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Temperature Sensitivity of Freshwater Denitrification and N₂O Emission—A Meta-Analysis

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Key Points:

- One degree warming enhances denitrification and N₂O emissions 8%–14 %. *In-situ* Q_{10,N₂O} is 2 times lower than temperature manipulation Q_{10,N₂O}
- Q_{10,denitrification} increases with nitrogen and O₂ availability. SOM correlations differ between *in-situ* and temperature manipulation studies
- Adapted Q_{10,EF5} is 6.6 times higher in the Australia and New Zealand region compared to regions on the northern hemisphere

Supporting Information:

Supporting Information may be found in the online version of this article.

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Abstract Freshwater denitrification removes a considerable amount of nitrogen from inland waters, which are under pressure from eutrophication and warming. However, incomplete denitrification can lead to the formation of N₂O, a potent greenhouse gas, which can amplify climatic warming. Although temperature effects on denitrification are well studied in individual habitats and experiments, global patterns in temperature-responses of denitrification and N₂O emissions remain to be elucidated. Here, we investigated the temperature sensitivity (Q₁₀) of denitrification and N₂O emissions in freshwater ecosystems worldwide, using a meta-analytic approach. To this end, Q₁₀ values from *in-situ* and temperature manipulation studies were related to environmental nutrient conditions, O₂, pH, sediment organic matter (SOM), and geographic location. Temperature sensitivity of denitrification displayed a strong positive correlation with environmental nitrogen concentrations, pH and O₂. Significant correlations with SOM and SOM:N ratios were observed as well, but the direction of the effect differed between *in-situ* and temperature manipulation studies. Surprisingly, temperature sensitivity of N₂O emissions did not correlate with pH, SOM, nutrient or O₂ conditions. Temperature sensitivity of the ratio between N₂O emission and NO₃⁻ concentration (adapted EF₅ values) was 6.6 times higher in Australia and New Zealand compared to other geographic regions. As global temperatures and nitrogen deposition in freshwater ecosystems are expected to increase over the coming decades, our results suggest enhanced future denitrification, which may present a natural way to balance eutrophication. The observed temperature sensitivity of N₂O emission factors, however, may indicate enhanced denitrification-derived N₂O emissions from freshwater ecosystems in a future warmer world.

1. Introduction

Humankind has altered a myriad of natural processes around the globe. Among its most impactful are the increase in global temperatures due to greenhouse gas emissions (Pachauri et al., 2014) and increase in nitrogen (N) fertilizer use (Steffen et al., 2015). Artificial nitrogen fertilizer is produced on an industrial scale using technological advancements such as the Haber-Bosch reaction (Smil, 2004). The usage of this N-fertilizer in agriculture enabled the steep human population growth since industrial times (Galloway et al., 2004). However, newly created nitrogen unintentionally also ends up in aquatic ecosystems (Seitzinger & Kroeze, 1998), where it contributes to eutrophication related issues such as cyanobacterial blooms, hypoxia, fish kills and reduced biodiversity (Smith et al., 1999). Global warming can further exacerbate the eutrophication-related issues in freshwater ecosystems, such as lakes (Moss et al., 2011).

Microbial denitrification presents a way to counteract the harmful effects of eutrophication, as it removes reactive nitrogen from the aquatic environment (Seitzinger et al., 2006). This redox process, mostly taking place in sediments, is a stepwise transformation to gaseous N₂. The reaction requires an electron donor—in most freshwaters organic carbon—and an oxidized nitrogen compound (NO₃⁻ and NO₂⁻) as electron acceptor. Incomplete denitrification however, leads to the production of N₂O (Knowles, 1982). Inland waters are estimated to emit between 148 and 277 Gg N-N₂O each year (Maavara et al., 2019). While denitrification is commonly referred to as the main source of N₂O in anoxic conditions, spikes in N₂O concentrations can also be the product of other (microbial and physical) processes, such as nitrification and transport of dissolved N₂O from groundwater (Jurado et al., 2017). N₂O emission to the atmosphere is problematic, as it is a potent greenhouse gas, with a 200–300 times stronger global warming potential than CO₂ (Neubauer & Megonigal, 2015; Seitzinger et al., 2000). Additionally, N₂O has the potential to deplete ozone and it is currently the dominant ozone-depleting substance in the stratosphere (Ravishankara et al., 2009).

Similar to other biological processes, denitrification is affected by temperature. Currently, the body of literature on the effects of global warming on denitrification in freshwater ecosystems is growing steadily (see for instance Palacin-Lizarbe et al. (2018) for a recent overview in lakes). While a straightforward increase in denitrification with warming is generally expected, examples of positive, negative and neutral responses can all be found in the literature (Brin et al., 2017; Tomaszek et al., 1997; Veraart et al., 2011). This strengthens the belief that the temperature sensitivity of denitrification is dependent on its surrounding environment. For example, denitrification takes place under hypoxic to anoxic conditions (Knowles, 1982), and temperature-regulated oxygen dynamics have been shown to amplify the effect of temperature on denitrification (de Klein et al., 2017; Veraart et al., 2011). Moreover, as canonical denitrification requires both nitrogen and organic matter (OM) as a substrate, the availability of both degradable OM and NO_3^- is expected to play a role in its response to elevated temperatures. Under low substrate availability, denitrification will be primarily constrained by substrate, minimizing temperature effects. At higher substrate availability, denitrification will be released from substrate limitation, possibly enabling a stronger response to increased temperatures.

Although the temperature effect on denitrification is well studied in individual habitats and experiments (see for instance Klaus et al., 2018; Messer & Brezonik, 1984; Veraart et al., 2011), less is known about the overarching patterns in temperature responses of denitrification and resulting N_2O emissions. Global patterns in denitrification rates across freshwaters can be mirrored by variations in nitrogen loading to the respective water bodies (Seitzinger et al., 2006) and related socio-economic factors such as human population density and use of nitrogen-rich fertilizers (Seitzinger et al., 2000). Modelled aquatic N_2O emissions are significantly higher in the northern hemisphere (Seitzinger et al., 2000). Projected temperature increases in temperate and polar regions by the end of the century are profoundly larger than those at lower latitudes (Pachauri et al., 2014). We therefore hypothesize that temperature sensitivity of denitrification in freshwater ecosystems displays spatial heterogeneity, following variation in nitrogen loading and temperatures.

To connect rate predictions to potential N_2O emission under a range of global change scenarios, it is important to take into account the amount of N_2O emitted relative to nitrogen load. This is expressed by the N_2O emission factor EF5, which is used by the Intergovernmental Panel on Climate Change (IPCC) to calculate indirect N_2O emissions from a suite of ecosystems (Well et al., 2005). In freshwater ecosystems, accurate estimates of nitrogen loading are not always available. Therefore, the N_2O -N/ NO_3^- -N mass ratio can be used as an adaptation to calculate EF5 values (Hama-Aziz et al., 2017). This ratio reflects the efficiency of N_2O production compared to the nitrogen substrate. Previous research indicates that EF5 values can differ substantially between sites (Clough et al., 2007) and can vary with environmental conditions such as temperature (Yang et al., 2021). However, to our knowledge there has been no global overview of this temperature sensitivity in freshwater ecosystems to date.

In this study, we summarize current knowledge on temperature sensitivity of global freshwater denitrification rates, N_2O emissions, and adapted EF5 values, using a meta-analytic approach. First, we constructed a database composed of published data gathered from a systematic digital literature search. Using this database, we tested the hypothesis that temperature sensitivity (expressed as Q10) of denitrification rates and N_2O emissions is dependent on substrate availability. Study-specific Q10 values were hypothesized to be positively correlated with nitrogen concentrations in the sediment and the water column and sediment organic matter. An additional positive correlation between temperature sensitivity and phosphorus (P) concentrations in the sediment and in the water column was anticipated, as P is indicative of eutrophic conditions that stimulate bacterial denitrification (Seitzinger & Nixon, 1985). We furthermore hypothesized a negative correlation between Q10 values and oxygen (O_2) concentrations.

