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Denitrifying bioreactors—An approach for reducing nitrate loads to receiving waters

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ABSTRACT

Low-cost and simple technologies are needed to reduce watershed export of excess nitrogen to sensitive aquatic ecosystems. Denitrifying bioreactors are an approach where solid carbon substrates are added into the flow path of contaminated water. These carbon (C) substrates (often fragmented wood-products) act as a C and energy source to support denitrification; the conversion of nitrate (NO_3^-) to nitrogen gases. Here, we summarize the different designs of denitrifying bioreactors that use a solid C substrate, their hydrological connections, effectiveness, and factors that limit their performance. The main denitrifying bioreactors are: denitrification walls (intercepting shallow groundwater), denitrifying beds (intercepting concentrated discharges) and denitrifying layers (intercepting soil leachate). Both denitrifcation walls and beds have proven successful in appropriate field settings with NO₃⁻ removal rates generally ranging from 0.01 to $3.6 \text{ g N m}^{-3} \text{ day}^{-1}$ for walls and $2-22 \text{ g N m}^{-3} \text{ day}^{-1}$ for beds, with the lower rates often associated with nitrate-limitations. Nitrate removal is also limited by the rate of C supply from degrading substrate and removal is operationally zero-order with respect to NO₃⁻ concentration primarily because the inputs of NO₃⁻ into studied bioreactors have been generally high. In bioreactors where NO₃⁻ is not fully depleted, removal rates generally increase with increasing temperature. Nitrate removal has been supported for up to 15 years without further maintenance or C supplementation because wood chips degrade sufficiently slowly under anoxic conditions. There have been few field-based comparisons of alternative C substrates to increase NO₃⁻ removal rates but laboratory trials suggest that some alternatives could support greater rates of NO_3^- removal (e.g., corn cobs and wheat straw). Denitrifying bioreactors may have a number of adverse effects, such as production of nitrous oxide and leaching of dissolved organic matter (usually only for the first few months after construction and start-up). The relatively small amount of field data suggests that these problems can be adequately managed or minimized. An initial cost/benefit analysis demonstrates that denitrifying bioreactors are cost effective and complementary to other agricultural management practices aimed at decreasing nitrogen loads to surface waters. We conclude with recommendations for further research to enhance performance of denitrifying bioreactors.

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1. Introduction

The nitrogen (N) cascade is an increasingly important global issue as N flowing through ecosystems has multiple impacts on terrestrial, aquatic and atmospheric environments (Galloway et al., 2003, 2008). In agricultural systems, N in excess of plant and animal needs can leach to shallow groundwater and ultimately enter surface waters through concentrated or diffuse discharges. Concen-

* Corresponding author. Tel.: +64 7 858 3700; fax: +64 7 858 4964. *E-mail address*: Schipper@waikato.ac.nz (L.A. Schipper). trated discharges in agricultural systems occur through under-field tile drainage and ditches (Duda and Johnson, 1985; Dinnes et al., 2002) while diffuse sources are typically through the discharge of shallow groundwater to surface waters. Nitrogen that is captured in biomass passes through the food chain and ends up in wastewater streams, which are ultimately discharged to surface waters.

A number of strategies are being implemented to reduce the N load to aquatic ecosystems. Sophisticated technologies have been developed to remove N from wastewater and are employed in municipal treatment plants and septic tank systems (Oakley et al., this issue). In agricultural ecosystems, many land management approaches have been proposed to reduce N losses, such as

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improved N use efficiency in crops, managing N inputs through soil tests, land management plans and controlled drainage (Sims et al., 1995; Drury et al., 1996; Dinnes et al., 2002; Jaynes et al., 2004). Integrating wetland and riparian buffers into the agricultural landscape have also been demonstrated as a potentially useful way to reduce N losses to surface waters (Hill, 1996; Kadlec, 2005).

The most widely understood process of permanent N removal from terrestrial and aquatic ecosystems is heterotrophic denitrification, which converts nitrate (NO_3^-) to N gases using a carbon (C) source as the electron donor and for growth (Seitzinger et al., 2006; Coyne, 2008; Rivett et al., 2008). In this review, we focus on heterotrophic denitrification but we recognise that other microbial processes such as anaerobic ammonium oxidation (Anammox) and chemo-autotrophic denitrification can also produce N gases (Burgin and Hamilton, 2007). The rates and controlling factors of these microbial processes deserve further attention, but are not covered in this review.

At the microbial scale, the rate of heterotrophic denitrification is controlled by concentrations of oxygen (O₂), NO₃⁻ and C (Seitzinger et al., 2006). The availability of a degradable C source to drive denitrification becomes critical where NO₃⁻ is present in excess, such as in many wastewater plants and agricultural settings. Aerobic microorganisms obtain energy through the oxidation of organic compounds, using O_2 as the electron acceptor, until the environment becomes energetically favourable for the use of NO₃⁻ as an electron acceptor. As such, organic C plays two key roles in promoting denitrification; firstly, to provide an anoxic environment and, secondly, to act as an electron donor for denitrification. In wastewater treatment systems, this C is part of the waste stream, but can also be supplemented with liquid C sources such as methanol (Oakley et al., this issue; Henze et al., 2008). In agricultural soils, denitrification can be limited by sufficient labile C to create an anoxic environment and provide energy for denitrification.

Here, we review passive technologies that have been recently developed to overcome the C limitation of denitrification for enhanced NO₃⁻ removal. A variety of carbonaceous solids and immiscible liquids have been successfully tested in denitrifying bioreactors (Hunter, 2005; Gibert et al., 2008), although many of C sources have only been tested in laboratory trials. To date, wood-particle media in particular, has been the most widely used material in field trials and has shown an ability to deliver consistent longer term (5–15 years) NO₃⁻ removal, while requiring minimum maintenance (Robertson et al., 2000, 2008, 2009; Schipper and Vojvodic-Vukovic, 2001; Fahrner, 2002; Schipper et al., 2005; Jaynes et al., 2008). Consequently, this review has a strong focus on the use of wood-particle media, although other carbonaceous solids, including some with higher reaction rates, could also play an important role as additional experience is gained. Our focus is on field trials of solid C substrates that have been used in agricultural and rural settings to enhance denitrification and decrease discharges of NO₃⁻ from either diffuse discharges (e.g., shallow groundwater) or concentrated discharges (e.g., effluents, tile drainage, small streams and ditches). Because these approaches are intended to be applied throughout the landscape at the scale of individual fields, septic systems and tributary streams it is important that the technologies are simple, passive and that maintenance requirements are minimal.

2. Terminology

There are multiple designs that use solid C sources for enhancing denitrification and are collectively referred to as denitrifying bioreactors (Figs. 1 and 2; Tables 1 and 2). Designs differ primarily in the hydrologic connection between water containing NO_3^- and C source and the ratio of source area:treatment area. Broadly, we use terminology first used by Robertson and Cherry (1995).

