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Publication Date
1996-08-01

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TECHNICAL COMPLETION REPORT

Project Number UCAL-WRC-W-837

August, 1996

University of California Water Resources Center

The research leading to this report was supported by the University of California Water Resources Center as part of Water Resources Center Project UCAL-WRC-W-837

ABSTRACT

Microbial Denitrification of Groundwater using Microporous Membranes
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Microbial denitrification, a frequently used and relatively inexpensive method of removing nitrate from wastewater, has been applied to the treatment of potable water supplies, on a limited scale, using packed bed reactors. However, two significant drawbacks exist in transferring wastewater denitrification technology to the treatment of domestic water supplies: (1) the water is intimately mixed with microbial cultures and (2) organic compounds must be supplied as an energy source to drive the denitrification reactions and residual organics can be a water quality problem. Process configurations used experimentally have included both packed beds and fluidized beds. Denitrifying microbial cultures have been supported on sand, ceramics, polymers, clay, alginate gel, and agar gel. Work with conventional support materials (sand, ceramics, polymers, clay) has been relatively straightforward in that the microbial cultures are grown on support surfaces and water containing nitrate is passed through the fixed or expanded/fluidized bed. Carbon and energy sources, nearly always organic compounds, are added to the water. Thus the problem outlined above - introduction of bacteria and organics - is characteristic of systems used to date.

The current work utilizes microporous membranes to separate the water being treated from the microorganisms carrying out the denitrification reactions. Nitrate passes through the 0.02 μm membrane pores by molecular diffusion. Water does not move through the pores and therefore contamination of the product water does not occur. Operation of microporous membrane systems can incorporate a biofilm on the reaction side of the membrane or utilize a suspended growth culture. Transport, and hence denitrification rates appear to be greater using suspended growth systems. In addition, suspended growth systems will have advantages in terms of minimization of biofouling of hollow fiber continuous flow units.

Measured nitrate diffusivities through the membrane pores was 3.5×10^{-6} cm^2/s for biofilm systems and 5.0×10^{-6} cm^2/s for suspended growth systems. Nitrate flux is dependent on the concentration gradient. Potential fluxes for concentration differences of 20 mg/L are in the range of 10 $g/m^2 \cdot day$.

KEY WORDS: Denitrification, Nitrate, Groundwater quality, Ground water remediation, Public health, Water quality, Water treatment

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INTRODUCTION

Human consumption of nitrate contaminated water may lead to health problems. Although nitrate toxicity to humans is not entirely understood, ingestion of high nitrate water and consequent reduction to nitrite in the gastrointestinal tract is known to produce methemoglobinemia or "blue baby syndrome" in infants [Shuval 1977] Additionally, an increased incidence of some forms of cancer due to consumption of high nitrate drinking water is reportedly being investigated, but no conclusive results have been reached [Weisenburger 1991, Crespi and Ramazzotti 1991]. The health problems associated with consumption of nitrate contaminated water have spurred the US Environmental Protection Agency (USEPA) and European Community to establish maximum drinking water contaminant levels (MCL) of 10 mg/L NO_3^--N[1] [Federal Register 1991] and 11.3 NO_3^--N [European Communities 1980], respectively.

Nitrate contamination is a nearly ubiquitous problem in groundwater supplies throughout the world. Nationally and worldwide, the problem of nitrate in municipal water supplies is severe with a significant fraction of ground waters currently used as municipal water supplies exceeding the maximum concentration limits [Anton et al. 1988, Bouchard et al. 1992, Power and Schepers 1989, Strebel et al. 1989]. Over 90 percent of the rural, and 50 percent of the total, population of North America obtain potable water from groundwater sources [Power and Schepers 1989]. Estimates from the National Pesticide Survey prepared by USEPA indicate that 5% of public and private wells in the U.S. have nitrate concentrations greater than the EPA maximum contaminant level [Bouchard et al. 1992]. Furthermore, in Europe many potable groundwater sources have shown an increase in nitrate concentration over the last two to three decades [Strebel et al. 1989]. Unless effective groundwater management measures are applied as populations grow and human activity expands, an increase in the number of nitrate contaminated groundwater supplies can be anticipated. Thus, an increase in the demand for efficient, economical nitrate removal technologies can be expected.

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[1] NO_3^--N is a standard terminology to indicate the value is reported as the mass of nitrogen in the form of the nitrate ion. Thus 10 mg NO_3^--N/L is equlal to 62 mg NO_3^-/L.

PROBLEM STATEMENT

Removal of nitrate from water is most commonly accomplished with desalinization technology; reverse osmosis or ion exchange. Both reverse osmosis and ion exchange are relatively non ion specific. In reverse osmosis most dissolved substances are retained in the brine which does not pass through the membrane. Because nitrate is nearly always a minor constituent the principal cost of nitrate removal is associated with the coincident removal of other solutes. The brine produced in reverse osmosis processes is rarely usable and the volume is generally between one-third and two-thirds of the total flow. Ion exchange resins highly selective for nitrate are not available. Resins having the highest affinities for nitrate have even higher affinities for sulfate, an ion that is often present at concentrations several times that of nitrate. Brine volumes produced during resin regeneration are lower in volume than those produced in reverse osmosis treatment. However, the total dissolved solids (TDS) concentrations of resin regenerates are very high and regenerate is unusable. As in the case of reverse osmosis, costs of nitrate removal by ion exchange are largely associated with removal of other ions.

Microbial Denitrification

Microbial denitrification, a frequently used and relatively inexpensive method of removing nitrate from wastewater, has been applied for the treatment of potable water supplies, on a limited scale, using packed bed reactors [Daigger et al. 1988, Liessens et al. 1993, Mateju, et al. 1992, Metcalf & Eddy 1991]. However, two significant drawbacks exist in transferring wastewater denitrification technology to the treatment of domestic water supplies: (1) the water is intimately mixed with microbial cultures and (2) organic compounds must be supplied as an energy source to drive the denitrification reactions and residual organics can be a water quality problem. Process configurations used experimentally have included both packed beds and fluidized beds [Green et al. 1994]. Denitrifying microbial cultures have been supported on sand, ceramics, polymers, clay, alginate gel, and agar gel. Work with conventional support materials (sand, ceramics, polymers, clay) has been relatively straightforward in that the microbial cultures are grown on support surfaces and water containing nitrate is passed through the fixed or expanded/fluidized bed.

Carbon and energy sources, nearly always organic compounds, are added to the water. Thus the problem outlined above - introduction of bacteria and organics - is characteristic of systems used to date.

