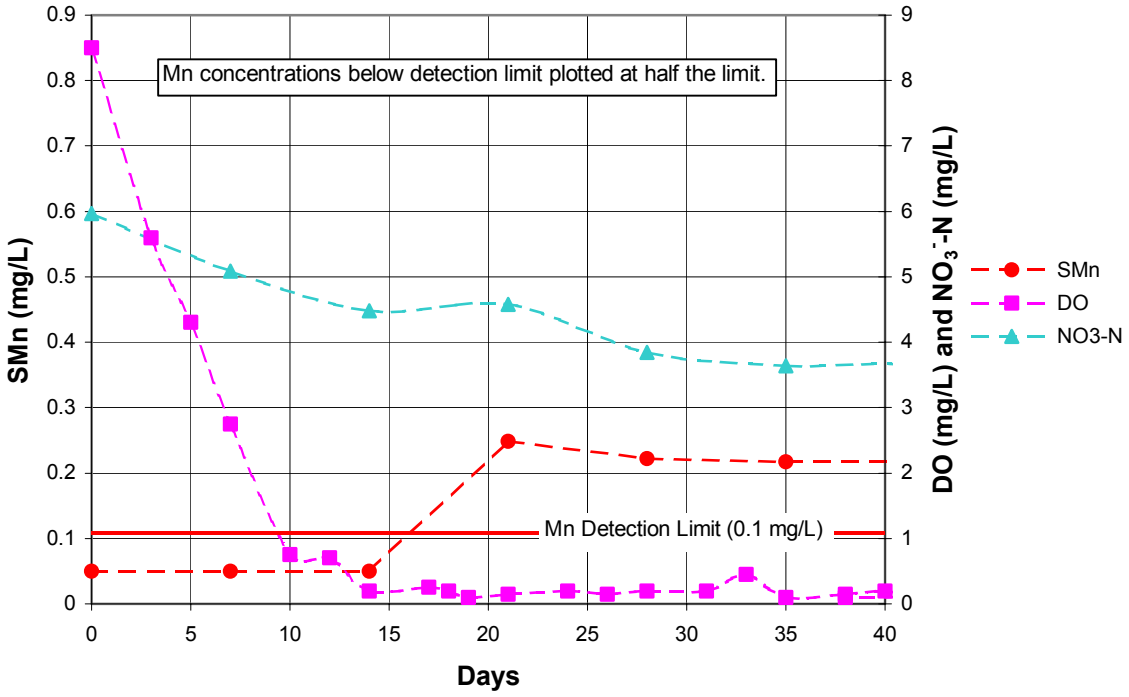

Figure 4-16. Time series of the sixth experiment soluble manganese (SMn), dissolved oxygen (DO), and nitrate (NO_3^-) in Reactor 1.

molecular oxygen, generating reactive manganese to continue the oxidation of organic matter. Therefore, if no oxygen is present, reduced manganese would be expected to be released to solution directly from the sediments, which was consistent with the observations of the present study.

Luther *et al.* also postulated that reduced manganese could chemically reduce nitrate to nitrogen gas, even in the presence of oxygen. This is a possible explanation for the decrease in nitrate concentrations observed in Reactor 1 Experiment 6 prior to the depletion of oxygen. Overall, the results of their study showed that sediments enriched with manganese were a sink for nitrate, while sediments low in manganese served as a source. They noted that this could be the reason why denitrification was sometimes observed in systems not devoid of oxygen.

The question remains as to why the apparent loss of nitrate in the presence of dissolved oxygen occurred only in the first reactor of Experiment 6 and not in the second reactor, or in any other reactors in all prior experiments. The major difference between the first reactor of Experiment 6 and the other reactors was that the former contained sediment taken from an upstream site near the reservoir tail waters. Unlike the lower part of the reservoir, the bottom waters in the upper reach generally remain aerobic throughout the year. In such a situation, it is possible that the reduced manganese diffusing to the sediment-water interface is rapidly re-oxidized, precipitates, and is re-incorporated into the surficial sediments. If this is the case, the amount of reactive manganese in the sediments of the upper reservoir would be expected to accumulate over time. In contrast, the reactive manganese in the lower reaches of the reservoir that might accumulate over the fall, winter, and spring seasons may be released to the water column during the summer period of anoxia. Some fraction of the released manganese in the lower reservoir would not be expected to re-precipitate onto the sediment surface following the introduction of oxygen at the fall circulation, because of prior removal from the reservoir by outflow, abstraction for treatment, or release for power generation. This scenario would explain the lack of significant accumulation of manganese on the sediment surfaces in the lower reaches of the reservoir, and is also a possible explanation of the failure to observe the aerobic reduction of nitrate in microcosm experiments with sediments from these reaches.

Experiment Seven

Experiment 7 was conducted with a single microcosm configured as a continuously mixed batch reactor (CMBR). The purpose was to determine if the above pattern, that is, the reduction of nitrate under aerobic conditions followed by the release of manganese under anoxic conditions, could be repeated, thereby lending weight to the hypothesis described in the previous paragraph. The reactor was initially configured with a mixture of UOSA discharge and Bull Run water computed to give an initial nitrate concentration of approximately 8 mg/L as N. In order to maximize the possibility of observing a phenomenon that was likely to be affected by exposed sediment area, sediment was placed in the reactor so that the entire cross-section was covered. A total of 1.5 L of wet sediment was used, and the final depth was 6 cm. The reactor was sealed, and periodic measurements made to determine the DO condition, and to withdraw water samples.

Figure 4-17 shows a time series of the concentrations of nitrogen species and soluble manganese. The figure is also annotated to show zones where oxygen was present (aerobic),

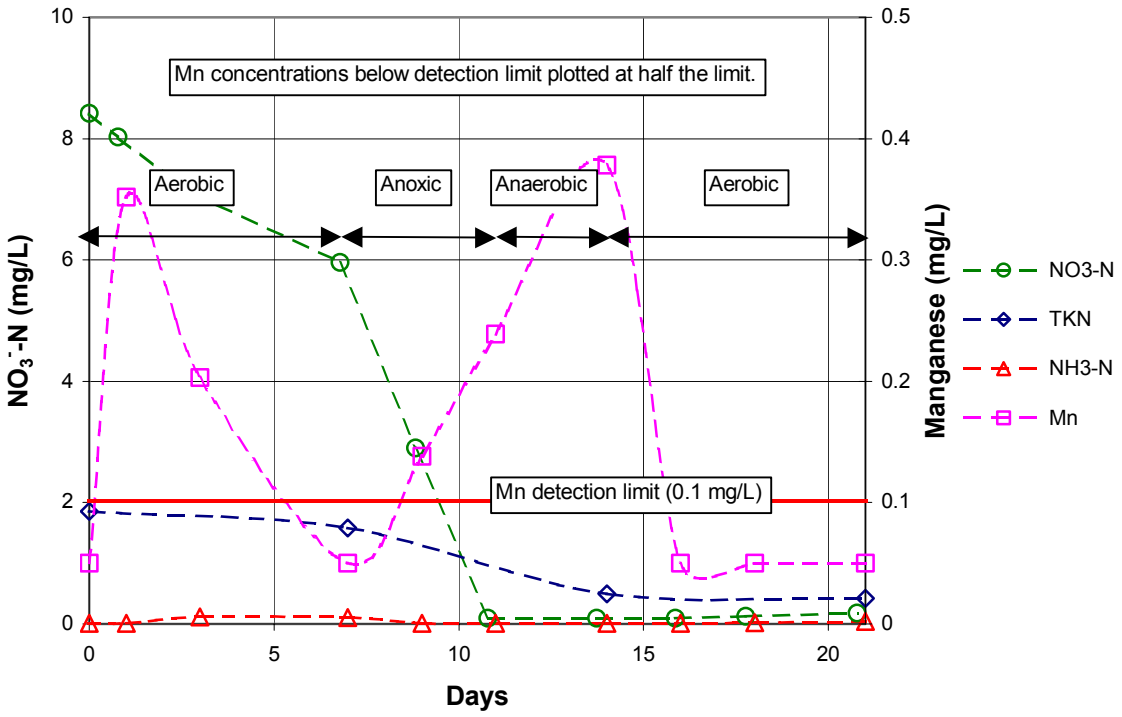

Figure 4-17. Time series of Experiment 7 nitrate (NO_3), total Kjeldahl nitrogen (TKN), ammonia (NH_3), and manganese (Mn) in the seventh experiment.

oxygen was excluded but nitrate was present (anoxic), and finally, where oxygen and nitrate were both absent (anaerobic).

