BIOLOGICAL REVIEWS
Cambridge Philosophical Society

Biol. Rev. (2021), **96**, pp. 692–715.
doi: 10.1111/brv.12673

Experimental nitrogen and phosphorus enrichment stimulates multiple trophic levels of algal and detrital-based food webs: a global meta-analysis from streams and rivers

Marcelo Ardón[1]*, Lydia H. Zeglin[2], Ryan M. Utz[3], Scott D. Cooper[4], Walter K. Dodds[2], Rebecca J. Bixby[5], Ayesha S. Burdett[6], Jennifer Follstad Shah[7], Natalie A. Griffiths[8], Tamara K. Harms[9], Sherri L. Johnson[10], Jeremy B. Jones[9], John S. Kominoski[11], William H. McDowell[12], Amy D. Rosemond[13], Matt T. Trentman[14], David Van Horn[5] and Amelia Ward[15]

[1]*Department of Forestry and Environmental Resources, North Carolina State University, Raleigh, NC, 27695, U.S.A.*
[2]*Division of Biology, Kansas State University, Manhattan, KS, 66506, U.S.A.*
[3]*Falk School of Sustainability, Chatham University, Pittsburgh, PA, 15232, U.S.A.*
[4]*Department of Ecology, Evolution, and Marine Biology and Marine Science Institute, University of California – Santa Barbara, Santa Barbara, CA, 93106, U.S.A.*
[5]*Department of Biology, University of New Mexico, Albuquerque, NM, 87131, U.S.A.*
[6]*River Bend Ecology, Wickliffe, VIC, 3379, Australia*
[7]*Environmental and Sustainability Studies Program/Department of Geography, University of Utah, Salt Lake City, UT, 84112, U.S.A.*
[8]*Climate Change Science Institute and Environmental Sciences Division, Oak Ridge National Laboratory, Oak Ridge, TN, 37831, U.S.A.*
[9]*Institute of Arctic Biology and Department of Biology and Wildlife, University of Alaska Fairbanks, Fairbanks, AK, 99775, U.S.A.*
[10]*Pacific Northwest Research Station, U. S. Forest Service, Corvallis, OR, 97731, U.S.A.*
[11]*Department of Biological Sciences and Southeast Environmental Research Center, Florida International University, Miami, FL, 33199, U.S.A.*
[12]*Department of Natural Resources and the Environment, University of New Hampshire, Durham, NH, 03824, U.S.A.*
[13]*Odum School of Ecology, University of Georgia, Athens, GA, 30602, U.S.A.*
[14]*Division of Biological Sciences, University of Notre Dame, Notre Dame, IN, 46556, U.S.A.*
[15]*Department of Biological Sciences, University of Alabama, Tuscaloosa, AL, 35487, U.S.A.*

ABSTRACT

Anthropogenic increases in nitrogen (N) and phosphorus (P) concentrations can strongly influence the structure and function of ecosystems. Even though lotic ecosystems receive cumulative inputs of nutrients applied to and deposited on land, no comprehensive assessment has quantified nutrient-enrichment effects within streams and rivers. We conducted a meta-analysis of published studies that experimentally increased concentrations of N and/or P in streams and rivers to examine how enrichment alters ecosystem structure (state: primary producer and consumer biomass and abundance) and function (rate: primary production, leaf breakdown rates, metabolism) at multiple trophic levels (primary producer, microbial heterotroph, primary and secondary consumers, and integrated ecosystem). Our synthesis included 184 studies, 885 experiments, and 3497 biotic responses to nutrient enrichment. We documented widespread increases in organismal biomass and abundance (mean response = +48%) and rates of ecosystem processes (+54%) to enrichment across multiple trophic levels, with no large differences in responses among trophic levels or between autotrophic or heterotrophic food-web pathways. Responses to nutrient enrichment varied with the nutrient added (N, P, or both) depending on rate

---

[*] Address for correspondence (Tel: +001 919 515 5574; E-mail: mlardons@ncsu.edu)

versus state variable and experiment type, and were greater in flume and whole-stream experiments than in experiments using nutrient-diffusing substrata. Generally, nutrient-enrichment effects also increased with water temperature and light, and decreased under elevated ambient concentrations of inorganic N and/or P. Overall, increased concentrations of N and/or P altered multiple food-web pathways and trophic levels in lotic ecosystems. Our results indicate that preservation or restoration of biodiversity and ecosystem functions of streams and rivers requires management of nutrient inputs and consideration of multiple trophic pathways.

Key words: lotic, eutrophication, nutrient criteria, primary and secondary production, ecosystem metabolism, decomposition

CONTENTS

I. INTRODUCTION

Nutrient concentrations are increasing in streams globally because of intensifying agricultural development, urbanization, and atmospheric deposition from fossil fuel combustion (Howarth *et al.*, 1996; Vitousek *et al.*, 1997; Grimm *et al.*, 2005). Nutrients (primarily nitrogen and phosphorous, hereafter N and P) often limit biological activity in stream and river ecosystems, so elevated concentrations of N or P can stimulate the productivity of primary producers (e.g. algae, cyanobacteria, vascular plants) and heterotrophic microorganisms (e.g. bacteria, fungi) that use terrestrially derived organic matter (detritus) as a carbon source (Elwood, Newbold & Stark, 1981; Grimm & Fisher, 1986; Elser *et al.*, 1996). However, few studies have considered multi-trophic level responses to nutrient enrichment in streams, limiting our knowledge of the effects of nutrient enrichment on the structure and function of lotic ecosystems (Palmer & Febria, 2012). Such considerations have significance for environmental management because the uptake and retention of nutrients in streams can ameliorate excessive nutrient loading to downstream ecosystems, yet excessive nutrient enrichment can overwhelm capacity for retention, exacerbate conservation and management challenges, and cause economic harm (Dodds *et al.*, 2009). Well-known consequences of riverine eutrophication include algal blooms that can be toxic to many organisms or hypoxia that leads to fish kills (Paerl *et al.*, 2004; Diaz & Rosenberg, 2008). Benthic toxic cyanobacteria can occur in streams and can be transported to downstream receiving waters (Fetscher *et al.*, 2015). Extensive research has found that nutrient enrichment leads to regime shifts in lakes, pushing clear macrophyte-dominated lakes to a turbid algae-dominated state (Scheffer *et al.*, 2001), but the widespread effects of N and P enrichment on whole-stream ecosystems have been less well recognized.

Previous meta-analyses of nutrient-enrichment experiments in rivers and streams focused on responses of algae (Elser, Marzolf & Goldman, 1990; Francoeur, 2001; Hillebrand, 2002; Keck & Lepori, 2012), and thus uncertainty remains regarding responses of other compartments and of whole ecosystems. Algal abundance and productivity typically increase in response to nutrient enrichment, although most meta-analyses have focused on small spatial scales studied using nutrient-diffusing substrata (NDS; Elser *et al.*, 1990, 2007; Francoeur, 2001; Hillebrand, 2002; Capps *et al.*, 2011; Keck & Lepori, 2012; Beck, Rugenski & Poff, 2017). Fewer efforts have focused on heterotrophic pathways, although one meta-analysis showed nutrient stimulation of leaf litter decomposition (Ferreira *et al.*, 2015). Despite broad trends towards increases in algal biomass and leaf breakdown rates with nutrient enrichment, a variety of environmental factors influence the magnitude of ecological responses (Francoeur, 2001; Ferreira *et al.*, 2015; Beck *et al.*, 2017). Responses to elevated nutrient concentrations may be muted if ambient nutrient concentrations are already high or if biotic activity is primarily limited by factors other than inorganic N or P concentrations [e.g. light, temperature (Rosemond, 1993; Dodds *et al.*, 2002; Tank & Dodds, 2003; Dodds, 2006; Eriksson, Rubach & Hillebrand, 2006; Elser *et al.*, 2007; Beck *et al.*, 2017)]. Geomorphic factors can introduce further among-site variation in biotic responses to enrichment, such as by reducing the bioavailability of added nutrients (Small *et al.*, 2016). Finally, variation in experimental approaches contributed to heterogeneity in enrichment responses in previous meta-analyses (Elser *et al.*, 1990; Ferreira *et al.*, 2015).

While much of the focus on excess nutrients has been on algal responses, reviews have shown that most river systems are net heterotrophic (Marcarelli *et al.*, 2011; Hall *et al.*, 2016), and thus understanding the relative responses of both heterotrophic and autotrophic metabolic pathways to nutrient enrichment is essential to characterize the consequences of eutrophication for whole river networks. The nature and magnitude of enrichment effects on autotrophic and heterotrophic organisms may differ due to the relative plasticity in elemental composition of each group (Sterner & Elser, 2002). Algae engage in luxury uptake of nutrients available in excess of demand, storing them within cells (Sterner & Elser, 2002). Accordingly, N:P ratios of autotrophic biofilms vary with nutrient availability (Stelzer & Lamberti, 2001). Nutrient enrichment to algal-dominated streams thus could lead to rapid changes in algal nutrient content, with slower changes in biomass or production rates (Rier *et al.*, 2016). Heterotrophic microbes on the other hand, tend to be more homeostatic than algae, maintaining elemental composition of biomass despite changes in nutrient availability (Persson *et al.*, 2010). However, some studies have observed elemental plasticity of aquatic bacteria (Chrzanowski & Kyle, 1996) and fungi (Gulis *et al.*, 2017). If heterotrophic microbes are more homeostatic than autotrophs, we would expect increased availability of limiting nutrients to stimulate biomass and activity to a greater extent in heterotrophic microbes.

Responses of upper trophic levels to nutrient enrichment may contrast with those of primary producers and decomposers. Inefficient energy transfer could dampen organismal production or biomass responses at higher trophic levels (Lindeman, 1942; Abrams, 1993). Nutrient enrichment can also cause shifts in algal resources to inedible or less-nutritional forms, with repercussions for upper trophic levels (Roll, Diehl & Cooper, 2005). Additionally, top-down effects may constrain the effects of nutrient enrichment (Davis *et al.*, 2010; Benstead *et al.*, 2014). For example, grazing insects suppressed a 'bottom-up' response of algae after 2 years of enrichment with P, the nutrient limiting primary production, in an Alaskan river (Peterson *et al.*, 1993). Individual studies suggest strong potential for interactive effects of nutrient enrichment and trophic dynamics to shape responses of upper trophic levels to eutrophication. Cross-site synthesis is needed to provide insight into the potential consequences of eutrophication for ecosystem services, such as fisheries, provided by upper trophic levels.

We conducted a global meta-analysis of responses of the biota of rivers and streams to experimental nutrient enrichment. Our objectives were to examine how: (*i*) experimental design (i.e. type of study: NDS, bottle, flume, whole stream), added nutrient (N, P, or both) and study duration modified the effects of nutrient enrichment on rate (productivity, leaf litter breakdown rates, metabolism) and state (biomass, abundance) variables; (*ii*) responses to experimental nutrient enrichment varied across trophic levels in detrital and algal food-web pathways; and (*iii*) abiotic conditions (light availability, ambient and experimental nutrient concentrations, temperature) modulated responses to nutrient enrichment. We hypothesized that: (*i*) experiment type, nutrient added, and study duration modulate responses, with larger-scale and longer-term studies, and studies that added both N and P, showing the strongest responses given that larger areas and longer timescales provide more space and time for biotic responses, and dual nutrient limitation is widespread (Elser *et al.*, 2007). (*ii*) The magnitude of biological responses to enrichment decreases with increasing trophic level due to the loss of energy and biomass with successive trophic transfers. (*iii*) Responses of autotrophic food-web components will be weaker than heterotrophic microbes, given that algae can carry out luxury uptake of nutrients and tend to be less homeostatic than microbial heterotrophs. (*iv*) Abiotic attributes of individual streams (light, nutrients, temperature) influence the magnitude of the responses due to the limitation of biotic responses by multiple factors in addition to nutrients.

II. METHODS

(1) Data harvest and database construction

We surveyed the primary literature for studies on the effects of experimentally added nutrients on stream or river biota. Results of a *Web of Science* database search using the criteria "TS = ((nutrient OR eutroph*) AND (lotic OR stream OR river)) AND TS = experiment*" were filtered to find studies that fitted the meta-analysis criteria, with the resulting list being supplemented by references in selected papers based

Table 1. Description of data types included as response variables, categorized by trophic level and rate or state variables

Trophic level	Rate variables	State variables
Primary producers (algae)	Algal cell growth Algal cell production Primary (O_2) production	Algal biovolume, abundance Chlorophyll-a concentration
Heterotrophic microbes (bacteria and fungi)	Fungal production Bacterial production	Fungal or bacterial biomass Fungal or bacterial abundance
Primary consumers (collector/gatherers, grazers, shredders)	Consumer growth Consumer production	Consumer biomass Consumer abundance
Secondary + tertiary consumers (invertebrate predators, salamanders, fish)	Consumer growth Consumer production	Consumer biomass Consumer abundance
Integrated ecosystem (multiple trophic levels included microbial + autotrophic respiration; leaf decomposition)	Leaf decomposition rate Community respiration (substrate-specific) Whole-stream respiration	Not applicable

on our collective knowledge (see online Supporting Information, Appendix S1, for a list of primary studies used in our meta-analysis). Criteria for inclusion of a study in the meta-analysis were: (*i*) paired control and experimental nutrient-addition treatments (i.e. land-use gradients, wastewater treatment plant inputs, or natural fertilization events were not included); (*ii*) N or P fertilization experiments only (no carbon or micronutrient enrichments were included); and (*iii*) experimental setting in a stream or in outdoor mesocosms containing lotic biota [e.g. whole-stream or artificial channel (flowing or recirculating, referred to as flume) experimental nutrient additions, nutrient-diffusing substrata (NDS), or short-term, non-flow (bottle) enrichments].

Database assembly was completed in 2018 and included studies published from 1980 to 2018. The final database included 184 published papers, 885 experiments conducted throughout the world, and 3497 dependent-variable responses (Fig. 1).

We harvested data from each study for quantitative meta-analyses that addressed each of our predictions. First, we recorded the mean control and response values for each response variable (Table 1), and the associated standard deviations (*s*) and number of replicates (*n*), when reported. We used this information to calculate the ln response ratio (LRR) and variance (V) for each experimental response, allowing statistical evaluation of the magnitude of responses

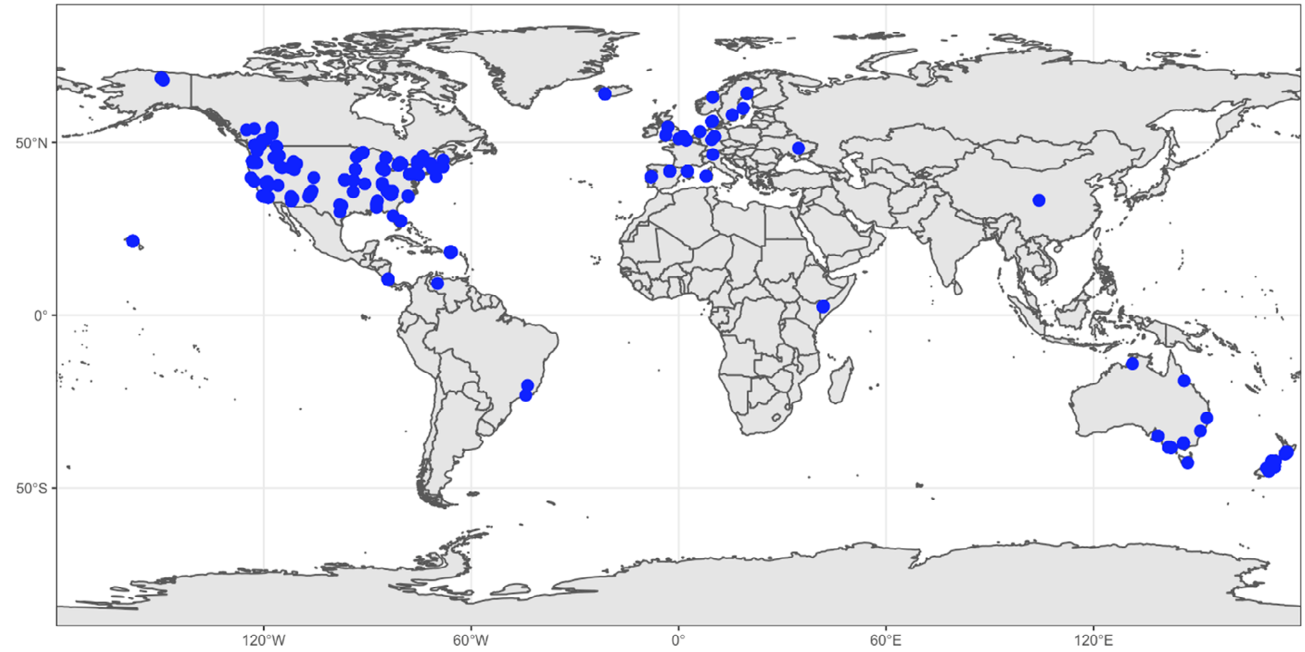

Fig 1. Global map with blue circles showing 885 locations of studies included in the meta-analysis.

across studies, trophic levels, and food-web pathways (Table 1). LRR and V were calculated for each pair as:

$$LRR = \ln\left(\frac{Y_1}{Y_2}\right) \tag{1}$$

$$V = \frac{s_1^2}{n_1 Y_1^2} + \frac{s_2^2}{n_2 Y_2^2} \tag{2}$$

Where Y_1 is the mean of the response to treatment and Y_2 is the mean of the control in a given experiment. Zero values for either control or response variables meant that LRR could not be calculated for 47 of the control/response pairs, leaving 3450 cases in the analysis; s and n were only available for 2613 of the control/response pairs (i.e. V was calculated for 76% of all LRR values).

We collected supporting information for each experiment when available, including experimental nutrient concentrations (soluble reactive phosphorus [SRP], total phosphorus [TP], ammonium [NH_4^+-N], nitrate [NO_3^--N], dissolved inorganic nitrogen [DIN], and total nitrogen [TN]), experimental area and duration, location, and environmental variables including temperature, photosynthetically active radiation (PAR), pH, ambient dissolved organic carbon (DOC), and nutrient concentrations. The coverage of supporting information differed for potential drivers, ranging from (as selected examples): 99% for experimental duration, 75% for both background NO_3^--N and SRP concentrations, 71% for water temperature, 40% for PAR, and 33% and 29% for experimental TN and TP concentrations, respectively. Experimental concentrations and addition rates are not easily estimated, and thus are not reported for NDS experiments.