"Denitrification walls" are where solid C material is incorporated vertically into shallow groundwater perpendicular to the flow. Darcian flow, saturated hydraulic conductivity, hydraulic gradient and the flow paths intercepted by the walls controls the flux of NO₃⁻ into the walls. Walls may intercept natural groundwater flow paths, or groundwater flow paths that have been altered by subsurface tile drainage systems or by the morphometry and relatively higher saturated hydraulic conductivity of C additions within the wall. The source area is roughly limited to the boundaries of the wall orthogonal to the flow direction. While the term "wall" suggests a barrier to flow, these walls are designed to sustain elevated hydraulic conductivities (i.e. >10 m dav $^{-1}$) conducive to substantial rates of shallow groundwater flow. Denitrification walls can be 100% wood chips (Farhner, 2002; Jaynes et al., 2008) or sawdust mixed with soil (Robertson and Cherry, 1995; Schipper and Vojvodic-Vukovic, 1998).

"Denitrification beds" are containers (sometimes lined) that are filled with wood chips and receive NO₃⁻ in concentrated discharges either from a range of wastewaters (Robertson et al., 2005a; Schipper et al., this issue) or tile/drain discharge (Blowes et al., 1994; Robertson et al., 2009; Robertson and Merkley, 2009). Denitrification beds have also been installed into existing stream beds or drainage ditches and are specifically referred to as "stream bed bioreactors" (Robertson and Merkley, 2009). The source area:treatment area ratio for beds is usually much greater than in wall designs, due to either natural or artificial drainage networks that intercept and funnel groundwater inputs to the bioreactor. Beds, referred to as "upflow bioreactors" have also been adapted for use along streambanks (van Driel et al., 2006a,b). These upflow bioreactors do not receive input from a specific point source, but have design features and placement requirements that induce focused flowpaths that resemble the high flux conditions associated with point sources. Upflow bed reactors rely on wood chips with high saturated hydraulic conductivity and create a favourable hydraulic gradient by lowering the water table within the bed through the placement of a drainage pipe near the top of the wall that discharges directly to the adjacent stream.

Finally, "denitrification layers" are horizontal layers of solid C material that have been installed under weeping tiles from septic tank drainage fields (Robertson and Cherry, 1995) or under effluent-irrigated topsoils (Schipper and McGill, 2008).

The key to selecting an appropriate bioreactors design depends on the hydrologic conditions and site constraints of the system of interest (Table 2, and see Section 4).

3. Removal rates and controlling factors

3.1. Removal rates

Nitrate removal rates have been reported for a wide range of denitrifying bioreactors (Table 3) using different units. For consistency, in this synthesis paper, NO_3^- removal rates are expressed as g NO_3 -N removed per m⁻³ reactor volume per day (g NO_3^- N m⁻³ day⁻¹). Where conversion of rate units was required for study comparisons (Table 3), an effective porosity value of 0.7 (van Driel et al., 2006a) was used in most cases. It is also possible to express NO_3^- removal rates based on surface area of the bioreactor; this can be useful when wanting to compare the performance of bioreactors to NO_3^- removal in wetlands or other terrestrial ecosystems (for example, where effluent is applied to land).

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Fig. 1. Schematic of different designs of denitrification walls: (A) plan view of a wall intercepting NO₃⁻ plume, (B) side view of wall installed to impervious layer forcing shallow groundwater through wall, (C) wall not installed to full depth to impervious layer allowing flow paths under, (D) high permeability layer installed in upper part of aquifer causing upwelling of groundwater containing NO₃⁻, and (E) denitrification walls installed on either side of a subsurface tile drain.

Sustained NO₃⁻ removal rates for denitrification beds incorporating wood, range from about 2 to 22 g N m⁻³ day⁻¹. Variation in rate is predominantly attributed to bed operating temperatures (typically from 2 to 20 $^\circ\text{C}$) and/or influent NO_3^- concentrations (see fuller discussion below). The highest sustained NO₃⁻ removal rates were measured by Blowes et al. (1994) and Robertson et al. (2000) in a denitrification bed (North Campus, Canada) using woodchips. Removal rates for this trial varied between 4 and $22 \text{ gN} \text{ m}^{-3} \text{ day}^{-1}$ depending on temperature (2–20 °C). Other denitrification bed studies (van Driel et al., 2006a,b; Schipper et al., this issue) utilizing a combination of sawdust and woodchip media report a slightly lower rate of NO3⁻ removal, varying between 2 and 20 g N m⁻³ day⁻¹. Robertson et al. (2005a) measured lower removal rates $(2-5 \text{ gN} \text{ m}^{-3} \text{ day}^{-1})$, during evaluation of the commercially available NitrexTM filter which contains a mixture of sawdust and woodchips; however, removal rates were nitrate-limited at these sites.

Nitrate removal rates for denitrification walls (Robertson et al., 2000; Schipper et al., 2005; Jaynes et al., 2008) containing wood media are generally an order of magnitude lower $(0.014-3.6 \text{ g N m}^{-3} \text{ day}^{-1})$ than denitrification beds. Nitrogen removal rates within walls may be limited by low rates of NO₃⁻ loading, as most walls removed virtually all the NO₃⁻ but also

because walls can have wood mixed with inert material such as soil or sand. The highest reported removal rate $(15 \text{ g N m}^{-3} \text{ day}^{-1})$ was for a 100% sawdust wall constructed in Western Australia (Fahrner, 2002), which received very high NO₃⁻ concentrations and had relatively high soil temperatures (Table 3); all of which would promote relatively high denitrification rates.

We have not addressed the importance of the dominant denitrifying organisms in the denitrifying bioreactors but these organisms are very wide-spread (Coyne, 2008), and bioreactors studied to date have not required inoculation as the denitrifiers respond rapidly to environmental drivers. However, microbial population diversity and dynamics deserve further attention as appropriate molecular tools are developed (Wallenstein et al., 2006).

3.2. Nitrate concentration

While denitrification is likely to obey Michelis–Menton kinetics with regard to NO_3^- concentration, most denitrifying bioreactors receive NO_3^- concentrations higher than Km of denitrifying bacteria (see Barton et al., 1999). Consequently, when considering whether NO_3^- removal follows either zero-order and first-order kinetics, the situation may be viewed as functionally similar to zero-order kinetics in many cases (Robertson et al., 2000; Schipper

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Fig. 2. Schematic of different designs of denitrification beds: (A) side view of bed treating concentrated discharges of effluent or drainage water, (B) plan view of bed installed to intercept drainage water from agricultural land, (C) plan view of bed installed to intercept drainage water leaching from septic tank drainage fields, and (D) bed installed into base of stream.

et al., 2005), although first order approaches have also been used in some studies (Chun et al., 2009; Leverenz et al., this issue). There is experimental evidence to support the notion that $NO_3^$ removal in denitrifying bioreactors is operationally zero-order. In a series of column tests using aged woodchip media (fresh to 7 years old), successive runs at increasing influent NO_3 -N concentrations (3.1–49 mg L⁻¹) did not result in increased NO_3^- removal (Robertson, this issue). This indicated that the reaction was controlled by an independent parameter (presumably the release rate of degradable C from the carbonaceous media) and thus zeroorder reaction kinetics would apply over this concentration range. A transition to first-order kinetics may occur at lower NO_3^- concentrations (such as might be found in stormwater), but in most settings, zero-order kinetics likely represents much of the $NO_3^$ transformation, and could be used for most design purposes.

