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Nitrogen fixation across the aquascape: current perspectives, future priorities

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Abstract Di-nitrogen (N_2) fixation rates, and the diversity of the organisms that fix N_2, remain largely unconstrained in the aquatic landscapes or aquascapes (e.g., lakes, wetlands, streams, rivers, estuaries) between land and sea. As a result, we lack a mechanistic understanding of the controls and contributions of N_2 fixation across disparate aquatic environments, and cannot accurately incorporate this process into local and global nitrogen (N) budgets. This special issue brings together papers highlighting current advances in understanding of N_2 fixation within and across all aquatic habitats, integrating novel methodology for studying N_2 fixation, quantification of N_2 fixation fluxes in understudied habitats, the role of N_2 fixation in biotic assemblages, and the rate and fate of fixed N in heterogeneous landscapes. Together, these papers address important gaps in understanding and highlight the frontiers of research on N_2 fixation in aquatic habitats.

Keywords Nitrogen fixation · Diazotroph · Aquatic · Research coordination network

Di-nitrogen (N_2) fixation, the microbial conversion of N_2 gas to biologically reactive ammonium, is a major component of the global nitrogen (N) cycle and has been extensively studied in open ocean and terrestrial ecosystems (Vitousek et al. 2013; Zehr and Capone 2021). Yet, rates and ecological dynamics remain poorly understood for the inland and coastal aquatic ecosystems (e.g., lakes, wetlands, rivers, streams, estuaries) that connect terrestrial and marine biomes (Fulweiler 2023; Marcarelli et al. 2022). Estimates of N inputs from N_2 fixation to the global N cycle have not been updated in over 3 decades (Zehr and Capone 2021). Warming surface temperatures (Woolway et al. 2022; but see Reinl et al. 2023), changes in nutrient use and loading to aquatic ecosystems (Manning et al. 2020; Oelsner and Stets 2019), and increasing occurrence of harmful cyanobacterial blooms (Burford et al. 2020) make it essential to understand the spatial

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and temporal controls on this key biogeochemical process.

The Aquatic N_2 Fixation Research Coordination Network (ANF RCN) was established in 2021 to cultivate a new paradigm of the fundamental, yet understudied, role of N_2 fixation in ecosystems across the freshwater to marine continuum. To that end, we have identified the following objectives to propel our understanding of aquatic N_2 fixation forward:

- Synthesize the current state of our knowledge of the rates and biodiversity of diazotrophs (organisms that fix N_2) for ecosystems along the freshwater-marine continuum.
- Derive conceptual and quantitative models that characterize the role of biological stoichiometry in constraining N_2 fixation at the population, community, and ecosystem scale, specifically comparing/contrasting the variable stoichiometries of diazotrophs with different metabolic strategies.
- Develop a set of common mathematical and statistical tools enabling the upscaling of N_2 fixation for comparison within and among diverse aquatic ecosystems and in regional and global N budgets.
- Identify major gaps in knowledge that are prohibiting research advancement and generate a plan for coordinating N_2 fixation research focused on these issues.
- Create a community for sharing data, conceptual and quantitative models, and other aquatic N_2 fixation information.

We are addressing the first 3 objectives through a series of workshops held between summer 2022 and spring 2025. The last two objectives are being met through grass-roots network building, including hosting a website (https://www.aquaticnfixation.com/) and email list for information sharing, and organizing a series of special sessions hosted at conferences, all of which are open to everyone interested in and actively working in the broad area of aquatic N_2 fixation research. To date we have held two of these special sessions: “Exploring Rates and Biological Diversity of Nitrogen Fixation Across Land- and Aquascapes” at the AGU Fall Meeting in New Orleans, Louisiana, USA in December 2021, and “Integrating perspectives on nitrogen fixation across the aquascape” at the Joint Aquatic Sciences Meeting hosted by the Consortium of Aquatic Science Societies in Grand Rapids, Michigan, USA in May 2022. This special issue includes papers contributed by authors who were part of these special sessions, as well as others who are part of the extended RCN network. Here, we introduce these papers and their key contributions, organized around four main themes that point to important unanswered questions and potential areas for future study of N_2 fixation across the aquascape.