2. Methods

To determine the global patterns in temperature-responses of denitrification and N_2O emissions, we compiled a database from the published literature on denitrification and N_2O emission rates in freshwater ecosystems under varying temperatures. We were specifically interested in two types of datasets: (a) *in-situ* measurements of temperature in freshwater ecosystems (e.g., lakes, reservoirs, wetlands, rivers and ditches) and (b) experimental temperature manipulations. This implied that the publication either included *in-situ* measurements of temperatures and denitrification rates or N_2O emissions (henceforth referred to as *in-situ* temperature dataset), or the publication included freshwater experiments in which temperature was manipulated and denitrification and/or

N₂O was measured (temperature manipulation dataset). For this purpose, we carried out a systematic literature review using Web of Science (<https://www.webofknowledge.com/>) on the 6th of March 2020 using the following query on paper titles, abstracts and keywords: ((freshwater* OR lake* OR aquatic OR pond* OR reservoir* OR lagoon* OR river* OR stream* OR creek* OR brook* OR ditch* OR surface water* OR wetland* OR *marsh*) AND (temperature* OR warming OR heating OR Q10 OR "Q(10)") AND (denitrification OR N₂O OR "nitrous oxide" OR "acetylene inhibition" OR denitrifying)). Initially, papers were screened for data on temperature - measured in the sediment or water -, denitrification rates and/or N₂O emissions in the graphs and tables. Studies were included if the *in-situ* sites were permanently water-bearing (or at least for the duration of the study period) and contained at least two combined measurements of denitrification rates or N₂O emissions and temperature. Whenever papers reported from several conducted experiments or measurements from distinct locations, these were considered as unique case-studies. To focus on freshwater studies, studies from salt marshes, estuaries or other tidal systems were not included. Studies on constructed wetlands and wastewater treatment systems were excluded as well. As our literature search rendered very few studies on the southern hemisphere, we supplemented the database with data on Uruguayan lakes from Veraart et al. (2012). Furthermore, the dataset was supplemented with a Finnish lake dataset (Rissanen et al., 2013). Data were extracted from publications using the Java program *Plot Digitizer* (Huwaldt & Stienhorst, 2013). When reported, data on sample size, nitrogen and phosphorus concentrations in the water column and/or sediment, sediment organic matter content (SOM), dissolved O₂ concentrations and pH were extracted as well. As denitrifiers may respond differently to short-term nutrient enrichment than long-term background nutrient concentrations, we recorded both responses separately. Total nutrient concentrations in the water column were calculated as the sum of nutrient enrichment and background concentrations. Not all studies reported on the same forms of nitrogen and phosphorus. Therefore, we improved comparability between studies by using the reported total nitrogen (TN) and total phosphorus (TP) concentrations as a proxy of lake N and P concentrations. In case-studies where TN and TP were not reported, TN concentrations were approximated by the sum of the individual N-species (NO₃⁻, NO₂⁻ and NH₄⁺) reported, and TP concentrations were equated to Soluble Reactive Phosphorus or PO₄³⁻ concentrations. If needed, we converted units to ensure comparability between studies. Additionally, information on the measurement method, system type (lake, river, pond, stream etc.) and its geographic location (latitude, longitude, geographic region, climate zone and country) was included. This resulted in a database with a total of 140 papers that contained 215 unique case-studies for the *in-situ* temperature dataset and 144 for the temperature manipulation dataset (Figure S1 in Supporting Information S1).

2.1. Calculations and Statistics

All calculations and statistics were carried out in R (R Core team 2013). Q10 values for denitrification rates and N₂O emissions were calculated for each dataset using the function *Q10* (package *respirometry* (Birk, 2017)). A cut-off of Q10 > 50 was used to remove extremely high Q10 values ($n = 21$; 5.9% of the analyzed dataset) from the *in-situ* dataset because these values might represent incorrect model fits. An overview of the studies incorporated in the two datasets can be found in Table S1 of Supporting Information S1. The overall system temperature coefficient θ_s was derived from the Q10 values. A doubling of reaction rates over a 10° temperature range corresponds with a θ_s of around 1.07 (Veraart et al., 2011). Differences in Q10 values between the two datasets were tested with a one-way ANOVA (function *aov*). To improve ANOVA-assumptions of normality and homogeneity of variance, data were log-transformed in the analysis. As Q10 values significantly differed between these datasets, other categorical variables (geographic region, climate zone, measurement method) were tested using two-way ANOVA with log-transformed Q10 values (function *aov*). Correlations between denitrification and N₂O emissions were determined by linear regression (function *lm*). Relationships between the Q10 values and nutrient conditions, pH, SOM, O₂ and latitude were investigated using generalized linear models (function *glm*) with a gamma distribution. When necessary, a log or inverse link was used in the glm models to improve normality of the residuals.

2.2. N₂O Emission Factors (EF5)

Indirect N₂O emission factors (EF5) were calculated according to Hama-Aziz et al. (2017). Ideally, N-N₂O concentrations are related to N-NO₃ concentrations for this calculation. However, we adapted the calculation due to limited data availability. To be specific, N-N₂O emissions (in $\mu\text{g m}^{-2} \text{h}^{-1}$) were divided by the dissolved

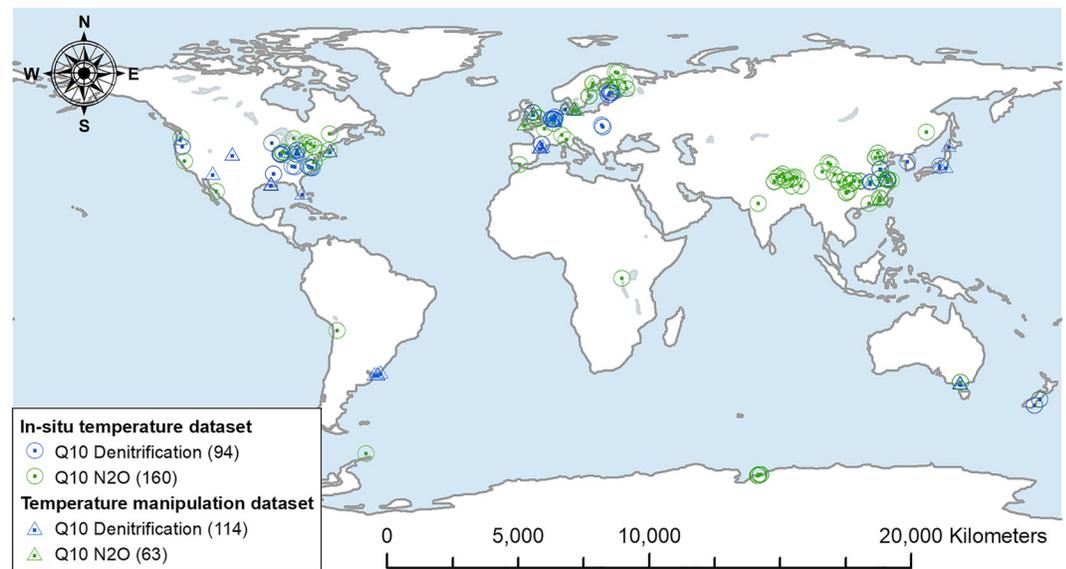


Figure 1. Global map showing the geographic locations of the *in-situ* temperature (○) and temperature manipulation (△) datasets. Locations of $Q10_{\text{denitrification}}$ values are indicated in blue and $Q10_{\text{N}_2\text{O}}$ in green. Sample sizes are indicated between brackets.

N-NO_3^- concentration (in $\mu\text{g L}^{-1}$). Our adapted EF5 values reflect the ratio of N_2O emissions to NO_3^- concentration and provide a relative measure for its conversion efficiency. Temperature sensitivity of these adapted EF5 values were calculated using the function $Q10$ (package *respirometry* (Birk, 2017)). A cut-off of $Q10 > 50$ was used to remove extremely high $Q10$ values ($n = 24$; 16% of the dataset). Relationships between the $Q10$ values and latitude were investigated using generalized linear models (function *glm*). The categorical variables "geographic region" and "climate zone" were tested using one-way ANOVA (function *aov*). Data was either log- or square root-transformed to improved normality of the residuals. In the *in-situ* dataset, the geographic regions South America, Central America, Southern Europe and Southern Asia were excluded from analysis due to low sample size ($n < 3$).

3. Results

Temperature sensitivity ($Q10$) values for denitrification tended to be higher in the temperature manipulation dataset compared to the *in-situ* dataset (3.6 ± 5.5 and 2.3 ± 4.7 , respectively, mean \pm SD), though this effect was not significant ($F_{(1,206)} = 3.15$, $P = 0.08$). These $Q10$ values correspond to system temperature coefficient (θ_s) values of 1.14 and 1.09 in the temperature manipulation and *in-situ* dataset. The $Q10$ values for N_2O emissions were significantly higher in the temperature manipulation dataset (4.5 ± 8.6 ; $\theta_s = 1.16$; $n = 63$) than those originating from the *in-situ* temperature dataset (2.2 ± 4.4 , $\theta_s = 1.08$; $n = 160$; $F_{(1,221)} = 7.25$, $P < 0.01$; Figure 2). Therefore, the *in-situ* temperature and temperature manipulation datasets are considered separately in further analyses. $Q10$ values calculated on denitrification rates were significantly higher than those calculated on N_2O emissions (one-way ANOVA; $F_{(1,429)} = 11.35$, $P < 0.001$).