3.3. Alternative C sources

Field-scale denitrification bed and wall studies have mainly used wood products (sawdust and wood chips) as a C source, generally because wood is commonly available at low cost, supports high permeability, has a high C:N ratio (ranging from 30:1 to 300:1 depending on source and type of wood material; Gibert et al., 2008; Vogan, 1993) and long durability (Robertson et al., 2009). There have been a number of smaller-scale laboratory experiments, which have examined the NO₃⁻ removal rates in a range of other C substrates (Table 4 and see also Gibert et al., 2008). Some of the highest reported NO₃⁻ removal rates (19–105 g N m⁻³ d⁻¹) are from the column studies of Vogan (1993) and Shao et al. (2009) using cellulose, alfalfa, wheat straw, and rice husk as the C substrate.

While more labile C sources (e.g., cracked corn, corn stalks, straw, etc.) may support higher removal rates than wood media, these may require more frequent replenishment because of rapid C depletion. For example, Stewart et al. (1979) found that humus rich soil was ineffective for long-term NO_3^- removal from septic tank effluent due to relatively rapid depletion of available C. Decreases in the saturated hydraulic conductivity of the more labile C sources may also occur as the C structure breaks down. Cameron and Schipper (this issue) found that while maize cob media sup-

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Removal of NO3- from

ground water prior to

Objective

Table 1

Nomenclature

Denitrification wall

rilysical description and potential settings for different defittinging bioreactor of	r designs.
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Physical description

upper 1–2 m of shallow

(i) C substrate placed into the

	groundwater in a trench perpendicular to groundwater flow path towards surface water.	surface water recharge	groundwater; at focal points of groundwater flow.	(Schipper et al., 2005), Australia (Fahrner, 2002)
	(ii) C substrate placed into the upper 1–2 m of shallow groundwater in a trench on either side of a tile drain	Removal of NO3 [–] from groundwater before entering drainage network	In subsurface drained agricultural land	Jaynes et al. (2008)
Denitrification bed	Container (varied length and breadth dimensions but typically 1–2 m deep) filled with solid C substrate. Effluent or drainage water enters and exits in pipes. Beds may be lined.	Removal of NO ₃ - from wastewaters or tile drainage from agricultural fields.	Concentrated discharges that have high NO ₃ - concentrations such as from tiles or treated wastewater	van Driel et al. (2006a) and Schipper et al. (this issue)
Upflow bioreactors (subset of denitrification beds)	C substrate in container with lined sides, open to groundwater flow at bottom. Groundwater flows towards C substrate with elevated saturated hydraulic conductivity and is discharged to adjacent stream via pipe.	Removal of NO ₃ from ground water prior to surface water recharge	Adjacent to surface water where groundwater is shallow and aquifers have lower conductivities than added C substrate	van Driel et al. (2006b)
Stream-bed bioreactor (subset of denitrification beds)	Container (varied length and breadth dimensions but typically 1–2 m deep) filled with solid C substrate installed in the base of a stream	Reducing NO₃ ⁻ concentrations in streams	Streams and drainage ditches	Robertson and Merkley (2009)
Denitrification layer	A horizontal layer of woodchips that receives nitrified effluent from above.	Reduce NO₃ [−] leaching vertically to groundwater	Below septic wastewater drainage field that passed through a sand/gravel filter or other land-based effluent treatment system	Canada (Robertson et al., 2000) New Zealand (Schipper and McGill, 2008)

ported a 6.5-fold higher NO_3^- removal rate than wood media over a 24-month period, the decline in saturated hydraulic conductivity was generally greater in the maize cob media.

The effect of media particle size on reaction rates has also been considered. Several studies have found no significant difference in the NO₃⁻ removal rates in wood-particle media of different particle sizes (Carmichael, 1994; van Driel et al., 2006a,b; Robertson et al., 2000; Cameron and Schipper, this issue), although Greenan et al. (2006) measured initially greater NO₃⁻ removal for both wood chips and cardboard when ground to <2 mm. In comparing two denitrification beds, one constructed with predominantly coarse-grained wood particles (woodchips, 1-50 mm) and the other constructed with fine grained wood particles (sawdust, 1-5 mm), van Driel et al. (2006a) measured NO₃⁻ removal rates of $5.9 \,\mathrm{gN}\,\mathrm{m}^{-3}\,\mathrm{day}^{-1}$ for coarse grained media and a similar rate of 5.5 g N m⁻³ day⁻¹ for fine grained media. Robertson et al. (2000) speculated that denitrification is associated with reaction rims that penetrate, by diffusion, into the carbonaceous solids, rather than being restricted to the grain surfaces. This was supported by examination of a denitrification bed after 4 years of operation, which revealed that the larger centimeter-sized wood particles exhibited dark coloured rims that extended several millimetres into the particles, while the centre of the particles remained light coloured and appeared unaltered from their original condition. They interpreted this to indicate that the dark rims represented the denitrification zone. The smaller woodchips were dark coloured throughout.

There is no evidence of a difference in the NO₃⁻ removal rate achieved by hardwood in comparison to softer, faster growing tree species (Gibert et al., 2008; Cameron and Schipper, this issue). It is possible that hardwood may maintain physical properties for longer due to greater density. However, hardwood is not as readily available in some areas and may be more expensive. Both hardwood and softwood species have been used successfully in field trials (Robertson et al., 2000; Schipper and Vojvodic-Vukovic, 1998; van Driel et al., 2006a,b; Jaynes et al., 2008) with little advantage indicated of one over the other.

While laboratory-scale experiments provide opportunity for relative comparison of the NO₃⁻ removal potential and hydraulic performance of different C substrates, these studies may not be a reliable indication of removal rates or hydraulic performance achievable in larger-scale field installations. This is due in part to the effect of dissolved O_2 (DO) content of the influent water on removal rates in small scale trials. Also laboratory trials tend to be of short duration, typically less than 6 months and NO₃⁻ removal rates tend to decline with time as labile C is reduced (Schipper and Vojvodic-Vukovic, 1998). The results of short-term experiments may not be reliable for assessing longer term sustainablity of removal rates (Cameron and Schipper, this issue). Multiyear field testing of wood media in bioreactors has shown that rates measured after about 1 year of operation are generally representative on long-term operation (Robertson et al., 2000; Schipper and Vojvodic-Vukovic, 2001; Jaynes et al., 2008). Recommended best practice would be field testing of favourable substrates identified from laboratory studies for a minimum of a year. Ultimately the selection of an appropriate C media is a balance between availability, cost and reaction rate.