We constructed statistical models to evaluate our hypotheses that experimental or stream characteristics could explain large amounts of the variation in the compiled LRR data. All data were extracted from published tables or figures, using digitization software when necessary. The final meta-analysis database is accessible on the Environmental Data Initiative (EDI) Data Portal (https://portal.edirepository.org/nis/home.jsp, DOI:10.6073/pasta/b674589d1a67589adadcb7762d928bba).

We used canopy cover data to evaluate our hypothesis regarding the influence of light on enrichment responses. Because PAR data were available for only a minority of experiments, we created a 'canopy cover' categorical variable to provide a qualitative evaluation of the influence of light on enrichment responses. Studies that reported <75% canopy cover were categorized as having an 'open' canopy, studies that reported ≥75% canopy cover were categorized as having a 'closed' canopy, and studies that did not report canopy cover were not categorized. This resulted in descriptions of canopy cover for 79% of studies.

To contrast the effects of nutrient enrichment on algal (autotrophic) *versus* detrital (heterotrophic)-based food webs, we created an 'autotrophic *versus* heterotrophic' response category (coded G for green or autotrophic, B for brown or heterotrophic). All algal response variables, and bacterial and consumer responses within algal-based food webs (i.e. periphyton-based or water column responses) were coded G; all fungal response variables, and bacterial and consumer responses within detrital-based food webs [i.e. responses of biota on particulate organic matter (POM) substrata] were coded B. G or B categories were designated for 96% of responses.

(2) Bias evaluation and univariate general linear models

Before addressing our hypotheses, we assessed the relationships between experimental scale (area) and duration (days) on biological responses to N and P enrichment (Osenberg *et al.*, 1999). Because nutrient enrichment should increase both growth rates and carrying capacities for stream biota populations relative to unenriched control populations, the LRR of organismal abundance or biomass will increase over time in short-duration experiments, before plateauing, while the LRR of time-specific response metrics may decrease over the duration of a longer experiment (Downing, Osenberg & Sarnelle, 1999). The scale of experimental measurement can also affect the sensitivity of measured responses, due to both detectability of differently sized organisms and to constraints on the influence of larger-scale interactions and feedbacks on the experiment's outcome (Englund, Cooper & Sarnelle, 2001). In addition to checking for systematic effects of experimental duration or size on LRR in our database, we binned all hypotheses-testing analysis in two ways to help minimize any bias in results due to experiment type. First, we separated NDS, bottle, flume, and whole-stream experiments (across the 885 experiments, mean ± SE areal extents were 0.01 ± 0.001, 0.03 ± 0.007, 1.5 ± 0.2, and 489 ± 241 m^2, respectively). In addition, we analysed rate and state variables separately for all statistical analyses, because of the expected differences in the response dynamics of state (e.g. biomass or density) and rate (e.g. production, growth, or mass loss) metrics to nutrient addition.

As detailed above, in addition to experiment type, we predicted that trophic level and food-web pathway, ambient temperature, light, and N, P, and carbon (DOC) concentrations drive or influence stream biotic responses to nutrient enrichment. We used general linear models (GLMs) to evaluate the statistical relationships of categorical and continuous predictor variables with the nutrient-enrichment effect size (LRR) across studies. Because non-NDS experiments reported the amount of limiting nutrient added, we also assessed the effects of experimental nutrient-amendment levels on LRR in these experiments (this cannot be quantified in NDS experiments due to differences in release rates). Thus, we assembled a series of GLMs, using data possibly predictive of rate and state enrichment responses in NDS and non-NDS experiments. If we identified a categorical variable as a predictor of LRR, a one-way analysis of variance using all data (*lm* and *aov* functions) with Tukey *post-hoc* tests (*glht* function in package *multcomp*) was used to identify differences among groups.

All primary GLM analyses were conducted using the *metafor* package in R (Viechtbauer, 2010). These models require an estimate of uncertainty for each data point, thus the 76% of all responses that included estimates of variance were included in these analyses. We ran all tests using random-effects models to account for the expected variability across individual studies that differed in methods or experimental designs, where among-study heterogeneity was modelled as a random effect. Latitude was evaluated as an absolute value, and PAR and all nutrient concentration data, including stream background and experimental and proportional increases, were $\log_{10}$ transformed before analysis to approximate parametric assumptions. All GLMs incorporated the mean and variance of individual LRR values, were calculated using the restricted maximum-likelihood estimator of heterogeneity, and applied the recommended adjustments to account for uncertainty in variance within studies (Knapp & Hartung, 2003). The effect of each categorical or continuous independent factor on LRR values was evaluated separately, owing to many missing variable values, using general linear models, each with a null hypothesis of no effect.

(3) Multivariate boosted regression tree models

We also used a complementary statistical approach to evaluate the relative influence of different environmental or experimental factors on responses against a background of natural environmental variation. We used boosted regression tree (BRT) modelling (De'ath, 2007; Elith, Leathwick & Hastie, 2008) to identify the environmental variables that most influenced LRR values, complementing the GLM models. BRTs combine a classification or regression decision-tree framework with boosting, an additive optimization procedure. Boosting in BRTs involves randomly selecting subsets of training data, then developing multiple decision trees that are sequentially combined to improve total model predictive performance over many (usually $\geq$1000) iterations until a stopping criterion, typically the minimization of prediction error, is met. We withheld 20% of the data at each iteration, then tested each model's performance. Early models were differentially weighted by their fit to test data in subsequent models to improve performance. The final product of a BRT analysis represents an ensemble of all iteratively adapted models with an optimized predictive performance. The BRT has multiple advantages, including its robustness to missing data, severe outliers, and irrelevant predictor variables, which are all useful attributes when modelling heterogeneous data compiled for meta-analysis (Leathwick *et al.*, 2006; Aertsen *et al.*, 2012).

We developed four separate BRT models, one each for rate and state variable in NDS and non-NDS experiments, to predict the combined influences of multiple independent variables on stream biotic responses to nutrient addition. To reduce model complexity and overfitting, redundant variables and variables with low data coverage were not included in BRT models. If two variables were correlated with *r* values exceeding 0.8, the predictor with the larger sample size was retained and the other (the redundant variable) omitted from subsequent analyses. The predictor terms in BRT analyses initially included the trophic level of the dependent variable (Table 1), canopy cover (open or closed), the nutrient added (N, P or both), trophic base of the system (auto- or heterotrophic), PAR, ambient pH, ambient DOC, NO_3^--N, and SRP concentrations, N:P molar ratios (DIN:SRP ratios), latitude, and water temperature. Continuous predictor variables were $\log_{10}$ transformed if distributions were heavily right-skewed (NO_3^--N, PAR, and SRP). Non-NDS BRT analyses also initially included experimental DIN and SRP concentrations and experiment type (bottle, flume, or whole-stream) as potential predictors.

Each BRT model was parameterized to optimize predictive performance as assessed by comparing observed and expected LRR values using linear regression analysis after at least 1000 trees were combined (Elith *et al.*, 2008). A Gaussian error distribution was assumed for the LRR values. Bag fraction, the proportion of model training data selected at each step, was set to 0.5 for all models. The learning rate (contribution of each tree to the ensemble model) and complexity (number of nodes in trees, which reflects interaction order) parameters were adjusted to minimize the deviance of ensemble model predictions from the data. Disparate sample sizes among models resulted in slightly different optimal learning rates and complexities: for NDS models, a learning rate of 0.001 and complexity of 3 were implemented, whereas for non-NDS models the respective parameters were 0.005 and 2.

We interpreted the relative influence of each independent variable on LRR values (expressed as percentage contribution to optimal model fit) and present partial dependency plots, which illustrate the effect of each independent variable on LRR after accounting for the average effect of all other model terms. We defined a threshold for the 'most influential' model terms as 100% divided by the total number of terms included (Müller, Leitão & Sikor, 2013). Although partial dependency plots can be difficult to interpret when independent variables interact or are correlated, these plots offer a clear way for evaluating model behaviour (Elith *et al.*, 2008). In addition, we estimated the least-squares regression slope of partial dependency change along gradients of continuous variables to summarize the magnitude and direction of the effect of each environmental variable on LRR values. We performed all BRT modelling operations with the *gbm* package for R (R Core Team, 2016; Elith & Leathwick, 2017).

III. RESULTS

(1) Overall effects of nutrients on biotic responses

The mean LRR for nutrient addition was 0.40 (median = 0.23, range = −5.25–6.91) across all 3450 experimental responses and, including only values with associated variances ($\mathcal{N}$ = 2613), was greater than zero (LRR = 0.36, $\mathcal{Z}$ = 22.3, $P < 0.0001$). Nutrient additions therefore stimulated the growth, activity, biomass, and abundance of all stream biota by an average of 49%. When all experiments were combined, LRR for rate (mean, median = 0.43, 0.27; mean increase = 54%) and state (mean, median = 0.39, 0.22; mean increase = 48%) variables were similar ($F = 2.85$, $P = 0.091$). When NDS and non-NDS experiments were treated separately, the rate LRRs (mean 0.18 = 20% increase) were slightly lower than state LRRs (mean 0.30 = 35% increase) in NDS experiments only ($P = 0.066$; Tables 2 and 3).

(2) Effects of experiment duration, type, and nutrient added

There was no relationship between rate LRR values and experiment duration, even when multi-year experiments (>365 days) were omitted from the analysis (overall rate LRR with and without multi-year studies: $F = 0.007$ and 0.005, $P = 0.93$ and 0.98, respectively). Inclusion of multi-year studies in the state LRR analysis did indicate a positive relationship between days of experiment duration and LRR (slope = 0.0002, $F = 11.1$, $P = 0.0009$); without multi-year studies, however, there was no relationship ($F = 1.42$, $P = 0.23$), suggesting that this relationship was driven by long-term studies rather than by short-term biomass accrual responses. Therefore, experiment duration was not considered as an independent variable in subsequent analyses.

On the other hand, while experimental area did not systematically affect LRR for either rate ($F = 0.70$, $P = 0.40$) or state ($F = 1.6$, $P = 0.21$) variables in non-NDS experiments, there was a clear difference in results among experiment types (rate $F = 13.0$, $P < 0.0001$, state $F = 21.5$, $P < 0.0001$, Fig. 2). Specifically, NDS experiments, which treat a small area, produced lower rate and state LRRs than flume or whole-stream experiments (Fig. 2C, D), particularly where P was added (Fig. 2E, F). NDS rate and state LRR had a mean of 0.18 and 0.30 (+20 and +35%), compared to a mean of 0.77 and 0.56 (+116 and +75%) for non-NDS rate

Table 2. Summary of weighted general linear model (GLM) and boosted regression tree (BRT) results for each independent variable predicted to affect ln response ratio (LRR) values from nutrient-diffusing substrata (NDS) experiments ($N = 2247$ total; 1811 with variance V). For each variable, the GLM test statistic [F: (H_0: mean LRR = 0 or H_0: mean LRRs are equal across different categories of class variables); or Z: (H_0: the standardized regression coefficient for the relationship between LRR and the independent variable = 0)], slope of relationship (only for the most important continuous variables), r^2 (expressed as %) and P value are shown. For rate *versus* state comparisons at the top of the table, mean LRR (1 SE) are reported. For BRT models, relative importance (%RI) values are listed, and redundant or low-N factors were not included (nd). Most important r^2 ($P < 0.05$) and %RI values [>100/N; after Müller *et al.* (2013)] are identified with an asterisk (*). Variables with predictive influence on LRR in both GLM and BRT models are highlighted in bold type

$F = 3.37$, $P = 0.066$
Rate LRR: $N = 725$, mean (median) = 0.183 (0.071); $N = 542$ with V, $Z = 6.25$, $P < 0.0001$*
State LRR, $N = 1522$, mean (median) = 0.302 (0.160) $N = 1271$ with V, $Z = 12.25$, $P < 0.0001$*

	Rate variables						State variables					
Rate *vs.* state	N	F or Z	P	Slope	r^2 (%)	%RI	N	F or Z	P	Slope	r^2 (%)	%RI
Trophic level	542	0.08	0.78		0.0	18.0*	1271	0.006	0.94		0.0	3.3
Hetero- *vs.* autotrophic	542	1.5	0.23		0.1	0.5	1271	0.02	0.89		0.0	0.0
N, P, or N + P	542	15.9	<0.0001*		3.6*	10.1*	1271	27.1	<0.0001*		2.6*	7.2*
Canopy	426	2.7	0.10		0.8	3.6	1056	24.6	<0.0001*		3.1*	3.7*
Latitude	428	0.26	0.61		0.0	10*	1149	13.4	0.0004*	0.011	1.2*	16.5*
Bg [NO_3^--N]	522	4.3	0.040*	−0.064	1.5*	23.5*	1103	105.4	<0.0001*	−0.28	10.3*	23.1*
Bg [SRP]	528	0.52	0.47		0.0	6.8*	1065	38.5	<0.0001*	−0.23	4.7*	15.1*
Temperature	454	5.0	0.026*	0.015	1.9*	5.8*	887	22.4	<0.0001*	0.018	3.5*	8.4*
Bg DIN:SRP	432	1.6	0.21		0.5	12.4*	822	45.2	<0.0001*	−0.22	6.2*	20.4*
PAR	385	4.2	0.042*	−0.065	1.4*	9.4*	561	30.7	<0.0001*	0.18	7.4*	2.3*
Bg [NH_4^+-N]	512	6.9	0.009*	−0.13	1.4*	nd	786	7.7	0.006*	−0.14	1.0*	nd
Bg [DIN]	442	4.8	0.029*	−0.094	1.8*	nd	856	69.6	<0.0001*	−0.27	8.0*	nd
Bg [TN]	420	3.1	0.08		0.2	nd	507	32.7	<0.0001*	−0.33	7.0*	nd
Bg [DOC]	320	4.1	0.044*	−0.18	1.5*	nd	339	17.8	<0.0001*	−0.42	6.8*	nd
Bg [TP]	122	1.0	0.32		0.0	nd	202	43.5	<0.0001*	−0.69	19.3*	nd
Bg DIN:TP	114	4.9	0.029*	−0.56	3.6*	nd	97	1.9	0.18		1.0	nd
Bg TN:TP	114	2.3	0.13		1.1	nd	113	3.2	0.08		2.1	nd
pH	20	0.003	0.96		0.0	nd	207	2.1	0.15		0.2	nd

Bg, background concentration; DIN, dissolved inorganic nitrogen; DOC, dissolved organic carbon; PAR, photosynthetically active radiation; SRP, soluble reactive phosphorus; TN, total nitrogen; TP, total phosphorus.

Table 3. Summary of weighted general linear model (GLM) and boosted regression tree (BRT) results for each independent variable predicted to affect ln response ratio (LRR) values from non-nutrient diffusing substrata (NDS) experiments ($N = 1249$ total; 801 with variance V). For each variable, the GLM test statistic [F: (H_0: mean LRR = 0 or H_0: mean LRRs are equal across different categories of class variables); or Z: (H_0: the standardized regression coefficient for the relationship between LRR and the independent variable = 0)], slope of relationship (only for the most important continuous variables), r^2 (expressed as %) and P value are shown. For rate *versus* state comparisons at the top of the table, mean LRR (1 SE) are reported. For BRT models, relative importance (%RI) values are listed, and redundant or low-N factors were not included (nd). Most important r^2 ($P < 0.05$) and %RI values [>100/N, after Müller *et al.* (2013)] are identified with an asterisk (*). Variables with predictive influence on LRR in both GLM and BRT models are highlighted in bold type

$F = 2.23$, $P = 0.112$
Rate LRR, $N = 510$, mean (median) = 0.771 (0.624); $N = 303$ with V, $Z = 13.30$, $P < 0.0001$*
State LRR, $N = 740$, mean (median) = 0.557 (0.319); $N = 497$ with V, $Z = 15.15$, $P < 0.0001$*

	Rate variables						State variables					
Rate *vs.* state	N	F or Z	P	Slope	r^2 (%)	%RI	N	F or Z	P	Slope	r^2	%RI
Trophic level	303	2.2	0.14		0.0	9.2*	497	12.2	0.0005*		0.0	5.8
Hetero- *vs.* autotrophic	266	0.15	0.69		0.0	0.4	470	2.0	0.16		0.0	0.3
N, P, or N + P	303	12.9	0.0004*		4.0*	7.3*	497	7.0	0.008*		1.2*	5.1
Experiment type	303	3.6	0.029*		2.7*	5.9	497	3.6	0.028*		1.7*	1.6
Canopy	234	0.24	0.63		0.0	0.0	387	0.62	0.43		0.0	3.5
Latitude	268	0.007	0.93		0.0	0	420	12.2	0.0005*	0.014	2.9*	0
Bg [NO_3^--N]	200	0.11	0.73		0.0	11.6*	352	0.44	0.51		0.0	14.0*
Bg [SRP]	282	5.3	0.02*	−0.16	2.4*	10.2*	416	16.4	<0.0001*	−0.24	4.1*	22.0*
Temperature	232	0.17	0.68		0.0	33.6*	362	2.6	0.11		0.3	18.3*
Bg DIN:SRP	152	2.5	0.11		0.0	6.9*	285	0.09	0.77		0.0	9.6
PAR	56	7.5	0.008*	0.35	12.6*	13.6*	87	1.1	0.30		0.9	19.8*
Bg [NH_4^+-N]	144	6.9	0.010*	−0.24	4.8*	nd	280	1.3	0.26		0.1	nd
Bg [DIN]	203	3.8	0.052	−0.20	2.0*	nd	376	11.5	0.0008*	−0.22	2.6*	nd
Bg [TN]	75	1.2	0.28		0.7	nd	175	8.3	0.004*	−0.38	3.7*	nd
Bg [DOC]	56	1.1	0.31		0.0	nd	64	0.14	0.71		0.0	nd
Bg [TP]	98	0.45	0.50		0.0	nd	175	0.01	0.91		0.0	nd
Bg DIN:TP	58	0.28	0.60		0.0	nd	114	1.0	0.31		0.1	nd
Bg TN:TP	73	0.21	0.65		0.0	nd	136	10.6	0.001*	−0.70	8.5*	nd
pH	150	1.5	0.22		0.3	nd	216	10.2	0.002*	0.15	5.6*	nd
Exp [DIN]	148	5.6	0.019*	−0.25	4.1*	nd	304	0.06	0.81		0.0	nd
Exp [SRP]	236	4.1	0.044	−0.13	2.8*	nd	330	0.95	0.33		0.03	nd
Exp [NO_3^--N]	141	1.4	0.24		0.3	nd	324	0.94	0.33		0.0	nd
Exp [NH_4^+-N]	45	1.0	0.32		0.0	nd	120	1.9	0.17		2.1	nd
Exp [TN]	33	0.04	0.85		0.0	nd	74	2.9	0.09		1.0	nd
Exp [TP]	19	3.7	0.07		11.5	nd	73	3.8	0.05		5.3	nd
Prop NO_3^--N +	101	1.6	0.21		1.9	nd	211	0.56	0.45		0.0	nd
Prop SRP +	142	1.7	0.20		2.7	nd	205	1.2	0.27		0.05	nd
Prop DIN +	69	1.1	0.29		0.0	nd	136	8.3	0.005*	0.38	7.1*	nd
Prop NH_4^+-N +	40	14.5	0.0005*	0.54	34.6*	nd	80	2.7	0.10		2.7	nd
Prop TN +	28	0.09	0.77		0.0	nd	22	0.57	0.46		0.0	nd
Prop TP +	5	6.9	0.08		64.1	nd	18	0.03	0.86		0.0	nd

Bg, background concentration; DIN, dissolved inorganic nitrogen; DOC, dissolved organic carbon; Exp, experimentally elevated nutrient concentration; PAR, photosynthetically active radiation; Prop +, proportion of nutrient added; SRP, soluble reactive phosphorus; TN, total nitrogen; TP, total phosphorus.

and state LRR. Because of these large quantitative and qualitative differences in enrichment responses, NDS and non-NDS experiments were treated separately in subsequent analyses (Tables 2 and 3).

