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Treatment of a low-carbon nitrified wastewater effluent in subsurface flow constructed wetlands

Ginachi Amah, I.H. (Mel) Suffet, Randal Orton, Jacqy Gamble and Michael K. Stenstrom

Abstract

A pilot-scale study investigated constructed wetlands to remove nitrogen from nitrified tertiary effluent. Two subsurface flow units, each with 3.25 m^3 volume, were planted with Typha sp. and operated at hydraulic residence times (HRT) of 18 and 48 hrs for 10 months after an initial startup period of six months. Nitrate removal averaged 50.6% and 20.8% at the 48- and 18-hr HRTs. The mass of nitrogen removed by both systems was 3.0 to 3.2 g/d. There was a modest variation in performance with changes in season. At the end of the experiment, sodium acetate was added to promote denitrification; and the response suggests that a denitrifying community of organisms was either absent, or not very large, implying that plant uptake is the predominant nitrate-removal mechanism. Phosphate removal was only 4% and 14.3% (18 and 48 hr HRT). Scale-up of these units suggests that removal rates of 1 g/m^2/d of nitrate are possible.

Key words: constructed wetlands, denitrification, nitrate removal, nitrogen uptake

INTRODUCTION

The detrimental effects of excess nitrogen and phosphorus compounds entering surface waters have become more evident with the increase in eutrophication and its associated problems. Wastewater treatment plant operators are seeking cost-effective means of nitrogen removal, since new policies and discharge regulations are limiting nutrient loading to receiving water bodies. Constructed wetlands are currently viewed as an effective and low-cost option, due to their simplicity of design, construction and operation. An important objective of many constructed wetlands for nutrient removal in the US has been ammonia and total nitrogen reduction (USEPA 1993; Wood 1995); however, some studies (Gersberg *et al*. 1984*a*), and an increasing number of more recent studies, are focusing on nitrate reduction as their goal (Bachand 1996; WERF 2000). One reason for more interest in nitrate removal is the existence of nitrified and only partially nitrified wastewater effluents.

The purpose of this study was to determine the extent of nitrate reduction possible in a subsurface flow (SSF) wetland, treating nitrified tertiary effluent in the absence of supplemental organic carbon. Performance at two hydraulic retention times was compared, and possible mechanisms of nitrate reduction were also investigated. Changes in phosphorus concentrations were also monitored.

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BACKGROUND

Wetlands may remove ammonia or nitrate from treated wastewaters by denitrification, plant uptake,

dissimilatory nitrate reduction to ammonia (DNRA), and ammonia volatilization. DNRA occurs under conditions of low dissolved oxygen, high carbon loading and low nitrate concentration, which are rarely observed in facilities or wetlands receiving treated wastewater. Volatilization of ammonia is generally insignificant at the neutral pH of most municipal wastewaters. Table 1 shows the results of previous studies conducted to ascertain the contribution of each removal mechanism in constructed wetlands. There is a general consensus among these results and other literature (Brix 1993; Hammer and Knight 1994; Ingersoll and Baker 1998) that denitrification is the dominant mechanism.

DENITRIFICATION

Denitrification is the process of nitrate reduction to nitrogen gas in the absence of oxygen; nitrate is used as the terminal electron acceptor in respiration. Organic carbon compounds serve as hydrogen donors as well as an energy source for growth of the denitrifying bacteria (Nichols 1983). The organic carbon requirement for denitrification varies between 2.5 and 10 g BOD per gram of nitrate, depending on the carbon source (Reed *et al*. 1995; Aesoy *et al*. 1998).

The nature of the carbon determines the efficiency of the reduction. Compounds that are easily biodegraded, such as simple sugars, alcohols, or organic acids, are oxidized quickly and promote high denitrification rates (Alexander 1961; Mitsch and Gosselink 1993). Acetate and methanol are effective artificial carbon sources in wastewater treatment when applied at weight ratios (mass compound/N) in excess of 2.7 and 1.9, respectively (Metcalf and Eddy Inc. 2003). Carbon contained in raw sewage has a moderate reaction rate, while reaction with carbon in treated wastewater and plant matter is slower (Mitsch and Gosselink 1993).

Dissolved oxygen interferes with denitrification by suppressing the enzyme system necessary for the process to occur (Metcalf and Eddy Inc. 2003). Diffusion of atmospheric oxygen is limited in an SSF wetland; therefore, the roots and rhizomes of the wetland plants are the main source of oxygen (Reed *et al*. 1995). The rate of oxygen production of these plants is estimated as 4.6–7.2 g/m^2/d (Gersberg *et al*. 1986; Armstrong *et al*. 1990; Brix and Schierup 1990), most of which is consumed by the respiratory demands of the plants (Brix and Schierup 1990). Even with oxygen present in the bulk water, the biofilms on the rhizomes and media are able to maintain anoxic internal regions where denitrification takes place (Williams *et al*. 1994).

The temperature for denitrification ranges from 15 to 65°C (Alexander 1961; Hammer and Knight 1994), with rates dropping off above and below this range. Within this range, the optimal temperature varies among wetlands (Wood *et al*. 1999; Ingersoll and Baker 1998). Pfenning and McMahon (1996) observed that potential denitrification rates at 4°C were about 77% lower than at 22°C in riverbed sediments. Stengel and Schultz-Hock (1989) demonstrated that the impact of low temperatures on denitrification rates in a wetland could be mitigated by the addition of supplemental carbon.

Table 1. Contribution of different mechanisms to nitrogen removal in constructed wetlands

Author	Method	DN	PU	DNRA	Comments
Gersberg *et al*. (1983)	Estimation from pilot-study results	84–88%	12–16%		SSF
Van Ostrom & Russel (1994)	^{15}N-labelling	95%	Inhibited by darkness	<5%	FWS
Cooke (1994)	Isotope (^{15}N) dilution	60–70%	5–10%	25–35%	FWS
Reed *et al*. (1995)	Harvesting trials	>90%	<10%		FWS
Raman *et al*. (1999)*	Estimations from literature values	44%	49%		FWS 8% volatilization

DN = denitrification, DNRA = dissimilatory nitrate reduction to ammonia, PU = plant uptake
Wetland types: FWS = Free water surface; SSF = Subsurface flow
* Influent nitrogen in the form of ammonia; all others were as nitrate