3.4. Temperature

In general, biological reactions rates increase with increasing temperature. Examination of reaction rates from a variety of studies where NO_3^- was non-limiting (Table 3) support a general

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Table 2

6

Hydrological connections, limitation and potential approaches to overcoming limitations in denitrifying bioreactors.

Nomenclature	Hydrological connections	Limitations	Potential for overcoming limitations
Denitrification wall	(i) Down-gradient of NO_3^- source. Flow into wall is governed by Darcian principles of groundwater flow. Pore water velocities are likely to be low (0.05–0.5 m day ⁻¹) and retention times within the wall are expected to be 3–10 days. Retention time within the wall is governed by incoming flow rate (passive). Hydrologic properties are likely to display high spatial (i.e., <i>K</i>) and temporal (i.e., hydraulic gradient) variability.	Requires site-specific analyses of hydraulic gradient, the depth and extent of NO ₃ ⁻ plumes. Removal of NO ₃ ⁻ limited to up-gradient source areas and within the upper 1–2 m of groundwater. If wall has lower saturated hydraulic conductivity than surrounding aquifer, NO ₃ ⁻ plumes may flow under or around the wall.	In aquifers > 2 m deep, upwelling of NO ₃ - laden groundwater into the interceptor wall may be enhanced by increasing the width (parallel to flow path) of the interceptor wall.
	(ii) For walls adjacent to tile drains, groundwater flow moves through bioreactors following drainage flow lines. Retention times are limited when water tables are highest and flow is greatest	For walls adjacent to tile drains, high flows with high NO3 [–] concentrations lead to short retention times resulting in large losses of NO3 [–] .	
Denitrification bed	For tile drains: Flow rate is governed by properties of subsurface drainage network including: area of drained land, pattern and extent of excess rainfall or irrigation, designed depth of water table decrease due to drainage system, and intensity of drainage network. Retention time within the bed is governed by incoming flow rate. <i>For wastewater</i> : flows controlled by upstream waste generation and wastewater plant man generation	Seasonality of flow rates can create high flow situations with limited retention rates within the bed, limiting NO3 ⁻ removal.	High flow bypass can be incorporated into design to minimize flooding and overflow within the ditch. Cellular designs can be coupled with flow diverters to optimize retention times at different flow regimes.
Upflow bioreactor	Groundwater upwells into open bottom of reactor: (i) C substrate has higher conductivity than the aquifer. (ii) Drainage pipes installed at upper portion of reactor discharges to adjacent stream, lowers water table within reactor and enhances hydraulic gradient to unflow reactor.	Very site specific applications. Stream side location may be prone to erosion; flow rate is effected by stream stage	'Rip-rap' can be used to control erosion
Stream-bed bioreactor	A bioreactor installed in bottom of stream with a down-gradient riffle creates a pressure gradient and stream water flows down through lower conductivity C substrate and through an exit pipe back into the lower reach of the stream	Seasonal flows over the top of the riffle resulting in partial treatment. Potential for siltation of surface inlet requiring cleaning	Rip rap cover and channel narrowing can minimize siltation
Denitrification layer	Loading is determined by the design of the septic system.	Extent of nitrification in preceding sand/gravel filter can limit N removal. Difficult to replace C substrate because underneath discharge.	Ensure appropriate pre-treatment, e.g., sand filter

Table 3

Rates of NO_3^- removal for a range denitrifying bioreactors in the field. In general average rates of NO_3^- removal are presented. Many of the systems recorded here had complete NO_3^- removal which would limit the rate of denitrification and consequently are likely underestimates of potential removal rate. Units are g N m⁻³ d⁻¹ where m⁻³ refers to volume of bioreactor.

System design	Study	Size of bioreactor (m ³)	Typical NO ₃ ⁻ inputs (g N m ⁻³)	Temperature annual average (°C)	Average rate of N removal $(gN m^{-3} d^{-1})$
Walls	Robertson et al. (2000)	1	50	14	1.7
	Schipper et al. (2005)	78	5–15	12	1.4 ^a
	Jaynes et al. (2008)	79	87	10	0.62
	Fahrner (2002)	160	>60	19	12.7
Beds	Robertson et al. (2000)	2	5	10	10
	Robertson et al. (2005a,b)	9	17	15	1.8 ^a
		108	38	15	2.4 ^a
		120	35	15	2.5ª
		360	14	15	5.1 ^a
	van Driel et al. (2006a) upflow reactors	0.7	9	9	2.1
	· · · ·	0.2	13	13	3.7
	Robertson and Merkley (2009)	40	5	8	3.2
	Robertson et al., 2009	17	10	7.7	3.4
	Schipper et al. (this issue)	83	53	15–25	1.4 ^a
		294	5.5		0–11 ^a
		1320	250	20	9.7
Layer	Robertson et al. (2000)	1	57	10	1.8

^a Nitrate removal rate limited by NO₃⁻ concentrations.

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Table 4

Laboratory-scale experiments (column and batch) that have tested a range of C sources for NO_3^- removal.

Carbon media Re	eference
Wood V(al al is	ogan (1993), Carmichael (1994), Healy et . (2006), Greenan et al. (2006), Gibert et . (2008) and Cameron and Schipper (this sue)
Cardboard G	reenan et al. (2006)
Newspaper Ve	olokita et al. (1996a,b)
Wheat straw Ve	ogan (1993), Soares and Abeliovich
(1	998) and Cameron and Schipper (this
is	sue)
Alfalfa V	ogan (1993)
Corn Fa	ay (1982)
Maize cobs Ca	ameron and Schipper (this issue)
Soil St	tewart et al. (1979) and Gibert et al.
(2	2008)
Soil and sawdust H	ealy et al. (2006)
Soil and jute pellets W	/akatsuki et al. (1993)
Cotton V	olokita et al. (1996b), Della Rocca et al.
(2	2005, 2006), Su and Puls (2007)
Compost, mulch or greenwaste G	ibert et al. (2008)
Ca	ameron and Schipper (this issue)
Seaweed O	vez et al. (2006)

positive relationship between NO₃⁻ removal and average annual temperature (Robertson et al., 2008, 2009; Cameron and Schipper, this issue). Field testing of near-surface reactors in Canada has shown that these continue to remove NO_3^- at a modest rate $(\sim 2 \text{ gN} \text{ m}^{-3} \text{ day}^{-1})$ even at effluent temperatures as low as 1–5 °C (Robertson and Merkley, 2009; Robertson et al., 2009; Elgood et al., this issue). In the latter study, the stream-bed bioreactor continued to operate and provided NO₃⁻ removal throughout the winter season even though the stream surface was periodically frozen. These studies were conducted using a range of wood-particle materials and it is likely that other factors also influenced the temperature response such as the degradability of different C substrates. For example, in a 23-month barrel study, Cameron and Schipper (this issue) found that Q_{10} (the increase in rate for a 10 °C increase in temperature) for NO₃⁻ removal differed between C substrates ranging from less than 0.8 to 2.3, between 14 and 24 °C. In the case of maize cobs, Cameron and Schipper (this issue) found that in the longer term NO₃⁻ removal was less at the higher temperature, presumably because labile C had been depleted more rapidly at higher temperature. While NO₃⁻ removal rate generally increases with increasing temperature, further information is needed for different substrates for design purposes.