Across all responses, LRR was highest where both N and P were added together; for rate variables, N + P addition was higher than either N or P addition alone [mean 0.24, 0.08, and 0.61 (+27, +8, and +84%) for N, P, and N + P respectively], and for state variables, N + P and N addition were higher than where P alone was added [mean 0.44, 0.11, and 0.49 (+55, +12, and +63%) for N, P, and N + P respectively; Fig. 2A, B]. This pattern was primarily driven by flume and whole-stream experiments for rate LRRs, and by NDS experiments for state LRRs, since state LRRs did not differ between N, P and N + P addition treatments in whole-stream experiments or flumes (Fig. 2E, F, Table 3).

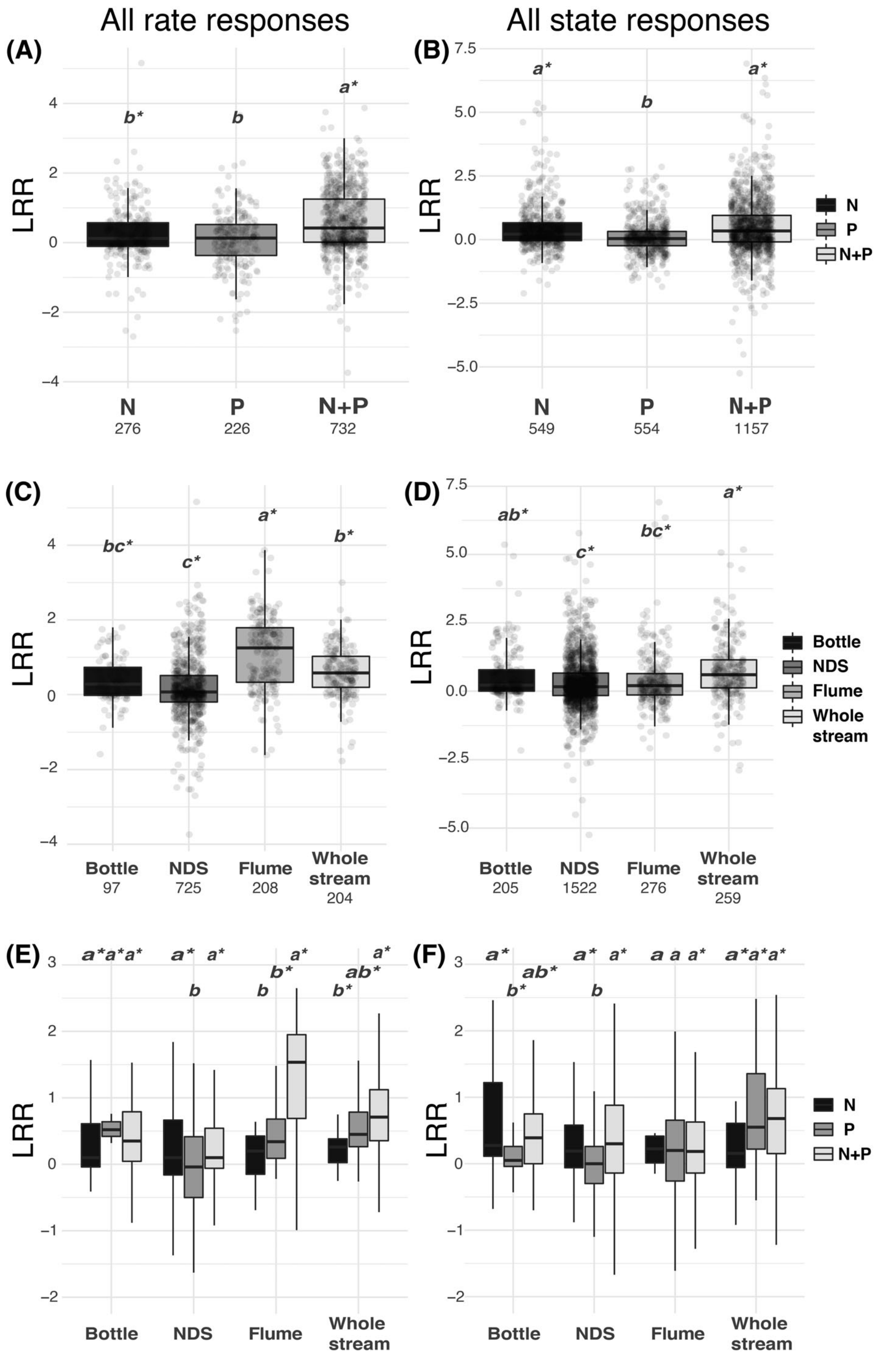

Fig 2. Boxplots of all ln response ratio (LRR) data for rate (A, C, E) and state (B, D, F) responses within categories of nutrient added (A, B), experiment type (C, D) and nutrient added by experiment type (E, F). Boxplots outline the median, interquartile range (IQR, 25–75th percentiles), and 1.5*IQR of all data in each category; all data points are shown in the single-category plots (A–D). Response distributions that differ from 0 ($P < 0.01$) are identified with an asterisk (*), and within each graph, group means with the same lowercase letters are not different ($P > 0.05$, Tukey *post-hoc* test). Sample size (*N* responses) is noted on the *x*-axis in graphs A–D. Sample sizes in E are: for bottle N (47), P (3), N + P (47); NDS N (170), P (174), N + P (381); flume N (27), P (22), N + P (160); and whole stream N (32), P (27), N + P (145). Sample sizes in F are: for bottle N (72), P (53), N + P (80); NDS N (418), P (399), N + P (705); flume N (20), P (71), N + P (185); and whole stream N (39), P (31), N + P (189). NDS, nutrient-diffusing substrata.

Similar to previous studies (Beck & Hall, 2018), P enrichment treatments in NDS experiments produced consistently low responses, as neither rate nor state LRR distributions differed from zero (Fig. 2E, F).

(3) Effects of nutrient enrichment across trophic levels

Responses of primary producers, heterotrophic microbes, primary consumers, and secondary consumers to nutrient additions were variable, but mean LRR values for all trophic levels were greater than zero (Fig. 3, Table S1, Fig. S1). In the subset of NDS rate responses, both primary producer and integrated ecosystem LRR distributions were different from 0 (mean 0.20 = +22%, and 0.14 = +15%, respectively), but did not differ from one another (Fig. 3A, Table 2). There was only one study ($N = 3$ responses) that measured NDS heterotrophic microbial respiration rate responses, and none that measured rates of production for consumers (Fig. 3A). In NDS state responses, both primary producer and heterotrophic microbe LRR distributions were different from 0 (mean 0.30 = +35%, and 0.34 = +40%, respectively), while primary consumer LRR was not, and there was only one experiment that measured higher-order consumer response (Fig. 3B). However, NDS state LRRs did not differ across trophic levels (Table 2).

In non-NDS experiments, mean rate LRRs for primary producers, heterotrophic microbes, primary consumers, secondary and tertiary consumers, and integrated ecosystems were all greater than 0 [LRR 1.03 (+180%), 0.47 (+60%),

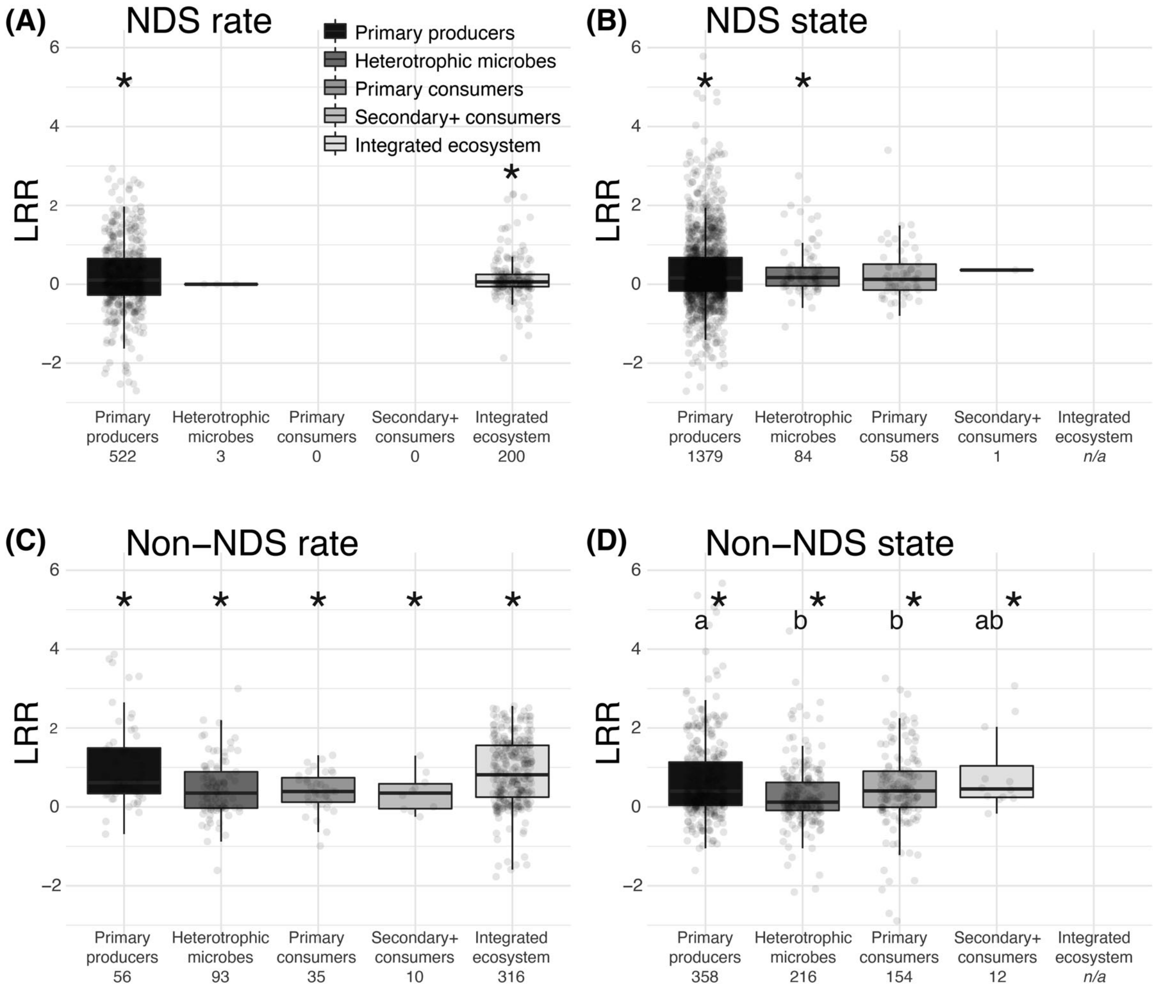

Fig 3. Boxplots of all ln response ratio (LRR) data for (A) rate and (B) state nutrient-diffusing substrata (NDS) responses, and (C) rate and (D) state non-NDS responses across trophic levels. Other details are as in Fig. 2. Three high outliers and four low outliers for primary producer data are outside the plot. Secondary+, secondary and tertiary consumers.

0.40 (+49%), 0.36 (+43%), and 0.87 (+139%), respectively; Fig. 3C], but did not differ across trophic levels (Table 3, Fig. 3C). For non-NDS state variables, mean LRRs for all trophic categories were greater than 0 (primary producer 0.74 = +109%, heterotrophic microbes 0.30 = +35%, primary consumers 0.47 = +60%, secondary and tertiary consumers 0.88 = +141%; Fig. 3D). In non-NDS state experiments primary producer LRR was higher than that of heterotrophic microbes and primary consumers, but similar to LRR of secondary and tertiary consumers (Table 3, Fig. 3D).

(4) Responses of detrital and algal food-web pathways

Both heterotrophic and autotrophic responses in all experiments were greater than 0 (Fig. 4): respectively, heterotrophic and autotrophic rate variables in NDS experiments had mean LRR of 0.29 and 0.17 (+34 and +19%, Fig. 4A), and in non-NDS rate experiments a mean LRR of 0.74 and 0.65 (+110 and +92%, Fig. 4C); and state variables in NDS experiments had a mean LRR of 0.35 and 0.30 (+42 and +35%, Fig. 4B) and in non-NDS experiments a mean

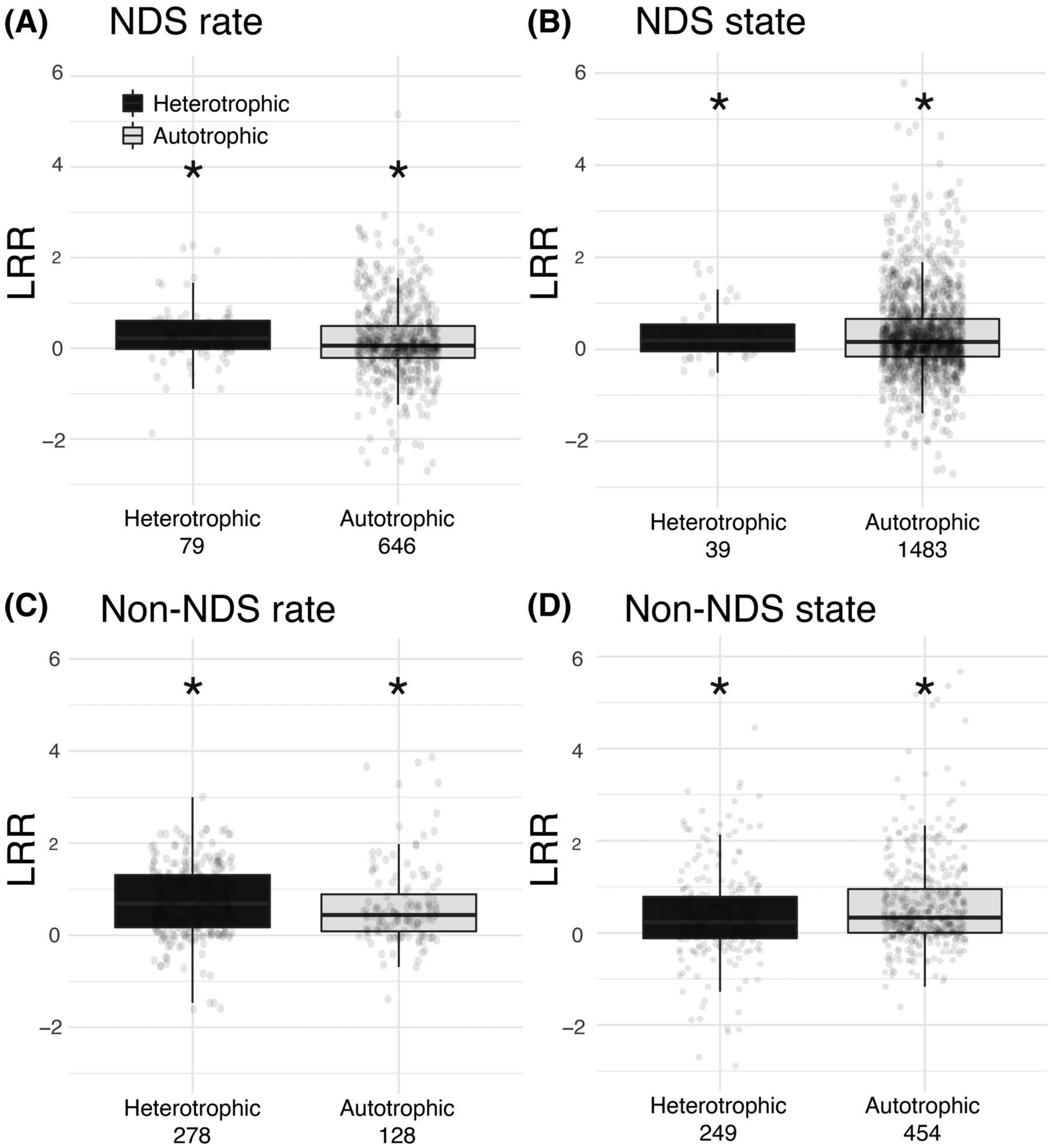

Fig 4. Boxplots of all ln response ratio (LRR) data for (A) rate and (B) state nutrient-diffusing substrata (NDS) responses, and (C) rate and (D) state non-NDS responses in heterotrophic and autotrophic food-web components. Other details are as in Fig. 2. Three high outliers and four low outliers for autotrophic data are outside the plot.

Table 4. Sample size, tree complexity, optimal tree number, and mean deviance for final boosted regression tree (BRT) models for each of the four BRT analyses [ln response ratio (LRR) values for rate and state variables in nutrient-diffusing substrata (NDS) and non-NDS experiments]. Regression slopes and r^2 values for relationships between observed and modelled LRR estimates are also provided

Parameter	NDS experiments		Non-NDS experiments	
	Rate variables	State variables	Non-NDS rate variables	Non-NDS state variables
Sample size	691	1518	501	739
Tree complexity*	3	3	2	2
Optimal number of trees	3050	3650	3700	9400
Final mean deviance	0.34	0.48	0.26	0.48
r^2	0.30	0.33	0.37	0.09
Regression slope	0.35	0.36	0.43	0.27

*Total sample size, 80% of which was randomly selected to train the model.