Table 2. Studies of denitrification in subsurface flow wetlands

Reference/ location	Dimensions (area x depth)	Media/plants	Type of waste	Retention time (days)	Influent nitrogen (mg/L)	Carbon source (mg/L)	Removal efficiency	Comments
Pilot scale								
Gersberg (1983) Santee, CA	65m^2 x 0.76m	*Scirpus* & *Typha* spp.	Nitrified 2° effluent	0.36	TN = 18 Nitrate = 17.3 NH_3 = 0.1	BOD: 2.0 Plant mulch 170gC/m^3	TN = 27% TN = 87% TIN = 91%	
Vymazal (1990) Prague, Czech.	Pilot 0.5m^2 x 1m	0.5–8mm gravel 0.5–3mm limestone *Phragmites* sp.	Diluted chicken waste	13	TON: 24.4–25.7 NH_3N: 155–270	BOD: 2340–3220	TON: >83% NH_3N: >91%	
Zhu & Sikora (1995) Alabama	0.14m^2 x 0.5m	5–10mm/ varied	Synthetic solutions	~5 ~2.1	TON: 48	Organic C: 12–30 Acetate: 118mgC/L	100% 100%	Short-term batch experiments 9 days
Laber *et al*. (1997) Austria	Small units for 2 farmhouses (VF) 5m^2 x 0.6m	Sand + gravel 1–4mm/ *Phragmites* sp.	Effluent from nitrifying wetland	n.a.	Nitrate: 36 36 45	Methanol	Nitrate: 89% 58% 72%	Intermittent feed continuous single batch
Green & Verhoeven (1999) New Zealand	2.53m^2 x 0.6m	Washed gravel/reeds	2° sewage effluent	0.25–5	TON: 12–18.5 NH_3N: 5.1–8.7	BOD: 11–19	TON: 29–64% NH_3N: 13.5–96%	Very short-term study
WERF (2000) New Hanover, NC	67.5m^2 x 0.6m	River rock/ varied	Treated landfill leachate	14	TN: 125 TON: 106 NH_3N: 12	BOD: 24±15	TN: 12–19% TON: 13–24% NH_3N: n.a.	Actual HRT reduced by short-circuiting
Full scale								
Gersberg *et al*. (1984*a*) Santee, CA	824m^2 x 0.76m & 742m^2 x 0.6m	*Scirpus* & *Typha* spp.	1: 2 mix of 1° & 2° effluent	0.33	TN = 21.3 TIN = 20 NH_3 = 7 Nitrate = 12.3	BOD: 51.5	TN = 80% TIN = 80%	
Gersberg *et al*. (1984*b*) Santee, CA	824m^2x 0.76m & 742m^2 x 0.6m	*Scirpus* & *Typha* spp.	Nitrified 2° effluent	0.3–0.7 0.24–0.3 0.5–0.7 0.3–0.4	TN = 19 TON = 18.3 NH_3 = 0.1	BOD: 3.3 Methanol MeOH:NO_3 = 4.5 C:NO_3 = 1.7 Plant mulch Plant = 23g/m^3 Plant = 13g/m^3	TN: 9–19% TN 94% TIN 97% TN 89% ± 7 TIN 95% ± 6 TN 70% ± 21% TIN 65% ± 18	A series of experiments to determine the effect of different carbon sources on nitrate removal
Reuter *et al*. (1989) Lake Tahoe, CA	660m^2	19mm gravel/ cattails	Urban run-off	4	TON: 0.114	n.a.	TON: 85–87%	
TVA (1990) Bear Creek, AL	2033m^2 x 0.3m	12.7mm river gravel/ *Typha* sp *Carex* sp.	2° sewage effluent		TON: 26 Org-N: 12 NH_3N: 10.7	BOD: 13	TON: 75% Org-N: 95% NH_3N: 83%	
Williams *et al*. (1995) UK	140m^2 x 0.2m	Flint gravel 10–20mm/ *Phragmites* sp.	Nitrified 2° sewage effluent	~0.25	TON: 16.7 NH_3N: 4.1	BOD: 21.5	TON: 53% NH_3N: 93%	Intermittent flow 12 hrs/day
Coombes & Collett (1995) Devon, UK	500m^2 x 0.4–0.5m	4–15mm basalt & limestone aggregate	2° sewage effluent	1.2–3.6*	TON: 14.7–19.6 NH_3N:0.6–4.3	BOD: 9.6–13.9	TON: 22% NH_3N: 15%	
Green & Verhoeven (1999) New Zealand	825m^2	n.a./ *Phragmites australis*	2° sewage effluent	1	TON: 18.8 TKN: 9.8	BOD: 24.6	TON: 76% TKN: 90%	Based on a 5-day and 3-day survey

* Calculated from available data; VF = vertical flow; n.a. = not available; TIN = Total inorganic nitrogen (NH_3 + NO_3 + NO_2); TON = Total oxidized nitrogen (NO_3 + NO_2); TN = Total nitrogen; TKN = Total Kjeldahl nitrogen
1° = primary, 2° = secondary

Plant uptake

Nitrate assimilation by plants can be another significant removal mechanism in constructed wetlands. This process varies seasonally, with high uptake rates in the spring and early summer. Senescence in the fall and winter months returns the nutrients to the wetland (Gersberg *et al*. 1983; Howard-Williams and Downes 1993; Kadlec 1995). Nitrogen uptake rates reported in the literature range from 0.16 to 0.71 g/m^2/d (van Oostrom and Russell 1994; Wood 1995; and Raman *et al*. 1999).

Experimental studies

Table 2 is a compilation of studies of nitrate removal in subsurface flow constructed wetlands, and contains thirteen references to previous bench- to full-scale research. The feed waters for these studies were mostly treated municipal effluent, but also included urban runoff (Reuter *et al*. 1989) and treated landfill leachate (WERF 2000). Nitrate reductions of 12 to 94% were achieved for influent nitrate concentrations of 0.1 to 105 mg/L. In some cases the authors reported total organic carbon concentration (TOC), biochemical oxygen demand (BOD) and chemical oxygen demand (COD). To create a basis for comparison, TOC was estimated by multiplying the BOD by 2.5. The TOC results will be discussed later.

Green and Verhoeven (1999) studied the effect of different hydraulic retention times (6, 12, 24, 48 and 120 hrs) on nitrate removal. They showed improvement with increasing residence time to 48 hrs, after which it was no longer beneficial. Gersberg (1983, 1984*a*,*b*) reported on the ability of different supplemental carbon sources to improve nitrate removal rates in SSF wetlands treating nitrified secondary effluent. Additions of methanol, plant mulch and primary effluent improved treatment performance to varying degrees. This work was concerned with determining