Novel methods for determining the rates and fate of N_2 fixation

A significant challenge facing researchers who study N_2 fixation are limitations of the commonly-used methods for quantifying process rates (Fulweiler et al. 2015; White et al. 2020). One of the most common methodological applications for measuring N_2 fixation is the acetylene reduction assay (ARA), which has been widely applied to diazotrophic symbionts with plants and free living diazotrophs in soils, sediments, biofilms, and waters. In this method, the nitrogenase enzyme reduces acetylene to ethylene proportionally to the rate by which N_2 is reduced to ammonium. However, a major drawback is that the proportion of ethylene produced to ammonium fixed tends to vary across diazotrophic species and the environments in which they inhabit and many studies do not include study specific conversion factors. Kunza and Hall (2023) test the variability in acetylene reduction compared with $^{15}N_2$ labeled water in benthic biofilms and *Nostoc* populations in four Wyoming streams. They found that the ratio of ethylene to ammonium produced in their assays varied substantially, and that ethylene production increased almost 2 times faster than $^{15}N_2$ uptake as stream water temperature increased, providing the potential for temperature to bias N_2 fixation rates determined using ARA.

Another increasingly used method for detecting N_2 fixation are N_2/Ar fluxes measured using membrane inlet mass spectrometry (MIMS). The idea is that N_2/Ar ratios below or above expected equilibrium concentrations result from processes that consume (e.g., N_2 fixation) or produce (e.g., denitrification) N_2. A large challenge for quantifying rates of N_2 fixation using this method lies in the fact that we are measuring changes in N_2 fluxes against a background of high concentrations - we are essentially flooded in N_2.

Great care is taken in the collection and incubation of water and sediment samples so that we can mimic natural conditions to best describe environmentally relevant fluxes. Such incubations introduce logistical challenges and limits replication. One possible solution lies in in situ measurements of N_2 fluxes, similar to the ecosystem metabolism techniques that use CO_2 or oxygen changes through time to estimate gross primary production, ecosystem respiration, and net primary production. Recent efforts have adapted ecosystem metabolism approaches to use N_2/Ar measurements to estimate daily denitrification (Reisinger et al. 2016) and N_2 removal (Nifong et al. 2020) in streams. In this volume, Taylor et al. (2023) used ecosystem-level experiments to combine in situ N_2/Ar ratios with other measurements of N_2 fixation (whole-ecosystem ^{15}N enrichments) and denitrification (intact sediment core incubations) in replicated ($n = 12$) independent mesocosms enriched with N and P. Overall they observed that mesocosms with NO_3–N concentrations less 0.1 mg L^{-1} had N_2/Ar saturation ratios below equilibrium indicative of net N_2 fixation, while mesocosms with NO_3-N concentrations greater than 1 mg L^{-1} had N_2/Ar values above equilibrium indicative of net denitrification. This study is a major step forward as it highlights that N_2/Ar grab samples are a helpful and complementary tool for a more complete understanding of N_2 fluxes in aquatic systems.

In addition to the difficulties in quantifying ecosystem-scale rates of N_2 fixation, it can be a challenge to identify which diazotrophs are contributing to those rates within and across environments. Here Geisler et al. (2023) apply a novel method which immunolabels nitrogenase (Geisler et al. 2019), and combine this with flow cytometry to quantify free-living unicellular diazotrotrophs. In this way they can determine what percentage of cells in a sample are actively synthesizing nitrogenase. Using this technique, Geisler et al. (2023) demonstrate that only 18% of the cells in a monoculture of *Vibrio natriegens* were synthesizing nitrogenase, and therefore that not all of the cells were fixing N_2.When mixed in co-culture with non-diazotropic *E. coli*, the percentage of *V. natriegnes* cells fixing N_2 stayed the same but overall N_2 fixation rates increased, suggesting the potential for synergetic interactions when diazotrophs are living in mixed microbial communities. Finally, the authors demonstrated the utility of this technique by estimating that diazotrophs ranged from 0.1 to 4.7% of the total bacterial abundance in samples collected from natural marine and freshwater locations. The applications of this technique are potentially widespread, from identifying and quantifying unicellular diazotrophs in habitats where they've been rarely studied, to estimating cell-specific rates of N_2 fixation for physiological studies of diazotrophs, to probing the community ecology of microbial communities where unicellular diazotrophs may be important keystone members.