As expected, denitrification rates correlated strongly with N_2O emissions (Figure 3, $R^2 = 0.81$, $P < 0.001$). There was no such correlation between the $Q10$ values of denitrification and N_2O emissions observed ($R^2 = -0.01$, $P = 0.39$). Most studies in the datasets originated from Europe ($n = 171$), Asia ($n = 112$) and North America ($n = 65$) and there was a general lack of data from both the southern hemisphere ($n = 42$) and tropical regions in particular ($n = 1$) (Figure 1). $Q10$ values for temperature manipulation datasets were not significantly different between climate zones or measurement methods (Table S2 in Supporting Information S1). *In-situ* $Q10_{\text{denitrification}}$ values, however, were significantly different between climate zones and measurement methods (Table S2 in Supporting Information S1). Tukey post-hoc tests indicated that these $Q10$ values were significantly lower in temperate compared to continental climatic regions (Table S3 in Supporting Information S1). Furthermore, *in-situ* $Q10_{\text{denitrification}}$ values measured with the acetylene inhibition method were significantly higher than those

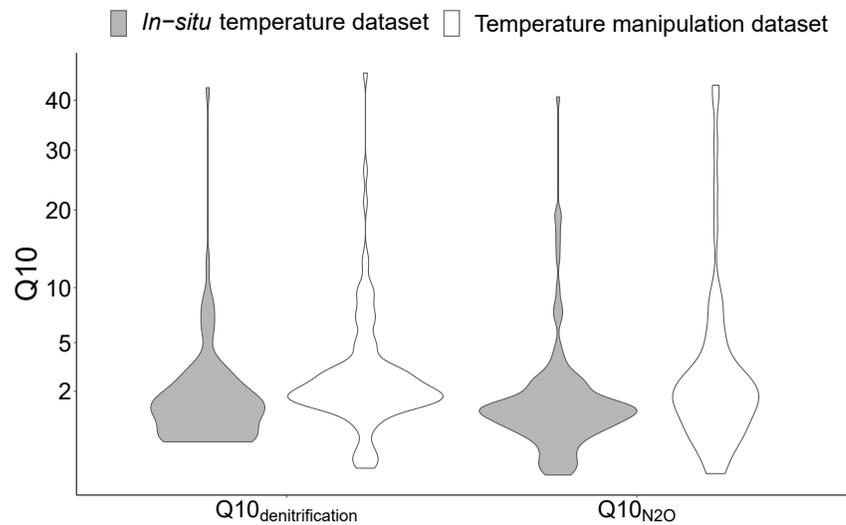


Figure 2. Violin plot depicting $Q10_{\text{denitrification}}$ and $Q10_{\text{N}_2\text{O}}$ values calculated from the *in-situ* temperature and the temperature manipulation dataset. Note that the Y-axis is square root-transformed to improve readability.

measured with the ^{15}N -tracer method (Table S3 in Supporting Information S1, $P = 0.03$), while no significant differences between the other methodologies were observed.

3.1. Relationships Between $Q10_{\text{denitrification}}$, $Q10_{\text{N}_2\text{O}}$ and Environmental Variables

In the *in-situ* temperature dataset, $Q10_{\text{denitrification}}$ values increased exponentially with N concentrations in the water column and sediment and with dissolved O_2 concentration, while $Q10_{\text{denitrification}}$ decreased with SOM and P concentration in the water column (Table 1, Figure 4). Similarly, we observed a negative correlation between $Q10_{\text{denitrification}}$ values with the SOM:N ratio in the sediment (Figure 4, Table 1). The correlation between N concentrations and $Q10_{\text{denitrification}}$ was not apparent for the individual N-species (Figure S2, Table S4 in Supporting Information S1). These correlations were not observed in the temperature manipulation dataset. In this dataset, $Q10_{\text{denitrification}}$ values correlated positively with SOM, pH values and sediment SOM:N ratio (Figure 4). No correlations between $Q10_{\text{denitrification}}$ values, N enrichment and sediment P concentrations were observed (Figure S3 in Supporting Information S1).

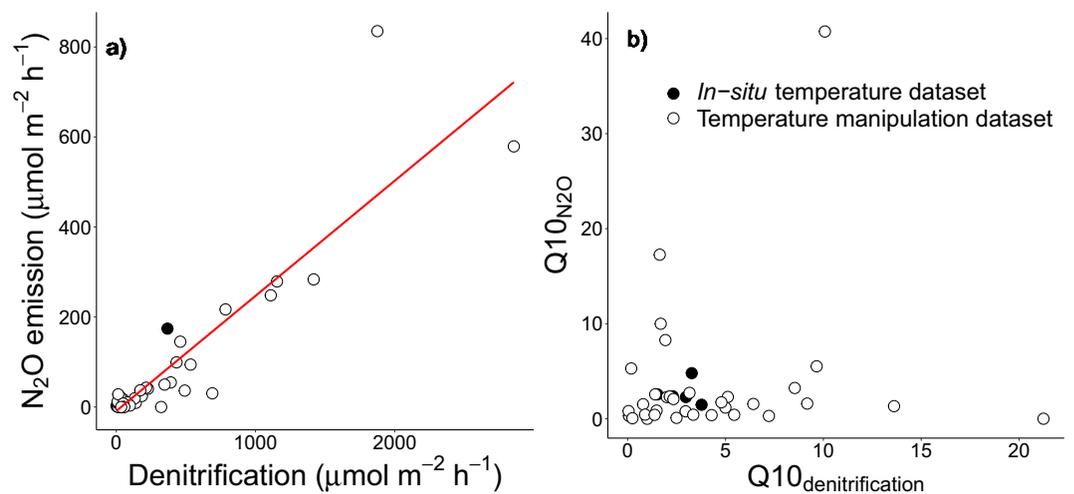


Figure 3. Correlation between study-specific mean denitrification rate and N_2O (a) and $Q10_{\text{denitrification}}$ and $Q10_{\text{N}_2\text{O}}$ (b). Data points from the *in-situ* temperature dataset are indicated in black and from the temperature manipulation dataset in white ($N = 5$ and $N = 41$, respectively). Significant linear regression is indicated, with $P < 0.001$ and $R^2 = 0.81$.

Table 1

Statistical Results, Showing the Correlation Coefficients From Generalized Linear Model (glm) Analysis for $Q10_{denitrification}$, $Q10_{N2O}$ and $Q10_{EF5}$ Values in the In-Situ Temperature ($n = 94, 160$ and 87 , Respectively) and Temperature Manipulation Datasets ($n = 112, 63$ and 35 , Respectively)

Factor	Model family	Sample size	Estimate	Standard error	t-value	P-value
<i>In-situ temperature dataset</i>						
$Q10_{denitrification}$						
N in water column	Log gamma	84	0.00139	0.000491	2.828	0.00589**
N in sediment	Log gamma	25	0.117	0.0385	3.052	0.00565**
P in water column	Gamma	42	-0.000687	0.000311	-2.212	0.0327*
P in sediment	Log gamma	13	0.631	0.502	1.258	0.234
SOM	Log gamma	51	-0.037	0.018	-2.035	0.047*
SOM:N ratio	Log gamma	18	-0.005	0.001	-4.987	0.0001***
pH	Log gamma	40	0.148	0.320	0.461	0.648
O₂	Log gamma	54	0.134	0.049	2.738	0.008**
Distance from equator	Log gamma	94	-0.016	0.020	-0.810	0.420
$Q10_{N2O}$						
N in water column	Log gamma	99	-0.00057	0.00079	-0.725	0.471
N in sediment	Gamma	44	0.0214	0.0364	0.586	0.561
P in water column	Log gamma	49	-0.0108	0.0108	-0.997	0.324
P in sediment	Gamma	3	10.086	7.292	1.383	0.399
SOM	Log gamma	10	-0.029	0.021	-1.385	0.203
SOM:N ratio	Log gamma	10	-0.031	0.021	-1.474	0.179
pH	Gamma	99	0.017	0.066	0.259	0.796
O ₂	Gamma	89	-0.007	0.039	-0.174	0.862
Distance from equator	Inverse gamma	160	0.005	0.005	1.036	0.302
$Q10_{EF5}$						
Distance from equator	Log gamma	87	-0.002	0.014	-0.122	0.903
<i>Temperature manipulation dataset</i>						
$Q10_{denitrification}$						
Total N in water column	Gamma	81	1.706e-5	3.144e-5	0.543	0.589
N enrichment in water column	Log gamma	112	-4.886e-5	1.065e-4	-0.459	0.647
N in sediment	Log gamma	46	-0.0684	0.0344	-1.992	0.0527*
Total P in water column	Log gamma	24	-0.004	0.003	-1.109	0.279
P enrichment in water column	Log gamma	3	-0.016	0.007	-2.396	0.252
P in sediment	Log gamma	5	-0.244	0.167	-1.464	0.239
SOM	Log gamma	58	0.030	0.015	2.04	0.046*
SOM:N ratio	Log gamma	40	0.000412	0.000101	4.072	0.000228***
pH	Gamma	51	0.116	0.035	3,317	0.00172**
O ₂	Log gamma	15	-0.012	0.112	-0.107	0.916
Distance from equator	Log gamma	114	-0.020	0.012	-1.633	0.105
$Q10_{N2O}$						
Total N in water column	Gamma	42	-5.998e-5	3.999e-5	-1.500	0.141
N enrichment in water column	Log gamma	63	0.00121	0.000770	1.573	0.121
N in sediment	Log gamma	26	0.126	0.0887	1.420	0.168
Total P in water column	Log gamma	24	0.0567	0.0284	1.998	0.0583*
P enrichment in water column	Log gamma	3	0.005	0.043	0.116	0.927

Table 1
Continued

Factor	Model family	Sample size	Estimate	Standard error	t-value	P-value
P in sediment	Log gamma	7	-0.095	0.364	-0.262	0.804
SOM	Log gamma	31	-0.004	0.046	-0.080	0.937
SOM:N ratio	Log gamma	22	-0.015	0.049	-0.296	0.771
pH	Log gamma	26	0.167	0.423	0.395	0.697
O ₂	Log gamma	7	-0.059	0.061	-0.971	0.376
Distance from equator	Log gamma	63	0.011	0.024	0.477	0.635
<i>Q10_{EF5}</i>						
Distance from equator	Log gamma	35	0.024	0.021	1.138	0.263

Note. SOM, sediment organic matter. Significant factors and values are indicated in boldface, with *** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$, $P < 0.10$.