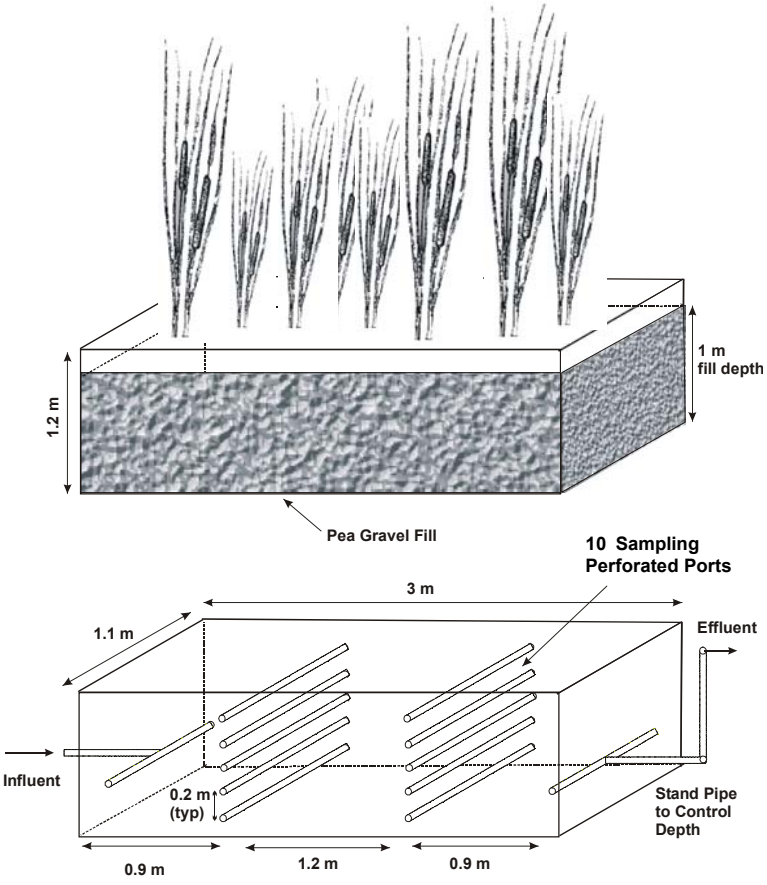

FIGURE 1. SCHEMATIC OF CONSTRUCTED PILOT WETLAND CELLS

the extent of nitrate removal possible in a subsurface flow wetland with nitrified tertiary effluent as influent, and the governing mechanism of nitrate reduction under these conditions.

MATERIALS, METHODS AND ANALYSIS

Materials

Two wetland cells (Figure 1) were constructed from plywood with dimensions of 3 m L × 1.1 m W × 1.2 m H each. Flow was controlled with 2.5 cm ball valves; a standpipe was used to control liquid level. Influent and effluent were distributed with perforated piping wrapped in nylon mesh that traversed the entire bed width. Sampling ports were located at depths of 0 cm, 20 cm, 41 cm, 61 cm and 81 cm from the top of the gravel bed. The interior of the boxes was lined with plastic, and pea-sized gravel (13 mm) was laid to a height of 1 m. The gravel was inoculated with organisms from the return sludge of a nitrifying activated sludge treatment plant. Porosity of the gravel was 40%. Dormant cattails (*Typha* sp.) were transplanted from a nearby pond at a density of 30 clumps per cell. The cells were located at the Tapia Wastewater Reclamation Facility (TWRF) in Calabasas, California.

Source of influent

Nitrified tertiary effluent obtained from the plant fed the pilot cells. The Tapia facility is a fully nitrifying 60 567 m^3/day (16 MGD) activated sludge plant. In addition to the nitrogen load in the influent wastewater, centrate from de-watering of the primary – and some part of the secondary – sludge is returned to the headworks for treatment. This centrate, which has ammonia concentrations from 500 to 800 mg/L and is pumped at the rate of 75 000 gallons per day, is a significant source of nitrogen.

Treatment at the Tapia Facility includes a single-stage nitrification process. In the summer months there is greater operational flexibility, and the process is modified to achieve partial denitrification. These modifications cannot be sustained during the wetter months when higher flows are received and the plant operates at full capacity. The continuous process modifications created variations in feed nitrate concentrations to the cells, which are shown later. Table 3 shows the composition of the plant's tertiary effluent for the period of study.

Table 3. Influent composition to the pilot wetland cells

Parameter	Concentration	Unit
pH	6.9	pH units
Temperature	21.4	deg C
Dissolved oxygen	7.5	mg/L
Biological oxygen demand	2.4	mg/L
Total organic carbon	9.0	mg/L
Total suspended solids	1.3	mg/L
Total dissolved solids	742	mg/L
Nitrate-N	10.5	mg/L
Nitrite-N	<0.01	mg/L
Ammonia-N	<0.2	mg/L
Organic nitrogen	0.6	mg/L
Phosphate	2.6	mg/L
Total coliform	<1.1	MPN/100 mL

Sampling

The wetland cells began operation in January 1999, once all the rhizomes were planted. Flow meters were initially used to control flow rate, but failed due to fouling and scale accumulation. A graduated bucket and stopwatch were later used to control flow. Standpipes at the effluent ports maintained water depth within the beds.

The two cells were operated at different hydraulic retention times (HRT) 18 and 48 hours ($Cell_{18}$ and $Cell_{48}$), and the flow rates were 1838 and 691 L/day, respectively. This resulted in different nutrient loading rates, as the nitrate concentrations for both influents were the same. These HRTs were selected based on results obtained from preliminary operation of the cells during March to September 1999. Beginning in October 1999, weekly composite samples of the influent and effluent were taken to determine nitrate removal. The sampling frequency increased to four 24-hour composite samples per week in January 2000. Phosphate and total organic carbon (TOC) analyses of influent and effluent began in February, with the same frequency as the nitrates. Samples for BOD analyses were collected a few times during the study

In order to determine concentration profiles of nitrate, ammonia, and TOC within the bed, samples were collected at all ten sampling ports on a bi-weekly basis, while phosphate samples were collected once per month. In addition, twice-weekly measurements of

grab samples for pH, temperature, dissolved oxygen and conductivity were taken. Daily maximum and minimum air temperatures were obtained from plant records, and, from January 2000, Optic Stowaway temperature loggers (Onset Computer Corp., Procasset MA) were placed within the gravel beds of the cells to continuously record temperature. Data from these loggers were downloaded using BoxCar Pro software (Onset Computer Corp., Pocasset, MA).

Acetate addition

Towards the end of the study period, acetate was used as a source of supplemental organic carbon, and was added to the pilot cells to determine its effect on performance, and to help shed light on the mechanisms involved in the nitrate removal. Sodium acetate trihy-