Contributing new insight on process rates in understudied habitats

Despite decades of research on N_2 fixation across the aquascape, there are still habitats and regions where we understand little about its ecological constraints and the relative rates compared to other biogeochemical processes (Marcarelli et al. 2022). This holds true even in marine ecosystems where most of the research on aquatic N_2 fixation has been conducted. N_2 fixation in N-replete marine environments such as upwelling regions has long been thought to be negligible. In this volume, Fernández Carrera et al. (2023) add to our limited knowledge on N_2 fixation in upwelling regions by studying two locations in the eastern tropical Atlantic Ocean: the seasonal Equatorial upwelling and the Guinea Dome. It has previously been hypothesized that N_2 fixation in the Equatorial upwelling is driven by the residual phosphorus left over after primary production (Subramaniam et al. 2013), while N_2 fixation in the Guinea Dome is driven by atmospheric deposition of iron from the Sahara Desert. Fernández Carrera et al. (2023) found that large colonies *Trichodesmium* were patchily distributed in the euphotic zone of the Guinea Dome, but were found in high abundance towards the edges, which coincided with the highest rates of N_2 fixation and a supply of phosphorus and iron. In contrast, *Trichodesmium* colonies were non-existent in the Equatorial upwelling study sites and N_2 fixation rates were low. Overall, this study aids our knowledge by highlighting the variable distribution of *Trichodesmium* colonies—suggesting that lower resolution sampling, typical of many oceanographic studies, might not correctly capture the spatial distribution of these diazotrophs. Further, the lack of *Trichodesmium* in the Equatorial upwelling implies that other diazotrophs might be responsible for the low rates measured here.

Relative to other habitats along the aquascape, rivers have been relatively understudied, in part because rivers receive and transport reactive N inputs from adjacent terrestrial habitats that should meet the need of microbial assemblages living in these dynamic habitats (Marcarelli et al. 2008). Yet, we know that cyanobacteria are important members of the biofilm assemblages in well-lit streams (Scott and Marcarelli 2012), and riverine algal blooms are increasing in frequency and concern in rivers worldwide (Paerl et al. 2018). In this volume, Valett et al. (2023) leverage a 21-year database of water chemistry and benthic algae data from the Upper Clark Fork River in Montana, USA to show that riverine algal blooms are moderated by snowmelt disturbance, and that late-successional blooms of N_2-fixing *Nostoc cf. pruniforme* are related to N:P ratios among sites. N_2 fixation rates measured in one year at this site show that rates can be exceptionally high during the peak of the bloom formation, although the blooms are short-lived and subside with the end of the growing season in this strongly seasonal, montane river. This study joins a growing body of literature demonstrating that low-nutrient, high-light streams can host benthic cyanobacteria that can fix at very high rates for short periods of time when temperature and nutrient conditions are favorable (e.g., Grimm and Petrone 1997; Kunza and Hall 2023). Further, these ephemeral contributions may be important for supporting productivity and ecological assemblages in these habitats at key times and places.

Role of N_2 fixation in biotic assemblages

How N_2 fixation affects ecological interactions among species in aquatic ecosystems remains poorly understood, partially because of the incredible biodiversity of free-living aquatic diazotrophs (Fernandez et al. 2020) and also because of the incredibly complicated scales of potential interactions (Marcarelli et al. 2022). One possibility is that diazotrophs actively leak recently fixed N into their surrounding environment that may be used by other heterotrophic or autotrophic microorganisms for growth (Mulholland et al. 2006). In contrast, Kunza and Hall (2023, this issue) leveraged their measurements of $^{15}N_2$ uptake to estimate that benthic biofilms in their study streams appeared to release less than 1% of their newly fixed N to the overlying water, although they acknowledge this could be because released N is quickly recycled into biofilms. Indeed, syntrophic relationships between diazotrophs and non-diazotrophs may be common and potentially explain observations of N_2 fixation in unexpected times and places (Fulweiler 2023). Moreover, the cost of N_2 fixation for some diazotrophs in terms of both energy and demography may make them susceptible to competition for other resources. For example, stoichiometric modeling has indicated that heterocytous cyanobacteria may be outcompeted by green algae for P due to their susceptibility to grazing while investing in heterocyte formation and nitrogenase production (Grover et al. 2022). Thus, the role of N_2 fixation in community ecology has tremendous potential for new research explorations.