Immobilized Bacterial Cell Denitrification Technology

A promising alternative to these conventional practices application of immobilized bacterial cell technology to denitrification. Bacterial cell immobilization may be defined as the physical confinement of intact cells to a selected location while preserving the desired micro biological activity [Liessens et al. 1993]. Desired microbial activity may include cell growth, fermentation, or denitrification. Conceptually, the microbial activity of immobilized bacterial cells can be compared to the microbial activity attainable in suspended cultures or fixed film cultures. The microbial activity of immobilized bacterial cells, for example cells contained behind a permeable membrane, can be considered to be influenced to a greater degree by substrate and waste product transport limitations than the microbial activity of a suspended bacterial culture (e.g., activated sludge). Conversely, the microbial activity of bacterial cells immobilized by a permeable membrane can be considered to be influenced to a lesser degree by mass transport limitations than the activity of a fixed biological film (e.g., trickling filter media slime). Flow regimes are typically laminar which give rise to unmixed boundary layers and lower rates of mass transport. For bacterial cells immobilized behind a permeable membrane, substrate and waste product transport is not prevented by a solid boundary, but only limited or reduced by the porous membrane. Turbulent flow patterns can be established on the "bacteria-free" side of the membrane without displacement of the cell culture. Thus, bacterial cell immobilization behind a porous membrane does not allow the ease of substrate or waste product transport available in a suspended culture medium, but does provide greater mass transport than a system with the biological film attached to an impermeable surface.

Gel Immobilized Cell Denitrification Bench-scale denitrification utilizing immobilized bacteria has been reported using systems with bacteria immobilization via agar-gel and agar-gel in combination with filter membranes.[Nilsson et al. 1980, Mattiasson et al. 1981, Nilsson and Ohlson 1982, Lemoine et al. 1988, Junter et al. 1990, Lemmoine et al. 1991a,b,c] The most

promising bench-scale gel/membrane "double flow reactor" [Junter et al. 1990, Lemmoine et al. 1991a,b] was reported to provide denitrification rates ranging from 800 - 1,400 mg/L NO_3-N/m^2/d, while maintaining segregation of the organic carbon energy source and microbial cells from the water being treated [Lemmoine et al. 1991a] In the gel/membrane reactor, ^{31} the carbon feed stream and high nitrate water were reportedly passed on opposite sides of a 3-mm thick agar-cell disk sandwiched between two 0.45 μm pore diameter microporous filters.

Microporous Membrane Systems McCleaf and Schroeder [1995] suggested that the problem of product water contamination with microorganisms, substrate, and metabolites could be solved by complete separation of the denitrification reactions from the water being treated by a microporous membrane. They proposed a system in which a denitrifying biofilm is established on one side of the membrane. Nitrate diffuses through the membrane from the "clean" water side and organic substrate and other nutrients are supplied from the biofilm side of the membrane, as shown in Figure 1. McCleaf and Schroeder investigated the concept using a two-cell batch reactor (Figure 2) and reported potential nitrate removal rates of up to 6.5 g/m^2•d using a 0.2 μm Nuclepore® filter microporous membrane. They reported values for effective NO_3 diffusivity through the membrane (0.34 x10^{-5} cm^2/s) and the composite membrane-biofilm (0.56 x10^{-5} cm^2/s)., considerably lower than the diffusivity of NO_3 in water of 1.5 x 10^{-5} cm^2/s. Some diffusion of methanol (the substrate used) from the reaction side to the clean water side of the membrane was observed. Because the experiments were focused on maximizing the NO_3 removal rate, McCleaf and Schroeder did not attempt to control methanol contamination. They also observed microbial growth occurring in the clean water side and suggested that the problem could be eliminated by (1) using smaller pore sizes and (2) UV disinfection of the water prior to treatment. Microbial transport across a 0.2 μm would not be surprising because many soil bacteria are smaller than 0.5 μm in size.

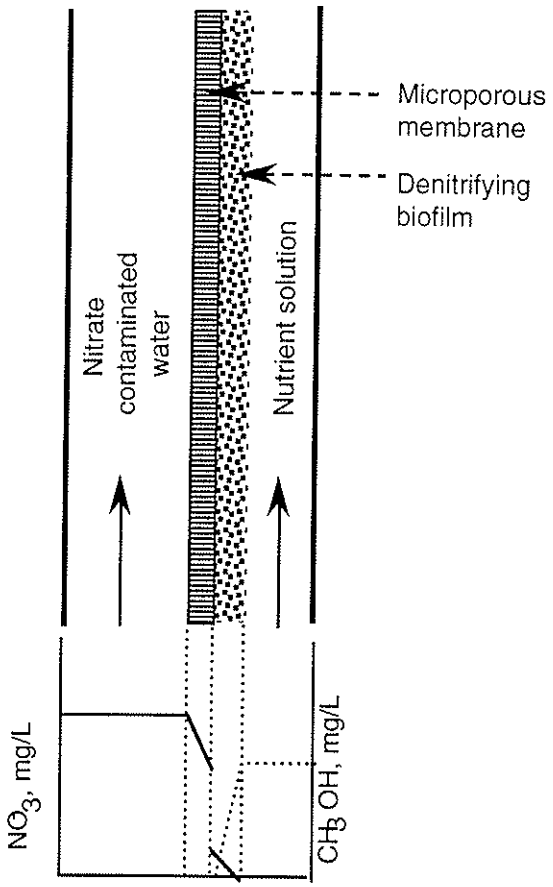

Figure 1.
Schematic of denitrifying biofilm system proposed by McCleaf and Schroeder [1995].

