

Nitrogen cycling processes within stormwater control measures: A review and call for research

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ABSTRACT

Stormwater control measures (SCMs) have the potential to mitigate negative effects of watershed development on hydrology and water quality. Stormwater regulations and scientific literature have assumed that SCMs are important sites for denitrification, the permanent removal of nitrogen, but this assumption has been informed mainly by short-term loading studies and measurements of potential rates of nitrogen cycling. Recent research concluded that SCM nitrogen removal can be dominated by plant and soil assimilation rather than by denitrification, and rates of nitrogen fixation can exceed rates of denitrification in SCM sediments, resulting in a net addition of nitrogen. Nitrogen cycling measurements from other human-impacted aquatic habitats have presented similar results, additionally suggesting that dissimilatory nitrate reduction to ammonium (DNRA) and algal uptake could be important processes for recycling nitrogen in SCMs. Future research should directly measure a suite of nitrogen cycling processes in SCMs and reveal controlling mechanisms of individual rate processes. There is ample opportunity for research on SCM nitrogen cycling, including investigations of seasonal variation, differences between climatic regions, and trade-offs between nitrogen removal and phosphorus removal. Understanding nitrogen dynamics within SCMs will inform more efficient SCM design and management that promotes denitrification to help mitigate negative effects of urban stormwater on downstream ecosystems.

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1. Introduction

Stormwater control measures (SCMs) are common across the urban landscape. These structures are used to mitigate the negative effects of watershed development, or increased impervious area, on

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watershed hydrology and stream water quality. The downstream effects of increasing impervious area (without mitigation) include streambed scour (Booth, 1990), increased nutrient loading (Paul and Meyer, 2001; Walsh et al., 2005), loss of stream macro-invertebrate diversity (Stranko et al., 2012), and flashier hydrology (O'Driscoll et al., 2010; Paul and Meyer, 2001; Walsh et al., 2005). Due to these observed consequences of development, most municipalities, counties, or states have permitting requirements for stormwater mitigation and suggested SCM design practices with new development (National Research Council, 2009).

The need to quantify the effectiveness and downstream impacts of SCMs for water quality management has resulted in predominantly concentration- or load-based studies of SCM nitrogen removal (Collins et al., 2010). These studies were typically conducted over short time scales (e.g., hours, days, weeks) and showed that while the average percent removal of different nitrogen species was positive in many different kinds of SCMs, there was large variability in the percent nitrogen removed within each kind of SCM (Koch et al., 2014). Also, percent removal of nitrogen calculated using loading measurements can be driven by stormwater volume reduction (e.g., infiltration) rather than actual treatment of nitrogen. Despite this documented variability in percent nitrogen removal and possible confounding effects of volume reduction on load-based measurements, assumptions have been made in policy, management, and research about the internal workings of SCMs based on loading mass-balance studies. The variability in SCM nitrogen removal may be, in part, caused by a lack of studies including direct measurements of nitrogen cycling processes occurring within SCMs. Recent research focuses more on internal SCM processes rather than mass balance between the inflow and outflow, but results range from effective nitrogen removal to results that are counter the stated water quality goals of the SCM (Datry et al., 2003; Duan et al., 2016; Gold et al., 2017a; Song et al., 2017; Williams et al., 2013). The large uncertainty in SCM effectiveness calls for in-depth mass balance and internal nitrogen cycling studies of SCMs.

A common assumption applied to SCM function from mass-balance studies is that denitrification removes a substantial amount of nitrogen in SCMs, especially in retention-based SCMs that never drain fully or drain slowly. Denitrification, the microbially-mediated transformation of nitrate (NO₃) to inert N₂ gas, is a removal mechanism for bioavailable nitrate from aquatic ecosystems (Seitzinger et al., 2006). The validity of this assumption has far-reaching implications for the condition of downstream waters because excess nitrogen can cause eutrophication of marine and coastal systems (Howarth and Marino, 2006) and freshwaters (Elser et al., 2007). If denitrification is less prevalent in SCMs than assumed, nitrogen inputs via stormwater could be exported downstream or recycled within SCMs.

If nitrogen in SCMs is not denitrified or exported immediately, it could be recycled internally through temporary uptake and remineralization by primary producers (Williams et al., 2013), buried through sedimentation of particles and organic matter (Passeport et al., 2013; Schroer et al., 2018; Sønderup et al., 2016), transformed via dissimilatory nitrate reduction to ammonium (DNRA) (Scott et al., 2008), or removed through anaerobic ammonium oxidation (anammox) (Burgin and Hamilton, 2007). Various factors could affect the importance of each of these processes within SCMs, as in other aquatic habitats, such as sediment carbon quality, water column N:P ratios, residence time, SCM depth, and SCM length:width ratios. Internal nitrogen processes have yet to be extensively measured in SCMs but they could explain variability in nitrogen removal efficacy observed by previous studies. Understanding the processing of nitrogen in SCMs may improve the ability of stormwater management plans to promote denitrification of nitrogen inputs and improve water quality in developed areas.

The purpose of this review is to present the current understanding of nitrogen cycling within multiple types of SCMs and suggest opportunities for research. The types of SCMs include many that have been promoted as effective sites of nitrogen removal or maintain permanent standing water such as stormwater wet ponds, extended detention dry ponds, stormwater wetlands, and bioretention cells.

The goals of this review article are to:

1. Present examples of the prevailing assumptions about nitrogen cycling in SCMs in the scientific literature and the reasoning behind them
2. Summarize recent studies focusing on nitrogen cycling within SCMs and their assumptions of denitrification
3. Discuss recent advances in nitrogen cycling measurements that can be applied to studies in SCMs
4. Highlight opportunities for future research of nitrogen cycling within SCMs

2. Methods

This review is based on studies that focused on nitrogen cycling processes within SCMs designed for the main purpose of collecting and treating stormwater derived from urban land uses. Studies were identified by searching various keywords related to stormwater control measures and nitrogen cycling (i.e., denitrification, DNRA, assimilation) in the Web of Science database (Table 1), and studies that fell within the scope of the review were summarized and sorted by method in an excel spreadsheet. This review identified a total of twelve studies that reported rates of denitrification (potential or direct measurement) (Table 2), two that reported rates or importance of DNRA (Messer et al., 2017; Payne et al., 2014), and three that reported rates or importance of assimilation within stormwater control measures (Messer et al., 2017; Norton et al., 2017; Payne et al., 2014).