In this issue, Marino et al. (2023) found unexpected high rates of N_2 fixation on epiphytes of seagrass (*Zostera marina*) in marine lagoons of the northeastern US. N_2 fixation by these phyllosphere diazotrophs was light dependent, but even low light saturated the communities to produce maximum rates. This suggests that the epiphytes were likely cyanobacteria and the relative importance of epiphytic N_2 fixation to the lagoon N budget suggested that these cyanobacteria provide a key ecosystem service to help maintain the health of seagrass communities. Similarly, Kohler et al. (2023) measured the importance of N_2 fixation by *Nostoc* in the N budget of Antarctic streams. They identified three common microbial mat types in pristine streams of the of the McMurdo Dry Valley: black mats typically dominated by *Nostoc*, orange mats dominated by other cyanobacteria such as *Leptolyngbya*, *Oscillatoria*, and *Phormidium*, and green mats dominated by chlorophytes. They found that the distribution of these different microbial mats was spatially segregated both locally and regionally, with black *Nostoc* mats generally occupying stream margins and becoming more dominant further from glacial meltwater sources that can supply dissolved inorganic N to streams. These results point toward a previously unrecognized importance of N_2 fixation as an N source for Antarctic stream communities. Collectively, these papers highlight the need for more research on the ecological role of N_2 fixation whereby diazotoph and non-diazotroph populations interact both positively and negatively due to the novel introduction of ‘new’ reactive N into their environment.

Landscape heterogeneity and connectivity

Although we mostly think of N_2 fixation as a source of N for specific diazotrophs or their close neighbors, fixed N_2 eventually enters the environment either via leakage (Mulholland et al. 2006) or via death and decomposition (Kohler et al. 2018). How that fixed N moves across landscapes and contributes to processes in adjacent or downstream ecosystems has been a particular focus of attention for temperate and tropical forests that host trees with diazotrophic symbionts (Compton et al. 2003; Hedin et al. 2009). In this volume, Ardon et al. (2023) link long-term studies of forest productivity and stream nutrient concentrations from La Selva Biological Station in Costa Rica to demonstrate that in-stream nitrate concentrations were positively related to N_2-fixing trees growing on slopes close to streams, but not to those growing elsewhere in the watershed. This suggests that landscape position and productivity of a few specific tree species may alter the N supply to downstream ecosystems, with consequences for processes like leaf litter decomposition in recipient streams. This suggests that understanding landscape heterogeneity will be important not only for understanding where and when N_2 fixation occurs, but also for understanding the fate of fixed N_2 in ecosystems.

Capturing the spatial variability in ecosystems for any process is complex, and this is especially true for quantifying N_2 fixation and denitrification across ecotones where conditions change rapidly in space and time, such as at the interface between wetlands, streams and lakes. Yet Eberhard et al. (2023) endeavor to do just this as they quantify net N_2 fluxes and environmental characteristics (e.g., temperature, nutrient concentrations, organic matter content) at five sites in Lake Huron and Lake Superior, USA. While they observed that median denitrification rates (914 $\mu g\ N\ m^{-2}\ h^{-1}$) were higher than median N_2 fixation rates (5.5 $\mu g\ N\ m^{-2}\ h^{-1}$) – they found that the denitrification and N_2 fixation were co-occurring in almost 50% of the measured sites across all five sampling locations and all habitats. Their predictive model suggests that organic matter was an important driver of both processes, and P concentrations could be important for N_2 fixation, but that N_2 fixation rates were not predicted by commonly assumed drivers like N availability, N:P ratios, or nutrient limitation status of the biofilms. This study highlights that the spatial scales at which we are able to measure environmental characteristics (e.g., nutrient limitation) may not be relevant to the spatial scales at which we are measuring rates of denitrification and N_2 fixation—that is, what we measure may not be what the microbial community observes. This has important implications for how we design future studies and how we understand the complexities of N cycling across habitats.

Looking forward

The studies in this volume demonstrate the complicated nature of N_2 fixation estimation in variable environments, illustrate the critical role of N_2 fixation in ecological communities, and highlight that the fate of fixed N remains a critical unresolved puzzle for understanding these ecological and biogeochemical dynamics. Together, they point to a variety of areas at the frontier of N_2 fixation research that can advance our understanding of this critical process in the coming decade, such as:

- Development of new tools and refinement of existing protocols to better quantify N_2 fixation rates in time and space, and to probe interactions between diazotrophs and non-diazotrophs in aquatic habitats.
- Quantification of N_2 fixation rates and diazotroph diversity in poorly studied habitats, particularly those that host non-cyanobacterial diazotrophs and those where study has been limited by geographic access, funding limitations and temperate bias.
- Determining the short-term fate of fixed N and how that may vary among different diazotrophs depending on taxonomic identity, metabolic strategy, habitat, and syntrophies with other microbes.
- Incorporating new N derived from N_2 fixation into regional budgets over mid-to-long term time scales, including the transport and remineralization of N incorporated into diazotroph biomass and consequences for adjacent and/or downstream ecosystems.

Advancing knowledge at these frontiers will require a robust and collaborative global network of researchers who are sharing insights, techniques, and knowledge. This special issue is an important reflection of the community of aquatic N_2 fixation researchers that exists, and which we hope will grow and thrive in the years to come.