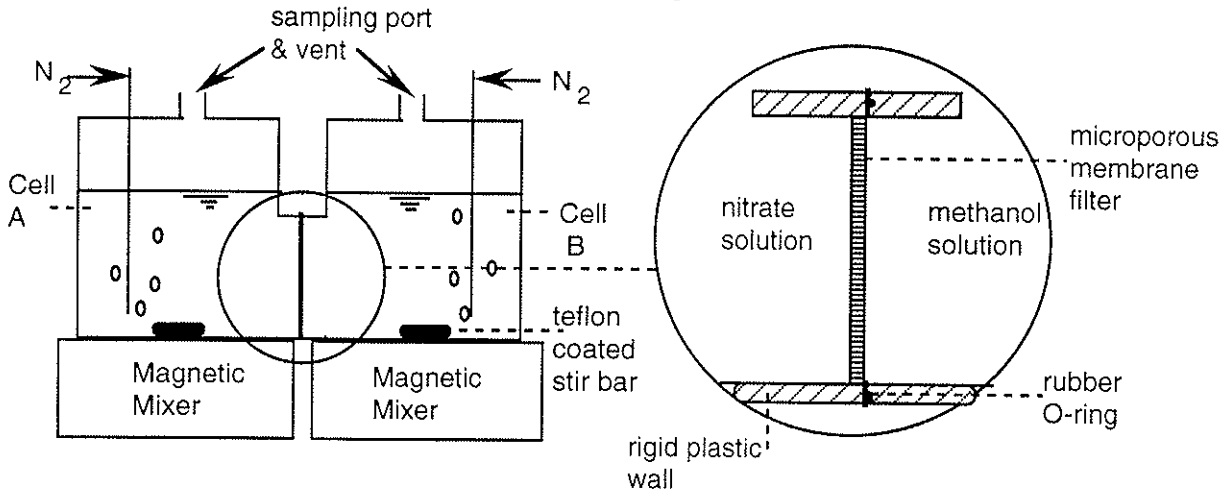

Figure 2.
Schematic diagram of the two-chambered biofilm reactor used by McCleaf and Schroeder (1995) and in work reported in this report. Reactor capacity was 2.8 L per cell.

RESEARCH OBJECTIVES

In this project results of experiments conducted for the purpose of addressing problems noted by McCleaf and Schroeder were addressed.

1. To minimize microbial contamination of the product water from the reaction side of the membrane, a 0.02 μm pore size membrane was used.
2. To study optimization of the removal rate, experiments were conducted using both biofilms and suspended growth cultures. Controlling diffusion of the organic substrate into the water being treated by controlled methanol addition was evaluated.
3. Deoxygenation with sulfite was evaluated as an alternative to stripping or adding excess substrate in two biofilm experiments.

MODEL OF MICROPOROUS MEMBRANE PROCESS

The NO_3^- removal process described schematically in Figure 1 is described mathematically for an idealized flat plate system in Equations 1 and 2. Assumptions made in writing Equations 1 and 2 were steady state conditions, negligible resistance to transport of NO_3^- in the lateral direction of the water being treated, and no reactions occurring in the water being treated or within the microporous membrane.

$$J_{NO_3^-} = \frac{Q}{W}\frac{dC_{Nb}}{dz} \tag{1}$$

$$J_{NO_3^-} = -D_{Nm}\frac{C_{Nb} - C_{Nbf}}{\delta_m} = \delta_{bf}\,\bar{r}_N \tag{2}$$

Where: $J_{NO_3^-}$ = mass flux of NO_3^--N through membrane, $g/m^2 \cdot s$

Q = volumetric flow rate of water being treated, m^3/s

W = width of membrane, m

z = longitudinal distance from entrance, m

C_{Nb} = bulk NO_3^--N concentration in the water being treated, g/m^3

C_{Nbf} = NO_3^--N concentration at the membrane-biofilm interface, g/m^3

D_{Nm} = molecular diffusivity of NO_3^- in the microporous membrane, m^2/s

δ_m = microporous membrane thickness, m

δ_{bf} = biofilm thickness, m

$\bar{r}_N$ = mean denitrification rate in biofilm, g NO_3^--N/m^3•s

The point denitrification rate within the biofilm, r_N, can probably be described using the Mondod model and a mass balance could be written for transport and reaction of NO_3^- in the biofilm. The result would be a set of equations which could be solved for "shallow" and "deep" conditions [Riemer and Harremoës, 1978, Rittman and McCarty, 1980]. However, based on the results of McCleaf and Schroeder, and those presented later in this paper the effective NO_3^--N concentration at the membrane-biofilm interface, C_{Nbf}, is substantially lower than the bulk concentration in the water being treated, but not negligible. Under the conditions studied to date, with initial bulk NO_3^--N concentrations in the 30 to 60 mg/L range, removal rates observed have been pseudo first order and can be described using a modified form of Equation 1 in which the membrane-biofilm NO_3^--N concentration, C_{Nbf}, is omitted and an effective diffusion coefficient, D_{me}, is substituted for the molecular diffusivity, D_{Nm}. The resulting expressions for removal and NO_3^--N concentration are then given by Equations 3 and 4.

$$\frac{Q}{L}\frac{dC_{Nb}}{dz} = -\frac{D_{me}}{\delta_m} C_{Nb} \tag{3}$$

$$C_{Nb} = C_{Nbo} \exp\left(-\frac{L D_{me}}{Q \delta_m} z\right) \tag{4}$$

Where: C_{Nbo} = initial NO_3^--N concentration of, g/m^3

MATERIALS AND METHODS

The experimental reactor used is shown in Figure 2 and was identical to that used by McCleaf and Schroeder [1995]. A polytetrafluoroethylene (PTFE) membrane material (W. L. Gore & Associates, Elkton, MD) with a nominal pore size of 0.02 μm, a pore fraction of 50% (by volume), and an average thickness of 80 μm was placed at the circular connection between the cells. Wetting of the hydrophobic membranes with methanol was the only preparation required to

allow the pores to fill with water. Liquid volume of each cell was 2.0 L. All experiments reported in this paper were run under batch conditions using UC Davis tap water containing a phosphate buffer composed of 1.74 g/L KH_2PO_4 and 2.14 g/L K_2HPO_4. Anoxic conditions were maintained using two methods; (1) bubbling N_2 gas through each cell, as shown in Figure 2, and (2) addition of 50 mg/L of SO_3^{-2}. Investigation of the use of SO_3^{-2} was of interest because stripping of oxygen from the water being treated is probably economically unfeasible and the only alternative is to overdose the systems with organic substrate. Relatively low concentrations of SO_4^{-2} result from addition of SO_3^{-2} (approximately 30 mg/L for a typical groundwater containing 5 mg/L dissolved oxygen).