3. Assumptions of denitrification and measurements within SCMs

Denitrification is an important process for mediating levels of bioavailable nitrogen in aquatic ecosystems (Howarth et al., 1996;

Table 1

List of keywords used for literature review. The search was conducted by searching the name of each process with additional keywords.

Process	Keywords	# of search results
Denitrification	Stormwater	148
	Retention, basin	144
	Stormwater, nitrogen	132
	Stormwater, wetland	58
	Bioretention	56
	Infiltration, basin	33
	Wet pond	25
	Detention, basin	12
	Stormwater	3
	Retention, basin	2
DNRA	Stormwater, wetland	0
	Bioretention	1
	Infiltration, basin	0
	Wet pond	1
	Detention, basin	0
	Stormwater	19
Nitrogen assimilation	Retention, basin	16
	Stormwater, wetland	14
	Bioretention	5
	Infiltration, basin	1
	Wet pond	2
	Detention, basin	1

Table 2
SCM studies that have measured denitrification (Adapted from Norton et al., 2017).

Area-based rates						
Source	SCM Type	Location	Time (frequency)	Method	Rate (Published units)	Rate (mg N m ⁻² h ⁻¹)
Norton et al. (2017)	Bioretention	Portland, OR, USA	October (1)	N ₂ :Ar	-63 to 520 μmol N m ⁻² h ⁻¹	-0.88 to 7.28
Payne et al. (2014)	Bioretention	Victoria, Australia	July (1), August (1), October (1)	¹⁵ N tracer	25–1800 μmol N m ⁻² h ⁻¹	0.35–25.2
Morse et al. (2017)	Dry ext. detention basin	Ithaca, NY, USA	April–October	N ₂ O	0.03 ± 0.0006 g N m ⁻² yr ⁻¹	3.36 × 10 ⁻³ ± 5.6 × 10 ⁻⁵
Zhu et al. (2004)	Infiltration basin	Phoenix, AZ, USA	July (1)	Soil core/C ₂ H ₂	3.3–57.6 mg N m ⁻² d ⁻¹	0.137–2.39
Newcomer Johnson et al. (2014)	Wetlands (inline)	Baltimore County, MD, USA	Summer (2), Winter (1)	“push-pull” ¹⁵ N tracer	132.3 ± 61.1 mg N m ⁻² d ⁻¹	5.513 ± 2.55
Lancaster et al. (2016)	Wetlands	Yale Myers Experimental Forest, CT	November (1) January (1)	¹⁵ N tracer	14–53 μmol N m ⁻² h ⁻¹	0.19–0.74
Morse et al. (2017)	Wet detention basin	Ithaca, New York, USA	April–October	N ₂ O	1.09 ± .02 g N m ⁻² yr ⁻¹	0.124 ± .0025
Gold et al. (2017a)	Wet pond	Jacksonville, NC	June (1), September (1)	N ₂ :Ar	-206 to 14.8 μmol N m ⁻² h ⁻¹	-2.88 to 0.207
Newcomer Johnson et al. (2014)	Wet pond	Baltimore County, MD, USA	Summer (2), Winter (1)	“Push-pull” ¹⁵ N tracer	248.2 ± 77.4 mg N m ⁻² d ⁻¹	10.34 ± 3.23
Mass-based rates						
Source	SCM Type	Location	Time (frequency)	Method	Rate (Published units)	Rate (mg N kg ⁻¹ h ⁻¹)
Norton et al. (2017)	Bioretention	Portland, OR, USA	October (1)	DEA	0.20 mg N kg ⁻¹ h ⁻¹	0.20
Waller et al. (2018)	Bioretention	MD, VA, NC	November/December (1)	DEA	0.7 ± 0.19 mg N kg ⁻¹ h ⁻¹	0.7 + 0.19
McPhillips and Walter (2015)	Dry detention basin	Ithaca, NY, USA	October (1)	DEA	0.23 mg N kg ⁻¹ h ⁻¹	0.23
Morse et al. (2017)	Dry detention basin	Ithaca, NY, USA	June (1)	DEA	0–0.3 mg N kg ⁻¹ h ⁻¹	0–0.3
Bettez and Groffman (2012)	Dry extended detention	Baltimore County, MD, USA	September (1)	DEA	1.43 mg N kg ⁻¹ h ⁻¹	1.43
Bettez and Groffman (2012)	Dry pond	Baltimore County, MD, USA	September (1)	DEA	1.49 mg N kg ⁻¹ h ⁻¹	1.49
Bettez and Groffman (2012)	Filtration basin	Baltimore County, MD, USA	September (1)	DEA	1.00 mg N kg ⁻¹ h ⁻¹	1.00
Bettez and Groffman (2012)	Infiltration basin	Baltimore County, MD, USA	September (1)	DEA	0.18 mg N kg ⁻¹ h ⁻¹	0.18
Zhu et al. (2004)	Infiltration basin	Phoenix, AZ, USA	July (1)	DEA	390–1151 ng N g ⁻¹ h ⁻¹	0.39–1.15
Harrison et al. (2011)	Wetlands	Baltimore County, MD, USA	June–August & November–December	“Push-pull” ¹⁵ N tracer	147 ± 29 μg N kg ⁻¹ d ⁻¹	0.00613 + 0.0012
McPhillips and Walter (2015)	Wet detention basin	Ithaca, NY, USA	October (1)	DEA	2.27 mg N kg ⁻¹ h ⁻¹	2.27
Morse et al. (2017)	Wet detention basin	Ithaca, NY, USA	June (1)	DEA	0–1.75 mg N kg ⁻¹ h ⁻¹	0–1.75
Blaszczak et al. (2018)	Wet pond	Baltimore, MD	June (1)	N ₂ :Ar (Potential)	0.42–35.9 mg N kg ⁻¹ h ⁻¹	0.42–35.9
Blaszczak et al. (2018)	Wet pond	Boston, MA	June (1)	N ₂ :Ar (Potential)	0.42–35.9 mg N kg ⁻¹ h ⁻¹	0.42–35.9
Blaszczak et al. (2018)	Wet pond	Durham, NC	June (1)	N ₂ :Ar (Potential)	0.42–35.9 mg N kg ⁻¹ h ⁻¹	0.42–35.9
Blaszczak et al. (2018)	Wet pond	Miami, FL	August (1)	N ₂ :Ar (Potential)	0.42–35.9 mg N kg ⁻¹ h ⁻¹	0.42–35.9
Blaszczak et al. (2018)	Wet pond	Minneapolis-St.Paul, MN	August (1)	N ₂ :Ar (Potential)	0.42–35.9 mg N kg ⁻¹ h ⁻¹	0.42–35.9
Blaszczak et al. (2018)	Wet pond	Phoenix-Scottsdale, AZ	August (1)	N ₂ :Ar (Potential)	0.42–35.9 mg N kg ⁻¹ h ⁻¹	0.42–35.9
Blaszczak et al. (2018)	Wet pond	Portland, OR	August (1)	N ₂ :Ar (Potential)	0.42–35.9 mg N kg ⁻¹ h ⁻¹	0.42–35.9
Blaszczak et al. (2018)	Wet pond	Salt Lake City, UT	July (1)	N ₂ :Ar (Potential)	0.42–35.9 mg N kg ⁻¹ h ⁻¹	0.42–35.9
Bettez and Groffman (2012)	Wet pond	Baltimore County, MD, USA	September (1)	DEA	1.07 mg N kg ⁻¹ h ⁻¹	1.07