During the aerobic period, approximately 25 percent of the nitrate (2.4 mg/L as N) was observed to disappear from solution, while TKN and ammonium remained at low, and nearly constant, concentrations. This could be explained by the nitrate diffusing into the sediment, or being reduced in the water column. No similar decrease was observed during the aerobic period of other experiments. Because the dissolved oxygen concentration remained above approximately 7.5 mg/L during the aerobic period, biologically mediated denitrification in the water column is an unlikely explanation for the nitrate loss. Chemical reduction by soluble manganese is another possible explanation, although under this scenario, nitrate would also compete with dissolved oxygen for electrons donated by reduced manganese species. In addition, stoichiometric calculations indicated that the observed loss of nitrate *via* chemical reduction would have required nearly 24 mg/L of manganese (II), which is vastly more than the observed water column manganese loss of 0.35 mg/L during the first aerobic period.

The exposed sediment surface area during Experiment 7 was greater by a factor of 4 - 8 than during prior experiments, which raises the possibility that increased area-based diffusion of nitrate into the sediment could be an explanation for the observed concentration decrease. As noted previously, the sediments used in Experiment 7 were obtained from the upper reservoir, specifically from a reach between Station ST40 (Bull Run at Yates Ford) and RE30 (Bull Run Marina) where anoxic conditions are rare. This has led to the previously-stated speculation that these sediments would be more likely to be enriched with reactive manganese. Luther *et al.* (1997) found that such sediments may serve as a sink for nitrate, while sediments not similarly enriched may serve as a source.

An additional note regarding the high soluble manganese concentration detected on Day 1 during the aerobic period is warranted. The presence of soluble manganese at the onset of an aerobic period was probably an artifact of anoxic pore water from the sediments mixing into the water column when the reactor sediment was disturbed during the filling process. The soluble manganese concentration on Day 0 was below the detection limit because the manganese sample was taken from the filling water, not directly from the reactor. Following the peak on day 1, the manganese concentrations decreased over time and declined below the

detection limit before the reactor was sealed on Day 7, most likely due to oxidation by either dissolved oxygen or nitrate present in the water column.

Further evidence of the reduction of reactive manganese on the sediment surface under aerobic conditions is provided by the reappearance of soluble manganese during the subsequent anoxic period. Once the reactor was sealed, oxygen depleted rapidly. Following the depletion of oxygen, soluble manganese concentrations were observed to increase in the water column, and ultimately reached a value similar to that observed immediately after filling the reactor. In the absence of oxygen, soluble manganese produced from the oxidation of organic matter on the sediment surface may be only partially re-oxidized by nitrates. The portion not oxidized would reach the water column as was observed during the anoxic phase of this experiment. A combination of biological denitrification and chemical reduction with manganese probably caused the nitrate to decrease at a higher rate than in the aerobic period. The nitrate was completely depleted by Day 11, and the manganese concentrations continued to increase under anaerobic conditions.

When oxygen was re-introduced to the reactor, the dissolved manganese was immediately oxidized and decreased to concentrations at or near the detection limit in the water column. Samples during this second aerobic phase were also analyzed for acid extracted manganese, and the concentrations were also at or below the detection limit. These results support the finding of Luther *et al.* (1997) that dissolved molecular oxygen may effectively oxidize reduced manganese.

Similar findings were reported by Gunnison *et al.* (1978) in their observation that manganese and nitrate were apparently reduced at the same time, providing evidence that microbial communities were able to use both species simultaneously. The manganese was added to a microcosm as an amorphous (nonstructural) manganese dioxide. Unfortunately, the investigators did not report the nitrate concentration under aerobic conditions. Otherwise, one could be more certain about the relationship.

Sorensen *et al.* (1987a) have speculated that some "chemo-denitrification" processes may be significant in the removal of nitrate in the water columns of perennially aerobic water bodies. The observed aerobic decrease of water column nitrate in Experiment 7 is difficult to

explain without the involvement of some sediment-based denitrification process, either biological or chemical. In the past, other researchers (Cavari and Phelps, 1977) attributed the removal of nitrate under aerobic conditions to denitrification in anaerobic microenvironments. However, near the end of the 1990's, Luther *et al.* (1997) stated that their microelectrode work did not support the existence of anaerobic microenvironments, and found any claims of the loss of nitrate under aerobic conditions to the existence of anaerobic microenvironments to be dubious.

Because the sediment manganese release under anoxic condition was repeatable, the existence of reactive manganese on the surface of sediments from the most upstream reservoir site appears to be certain. However, the longitudinal profile of reactive manganese in sediments from the Occoquan Reservoir, as shown in Figure 4-18, shows higher manganese content in the sediments from the downstream sites. It is possible that this anomaly could be a result of the failure of the acid extraction to fractionate the non-crystalline (reactive) manganese from the crystalline manganese. In future work, more useful information might be obtained from a sediment depth profile of manganese as made by Luther *et al.* (1997).

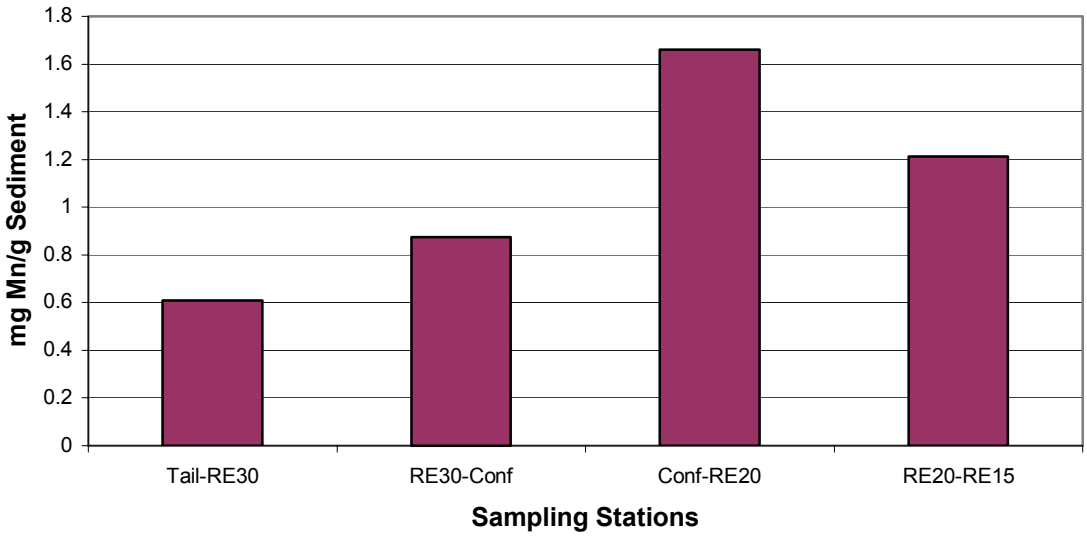

Figure 4-18. Profile of sediment reactive manganese along the Occoquan Reservoir from the tail waters on Bull Run arm to RE15.

Oxidation Reduction Potential *versus* Sediment-Water Interaction

Oxidation Reduction Potential (ORP) measurements were made in both the water column and at the sediment surface in all experiments. However, because of a number of problems with the measurement technique, as was previously discussed in the *Materials and Methods* section, the data from the five earliest experiments were determined to be unreliable and have been omitted from this discussion. Moreover, due to lack of chemistry data in the interstitial water, changes in sediment ORP observed in the study described herein cannot be interpreted without speculation on the fate of reduced and oxidized species in the sediment, and as a result the sediment ORP data have also been omitted from the discussion. The data have, nevertheless, been included in Appendix F in the interest of providing a complete record. The discussion in this section will be limited to the water column ORP data from Experiments 6 and 7.