Analytical Methods

All samples were initially filtered through a glass fiber filter with an effective retention of 1.2 μm (GF/C, Whatman, Clifton, NJ) and refrigerated at 4°C until analyzed. Nitrate concentrations (as nitrate nitrogen) were measured using an nitrate probe (Orion Research, Boston, MA) consisting of a nitrate ion-selective electrode (model 93-07) and a double junction reference electrode (model 90-02). For the diffusion experiments, nitrate standards were prepared using potassium nitrate and deionized water. In the case of the denitrification experiments nitrate standards were prepared in tap water containing approximately the same concentrations of the buffering chemicals to overcome interferences. Nitrite concentrations were assumed to be insignificant based on the results of McCleaf and Schroeder [1995]. Total organic carbon (TOC) concentrations were measured using a Shimadzu model 5050 TOC analyzer (Shimadzu Instruments, Kyoto, Japan) For selected samples in the suspended culture experiments, triplicate total suspended solids (TSS) measurements (mg dry cells/L) were made [American Public Health Association 1992].

Membrane Diffusivity Determination

To determine the diffusion coefficient through the membrane, Cell A was filled with 2 liters of deionized water containing 1000 mg/L NO_3-N while Cell B was filled with the same volume of water containing without NO_3. Samples were collected from both cells at selected time intervals

over a four hour period. Each sample was diluted 10:1 with deionized water and the NO_3^--N concentration was determined using the nitrate probe.

Because the cells were of equal volume and well stirred, a mass balance on cell A, the cell to which NO_3^- was added, results in Equations 5 and 6:

$$C_{NB} = C_{NAo} - C_{NA} \tag{5}$$

$$V\frac{dC_{NA}}{dt} = -A_{mp}D_m\left(\frac{C_{NA} - C_{NB}}{\delta_m}\right) = -A_{mp}D_m\left(\frac{2C_{NA} - C_{NAo}}{\delta_m}\right) \tag{6a}$$

$$V\frac{dC_{NB}}{dt} = A_{mp}D_m\left(\frac{C_{NA} - C_{NB}}{\delta_m}\right) = A_{mp}D_m\left(\frac{C_{NAo} - 2C_{NB}}{\delta_m}\right) \tag{6b}$$

where:

V = individual volume of cells, m^3

C_{NA}, C_{NB} = NO_3^--N concentration in Cell A and B, respectively, g/m^3

C_{NAo} = NO_3^--N concentration in Cell A at time = 0, g/m^3

t = time elapsed, seconds

A_{mp} = pore area of membrane = $\pi r^2 \varepsilon$, m^2

ε = nominal pore fraction

r = membrane radius, m

D_m = nitrate diffusion coefficient through membrane material, m^2/s

δ_m = nominal thickness of membrane, m

Integration of Equation 6 and solution for the diffusivity, D_m, gives:

$$D_m = \frac{\delta_m V}{2A_{mp}t}\ln\left(\frac{2C_{NA} - C_{NAo}}{C_{NAo}}\right) = \frac{\delta_m V}{2A_{mp}t}\ln\left(\frac{C_{NAo} - 2C_{NB}}{C_{NAo}}\right) \tag{7}$$

The diffusivity through the membrane was determined by plotting the transformed data from both cells were used in calculating the slope of the regressed line.

Denitrifying Enrichment Culture

A denitrifying enrichment culture was developed from activated sludge taken from the UC Davis wastewater treatment plant. Methanol (CH_3OH), and potassium nitrate were added to the culture in a ratio of 3.0 grams (3.75 mL) of methanol per gram of NO_3^--N added as suggested by Mateju *et al.* [1992] with phosphate buffer and UC Davis tap water. The culture was maintained in

a stirred, sealed flask. At two day intervals, the culture was allowed to settle, half the liquid volume was decanted, and the flask was re-filled with tap water to which nitrate, methanol, and phosphate buffer were added.

Biofilm Experiments

Biofilms were established on the Cell B side of the membrane over a period of 1-2 weeks prior to experiments. Initially, a membrane was placed in the reactor and wetted with methanol to allow passage of liquid through the membrane. Tap water containing potassium nitrate was placed in Cell A, while denitrifying organisms and methanol were added to Cell B. Bacteria accumulated on the Cell B side of the membrane (the interface between nitrate and carbon sources). Wall growth in Cell B was periodically brushed off to maximize growth on the membrane. Before each rate experiment both cells were emptied and all walls were cleaned. Cell A was then filled with 2000 mL of buffered UC Davis tap water containing 45 to 50 mg/L of NO_3^--N and cell B was filled with buffered UC Davis tap water containing 120 to 160 mg/L CH_3OH (45 to 60 mg/L TOC). Oxygen was stripped from water with N_2 gas and anoxic conditions were maintained by either continuous addition of N_2 or by the addition of 50 mg/L SO_3^{2-} and maintaining an oxygen free headspace using N_2 gas.

As noted above, observed removal rate behavior of the batch systems has been pseudo-first order with respect to the bulk NO_3^--N concentration. The batch equation for the biofilm experiments that corresponds to Equation 4 is:

$$C_{NA} = C_{NAo} \exp\left(- \frac{A_{mp} D_{me}}{V \quad \delta_m} t\right) \tag{8}$$

Suspended Growth Experiments

The suspended culture experiments were conducted in the same manner as the biofilm experiments except that the pre-experiment growth period was not required. Cell B was prepared by adding a selected volume of denitrifying bacteria from the stock culture and filled to a 2.0 L

volume with buffered tap water and methanol. Initial NO_3-N-N concentrations were the same as in the biofilm experiments and initial biological solids concentrations ranged from 50 to 250 mg/L.

Because of the high concentration of CH_3OH at the beginning of the suspended growth experiments and the lack of reaction and diffusion resistance associated with the biofilm, significant substrate transport across the membrane occurred. Over the course of the experiments the CH_3OH accumulated in cell A and then diffused back across the membrane into cell B where it was consumed in the denitrification reactions. Although continuous flow processes would not be affected in the same manner, the problem was addressed in these experiments by incremental addition of methanol in four steps.

Samples were taken from both cells at selected time intervals during the 48 hour experiments. Suspended solids were measured in the samples taken at the initial time, after 24 hours, and after 48 hours to monitor bacterial growth over the experimental period. Anoxic conditions were maintained by bubbling nitrogen gas through both cells, which also maintained the oxygen free conditions in the head space.

To determine denitrification rates, the same procedure was used as in the biofilm experiments if the concentration in Cell B remained at zero. In the experiments in which NO_3-N accumulated in Cell B, a modified form of Equation 6 was applied:

$$V\frac{\Delta C_{NA}}{\Delta t} = V\frac{C_{NA_{t+\Delta t}} - C_{NA_t}}{\Delta t} = -A_{mp}D_m\frac{\bar{C}_{NA} - \bar{C}_{NB}}{\delta_m} \qquad (9)$$

where: ΔC_{NA} = change in NO_3-N concentration in Cell A over time increment, mg/L,

$\bar{C}_{NA}$ = average NO_3-N concentration in Cell A over time increment, mg/L,

$\bar{C}_{NB}$ = average NO_3-N concentration in Cell B over time increment, mg/L,

Δt = time elapsed during time increment, min,

and all other variables are as defined previously.