Note: DEA = denitrification enzyme assay, Wetlands (inline) = constructed wetlands that replaced a stream channel, N₂:Ar (Potential) = potential denitrification assays that used N₂:Ar rather than acetylene reduction.

Seitzinger et al., 2006). Denitrification occurs under anaerobic or low-oxygen conditions and requires a suitable carbon source and available nitrate either in the overlying water or produced via nitrification of ammonium from the sediments (Eyre et al., 2013; Kana et al., 1994; Seitzinger et al., 2006; Seitzinger, 1988). Based on

these conditions, SCMs appear to be ideal locations for denitrification to occur. Since most SCMs only have inflow during and after storm events, their residence times can range from hours to weeks (Jefferson et al., 2015), and longer residence times are positively correlated with the magnitude of nitrogen removal from overlying

waters due mainly to denitrification promoted by increased exposure to the sediment-water interface (Bettez and Groffman, 2012; Klocker et al., 2009; Mallin et al., 2002; Nixon et al., 1996; Passeport et al., 2013). Over time, some SCMs fill in with sediment and organic matter (Gold et al., 2017a; Merriman et al., 2017; Moore and Hunt, 2012; Schroer et al., 2018), which could provide a carbon source for denitrification and increase the incidence of anaerobic conditions due to decomposition. Denitrification can also be promoted in some SCMs through soil amendments and certain design specifications, such as elevating underdrains in bioretention cells to increase low-oxygen conditions (reviewed in Hunt et al., 2012). Many studies have measured low oxygen conditions in the bottom water of stormwater ponds (Duan et al., 2016; Gold et al., 2017a; Newcomer Johnson et al., 2014), further indicating that these SCMs could be important sites for denitrification based on the favorable combination of factors (Bettez and Groffman, 2012; Newcomer Johnson et al., 2014; Zhu et al., 2004).

A number of published articles that utilized mass-balance experimental designs (i.e., load-based measurements) have suggested that SCMs are important locations for denitrification (summarized in Collins et al., 2010). These load-based studies that have hypothesized about the importance of denitrification in SCMs often attribute reduced loads of nitrogen from SCMs to denitrification, when, in fact, the mechanism for nitrogen removal is unknown. Some studies have measured denitrification within SCMs, and most of these studies have assessed denitrification using various proxy-based or potential-based methods, such as denitrification enzyme assays (DEA; Groffman et al., 1999), acetylene-block intact sediment core incubations (described in Groffman et al., 2006), groundwater “push-pull” ^{15}N tracers (Addy et al., 2002), and N_2O flux measurements that are converted into denitrification rates (Schlesinger, 2009) (Fig. 1, Table 2).

The first investigation of denitrification within SCMs utilized DEA and intact core acetylene-block methods in an infiltration basin and found rates of potential denitrification and *in situ* denitrification that were similar to the highest rates measured in other aquatic environments (Zhu et al., 2004) (Fig. 1, Table 2). This study also found a positive relationship between sediment organic matter (SOM) and potential denitrification, indicating that the settling of sediment and organic matter particles within the SCM may promote denitrification. Later studies found higher rates of potential denitrification in SCMs compared to reference riparian and upland areas and showed positive relationships between potential denitrification and both SOM and inundation time (Bettez and Groffman, 2012; McPhillips and Walter, 2015) (Fig. 1, Table 2). Additionally, these studies concluded that the wetter, retention-based SCMs, for the most part, had higher rates of potential denitrification than other SCMs, possibly due to constant inundation and longer residence times (Bettez and Groffman, 2012; McPhillips and Walter, 2015) (Fig. 1, Table 2). An extensive study of stormwater ponds in 8 US cities found higher rates of potential denitrification than previous studies that used DEA methods, but the influence of environmental and landscape controls were unclear (Blaszczak et al., 2018) (Fig. 1, Table 2). High rates of groundwater denitrification were measured in wet ponds and stormwater wetlands using groundwater “push-pull” ^{15}N tracers, and these rates were comparable to rates in hydrologically-connected floodplains (Harrison et al., 2011; Newcomer Johnson et al., 2014) (Fig. 1, Table 2). Measurements of N_2O fluxes and an SCM nitrogen loading mass-balance determined that a wet detention basin was able to denitrify up to 58% of the dissolved inorganic nitrogen (DIN) that flowed into it, and a dry detention basin was only able to denitrify 1% of incoming DIN (Morse et al., 2017) (Fig. 1, Table 2). Studies utilizing molecular methods have found a positive relationship

between inundation time and the abundance of denitrifier functional genes such as *nar* (nitrate reductase), *nirK*, *nirS* (nitrite reductase), *cnor*, *qnor*, *norB* (nitric oxide reductase), and *nosZ* (nitrous oxide reductase) (Chen et al., 2013; Morse et al., 2017). Aligning with the prevailing knowledge about SCM nitrogen removal, these studies suggest that SCMs, especially SCMs with long residence times, are important sites of denitrification and could help reduce nitrogen export from urban watersheds.