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Declarations

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References

Ardon M, Clark DA, Marzolf NS, Ramirez A, Pringle CM (2023) Can we see the nitrate from the trees? long-term linages between tropical forest productivity and stream nitrogen concentrations. Biogeochemistry 163:201–218. https://doi.org/10.1007/s10533-023-01030-1

Burford MA, Carey CC, Hamilton DP, Huisman J, Paerl HW, Wood SA, Wulff A (2020) Perspective: advancing the research agenda for improving understanding of *Cyanobacteria* in a future of global change. Harmful Algae 91:101601. https://doi.org/10.1016/j.scitotenv.2018.10.437

Compton JE, Church MR, Larned ST, Hogsett WE (2003) Nitrogen export from forested watersheds in the Oregon Coast range: the role of N_2-fixing red alder. Ecosystems 6:773–785. https://doi.org/10.1007/s10021-002-0207-4

Eberhard EK, Kane ES, Marcarelli AM (2023) Heterogeneity in habitat and nutrient availability facilitate the co-occurrence of N_2 fixation and denitrification across wetland– stream–lake ecotones of Lakes Superior and Huron. Biogeochemistry. https://doi.org/10.1007/s10533-023-01090-3

Fernandez L, Peura S, Eiler A, Linz AM, McMahon KD, Bertilsson S (2020) Diazotroph genomes and their seasonal dynamics in a stratified humic bog lake. Front Microbiol 11:1500. https://doi.org/10.3389/fmicb.2020.01500

Fernandez-Carrera A, Kiko R, Hauss H, Hamilton DS, Achterberg EP, Montoya JP, Dengler M, Brandt P, Subramaniam A (2023) Nitrogen fixation rates in the Guinea Dome and the equatorial upwelling regions in the Atlantic Ocean. Biogeochemistry. https://doi.org/10.1007/s10533-023-01089-w

Fulweiler R (2023) More foxes than hedgehogs: the case for nitrogen fixation in coastal marine sediments. Global Biogeochem Cycles. https://doi.org/10.1029/2023GB007777.e2023GB007777

Fulweiler RW, Heiss EM, Rogener MK, Newell SE, LeCleir GR, Kortebein SM, Wilhelm SM (2015) Examining the impact of acetylene on N-fixation and the active sediment microbial community. Front Microbiol 6:418. https://doi.org/10.3389/fmicb.2015.00418

Geisler E, Bogler A, Rahav E, Bar-Zeev E (2019) Direct detection of heterotrophic diazotrophs associated with planktonic aggregates. Sci Rep 9:9288. https://doi.org/10.1038/s41598-019-45505-4

Geisler E, Siebner H, Rahav E, Bar-Zeev E (2023) Quantification of aquatic unicellular diazotrophs by immunolabeled flow cytometry. Biogeochemistry 164:509–520. https://doi.org/10.1007/s10533-023-01025-y

Grimm NB, Petrone KC (1997) Nitrogen fixation in a desert stream ecosystem. Biogeochemistry 37:33–61. https://doi.org/10.1023/A:1005798410819

Grover JP, Scott JT, Roelke DL, Brooks BW (2022) Competitive superiority of N-fixation *Cyanobacteria* when fixed N is scarce: reconsiderations based on a model with heterocyst differentiation. Ecol Mod 466:109904. https://doi.org/10.1016/j.ecolmodel.2022.109904

Hedin LO, Brookshire ENJ, Menge DNL, Barron AR (2009) The nitrogen paradox in tropical forest ecosystems. Ann Rev Ecol Evol Syst 40:613–635. https://doi.org/10.1146/annurev.ecolsys.37.091305.110246

Kohler TJ, Stanish LF, Liptzin D, Barrett JE, McKnight DM (2018) Catch and release: hyporheic retention and mineralization of N-fixing *Nostoc* sustains downstream microbial mat biomass in two polar desert streams. Limnol Oceanogr Lett 3:357–364. https://doi.org/10.1002/lol2.10087

Kohler TJ, Singley JG, Wlostowski AN, McKnight DM (2023) Nitrogen fixation facilitates stream microbial mat biomass across the McMurdo Dry valleys. Antarctica Biogeochemistry. https://doi.org/10.1007/s10533-023-01069-0

Kunza LA, Hall RO Jr (2023) Temperature affects the ratio of ethylene produced to N_2-fixed more than immediate release of fixed N in streams. Biogeochemistry. https://doi.org/10.1007/s10533-023-01068-1