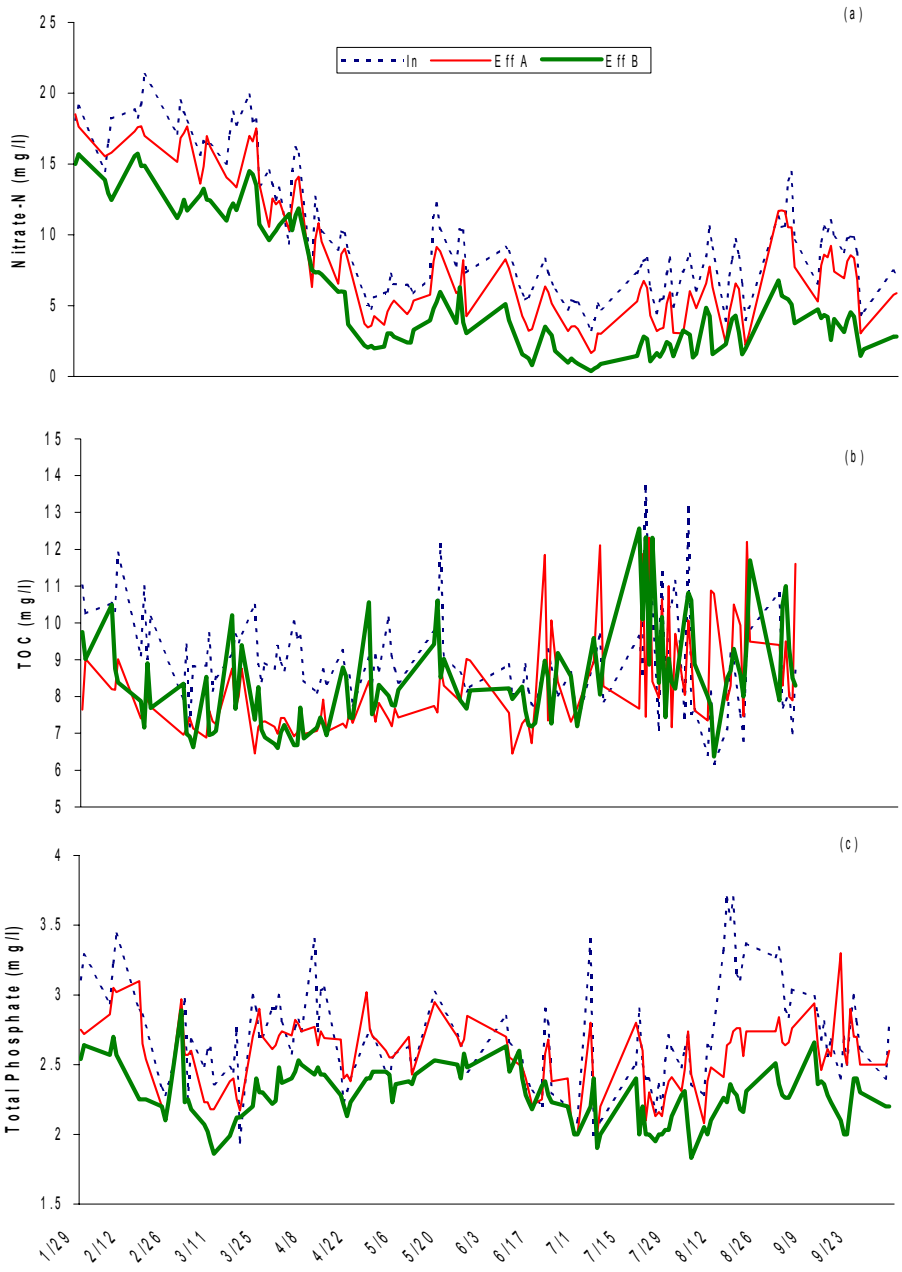

FIGURE 2. INFLUENT AND EFFLUENT CONCENTRATIONS OVER TIME: (A) NITRATE, (B) TOC, (C) TOTAL PHOSPHATE.

drate (NaAc) crystals dissolved in de-ionized water were used. The amount of acetate added for denitrification was 7.1 g NaAc/gN, and was based on stoichiometry (Constantin and Fick 1997). With an average influent of 8 mg/L NO_3 and 4.5 mg/L DO, 124.4 g NaAc/d and 46.7 g NaAc/day were estimated for the cells with HRTs of 18 and 48 hrs respectively.

Concentrated NaAc solution was placed in plastic containers and applied to each bed at the influent (Figure 1). The feed rate was 5 mL/min continuously for three weeks. During this period, profile samples were collected for nitrate analysis, while dissolved oxygen measurements were taken daily during tl last two weeks.