RESULTS

Results of the membrane diffusivity, biofilm and suspended culture experiments are presented separately. Overall the potential flux of nitrate through the membrane appears to be considerably greater using suspended cultures. Operation with a biofilm removal process results in a decrease in the effective diffusivity of approximately 25 percent relative to the use of suspended cultures. Effective diffusivities observed for the suspended culture systems were equal to the sterile system membrane diffusivities, which are presumed to be the maximum attainable.

Membrane Diffusivity

The results of the six membrane diffusion experiments conducted are summarized using pooled data in Figure 3. Each of the experiments was begun with an initial concentration of approximately 1000 NO_3^--N mg/L and samples were taken over a three to five hour period. Using Equation 7, and the regression slope value calculated for the data shown in Figure 3, the NO_3^--N membrane diffusivity, D_m, is given by Equation 10. The pore area was assumed to be 50 percent of the membrane cross section on the basis of the membrane void fraction.

$$D_m = 0.00086 \frac{\delta_m V}{2A_{mp}} = 0.00086 \frac{(8 \times 10^{-5}\ m)(2 \times 10^{-3}\ m^3)}{2(0.5)(4.56 \times 10^{-3}\ m^2)}$$

$$= 3.02 \times 10^{-8}\ m^2/min = 5.03 \times 10^{-6}\ cm^2/s \qquad (10)$$

Data from the six experiments were pooled to show the amount of variation between experiments. Concentrations measured in both cells were used in the calculations, giving a check on the mass balances and providing an additional five sets of data. Calculation of diffusivities for the individual experiments resulted values ranging from 3.02×10^{-6} to 5.8×10^{-6} cm^2/s with R^2 values, with one exception, greater than 0.98.

Biofilm Diffusivities

Results of the six biofilm experiments conducted to provide a comparison with the 0.2 μm pore size membrane results of McCleaf and Schroeder [1995] are summarized in Table 1 and typical biofilm system response is shown in Figures 4 and 5. The diffusivities given in Table 1 are

averages of 9 to 11 values calculated for individual samples using Equation 8. Calculations were based on the assumption that the NO_3^--N concentration at the membrane-biofilm interface was small relative the bulk concentration in cell A. The assumption was used in the derivation of Equation 8 and validity of the assumption is supported by the fit shown in Figure 5. Diffusivity values were calculated using the membrane thickness (80 μm) and somewhat larger diffusivity values would be estimated if diffusion and reaction in the biofilm were included. The fact that the estimated diffusivities were approximately 25 percent lower than the sterile system diffusivity calculated above suggests that a biofilm effect occurred and that the actual interface concentration, CN_{bf}, was not negligible. Effective depth of the biofilm could not be determined in experiments and the good fit of the data to a first order expression provides an adequate empirical model for the system, however.

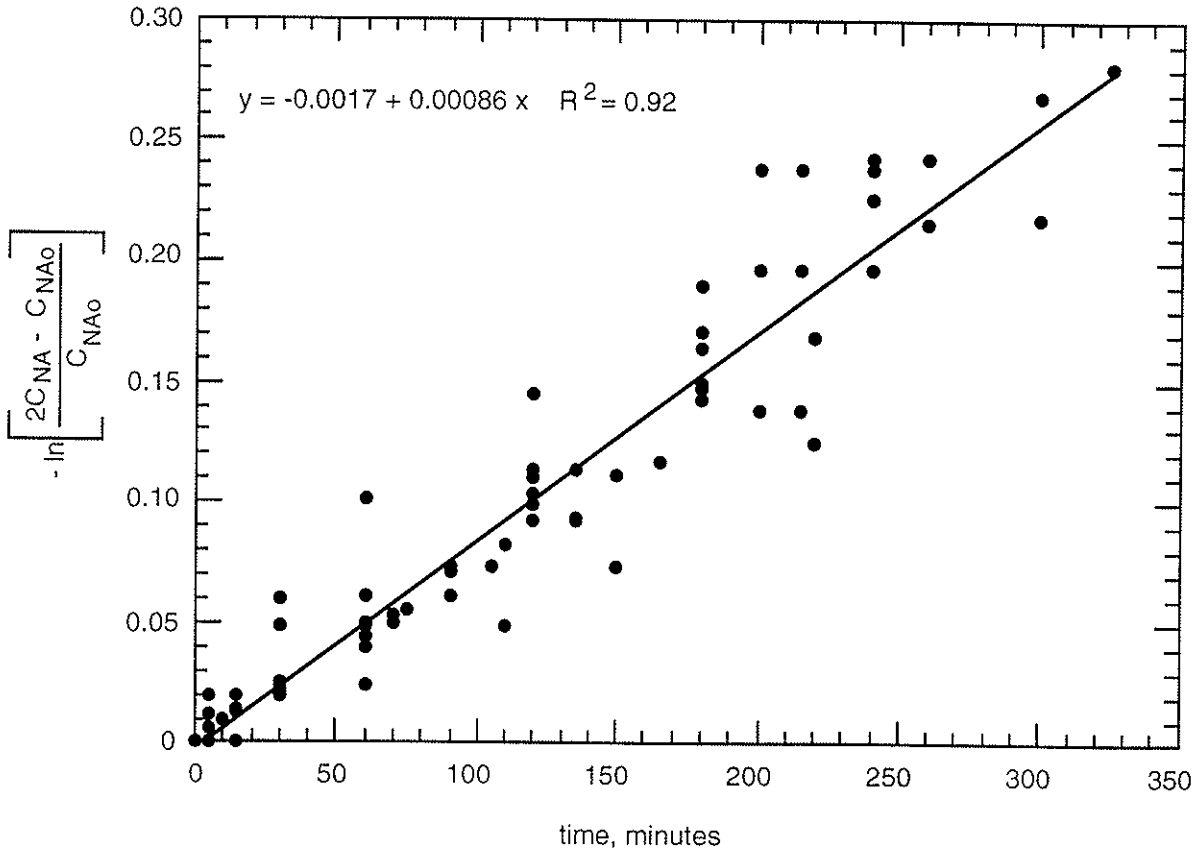

Figure 3
Combined data from six diffusion experiments using sterile, batch systems and a 0.02 μm nominal pore diameter membrane having a 50 % porosity

Table 1.
Composite diffusivities based on a membrane thickness of 80 μm. Standard deviations (σ) given are based on 9 to 11 values in individual experiments.