Experiment 6

As previously described, Experiment 6 was conducted with a microcosm system configured with two-CSTRs-in-series. Figure 4-19 shows a time series of ORP in the water column in both reactors. The ORP in Reactor 1 consistently increased for a few days after the start of the experiment, and then the values began to decline, corresponding to the declining DO. A few days after the DO had been depleted on Day 14, the ORP decreased to the minimum recorded value of approximately -10 mV on Day 22. The minimum was followed by a recovery back to a value slightly above 300 mV by Day 40. Reference back to Figure 4-12 will show that, during that period of low ORP, the nitrate in Reactor 1 had not been depleted, and in fact, remained at a concentration only slightly below 4 mg/L as N. A number of researchers have found that the presence of nitrate would poise anoxic systems at a variety of ORP values, all of which are substantially well above the minimum value observed on Day 22:

- 400 - 450 mV (E_7) (Mortimer, 1941, 1942)
- 300 - 350 mV (E_7) (Greatz *et al.*, 1973)
- 200 mV (E_7) (Bell, 1969; Bailey and Beauchamp, 1970)

The minimum value observed in the present study between Days 20 and 30 was over 200 mV lower than any values from the sources cited above, which suggests that some other redox

couple was affecting the measurement even though nitrate-N still persisted in the water column.

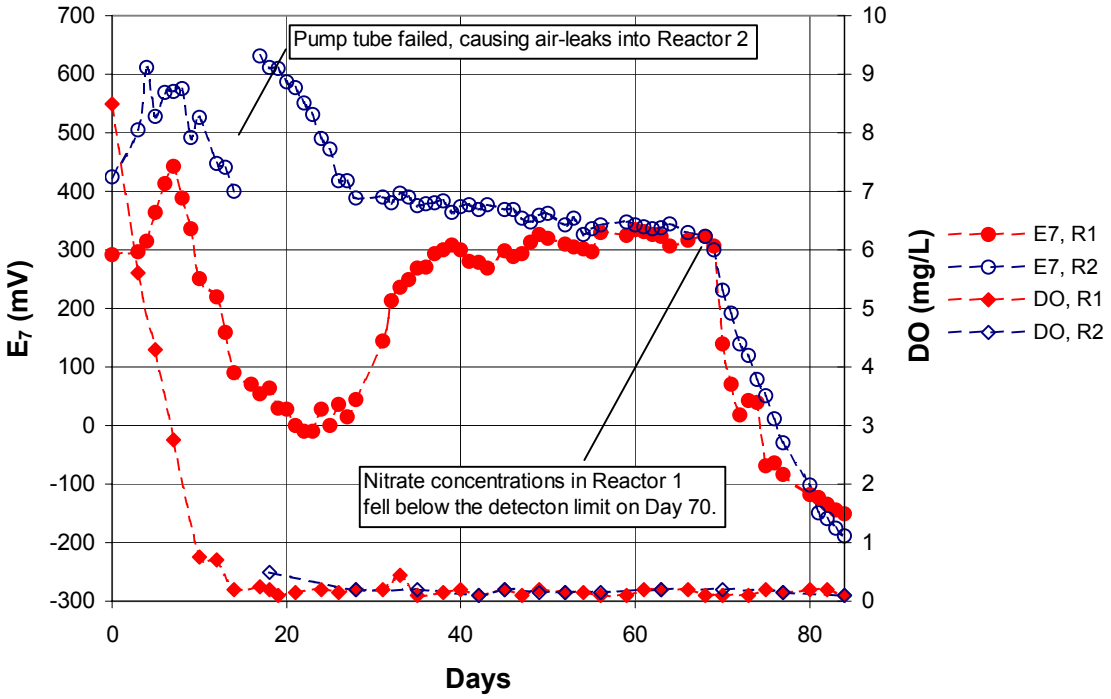

Figure 4-19. Time series of sixth experiment dissolved oxygen (DO) concentrations and E_7 in the water column of the first (R1) and second (R2) reactors.

Just prior to the ORP reaching the minimum observed value, dissolved manganese concentrations were observed to begin increasing in the water column, as may be seen in the time series presented in Figure 4-20. The manganese concentrations rose from below the detection limit of 0.1 mg/L to a value slightly above 0.3 mg/L even though nitrate still persisted, and did not begin to decline again until about Day 42. The concentration increase, as indicated in the previous section, could be attributed to the chemical reduction or microbially mediated reduction of previously precipitated reactive manganese on the sediment surface. Following the depletion of oxygen on Day 14, it is possible that nitrate alone could not re-oxidize all the reduced manganese diffusing from the sediment surface. This may be an artifact of differences in the kinetics of manganese oxidation by oxygen and nitrate, respectively.

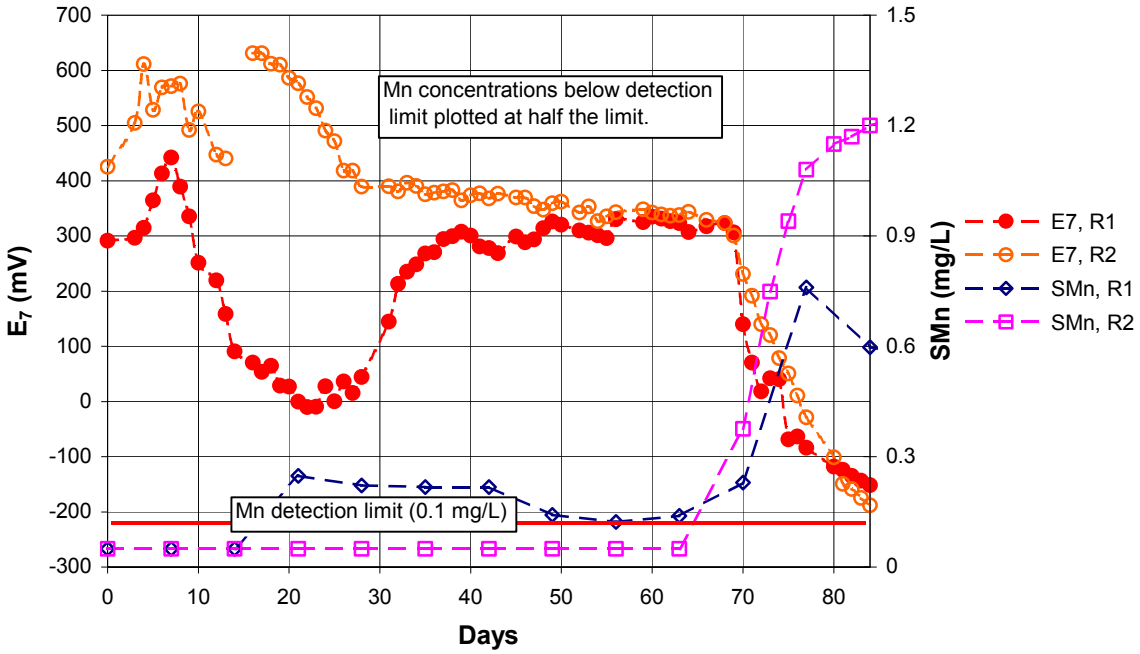

Figure 4-20. Time series of Experiment 6 E_7 and soluble manganese (SMn) in the water column of the first (R1) and second (R2) reactors.

Although there is an apparent contradiction between the existence of nitrate and the observed ORP negative values, one must consider the potential effect of redox couple(s) set up by soluble manganese entering the water column from the sediment. Following the depletion of molecular oxygen, even in the presence of nitrate, it is possible for the newly present manganese redox couples to have such an effect. This is because the electrochemical measurement of ORP is a mixed potential, which is essentially a weighted average of the potentials of all redox couples present in the system (Bohn, 1971). The weight of each couple depends on its exchange current, that is the rate of electron transfer between oxidized and reduced species (Bohn, 1971). Because manganese redox couple(s) are electroactive, while nitrate transformation is not (Snoeyink and Jenkins, 1980), the exchange currents of the manganese redox couple(s) may be expected to exert a much greater influence on measured ORP than those associated with nitrate transformation. Therefore, the measured potential may basically be determined by the potential of the manganese redox couple(s) present in the system, which, as suggested by Sigg (2000), are lower than those for nitrate transformation.