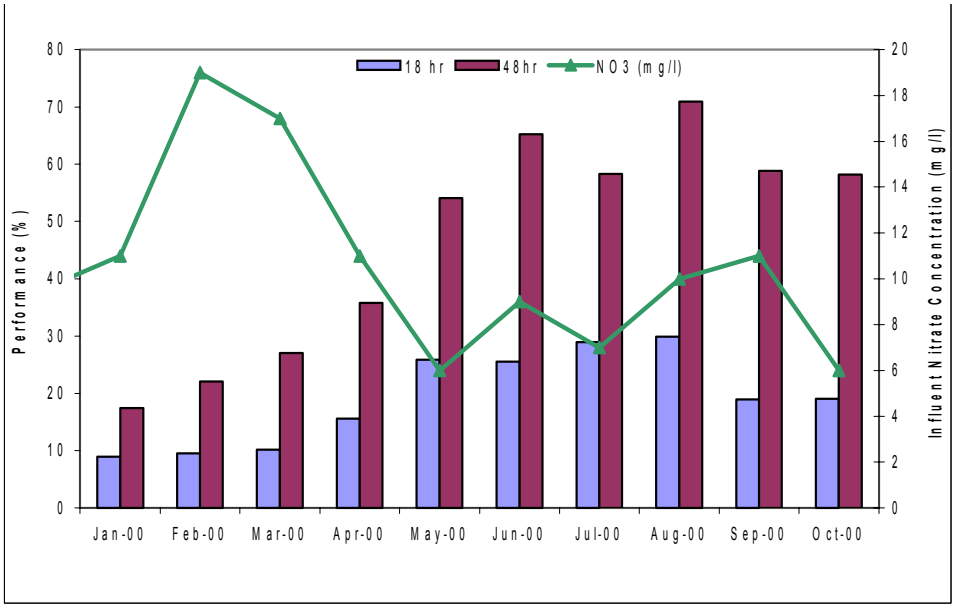

Figure 3a: Nitrate removal performance ot wetland cells at 18- and 48-hr HRTs

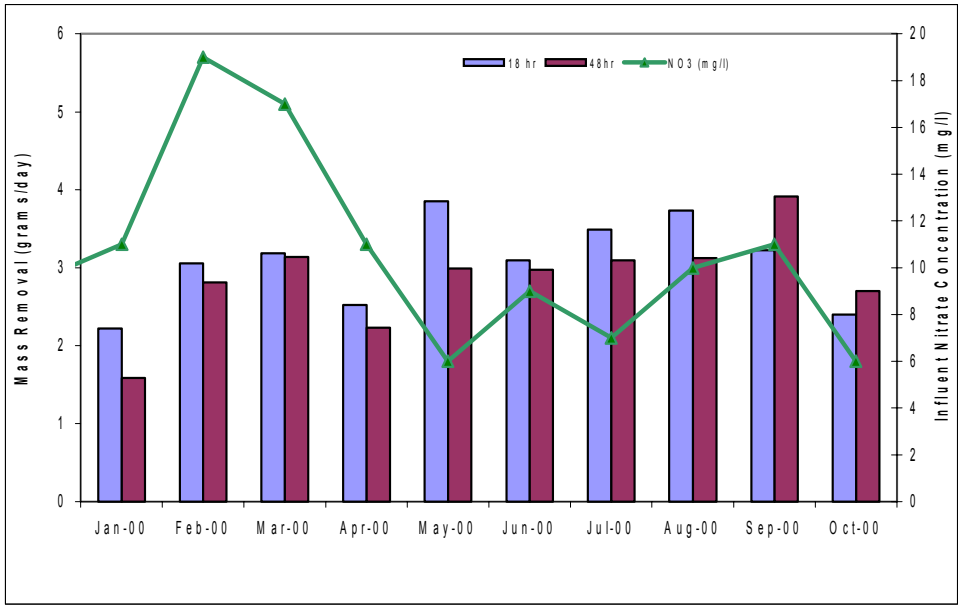

Figure 3b: Nitrate mass removal rates for wetland cells with 18hr- and 48hr HRTs

Table 4. Results of plant tissue analysis of cattails in pilot cells. N = 5 for each cell

Elements	$Cell_{18}$ plant tissue conc. (mg/kg) d.w.*	$Cell_{48}$ plant tissue conc. (mg/kg) d.w.	Influent conc. (mg/L)
Essential nutrients			
Nitrogen	10 092 ± 2168	5640 ± 3769	10.0
Phosphorus	2232 ± 691	1604 ± 565	2.7
Potassium	36 341 ± 9682	18 181 ± 5943	12.5
Priority pollutants			
Arsenic	<0.26	<0.26	<0.010
Barium	7.6 ± 3.2	13.7 ± 7.6	0.015
Cadmium	0.30 ± 0.07	0.18 ± 0.07	<0.010
Chromium VI	13.7 ± 12.2	17.7 ± 20.3	<0.010
Cobalt	0.73 ± 0.70	0.98 ± 0.95	<0.010
Copper	7.4 ± 2	5.4 ± 2	<0.008
Lead	< 0.62	<0.62	<0.010
Mercury	< 0.6	< 0.6	<0.010
Nickel	5.6 ± 4.5	6.6 ± 6.9	<0.010
Selenium	<0.98	<0.98	<0.010
Silver	<0.03	<0.03	<0.010
Zinc	238 ± 35	99 ± 46	0.058

* d.w.: dry weight

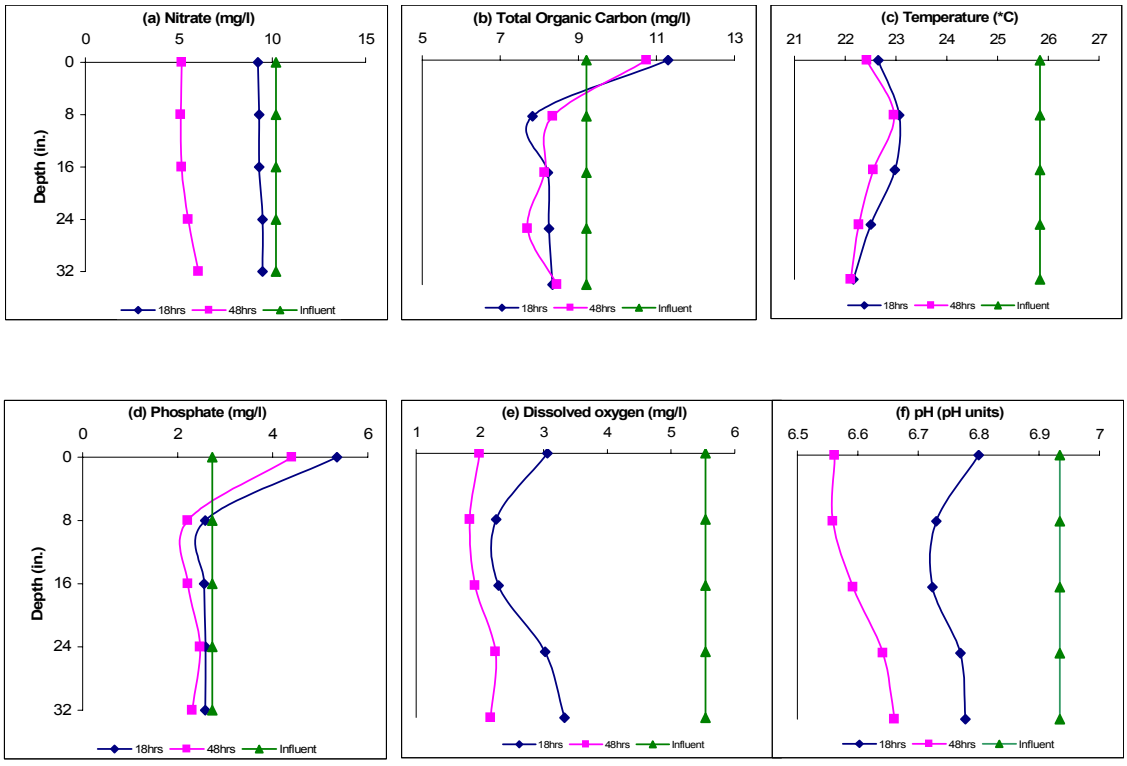

FIGURE 4. DEPTH PROFILES OF (A) NITRATE, (B) TOC, (C) TEMPERATURE, (D) PHOSPHATE, (E) DISSOLVED OXYGEN, AND (F) PH.