A growing number of studies have directly measured denitrification in SCMs, utilizing either the $\text{N}_2:\text{Ar}$ method to measure net N_2 fluxes (described in Kana et al., 1994) or lab-based, mass-balance ^{15}N tracers (described in Payne et al., 2014). Direct methods, in this review, are defined as methods that measure the end-product of a microbial process or net production of opposite processes over time (e.g., denitrification vs. nitrogen fixation) rather than proxies or potential measurements. Direct methods are key to understanding the importance of denitrification and other nitrogen removal pathways within SCMs. Recent work utilizing ^{15}N mass-balance methods found that assimilation in plants and soils was the dominant nitrate removal pathway in bioretention and stormwater wetland mesocosms (Messer et al., 2017; Morse et al., 2018; Payne et al., 2014). Another study measured rates of net N_2 flux ranging from -63 (net nitrogen fixation) to 520 (net denitrification) $\mu\text{mol N m}^{-2} \text{h}^{-1}$ in bioretention mesocosms (Fig. 1, Table 2), and denitrification accounted for a maximum of 23% of dissolved inorganic nitrogen removal (Norton et al., 2017). This study also concluded through concurrent measurements of DEA and simple nitrogen mass balances that these latter methods might vastly overestimate the net rate and importance of denitrification (Norton et al., 2017). The only published study to measure net N_2 fluxes from SCMs in the field (rather than mesocosms) reported substantial rates of net nitrogen fixation in unamended sediments of wet ponds during the summer (Gold et al., 2017a), with net N_2 fluxes ranging from -206 and $14.8 \mu\text{mol N m}^{-2} \text{h}^{-1}$ (Fig. 1, Table 2). Further, in this study the response of sediments to a nitrate addition varied based on wet pond age, where younger ponds took up nitrate and switched to net denitrification while older ponds also took up nitrate but did not utilize it for denitrification (Gold et al., 2017a). The measured rates of net nitrogen fixation coincident with uptake of nitrate in older ponds supports conclusions of other recent studies that alternative nitrogen pathways, such as DNRA or assimilation in plants and soils, could be more important than denitrification in SCMs.

This new body of evidence utilizing direct methods such as $\text{N}_2:\text{Ar}$ and mass-balance ^{15}N methods provides context for previous work in SCMs and raises questions about the importance assigned to denitrification in SCMs based on DEA and field-based ^{15}N tracers. While DEA does, indeed, measure the ability of the microbial community to denitrify given ideal conditions, it is a measure of potential rates of denitrification without a concurrent measure of nitrogen fixation, which could be occurring at a higher rate than denitrification (Foster and Fulweiler, 2014; Fulweiler et al., 2007, 2013; Gold et al., 2017a; Newell et al., 2016a). Regardless of the absolute value of denitrification, if the process of nitrogen fixation is occurring at a higher rate than denitrification, nitrogen is being created faster than it is being removed and there is a net addition of nitrogen to the system. Furthermore, bottle effects from the DEA methodology can change the microbial community (Hartzog et al., 2017), and the method underestimates coupled nitrification-denitrification (Seitzinger et al., 1993) due to the inhibition of nitrification by acetylene (Hynes and Knowles, 1982, 1978; Mosier, 1980; Walter et al., 1979). Potential denitrification is reported as a sediment mass-based rate, which increases the complexity of scaling up field measurements and comparing results to area-based rates measured with other methods. Field-based measurements of