It may be possible to explain the increase in redox potential between Day 22 and Day 40 by the oxidation of reduced manganese, which diminished the amount of reduced

manganese and increased the amount of oxidized manganese. According to the Nernst equation, the redox potential will increase if the ratio of oxidized to reduced manganese increases. It is not known if the oxidation of manganese was caused by nitrate, as proposed by Luther *et al.* (1997), or if it was a biologically-mediated system where, under anoxic conditions, autotrophic organisms employed the reduced manganese as an electron donor, and the nitrate as a terminal electron acceptor. Either mechanism would not only result in an increase in the concentration of oxides of manganese, but also a reduction in NO_3^-. Although oxides of manganese are known to be insoluble (Weast, 1970), it is possible that oxidation of manganese with nitrate may have Mn(III) as an intermediate (Kostka, 1995; and Yv, 1994). The intermediate Mn(III) may complex with dissolved organic matter (Luther, 1994), such as humic and fulvic acids to form soluble manganese species (Snoeyink and Jenkins, 1980; McGhee, 1991). If this occurs, it is possible that an oxidation of reduced manganese with nitrate may result in a higher ratio of soluble oxidized to reduced manganese.

As a result of whatever reduction processes occurred, by approximately Day 70, the nitrate in Reactor 1 was depleted, and was immediately followed by a rapid decline in the ORP in Reactor 1, which continued until the end of the experiment. The value at the end of the experiment was -152 mV. This trend has been commonly observed by many researchers (Mortimer, 1941 and 1942; Sherman, 1983, Greatz *et al.*, 1973; Bell, 1969; Bailey and Beauchamp, 1970) as a system shifts, sequentially, from denitrification to manganese reduction, to iron reduction, to sulfate reduction, to methane production, and to fermentation (Stumm and Morgan, 1996). The increases in soluble manganese and iron concentrations as shown in Figure 4-20 and 5-21, respectively, during the final ORP decrease support the proposition that manganese and iron were being reduced.

During Experiment 6, the water column ORP in Reactor 2 exhibited a similar pattern to that observed in Reactor 1, as also may be seen in the time series data presented in Figure 4-19. The ORP displayed an initial increase, followed by a decrease, just as was observed in Reactor 1, and which may be explained in the same manner. The DO in Reactor 2 was not measured until Day 18 because, during the early stage of the experiment, it was suspected that the DO measurement could introduce a significant amount of oxygen into the reactor. After a protocol was worked out to prevent introduction of atmospheric oxygen, DO measurements

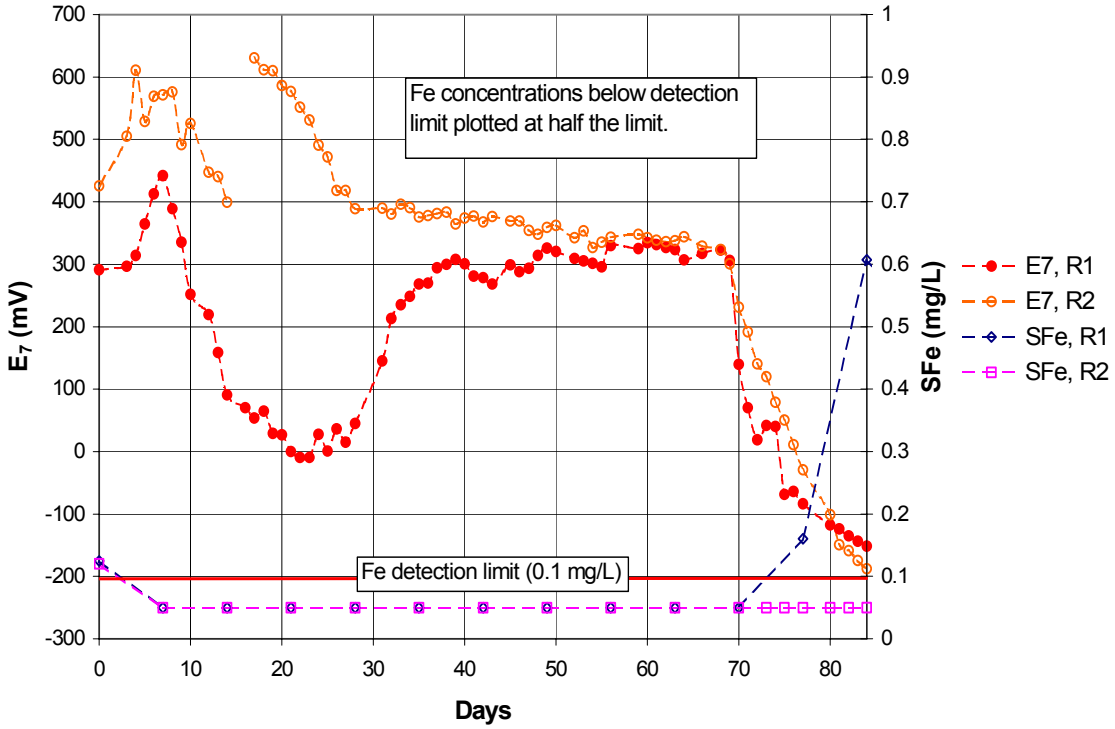

Figure 4-21. Time series of Experiment 6 E_7 and soluble iron (SFe) in the solution of first reactor (R1), and second (R2) reactors.

were resumed. On Day 16, a peristaltic pump for the Reactor 2 effluent failed, and the reactor contents were siphoned out at a rate greater than the inflow rate, creating a void space, along with a negative pressure head, which resulted in ambient air being sucked into the reactor. No water column measurement of ORP was possible on that day, because the water level was lower than the tip of the reference electrode.

After re-setting the reactor volume, on the following day the ORP responded to the introduction of oxygen with a rapid increase, followed by a resumption of the decline previously observed (along with DO). The DO in Reactor 2 was depleted by Day 28. Following the disappearance of oxygen, the ORP in Reactor 2 did not exhibit the same pattern observed in Reactor 1. The values declined slowly from about 400 to 300 mV, and then decreased more rapidly when the effluent nitrate concentrations from Reactor 1 fell below the detection limit (0.1 mg/L NO_3^--N). The value at the end of experiment was –188 mV.

Unlike Reactor 1, as described in the previous section, the sediment in Reactor 2 was obtained from a reservoir reach that probably did not contain reactive manganese, and

manganese was not released from the sediment during the anoxic period between Days 28 and 70. In a study of ORP and sediment water interactions with a batch reactor containing sediment from the Occoquan Reservoir, Sherman (1983) did not observe release of manganese under anoxic conditions. It is possible that this was because the sediments used were not enriched with newly precipitated manganese. For the sediments used in her study, Sherman observed that nitrate poised the water column ORP at a level that prevented reduction of manganese and iron at the sediment surface until nitrate was depleted.

This may be similar to the situation observed in the second reactor of Experiment 6. The poising effect of nitrate has been observed by many other investigators, and the values have been given earlier in this section. Furthermore, it should be noted that the ORP in Reactor 2 did not respond to the disappearance of nitrate, indicating that the small amount of nitrate coming from Reactor 1 was sufficient to poise the ORP in Reactor 2.

As observed in Reactor 1, after the nitrate in both reactors was completely depleted by denitrification and/or the oxidation of manganese, an accelerated decrease of reactor ORP commenced and continued until the end of the experiment as shown in Figure 4.21. However, in contrast to Reactor 1, soluble iron was not observed during this final ORP decrease. This may be an artifact of higher available manganese in the sediment of Reactor 2 than that in Reactor 1. Figure 4-20 shows that more manganese was released from the sediment in Reactor 2 than from the sediment in Reactor 1. It has been established that sediment iron will not serve as an electron acceptor until all available manganese is exhausted (Stumm and Morgan, 1996). The continuing increase in the manganese concentrations in Reactor 2 from Day 70 until the end of the experiment suggests that the available sediment manganese had not been depleted, thereby delaying the sediment iron reduction. In Reactor 1, on the other hand, the increase in the reactor iron concentrations coincided with the decrease in the manganese concentrations, which is a sign that all the available sediment manganese had been utilized.

Experiment 7

Because the manganese domination over ORP observed in Reactor 1 of Experiment 6 was so unusual, another experiment was designed to insure that the phenomenon was reproducible. Experiment 7 was conducted with a batch reactor containing sediment from the site where it was speculated that the sediment was enriched with reactive manganese.