In contrast to $Q10_{\text{denitrification}}$, $Q10_{\text{N}_2\text{O}}$ values did not correlate with any of the selected environmental variables in either the temperature manipulation or *in-situ* temperature datasets (Table 1, Figure S4 in Supporting Information S1). In the temperature manipulation dataset, $Q10_{\text{N}_2\text{O}}$ values tended to increase with P concentration in the water column, but this correlation was non-significant ($P = 0.06$).

3.2. Temperature Sensitivity of N₂O Emission Factors

The temperature sensitivity of adapted emission factors (EF5, see methods) was calculated for the studies that reported both N₂O emissions and dissolved NO₃⁻ concentration. In the *in-situ* dataset, $Q10_{\text{EF5}}$ values were 1.82 ± 2.87 (mean \pm SD, $n = 87$) and significantly differed between geographic regions (Table S5 in Supporting Information S1, Figure 5). To be specific, $Q10_{\text{EF5}}$ values were 6.6 times higher in the region Australia and New Zealand compared to regions on the northern hemisphere (Tukey-HSD test, $P < 0.05$, Table S6 in Supporting Information S1). Furthermore, no significant differences between climate zones were observed for the $Q10_{\text{EF5}}$ values calculated on the *in-situ* dataset. No correlations between $Q10_{\text{EF5}}$ values and distance from equator were observed in either *in-situ* and temperature manipulation dataset (Table 1, Figure S5 in Supporting Information S1).

In the temperature manipulation dataset, average $Q10_{\text{EF5}}$ values were 6.90 ± 10.98 (mean \pm SD, $n = 35$). No significant differences between climate zones nor geographic regions were observed (Table S5 in Supporting Information S1).

4. Discussion

This meta-analysis compiles and synthesizes current knowledge on temperature sensitivity of freshwater denitrification and N₂O emissions. A total number of 359 $Q10$ values from 144 temperature manipulation case-studies and 215 *in-situ* case-studies of temperature and denitrification/N₂O could be incorporated into the analysis. Our results showed a positive correlation between temperature sensitivity of denitrification and environmental nitrogen concentrations, pH and O₂ (see Figure 6 for a conceptual overview of our meta-analytic results). Significant correlations with SOM and SOM:N ratios were observed as well, but the direction of the effect differed between *in-situ* and temperature manipulation datasets. Surprisingly, temperature sensitivity of N₂O emissions did not correlate with any environmental variables included in the analysis. Possibly, this could be due to the multitude of physical processes and microbial guilds producing N₂O, which can all directly or indirectly be affected by various environmental conditions. Additionally, various inhibiting factors for N₂O consumption, such as oxygen (Ji et al., 2018) and pH (Blum et al., 2018), could also be a factor in this discrepancy. Below, we discuss the observed relationships between the temperature sensitivity of freshwater denitrification, N₂O emissions, N₂O emission factors and environmental conditions.

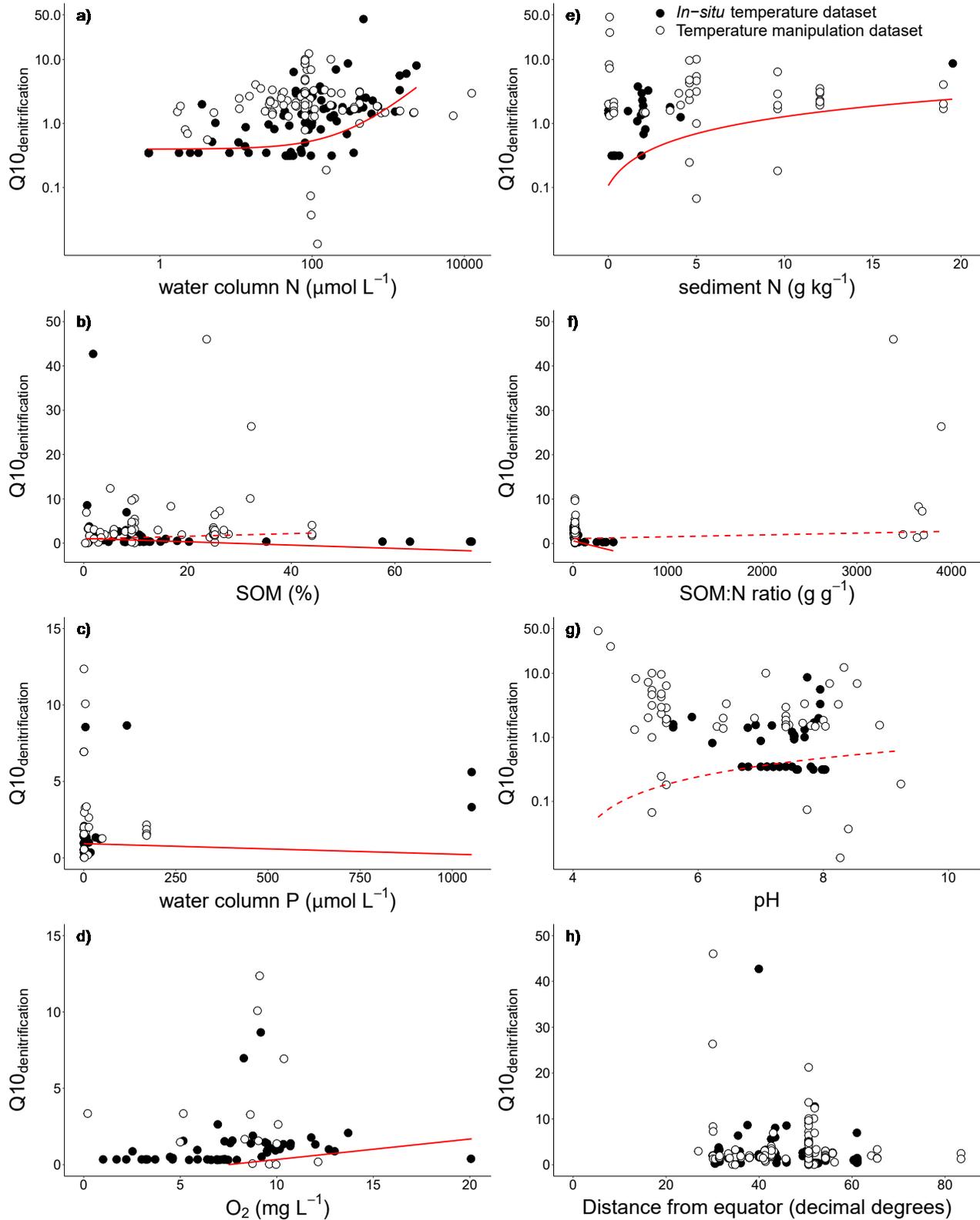


Figure 4. Correlation between Q_{10} denitrification values and physical-chemical conditions in water and sediment. Data from the *in-situ* temperature dataset are depicted in black and data from the temperature manipulation dataset in white. Red lines show significant results from *glm* analysis, with solid and dashed lines for *in-situ* measurements and temperature-manipulation datasets, respectively. Note that the X-axis of panel a and the Y-axis of panels a, e and g are log-transformed to improve readability.

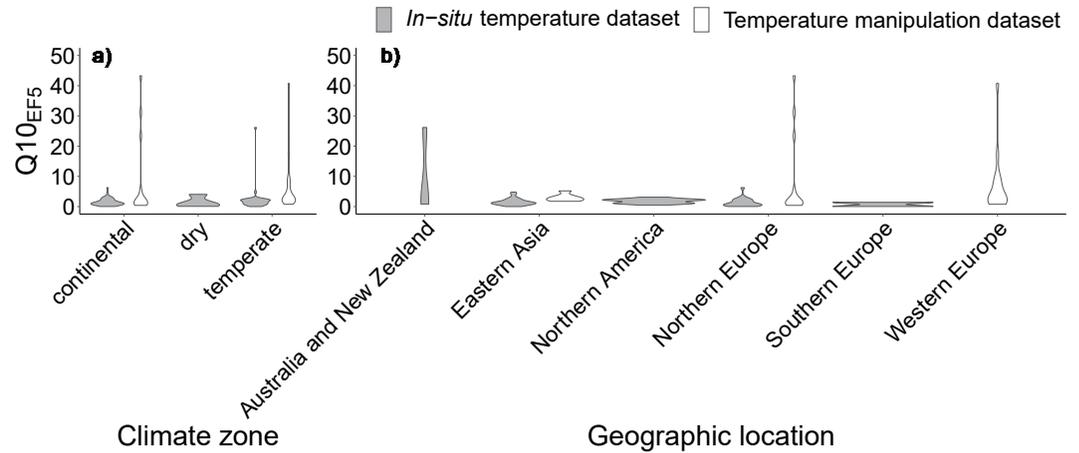


Figure 5. Violin plot depicting $Q10_{EF5}$ values calculated from the *in-situ* temperature and temperature manipulation dataset across climate zones (a) and geographic regions (b). Statistical output is reported in Table S5 of Supporting Information S1.