LRR of 0.39 and 0.65 (+48 and +92%, Fig. 4D). However, there were no statistical differences in LRRs between heterotrophic and autotrophic food web components in any category (Tables 2 and 3).

(5) Environmental drivers of the effects of nutrient enrichment

A number of environmental and experimental factors influenced nutrient enrichment (LRRs) in both NDS and non-NDS experiments. Generally, in NDS experiments, state and rate LRRs decreased with increasing background inorganic N concentrations, and state LRRs additionally decreased with increasing background SRP concentrations (Table 2). In NDS experiments, PAR modulated an increase in state LRRs and a decrease in rate LRRs (Table 2). Background DOC concentrations were negatively related to NDS state and rate LRR values, and a closed canopy was negatively associated with NDS state variable LRRs (Table 2). Temperature was positively correlated with both rate and state LRRs in NDS experiments (Table 2).

In non-NDS experiments, both rate and state LRRs decreased with background SRP and DIN concentrations (Table 3). In non-NDS experiments, the concentrations and proportional increases in added nutrients (SRP, TP, NH_4^+, NO_3^-, DIN, TN) were generally not related to LRRs, with the exceptions that proportional increases in DIN and NH_4^+ were positively related to LRRs for state and rate variables, respectively, and experimental concentrations of DIN and SRP were negatively related to LRRs for rate responses (Table 3). PAR modulated an increase in rate LRR, but not in state LRRs (Table 3). pH was positively related to non-NDS state variable LRRs (Table 3). Overall, in both NDS and non-NDS experiments background nutrient concentrations, canopy cover (light availability), and temperature were related to the response magnitude of state and rate variables, although relationships were weaker for non-NDS experiments (see Table 3).

BRT models accounted for 30–37% of the variation in LRR values across the three groups of analyses (rate and state LRRs for NDS experiments and rate LRRs for non-NDS experiments), but less than 10% of the variation in non-NDS state variables (Table 4). All BRT models tended to underestimate the largest LRR values, because slopes for the relationships between observed and modelled LRR estimates were consistently below 0.5. The final model for state variable responses in NDS experiments revealed that high LRR values occurred at low ambient NO_3^--N concentrations, higher latitudes, and warmer water temperatures, with background NO_3^--N concentrations exerting the greatest relative influence on biotic responses, although partial dependency plots revealed non-monotonic relationships for SRP and DIN:SRP gradients (Fig. 5). By contrast, relationships between partial dependencies for rate LRR *versus* environmental variables in NDS experiments were usually non-monotonic, but positive slopes for background NO_3^--N, SRP, and temperature were significant (Fig. 6). Here and in other partial dependency plots, the lack of monotonic relationships suggests unpredictable outcomes among studies across environmental gradients (Figs 6, S2). For non-NDS experiments, the BRT model for rate-based responses accounted for 37% of the variation in LRR values (Table 4), but most partial dependency plots showed non-monotonic relationships with environmental gradients (Fig. S3) and only the negative slope for DIN:SRP ratio was different from 0 (Fig. 6). The final BRT model for state variables in non-NDS experiments could account for only 9% of the variation in LRR values (Table 4), but LRR values tended to increase with increasing light availability and slightly higher SRP concentrations (Figs 6 and S4). However, the relative influence of these variables was low, potentially due to gaps along these gradients and smaller sample sizes (Figs 6 and S4). Across all models, the most important environmental influences on biotic responses to nutrient enrichment were related to ambient nutrient concentrations, light, and water temperature. Food-web pathways (autotrophic *versus* heterotrophic) and canopy cover were not important predictors in BRT models.

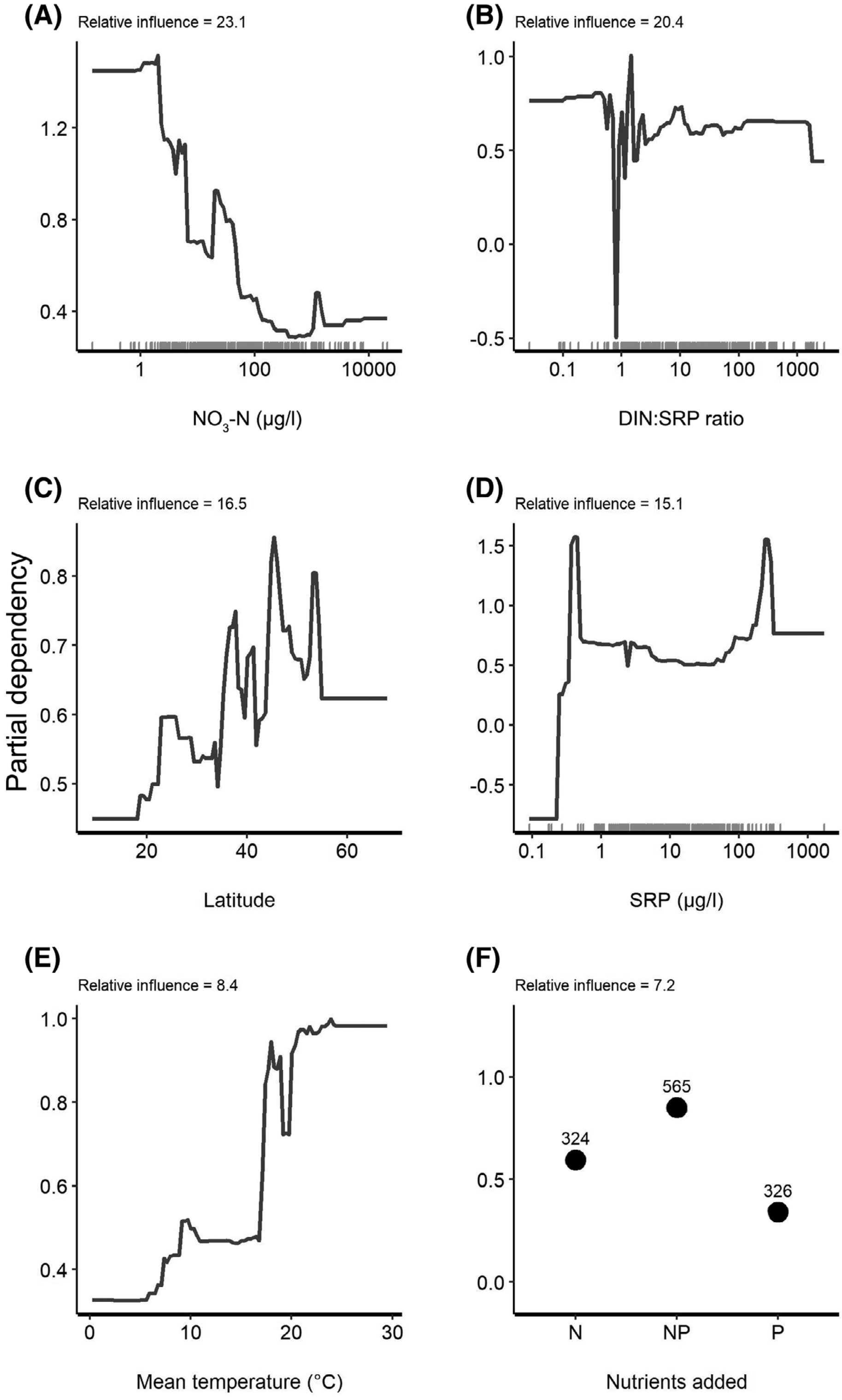

Fig 5. Partial dependency plots for state variables in nutrient-diffusing substrata (NDS) experiments. Partial dependency coefficients *versus* values (or gradients) of independent variables that exceeded 5% relative influence in the boosted regression tree (BRT) model are shown. Vertical grey hash marks above the *x*-axis represent individual observations along the gradient, and the relative influence (%) of each term is provided in each plot. Sample sizes are shown above the points for categorical variables. SRP, soluble reactive phosphorus.

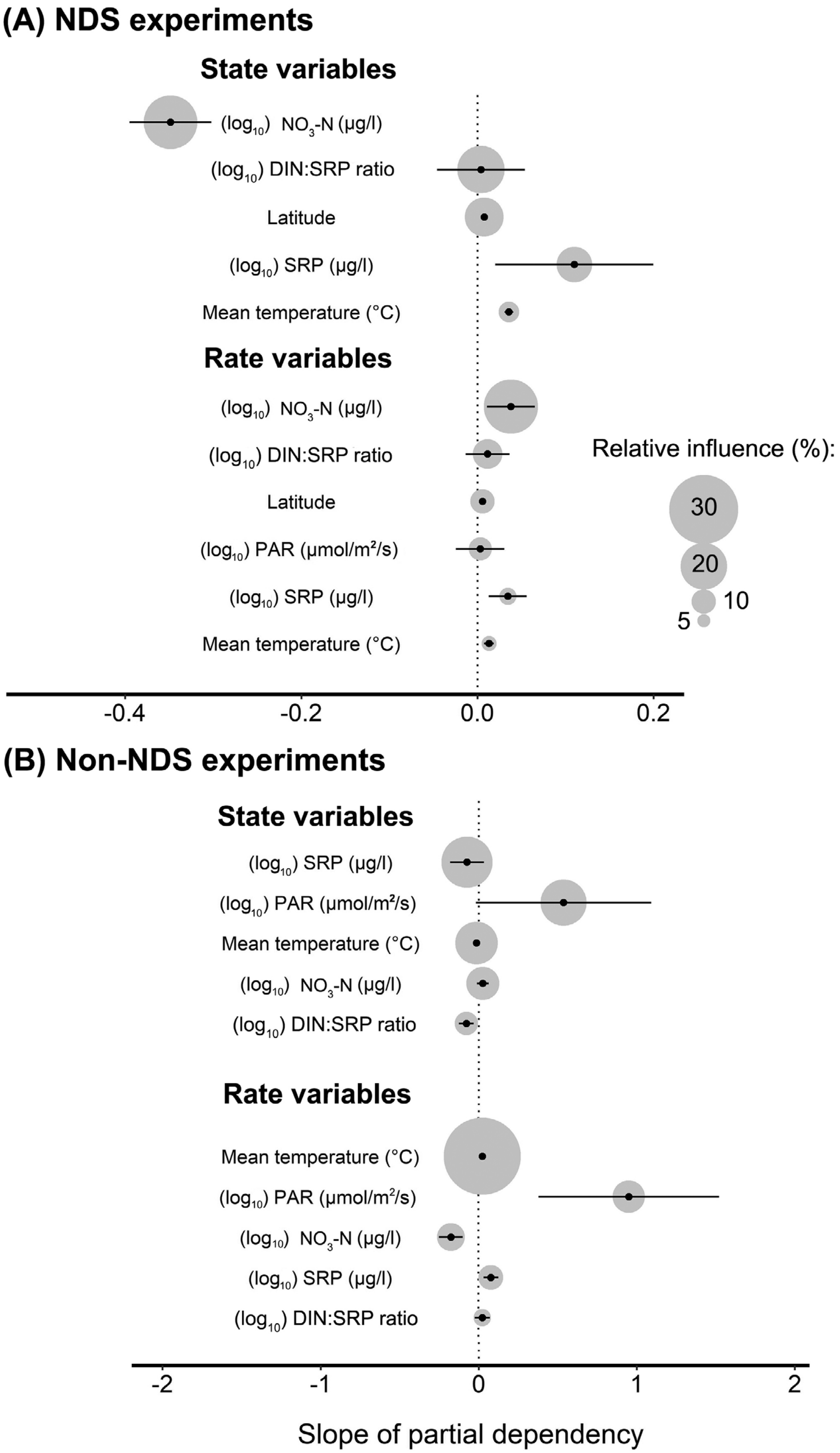

Fig 6. Slopes of relationships between partial dependency values derived from boosted regression tree (BRT) models and values for independent variables with >5% relative influence. Slope estimates ±95% confidence intervals are shown. The relative influence of each variable is depicted by the size of the grey bubble. DIN, dissolved inorganic nitrogen; NDS, nutrient-diffusing substrata; PAR, photosynthetically active radiation; SRP, soluble reactive phosphorus.

IV. DISCUSSION

(1) Overall effects of nutrients on biotic responses

Our results expand on previous meta-analyses of nutrient-enrichment experiments by showing that elevated nutrients increased autotrophic and heterotrophic rate and state variables across multiple trophic levels in streams and rivers (Figs 2, 3 and 4). These results support our first hypothesis that experiment type (i.e. NDS *versus* non-NDS), spatial scale, and nutrient type (N, P, or both), all modulated the strength of nutrient-enrichment effects on stream biota. We did not find conclusive support for our hypotheses that the magnitude of the response declines with increasing trophic level, or that heterotrophic responses are stronger than autotrophic responses (Figs 3 and 4). We did find support for our hypothesis that site environmental conditions (such as ambient nutrient concentrations, temperature, and light availability) modulate the response to nutrient enrichment (Fig. 6). Our results show that nutrient enrichment of rivers and streams has widespread stimulating effects on both rate and state variables, across multiple trophic levels, and in autotrophic and heterotrophic organisms, food webs, and ecosystems.

(2) Effects of experiment duration, type, and nutrient added

Our analyses revealed contrasts in biotic responses in NDS and non-NDS enrichment experiments, corroborating the conclusions of previous meta-analyses that called for caution when interpreting the results of NDS experiments in assessing nutrient limitation in streams (Keck & Lepori, 2012; Beck *et al.*, 2017; Beck & Hall, 2018). Although we do not know the reasons for the higher (2–5 times) LRR values obtained in flume and whole-stream experiments compared to NDS experiments (Fig. 2), we postulate that: (*i*) NDS may not be suitable for natural levels of algal establishment and growth, for example Klose, Cooper & Leydecker (2012) found that *Cladophora* that were abundant in the surrounding stream did not colonize NDS; (*ii*) NDS experiments are much smaller than flume and stream experiments (0.01 *versus* 10–100 m^2, respectively) and NDS often diverge from natural stream bottoms in physical and chemical characteristics (e.g. flow, primary succession, habitat characteristics); and (*iii*) P addition in commonly used NDS designs often elicit inhibitory responses (Sanderson *et al.*, 2009; Klose *et al.*, 2012; Beck *et al.*, 2017; Beck & Hall, 2018). The latter could be an artefact owing to the production of toxic hydrogen peroxide when agar is autoclaved together with phosphate (Tanaka *et al.*, 2014; Beck & Hall, 2018), a commonly used procedure when preparing NDS experiments that is now discouraged (Tank, Reisinger & Rosi, 2017), or may be due to differences in pH that result from different phosphate salts (Beck & Hall, 2018). Our meta-analysis includes NDS experiments from many continents and a wide range of streams in North America (e.g. Johnson, Tank & Dodds, 2009), reinforcing the conclusions of Beck *et al.* (2017) regarding the confounding effects of P inhibition, and also demonstrating that these effects are widespread. NDS experiments can mimic natural small-scale, point-source nutrient enrichment, such as local patches caused by chironomid pupal cases (Pringle & Bowers, 1984), but the exact level of nutrient enrichment in NDS is unknown in most experiments. We suggest that NDS experiments be combined with or replaced by larger spatial-scale experiments to interpret biotic responses to nutrient enrichment more accurately.

Our results indicated no short-term effect of the duration of experimental enrichment on biotic responses, although our data set included few studies of duration greater than 60 days. By contrast, experiments and observations spanning multiple seasons or years indicate substantial intra- and inter-annual variation in responses of stream biota to fertilization. Such variation could result from organismal attributes including size, lifespan, life cycle, growth, and movement (Peterson *et al.*, 1993;Cross *et al.*, 2006; Davis *et al.*, 2010) and interactions of biotic attributes with fertilization (O'Brien & Dodds, 2010; Suberkropp *et al.*, 2010). Changes in abiotic conditions, such as light, and thermal or discharge regimes could further amplify or mute the biotic responses to fertilization (Slavik *et al.*, 2004; Greenwood & Rosemond, 2005), as suggested by strong cross-site patterns attributed to abiotic conditions (Tables 2 and 3, Fig. 6).

Long-term, chronic nutrient enrichment can also produce ecological surprises. The longest running nutrient-enrichment experiments were conducted in a primary producer-based ecosystem in the Arctic (16 years; Slavik *et al.*, 2004) and in a detrital-based ecosystem in the temperate zone (5 years; Rosemond *et al.*, 2015). In the Arctic stream, moss replaced algae as the dominant primary producer after eight consecutive summers of phosphorus addition, a change not predicted by experiments of shorter duration (Slavik *et al.*, 2004). In the temperate stream, nutrient enrichment stimulated heterotrophic microbes and initially stimulated production of detritivores and invertebrate predators; however, over longer time periods, bottom-up effects on higher trophic levels were constrained by gape limitation of predators (Cross *et al.*, 2006; Davis *et al.*, 2010). In lakes and terrestrial ecosystems, where more long-term nutrient-enrichment experiments have been conducted, such non-linear and unexpected responses to fertilization have been documented repeatedly, engendering theories related to nutrient saturation and thresholds in ecosystem responses to fertilization (Aber & Melillo, 2001; Scheffer *et al.*, 2001). Thus, the relatively short-term nature of the majority of the experiments included in our meta-analysis, combined with documented contrasts between short- and long-term effects in a small number of studies, show that stream ecology research will benefit from more long-term fertilization experiments (Dodds, 2006; Groffman *et al.*, 2006).

(3) Effects of nutrient enrichment across trophic levels

Our hypothesis that enrichment effects would attenuate across trophic levels was based on the notion of energy transfer efficiency (Lindeman, 1942; Abrams, 1993). However, we found mixed support for this hypothesis. GLM analyses detected an effect of trophic level only on state variables in non-NDS experiments (Table 3) and BRT analyses included trophic level only in models explaining rate variables (Tables 2 and 3), and these differences did not fully reflect an attenuation pattern. There was a mean increase of 43% in secondary and tertiary consumers growth rates in non-NDS experiments, which included responses from diverse organisms and limited evidence of trophic buffering of enrichment effects. Mechanisms that give rise to such effects may include promotion of algal versus detrital pathways that propagate to affect diets of higher order consumers, such as in nutrient-stimulated growth of salamanders (Bumpers *et al.*, 2015; Bumpers *et al.*, 2017). We only found significant differences across trophic levels in non-NDS state variables (Fig. 3D), with heterotrophic microbes and primary consumers having lower LRRs than primary producers and secondary and tertiary consumers. These results could suggest a stronger response in higher trophic levels (secondary consumers), but the low number of responses for secondary consumers ($N = 12$) makes it difficult to draw strong inferences. Species diversity of fishes and invertebrates has previously been linked to nutrient concentrations (Bourassa & Cattaneo, 1998; Wang, Robertson & Garrison, 2007; Evans-White *et al.*, 2009), although these studies generally found no clear link between animal biomass and nutrient levels. Our meta-analysis highlights that nutrient-enrichment effects can propagate through higher trophic levels, and this topic is in need of further research.