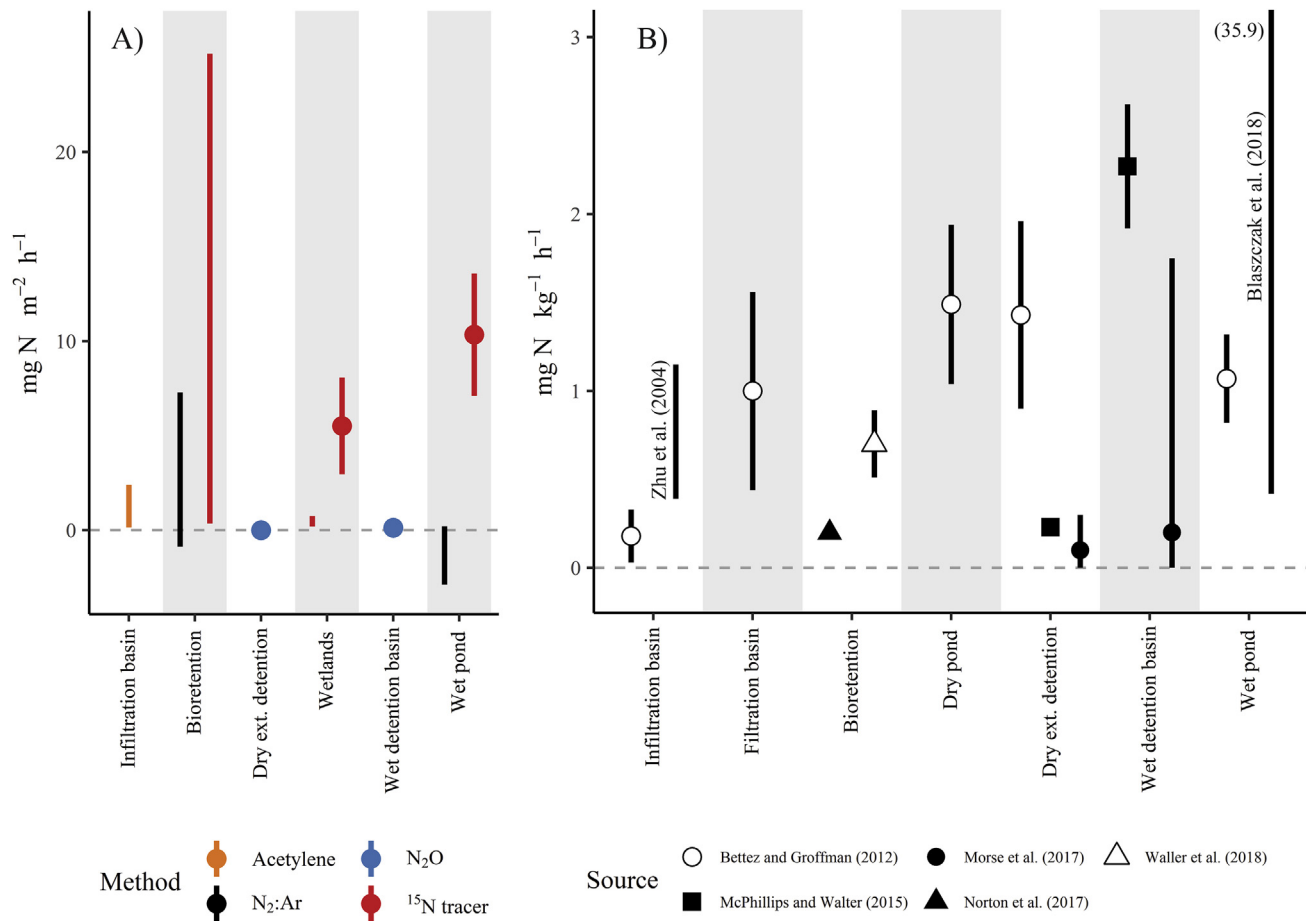


Fig. 1. **A)** Areal rates of denitrification or net N₂ flux (N₂:Ar) and **B)** rates of denitrification based on denitrification enzyme assay (DEA) from SCM nitrogen cycling studies (Table 2). SCM types for each panel are ordered from less frequently inundated (left) to more frequently inundated (right). Some rates were converted from published units to above units reported in the majority of studies. ¹⁵N tracer studies that reported only mass-based rates of denitrification are not shown (n = 1).

denitrification with ¹⁵N tracers are an effective way to measure denitrification (Groffman et al., 2006), but they also do not assess nitrogen fixation and have some limitations due to underestimates of denitrification from water column nitrate (Seitzinger et al., 1993). Conversions of N₂O fluxes to denitrification rates introduce large amounts of variability because these ratios vary greatly within aquatic (Seitzinger, 1988) and terrestrial ecosystems (Schlesinger, 2009). N₂:Ar methods and mass-balance ¹⁵N tracers have some drawbacks as well because, as stated in Groffman et al. (2006), denitrification “is a miserable process to measure”. Mass-balance ¹⁵N tracers may underestimate nitrogen fixation by almost half (Newell et al., 2016a) and are typically limited to mesocosm experiments. N₂:Ar methods are also typically constrained to mesocosm experiments, and they can also take longer to conduct and may not capture heterogeneity within the sampled ecosystem (Groffman et al., 2006).

In light of recent research that questions the relative importance of denitrification to SCM nitrogen removal, N₂:Ar methods or mass balance ¹⁵N tracers should be used to determine the balance of nitrogen fluxes in SCM sediments and distinguish between temporary nitrate removal (e.g., assimilation, DNRA) and permanent nitrogen removal (e.g., denitrification, anammox). Nitrogen fixation and denitrification co-occur (Fulweiler et al., 2013), so measuring the net effects of these processes and the rates of other nitrogen cycling processes is key to understanding if SCMs are sources or sinks for nitrogen.

4. Heterotrophic nitrogen fixation in nitrogen cycling studies

Nitrogen fixation, the conversion of N₂ gas to NH₄⁺ by heterotrophic bacteria, has been measured using acetylene reduction assays (Hardy et al., 1968) for almost fifty years, and these measurements informed thinking that nitrogen fixation was not an important nitrogen input in most aquatic environments. We now know that these acetylene reduction assays can significantly alter the sediment microbial community (Fulweiler et al., 2015), renewing questions about the importance of nitrogen fixation in aquatic ecosystems. Recent studies have measured large rates of net nitrogen fixation in estuarine environments using N₂:Ar measurements, indicating that heterotrophic nitrogen fixation may play a larger role in aquatic nitrogen cycling than previously thought (Foster and Fulweiler, 2014; Fulweiler et al., 2013, 2007; Newell et al., 2016a, 2016b; Rao and Charette, 2012).

The rapidly increasing use of microbial methods for identifying active microbial communities and quantifying functional gene expression through quantitative PCR (qPCR) and metagenomics testing has allowed for more precise measurements of bacteria and archaea community structure. This methodology is especially useful for studying the nitrogen cycle because these microbial organisms are responsible for modulating each step of the nitrogen cycle. Recent genetic work measuring potential nitrogen fixation by targeting *nifH* gene expression or measuring total abundance of *nifH* has provided evidence of the importance of nitrogen fixation

in certain areas of the estuarine environment (Andersson et al., 2014; Fulweiler et al., 2013; Newell et al., 2016b), and these genetic results have been corroborated with direct measurements of net N_2 fluxes from sediment cores (Fulweiler et al., 2013; Newell et al., 2016b). These studies have determined that a small number of heterotrophic nitrogen fixing bacteria can dominate and outcompete denitrifiers in organic-rich sediments (Newell et al., 2016b). Poor carbon quality and low-oxygen conditions may also allow heterotrophic nitrogen fixers to outcompete denitrifiers in aquatic sediments (Eyre et al., 2013; Fulweiler et al., 2013, 2007). The dominance of heterotrophic nitrogen fixers over denitrifiers in SCM sediments would lead to more nitrogen fixation than denitrification, essentially flipping sediments from nitrogen sinks to sources. The techniques for measuring denitrifier and nitrogen-fixer genes have not yet been applied together to study SCM nitrogen cycling but could be utilized together in the future. Functional genes and molecular metrics associated with parts of the nitrogen cycle (e.g., denitrification, nitrification) have been studied in SCMs (Chen et al., 2013; Morse et al., 2018, 2017; Waller et al., 2018), but none have measured the nitrogen fixing community. Along with measurements of net N_2 fluxes and mass-balance ^{15}N tracers, microbial methods should be used to better understand the balance of nitrogen cycling processes in SCMs. Without the measurement of nitrogen fixation or the heterotrophic nitrogen-fixing community that can add new nitrogen to the system, studies are likely capturing only part of the picture, possibly inflating the importance of SCMs as hot spots of net nitrogen removal via denitrification.