4.1. Substrate Availability Drives Temperature Sensitivity of Denitrification

Worldwide, lakes are estimated to cover about 5×10^6 km² (Verpoorter et al., 2014) and rivers and streams 0.8×10^6 km² (Allen & Pavelsky, 2018). Understanding the impact of global warming on the ecological processes in freshwater ecosystems is important to predict the behaviour of these ecosystems in the future. The results of our study demonstrate that temperature sensitivity of freshwater denitrification relates positively to the surrounding nitrogen availability in both sediment and water. Specifically, we observed a positive correlation between $Q10_{denitrification}$ and nitrogen concentrations in both the water column and the sediment in the *in-situ* temperature dataset. This synergistic interaction between nitrogen availability and temperature responses is commonly observed in a myriad of ecosystems, ranging from lakes (Palacin-Lizarbe et al., 2018) and estuaries (Nowicki, 1994) to alpine meadows (Chen et al., 2017). In addition, earlier work of Palacin-Lizarbe et al. (2018)

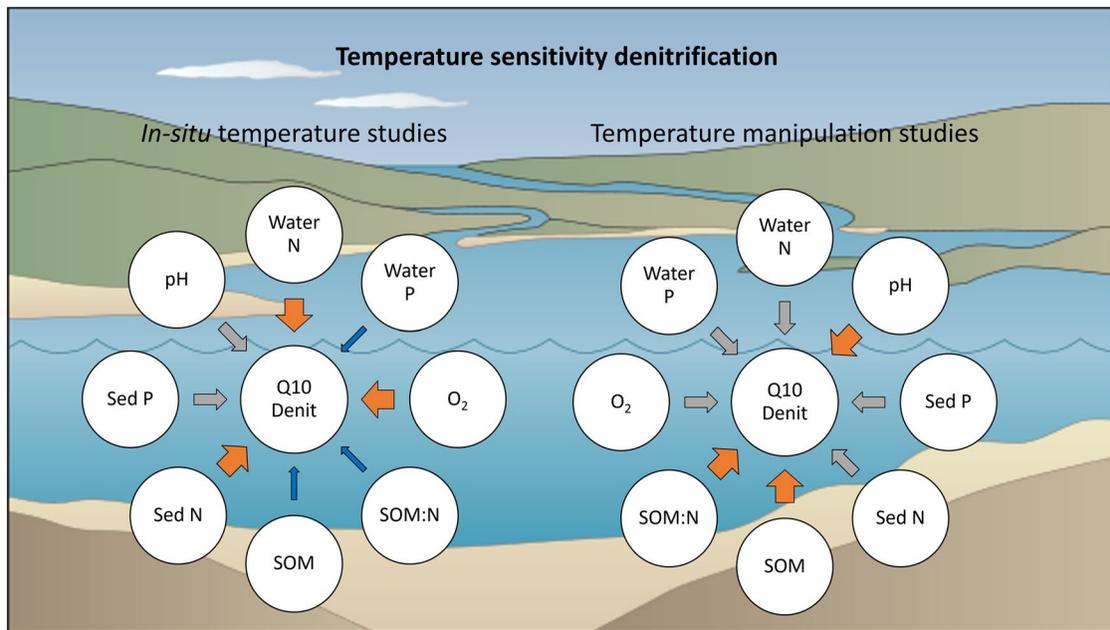


Figure 6. Conceptual overview depicting the relationships between $Q10_{denitrification}$ ($Q10_{denit}$) and the environmental variables included in this meta-analysis for both *in-situ* temperature and temperature manipulation datasets. The arrows show the directionality of the relationship, with blue arrows indicating a negative correlation, orange arrows a positive correlation and gray arrows indicate no observed correlation between the variables. Sed and water indicate the concentrations of the respective nutrients (N and P) in the sediment and water column, while sediment organic matter (SOM) indicates Soil Organic Matter.

has shown that denitrification activation energy can inversely correlate with nitrate concentrations, further supporting our meta-analytic results. The commonality between our results and other studies in freshwater and terrestrial ecosystems highlights the synergistic link between nitrogen as a driver of instantaneous denitrification rates and temperature as a regulator of the enzymatic process (Wallenstein et al., 2006).

In the *in-situ* denitrification dataset, this positive correlation with N conditions co-occurred with a negative correlation with sediment organic matter (SOM). SOM encompasses all organic material in the sediment and consists of a combination of recalcitrant and labile organic carbon, of variable nitrogen content (Schnitzer & Khan, 1975). We anticipated a positive correlation between SOM and temperature sensitivity as organic carbon is an electron donor in canonical denitrification and can thus limit denitrification rates. This positive correlation was observed in the temperature manipulation experiments, but we observed a negative correlation between SOM and $Q_{10_{\text{denitrification}}}$ in the *in-situ* dataset. This discrepancy highlights potential differences between conclusions drawn on laboratory experiments and *in-situ* measurements and show the difficulty in interpreting and upscaling experimental biogeochemical data to field scenarios. Possibly, competition between denitrification and other microbial processes might be more prominent under *in-situ* conditions. For instance, dissimilatory nitrate reduction to ammonium (DNRA) competes for organic carbon and nitrogen sources (Burgin & Hamilton, 2007). Following denitrification and DNRA stoichiometry, the ratio between SOM and N availability in the sediment can be a strong factor in determining the outcome of this competition, favouring DNRA at higher SOM:N ratios (Burgin & Hamilton, 2007; van de Leemput et al., 2011). In our work, we indeed observed that *in-situ* temperature sensitivity of denitrification was negatively correlated with sediment SOM:N ratios. Moreover, a study by Rahman, Roberts, et al. (2019) conducted in urban wetlands showed DNRA benefited more from the addition of organic carbon than denitrification did. In a follow-up study, these authors also showed that DNRA is more sensitive to temperature changes than denitrification (Rahman, Grace, et al., 2019). Therefore, while DNRA and denitrifiers can coexist under a variety of C:N ratios (van den Berg et al., 2016), the temperature response of denitrification may be limited by competition with DNRA at high SOM:N ratios.

In contrast to our hypotheses, we observed a positive correlation between temperature sensitivity of denitrification and O_2 concentration in this meta-analytic dataset. We expected that this temperature sensitivity would decrease with higher oxygen availability, as denitrification rates increase with temperature and hypoxia (de Klein et al., 2017; Veraart et al., 2011) and riverine work of Li and colleagues showed a reverse correlation of N_2O concentration with oxygen (Li et al., 2021). However, effects of O_2 on N_2O emission are inherently more difficult to predict, due to the contribution of both aerobic nitrification and anaerobic denitrification. For example, work of Liikanen et al. (2002), included in our dataset, showed higher N_2O production in oxic than anoxic conditions, hinting at a larger role for nitrification than denitrification in N_2O production in their eutrophic systems. At the same time, the enzyme responsible for the reduction of N_2O to N_2 (*NosZ*) is known to be sensitive to O_2 inhibition (Dalsgaard et al., 2014), which could indicate reduced transformation rates of N_2O to N_2 under high O_2 concentrations. The contrasting findings in studies included in our meta-analysis may be explained by site-specific characteristics affecting the balance between underlying processes driving both denitrification and N_2O emission. In addition, reported correlations between O_2 and denitrification *in-situ* also depend on where and how O_2 is monitored, and denitrification is measured. Reporting changes in O_2 profiles when measuring temperature effects on denitrification will provide better insight in their interactive dynamics.

4.2. Global Heterogeneity in Temperature Sensitivity of N_2O Emission Factors

Ecosystems near the poles, especially in the Arctic, are warming at least twice as fast as ecosystems at lower latitudes (polar amplification; Pachauri et al., 2014; Smith et al., 2019). Globally, nitrogen inputs in aquatic ecosystems are increasing simultaneously (Ackerman et al., 2019; Reay et al., 2008), and indications of nitrogen effects on the biogeochemistry of lakes have been documented as far north as 70° (Wolfe et al., 2006). In our meta-analysis, no latitudinal patterns in any of the temperature sensitivity values were observed. Nonetheless, the temperature sensitivity of N_2O emission factors (EF5) exhibited spatial heterogeneity. Specifically, *in-situ* $Q_{10_{\text{EF5}}}$ values were significantly higher in the region of Australia and New Zealand compared to other geographic regions. Though sample sizes were small, this may indicate that N_2O emission factors are sensitive to changes in surrounding temperatures and that this sensitivity differs spatially. IPCC methodology of EF5 values is aimed to determine an average N_2O emission factor per ecosystem. In our analysis, we showed that EF5 values can increase with a factor 1.8–6.9 with a 10° temperature increase. As the IPCC has reduced their EF5 estimates for rivers and

lakes between their 1997 and 2006 reports (IPCC, 1997, 2006; Tian et al., 2019), we would advise to take the temperature-dependence into account in future decision making as the world is continuing to warm.

We should note that most of the studies included in our meta-analysis were conducted in Europe, Northern America and Asia. A knowledge gap therefore exists for tropical ecosystems and ecosystems south of the equator in general, as only one study from South America (Veraart et al., 2012), one from Africa (Roland et al., 2017) and few from Australia and New Zealand were identified (see for instance Crawshaw et al. (2019), an overview is presented in Table S1 of Supporting Information S1). While additional datasets may have been published (see for instance Borges et al. (2015) for an overview on N₂O emissions from African rivers), these were not picked up by our systematic literature search. Studies on temperature sensitivity in some ecosystems may have been published in other languages than English. Because our literature search focused on English literature, these potential studies are currently not considered in our analysis. Taking non-English studies into account can alter meta-analytic outcomes significantly (see e.g., Konno et al. (2020)). This knowledge gap should be considered when interpreting our meta-analytic results.