Descriptions of the experiment have been given in the *Materials and Methods* section. The aerobic-anoxic-anaerobic sequence for Experiment 7 was decribed previously in this section and may be seen in Figures 4-17 and 4-22.

Once the reactor was sealed from the atmosphere, the redox potential declined rapidly to the minimum observed value and did not decrease further, even when nitrate was depleted on Day 11 as shown in Figure 4-22. This pattern seems to suggest that nitrate did not retard the ORP decrease. The redox potential did not swing back as had been observed in Experiment 6 because there was no nitrate remaining to oxidize reduced manganese, which was increasing over time as shown in Figure 4-23. As in Reactor 2 of Experiment 6, the soluble manganese concentrations were still increasing at the end of the anaerobic phase, providing additional evidence that the sediment manganese supply had not been depleted. Consequently, sediment iron had not yet been reduced and no soluble iron was observed in the water column, as shown in Figure 4-23. Following the re-introduction of oxygen into the reactor, the ORP increased immediately to approximately the values observed during the first aerobic phase. The dissolved manganese was re-oxidized and disappeared within 2 days.

Gunninson *et al.* (1978) observed a comparable decrease in oxidation-reduction potential without any retardation by nitrate. They added manganese to their system in form of an amorphous manganese oxide, which is non-structural and reactive. Once they stopped the supply of oxygen, the oxygen concentrations depleted rapidly to zero, followed by the reduction of both nitrate and manganese, as evidenced by the detection of soluble manganese in the water column. The ORP was found to decrease to the minimum observed value with no sign of retardation by nitrate. This supports the finding of the present study that the ORP poising effect of nitrate could not be observed in the presence of certain reduced species such as manganese.

Due to the mixed potential nature of electrochemical ORP, quantitative interpretation is difficult (Stumm and Morgan, 1996). Nonetheless, ORP measurements may be useful in studies of aquatic systems if one uses it as an "operational"

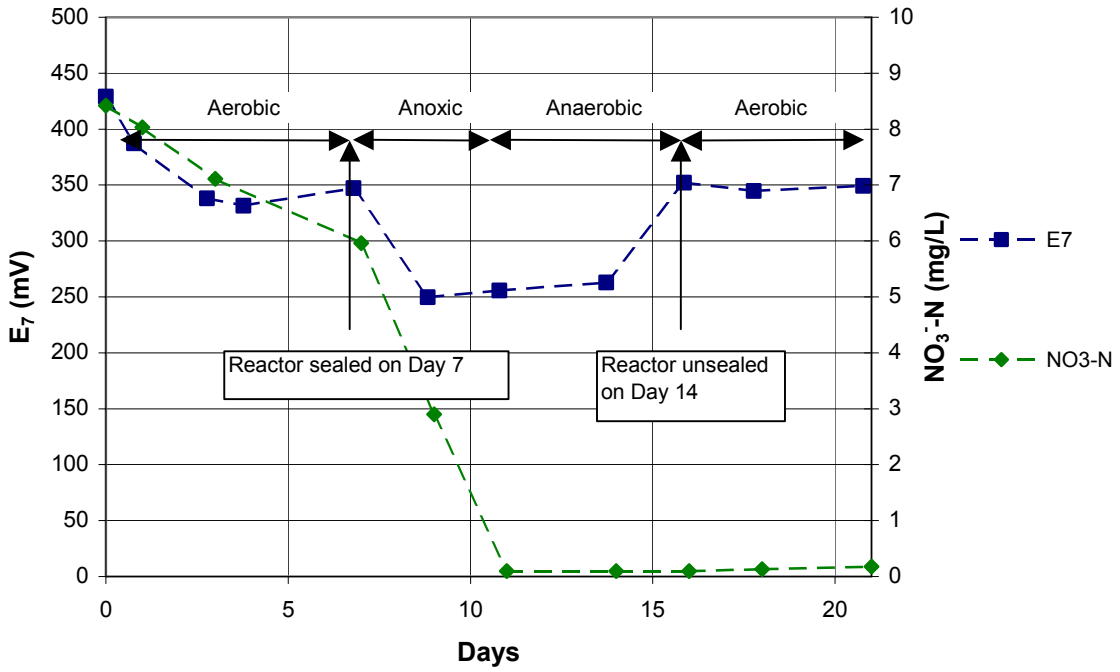

Figure 4-22. Time series of Experiment 7 E_7 and nitrate (NO_3^-) concentrations in the solution.

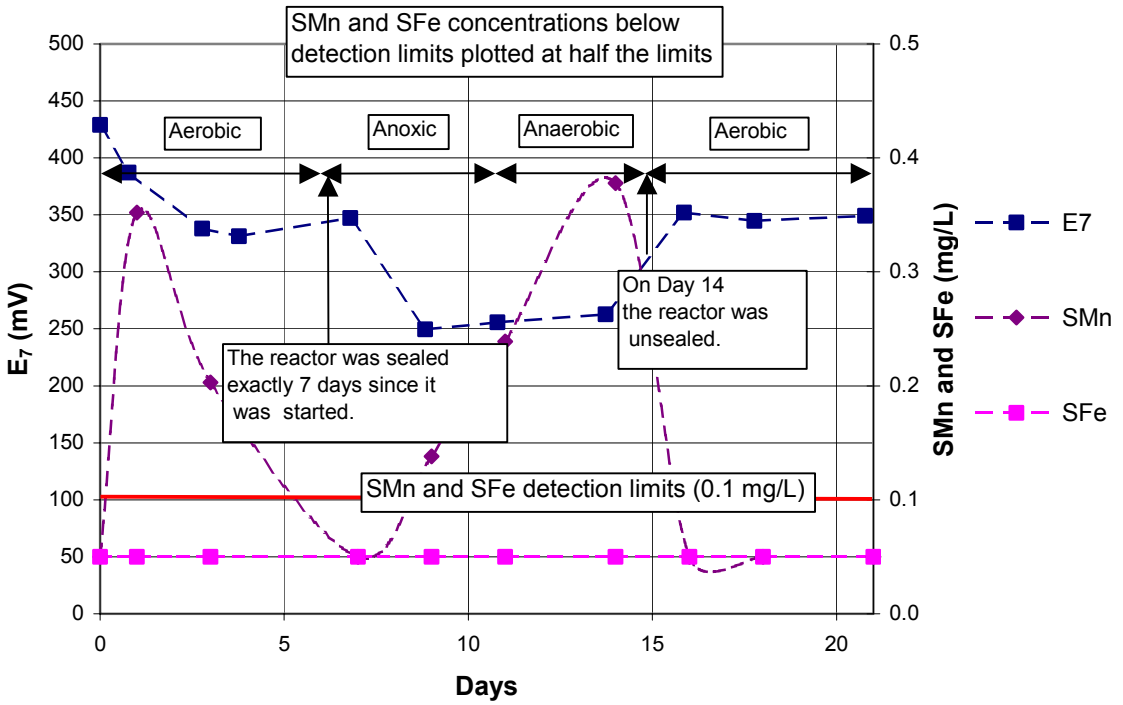

Figure 4-23. Time series of Experiment 7 E_7, soluble manganese (SMn), and soluble iron (SFe) in the reactor solution.

parameter as suggested by Whitefield (1969) and Hargrave (1972). Whitefield (1969) recommended using E_h as an indicator of the degree of stagnation in a water body, while Hargrave (1972) recommended using it to separate between reduced and oxidized sediments. In the present study, the E_h served to reveal the probable existence of reactive manganese, which might have been ignored had not the absence of the nitrate poising effect on ORP been noted.

However, it should be noted that judgments based on E_h for a particular system should be made considering prior knowledge about E_h of the same system. For example, in the present study, the potential in Reactor 2 of Experiment 6 was poised by nitrate at approximately 330 - 400 mV (E_7). Other investigators reported the poising values ranging from 200 - 450 mV (E_7) (Mortimer, 1941, 1942; Bell, 1969; Bailey and Beauchamp, 1970; and Greatz *et al.*, 1973). Because the reported ranges are quite wide, they may overlap with ranges where other oxidants have been observed to be active:

- 300 – 630 mV (E_7) for O_2 (from the present study)
- 200 - 300 mV (E_7) for ferric complexes (Mortimer, 1941, 1942)

As a result, predicting the predominating redox couple(s) from ORP measurement only may not be possible, or may be applicable only on the system from which the ranges were obtained. For example, the ORP attained during Experiment 7 in the first aerobic phase was the same as during the second aerobic phase.