The magnitude of integrated ecosystem responses was similar to those of primary producers in NDS and non-NDS rate studies (Fig. 3A, C). In NDS rate studies the integrated ecosystem responses included measurements of community respiration and primary productivity on small substrates, so it is not surprising that LRR values were similar. In the non-NDS rate studies the integrated ecosystem responses include whole-stream respiration measurements (measured using changes in dissolved oxygen levels), leaf litter breakdown rates, and substrate-specific respiration and productivity, making it more surprising that the responses were similar (180% increase in primary producers, 139% increase in integrative ecosystem). Our results support previous calls for studies to include ecosystem process rates to assess the health of ecosystems (Palmer & Febria, 2012; Woodward *et al.*, 2012).

(4) Responses of detrital and algal food-web pathways

Contrary to our hypothesis that detrital food-web pathways would respond more strongly to nutrient enrichment, we found similar LRRs in heterotrophic and autotrophic pathways in rate and state variables in both NDS and non-NDS experiments (Fig. 4). One potential explanation is that, given the short duration of most studies, 'luxury' algal uptake was not involved in dampening the response of algae to nutrient enrichment. Both autotrophic and heterotrophic resources support most stream and river ecosystems (Dodds, 2006; Lau, Leung & Dudgeon, 2009). Increased nutrient loading can strongly increase algal production and biomass (Biggs, 2000; Francoeur, 2001; Hillebrand, 2002; Keck & Lepori, 2012), as well as heterotrophic microbial activity and leaf litter breakdown (Ferreira *et al.*, 2015; Rosemond *et al.*, 2015). Our results expand the limited perspective of algal biomass as the main indicator of nutrient enrichment by demonstrating a widespread sensitivity of detrital food webs to nutrient addition (Dodds, 2007; Ferreira *et al.*, 2015; Rosemond *et al.*, 2015).

(5) Environmental influences on biotic responses to nutrient enrichment

The range of response magnitude across all rate and state variables and all trophic levels was high (Fig. S1, Table S1), reflecting the many environmental and experimental factors that influence biological responses to nutrient enrichment (LRRs). The results support our hypothesis that biotic responses to nutrient enrichment were moderated by water temperature, background concentrations of inorganic N and P, and light availability (Tables 2 and 3, Figs 5, 6, S2–S4). The partial dependency values depict the strength and directionality of responses to independent gradients, but must be contextualized with corresponding relative influence values (which convey how influential the gradient is among all others) and the frequency of observations along the gradient (depicted by the grey ticks on the plots; Elith *et al.*, 2008). Variables exhibiting univariate relationships with both high influence and even distribution of observations along the gradient are most likely to consistently predict how ecological variables will respond to experimental eutrophication. Therefore, for example, the monotonic relationship between rate LRRs and PAR levels observed in non-NDS experiments (Fig. S3) suggests an important ability of light to alter biological responses to nutrient addition, a finding stressed elsewhere and in diverse contexts (Hill & Knight, 1988; Taulbee, Cooper & Melack, 2005; Hill, Rinchard & Czesny, 2011*a*; Klose, Cooper & Bennett, 2015). While temperature was the most influential variable for the rate variable, non-NDS model (Fig. S3) and exhibited a mostly univariate positive relationship, observations along the gradient were uneven and skewed towards temperate streams, thus we cannot conclusively state that temperature represents a consistently influential variable. For NDS state variables, LRRs exhibited a very strong univariate decreasing relationship with ambient NO_3^--N concentrations (Fig. 5). This clearly supports a mechanism of nutrient saturation of biotic response; a pattern congruent with previous results (Keck & Lepori, 2012; Klose *et al.*, 2012). While ambient NO_3^--N concentrations were not found to be correspondingly influential in non-NDS experiments (Figs S3, S4), this could be due to a paucity of experiments with low NO_3^--N (<10 μg l^{-1}) levels. Still, the non-NDS state variables also exhibited a mostly univariate, negative relationship with NO_3^--N like that observed in the state variable NDS gradient. The overall lack of concordance observed between NDS and non-NDS models illustrated here highlights the potential limitation of using NDS as a means of assessing ecosystem-scale responses identified elsewhere (Capps *et al.*, 2011; Beck & Hall, 2018).

Our data highlight possible thresholds of ecological responses to gradients of abiotic or nutrient variables. Partial dependency plots using BRT models can illustrate where threshold-like non-linearities occur, especially for thresholds that cannot be detected using other techniques. However, statistically detecting thresholds may be compromised by data gaps or uneven data coverage. As an example of a possible threshold, the relative importance of NO_3^--N for NDS state LRRs decreased with ambient NO_3^--N concentration, apparently reaching a lower asymptote at *ca.* 300 μg l^{-1} (Fig. 5). The lack of an increasing or decreasing trend in partial dependency above this value suggests that NDS experiments conducted in streams with ambient NO_3^--N levels above this concentration will have little or no effect. Ecological threshold responses to NO_3^--N of a similar magnitude have been reported elsewhere (Keck & Lepori, 2012; Smith *et al.*, 2013), although the potential thresholds reported here are not tailored to a specific dependent parameter (such as productivity or biomass accrual), thus comparisons must be made in this context. Such patterns are most credible where: (*i*) there is comprehensive data coverage along the gradient of interest, and (*ii*) the variable is highly influential in predicting LRR values, as is the case for ambient NO_3^--N in the NDS state variables analysed here (Fig. 5). Heterogeneity in the relative importance and consistency of thresholds for specific factors across different physicochemical settings has been observed elsewhere, rendering general conclusions difficult (Black, Moran & Frankforter, 2011). One fundamental challenge in identifying single-gradient thresholds is that ecological properties respond to myriad environmental changes simultaneously (Groffman *et al.*, 2006). Partial dependencies reported by BRT models attempt to address this challenge by depicting the effect of the environmental gradient after accounting for the average effects of all other variables in the model (Elith *et al.*, 2008). Other potential thresholds on LRRs that might be drawn from our findings, but that must be contextualized within the distribution of observations along the gradient, include a positive effect on non-NDS rate variables above ~20°C mean temperature and ~150 μmol m^{-2} s^{-1} PAR (Fig. S3). By contrast, the most influential variable in non-NDS state data (SRP, see Fig. S4) exhibited non-monotonicity, which indicates that a subset of studies likely exerted disproportionate influence on the model. Furthermore, overrepresentation of ecological investigations in temperate regions (Stroud & Feeley, 2017) very likely limited the distributions of several variables in our models, including mean water temperature and latitude. Therefore, targeting tropical and high-latitude sites for future nutrient-enrichment studies may allow detection of thresholds that our data are not currently able to resolve.

(6) Implications for water nutrient management

Our results are relevant to the ongoing development of numerical nutrient criteria to prevent or reduce the impairment of beneficial human uses of water bodies in the U.S.A. and elsewhere (Stoner, 2011). One of the challenges facing groups developing regional nutrient criteria is identifying biological indicators that are sensitive to nutrient pollution (EPA, 2013). Our results show that both algae and heterotrophic microbes respond to similar extents (34–43% increase) to nutrient enrichment. Work in lakes has shown a strong relationship between chlorophyll a concentration, a common index of nuisance algal levels, and TN and TP concentrations (McCauley, Downing & Watson, 1989). Although similar relationships have been developed for streams and rivers (Van Nieuwenhuyse & Jones, 1996; Biggs, 2000) these relationships were not as strong, perhaps because of interactive effects of nutrients, light, turbidity (and N and P associated with suspended sediments), and flow on algal biomass in lotic systems (Dodds, Jones & Welch, 1998). Evidence is growing that the biomass of primary producers, and of primary and secondary/tertiary consumers, leaf litter breakdown rates, and microbial biomass and activity are sensitive to nutrient pollution in streams and rivers (present study; Woodward *et al.*, 2012; Ferreira *et al.*, 2015; Rosemond *et al.*, 2015).

Although responses of higher trophic levels can be used as indicators of nutrient pollution (Woodward *et al.*, 2012), it is clear that more data are needed to quantify animal responses to nutrient enrichment at large spatial and temporal scales. Our results suggest that moderate nutrient inputs can stimulate fish production, but we caution that excessive nutrient levels can drive algal blooms and increase microbial activity leading to depleted oxygen levels and possible fish kills (Paerl *et al.*, 2004), and that nutrient enrichment may shift fish communities to less-desirable species (Wang *et al.*, 2007). The similar LRRs reported here in rate (LRR range 0.28–0.58) and state (0.30–0.65) variables across multiple trophic levels suggest that development of numerical nutrient criteria could move beyond reliance on chlorophyll a, and use multiple trophic levels and process rates.

Our analyses indicate that streams with lower DIN and higher light and temperature levels often show stronger biotic responses to increased nutrient loading. Hence, streams with these characteristics may be the most vulnerable to increased nutrient loading. Our results suggest that several approaches have the potential to mitigate the effects of nutrient pollution, including reduction of nutrient inputs and actions that increase shading or decrease water temperature (such as planting of riparian vegetation, reduction of urban runoff). Furthermore, our results showed that increased nutrient inputs affected both algal and detrital-based food webs similarly, suggesting that both must be addressed in regulatory actions (Rosemond *et al.*, 2015).

Recent debate has centred on whether regulation of both N and P are equally effective strategies in protecting inland waters from eutrophication (Schindler *et al.*, 2008; Conley *et al.*, 2009), but our results, including those from whole-stream experiments, suggest that both N and P individually and in combination influence multiple trophic levels in streams. Similar to previous studies, we found the strongest enrichment responses when N and P were added together (Francoeur, 2001; Elser *et al.*, 2007; Beck *et al.*, 2017), supporting calls for numerical criteria for both N and P (Paerl *et al.*, 2004; Lewis, Wurtsbaugh & Paerl, 2011; Wurtsbaugh, Paerl & Dodds, 2019).

V. CONCLUSIONS

(1) Increased nutrient loading to lotic ecosystems causes an average 49% increase in biomass, abundance, and activity rates across multiple trophic levels in both autotrophic and heterotrophic food webs.

(2) Experiment type modulated the response of biotic components to nutrient enrichment, with nutrient-diffusing substrata (NDS) experiments showing weaker responses than whole-stream or flume experiments. We suggest that NDS be combined with experiments at larger scales to increase our understanding of the mechanisms driving biotic responses to excess nutrients.

(3) Nutrient added (N, P, or both) affected the magnitude of the response from biotic components, with the addition of N and P together generally leading to the strongest responses. Our results provide strong support for calls to consider both N and P in strategies to protect aquatic ecosystems from eutrophication (Conley *et al.*, 2009).

(4) While experiment duration was not a clear factor explaining response magnitude, most studies were less than 60 days in duration. Given the surprises found in the few multiyear nutrient-enrichment experiments to date, we suggest that longer studies that follow the responses of multiple trophic levels (primary producers, heterotrophic microbes, primary consumers, secondary and tertiary consumers, and integrated ecosystems) are necessary.

(5) Contrary to our initial hypotheses, responses to nutrient enrichment were similar across trophic levels, and in autotrophic and heterotrophic food-web components. Increased nutrient loading will alter multiple food-web components in rivers and streams.

(6) In support of our initial hypotheses, the magnitude of enrichment responses is highly contingent on environmental conditions, such as ambient temperature, light, and background nutrient levels. This information could help to identify rivers or streams that are particularly vulnerable to increased nutrient loading, or identify management practices that could help mitigate the biotic responses to excess nutrients.

(7) Future studies should examine how increases in nutrients and other environmental variables (such as temperature, light availability, organic matter quantity and quality) interact to affect autotrophic and heterotrophic food webs. Humans are increasing nutrient loading to rivers and streams and changing many other factors, thus a more mechanistic understanding of possible interactions among stressors is needed.

VI. ACKNOWLEDGEMENTS

This work was conducted as a part of the ‘Synthesis of stream ecosystem responses to nutrient enrichment at multiple trophic levels’ Working Group funded by the National Science Foundation under grant DEB-0832652, through the Long Term Ecological Research Network Office (LNO), University of New Mexico. L.H.Z. was partially funded by this grant as initial lead for the Working Group. M.A. acknowledges funding from NSF DEB-1713502 during the writing of the manuscript. N.A.G. was supported by the Department of Energy’s Office of Science, Biological and Environmental Research. Oak Ridge National Laboratory is managed by UT-Battelle, LLC, for the US DOE under contract DE-AC05-00OR22725.

VII. REFERENCES

Data for meta-analyses were extracted from papers denoted with *.

*Abelho, M. & Graca, M. A. S. (2006). Effects of nutrient enrichment on decomposition and fungal colonization of sweet chestnut leaves in an Iberian stream (Central Portugal). *Hydrobiologia* **560**, 239–247.

*Abelho, M., Moretti, M., Franca, J. & Callisto, M. (2010). Nutrient addition does not enhance leaf decomposition in a southeastern Brazilian stream (Espinhaco mountain range). *Brazilian Journal of Biology* **70**, 747–754.

Aber, J. D. & Melillo, J. M. (2001). *Terrestrial Ecosystems*, 2nd Edition (). Academic Press, San Diego.

Abrams, P. A. (1993). Effect of increased productivity on the abundances of trophic levels. *The American Naturalist* **141**, 351–371.

Aertsen, W., Kint, V., De Vos, B., Deckers, J., van Orshoven, J. & Muys, B. (2012). Predicting forest site productivity in temperate lowland from forest floor, soil, and litterfall characteristics using boosted regression trees. *Plant and Soil* **354**, 157–172.

*Allen, N. S. & Hershey, A. E. (1996). Seasonal changes in chlorophyll alpha response to nutrient amendments in a north shore tributary of Lake Superior. *Journal of the North American Benthological Society* **15**, 170–178.

*Anderson, E. L., Welch, E. B., Jacoby, J. M., Schimek, G. M. & Horner, R. R. (1999). Periphyton removal related to phosphorus and grazer biomass level. *Freshwater Biology* **41**, 633–651.

*Ardon, M. & Pringle, C. M. (2007). The quality of organic matter mediates the response of heterotrophic biofilms to phosphorus enrichment of the water column and substratum. *Freshwater Biology* **52**, 1762–1772.

*Ardon, M., Stallcup, L. A. & Pringle, C. M. (2006). Does leaf quality mediate the stimulation of leaf breakdown by phosphorus in Neotropical streams? *Freshwater Biology* **51**, 618–633.

*Artigas, J., Romani, A. M. & Sabater, S. (2008). Effect of nutrients on the sporulation and diversity of aquatic hyphomycetes on submerged substrata in a Mediterranean stream. *Aquatic Botany* **88**, 32–38.

*Artigas, J., Romaní, A. M. & Sabater, S. (2015). Nutrient and enzymatic adaptations of stream biofilms to changes in nitrogen and phosphorus supply. *Aquatic Microbial Ecology* **75**, 91–102.

*Back, J. A., Taylor, J. M., King, R. S., Fallert, K. L. & Hintzen, E. H. (2008). Ontogenic differences in mayfly stoichiometry influence growth rates in response to phosphorus enrichment. *Fundamental and Applied Limnology* **171**, 233–240.

*Bækkelie, K. A. E., Schneider, S. C., Hagman, C. H. C. & Petrin, Z. (2017). Effects of flow events and nutrient addition on stream periphyton and macroinvertebrates: an experimental study using flumes. *Knowledge & Managment of Aquatic Ecosystems* **418**, 47.

*Baker, P. D., Brookes, J. D., Burch, M. D., Maier, H. R. & Ganf, G. G. (2000). Advection, growth and nutrient status of phytoplankton populations in the lower river Murray, South Australia. *Regulated Rivers-Research & Management* **16**, 327–344.

Beck, W. S. & Hall, E. K. (2018). Confounding factors in algal phosphorus limitation experiments. *PLoS One* **13**(10), e0205684. https://doi.org/10.1371/journal.pone.0205684.

Beck, W. S., Rugenski, A. T. & Poff, N. L. (2017). Influence of experimental, environmental, and geographic factors on nutrient-diffusing substrate experiments in running waters. *Freshwater Biology* **62**, 1667–1680.

*Benstead, J. P., Deegan, L. A., Peterson, B. J., Huryn, A. D., Bowden, W. B., Suberkropp, K., Buzby, K. M., Green, A. C. & Vacca, J. A. (2005). Responses of a beaded Arctic stream to short-term N and P fertilisation. *Freshwater Biology* **50**, 277–290.

*Benstead, J. P., Rosemond, A. D., Cross, W. F., Wallace, J. B., Eggert, S. L., Suberkropp, K., Gulis, V., Greenwood, J. L. & Tant, C. J. (2009). Nutrient enrichment alters storage and fluxes of detritus in a headwater stream ecosystem. *Ecology* **90**, 2556–2566.

Benstead, J. P., Hood, J. M., Whelan, N. V., Kendrick, M. R., Nelson, D., Hanninen, A. F. & Demi, L. M. (2014). Coupling of dietary phosphorus and growth across diverse fish taxa: a meta-analysis of experimental aquaculture studies. *Ecology* **95**, 2768–2777.

*Bergey, E. A. & Resh, V. H. (1994). Interactions between a stream caddisfly and the algae on its case - factors affecting algal quantity. *Freshwater Biology* **31**, 153–163.

*Bergfur, J. & Friberg, N. (2012). Trade-offs between fungal and bacterial respiration along gradients in temperature, nutrients and substrata: experiments with stream derived microbial communities. *Fungal Ecology* **5**, 46–52.

*Berggren, M., Laudon, H. & Jansson, M. (2007). Landscape regulation of bacterial growth efficiency in boreal freshwaters. *Global Biogeochemical Cycles* **21**, GB4002.

*Berggren, M., Laudon, H., Jonsson, A. & Jansson, M. (2010). Nutrient constraints on metabolism affect the temperature regulation of aquatic bacterial growth efficiency. *Microbial Ecology* **60**, 894–902.

*Bernhardt, E. S. & Likens, G. E. (2004). Controls on periphyton biomass in heterotrophic streams. *Freshwater Biology* **49**, 14–27.