3.5. Processes competing for available C

A key determinant for NO₃⁻ removal is the availability of C to denitrifiers and any microbial process that out-competes denitrifiers for this C will reduce NO₃⁻ removal in denitrifying bioreactors. Dissolved O₂ in either groundwater or in point source discharges may allow aerobes to out-compete denitrifiers for available C (Rivett et al., 2008). This is most likely an issue when retention times are short but less likely of concern in large bioreactors with long retention. Laboratory column tests have shown that the time required to deplete the dissolved O₂ in DO saturated water is approximately 1 h in aged, 2-year-old woodchip media (Robertson, this issue) and field trials have indicated similar rates of DO removal in wood particle reactors (Down, 2001; Robertson et al., 2009). Data in Table 5 shows the percent of dissolved organic C (DOC) that would theoretically be consumed by denitrification and aerobic respiration in DO saturated water, for a range of NO₃⁻ removal amounts. In bioreactors where NO₃-N removal is less than about

Table 5

Theoretical percent of dissolved organic C (DOC) that is utilized for denitrification when dissolved O_2 (DO) reduction occurs first. Calculation uses the stoichiometry of Robertson and Merkley (2009) for the denitrification and aerobic respiration reactions, and assumes initial DO of 8 mg L⁻¹ (saturation value at 25 °C).

NO ₃ -N removed (g m ⁻³)	DOC utilized for denitrification (%)
1	22
3	46
10	74
30	90

 3 g N m^{-3} , either because of short retention times (less than several hours) or because of low NO₃⁻ concentrations, consumption of available C by aerobes can exceed that by denitrifying bacteria. Initially poor NO₃⁻ removal in the denitrification bed field trial of Healy et al. (2006) was attributed to high DO concentration (3.7–7.3 mg L⁻¹) and the relatively short retention time.

Sulfate can also be present in wastewaters and groundwaters and act as an alternative electron acceptor when more reducing conditions develop. However, denitrifying organisms generally out-compete sulfate reducers for available C (Appelo and Postma, 1994), consequently sulfate reduction normally only occurs when NO₃⁻ concentrations have been substantially depleted (Vogan, 1993; Robertson and Merkley, 2009; Robertson et al., 2009; Robertson, this issue; Woli et al., this issue; Elgood et al., this issue). Sulfate-reducing conditions are often also accompanied by increasing DOC concentrations (Vogan, 1993; Robertson, this issue). Thus, reactors that have excessively long retention times, beyond that required to fully deplete NO₃⁻, risk generating high levels of DOC and undesirable reaction by-products such as hydrogen sulfide. If sulfate reduction is significant there is the possibility of production of toxic methyl mercury (Woli et al., this issue) by sulfate reducing bacteria; however, this production has yet to be measured.

4. Hydrology

The application of denitrifying bioreactors requires an understanding of the specific hydrologic settings and the spatial and temporal patterns of NO_3^- flux at the site (Table 2). Consequently, site investigations differ between walls, beds and layers and are site-specific.

For denitrification walls, the NO_3^- flux relies on both Darcian principles and the extent of groundwater NO_3^- contamination. If a denitrification wall is located in an area with either low NO_3^- concentrations or low groundwater flow rates, the removal rates will be quite low due to NO_3^- limitations. Hence, a number of aquifer characteristics must be investigated to determine site suitability and design parameters, including:

- Depth to the water table. Low cost wall construction usually precludes placing C material deeper than 4–5 m depth, therefore the water table needs to be within 2–3 m of the ground surface in most cases.
- Pattern and values of saturated hydraulic conductivity. The wall should be placed in media that is conducive to groundwater flow (K>1 m day⁻¹).
- Depth to a restrictive layer with low saturated hydraulic conductivity.
- Direction and gradient of groundwater flow. In many areas, hydraulic gradient undergoes marked seasonal changes, thus site investigations should target both high and low water table conditions.
- Spatial pattern of shallow groundwater NO₃⁻ concentrations. Where walls are to be placed near localized sources of NO₃⁻ input, such as septic systems or animal waste disposal sites, the

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potential for narrow plumes warrants field investigations with multiple monitoring wells to optimize site location.

Bioreactor designs that treat concentrated flows (e.g., tile drains or wastewater) or surface flows, avoid the necessity for site specific studies of shallow groundwater flow conditions, and therefore can have a considerable cost advantage in many cases.

Differences between the saturated hydraulic conductivity of the denitrifying bioreactor and the surrounding aquifer can either diminish or enhance NO_3^- flux into the wall. Schipper et al. (2004) found that in situ mixing of sawdust into saturated sands substantially lowered the saturated hydraulic conductivity of the wall compared to the coarse sand media that composed the shallow aquifer. Consequently, some shallow groundwater bypassed below the wall, substantially decreasing overall NO₃⁻ removal. Barkle et al. (2008) demonstrated that mixing of sands that were saturated while constructing a denitrification wall caused a decline in saturated hydraulic conductivity, presumably because of particle resorting. Furthermore, they showed that the addition of larger particles of organic material (garden mulch) did not improve saturated hydraulic conductivity when mixed with sand. However, walls composed entirely of wood chips can have very high hydraulic conductivities in excess of 100 m day⁻¹ and can induce groundwater flow convergence and upwelling particularly as the width of the wall in the direction of flow is increased (Robertson et al., 2005b). Several studies have used model simulations to provide insight into the nature of groundwater flow in and around reactive barriers considering a variety of geometries and permeability characteristics (Benner et al., 2001: Robertson et al., 2005b, 2007). These studies were also accompanied by field tests with detailed monitoring in order to validate model simulations. Nitrate flux into walls can also be enhanced by their placement at locations where ambient hydraulic gradients have been enhanced. For example Jaynes et al. (2008), placed denitrification walls parallel to tile drains in artificially drained fields. Current studies have been mostly small scale and more work is required to better understand the nature of groundwater flow in and around reactive barriers particularly as they are scaled up in size and a wider range of configurations and media types are tested.

Design of denitrification beds requires information on the seasonal variability in flow rate and NO₃⁻ flux. Bypass systems that divert flows around a denitrification bed can be installed where flow rates vary greatly or are flashy with rainfall.