Experiment	D_{me} cm^2/s	σ cm^2/s
BF 1	3.7×10^{-6}	1.8×10^{-7}
BF 2	3.6×10^{-6}	5.8×10^{-7}
BF 3*	3.4×10^{-6}	5.7×10^{-7}
BF 4	2.7×10^{-6}	5.3×10^{-7}
BF 5	3.4×10^{-6}	3.8×10^{-7}
BF 6	4.0×10^{-6}	3.3×10^{-7}
average	3.5×10^{-6}	

*initial point of 10 points omitted in calculations

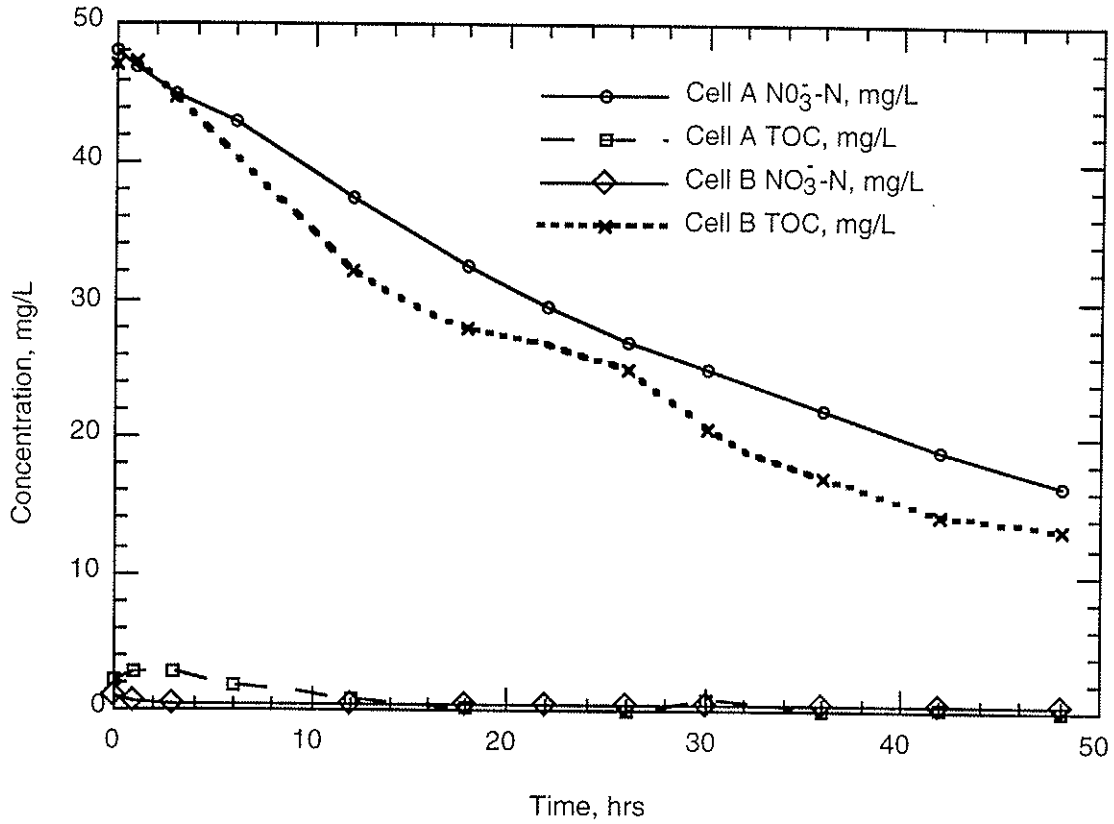

Figure 4
Typical variation of NO_3^--N and TOC concentrations with time in biofilm experiments.

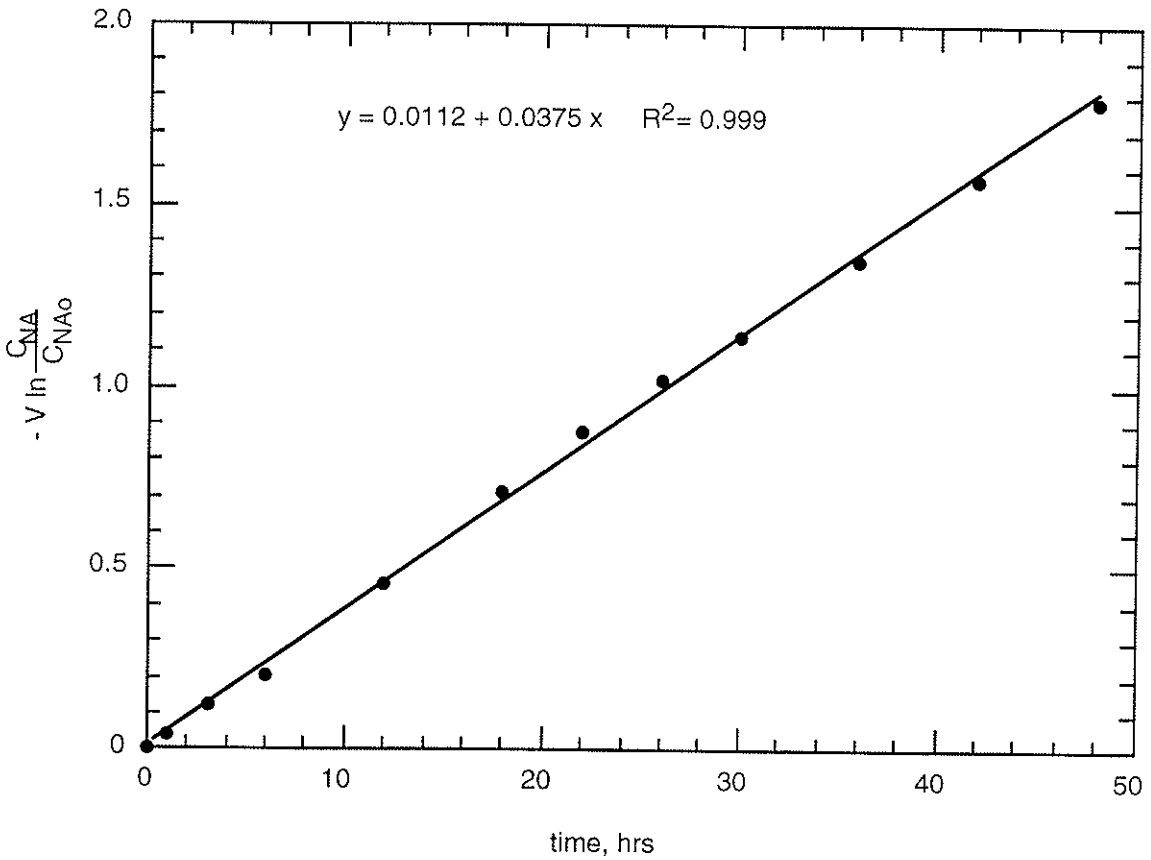

Figure 5.
First order fit of biofilm data from experiment BF 1. Goodness of fit was similar for all of the biofilm experiments with R^2 values ranging from 0.983 to 0.999