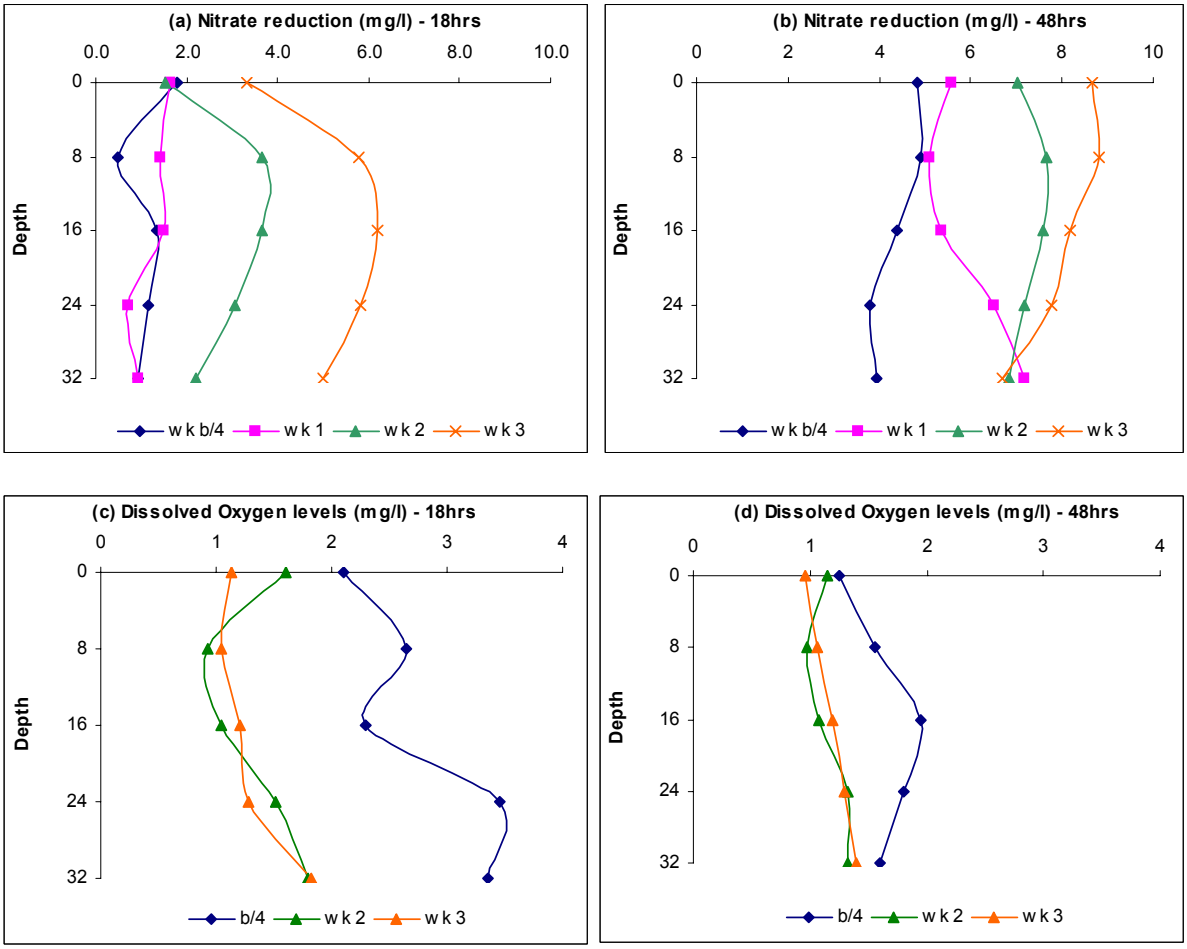

FIGURE 5. EFFECT OF SODIUM ACETATE ADDITION ON (A), (B) NITRATE REDUCTION, AND (C), (D), DO DEPLETION AT DIFFERENT HRTS

Water sample analysis

Samples were analysed according to Standard Methods (SM 1999) or EPA approved methodology. Nitrate- and nitrate-N were determined by the automated cadmium reduction method (SM 4500NO3 F) using a Technicon Auto-analyzer II system. Ammonia-N was analysed using the automated phenate method (SM 4500NH3 H) with the same Technicon system. Total phosphorus was determined using the colorimetric, ascorbic acid, two-reagent method (EPA 365.3). Samples for BOD analysis were filtered and then analysed using a five-day incubation at 20ºC (SM 5210 B). The combustion infrared method (SM 5310 B), using a total organic carbon analyser, was used to determine TOC concentrations. The TWRF laboratory performed all analyses. Dissolved oxygen and temperature were measured using a YSI model 57 Oxygen Meter.

Plant tissue analysis

Samples of the dormant cattails were collected for analysis during transplanting in January 1999. At the end of the study, in October 2000, more plant samples were collected from both pilot cells. Tissue analysis was also performed on cattails obtained from a natural site upstream of the treatment plant. Wallace Laboratories in El Segundo, California analysed all samples. About 10% of the total tissue representing the entire plant sample (i.e. the stems, leaves, roots and rhizomes) was cut into about two- to three-centimetre segments. The cut tissue was dried at 70ºC to a constant weight, and ground with a stainless-steel Waring blender and then reground with a Wiley mill. The tissue was digested using the EPA 3051 procedure. Elemental and heavy metals analysis was conducted on the digested tissue with a Thermo Jarrell Ash IRIS ICP (EPA 6010).

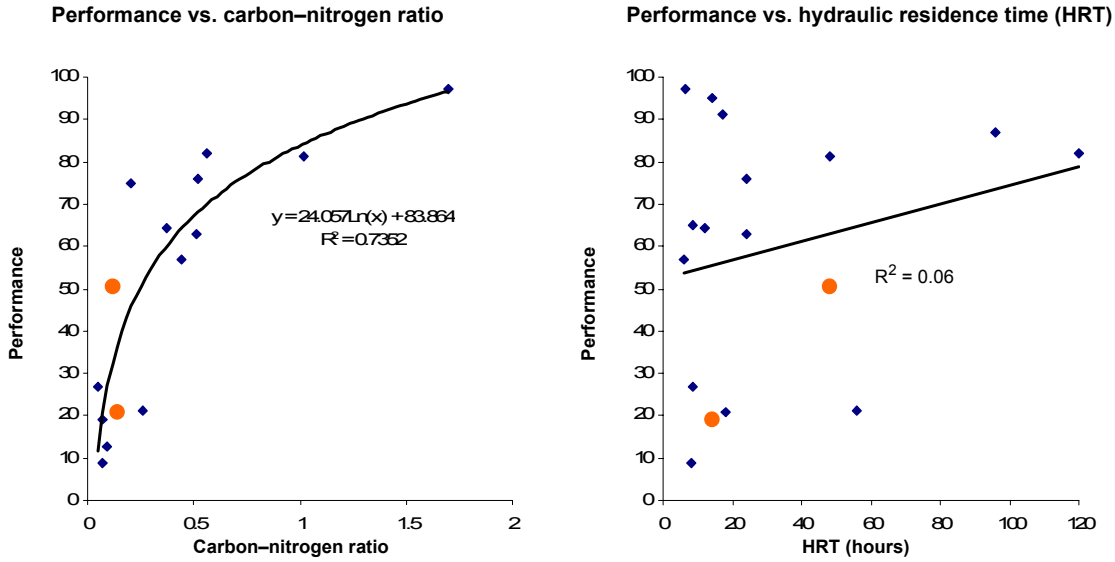

FIGURE 6. (A) IMPACT OF C:N RATIO ON PERFORMANCE; (B) IMPACT OF TEMPERATURE ON PERFORMANCE. DATA OBTAINED FROM STUDIES REPORTED IN TABLE 2. LARGE CIRCULAR DATA POINTS SHOW RESULTS FROM THIS STUDY.

Carbon and nitrogen were measured with a Carlo Erba 2500 NA analyser.