In many cases it may be useful to adopt a mass removal approach when considering bioreactor use. If the selected bioreactor design can provide adequate flows and NO₃⁻ levels to avoid NO₃⁻-limiting conditions, then the amount of nitrate removal can be easily estimated simply by considering the volume of media utilized and the published reaction rates (Section 3.1). This approach could become very attractive if (when) programs of nutrient trading are adopted. With trading incentives, bioreactors would be preferentially deployed at the most efficient, lowest cost locations, for example in shallow sand and gravel aquifers with high groundwater fluxes in intensively cultivated landscapes with high groundwater NO₃ concentrations.

5. Mechanism for nitrate removal

In most studies of denitrifying bioreactors, it is assumed that conventional heterotrophic denitrification is the dominant mechanism of NO₃⁻ removal. Other possible fates for NO₃⁻ include N immobilisation into organic matter, dissimilatory nitrate reduction to ammonium (DNRA), and Anammox (Burgin and Hamilton, 2007). There is evidence in both laboratory and field studies that denitrification is the dominant mechanism of NO₃⁻ removal in denitrifying bioreactors. Greenan et al. (2006) added ¹⁵N-labelled NO₃into wood chip columns and laboratory incubations and found that immobilisation and DNRA accounted for less than 4% of total NO3removed. Similarly, Gibert et al. (2008) concluded that less than 10% of NO₃⁻ removed was attributable to DNRA (and generally this was less than 5%). Further evidence against the occurrence of DNRA is the lack of significant ammonium (NH₄⁺) production observed in most denitrification walls and beds (Schipper and Vojvodic-Vukovic, 1998; Robertson et al., 2000, 2005a, 2007; Schipper et al., this issue). Although, there is little evidence for DNRA occurring in wood particle barriers, it could be more important when considering other more labile carbonaceous substrates. Vogan (1993) observed an NH₄⁺ increase of 5 mg NL⁻¹ in a laboratory column test utilizing alfalfa straw as a C source. Because the NH_4^+ increase occurred in the same column zone where $NO_3^$ was being depleted and not further along the column where NO_3^{-1} was fully depleted and DOC was still increasing, it was concluded that the NH₄⁺ increase was probably the result of DNRA, rather than from mineralization of organic N from the media. Longterm accumulation of N into organic matter is difficult to assess because even large amounts of N accumulation in added C source would only result in small (and likely undetectable) declines in C:N ratio.

In support of active denitrification in bioreactors, elevated levels of denitrifying enzyme activity (DEA) have commonly been measured in denitrification walls and layers (Schipper et al., 2004, 2005; Schipper and McGill, 2008; Barkle et al., 2008; Moorman et al., this issue). However, elevated DEA does not always mean that significant denitrification is occurring, Schipper and McGill (2008) did not measure much NO_3^- removal in a denitrification layer despite increased DEA. In this case, leaching rates were very high and residence times short (<1 day). Further evidence to support denitrification as a major mechanism for NO₃- removal comes from a field study conducted by Schipper and Vojvodic-Vukovic (2000) who calculated NO₃⁻ removal in a denitrification wall using hydraulic gradients, saturated conductivity and NO₃⁻ concentrations and compared these calculations to laboratory measures of denitrification rate. Schipper and Vojvodic-Vukovic (2000) found that denitrification rates $(0.6-18.1 \text{ ng N cm}^{-3} \text{ h}^{-1})$ were sufficiently high to account for NO₃⁻ removal from shallow groundwater $(0.8-12.8 \text{ ng N cm}^{-3} \text{ h}^{-1})$; however, error bars for both measurement approaches were large. A subsequent study at the same site, where NO₃⁻ was injected into the denitrification wall, measured greater rates of NO₃⁻ removal than could be accounted for by laboratory measurements of denitrification (Schipper et al., 2005). Moorman et al. (this issue) measured denitrifcation potential in bioreactors to be comparable to activity in the surface 15 cm of an organic rich field soil. In contrast to the soil, enrichment with glucose did not increase denitrification potential in the bioreactor indicating that the woodchips provided sufficient C substrate for the denitrifiers.

Another line of evidence supporting denitrification activity is enrichment of ¹⁵N in the residual NO₃⁻, because denitrifying bacteria preferentially consume the lighter isotope (¹⁴N). In contrast, immobilisation of NO3⁻ into organic pools does not discriminate between isotopes (Mariotti et al., 1982). In laboratory studies using woodchips of varying age (fresh to 7-year-old), Robertson (this issue) observed progressive enrichment of $NO_3^{-15}N$ as depletion proceeded. Enrichment was similar in all four woodchip types and was consistent with a Rayleigh-type distillation process with isotopic fractionation factor of -13 per mil. Similar fractionation has been reported for denitrification in groundwater (Robertson, this issue). Enrichment of NO₃⁻⁻¹⁵N has also been observed in field bioreactors (Down, 2001; Robertson et al., 2000) In each of these

studies NH_4^+ concentrations remained low (<1 mg N L⁻¹) making it unlikely that DNRA or Anammox caused the enrichments.

6. Potential adverse effects of using denitrifying bioreactors

A general concern associated with enhancing denitrification is that nitrous oxide (N₂O), a greenhouse gas, can also be produced as a by-product, but so far there are only a few studies of N₂O fluxes from denitrifying bioreactors. In column studies, Greenan et al. (2009) measured N₂O emissions that were between 0.003 and 0.028% of the total NO_3^- removed. This was much less than IPCC default value for production of N₂O assumed to ultimately arise from NO₃⁻ leaching from soil (0.75%; Mosier et al., 1998). N₂O fluxes were also measured from a denitrification wall and adjacent pasture using closed chamber techniques for 2-year period and fluxes were significantly greater (P < 0.05) from the wall (average $0.31 \text{ gN} \text{ ha}^{-1} \text{ h}^{-1}$) than the adjacent pasture (average $0.05 \text{ g N} \text{ ha}^{-1} \text{ h}^{-1}$) (Schipper and Vojvodic-Vukovic, unpublished data). It is likely that N₂O emissions will be lowest from denitrifying bioreactors with complete or near-complete NO₃⁻ removal. This was the case for N₂O flux from a stream-bed reactor (Avon site, Canada, Elgood et al., this issue) where dissolved N₂O was found to be much lower when NO₃⁻ removal was complete $(0-5 \mu g N L^{-1})$ compared to when NO₃⁻ removal was incomplete $(10-35 \mu g N L^{-1})$. Overall dissolved N₂O production amounted to 0.5% of NO₃⁻ removed over a 1-year period. Similarly, Moorman et al. (this issue) estimated N₂O production at 0.62% of NO₃⁻ removed in denitrification walls on either side of a tile drain.