In biofilm experiments one through four deoxygenation was by stripping with N_2 and in experiments 5 and 6 deoxygenation was by SO_3^{-2} addition. Nitrate removal rates did not vary significantly with the method of deoxygenation.

Suspended Culture Experiments

The suspended culture experiments were conducted to determine if higher rates of removal could be attained in a suspended growth system. Minimizing microbial fouling would be an additional advantage of such a system. Five experiments were conducted, two in which methanol was added at the beginning and three in which methanol was added in four steps at 12 hour intervals in an attempt to minimize transport into the clean water cell. In the first suspended culture experiment the initial solids concentration was 51 mg/L and NO_3 accumulated in Cell B.

Increasing the initial solids concentration to 245 mg/L eliminated nitrate accumulation in Cell B but did not prevent transport of methanol across the membrane into Cell A, as shown in Figure 6. Incremental methanol feeding decreased but did not eliminate the accumulation of methanol in Cell A as shown in Figure 7. Transient accumulation of NO_3-N also occurred in Cell B during the incremental feed experiments, with the maximum concentration being approximately 4 mg/L.

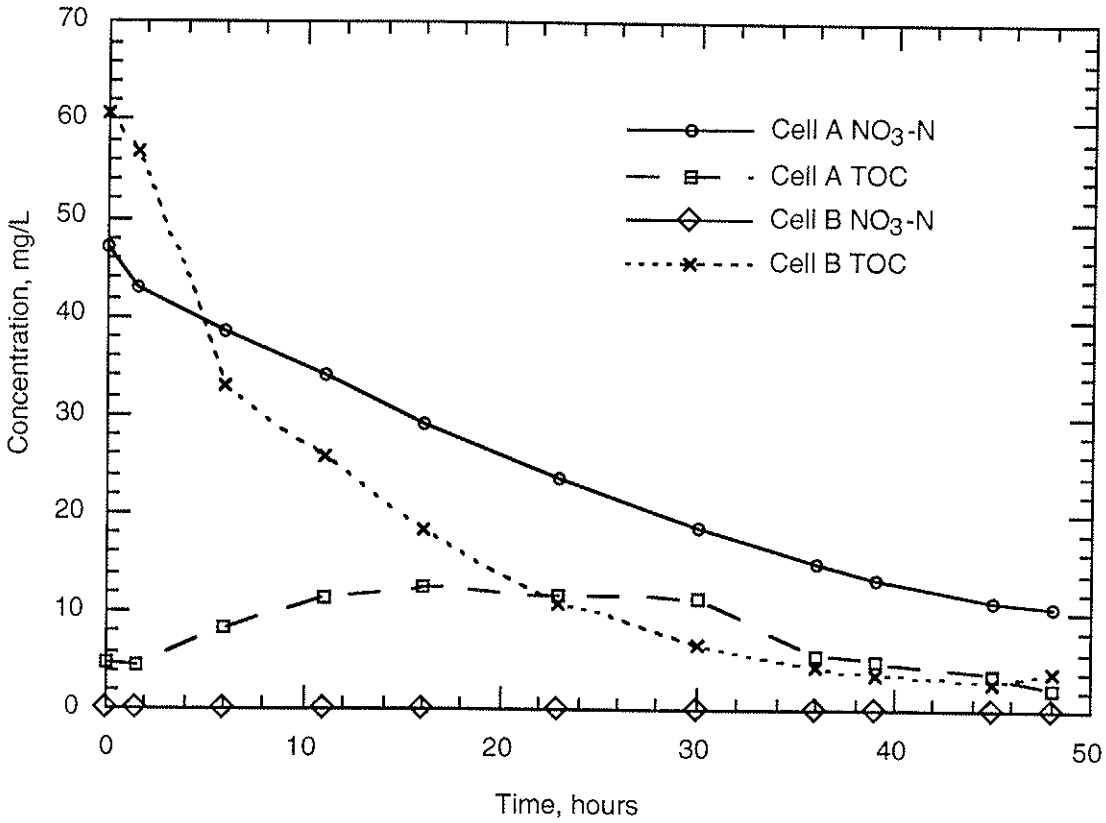

Figure 6.
Variation of NO_3-N and TOC in slug fed suspended Culture Experiment 2.

Support for a first order removal model is provided by the data shown for suspended culture experiment 2 shown in Figure 9. Diffusivities calculated from the suspended culture experiments averaged 4.7 x 10^{-6} cm^2/s with the two slug fed systems having diffusivities greater than 5 x 10^{-6} cm^2/s. Thus biofilm formation appears to be minimal and diffusivities approaching sterile system values can be attained in the suspended culture systems. Continuous flow systems, such as that schematically shown in Figure 2, could be operated using higher cell concentrations that provided adequate control of methanol contamination by maintaining high denitrification rates

at low methanol concentrations. Note that the objective of maintaining very low NO_3^--N concentrations on the reaction side of the membrane is to maximize the concentration gradient, and consequently the nitrate flux across the membrane. Such a system is currently under evaluation in the laboratories of the Center for Environmental and Water Resources Engineering.