RESULTS

Nitrate removal

Figure 2a shows influent and effluent nitrate concentrations over the period of the study. Influent nitrate varied considerably according to season (3.3–21.3 mg/L) and averaged 10 mg/L. Concentrations were highest in the winter through early spring, but dropped in late spring, and remained low through the fall season. There were corresponding variations in the effluent nitrate concentrations (1.6–18.9 mg/L for $Cell_{18}$ and 0.4–15.7 mg/L for $Cell_{48}$). In a few instances during rainstorms, the effluent nitrate concentrations matched or surpassed that of the influent. Reduction in nitrate concentration was lowest in the winter, highest in September, and mostly constant throughout spring and most of sum-

Table 5. Nitrate budget for the wetland cells

	Cell 18		Cell 48	
	Low carbon	Supplemental carbon	Low carbon	Supplemental carbon
Input (grams/day)				
Cell influent	18.1	16	6.8	6.0
Removal (grams/day)				
Plant uptake	3.2	3.2	3.0	3.0
Denitrification	0	3.6	0	1.9
Output (grams/day)				
Cell effluent	14.9	9.2	3.8	1.1

mer. A regression of effluent concentration as a function of influent concentration for both cells (not shown) yielded the following relationships:

$$\text{Effluent}_{(18hr)} = 0.98\ (\text{Influent}) - 1.53\ (R^2 = 0.94,\ n = 127)$$

$$\text{Effluent}_{(48hr)} = 0.89\ (\text{Influent}) - 3.34\ (R^2 = 0.87,\ n = 127)$$

Per cent nitrate reduction was used to compare performance between the two cells. Figure 3a shows the monthly performance of both units over time. The variations are a result of the changing influent concentrations; low performance values coincided with high influent nitrate and vice versa. $Cell_{48}$ (50.2 ±15%) consistently out-performed $Cell_{18}$ (20.8 ±10%).

The beds were loaded at different rates: 18 g/d average and 7 g/d for $Cell_{18}$ and $Cell_{48}$, respectively. However, removal rates were similar for both units – 3.1 grams/day for $Cell_{18}$ and 3.0 grams/day for $Cell_{48}$ (Figure 3b). The difference between the removal rates was not statistically significant (95% CL). Rates were lowest in January, increased through May, and remained relatively constant throughout the rest of the study.

Nitrate profiles within the bed show similar concentrations throughout in $Cell_{18}$ (Figure 4a). However, in $Cell_{48}$, nitrate levels were lowest at the top of the bed and increased gradually towards the bottom. Levels of ammonia in all samples from both cells always remained below the detection limit (0.2 mg/L)

Carbon

The total organic carbon in the influent ranged from 6.2 to 13.7 mg/L, with an average of 9.0 mg/L. Effluent concentrations showed similar variation – 6.4 to 12.6 mg/L ($8.2_{ave.}$) for $Cell_{18}$ and 6.4 to 12.6 mg/L ($8.4_{ave.}$) for $Cell_{48}$ (Figure 2b). While influent values were mostly greater than effluent values for the first part of the study, from mid-June, the effluent TOC concentrations of both cells exceeded that of the influent several times. The average overall change in TOC concentration throughout the study was +0.8 mg/L for $Cell_{18}$ and +0.6 mg/L for $Cell_{48}$. T-tests showed no significant difference between the TOC levels in the effluents of the two cells (95% CL). Analysis of the profiles within the bed showed TOC up to 2 mg/L greater than influent values at the top of the bed (Figure 4b). These levels were reduced by 3–4 mg/L at the other depths. BOD values were generally equal to or less than 3 mg/L for both influent and effluent.

Phosphate

Figure 2c is a plot of influent and effluent total phosphate concentrations for both cells. Influent concentrations of phosphate ranged from 1.9 to 3.7 mg/L, with an average value of 2.7 mg/L. Phosphate removal was low in both cells. $Cell_{18}$ produced effluents with concentrations ranging from 2.3 to 3.3 mg/L (2.6 mg/L average), while $Cell_{48}$ had phosphate levels ranging from 1.8 to 2.9 mg/L (2.3 mg/L average). Overall, removal was consistently better in $Cell_{48}$ (95% CL). Phosphate profiles showed high concentrations at the top of the bed that declined with depth (Figure 4d).

Dissolved oxygen

Figure 4e shows the relationship between dissolved oxygen (DO) in the influent and within the cells. The drop in dissolved oxygen ranged from 3 to 4 mg/L (60–80%) in $Cell_{48}$ and 2 to 3 mg/L (40–60%) in $Cell_{18}$. These values did not drop below 0.5 mg/L at any point, therefore the water column was never anoxic. However, anoxic zones do occur in wetlands, for example within microbial films. In both cells, there is a reduction towards the middle of the bed, followed by an increase towards the bottom.

Temperature and pH

Temperatures within the bed were always 3 to 4ºC degrees cooler than the influent (Figure 4c). Figure 4f shows the changes in pH with depth over time, and the changes in relation to the influent value (pH/pHo) for both cells. There was a 0.2 to 0.4 drop in pH in $Cell_{48}$. The difference in $Cell_{18}$ was less, ranging from 0.1 to 0.2 pH units.

Sodium acetate (NaAc) addition

During the period of supplemental carbon addition, influent nitrate concentrations averaged 8.7 mg/L, and average effluent concentrations of $Cell_{18}$ and $Cell_{48}$ were 5 mg/L and 1.6 mg/L, respectively. There was a delay in reaction time once NaAc addition began. Initially, removal rates were 1.2 mg/L for $Cell_{18}$ and 5.9 mg/L for $Cell_{48}$. This increased to 5.2 mg/L and 8 mg/L, respectively, by the third week. Complete denitrification was not achieved, since a sudden change in plant

operations resulted in higher nitrate levels than had been estimated for carbon addition. In addition to this, it is likely that some of the carbon was used for processes other than denitrification, e.g. plant growth.

Mass removal rates of 6.9 g/d and 4.8 g/d were observed for $Cell_{18}$ and $Cell_{48}$, respectively. Figures 5a–b profile the effect of NaAc addition on nitrate removal in the pilot cells. The increase in nitrate removal indicates the systems' response to the availability of supplemental carbon. Changes in dissolved oxygen levels were also profiled. Significant reductions were observed in $Cell_{18}$ while reductions in $Cell_{48}$ were more modest (Figure 5c–d).

Plant analysis

Table 4 presents the results of the plant tissue analysis of the cattails in the pilot units, along with matching parameters of the cells' influent. Higher concentrations of nitrogen, phosphorus and potassium were found in plants from the cell with the 18-hr HRT when compared to plants from the 48-hr HRT cell. This is likely a result of the higher mass loading rate of $Cell_{18}$. All non-detectable metals in the plant tissues were non-detectable in the influent to the cells. However, cadmium, chromium VI, copper, and nickel, while below the detection limit in the influent, were detectable in the plant tissues. Concentrations of zinc were noticeably high in plant tissue from $Cell_{18}$. All the elements in the pilot cell plant samples occurred in concentrations comparable with or lower than the amounts detected in naturally occurring samples obtained from the creek adjacent to the treatment plant.