Biggs, B. J. F. (2000). Eutrophication of streams and rivers: dissolved nutrient-chlorophyll relationships for benthic algae. *Journal of the North American Benthological Society* **19**, 17–31.

*Biggs, B. J. F. & Lowe, R. L. (1994). Responses of two trophic levels to patch enrichment along a New Zealand stream continuum. *New Zealand Journal of Marine and Freshwater Research* **28**, 119–134.

*Biggs, B. J. F., Kilroy, C. & Lowe, R. L. (1998). Periphyton development in three valley segments of a New Zealand grassland river: test of a habitat matrix conceptual model within a catchment. *Archiv Fur Hydrobiologie* **143**, 147–177.

*Biggs, B. J. F., Tuchman, N. C., Lowe, R. L. & Stevenson, R. J. (1999). Resource stress alters hydrological disturbance effects in a stream periphyton community. *Oikos* **85**, 95–108.

Black, R. W., Moran, P. W. & Frankforter, J. D. (2011). Response of algal metrics to nutrients and physical factors and identification of nutrient thresholds in agricultural streams. *Environmental Monitoring and Assessment* **175**, 397–417.

*Bondar-Kunze, E., Maier, S., Schönauer, D., Bahl, N. & Hein, T. (2016). Antagonistic and synergistic effects on a stream periphyton community under the influence of pulsed flow velocity increase and nutrient enrichment. *Science of the Total Environment* **573**, 594–602.

*Bothwell, M. L. (1989). Phosphorus limited growth dynamics of lotic periphytic diatom communities - areal biomass and cellular growth-rate responses. *Canadian Journal of Fisheries and Aquatic Sciences* **46**, 1293–1301.

*Bothwell, M. L. & Kilroy, C. (2011). Phosphorus limitation of the freshwater benthic diatom *Didymosphenia geminata* determined by the frequency of dividing cells. *Freshwater Biology* **56**, 565–578.

Bourassa, N. & Cattaneo, A. (1998). Control of periphyton biomass in Laurentian streams (Quebec). *Journal of the North American Benthological Society* **17**, 420–429.

*Bourassa, N. & Cattaneo, A. (2000). Responses of a lake outlet community to light and nutrient manipulation: effects on periphyton and invertebrate biomass and composition. *Freshwater Biology* **44**, 629–639.

*Bowes, M. J., Smith, J. T., Hilton, J., Sturt, M. M. & Armitage, P. D. (2007). Periphyton biomass response to changing phosphorus concentrations in a nutrient-impacted river: a new methodology for phosphorus target setting. *Canadian Journal of Fisheries and Aquatic Sciences* **64**, 227–238.

*Bowes, M. J., Ings, N. L., McCall, S. J., Warwick, A., Barrett, C., Wickham, H. D., Harman, S. A., Armstrong, L. K., Scarlett, P. M., Roberts, C., Lehmann, K. & Singer, A. C. (2012). Nutrient and light limitation of periphyton in the river Thames: implications for catchment management. *Science of the Total Environment* **434**, 201–212.

Bumpers, P. M., Maerz, J. C., Rosemond, A. D. & Benstead, J. P. (2015) Salamander growth rates increase along an experimental stream phosphorus gradient. *Ecology*, **96**, 2994–3004.

Bumpers, P. M., Rosemond, A. D., Maerz, J. C. & Benstead, J. P. (2017) Experimental nutrient enrichment of forest streams increases energy flow to predators along greener food-web pathways. *Freshwater Biology*, **62**, 1794–1805.

*Bushong, S. J. & Bachmann, R. W. (1989). In situ nutrient enrichment experiments with periphyton in agricultural streams. *Hydrobiologia* **178**, 1–10.

*Busse, L. B., Simpson, J. C. & Cooper, S. D. (2006). Relationships among nutrients, algae, and land use in urbanized southern California streams. *Canadian Journal of Fisheries and Aquatic Sciences* **63**, 2621–2638.

Capps, K. A., Booth, M. T., Collins, S. M., Davison, M. A., Moslemi, J. M., El-sabaawi, R. W., Simonis, J. L. & Flecker, A. S. (2011). Nutrient diffusing substrata: a field comparison of commonly used methods to assess nutrient limitation. *Journal of the North American Benthological Society* **30**, 522–532.

*Castillo, M. M., Kling, G. W. & Allan, J. D. (2003). Bottom-up controls on bacterial production in tropical lowland rivers. *Limnology and Oceanography* **48**, 1466–1475.

*Chadwick, M. A. & Huryn, A. D. (2003). Effect of a whole-catchment N addition on stream detritus processing. *Journal of the North American Benthological Society* **22**, 194–206.

*Chadwick, M. A. & Huryn, A. D. (2005). Response of stream macroinvertebrate production to atmospheric nitrogen deposition and channel drying. *Limnology and Oceanography* **50**, 228–236.

*Chase, J. W., Benoy, G. A. & Culp, J. M. (2017). Combined effects of nutrient enrichment and inorganic sedimentation on benthic biota in an experimental stream system. *Water Quality Research Journal* **52**, 151–165.

*Chessman, B. C., Hutton, P. E. & Burch, J. M. (1992). Limiting nutrients for periphyton growth in sub-alpine, forest, agricultural and urban streams. *Freshwater Biology* **28**, 349–361.

Chrzanowski, T. H. & Kyle, M. (1996). Ratios of carbon, nitrogen, and phosphorus in *Pseudomonas fluorescens* as a model for bacterial element ratios and nutrient regeneration. *Aquatic Microbial Ecology* **10**, 115–122.

*Chung, N. & Suberkropp, K. (2008). Influence of shredder feeding and nutrients on fungal activity and community structure in headwater streams. *Fundamental and Applied Limnology* **173**, 35–46.

Conley, D. J., Paerl, H. W., Howarth, R. W., Boesch, D. F., Seitzinger, S. P., Havens, K. E., Lancelot, C. & Likens, G. E. (2009). Controlling eutrophication: nitrogen and phosphorus. *Science* **323**, 1014–1015.

R Core Team (2016). *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna. https://www.r-project.org/.

*Corkum, L. D. (1996). Responses of chlorophyll-a, organic matter, and macroinvertebrates to nutrient additions in rivers flowing through agricultural and forested land. *Archiv Fur Hydrobiologie* **136**, 391–411.

*Cross, W. F., Johnson, B. R., Wallace, J. B. & Rosemond, A. D. (2005). Contrasting response of stream detritivores to long-term nutrient enrichment. *Limnology and Oceanography* **50**, 1730–1739.

*Cross, W. F., Wallace, J. B., Rosemond, A. D. & Eggert, S. L. (2006). Whole-system nutrient enrichment increases secondary production in a detritus-based ecosystem. *Ecology* **87**, 1556–1565.

*Cross, W. F., Wallace, J. B. & Rosemond, A. D. (2007). Nutrient enrichment reduces constraints on material flows in a detritus-based food web. *Ecology* **88**, 2563–2575.

*Dalton, R. L., Boutin, C. & Pick, F. R. (2015). Determining in situ periphyton community responses to nutrient and atrazine gradients via pigment analysis. *Science of the Total Environment* **515–516**, 70–82.

*Datri, C. W., Pray, C. L., Zhang, Y. & Nowlin, W. H. (2015). Nutrient enrichment scarcely affects ecosystem impacts of a non-native herbivore in a spring-fed river. *Freshwater Biology* **60**, 551–562.

*Davies, J. M. & Bothwell, M. L. (2012). Responses of lotic periphyton to pulses of phosphorus: P-flux controlled growth rate. *Freshwater Biology* **57**, 2602–2612.

*Davis, J. M., Rosemond, A. D., Eggert, S. L., Cross, W. F. & Wallace, J. B. (2010). Long-term nutrient enrichment decouples predator and prey production. *Proceedings of the National Academy of Sciences of the United States of America* **107**, 121–126.

*Deans, C. A., Behmer, S. T., Kay, A. & Voelz, N. (2015). The importance of dissolved N:P ratios on mayfly (Baetis spp.) growth in high-nutrient detritus-based streams. *Hydrobiologia* **742**, 15–26.

De'ath, G. (2007). Boosted trees for ecological modeling and prediction. *Ecology* **88**, 243–251.

*Death, R. G., Death, F. & Ausseil, O. M. N. (2007). Nutrient limitation of periphyton growth in tributaries and the mainstem of a central North Island river, New Zealand. *New Zealand Journal of Marine and Freshwater Research* **41**, 273–281.

*Demi, L. M., Benstead, J. P., Rosemond, A. D. & Maerz, J. C. (2018). Litter P content drives consumer production in detritus-based streams spanning an experimental N:P gradient. *Ecology* **99**, 347–359.

Diaz, R. J. & Rosenberg, R. (2008). Spreading dead zones and consequences for marine ecosystems. *Science* **321**, 926–929.

Dodds, W. K. (2006). Eutrophication and trophic state in rivers and streams. *Limnology and Oceanography* **51**, 671–680.

Dodds, W. K. (2007). Trophic state, eutrophication and nutrient criteria in streams. *Trends in Ecology & Evolution* **22**, 669–676.

Dodds, W. K., Jones, J. R. & Welch, E. B. (1998). Suggested classification of stream trophic state: distributions of temperate stream types by chlorophyll, total nitrogen, and phosphorus. *Water Research* **32**, 1455–1462.

Dodds, W. K., Lopez, A. J., Bowden, W. B., Gregory, S., Grimm, N. B., Hamilton, S. K., Hershey, A. E., Marti, E., McDowell, W. H., Meyer, J. L., Morrall, D., Mulholland, P. J., Peterson, J. B., Tank, J. L., Valett, H. M., et al. (2002). N uptake as a function of concentration in streams. *Journal of the North American Benthological Society* **21**, 206–220.

Dodds, W. K., Bouska, W. W., Eitzmann, J. L., Pilger, T. J., Pitts, K. L., Riley, A. J., Schloesser, J. T. & Thornbrugh, D. J. (2009). Eutrophication of US freshwaters: analysis of potential economic damages. *Environmental Science & Technology* **43**, 12–19.

Downing, J. A., Osenberg, C. W. & Sarnelle, O. (1999). Meta-analysis of marine nutrient-enrichment experiment: variation in the magnitude of nutrient limitation. *Ecology* **80**, 1157–1167.

*Eckert, R. A. & Carrick, H. J. (2014). Evidence for consumer regulation of biofilm–nutrient interactions among hardwater streams (Pennsylvania, USA). *Hydrobiologia* **722**, 183–198.

*Elbrecht, V., Beermann, A. J., Goessler, G., Neumann, J., Tollrian, R., Wagner, R., Wlecklik, A., Piggott, J. J., Matthaei, C. D. & Leese, F. (2016). Multiple-stressor effects on stream invertebrates: a mesocosm experiment manipulating nutrients, fine sediment and flow velocity. *Freshwater Biology* **61**, 362–375.

Elith, J. & Leathwick, J. (2017). Boosted Regression Trees for ecological modeling. R. Package. https://cran.r-project.org/web/packages/dismo/vignettes/brt.pdf

Elith, J., Leathwick, J. R. & Hastie, T. (2008). A working guide to boosted regression trees. *Journal of Animal Ecology* **77**, 802–813.

*Elsdon, T. S. & Limburg, K. E. (2008). Nutrients and their duration of enrichment influence periphyton cover and biomass in rural and urban streams. *Marine and Freshwater Research* **59**, 467–476.

Elser, J. J., Marzolf, E. R. & Goldman, C. R. (1990). Phosphorus and nitrogen limitation of phytoplankton growth in the freshwaters of North America: a review and critique of experimental enrichments. *Canadian Journal of Fisheries and Aquatic Sciences* **47**, 1468–1477.

Elser, J. J., Dobberfuhl, D. R., MacKay, N. A. & Schampel, J. H. (1996). Organism size, life history, and N:P stoichiometry: toward a unified view of cellular and ecosystem processes. *Bioscience* **46**, 674–684.

*Elser, J. J., Bracken, M. E. S., Cleland, E. E., Gruner, D. S., Harpole, W. S., Hillebrand, H., Ngai, J. T., Seabloom, E. W., Shurin, J. B. & Smith, J. E. (2007). Global analysis of nitrogen and phosphorus limitation of primary producers in freshwater, marine and terrestrial ecosystems. *Ecology Letters* **10**, 1135–1142.

*Elwood, J., Newbold, J. D. & Stark, R. W. (1981). The limiting role of phosphorus in a woodland stream ecosystem: effects of P enrichment on leaf decomposition and primary producers. *Ecology* **62**, 146–158.

Englund, G., Cooper, S. D. & Sarnelle, O. (2001). Application of a model of scale dependence to quantify scale domains in open predation experiments. *Oikos* **92**, 501–514.

EPA (2013). U.S. E.P.A Expert Workshop: Nutrient Enrichment Indicators in Streams. https://www.epa.gov/sites/production/files/2013-09/documents/indicatorsworkshop.pdf

Eriksson, B. K., Rubach, A. & Hillebrand, H. (2006). Biotic habitat complexity controls species diversity and nutrient effects on net biomass production. *Ecology* **87**, 246–254.

Evans-White, M. A., Dodds, W. K., Huggins, D. G. & Baker, D. S. (2009). Thresholds in macroinvertebrate biodiversity and stoichiometry across water-quality gradients in Central Plains (USA) streams. *Journal of the North American Benthological Society* **28**, 855–868.

*Ferreira, V. & Graca, M. A. S. (2007). Fungal activity associated with decomposing wood is affected by nitrogen concentration in water. *International Review of Hydrobiology* **92**, 1–8.

*Ferreira, V., Gulis, V. & Graca, M. A. S. (2006). Whole-stream nitrate addition affects litter decomposition and associated fungi but not invertebrates. *Oecologia* **149**, 718–729.

Ferreira, V., Castagneyrol, B., Koricheva, J., Gulis, V., Chauvet, E. & Graca, M. (2015). A meta-analysis of the effects of nutrient enrichment on litter decomposition in streams. *Biological Reviews* **90**, 669–688.

Fetscher, A. E., Howard, M. G., Stancheva, R. G., Kudela, M., Stein, E. D., Sutula, M. A., Busse, L. B. & Sheath, R. G. (2015). Wadeable streams as widespread sources of benthic cyanotoxins in California, USA. *Harmful Algae* **49**, 105–116.

*Findlay, S. & Sinsabaugh, R. L. (2003). Response of hyporheic biofilm metabolism and community structure to nitrogen amendments. *Aquatic Microbial Ecology* **33**, 127–136.

*Flecker, A. S., Taylor, B. W., Bernhardt, E. S., Hood, J. M., Cornwell, W. K., Cassatt, S. R., Vanni, M. J. & Altman, N. S. (2002). Interactions between herbivorous fishes and limiting nutrients in a tropical stream ecosystem. *Ecology* **83**, 1831–1844.

*Follstad Shah, J. J., Kominoski, J. S., Ardón, M., Dodds, W. K., Gessner, M. O., Griffiths, N. A., Hawkins, C. P., Johnson, S. L., Lecerf, A., LeRoy, C. J., Manning, D. W. P., Rosemond, A. D., Sinsabaugh, R. L., Swan, C. M., Webster, J. R. & Zeglin, L. H. (2017). Global synthesis of the temperature sensitivity of leaf litter breakdown in streams and rivers. *Global Change Biology* **23**, 3064–3075.

*Forrester, G. E., Dudley, T. L. & Grimm, N. B. (1999). Trophic interactions in open systems: effects of predators and nutrients on stream food chains. *Limnology and Oceanography* **44**, 1187–1197.

Francoeur, S. E. (2001). Meta-analysis of lotic nutrient amendment experiments: detecting and quantifying subtle responses. *Journal of the North American Benthological Society* **20**, 358–368.

*Francoeur, S. N., Biggs, B. J. F., Smith, R. A. & Lowe, R. L. (1999). Nutrient limitation of algal biomass accrual in streams: seasonal patterns and a comparison of methods. *Journal of the North American Benthological Society* **18**, 242–260.

*Gafner, K. & Robinson, C. T. (2007). Nutrient enrichment influences the responses of stream macroinvertebrates to disturbance. *Journal of the North American Benthological Society* **26**, 92–102.

*García, L., Pardo, I., Cross, W. F. & Richardson, J. S. (2017). Moderate nutrient enrichment affects algal and detritus pathways differently in a temperate rainforest stream. *Aquatic Sciences* **79**, 941–952.

*Gaudes, A., Ocana, J. & Munoz, I. (2012). Meiofaunal responses to nutrient additions in a Mediterranean stream. *Freshwater Biology* **57**, 956–968.

*Gibeau, G. G. & Miller, M. C. (1989). A micro-bioassay for epilithon using nutrient-diffusing artificial substrata. *Journal of Freshwater Ecology* **5**, 171–176.

*Grattan, R. M. & Suberkropp, K. (2001). Effects of nutrient enrichment on yellow poplar leaf decomposition and fungal activity in streams. *Journal of the North American Benthological Society* **20**, 33–43.

*Greenwood, J. L. & Rosemond, A. D. (2005). Periphyton response to long-term nutrient enrichment in a shaded headwater stream. *Canadian Journal of Fisheries and Aquatic Sciences* **62**, 2033–2045.

*Greenwood, J. L., Rosemond, A. D., Wallace, J. B., Cross, W. F. & Weyers, H. S. (2007). Nutrients stimulate leaf breakdown rates and detritivore biomass: bottom-up effects via heterotrophic pathways. *Oecologia* **151**, 637–649.

*Grimm, N. B. & Fisher, S. G. (1986). Nitrogen limitation in a Sonoran Desert stream. *Journal of the North American Benthological Society* **5**, 2–15.

*Grimm, N. B., Sheibley, R. W., Crenshaw, C. L., Dahm, C. N., Roach, W. J. & Zeglin, L. H. (2005). N retention and transformation in urban streams. *Journal of the North American Benthological Society* **24**, 626–642.

Groffman, P. M., Baron, J. S., Blett, T., Gold, A. J., Goodman, I., Gunderson, L. H., Levinson, B. M., Palmer, M. A., Paerl, H. W., Peterson, G. D., Poff, N. L., Rejeski, D. W., Reynolds, J. F., Turner, M. G., Weathers, K. C. & Wiens, J. (2006). Ecological thresholds: the key to successful environmental management or an important concept with no practical application. *Ecosystems* **9**, 1–13.