Other gases will also be released from the denitrifying bioreactors including CO_2 and potentially CH_4 both derived from decaying organic matter. The emission of CO_2 does not result in a net increase in CO_2 emissions as the C substrate used in the bioreactor would have decayed if used for other purposes. Elgood et al. (this issue) collected gas bubbles erupting from the surface of the Avon streambed reactor, and found these to contain substantial amounts of CH_4 (25–45%). Methane was also detected during early operation of the bioreactors described by Jaynes et al. (2008), but disappeared after a few months presumably as highly labile C in the wood chips was consumed. Theoretically, the fluxes of CH_4 from bioreactors should be low when NO_3^- concentrations remain sufficiently high to suppress methanogens; however, this concept requires validation.

During start-up of denitrifying bioreactors there is also the potential for the release of soluble C compounds (measured as biochemical oxygen demand, BOD) associated with fresh wood material or other solid C sources. Release of BOD can reduce DO in receiving waters and adversely affect biota. Fresh wood contains 1-2 wt% soluble organic constituents such as tannic acids (Vogan, 1993), which can rapidly leach from wood-particle media during start-up. Initial reactor effluent is normally dark coloured and can have DOC concentrations of hundreds of mgL^{-1} . This is similar to the leachate that occurs at sawmills and in log storage yards where control measures focus on high DOC, trace metals and phenolic compound levels (Taylor et al., 1996). In wood particle reactors, the duration and magnitude of soluble DOC leaching during start-up is dependant upon the reactor retention time. Several field studies have observed dissipation of the initial DOC spike over the first 3-6 months of reactor operation (Robertson and Cherry, 1995; Robertson et al., 2005b; Leverenz et al., this issue; Schipper et al., this issue). In other reactor studies with shorter retention times, more rapid dissipation of the initial DOC spike has been observed. In column tests with sand and sawdust media and one day retention time, Vogan (1993) observed that the initial effluent DOC of 33 mg L^{-1} declined to $< 10 \text{ mg L}^{-1}$ after 10 days of column operation. However, substantial NO₃-N concentrations (> 10 mg L^{-1}) remained in the column effluent which may have assisted with DOC consumption. Likewise, Carmichael (1994) observed DOC depletion to $<15 \text{ mg L}^{-1}$ after 25 pore volumes in a laboratory column utilizing 100% woodchips with a 2-day retention time. Interesting, DOC declined from 43 mg L⁻¹ at pore volume 22 down to 11 mg L⁻¹ at pore volume 26, coincident with the breakthrough of NO₃⁻ in the column effluent. Thus, at sites where high initial DOC concentrations may be unacceptable, control measures might include maintaining high flow rates during start-up, installing postbioreactor treatment (e.g., sandfilter) or collection of the initial effluent for disposal elsewhere. In agricultural terrain, a practical solution may be to use the initial effluent water for irrigating adjacent fields. Another option is to pre-leach the media prior to use, but this is likely to be logistically difficult adding to costs and has not, as yet, been attempted. Robertson and Merkley (2009) observed that slightly elevated concentrations of total phenolic compounds $(4 \mu g L^{-1})$ persisted in the Avon streambed reactor into its second year of operation. Similarly, water passing through a denitrifying bioreactor will have DO removed by microbes and this deoxygenated water could have adverse effects on biota of receiving waters. Currently, there is little information on this potential impact, but presumably this would depend on the relative flow rates of the reactor and the receiving waters and the nature of the receiving waters.

7. Longevity of nitrate removal and maintenance of saturated hydraulic conductivity

The longevity of NO_3^- removal in denitrifying bioreactors is not fully known because currently there appear to be no examples of reactors that have failed due to C depletion (Robertson et al., 2008). Two factors will affect longevity—the continued supply of C to denitrifiers and the maintenance of adequate saturated hydraulic conductivity. Decomposition of solid C sources (e.g., woodchips, sawdust) are greatly slowed when water saturated conditions are maintained and, in most cases, only slow rates of C decomposition are needed to support NO_3^- removal because there is generally a large amount of C relative to NO_3^- inputs.

Currently, the most long-lived bioreactor is the denitrification wall of Robertson and Cherry (1995), constructed in 1992 to treat a septic system plume. In a recent re-examination of this wall in 2007 (year 15, Robertson et al., 2008), core samples of the reactive media provided a NO₃⁻ removal rate in laboratory tests, of $\sim 4 g N m^{-3} day^{-1}$, which was only about 50% lower than the rate measured in year 1 (7 g N m⁻³ day⁻¹). Schipper et al. (2005) reported ongoing NO₃⁻ removal in a denitrification wall over 7 years of operation, and found that denitrification continued to be nitrate-limited, rather than C-limited, throughout this period. In the denitrification walls constructed by Jaynes et al. (2008), more than 60% of the NO₃⁻ was removed during the first 2 years of operation while removal was slightly more than 50% on average in the following 6 years (Moorman et al., this issue). Moorman et al. (this issue) measured an exponential decrease in the C content over the first 8 years for wood chips located at the saturated/unsaturated interface of the bioreactor. After 8 years, only about 25% of the C content of these woodchips remained with an estimated half life of 4.6 years. However, for woodchips that were deeper in the bioreactor and below the water table for a greater fraction of the year, more than 80% of the C still remained (estimated half life of 36.6 years), which accounted for the continued NO₃⁻ removal efficacy of the bioreactor. In a comparative study of woodchip media of varying age, Robertson (this issue) measured reaction rates in core samples

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10 **Table 6**

Cost/benefit analysis of N removal of a denitrifying bioreactor (Robertson et al., 2009) installed for treatment of tile drainage in comparison to other agricultural approaches for reducing N.

Practice	US $(kg-N^{-1})$	Source
Bioreactor	2.39-15.17	This paper
Soil testing and side dressing N fertilizer	1.15	Saleh et al. (2007)
Drainage water management	2.71	Jaynes and Thorp (2008)
Wetlands	3.26	Hyberg (2007)
Fall cover crops	11.06	Saleh et al. (2007)

from two reactors that were 2 and 7 years old, and found that NO_3^- removal rates remained within 50–75% of rates measured in fresh woodchips.

With such slow degradation rates, woodchip bioreactors have demonstrated an ability to remain highly permeable over a number of years with no deterioration in saturated hydraulic conductivity (\sim 100 m day⁻¹) evident (Robertson et al., 2009). However, the bioreactor in the latter study was a surface installation that was subject to minimal overburden pressures. Subsurface bioreactors that experience greater overburden pressures could potentially be prone to greater deterioration in saturated hydraulic conductivity with time. However, little long-term saturated hydraulic conductivity data is currently available for subsurface bioreactors.

8. Cost-benefit analysis

While it is clear that denitrifying bioreactors can remove NO_3^- from concentrated or diffuse discharges, there are other technologies/approaches that are also effective. To be applied in the "real world", denitrifying bioreactors need to be cost effective when compared to these other approaches for managing NO_3^- , e.g., installing wetland and riparian buffers, or more intensive management of N application.