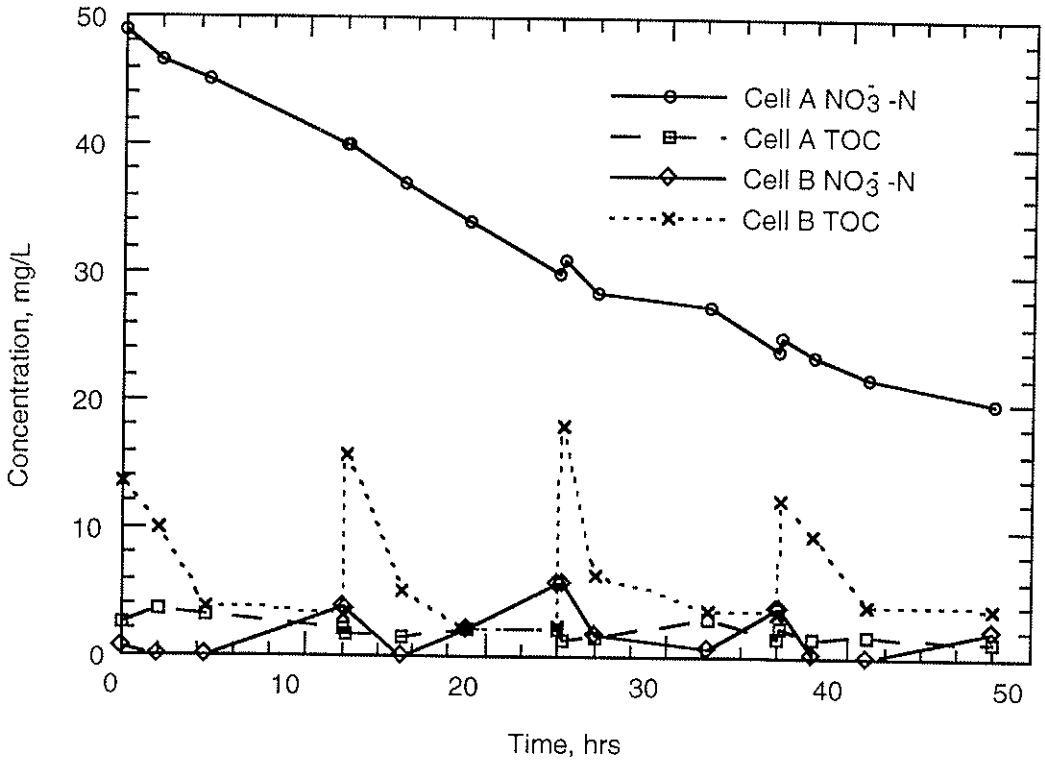

Figure 7.
Variation of NO_3^--N and TOC in incrementally fed suspended Culture Experiment 2.

Methanol Requirement

The stoichiometric methanol requirement is approximately 2.5 g methanol/gram NO_3^--N removed, or 0.92 g carbon/gram NO_3^--N removed. McCleaf and Schroeder [1995] reported a methanol requirement of 2.2 g organic carbon; more than twice the stoichiometric value. Methanol stripping was expected to occur in the experimental system (Figure 2) because of the free surface and the use of diffused N_2 to maintain anaerobic conditions. An attempt to lower the methanol requirement by decreasing the diffused N_2 flow rate was successful. For the 11 experiments reported here (six biofilm and five suspended culture) the average methanol requirement was 1.4 g

organic carbon/g N removed. Because free surfaces will not exist in prototype systems, the methanol requirement should decrease to a value approaching stoichiometric.

DISCUSSION AND CONCLUSIONS

The possibility of designing a microbial denitrification system using microporous membranes to separate the water under treatment from the microbial culture has been established. Using a suspended culture system, diffusivities of 5×10^{-6} cm^2/s appear to be realistic and improved membranes may be possible that have diffusivities more closely approaching that of NO_3^- in water ($\approx 1.5 \times 10^{-5}$ cm^2/s). The necessary size of prototype unit having membrane properties similar to those of the membranes used in this study and treating one million gallons of water per day can be estimated using Equation 8. Reducing the NO_3^--N concentration from 30 mg/L to 10 mg/L (the drinking water limit) with a system of 2 cm diameter tubes would require a cross sectional area (including spacing between tubes) of approximately 50 m^2 and a length of 10 m. Thus the size of the necessary facilities for ground water treatment are not excessive. Cost estimates will require more knowledge of unit construction than is currently available.

RECOMMENDATIONS

Denitrification of drinking water using microporous membranes appears to be extremely promising. Four principal recommendations can be formulated from the work completed:

1. Development of a continuous flow, microporous membrane denitrification process is necessary if the approach is to be useful on a large scale.

2. Experience with the long term performance of the membranes is necessary. Because microbial fouling of the pores is not expected to greatly affect the nitrate flux, as demonstrated in the biofilm studies, long term performance is expected to be good.

3. Autotrophic denitrification using microporous membrane systems needs to be investigated as soon as possible. A proposal to carry out this work is in preparation.

4. Pilot scale evaluation of the microporous membrane process needs to be scheduled as soon as possible.

SUMMARY

The specific objectives stated in the project proposal were:

1. Determination of diffusion and overall transfer rates using 0.02 μm pore size membrane,
2. Development of a method for controlling dissolved oxygen in the reactor,
3. Determination of operation protocol to control film growth,
4. Investigation and comparison of Methanol of H_2 as energy sources

Objectives one through three have been met. The results using the 0.02 μm membranes were even more promising than the results from the larger pore size experiments of McCleaf and Schroeder. Use of the smaller pore size eliminated microbial contamination of the clean water and flux of nitrate was somewhat greater than reported in the earlier experiments. Two methods of controlling dissolved oxygen concentrations were successful, stripping with an inert gas and addition of sulfite. Use of suspended growth cultures appear to be a suitable method of controlling biofouling and transport of the organic feed across the membrane into the water being treated. Moreover, suspended growth systems will have maximum concentration gradients and higher nitrate fluxes across the membrane.

The fourth objective was not met. However, recent work at the University of Nevada, Reno [Ahmed, 1996] on fluidized bed, autotrophic denitrification is extremely promising. In that work H_2 was used as the electron donor and observed rates were comparable to rates with organic feeds. Conversion between heterotrophic and autotrophic operation was not difficult and very rapid.

Work on continuous flow microporous membrane denitrification has begun in the CEWRE laboratories. A Proposal for funding further work have been submitted to the USEPA. Proposals for funding related microporous membrane denitrification research are being prepared for

submission to the National Science Foundation and the Government of Israel (in cooperation with Professor Asher Brenner of the Ben-Gurion University of the Negev).

ACKNOWLEDGEMENT

The participation of Professor Asher Brenner of the J. Blaustein Institute for Desert Research , Ben-Gurion University of Negev, Beer-Sheva, Israel during his 1995-96 sabbatical year was of great importance to the final year of the project and was greatly appreciated.

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