*Guasch, H., Marti, E. & Sabater, S. (1995). Nutrient enrichment effects on biofilm metabolism in a mediterranean stream. *Freshwater Biology* **33**, 373–383.

*Gudmundsdottir, R., Olafsson, J. S., Palsson, S., Gislason, G. M. & Moss, B. (2011). How will increased temperature and nutrient enrichment affect primary producers in sub-Arctic streams? *Freshwater Biology* **56**, 2045–2058.

*Gulis, V., Ferreira, V. & Graca, M. A. S. (2006). Stimulation of leaf litter decomposition and associated fungi and invertebrates by moderate eutrophication: implications for stream assessment. *Freshwater Biology* **51**, 1655–1669.

Gulis, V., Kuehn, K. A., Schoettle, L. N., Leach, D., Benstead, J. P. & Rosemond, A. D. (2017). Changes in nutrient stoichiometry, elemental homeostasis and growth rate of aquatic litter-associated fungi in response to inorganic nutrient supply. *ISME Journal* **11**, 2729–2739.

Hall, R. O., Tank, J. L., Baker, M. A., Rosi-Marshall, E. J. & Hotchkiss, E. R. (2016). Metabolism, gas exchange, and carbon spiraling in rivers. *Ecosystems* **19**, 73–86.

*Halvorson, H. M., Scott, E. E., Entrekin, S. A., Evans-White, M. A. & Scott, J. T. (2016). Light and dissolved phosphorus interactively affect microbial metabolism, stoichiometry and decomposition of leaf litter. *Freshwater Biology* **61**, 1006–1019.

*Hart, D. D. & Robinson, C. T. (1990). Resource limitation in a stream community - phosphorus enrichment effects on periphyton and grazers. *Ecology* **71**, 1494–1502.

*Hepinstall, J. A. & Fuller, R. L. (1994). Periphyton reactions to different light and nutrient levels and the response of bacteria to these manipulations. *Archiv Fur Hydrobiologie* **131**, 161–173.

*Hill, W. R. & Fanta, S. E. (2008). Phosphorus and light colimit periphyton growth at subsaturating irradiances. *Freshwater Biology* **53**, 215–225.

*Hill, W. R. & Knight, A. W. (1988). Nutrient and light limitation of algae in two northern California streams. *Journal of Phycology* **24**, 125–132.

*Hill, W. R., Boston, H. L. & Steinman, A. D. (1992). Grazers and nutrients simultaneously limit lotic primary productivity. *Canadian Journal of Fisheries and Aquatic Sciences* **49**, 504–512.

*Hill, W. R., Fanta, S. E. & Roberts, B. J. (2008). C-13 dynamics in benthic algae: effects of light, phosphorus, and biomass development. *Limnology and Oceanography* **53**, 1217–1226.

Hill, W. R., Rinchard, J. & Czesny, S. (2011a). Light, nutrients and the fatty acid composition of stream periphyton. *Freshwater Biology* **56**, 1825–1836.

*Hill, W. R., Roberts, B. J., Francoeur, S. N. & Fanta, S. E. (2011b). Resource synergy in stream periphyton communities. *Journal of Ecology* **99**, 454–463.

Hillebrand, H. (2002). Top-down versus bottom-up control of autotrophic biomass—a meta-analysis on experiments with periphyton. *Journal of the North American Benthological Society* **21**, 349–369.

*Holland, D., Roberts, S. & Beardall, J. (2004). Assessment of the nutrient status of phytoplankton: a comparison between conventional bioassays and nutrient-induced fluorescence transients (NIFTs). *Ecological Indicators* **4**, 149–159.

Howarth, R. W., Billen, G., Swaney, D., Townsend, A., Jaworski, N., Lajtha, K., Downing, J. A., Elmgren, R., Caraco, N., Jordan, T., Berendse, F., Freney, J., Kudeyarov, V., Murdoch, P. & Zhu, Z. L. (1996). Regional nitrogen budgets and riverine N&P fluxes for the drainages to the North Atlantic Ocean: natural and human influences. *Biogeochemistry* **35**, 75–139.

*Irvine, R. L. & Jackson, L. J. (2006). Spatial variance of nutrient limitation of periphyton in montane, headwater streams (McLeod River, Alberta, Canada). *Aquatic Ecology* **40**, 337–348.

*Jabiol, J., Cornut, J., Tlili, A. & Gessner, M. O. (2018). Interactive effects of dissolved nitrogen, phosphorus and litter chemistry on stream fungal decomposers. *FEMS Microbiology Ecology* **94**, fiy151.

*Johnson, L. T., Tank, J. L. & Dodds, W. K. (2009). The influence of land use on stream biofilm nutrient limitation across eight north American ecoregions. *Canadian Journal of Fisheries and Aquatic Sciences* **66**, 1081–1094.

Keck, O. I. S. & Lepori, F. (2012). Can we predict nutrient limitation in streams and rivers? *Freshwater Biology* **57**, 1410–1421.

*Keithan, E. D., Lowe, R. L. & Deyoe, H. R. (1988). Benthic diatom distribution in a Pennsylvania stream - role of pH and nutrients. *Journal of Phycology* **24**, 581–585.

*Keldsen, K. (1996). Regulation of algal biomass in a small lowland stream: field experiments on the role of invertebrate grazing, phosphorus and irradiance. *Freshwater Biology* **36**, 535–546.

*Kendrick, M. R. & Benstead, J. P. (2013). Temperature and nutrient availability interact to mediate growth and body stoichiometry in a detritivorous stream insect. *Freshwater Biology* **58**, 1820–1830.

*Kiffney, P. M. & Richardson, J. S. (2001). Interactions among nutrients, periphyton, and invertebrate and vertebrate (*Ascaphus truei*) grazers in experimental channels. *Copeia* **2001**, 422–429.

*Kilroy, C. & Larned, S. T. (2016). Contrasting effects of low-level phosphorus and nitrogen enrichment on growth of the mat-forming alga *Didymosphenia geminata* in an oligotrophic river. *Freshwater Biology* **61**, 1550–1567.

*Klose, K., Cooper, S. D. & Leydecker, A. D. (2012). Relationships among catchment land use and concentrations of nutrients, algae, and dissolved oxygen in a southern California river. *Journal of the North American Benthological Society* **31**, 908–927.

*Klose, K., Cooper, S. D. & Bennett, D. M. (2015). Effects of wildfire on stream algal abundance, community structure, and nutrient limitation. *Freshwater Science* **34**, 1494–1509.

Knapp, G. & Hartung, J. (2003). Improved tests for a random effects meta-regression with a single covariate. *Statistics in Medicine* **22**, 2693–2710.

*Kominoski, J. S., Rosemond, A. D., Benstead, J. P., Gulis, V., Maerz, J. C. & Manning, D. W. P. (2015). Low-to-moderate nitrogen and phosphorus concentrations accelerate microbially driven litter breakdown rates. *Ecological Applications* **25**, 856–865.

*Kominoski, J. S., Rosemond, A. D., Benstead, J. P., Gulis, V. & Manning, D. W. P. (2018). Experimental nitrogen and phosphorus additions increase rates of stream ecosystem respiration and carbon loss. *Limnology and Oceanography* **63**, 22–36.

*Lang, D. A., King, R. S. & Scott, J. T. (2012). Divergent responses of biomass and enzyme activities suggest differential nutrient limitation in stream periphyton. *Freshwater Science* **31**, 1096–1104.

*Larned, S. T. & Santos, S. R. (2000). Light- and nutrient-limited periphyton in low order streams of Oahu, Hawaii. *Hydrobiologia* **432**, 101–111.

Lau, D. C. P., Leung, K. M. Y. & Dudgeon, D. (2009). Are autochthonous foods more important than allochthonous resources to benthic consumers in tropical headwater streams? *Journal of the North American Benthological Society* **28**, 426–439.

Leathwick, J. R., Elith, J., Francis, M. P., Hastie, T. & Taylor, P. (2006). Variation in demersal fish species richness in the oceans surrounding New Zealand: an analysis using boosted regression trees. *Marine Ecology Progress Series* **321**, 267–281.

Lewis, W. M., Wurtsbaugh, W. A. & Paerl, H. W. (2011). Rationale for control of anthropogenic nitrogen and phosphorus to reduce eutrophication of inland waters. *Environmental Science & Technology* **45**, 10300–10305.

*Liess, A. & Hillebrand, H. (2006). Role of nutrient supply in grazer-periphyton interactions: reciprocal influences of periphyton and grazer nutrient stoichiometry. *Journal of the North American Benthological Society* **25**, 632–642.

*Liess, A. & Kahlert, M. (2009). Gastropod grazers affect periphyton nutrient stoichiometry by changing benthic algal taxonomy and through differential nutrient uptake. *Journal of the North American Benthological Society* **28**, 283–293.

*Liess, A., Lange, K., Schulz, F., Piggott, J. J., Matthaei, C. D. & Townsend, C. R. (2009). Light, nutrients and grazing interact to determine diatom species richness via changes to productivity, nutrient state and grazer activity. *Journal of Ecology* **97**, 326–336.

*Lin, Y. J., He, Z. L., Yang, Y. G., Stoffella, P. J., Phlips, E. J. & Powell, C. A. (2008). Nitrogen versus phosphorus limitation of phytoplankton growth in ten Mile Creek, Florida, USA. *Hydrobiologia* **605**, 247–258.

Lindeman, R. L. (1942). The trophic-dynamic aspect of ecology. *Ecology* **23**, 399–418.

*Lock, M. A., Ford, T. E., Hullar, M. A. J., Kaufman, M., Vestal, J. R., Volk, G. S. & Ventullo, R. M. (1990). Phosphorus limitation in an arctic river biofilm - a whole ecosystem experiment. *Water Research* **24**, 1545–1549.

*Lohman, K., Jones, J. R. & Baysingerdaniel, C. (1991). Experimental-evidence for nitrogen limitation in a northern Ozark stream. *Journal of the North American Benthological Society* **10**, 14–23.

*Lourenço-Amorim, C., Neres-Lima, V., Moulton, T. P., Sasada-Sato, C. Y., Oliveira-Cunha, P. & Zandonà, E. (2014). Control of periphyton standing crop in an Atlantic Forest stream: the relative roles of nutrients, grazers and predators. *Freshwater Biology* **59**, 2365–2373.

*Lowe, R. L., Golladay, S. W. & Webster, J. R. (1986). Periphyton responses to nutrient manipulation in streams draining clearcut and forested watersheds. *Journal of the North American Benthological Society* **5**, 221–229.

*Lowell, R. B., Culp, J. M. & Wrona, F. J. (1995). Stimulation of increased short-term growth and development of mayflies by pulp-mill effluent. *Environmental Toxicology and Chemistry* **14**, 1529–1541.

*Mallin, M. A., McIver, M. R., Ensign, S. H. & Cahoon, L. B. (2004). Photosynthetic and heterotrophic impacts of nutrient loading to Blackwater streams. *Ecological Applications* **14**, 823–838.

*Mallory, M. A. & Richardson, J. S. (2005). Complex interactions of light, nutrients and consumer density in a stream periphyton-grazer (tailed frog tadpoles) system. *Journal of Animal Ecology* **74**, 1020–1028.

*Manning, D. W. P., Rosemond, A. D., Kominoski, J. S., Gulis, V., Benstead, J. P. & Maerz, J. C. (2015). Detrital stoichiometry as a critical nexus for the effects of streamwater nutrients on leaf litter breakdown rates. *Ecology* **96**, 2214–2224.

*Manning, D. W. P., Rosemond, A. D., Gulis, V., Benstead, J. P., Kominoski, J. S. & Maerz, J. C. (2016). Convergence of detrital stoichiometry predicts thresholds of nutrient-stimulated breakdown in streams. *Ecological Applications* **26**, 1745–1757.

*Manning, D. W. P., Rosemond, A. D., Gulis, V., Benstead, J. P. & Kominoski, J. S. (2018). Nutrients and temperature additively increase stream microbial respiration. *Global Change Biology* **24**, e233–e247.

Marcarelli, A. M., Baxter, C. V., Mineau, M. & Hall, R. O. (2011). Quantity and quality: unifying food web and ecosystem perspectives on the role of resource subsidies in freshwaters. *Ecology* **92**, 1215–1225.

*Marcus, M. D. (1980). Periphytic community response to chronic nutrient enrichment by a reservoir discharge. *Ecology* **61**, 387–399.

*Marinelarena, A. J. & Di Giorgi, H. D. (2001). Nitrogen and phosphorus removal by periphyton from agricultural wastes in artificial streams. *Journal of Freshwater Ecology* **16**, 347–353.

*Marks, J. C. & Power, M. E. (2001). Nutrient induced changes in the species composition of epiphytes on *Cladophora glomerata* Kutz. (Chlorophyta). *Hydrobiologia* **450**, 187–196.

*Matthaei, C. D., Piggott, J. J. & Townsend, C. R. (2010). Multiple stressors in agricultural streams: interactions among sediment addition, nutrient enrichment and water abstraction. *Journal of Applied Ecology* **47**, 639–649.

McCauley, E., Downing, J. A. & Watson, S. (1989). Sigmoid relationships between nutrients and chlorophyll among lakes. *Canadian Journal of Fisheries and Aquatic Sciences* **46**, 1171–1175.

*McCormick, P. V. (1994). Evaluating the multiple mechanisms underlying herbivore algal interactions in streams. *Hydrobiologia* **291**, 47–59.

*McCormick, P. V. & Stevenson, R. J. (1989). Effects of snail grazing on benthic algal community structure in different nutrient environments. *Journal of the North American Benthological Society* **8**, 162–172.

*McCormick, P. V. & Stevenson, R. J. (1991). Grazer control of nutrient availability in the periphyton. *Oecologia* **86**, 287–291.

*Miller, M. C., Deoliveira, P. & Gibeau, G. G. (1992). Epilithic diatom community response to years of PO4 fertilization - Kuparuk River, Alaska (68 N lat.). *Hydrobiologia* **240**, 103–119.

*Mitrovic, S. M., Bowling, L. C. & Buckney, R. T. (2001). Responses of phytoplankton to in-situ nutrient enrichment; potential influences on species dominance in a river. *International Review of Hydrobiology* **86**, 285–298.

*Mohamed, M. N. & Robarts, R. D. (2003). Sestonic bacterial nutrient limitation in a northern temperate river and the impact of pulp-mill effluents. *Aquatic Microbial Ecology* **33**, 19–28.

*Molinos, J. G. & Donohue, I. (2010). Interactions among temporal patterns determine the effects of multiple stressors. *Ecological Applications* **20**, 1794–1800.

*Morris, L. & Nicholson, G. (2015). Can moderate increases in nutrient loads cause ecological effects in rivers already impacted by nutrients? *Hydrobiologia* **749**, 213–229.

*Mosisch, T. D., Bunn, S. E., Davies, P. M. & Marshall, C. J. (1999). Effects of shade and nutrient manipulation on periphyton growth in a subtropical stream. *Aquatic Botany* **64**, 167–177.

*Mulholland, P. J. & Brawley, S. H. (2000). Seasonally shifting limitation of stream periphyton: response of algal populations and assemblage biomass and productivity to variation in light, nutrients, and herbivores. *Canadian Journal of Fisheries and Aquatic Sciences* **57**, 66–75.

*Mulholland, P. J., Steinman, A. D., Palumbo, A. V., Elwood, J. W. & Kirschtel, D. B. (1991). Role of nutrient cycling and herbivory in regulating periphyton communities in laboratory streams. *Ecology* **72**, 966–982.

Müller, D., Leitão, P. J. & Sikor, T. (2013). Comparing the determinants of cropland abandonment in Albania and Romania using boosted regression trees. *Agricultural Systems* **117**, 66–77.

*Neif, É. M., Graeber, D., Rodrigues, L., Rosenhøj-Leth, S., Jensen, T. M., Wiberg-Larsen, P., Landkildehus, F., Riis, T. & Baattrup-Pedersen, A. (2017). Responses of benthic algal communities and their traits to experimental changes in fine sediments, nutrients and flow. *Freshwater Biology* **62**, 1539–1550.

*Nelson, C. E., Bennett, D. M. & Cardinale, B. J. (2013). Consistency and sensitivity of stream periphyton community structural and functional responses to nutrient enrichment. *Ecological Applications* **23**, 159–173.

*Newbold, J. D., Elwood, J. W., Schulze, M. S., Stark, R. W. & Barmeier, J. C. (1983). Continuous ammonium enrichment of a woodland stream - uptake kinetics, leaf decomposition, and nitrification. *Freshwater Biology* **13**, 193–204.

*Notestein, S. K., Frazer, T. K., Hoyer, M. V. & Canfield, D. E. (2003). Nutrient limitation of periphyton in a spring-fed, coastal stream in Florida, USA. *Journal of Aquatic Plant Management* **41**, 57–60.

O'Brien, J. M. O. & Dodds, W. K. (2010). Saturation of NO_3^{2-} uptake in prairie streams as a function of acute and chronic N exposure. *Journal of the North American Benthological Society* **29**, 627–635.

Osenberg, C. W., Sarnelle, O., Cooper, S. D. & Holt, R. D. (1999). Resolving ecological questions through meta-analysis: goals, metrics and models. *Ecology* **80**, 1105–1117.

*Paaby, P. & Goldman, C. R. (1992). Chlorophyll, primary productivity, and respiration in a lowland Costa Rican stream. *Revista De Biologia Tropical* **40**, 185–198.

Paerl, H. W., Valdes, L. M., Joyner, A. R. & Piehler, M. F. (2004). Solving problems resulting from solutions: evolution of a dual nutrient management strategy for the eutrophying Neuse river estuary, North Carolina. *Environmental Science & Technology* **38**, 3068–3073.

Palmer, M. A. & Febria, C. M. (2012). The heartbeat of ecosystems. *Science* **336**, 1393–1394.

*Pan, Y., Deng, G., Wang, L., Cao, Y., Pang, W., Wang, Q., Li, B., Wang, B., Zhang, J. & Xu, R. (2017). Effects of in situ phosphorus enrichment on the benthos in a subalpine karst stream and implications for bioassessment in nature reserves. *Ecological Indicators* **73**, 274–283.

*Pearson, R. G. & Connolly, N. M. (2000). Nutrient enhancement, food quality and community dynamics in a tropical rainforest stream. *Freshwater Biology* **43**, 31–42.

*Perrin, C. J. & Richardson, J. S. (1997). N and P limitation of benthos abundance in the Nechako River, British Columbia. *Canadian Journal of Fisheries and Aquatic Sciences* **54**, 2574–2583.