4.3. Technical Recommendations—Upscaling, FAIR Data and New Scientific Discoveries

It is extremely difficult, or even impossible, to mimic the complexity of a real ecosystem in a laboratory setting. Experimental set-ups allow us to reduce environmental complexity while simultaneously increasing control. In laboratory settings, ecological interactions are greatly reduced relative to natural ecosystems, which can alter observed responses (Englund & Cooper, 2003; Goldenberg et al., 2018). This may explain the difference in temperature sensitivity between the datasets, as Q10 values were higher in temperature-manipulation experiments than in *in-situ* datasets. While experimental studies are extremely valuable and necessary to gain mechanistic understanding of the influence of environmental pressures, considerable care should be taken when extrapolating these findings to natural conditions. Another platform for the potential upscaling of denitrification studies is modeling studies. Nitrogen processing models such as Global NEWS (Mayorga et al., 2010) and nutrient loading models such as PCLake+ (Janssen et al., 2019) and VEMALA (Huttunen et al., 2016), often include denitrification as a nitrogen conversion term. Moreover, other ecosystem models such as the MyLake-Sediment model (Markelov et al., 2019) and the terrestrial NEMIS model (Hénault & Germon, 2000), even use (derivatives of) Q10-values to describe the temperature sensitivity of denitrification. Our meta-analytic results provide an overview of *in-situ* Q10-values for a wide range of ecosystems (Table S1 in Supporting Information S1), which can potentially aid the modeling of the temperature sensitivity of denitrification for specific freshwater ecosystems.

It is quite common in scientific publications to include only the measured variables that are valuable for the scientific question of the paper. In meta-analyses, however, auxiliary data, combined with meta-data, which are not crucial for the specific paper of interest become valuable to address new research questions. The publication of auxiliary data, either in Supporting Information S1 or in an online repository, has become more and more common in ecological research (Culina et al., 2020). In our opinion, denitrification research, which has been an active field of investigation for over a century, could rapidly move forward by FAIR (Findable, Accessible, Interoperable Reproducible) data practises (Alves et al., 2018; Wilkinson et al., 2016).

The field of environmental denitrification research is continuously advancing. For instance, scientific discoveries in the nitrogen cycle such as anammox (Strous et al., 1999), comammox (Daims et al., 2015; van Kessel et al., 2015) and non-denitrifying N₂O reduction (Jones et al., 2013; Sanford et al., 2012) can shed new light on previous scientific publications. A limitation of meta-analytic studies is the description of processes in the original paper, which may be partly outdated after these new scientific discoveries. In this study, we used a broad definition of denitrification (being the reduction of nitrate and nitrite to N₂O and/or N₂) and a range of methodologies. Overall, no strong differences in Q10-values were observed between methods (Table S2 in Supporting Information S1). However, in the *in-situ* denitrification dataset, acetylene inhibition studies reported higher temperature sensitivity than studies that used a ¹⁵N-tracer method. The ¹⁵N-tracer method cannot always distinguish between denitrification and coupled DNRA-anammox pathways and the balance of these pathways is a current hot topic in research. The contribution of anammox to N₂ production in lake sediments can be substantial (Crowe et al., 2017) and depends on environmental conditions such as oxygen and light (Valiente et al., 2022). Recent work by Tan et al. (2020) suggests that sediment denitrifiers are more thermotolerant than anammox bacteria, which may explain our observed lower temperature sensitivity in ¹⁵N-tracer studies. Overall, we cannot

guarantee that the ¹⁵N-tracer studies included in our meta-analysis did not include the DNRA-anammox pathway and think this is a valuable and interesting research avenue for further exploration.

5. Conclusion

Using a meta-analytic approach, we show that temperature sensitivity of denitrification is positively correlated with nitrogen concentrations and that this increase in temperature sensitivity is non-linear. In general, one degree temperature increase is expected to amplify denitrification and N₂O emissions between 8 and 14%. Both global temperatures and nitrogen deposition in freshwater ecosystems are expected to increase over the coming years (Brunner et al., 2020; Pachauri et al., 2014; Vuuren et al., 2011). Our analysis therefore predicts overall increased freshwater microbial denitrification rates in the near future, which may present a way to naturally counteract the harmful effects of eutrophication. However, temperature sensitivity of N₂O emission factors was observed as well, potentially indicating that warming and eutrophication may enhance future denitrification-derived N₂O emissions from freshwater ecosystems.

Data Availability Statement

The data and R scripts that support the findings of this study are publicly available after the 6th of August, 2022 at the DANS EASY archive (<https://easy.dans.knaw.nl>) via <https://doi.org/10.17026/dans-xy8-mydc> (dataset 246823) under the creative commons zero waver (CC0) license.

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References

- Ackerman, D., Millet, D. B., & Chen, X. (2019). Global estimates of inorganic nitrogen deposition across four decades. *Global Biogeochemical Cycles*, 33(1), 100–107. <https://doi.org/10.1029/2018gb005990>
- Allen, G. H., & Pavelsky, T. M. (2018). Global extent of rivers and streams. *Science*, 361(6402), 585–588. <https://doi.org/10.1126/science.aat0636>
- Alves, C., Castro, J. A., Ribeiro, C., Honrado, J. P., & Lomba, A. (2018). Research data management in the field of Ecology: An overview. In *International conference on dublin Core and metadata applications* (pp. 87–94).
- Birk, M. (2017). *Respirometry: Tools for conducting respirometry experiments*. R package. Version 0.4. 0.
- Blum, J.-M., Su, Q., Ma, Y., Valverde-Pérez, B., Domingo-Félez, C., Jensen, M. M., & Smets, B. F. (2018). The pH dependency of N-converting enzymatic processes, pathways and microbes: Effect on net N₂O production. *Environmental Microbiology*, 20(5), 1623–1640. <https://doi.org/10.1111/1462-2920.14063>
- Borges, A. V., Darchambeau, F., Teodoru, C. R., Marwick, T. R., Tamooh, F., Geeraert, N., et al. (2015). Globally significant greenhouse-gas emissions from African inland waters. *Nature Geoscience*, 8, 637–642. <https://doi.org/10.1038/ngeo2486>
- Brin, L. D., Giblin, A. E., & Rich, J. J. (2017). Similar temperature responses suggest future climate warming will not alter partitioning between denitrification and anammox in temperate marine sediments. *Global Change Biology*, 23(1), 331–340. <https://doi.org/10.1111/gcb.13370>
- Brunner, L., Pendergrass, A. G., Lehner, F., Merrifield, A. L., Lorenz, R., & Knutti, R. (2020). Reduced global warming from CMIP6 projections when weighting models by performance and independence. *Earth System Dynamics*, 11(4), 995–1012. <https://doi.org/10.5194/esd-11-995-2020>
- Burgin, A. J., & Hamilton, S. K. (2007). Have we overemphasized the role of denitrification in aquatic ecosystems? A review of nitrate removal pathways. *Frontiers in Ecology and the Environment*, 5(2), 89–96. [https://doi.org/10.1890/1540-9295\(2007\)5\[89:hwotro\]2.0.co;2](https://doi.org/10.1890/1540-9295(2007)5[89:hwotro]2.0.co;2)
- Chen, X. P., Wang, G. X., Huang, K. W., Hu, Z. Y., Song, C. L., Liang, Y. M., et al. (2017). The effect of nitrogen deposition rather than warming on carbon flux in alpine meadows depends on precipitation variations. *Ecological Engineering*, 107, 183–191. <https://doi.org/10.1016/j.ecoleng.2017.07.018>
- Clough, T. J., Buckthought, L. E., Kelliher, F. M., & Sherlock, R. R. (2007). Diurnal fluctuations of dissolved nitrous oxide (N₂O) concentrations and estimates of N₂O emissions from a spring-fed river: Implications for IPCC methodology. *Global Change Biology*, 13(5), 1016–1027. <https://doi.org/10.1111/j.1365-2486.2007.01337.x>
- Crawshaw, J. A., Schallenberg, M., & Savage, C. (2019). Physical and biological drivers of sediment oxygenation and denitrification in a New Zealand intermittently closed and open lake lagoon. *New Zealand Journal of Marine & Freshwater Research*, 53(1), 33–59. <https://doi.org/10.1080/00288330.2018.1476388>
- Crowe, S. A., Treusch, A. H., Forth, M., Li, J., Magen, C., Canfield, D. E., et al. (2017). Anammox bacteria and nitrogen loss from Lake Superior. *Scientific Reports*, 7(1), 13757. <https://doi.org/10.1038/s41598-017-12270-1>
- Culina, A., van den Berg, I., Evans, S., & Sánchez-Tójar, A. (2020). Low availability of code in ecology: A call for urgent action. *PLoS Biology*, 18(7), e3000763. <https://doi.org/10.1371/journal.pbio.3000763>
- Daims, H., Lebedeva, E. V., Pjevac, P., Han, P., Herbold, C., Albertsen, M., et al. (2015). Complete nitrification by Nitrospira bacteria. *Nature*, 528(7583), 504–509. <https://doi.org/10.1038/nature16461>
- Dalsgaard, T., Stewart, F. J., Thamdrup, B., De Brabandere, L., Revsbech, N. P., Ulloa, O., et al. (2014). Oxygen at nanomolar levels reversibly suppresses process rates and gene expression in anammox and denitrification in the oxygen minimum zone off northern Chile. *mBio*, 5(6). <https://doi.org/10.1128/mbio.01966-14>
- de Klein, J. J. M., Overbeek, C. C., Juncher Jørgensen, C., & Veraart, A. J. (2017). Effect of temperature on oxygen profiles and denitrification rates in freshwater sediments. *Wetlands*, 37(5), 975–983. <https://doi.org/10.1007/s13157-017-0933-1>
- Englund, G., & Cooper, S. D. (2003). Scale effects and extrapolation in ecological experiments. *Advances in Ecological Research*, 33, 161–213. [https://doi.org/10.1016/S0065-2504\(03\)33011-9](https://doi.org/10.1016/S0065-2504(03)33011-9)
- Galloway, J. N., Dentener, F. J., Capone, D. G., Boyer, E. W., Howarth, R. W., Seitzinger, S. P., et al. (2004). Nitrogen cycles: Past, present, and future. *Biogeochemistry*, 70(2), 153–226. <https://doi.org/10.1007/s10533-004-0370-0>