To estimate the cost effectiveness of bioreactors, the bed reactor described in Robertson et al. (2009) will be used as an example. This bioreactor was 13 m long × 1.2 m wide × 1.1 m deep for a total volume of 17.2 m³. It removed an average of 11.3 kg N year⁻¹ from an agricultural field drain. Assuming a conservative 20-year life expectancy for this bioreactor, a total of 226 kg N will be removed. In the central US, woodchips can be purchased for US\$ 26.50 m⁻³. Hauling to a site within 35 km would cost an additional US\$ 65. Rental of a backhoe for installation would range from US\$ 500 to 1000 and incidental expenses would add another US\$ 50. Assuming 4% annual interest for the "time value of money" for the installation costs gives a total cost of installation of US\$ 3249 or a cost per removal of NO₃⁻ of US\$ 15.17 kg-N⁻¹.

However, many bioreactors might be installed on farms where farmers have access to both a backhoe and wood from wind breaks or other local sources, greatly reducing the cost of installation. In addition, the bioreactor described in Robertson et al. (2009) only removed NO₃⁻ for about 70% of the year when the field tile was draining. Connecting a bioreactor to a NO₃⁻ source that flowed year round would increase its efficiency. Thus, for a farmer installing this bioreactor onto a year-round source the cost of NO₃⁻ removal would be reduced to US\$ 2.39 kg-N⁻¹. This cost of removal range (US\$ 2.39-15.17) compares favourably with estimates of other NO₃⁻ removal technologies as shown in Table 6. Cost efficiencies for bioreactors of other designs would of course vary, but bioreactors can be cost efficient alternatives for removing NO₃⁻ when compared against other commonly promoted approaches for managing N. In many cases, denitrifying bioreactors are complementary with these other practices and do not preclude the use of multiple mitigation approaches (Woli et al., this issue).

9. Treatment of other contaminants

The majority of data on the performance of denitrifying bioreactors has logically focused on NO3- removal but there are other redox sensitive contaminants in wastewaters and shallow groundwater that might be treated using modified bioreactors. These include pathogens and pharmaceutical compounds in wastewaters, pesticides in agricultural drainage and industrial contaminants such as perchlorate which is associated with explosives and rocket fuel manufacturing. Monitoring of four sawdust beds treating septic tank effluent in Ontario (Robertson et al., 2005a), showed that over several years of operation, Escherichia coli levels generally remained below detection in the reactor effluents (<10 cfu 100 mL⁻¹). However, several breakouts did occur, up to \sim 1000 cfu 100 mL⁻¹, particularly at the site with the highest loading rate. Robertson et al. (2007) reported complete attenuation of trace levels of perchlorate (ClO₄) occurring in agriculturally impacted groundwater in Ontario, during migration through a woodchip layer. In a subsequent experiment, Robertson et al. (2009) immersed a highway safety flare containing ClO₄ in the inlet pipe of a woodchip reactor treating agricultural drainage and the elevated influent ClO₄ concentration (up to 33 μ g L⁻¹) was entirely attenuated in the reactor. It was noted that ClO₄ attenuation did not commence until NO₃⁻ was first depleted, demonstrating that NO₃⁻ inhibits the degradation of ClO₄.

Of the N forms (NO_3^- , NH_4^+ , organic N), only NO_3^- and NO_2^- seem to be removed with little, if any, removal of organic N or ammonium (Robertson et al., 2005a; Schipper et al., this issue). This reinforces the need for a nitrification step of wastewater prior to being loaded into a denitrifying bioreactor.

Phosphate is often at low concentrations in groundwater but can be significant in effluents. To date, there has been little evidence that wood-based bioreactors remove phosphate from effluents (Robertson et al., 2005a; Jaynes et al., 2008; Schipper et al., this issue). However, the addition of other amendments to bioreactors (e.g., iron slag), has resulted in considerable phosphate removal from treated septic tank effluent (Baker et al., 1998; Robertson, 2000) and streams (McDowell et al., 2008).

Emerging contaminants such as pharmaceutical compounds also have the potential to be attenuated in denitrifying bioreactors but monitoring data is, as yet, lacking.

10. Conclusions and future work

To date, field studies have demonstrated that denitrifying bioreactors are capable of substantial NO_3^- removal in a number of watershed settings. Major advantages of denitrifying bioreactors are their simplicity with low maintenance requirements and the ability to tailor designs to fit hydrological site criteria. It is not clear how long these systems will continue to remove NO_3^- because no studied systems has yet been observed to fail—consequently we can only conclude that denitrifying bioreactors could last for a minimum of 15 years (Robertson et al., 2008). Moorman et al. (this issue) suggest that sustainability of wood chips to support $NO_3^$ removal was dependent on the time that wood chips remained water saturated (and presumably anoxic). Halflives varied between 4.6 and 36.6 years for woodchip either periodically saturated or permanently saturated, respectively.

Where NO_3^- loading is high, NO_3^- removal is dependent on temperature and availability of C. The high loads of NO_3^- entering denitrifying bioreactors mean that often NO_3^- removal rates are zero-order. Organic N and NH_4^+ are not removed in denitrifying bioreactors and a nitrification step may be required before wastewater enters the bioreactor. Development of simple nitrifi-

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cation pre-treatment systems coupled with bioreactors should be encouraged such as those described by Oakley et al. (this issue).

There is some laboratory and field evidence that denitrification is the main mechanism for NO₃⁻ removal but further work on the microbial ecology of these systems is needed and may lead to approaches for increasing nitrate removals rates and longevity. Field-scale research is needed to determine the effectiveness, costs, and factors controlling the rate of NO₃⁻ removal and denitrification in different bioreactors, particularly the suitability of alternative C sources. Management approaches still need to be developed for decreasing unwanted side effects, such as the production of N₂O and initial leaching of dissolved organic matter. Design criteria and demonstration sites are warranted to test alternative designs that merge bioreactors with constructed wetlands to provide co-benefits of biodiversity and aesthetics (Leverenz et al., this issue). Integration of bioreactors with other approaches for reducing NO₃⁻ loads to surface water, such as riparian zones or controlled drainage, would also be beneficial.

While there are unanswered questions about performance of denitrifying bioreactors, there is sufficient information available to utilize bioreactors for reducing NO_3^- fluxes in a variety of settings. Design manuals should be developed that address site evaluation, provide detailed construction approaches that integrate with local hydrology while meeting policy directives and performance goals of different countries and regions. These design manuals might be developed for engineers but, where appropriate, also for farmers and farm advisors. Finally, of particular importance is determining linkages between hydrological flow paths, retention time in the bioreactors and NO_3^- removal efficiency, thus we recommend interdisciplinary research combining the skills of hydrologists, hydrogeologists, engineers, biogeochemists, and land managers.

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