5. Dissimilatory nitrate reduction to ammonium (DNRA)

Dissimilatory nitrate reduction to ammonium (DNRA) is often overlooked as a fate of nitrate in SCMs, but some types of SCMs may have conditions favorable for DNRA - low nitrate concentrations, organic-rich sediments, low-oxygen conditions, and high iron concentrations (Burgin and Hamilton, 2007; Kessler et al., 2018). The conditions that support DNRA are similar to those which are needed for denitrification, and the ratio of nitrate to carbon seems to determine which pathway reduces nitrate (Burgin and Hamilton, 2007; Kessler et al., 2018; Morrissey et al., 2013). There is some indirect evidence for DNRA in SCMs from nitrogen loading mass-balance studies. For example, some SCMs, especially deeper ponds that are designed to settle suspended particles and enhance denitrification, have been shown to increase ammonium concentrations based on short-term loading studies (Koch et al., 2014). However, this could also be due to decreased nitrification in low-oxygen conditions (Koch et al., 2014). Also, nitrate uptake in sediments of older ponds that also exhibited net nitrogen fixation suggests that DNRA could have occurred (Gold et al., 2017a). Of the few studies that have directly measured rates and the relative importance of DNRA in SCMs or man-made aquatic ecosystems, DNRA can range from constituting a relatively minor nitrate reduction pathway in wetlands and ponds (Messer et al., 2017; Nogaro and Burgin, 2014; Scott et al., 2008) to exceeding rates of denitrification in urbanized tidal creeks (Dunn et al., 2013), groundwater of constructed wetlands treating wastewater (Jahangir et al., 2017), and other freshwater ecosystems (Burgin and Hamilton, 2007).

Rates of DNRA have typically been measured using ^{15}N tracers, but similar to techniques described for denitrification and nitrogen fixation, genetic methods can be used to measure the prevalence of genes that encode the enzyme responsible for DNRA, *nrfA*. The abundance of DNRA communities, as measured by *nrfA* abundance via qPCR, correlates with rates of DNRA (Smith et al., 2015; Song et al., 2014). Future research should attempt to quantify DNRA in SCMs through the use of ^{15}N mass balances and genetic

measurements. This future research should also examine the relationships between DNRA and environmental controls on DNRA found in other aquatic ecosystems (e.g., carbon quality, oxygen concentrations, nitrate concentrations) so that management actions can be directed to promote denitrification over DNRA.

6. Biotic assimilation and remineralization

Transformations of nitrogen in SCMs can be biological rather than chemical, with nitrogen in SCMs being assimilated by soil microbes (Messer et al., 2017; Payne et al., 2014), algae (DeLorenzo et al., 2012; Gold et al., 2017b; Lewitus et al., 2008; Reed et al., 2016), and vegetation (Lenhart et al., 2012; Messer et al., 2017; Payne et al., 2014). Plant, algal, and microbial uptake of nitrogen can be important sinks of nitrogen in SCMs (Lenhart et al., 2012; Messer et al., 2017; Morse et al., 2018; Payne et al., 2014) and can supply organic matter to the sediments of SCMs (Merriman et al., 2017). Organic matter accumulation in SCMs likely has an equivocal effect on denitrification rates in SCMs depending on the depth. In shallower or intermittently inundated SCMs, denitrifying bacteria could utilize accumulated organic matter as a substrate and the concurrent remineralization of organic matter could produce microsites for coupled nitrification-denitrification to occur. Denitrification of nitrate from the water column, however, may be lower in shallower SCMs that are densely vegetated because of competition for nitrate with plants (Morse et al., 2018). In deeper SCMs that have longer residence times, high levels of organic matter could produce extended anoxic conditions in the bottom water. Stratification can occur during warm temperatures in deep SCMs (Song et al., 2013) and can contribute to phosphorus release (Duan et al., 2016; Gold et al., 2017a; Song et al., 2013), driving down N:P ratios in the water column and promoting nitrogen limitation observed in deep SCMs (Gold et al., 2017a; Reed et al., 2016). In some deep SCMs, high rates of remineralization promoted by alternating low and high-oxygen conditions caused by seasonal or storm-based mixing are likely to result in more recalcitrant (i.e., High C:N) carbon pool, which could favor nitrogen fixers over denitrifiers (Eyre et al., 2013; Fulweiler et al., 2013, 2007). Low quality carbon in SCMs could also originate from terrestrial sources (Schroer et al., 2018), and help promote the dominance of nitrogen fixers over denitrifiers. Low-nitrogen and low-oxygen water columns and organic-rich sediments from intense remineralization also could promote DNRA (Burgin and Hamilton, 2007; Kessler et al., 2018), leading to increased internal loading of nitrogen.

Unless plant or algal material is buried, biotic assimilation is only a temporary sink of nitrogen because of the potential for remineralization of nitrogen contained in organic matter. Assimilated nitrogen in SCMs is eventually either buried in sediments, exported downstream, or remineralized and transformed through further biological or chemical transformations. For this reason, traditional studies of SCM nitrogen removal that utilize load-based measurements should aim for sampling periods that span periodicity of both plant and algae growth and senescence and also aim to measure sediment properties (e.g., C:N, organic matter %, etc.). Studies that utilize isotopic tracer methods to partition assimilation between soils, algae, and plants will be especially helpful in better understanding the role of biotic assimilation and remineralization. The process of organic matter accumulation and remineralization should continue to be characterized in SCMs due to its role in internal nutrient loading and nitrogen cycling.

7. Opportunities for research

There is ample opportunity for research on nitrogen cycling within SCMs. This review identified a total of twelve studies that

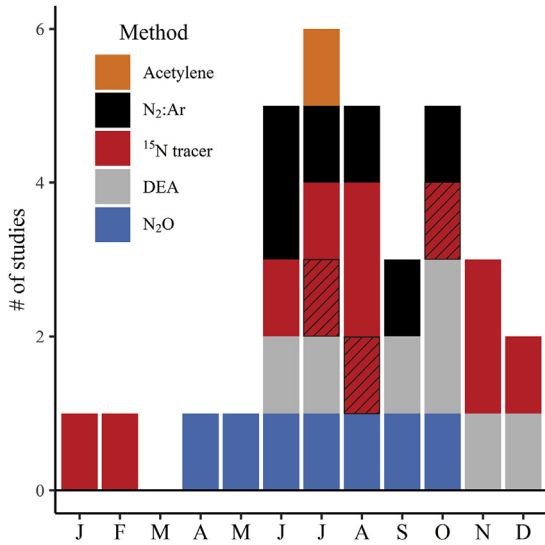


Fig. 2. Histogram of sampling dates from SCM nitrogen cycling studies by month. Cross-hatched bars indicate studies conducted in the southern hemisphere.

reported rates of denitrification within stormwater control measures, of which only four utilized direct measurements (Table 2). This review also found two studies that reported rates or relative importance of DNRA and three that reported rates or relative importance of biotic assimilation. There exists an opportunity to characterize nitrogen cycling processes over all seasons and

locations.