Nitrate and Water Quality

Although nitrate is listed in the national primary drinking water standards as one of the inorganic contaminants (USEPA, 2001) for which a maximum contaminant level (MCL) in finished drinking water has been assigned (10 mg/L as N), it has proven quite beneficial in improving water quality conditions in the Occoquan Reservoir (OWML, 1997).

Under the anaerobic conditions commonly experienced in the bottom waters of deep reservoirs during thermal stratification, phosphorus is often released from the deposited sediments (To, 1974; McLaughlin, 1981; Holdren and Armstrong, 1980; and Patrick and Kjalid, 1974). Because phosphorus is also usually the primary limiting algal nutrient in fresh waters (Lee *et al.*, 1978), it is of great concern to those charged with the management of water supply reservoirs. Sherman (1983) found that nitrate addition could rapidly raise the oxidation-reduction potential (ORP) of an anaerobic system to a point where oxidizing conditions could be maintained, and only a small amount of nitrate (0.2 mg/L as N) would be required to prevent the release of phosphate from bottom sediments. Ripl (1976) successfully reduced phosphorus concentrations in the bottom water of Lake Lillesjon in Sweden by mixing calcium nitrate with the lake sediment. Tiren and Pettersson (1985) concluded from their laboratory experiments and reviews of other investigators' results that the addition of nitrate to lakes would maintain high redox potentials at the sediment surface, and thus prevent, decrease, or delay the release of iron-bound phosphorus.

Sediment phosphorus release at the rates reported by McLaughlin (1981) was not observed in this study, but this may be related to the somewhat more realistic microcosm system design and operation during the current experiments. McLaughlin operated the microcosm in a batch mode, and also configured the reactor with sediment over the entire bottom, which resulted in a sediment surface of 286 sq.cm., which was about 3.8 times the sediment area exposed in the microcosm for Experiment 6 of the present study. In addition, the computed values of effective depth (water volume:sediment area) were 0.31 m and 1.27 m for the study by McLaughlin, and in the present study, respectively. Operationally, this provided for a larger, and more realistic, water volume per unit sediment area in the present study, thereby reducing the observed water column phosphorus concentrations. Finally, because the earlier experiments by McLaughlin were operated in a batch mode instead of

continuous flow, all the phosphorus liberated from the sediments during anaerobic conditions was retained in the reactor, and contributed cumulatively to the concentrations measured over the duration of the experiment. Even if one assumes that the actual sediment release rates are similar to those observed by McLaughlin (1981), each of these differences in experimental design would result in substantially lower water column concentrations of phosphorus being observed in the present study.

Figure 4-24 shows a time series of water column nitrate, phosphorus, soluble iron, and soluble manganese in Reactor 1 of Experiment 6. As may be seen, following the depletion of nitrate, the metals concentrations increased rapidly in the water column as they were reduced to the soluble Mn^{2+} and Fe^{2+} forms and liberated from the sediments. The lack of observed phosphorus release is puzzling, particularly with the well-established understanding of the release of sediment phosphorus under anaerobic conditions. It is possible that phosphorus was actually released, but that the resulting concentrations remained below the detection limit. This may be shown to be plausible if one assumes rates of total and orthophosphate phosphorus release about 4 times lower than the rate of 8.7 $mg/m^2/d$ reported by McLaughlin (1981).

Using the Experiment 6 reactor set-up and flow rates, one may determine the total phosphorus release rate that would have resulted in a steady state water column concentration equal to the analytical detection limit of 0.01 mg/L as P. The release rate was computed to be 1.9 $mg/m^2/d$, which is a factor 4.6 less than the rate observed by McLaughlin (1981). The predicted time series plots, for both cases, which were based on an assumed zero initial concentration and no other phosphorus sources or sinks, are shown in Figure 4-25.

Such an observed decrease in the phosphorus release rate would be consistent with a prediction made by To (1974) during an earlier study of the Occoquan Reservoir. He concluded that release of existing sediment phosphorus, coupled with better removal of sources in the watershed, such as wastewater and stormwater runoff, would eventually result in an overall reduction of the sediment as a source.

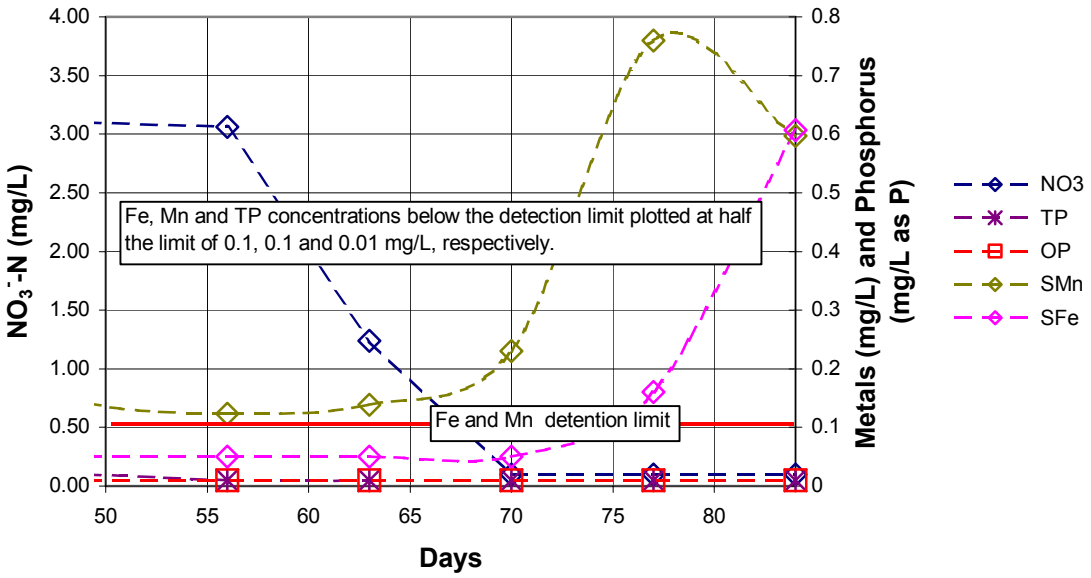

Figure 4-24. Time series of nitrate (NO_3^--N), total phosphorus (TP), orthophosphate phosphorus (OP), soluble manganese (SMn), and soluble iron (SFe) in Reactor 1 of Experiment 6.

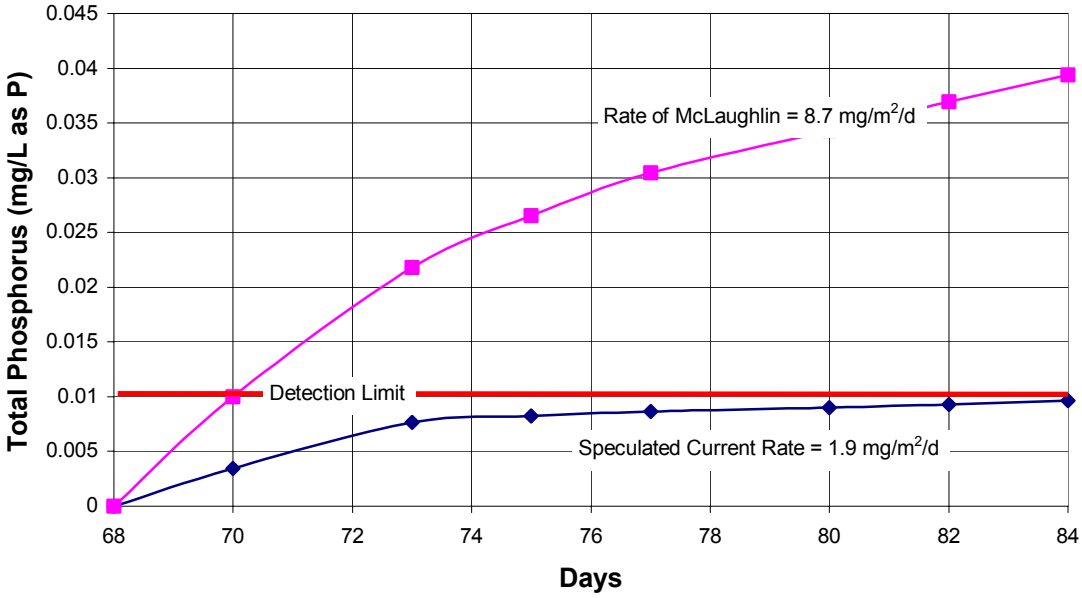

Figure 4-25. Paired time series of predicted total phosphorus water column concentrations using the release rate reported by McLaughlin (1981) and the speculated current release rate.