Persson, J., Fink, P., Goto, A., Hood, J. M., Jonas, J. & Kato, S. (2010). To be or not to be what you eat: regulation of stoichiometric homeostasis among autotrophs and heterotrophs. *Oikos* **119**, 741–751.

*Peterson, C. G. & Grimm, N. B. (1992). Temporal variation in enrichment effects during periphyton succession in a nitrogen-limited desert stream ecosystem. *Journal of the North American Benthological Society* **11**, 20–36.

*Peterson, B. J., Hobbie, J. E., Hershey, A. E., Lock, M. A., Ford, T. E., Vestal, J. R., McKinley, V. L., Hullar, M. A. J., Miller, M. C., Ventullo, R. M. & Volk, G. S. (1985). Transformation of a tundra river from heterotrophy to autotrophy by addition of phosphorus. *Science* **229**, 1383–1386.

*Peterson, B. J., Deegan, L., Helfrich, J., Hobbie, J. E., Hullar, M., Moller, B., Ford, T. E., Hershey, A., Hiltner, A., Kipphut, G., Lock, M. A., Fiebig, D. M., McKinley, V., Miller, M. C., Vestal, J. R., Ventullo, R. & Volk, G. (1993). Biological responses of a tundra river to fertilization. *Ecology* **74**, 653–672.

*Piggott, J. J., Lange, K., Townsend, C. R. & Matthaei, C. D. (2012). Multiple stressors in agricultural streams: a mesocosm study of interactions among raised water temperature, sediment addition and nutrient enrichment. *PLoS One* **7**, e49873.

*Piggott, J. J., Niyogi, D. K., Townsend, C. R. & Matthaei, C. D. (2015a). Multiple stressors and stream ecosystem functioning: climate warming and agricultural stressors interact to affect processing of organic matter. *Journal of Applied Ecology* **52**, 1126–1134.

*Piggott, J. J., Salis, R. K., Lear, G., Townsend, C. R. & Matthaei, C. D. (2015b). Climate warming and agricultural stressors interact to determine stream periphyton community composition. *Global Change Biology* **21**, 206–222.

*Piggott, J. J., Townsend, C. R. & Matthaei, C. D. (2015c). Climate warming and agricultural stressors interact to determine stream macroinvertebrate community dynamics. *Global Change Biology* **21**, 1887–1906.

*Pringle, C. M. (1987). Effects of water and substratum nutrient supplies on lotic periphyton growth - an integrated bioassay. *Canadian Journal of Fisheries and Aquatic Sciences* **44**, 619–629.

*Pringle, C. M. (1990). Nutrient spatial heterogeneity - effects on community structure, physiognomy, and diversity of stream algae. *Ecology* **71**, 905–920.

*Pringle, C. M. & Bowers, J. (1984). An in situ substratum fertilization technique: diatom colonization on nutrient-enriched sand substrata. *Canadian Journal of Fisheries and Aquatic Sciences* **41**, 1247–1251.

*Pringle, C. M., Paabyhansen, P., Vaux, P. D. & Goldman, C. R. (1986). In situ nutrient assays of periphyton growth in a lowland Costa Rican stream. *Hydrobiologia* **134**, 207–213.

*Ramirez, A. & Pringle, C. M. (2006). Fast growth and turnover of chironomid assemblages in response to stream phosphorus levels in a tropical lowland landscape. *Limnology and Oceanography* **51**, 189–196.

*Rier, S. T. & Stevenson, R. J. (2002). Effects of light, dissolved organic carbon, and inorganic nutrients on the relationship between algae and heterotrophic bacteria in stream periphyton. *Hydrobiologia* **489**, 179–184.

Rier, S. T., Kinek, K. C., Hay, S. E. & Francoeur, S. N. (2016). Polyphosphate plays a vital role in the phosphorus dynamics of stream periphyton. *Freshwater Science* **32**, 490–502.

*Roll, S. K., Diehl, S. & Cooper, S. D. (2005). Effects of grazer immigration and nutrient enrichment on an open algae-grazer system. *Oikos* **108**, 386–400.

*Romani, A. M., Giorgi, A., Acuna, V. & Sabater, S. (2004). The influence of substratum type and nutrient supply on biofilm organic matter utilization in streams. *Limnology and Oceanography* **49**, 1713–1721.

*Rosemond, A. D. (1993). Interactions among irradiance, nutrients, and herbivores constrain a stream algal community. *Oecologia* **94**, 585–594.

*Rosemond, A. D., Mulholland, P. J. & Elwood, J. W. (1993). Top-down and bottom-up control of stream periphyton - effects of nutrients and herbivores. *Ecology* **74**, 1264–1280.

*Rosemond, A. D., Pringle, C. M., Ramirez, A. & Paul, M. J. (2001). A test of top-down and bottom-up control in a detritus-based food web. *Ecology* **82**, 2279–2293.

*Rosemond, A. D., Swan, C. M., Kominoski, J. S. & Dye, S. E. (2010). Non-additive effects of litter mixing are suppressed in a nutrient-enriched stream. *Oikos* **119**, 326–336.

Rosemond, A. D., Benstead, J. P., Bumpers, P. M., Gulis, V., Kominoski, J. S., Manning, D. W. P., Suberkropp, K. & Wallace, J. B. (2015). Experimental nutrient additions accelerate terrestrial carbon loss from stream ecosystems. *Science* **347**, 1142–1145.

*Rossi, F., Pesce, S., Mallet, C., Margoum, C., Chaumot, A., Masson, M. & Artigas, J. (2018). Interactive effects of pesticides and nutrients on microbial communities responsible of litter decomposition in streams. *Frontiers in Microbiology* **9**, 2437.

*Royer, T. V. & Minshall, G. W. (2001). Effects of nutrient enrichment and leaf quality on the breakdown of leaves in a hardwater stream. *Freshwater Biology* **46**, 603–610.

*Sabater, S., Acuna, V., Giorgi, A., Guerra, E., Munoz, I. & Romani, A. M. (2005). Effects of nutrient inputs in a forested Mediterranean stream under moderate light availability. *Archiv Fur Hydrobiologie* **163**, 479–496.

*Sabater, S., Artigas, J., Gaudes, A., Munoz, I., Urrea, G. & Romani, A. M. (2011). Long-term moderate nutrient inputs enhance autotrophy in a forested Mediterranean stream. *Freshwater Biology* **56**, 1266–1280.

*Sanderson, B. L., Coe, H. J., Tran, C. D., Macneale, K. H., Harstad, D. L. & Goodwin, A. B. (2009). Nutrient limitation of periphyton in Idaho streams: results from nutrient diffusing substrate experiments. *Journal of the North American Benthological Society* **28**, 832–845.

*Sanpera-Calbet, I., Ylla, I., Romaní, A. M., Sabater, S. & Muñoz, I. (2017). Biochemical quality of basal resources in a forested stream: effects of nutrient enrichment. *Aquatic Sciences* **79**, 99–112.

Scheffer, M., Carpenter, S. R., Foley, J. A., Folke, C. & Walker, B. (2001). Catastrophic shifts in ecosystems. *Nature* **413**, 591–596.

Schindler, D. W., Hecky, R. E., Findlay, D. L., Stainton, M. P., Parker, B. R., Paterson, M. J., Beaty, K. G., Lyng, M. & Kasian, S. E. M. (2008). Eutrophication of lakes cannot be controlled by reducing nitrogen input: results of a 37-year whole-ecosystem experiment. *Proceedings of the National Academy of Sciences of the United States of America* **105**, 11254–11258.

*Scott, J. T., Lang, D. A., King, R. S. & Doyle, R. D. (2009). Nitrogen fixation and phosphatase activity in periphyton growing on nutrient diffusing substrata: evidence for differential nutrient limitation in stream periphyton. *Journal of the North American Benthological Society* **28**, 57–68.

*Scrimgeour, G. J. & Chambers, P. A. (1997). Development and application of a nutrient-diffusing bioassay for large rivers. *Freshwater Biology* **38**, 221–231.

*Sharifi, M. & Ghafori, M. (2005). Effects of added nutrients on dry mass, AFDM, chlorophyll a and biovolume of periphyton algae in artificial streams. *Iranian Journal of Science and Technology Transaction a-Science* **29**, 29–38.

*Slavik, K., Peterson, B. J., Deegan, L. A., Bowden, W. B., Hershey, A. E. & Hobbie, J. E. (2004). Long-term responses of the Kuparuk River ecosystem to phosphorus fertilization. *Ecology* **85**, 939–954.

Small, G. E., Ardón, M., Duff, J. H., Jackman, A. P., Ramírez, A., Triska, F. J. & Pringle, C. M. (2016). Phosphorus retention in a lowland Neotropical stream following an eight-year enrichment experiment. *Freshwater Science* **35**, 1–11.

Smith, A. J., Thomas, R. L., Nolan, J. K., Velinsky, D. J., Klein, S. & Duffy, B. T. (2013). Regional nutrient thresholds in wadeable streams of New York state protective of aquatic life. *Ecological Indicators* **29**, 455–467.

*Snyder, E. B., Robinson, C. T., Minshall, G. W. & Rushforth, S. R. (2002). Regional patterns in periphyton accrual and diatom assemblage structure in a heterogeneous nutrient landscape. *Canadian Journal of Fisheries and Aquatic Sciences* **59**, 564–577.

*Stallcup, L. A., Ardón, M. & Pringle, C. M. (2006). Does nitrogen become limiting under high-P conditions in detritus-based tropical streams? *Freshwater Biology* **51**, 1515–1526.

*Stanley, E. H., Short, R. A., Harrison, J. W., Hall, R. & Wiedenfeld, R. C. (1990). Variation in nutrient limitation of lotic and lentic algal communities in a Texas (USA) river. *Hydrobiologia* **206**, 61–71.

*Steinman, A. D., Mulholland, P. J., Palumbo, A. V., Flum, T. F., Elwood, J. W. & DeAngelis, D. L. (1990). Resistance of lotic ecosystems to a light elimination disturbance - a laboratory stream study. *Oikos* **58**, 80–90.

*Stelzer, R. A. & Lamberti, G. A. (2001). Effects of N: P ratio and total nutrient concentration on stream periphyton community structure, biomass, and elemental composition. *Limnology and Oceanography* **46**(2), 357–367.

*Stelzer, R. S., Heffernan, J. & Likens, G. E. (2003). The influence of dissolved nutrients and particulate organic matter quality on microbial respiration and biomass in a forest stream. *Freshwater Biology* **48**, 1925–1937.

*Stephens, S. H., Brasher, A. M. D. & Smith, C. M. (2012). Response of an algal assemblage to nutrient enrichment and shading in a Hawaiian stream. *Hydrobiologia* **683**, 135–150.

Sterner, R. W. & Elser, J. J. (2002). *Ecological Stoichiometry: The Biology of Elements from Molecules to the Biosphere.* Princeton University Press, Princeton.

*Stevenson, R. J., Peterson, C. G., Kirschtel, D. B., King, C. C. & Tuchman, N. C. (1991). Density-dependent growth, ecological strategies, and effects of nutrients and shading on benthic diatom succession in streams. *Journal of Phycology* **27**, 59–69.

Stoner, N. (2011). EPA. Working in partnership with states to address phosphorus and nitrogen pollution through use of a framework for state nutrient reductions. https://www.epa.gov/nutrient-policy-data/working-partnership-states-address-phosphorus-and-nitrogen-pollution-through

Stroud, J. T. & Feeley, K. J. (2017). Neglect of the tropics is widespread in ecology and evolution: a comment on Clarke et al. *Trends in Ecology & Evolution* **32**, 626–628.

*Suberkropp, K. (2003). Leaf litter decomposition and microbial activity in nutrient-enriched and unaltered reaches of a headwater stream. *Freshwater Biology* **48**, 123–134.

Suberkropp, K., Gulis, V., Rosemond, A. D. & Benstead, J. P. (2010). Ecosystem and physiological scales of microbial responses to nutrients in a detritus-based stream: results of a 5-year continuous enrichment. *Limnology and Oceanography* **55**, 149–160.

*Sumner, W. T. & McIntire, C. D. (1982). Grazer - periphyton interactions in laboratory streams. *Archiv Fur Hydrobiologie* **93**, 135–157.

Tanaka, T., Kawasaki, K., Daimon, S., Kitagawa, W., Yamamoto, K., Tamaki, H., Tanaka, M., Nakatsu, C. H. & Kamagata, Y. (2014). A hidden pitfall in the preparation of agar media undermines microorganism cultivability. *Applied and Environmental Microbiology* **80**, 7659–7666.

*Tank, J. L. & Dodds, W. K. (2003). Nutrient limitation of epilithic and epixylic biofilms in ten north American streams. *Freshwater Biology* **48**, 1031–1049.

Tank, J. L., Reisinger, A. J. & Rosi, E. J. (2017). Nutrient limitation and uptake. In *Methods in Stream Ecology*, pp. 147–171. Elsevier, Amsterdam.

*Tant, C. J., Rosemond, A. D., Mehring, A. S., Kuehn, K. A. & Davis, J. M. (2015). The role of aquatic fungi in transformations of organic matter mediated by nutrients. *Freshwater Biology* **60**, 1354–1363.

*Tate, C. M. (1990). Patterns and controls of nitrogen in tallgrass prairie streams. *Ecology* **71**, 2007–2018.

*Taulbee, W. K., Cooper, S. D. & Melack, J. M. (2005). Effects of nutrient enrichment on algal biomass across a natural light gradient. *Archiv Fur Hydrobiologie* **164**, 449–464.

*Taylor, J. M., Back, J. A. & King, R. S. (2012). Grazing minnows increase benthic autotrophy and enhance the response of periphyton elemental composition to experimental phosphorus additions. *Freshwater Science* **31**, 451–462.

*Toetz, D. & Payton, M. E. (2004). Synergism of nutrients and humic acid in accrual of periphytic biomass in a subalpine stream, Colorado front range. *Journal of Freshwater Ecology* **19**, 35–40.

*Townsend, S. A., Garcia, E. A. & Douglas, M. M. (2012). The response of benthic algal biomass to nutrient addition over a range of current speeds in an oligotrophic river. *Freshwater Science* **31**, 1233–1243.

Van Nieuwenhuyse, E. E. & Jones, J. R. (1996). Phosphorus-chlorophyll relationship in temperate streams and its variation with stream catchment area. *Canadian Journal of Fisheries and Aquatic Sciences* **53**, 99–105.

*Veraart, A. J., Romani, A. M., Tornes, E. & Sabater, S. (2008). Algal response to nutrient enrichment in forested oligotrophic stream. *Journal of Phycology* **44**, 564–572.

Viechtbauer, W. (2010). Conducting meta-analyses in R with the metafor package. *Journal of Statistical Software* **36**, 1–48.

Vitousek, P. M., Aber, J. D., Howarth, R. W., Likens, G. E., Matson, P. A., Schindler, D. W., Schlesinger, W. H. & Tilman, G. D. (1997). Human alteration of the global nitrogen cycle: sources and consequences. *Ecological Applications* **7**, 737–750.

*Von Schiller, D., Marti, E., Riera, J. L. & Sabater, F. (2007). Effects of nutrients and light on periphyton biomass and nitrogen uptake in Mediterranean streams with contrasting land uses. *Freshwater Biology* **52**, 891–906.

*Walton, S. P., Welch, E. B. & Horner, R. R. (1995). Stream periphyton response to grazing and changes in phosphorus concentration. *Hydrobiologia* **302**, 31–46.

Wang, L., Robertson, D. M. & Garrison, P. J. (2007). Linkages between nutrients and assemblages of macroinvertebrates and fish in wadeable streams: implication to nutrient criteria development. *Environmental Management* **39**, 194–212.

*Wellnitz, T. A., Rader, R. B. & Ward, J. V. (1996). Importance of light and nutrients in structuring an algal community in a Rocky Mountain stream. *Journal of Freshwater Ecology* **11**, 399–413.

*Winterbourn, M. J. (1990). Interactions among nutrients, algae and invertebrates in a New Zealand mountain stream. *Freshwater Biology* **23**, 463–474.

*Winterbourn, M. J. & Fegley, A. (1989). Effects of nutrient enrichment and grazing on periphyton assemblages in some spring-fed, South Island streams. *New Zealand Natural Sciences* **16**, 57–65.

*Winterbourn, M. J., Hildrew, A. G. & Orton, S. (1992). Nutrients, algae and grazers in some british streams of contrasting pH. *Freshwater Biology* **28**, 173–182.

*Wold, A. P. & Hershey, A. E. (1999). Spatial and temporal variability of nutrient limitation in 6 north shore tributaries to Lake Superior. *Journal of the North American Benthological Society* **18**, 2–14.

Woodward, G., Gessner, M. O., Giller, P. S., Gulis, V., Hladyz, S., Lecerf, A., Malmqvist, B., McKie, B. G., Tiegs, S. D., Cariss, H., Dobson, M., Elosegi, A., Ferreira, V., Graca, M. A. S., Fleituch, T., Lacoursiere, J. O., Nistorescu, M., Pozo, J., Risnoveanu, G., Schindler, M., Vadineanu, A., Vought, L. B. M. & Chauvet, E. (2012). Continental-scale effects of nutrient pollution on stream ecosystem functioning. *Science* **336**, 1438–1440.

Wurtsbaugh, W. A., Paerl, H. W. & Dodds, W. K. (2019). Nutrients, eutrophication and harmful algal blooms along the freshwater to marine continuum. *Wiley Interdisciplinary Reviews: Water* **6**, e1373.

VIII. Supporting information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Appendix S1. List of primary studies used in the meta-analyses.

Table S1. Summary of ln response ratio (LRR) and variance (V) values across all 184 studies, 885 experiments, and 3450 numerical responses, with Z and P value statistics for each group.

Fig. S1. Distribution of effect size (ln response ratio, LRR) and variance for all control–treatment pairs, binned by response category and sorted by effect size.

Fig. S2. Partial dependency plots for the most important factors for boosted regression tree (BRT) models of rate variables in nutrient-diffusing substrata (NDS) experiments.

Fig. S3. Partial dependency plots for the most important factors for boosted regression tree (BRT) models of rate variables in non-nutrient-diffusing substrata (non-NDS) experiments.

Fig. S4. Partial dependency plots for the most important factors for boosted regression tree (BRT) models of state variables in non-nutrient-diffusing substrata (non-NDS) experiments.

(*Received 2 July 2020; revised 16 November 2020; accepted 19 November 2020; published online 17 December 2020*)