The effectiveness of nitrogen removal from stormwater ponds (via loading measurements) can vary seasonally (Rosenzweig et al., 2011), so seasonal investigations of nitrogen cycling within SCMs are critical for characterizing nitrogen cycling processes and understanding controlling mechanisms. This information is also important for predicting how SCMs will function in the future with increasing effects from climate change. Most studies of nitrogen cycling within SCMs have been conducted during the summer and early fall (Fig. 2), leaving the late fall, winter, and spring months relatively understudied. It should be noted, however, that three ¹⁵N sampling events (Payne et al., 2014) (July, August, and October) were conducted in the southern hemisphere, so this experiment adds resolution to “winter” months (cross-hatched in Fig. 2). Future research should address this gap in knowledge by conducting seasonal or monthly experiments to characterize nitrogen cycling in SCMs throughout the year.

Most of the studies investigating nitrogen cycling in SCMs have taken place in the eastern US (n = 8) (Fig. 3). Two studies were located in the western US in distinct climatic regions (Portland, OR & Phoenix, AZ), one study spanned 8 cities across the US, and one study took place in southeastern Australia (Victoria) (Fig. 3). Due to the varying climatic regions of the study sites and differences in SCM types, seasonal dynamics of nitrogen cycling in SCMs remain unclear at any single site. The types of SCMs that are implemented in different climatic regions can differ drastically (McPhillips and Matsler, 2018), so studies on SCM nitrogen cycling in under-sampled or un-sampled locations (Fig. 3) will be important for characterizing nitrogen cycling for the entire range of SCM types. It

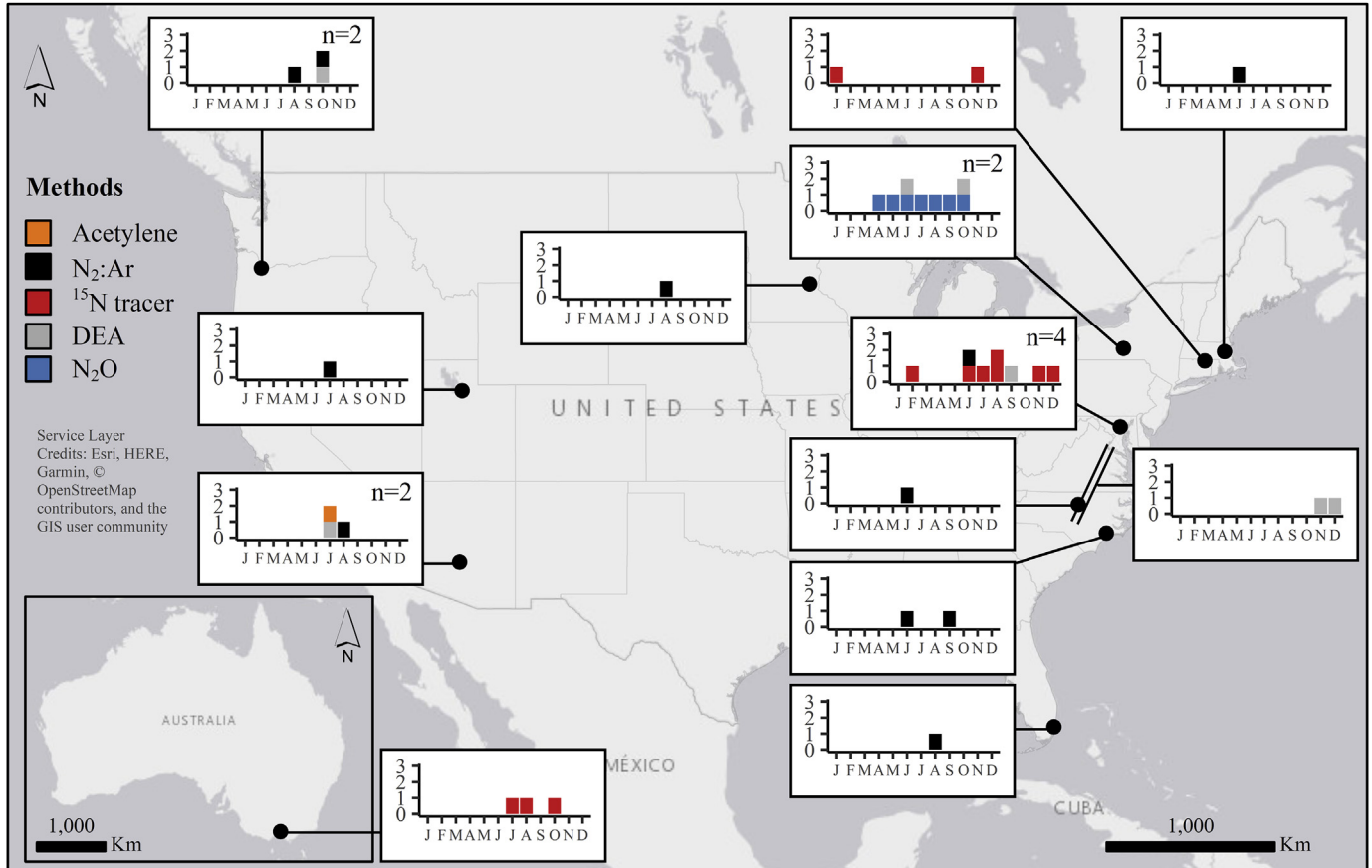


Fig. 3. Study sites of SCM nitrogen cycling studies and histograms of sampling event timing by month.

is important to note that the studies reviewed and topics discussed in this article may be inherently biased towards SCMs that are more prevalent in the eastern US due to the locations of existing SCM nitrogen cycling studies. The in-depth characterization of nitrogen cycling within SCMs at any location, even in systems where nitrogen is not limiting (higher N:P ratio), would be a worthwhile endeavor due to the scarcity of studies on the topic.