In spite of the predictions, however, OWML monitoring data show that the phosphorus concentrations in the bottom waters of the reservoir have not changed appreciably, with the exception of expected seasonal variations, from 1982 until the present time. A time series plot illustrating this conclusion is shown in Figure 4-26. The slope of linear regression line of RE02 data was found to be not significantly different from zero, with a probability of 0.62 when the null hypothesis was that the slope was zero.

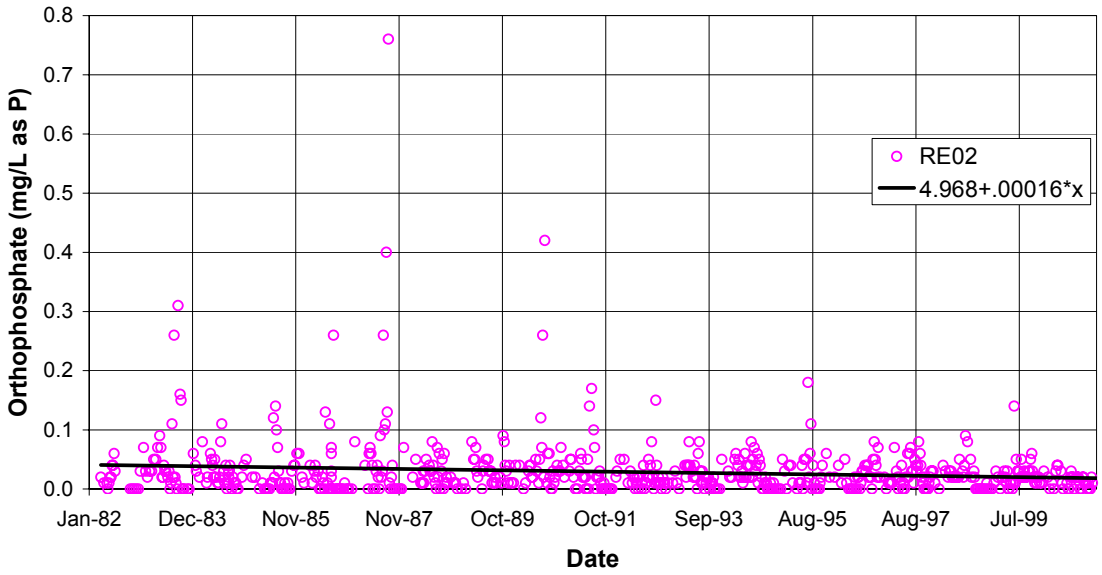

Figure 4-26. Historic orthophosphate data in the bottom waters of the reservoir at Station RE02. Source: OWML (2000b).

Moreover, release of phosphorus in summer was observed in the reservoir at RE02 in 1998, when oxidized nitrogen concentrations were low, as shown on Figure 4-27. In addition, observed concentrations were lower in 1999 and 2000 when the oxidized nitrogen concentrations were relatively high. This is further evidence of the effectiveness of nitrate, the dominant form of oxidized nitrogen, in retarding sediment phosphorus release.

Release rates observed in the current experiments might also have been affected because the sediments used were obtained from the upper reaches of the reservoir, and may have been depleted of phosphorus prior to retrieval. Some insight may be obtained if the monitoring data are examined from the stations near the sediment sampling locations.

The *in situ* total phosphorus concentrations were not high at RE30, possibly due to the presence of high oxidized nitrogen concentrations as shown in Figure 4-28. However, high

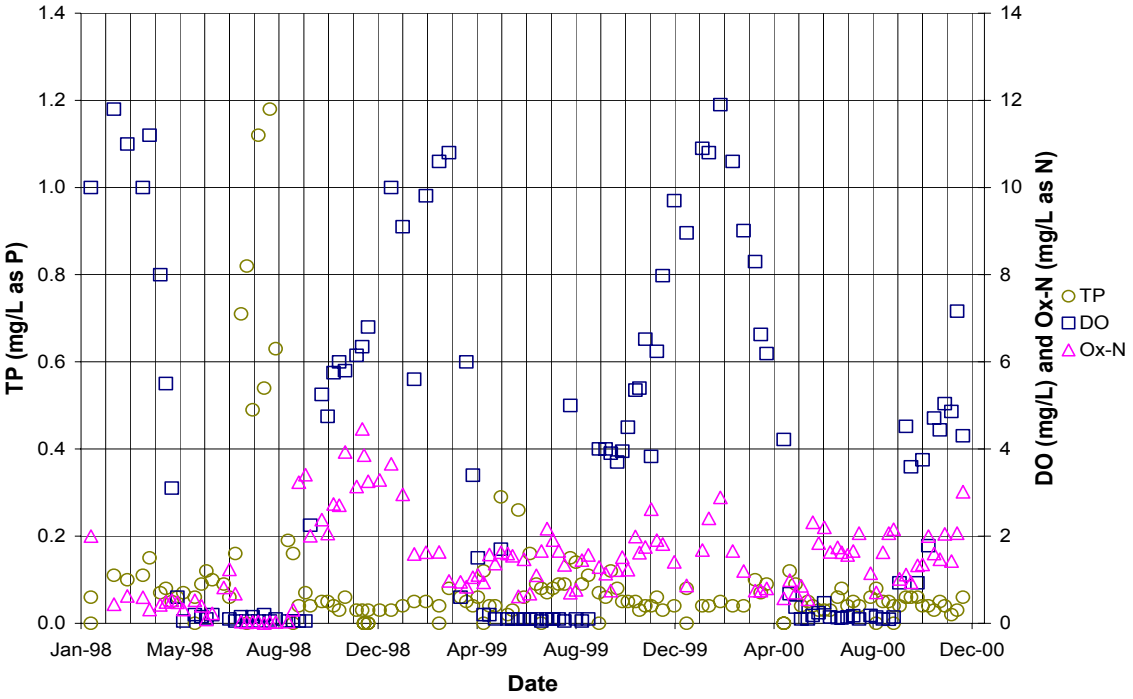

Figure 4-27. Time series of total phosphorus (TP), DO, and oxidized nitrogen (Ox-N) in the bottom waters at RE02 from 1998 to 2000. Source: OWML (2000b).