- Goldenberg, S. U., Nagelkerken, I., Marangon, E., Bonnet, A., Ferreira, C. M., & Connell, S. D. (2018). Ecological complexity buffers the impacts of future climate on marine consumers. *Nature Climate Change*, 8(3), 229–233. <https://doi.org/10.1038/s41558-018-0086-0>
- Hama-Aziz, Z. Q., Hiscock, K. M., & Cooper, R. J. (2017). Indirect nitrous oxide emission factors for agricultural field drains and headwater streams. *Environmental Science & Technology*, 51(1), 301–307. <https://doi.org/10.1021/acs.est.6b05094>
- Hénault, C., & Germon, J. C. (2000). NEMIS, a predictive model of denitrification on the field scale. *European Journal of Soil Science*, 51(2), 257–270. <https://doi.org/10.1046/j.1365-2389.2000.00314.x>
- Huttunen, I., Huttunen, M., Piirainen, V., Korppoo, M., Lepistö, A., Räike, A., et al. (2016). A national-scale nutrient loading model for Finnish watersheds—VEMALA. *Environmental Modeling & Assessment*, 21(1), 83–109. <https://doi.org/10.1007/s10666-015-9470-6>
- Huwaldt, J., & Stienhorst, S. (2013). *Plot digitizer*. Version 2.6.8.
- IPCC. (1997). Revised 1996 IPCC guidelines for national greenhouse gas inventories. *Reference manual, volume 3* (p. 4.105–4.108). Institute for Global Environmental Strategies, Kamiyamaguchi, Japan.
- IPCC. (2006). N₂O emissions from managed soils, and CO₂ emissions from lime and urea application. In: *IPCC guidelines for national greenhouse gas inventories, volume 4* (p. 11.1–11.54). Institute for Global Environmental Strategies, Hayama, Japan.
- Janssen, A. B., Teurlinx, S., Beusen, A. H., Huijbregts, M. A., Rost, J., Schipper, A. M., et al. (2019). PCLake+: A process-based ecological model to assess the trophic state of stratified and non-stratified freshwater lakes worldwide. *Ecological Modelling*, 396, 23–32. <https://doi.org/10.1016/j.ecolmodel.2019.01.006>
- Ji, Q., Buitenhuis, E., Suntharalingam, P., Sarmiento, J. L., & Ward, B. B. (2018). Global nitrous oxide production determined by oxygen sensitivity of nitrification and denitrification. *Global Biogeochemical Cycles*, 32(12), 1790–1802. <https://doi.org/10.1029/2018gb005887>
- Jones, C. M., Graf, D. R. H., Bru, D., Philippot, L., & Hallin, S. (2013). The unaccounted yet abundant nitrous oxide-reducing microbial community: A potential nitrous oxide sink. *The ISME Journal*, 7(2), 417–426. <https://doi.org/10.1038/ismej.2012.125>
- Jurado, A., Borges, A. V., & Brouyère, S. (2017). Dynamics and emissions of N₂O in groundwater: A review. *The Science of the Total Environment*, 584–585, 207–218. <https://doi.org/10.1016/j.scitotenv.2017.01.127>
- Klaus, M., Geibrink, E., Jonsson, A., Bergstrom, A. K., Bastviken, D., Laudon, H., et al. (2018). Greenhouse gas emissions from boreal inland waters unchanged after forest harvesting. *Biogeosciences*, 15(18), 5575–5594. <https://doi.org/10.5194/bg-15-5575-2018>
- Knowles, R. (1982). Denitrification. *Microbiological Reviews*, 46(1), 43–70. <https://doi.org/10.1128/mr.46.1.43-70.1982>
- Konno, K., Akasaka, M., Koshida, C., Katayama, N., Osada, N., Spake, R., & Amano, T. (2020). Ignoring non-English-language studies may bias ecological meta-analyses. *Ecology and Evolution*, 10(13), 6373–6384. <https://doi.org/10.1002/ece3.6368>
- Li, M., Peng, C., Zhang, K., Xu, L., Wang, J., Yang, Y., et al. (2021). Headwater stream ecosystem: An important source of greenhouse gases to the atmosphere. *Water Research*, 190, 116738. <https://doi.org/10.1016/j.watres.2020.116738>
- Liikanen, A., Murtoniemi, T., Tanskanen, H., Vaisanen, T., & Martikainen, P. J. (2002). Effects of temperature and oxygen availability on greenhouse gas and nutrient dynamics in sediment of a eutrophic mid-boreal lake. *Biogeochemistry*, 59(3), 269–286. <https://doi.org/10.1023/a:1016015526712>
- Maavara, T., Lauerwald, R., Laruelle, G. G., Akbarzadeh, Z., Bouskill, N. J., Van Cappellen, P., & Regnier, P. (2019). Nitrous oxide emissions from inland waters: Are IPCC estimates too high? *Global Change Biology*, 25(2), 473–488. <https://doi.org/10.1111/gcb.14504>
- Markelov, I., Couture, R. M., Fischer, R., Haande, S., & Van Cappellen, P. (2019). Coupling water column and sediment biogeochemical dynamics: Modeling internal phosphorus loading, climate change responses, and mitigation measures in Lake Vansjø, Norway. *Journal of Geophysical Research: Biogeosciences*, 124(12), 3847–3866. <https://doi.org/10.1029/2019jg005254>
- Mayorga, E., Seitzinger, S. P., Harrison, J. A., Dumont, E., Beusen, A. H. W., Bouwman, A. F., et al. (2010). Global nutrient Export from Watersheds 2 (NEWS 2): Model development and implementation. *Environmental Modelling & Software*, 25(7), 837–853. <https://doi.org/10.1016/j.envsoft.2010.01.007>
- Messer, J. J., & Brezonik, P. L. (1984). Laboratory evaluation of kinetic parameters for lake sediment denitrification models. *Ecological Modelling*, 21(4), 277–286. [https://doi.org/10.1016/0304-3800\(84\)90064-4](https://doi.org/10.1016/0304-3800(84)90064-4)
- Moss, B., Kosten, S., Meerhoff, M., Battarbee, R. W., Jeppesen, E., Mazzeo, N., et al. (2011). Allied attack: Climate change and eutrophication. *Inland Waters*, 1(2), 101–105. <https://doi.org/10.5268/iw-1.2.359>
- Neubauer, S. C., & Megonigal, J. P. (2015). Moving beyond global warming potentials to quantify the climatic role of ecosystems. *Ecosystems*, 18(6), 1000–1013. <https://doi.org/10.1007/s10021-015-9879-4>
- Nowicki, B. L. (1994). The effect of temperature, oxygen, salinity, and nutrient enrichment on estuarine denitrification rates measured with a modified nitrogen gas flux technique. *Estuarine, Coastal and Shelf Science*, 38(2), 137–156. <https://doi.org/10.1006/ecss.1994.1009>
- Pachauri, R. K., Allen, M. R., Barros, V. R., Broome, J., Cramer, W., Christ, R., et al. (2014). *Climate change 2014: Synthesis report. Contribution of working Groups I, II and III to the fifth assessment report of the intergovernmental panel on climate change*. IPCC.
- Palacin-Lizarbe, C., Camarero, L., & Catalan, J. (2018). Denitrification temperature dependence in remote, cold, and N-poor lake sediments. *Water Resources Research*, 54(2), 1161–1173. <https://doi.org/10.1002/2017wr021680>
- R Core team. (2013). *R: A language and environment for statistical computing*.
- Rahman, M., Grace, M. R., Roberts, K. L., Kessler, A. J., & Cook, P. L. M. (2019). Effect of temperature and drying-rewetting of sediments on the partitioning between denitrification and DNRA in constructed urban stormwater wetlands. *Ecological Engineering*, 140, 105586. <https://doi.org/10.1016/j.ecoleng.2019.105586>
- Rahman, M. M., Roberts, K. L., Grace, M. R., Kessler, A. J., & Cook, P. L. M. (2019). Role of organic carbon, nitrate and ferrous iron on the partitioning between denitrification and DNRA in constructed stormwater urban wetlands. *The Science of the Total Environment*, 666, 608–617. <https://doi.org/10.1016/j.scitotenv.2019.02.225>
- Ravishankara, A. R., Daniel, J. S., & Portmann, R. W. (2009). Nitrous oxide (N₂O): The dominant ozone-depleting substance emitted in the 21st century. *Science*, 326(5949), 123–125. <https://doi.org/10.1126/science.1176985>
- Reay, D. S., Dentener, F., Smith, P., Grace, J., & Feely, R. A. (2008). Global nitrogen deposition and carbon sinks. *Nature Geoscience*, 1(7), 430–437. <https://doi.org/10.1038/ngeo230>
- Rissanen, A. J., Tiirola, M., Hietanen, S., & Ojala, A. (2013). Interlake variation and environmental controls of denitrification across different geographical scales. *Aquatic Microbial Ecology*, 69, 1–16. <https://doi.org/10.3354/ame01619>
- Roland, F. A. E., Darchambeau, F., Morana, C., & Borges, A. V. (2017). Nitrous oxide and methane seasonal variability in the epilimnion of a large tropical meromictic lake (Lake Kivu, East-Africa). *Aquatic Sciences*, 79(2), 209–218. <https://doi.org/10.1007/s00027-016-0491-2>
- Sanford, R. A., Wagner, D. D., Wu, Q., Chee-Sanford, J. C., Thomas, S. H., Cruz-García, C., et al. (2012). Unexpected nondenitrifier nitrous oxide reductase gene diversity and abundance in soils. *Proceedings of the National Academy of Sciences*, 109(48), 19709–19714. <https://doi.org/10.1073/pnas.1211238109>
- Schnitzer, M., & Khan, S. U. (1975). *Soil organic matter*. Elsevier.