Literature data

Figures 6a and b show plots of HRT and carbon–nitrogen (C:N) ratios versus performance, using data from Table 2. Carbon values were derived from BOD concentrations provided by the literature. The analysis included data with influent nitrate concentrations of 12.6 to 26 mg/L and HRTs of 6 to 120 hrs. Figure 6a shows a modest correlation of $R^2 = 0.74$, suggesting that the C:N ratio affects performance to some degree. Hydraulic residence time showed no correlation ($R^2 = 0.06$) with performance when comparing different systems. Inclusion of the results of this study in the analysis showed that our systems' performance coincided with those studies with lower C:N ratios. The data were insufficient to perform multiple regression analysis.

DISCUSSION

Nitrate removal

Nitrate reduction is possible to some extent throughout the year in these wetland cells. Since the nitrogen is predominantly in the form of nitrate, denitrification and/or plant uptake should be the major removal mechanisms involved. However, to achieve the average removals observed in the cells by denitrification, 9 mg/L and 22 mg/L BOD would be required in $Cell_{18}$ and $Cell_{48}$, respectively. Influent analysis showed BOD levels ranging between <2 to 3 mg/L, which are insufficient to support this removal mechanism.

In addition, reductions of this magnitude were not observed upon analysis of effluent TOC. Samples collected over time revealed little change between influent and effluent concentrations for both 18- and 48-hour detention times. The type of carbon present in the highly polished influent entering the bed is less prone to degradation, and hence is not able to facilitate denitrification. There was also a possibility that organic carbon is being produced within the bed, from rhizome secretions and decomposition of plant material. However, continuous analysis of samples obtained from within the cells did not show significant increases in TOC levels.

With the addition of NaAc, it took over seven days for significant changes in nitrate removal to occur. This suggests that there was little denitrification activity, and that a denitrifying population needed time to develop in the presence of increased carbon availability. These results also show the potential for increase in denitrification rates within the bed when a carbon source is made available.

In the absence of a usable carbon source for denitrification, uptake by plants is the more likely removal mechanism. Average annual rates of nitrogen uptake have been reported in the range of 0.16–0.71 $g/m^2/d$ (DeBusk and Reddy 1987; Howard-Williams and Downes 1993; van Oostrom and Russell 1994; Wood 1995; Raman *et al*. 1999). Removal rates in this study averaged 1 $g/m^2/d$, obtained by dividing the mass removal rates by the surface areas of the wetland cells.

The lower values in the winter and higher removal rates in late spring and summer, are consistent with the literature stating that the highest uptake rates occur in spring and early summer (Howard-Williams and Downes 1993; Kadlec 1995). However, there was no sign of the release, which is believed to occur during fall and winter. This could be explained by the continuous production of new shoots within the cells during the fall and, at a slower rate, during the relatively warm southern Californian winter.

In a series of small-scale greenhouse experiments using labelled ^{15}N, Zhu and Sikora (1995) observed that plant uptake was the predominant nitrate removal process (70–85%) for cells planted with bulrush (*Scirpus* sp.) and cattails (*Typha* sp.) in the absence of carbon. With supplemental carbon, denitrification and immobilization became the major mechanisms (55–70%). Gersberg *et al*. (1983) observed that nitrate removal efficiencies were greatest in their vegetated beds during the spring and summer period when plant growth and nutrient uptake were greatest. However, there was also a corresponding increase in the unvegetated cells during the same period, which implied that plant uptake was negligible. Algal production of carbon (Zhu and Sikora 1995; Wood *et al*. 1999) and immobilization (Zhu and Sikora 1995) were suggested as mechanisms of nitrate removal in unplanted cells without supplemental carbon.

Wood *et al*. (1999) were able to achieve a 90% reduction in synthetic solutions containing 100 mg/L nitrate in bench-scale SSF constructed wetlands in the absence of carbon. They attributed this to the availability of sufficient internal carbon (including plant root exudates and decomposing plant material) provided by the plants (*Iris pseudocoras*). Such high performance could be a result of the rapid plant uptake that occurs in new wetland systems as the plants become established. A similar effect was observed during the first year of this study.

These results can be summarized in a nitrate budget for the wetland cells, based on the following equation:

$$O = I - PU - DN$$

where O is the outflow of nitrate (grams/day) from the cells, I is the inflow of nitrate, PU is plant uptake, and DN is denitrification. The budget for nitrate is presented in Table 5. The inflow and outflow values were derived from influent and effluent nitrate concentrations and flow volumes. Atmospheric deposition was not included in the input, as it was determined to be insignificant (<0.1% and <0.4% of inputs to $Cell_{18}$ and $Cell_{48}$ respectively). These values were based on estimates of wet and dry deposition rates for nitrogen in the Malibu Creek watershed (Tetra Tech 2001). As discussed earlier, plant uptake was the dominant removal mechanism when there was insufficient carbon. Upon addition of NaAc, the impact on denitrification is evident. However, since the addition of carbon was only for a limited time, the full denitrification potential was not reached.

The nitrate data were not fitted to a denitrification model, as the predominant removal mechanism within the bed was plant uptake, which is a constant. This constant is evident from the regression equation presented in the results section.

Phosphate removal

The low phosphate removal by the cells is consistent with what has been reported in literature. Removal mechanisms for phosphates in subsurface flow wetlands include sorption on to substrate, plant uptake, and bacterial assimilation. In subsurface flow beds, sorption is limited by the large size of the media that results in reduced available surface area, and saturation is soon reached.

The ability of $Cell_{48}$ to remove more phosphate may have been due to the additional time that would allow for additional plant uptake or adsorption. It is interesting to note that high concentrations of phosphate, at the top of the bed, indicate some form of release from the plant material However, there was no corresponding increase in nitrogen, and therefore, the source of this increase is unclear.

To effectively remove phosphorus, media with smaller grain sizes, such as clay, could be used, but these result in significant hydraulic problems. Addition of lime and alum (Davies and Cottingham 1993), and coating gravel with iron or aluminium oxides (Mann 1990) have been suggested as other means of improving phosphorus removal performance.

CONCLUSIONS

These results indicate that a constructed wetland fed with low-carbon tertiary effluent can achieve nitrate reduction. In systems such as these, with minimal supplemental carbon, plant uptake is likely the major mechanism involved in nitrate reduction. While there is probably some denitrification occurring, the lag time observed in both cells' response to supplemental carbon addition indicates that if any denitrification population was present, it was not very large. With the passage of time and the build-up of organic carbon within the beds, denitrification would play a more significant role in nitrate removal. This would occur when the vegetation has come to equilibrium and plant uptake rates match release rates.

With a nitrate removal rate of 1 g/m^2/d, the available land area could remove up to 2.6 kg/d of nitrogen without supplemental carbon. In the event that greater removal is required, increasing the detention time of the system should result in improved treatment. However, this may be uneconomical in areas where land acquisition costs have to be taken into consideration. No significant reduction in phosphorus is expected for this system.

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