Another opportunity for research is the possible tradeoff between the management of nitrogen and phosphorus using SCMs and the interactions between nitrogen and phosphorus cycling in SCMs. The conditions that may promote denitrification in SCMs, such as low-oxygen conditions and a suitable carbon source, can promote the release of inorganic phosphorus from SCM sediments and its export downstream (Collins et al., 2010; Duan et al., 2016; Gold et al., 2017a; Song et al., 2017, 2013). On the other hand, aerobic conditions would decrease phosphorus release from sediments but could discourage denitrification and promote mineralization and nitrification. Rooted vegetation can oxygenate soils and prevent the release of sediment-bound phosphorus, and some studies have recommended that more rooted vegetation can be used to promote phosphorus retention in SCMs (Duan et al., 2016; Mallin et al., 2002). With regard to nitrogen, rooted vegetation in SCMs can increase nitrogen retention within SCMs due to plant assimilation (Messer et al., 2017; Morse et al., 2018; Payne et al., 2014), but the effects on denitrification can vary greatly in aquatic environments from decreased denitrification caused by competition for nitrate with plants (Morse et al., 2018) to increased denitrification due to the stimulation of nitrification-denitrification by plant roots (Kreiling et al., 2011). Also, as noted in section 6, the release of inorganic phosphorus from SCM sediments could promote nitrogen limitation by increasing the N:P ratio, possibly increasing rates of nitrogen fixation or DNRA. Measuring phosphorus cycling in SCMs along with the nitrogen cycling measurements suggested in this review would provide a more complete understanding of how SCMs function and how they can affect downstream water quality.

8. Conclusions

Denitrification is assumed to be an important process of nitrogen removal within SCMs, but very few studies have directly measured this process. Recent studies that utilized direct measurements in SCMs showed that denitrification was less important than previously assumed, and research from other aquatic environments suggests that other nitrogen cycling processes may be important in SCMs.

Nitrogen fixation can exceed denitrification in some open-water SCM sediments. A working hypothesis is that conditions within some open-water SCMs, such as stormwater ponds, could promote the dominance of heterotrophic nitrogen-fixing bacteria over denitrifiers because of the presence of extended anoxic conditions, extreme nitrogen limitation, and poor sediment carbon quality (Fig. 4). Research from estuarine environments has observed the same phenomenon of net sediment nitrogen fixation under similar conditions. Conditions within deeper open-water SCMs, such as organic matter amount and quality, could also promote DNRA or assimilation over denitrification when nitrate is available (Fig. 4), but this area requires additional research. Nitrate uptake in shallow or intermittently inundated SCMs can be dominated by assimilation by plants and soils rather than by denitrification, but assimilation is likely more dominant when DIN is low (Fig. 4). Microbial studies within SCMs have determined that organic matter quality may also play a part in determining the pathway of nitrate reduction (i.e., denitrification vs. DNRA) (Fig. 4). Large amounts of algae reported in open-water SCMs suggests that algal uptake could be

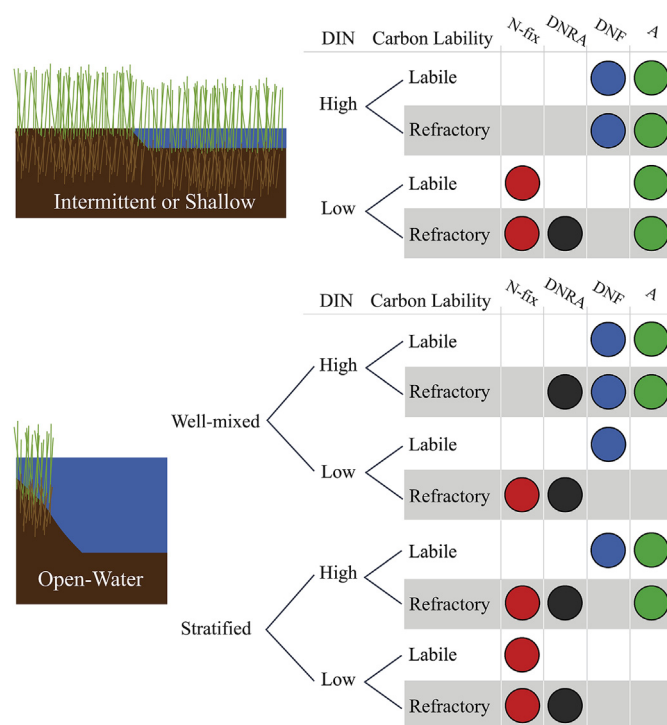


Fig. 4. Conceptual diagram showing nitrogen cycling processes that are hypothesized to be important in different types of SCMs. This diagram does not show hypothesized fate of nitrogen as nitrogen fixation adds nitrogen to the system and both DNRA and assimilation are temporary transformations of nitrogen that can then lead to remineralization, denitrification, burial, or export from the SCM. Note: DIN = Dissolved inorganic nitrogen, N-fix = Nitrogen fixation, DNF = Denitrification, A = Plant, algal, and soil assimilation.

an important pathway of temporary nitrogen removal that can be exported, buried, internally recycled, or denitrified (Fig. 4).

The scarcity of nitrogen cycling measurements within SCMs means that there is abundant opportunity for research. Seasonal variation in SCM function has been poorly characterized in any single location, and the eastern US by far has had the most studies. Seasonal studies of various types of SCMs will be essential for effective stormwater management and water quality improvement in urban areas, especially to plan for the effects of climate change. The connection between nitrogen cycling and phosphorus cycling has not been characterized in SCMs, but future work on this topic is necessary to understand the impacts of nitrogen removal on phosphorus removal and vice versa. Nitrogen cycling in SCMs should be measured by utilizing direct measurements of $N_2:Ar$, mass-balance ^{15}N tracer experiments that capture both nitrogen removal and addition, or microbial methods that include a measure of *nifH* expression with the more commonly measured denitrification genes. The process of nitrogen fixation must be accounted for in future studies because it produces new bioavailable nitrogen to the system. Ideally, a combination of these methods would be used to examine specific nitrogen removal pathways as well as the balance between them. DNRA, assimilation, remineralization (or sediment carbon quantity and quality), and the controlling factors for these processes should also be analyzed so that managers can promote denitrification in SCMs.

SCMs have shown potential to mitigate negative effects of urbanization on hydrology and water quality, but their ability to remove nitrogen has been extremely variable. The internal nitrogen cycling processes that occur in SCMs should be characterized so that managers can implement efficient management strategies that improve SCM function and downstream water quality.

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