- Seitzinger, S., Harrison, J. A., Böhlke, J. K., Bouwman, A. F., Lowrance, R., Peterson, B., et al. (2006). Denitrification across landscapes and waterscapes: A synthesis. *Ecological Applications*, *16*(6), 2064–2090. [https://doi.org/10.1890/1051-0761\(2006\)016\[2064:dalawa\]2.0.co;2](https://doi.org/10.1890/1051-0761(2006)016[2064:dalawa]2.0.co;2)
- Seitzinger, S. P., & Kroeze, C. (1998). Global distribution of nitrous oxide production and N inputs in freshwater and coastal marine ecosystems. *Global Biogeochemical Cycles*, *12*(1), 93–113. <https://doi.org/10.1029/97gb03657>
- Seitzinger, S. P., Kroeze, C., & Styles, R. V. (2000). Global distribution of N₂O emissions from aquatic systems: Natural emissions and anthropogenic effects. *Chemosphere - Global Change Science*, *2*(3–4), 267–279. [https://doi.org/10.1016/s1465-9972\(00\)00015-5](https://doi.org/10.1016/s1465-9972(00)00015-5)
- Seitzinger, S. P., & Nixon, S. W. (1985). Eutrophication and the rate of denitrification and N₂O production in coastal marine sediments. *Limnology & Oceanography*, *30*(6), 1332–1339. <https://doi.org/10.4319/lo.1985.30.6.1332>
- Smil, V. (2004). *Enriching the earth: Fritz Haber, Carl Bosch, and the transformation of world food production*. MIT press.
- Smith, D. M., Screen, J. A., Deser, C., Cohen, J., Fyfe, J. C., García-Serrano, J., et al. (2019). The Polar Amplification Model Intercomparison Project (PAMIP) contribution to CMIP6: Investigating the causes and consequences of polar amplification. *Geoscientific Model Development*, *12*(3), 1139–1164. <https://doi.org/10.5194/gmd-12-1139-2019>
- Smith, V. H., Tilman, G. D., & Nekola, J. C. (1999). Eutrophication: Impacts of excess nutrient inputs on freshwater, marine, and terrestrial ecosystems. *Environmental Pollution*, *100*(1–3), 179–196. [https://doi.org/10.1016/s0269-7491\(99\)00091-3](https://doi.org/10.1016/s0269-7491(99)00091-3)
- Steffen, W., Richardson, K., Rockström, J., Cornell, S. E., Fetzer, I., Bennett, E. M., et al. (2015). Planetary boundaries: Guiding human development on a changing planet. *Science*, *347*(6223), 1259855. <https://doi.org/10.1126/science.1259855>
- Strous, M., Fuerst, J. A., Kramer, E. H. M., Logemann, S., Muyzer, G., van de Pas-Schoonen, K. T., et al. (1999). Missing lithotroph identified as new planctomycete. *Nature*, *400*(6743), 446–449. <https://doi.org/10.1038/22749>
- Tan, E., Zou, W., Zheng, Z., Yan, X., Du, M., Hsu, T.-C., et al. (2020). Warming stimulates sediment denitrification at the expense of anaerobic ammonium oxidation. *Nature Climate Change*, *10*(4), 349–355. <https://doi.org/10.1038/s41558-020-0723-2>
- Tian, L., Cai, Y., & Akiyama, H. (2019). A review of indirect N₂O emission factors from agricultural nitrogen leaching and runoff to update of the default IPCC values. *Environmental Pollution*, *245*, 300–306. <https://doi.org/10.1016/j.envpol.2018.11.016>
- Tomaszek, J. A., Gardner, W. S., & Johengen, T. H. (1997). Denitrification in sediments of a Lake Erie coastal wetland (Old Woman Creek, Huron, Ohio, USA). *Journal of Great Lakes Research*, *23*(4), 403–415. [https://doi.org/10.1016/s0380-1330\(97\)70922-5](https://doi.org/10.1016/s0380-1330(97)70922-5)
- Valiela, N., Jirsa, F., Hein, T., Wanek, W., Prommer, J., Bonin, P., & Gómez-Alday, J. J. (2022). The role of coupled DNRA-Anammox during nitrate removal in a highly saline lake. *The Science of the Total Environment*, *806*, 150726. <https://doi.org/10.1016/j.scitotenv.2021.150726>
- van de Leemput, I. A., Veraart, A. J., Dakos, V., de Klein, J. J. M., Strous, M., & Scheffer, M. (2011). Predicting microbial nitrogen pathways from basic principles. *Environmental Microbiology*, *13*(6), 1477–1487. <https://doi.org/10.1111/j.1462-2920.2011.02450.x>
- van den Berg, E. M., Boleij, M., Kuenen, J. G., Kleerebezem, R., & van Loosdrecht, M. C. M. (2016). DNRA and denitrification coexist over a broad range of acetate/N-NO₃⁻ ratios, in a chemostat enrichment culture. *Frontiers in Microbiology*, *7*, 1842. <https://doi.org/10.3389/fmicb.2016.01842>
- van Kessel, M. A. H. J., Speth, D. R., Albertsen, M., Nielsen, P. H., Op den Camp, H. J. M., Kartal, B., et al. (2015). Complete nitrification by a single microorganism. *Nature*, *528*(7583), 555–559. <https://doi.org/10.1038/nature16459>
- Veraart, A. J., de Klein, J. J. M., & Scheffer, M. (2011). Warming can boost denitrification disproportionately due to altered oxygen dynamics. *PLoS One*, *6*(3), e18508. <https://doi.org/10.1371/journal.pone.0018508>
- Veraart, A. J., Kosten, S., Lacerot, G., & de Klein, J. J. M. (2012). Influence of warming and macrophyte presence on denitrification in temperate and subtropical shallow lakes. *Denitrification in ditches, streams and shallow lakes*. PhD thesis. Wageningen University.
- Verpoorter, C., Kutser, T., Seekell, D. A., & Tranvik, L. J. (2014). A global inventory of lakes based on high-resolution satellite imagery. *Geophysical Research Letters*, *41*(18), 6396–6402. <https://doi.org/10.1002/2014gl060641>
- Vuuren, D. P. v., Bouwman, L. F., Smith, S. J., & Dentener, F. (2011). Global projections for anthropogenic reactive nitrogen emissions to the atmosphere: An assessment of scenarios in the scientific literature. *Current Opinion in Environmental Sustainability*, *3*(5), 359–369. <https://doi.org/10.1016/j.cosust.2011.08.014>
- Wallenstein, M. D., Myrold, D. D., Firestone, M., & Voytek, M. (2006). Environmental controls on denitrifying communities and denitrification rates: Insights from molecular methods. *Ecological Applications*, *16*(6), 2143–2152. [https://doi.org/10.1890/1051-0761\(2006\)016\[2143:ecodca\]2.0.co;2](https://doi.org/10.1890/1051-0761(2006)016[2143:ecodca]2.0.co;2)
- Well, R., Weymann, D., & Flessa, H. (2005). Recent research progress on the significance of aquatic systems for indirect agricultural N₂O emissions. *Environmental Sciences*, *2*(2–3), 143–151. <https://doi.org/10.1080/15693430500393334>
- Wilkinson, M. D., Dumontier, M., Aalbersberg, I. J., Appleton, G., Axton, M., Baak, A., et al. (2016). The FAIR guiding principles for scientific data management and stewardship. *Scientific Data*, *3*(1), 160018. <https://doi.org/10.1038/sdata.2016.18>
- Wolfe, A. P., Cooke, C. A., & Hobbs, W. O. (2006). Are current rates of atmospheric nitrogen deposition influencing lakes in the eastern Canadian Arctic? *Arctic Antarctic and Alpine Research*, *38*(3), 465–476. [https://doi.org/10.1657/1523-0430\(2006\)38\[465:acroan\]2.0.co;2](https://doi.org/10.1657/1523-0430(2006)38[465:acroan]2.0.co;2)
- Yang, P., Huang, J., Tan, L., Tong, C., Jin, B., Hu, B., et al. (2021). Large variations in indirect N₂O emission factors (EF₅) from coastal aquaculture systems in China from plot to regional scales. *Water Research*, *200*, 117208. <https://doi.org/10.1016/j.watres.2021.117208>