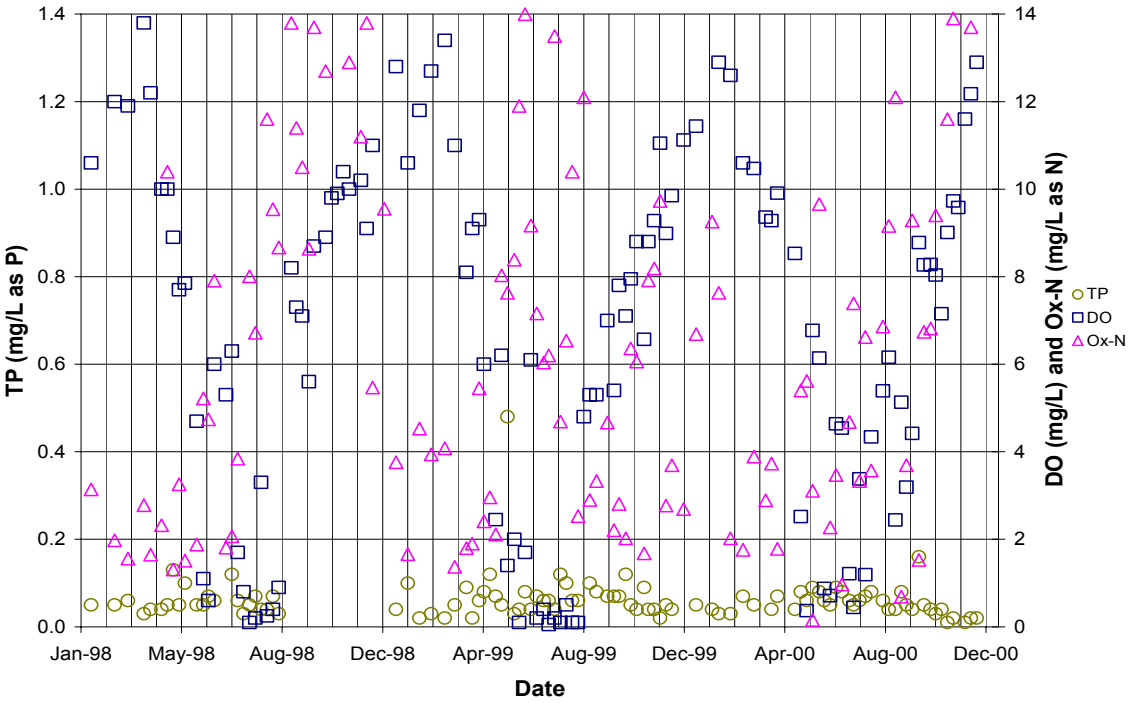

Figure 4-28. Time series of total phosphorus (TP), DO, and oxidized nitrogen (Ox-N) in the bottom waters at RE30 from 1998 to 2000. Source: OWML (2000b).

bottom water phosphorus concentrations were observed at RE15 in 1999, as shown in Figure 4-29.

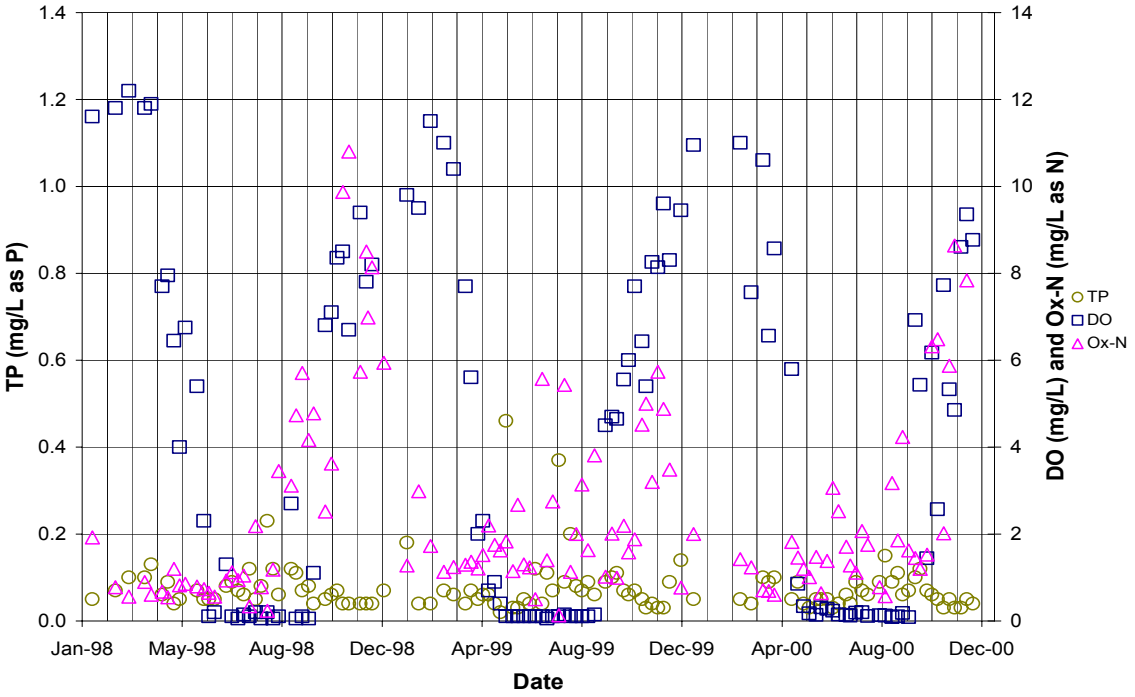

Figure 4-29.Time series of total phosphorus (TP), DO, and oxidized nitrogen (Ox-N) in the bottom waters at RE15 from 1998 to 2000. Source: OWML (2000b).

OWML monitoring data and the findings of other investigators, as described above, suggest that phosphorus must have been released in Experiment 6 but that it was not detected. Assuming this to be the case, a possible experimental flaw might explain the absence. APHA (1998) recommended that samples containing low phosphorus concentrations should not be stored in plastic bottles, unless kept in a frozen state, because phosphates may sorb to the plastic. Even at the mean release rate reported by McLaughlin (1981), the maximum steady state water column total phosphorus concentrations that would be expected in a reactor operated as described in Experiment 6, would have been only 0.04 mg/L as P. Such a small amount of phosphorus may have sorbed to the Plexiglas reactor walls.

Holdren and Armstrong (1980) found that 90 percent of an orthophosphate phosphorus solution of 0.05 mg/L P could sorb to the walls of a Pyrex beaker within 4 days. In their experiment, the beaker was initially soaked in a 2 mg/L orthophosphate phosphorus solution for 3 days. One might expect an untreated plastic wall, as in the present study, to sorb

even more phosphorus. In fact, if such losses occur through sorption to the reactor walls, it is possible that release rates as high as those observed by McLaughlin (1981) could have been experienced without detectable phosphorus concentrations being observed in the water column.

Future work to quantify sediment phosphorus release should employ an experimental design that eliminates, or accounts for, sorption to the reactor walls. This is particularly true if the experiment is conducted with continuous flow systems that maintain more realistic hydraulics and constituent concentrations. It should be noted that phosphate sorption to the plastic sampling tube and syringe may most probably be neglected because the samples were exposed to them for a very short time before being frozen.

The current magnitude of sediment phosphorus release cannot be determined from the microcosm experiments conducted for this study. However, OWML *in situ* monitoring data show that sediment release continues to occur once oxygen and nitrate are depleted. The possibility of large amounts of stored phosphorus becoming available to algae in the reservoir water column should continue to be of great concern to water quality managers. Should sediment release occur up to the potential rates postulated by Sherman (1983) and McLaughlin (1981), a return to massive summer algal blooms might be envisioned. Should such bloom conditions again become commonplace, it is also expected that other related water quality impacts would be experienced, including loss of oxygen, fish kills, formation of trihalomethane precursor compounds, and increased taste and odor episodes at the water treatment works.

In addition to preventing sediment phosphorus release, nitrate may also retard the release of manganese and iron from the deposited sediments. Although these two species do not cause health problems in the concentration ranges likely to be experienced, they can cause taste and odor problems, as well as discolored finished water (McGhee, 1991).

Manganese release was found to be particularly significant in this study because the maximum concentration observed in Reactor 2 of Experiment 6 was more than 70 times the secondary maximum contaminant level (SMCL) of 0.05 mg/L set by EPA (USEPA, 2000). Experiencing such concentrations in the raw water withdrawn from the reservoir would result in additional treatment costs being incurred by FCWA.

Consequently, mandating UOSA to remove nitrogen, which would ultimately result in a reduced supply of nitrate being delivered to the Occoquan Reservoir, would very likely result in some combination of the water quality problems indicated above. The maintenance of a nitrate supply to the reservoir would appear to be a prudent mean of continuing to retard the release of phosphorus, iron, and manganese from the deposited sediments. Given that the principal source of nitrate is the reclaimed water discharge from UOSA, this mode of operation should continue, except under those conditions where adequate nitrate reduction is not likely to occur *in situ*. Previous observations (Grizzard, 2002) in the reservoir have shown that extended drought conditions during periods when the reservoir water column is well oxygenated may result in high nitrate concentrations penetrating further downstream to the vicinity of the raw water intake. Under such conditions, the UOSA water reclamation plant must be equipped to shift to a biological denitrification operational mode so that the drinking water MCL of 10 mg/L NO_3^--N is not exceeded.