Manning DWP, Rosemond AD, Benstead JP, Bumpers PM, Kominoski JS (2020) Transport of N and P in US streams and rivers differs with land use and between dissolved and particulate forms. Ecol Appl 30:e02130. https://doi.org/10.1002/eap.2130

Marcarelli AM, Baker MA, Wurtsbaugh WA (2008) Is in-stream N_2 fixation an important N source for benthic communities and stream ecosystems? J N Am Benthol Soc 27:186–211. https://doi.org/10.1899/07-027.1

Marcarelli AM, Fulweiler RW, Scott JT (2022) Nitrogen fixation: a poorly understood process along the freshwater-marine continuum. Limnol Oceanogr Lett 7:1–10. https://doi.org/10.1002/lol2.10220

Marino R, Hayn M, Howarth RW, Giblin AE, McGlathery KJ, Berg P (2023) Nitrogen fixation associated with epiphytes on the seagrass *Zostera marina* in a temperate lagoon with moderate to high nitrogen loads. Biogeochemistry. https://doi.org/10.1007/s10533-023-01083-2

Mulholland MR, Bernhardt PW, Heil CA, Bronk DA, O'Neil JM (2006) Nitrogen fixation and release of fixed nitrogen by *Trichodesmium* spp. in the Gulf of Mexico. Limnol Oceanogr 51:1762–1776. https://doi.org/10.4319/lo.2006.51.4.1762

Nifong RL, Taylor JM, Adams G, Moore MT, Farris JL (2020) Recognizing both denitrification and nitrogen consumption improves performance of stream diel N-2 flux models. Limnol Oceanogr-Meth 18:169–182. https://doi.org/10.1002/lom3.10361

Oelsner GP, Stets EG (2019) Recent trends in nutrient and sediment loading to coastal areas of the conterminous U.S.L insights and global context. Sci Tot Environ 654:1225–1240. https://doi.org/10.1016/j.scitotenv.2018.10.437

Paerl HW, Otten TG, Kudela R (2018) Mitigating the expansion of harmful algal blooms across the freshwater-to-marine continuum. Environ Sci Technol 52:5519–5529. https://doi.org/10.1021/acs.est.7b05950

Reinl KL, Harris TD, North RL et al (2023) Blooms also like it cold. Limnol Oceanogr Lett 8:546–564. https://doi.org/10.1002/lol2.10316

Reisinger AJ, Tank JL, Hoellein TJ, Hall RO (2016) Sediment, water column, and open-channel denitrification in rivers measured using membrane-inlet mass spectrometry. J Geophys Res-Biogeosci 121:1258–1274. https://doi.org/10.1002/2015jg003261

Scott JT, Marcarelli AM (2012) *Cyanobacteria* in freshwater benthic environments. In: Whitton BA (ed) Ecology of *Cyanobacteria* II. Springer, Dordrecht

Subramaniam A, Mahaffey C, Johns W, Mahowald N (2013) Equatorial upwelling enhances nitrogen fixation in the Atlantic Ocean. Geophys Res Lett 40:1766–1771. https://doi.org/10.1002/grl.50250

Taylor JM, Andersen IM, Hoke AK, Kelly PT, Scott JT (2023) In-situ N_2: Ar ratios describe the balance between nitrogen fixation and denitrification in shallow eutrophic experimental lakes. Biogeochemistry. https://doi.org/10.1007/s10533-023-01063-6

Valett HM, Feijó de Lima R, Peipoch M, Engstrom RC (2023) Bloom succession and nitrogen dynamics during snowmelt in a mid-order montane river. Biogeochemistry. https://doi.org/10.1007/s10533-023-01080-5

Vitousek PM, Menge DNL, Reed SC, Cleveland CC (2013) Biological nitrogen fixation: patterns and ecological controls in terrestrial ecosystems. Phil Trans R Soc B 368:20130119. https://doi.org/10.1098/rstb.2013.0119

White AE, Granger J, Selden C et al (2020) A critical review of the 15N2 tracer method to measure diazotrophic production in pelagic ecosystems. Limnol Oceanogr-Meth 18:129–147. https://doi.org/10.1002/lom3.10353

Woolway RI, Sharma S, Smol JP (2022) Lakes in hot water: the impacts of a changing climate on aquatic ecosystems. Bioscience 72:1050–1061. https://doi.org/10.1093/biosci/biac052

Zehr J, Capone DG (2021) Marine nitrogen fixation. Springer, Switzerland