



HAL
open science

Global Patterns and Controls of Nutrient Immobilization on Decomposing Cellulose in Riverine Ecosystems

David Costello, Scott Tiegs, Luz Boyero, Cristina Canhoto, Krista Capps,
Michael Danger, Paul Frost, Mark Gessner, Natalie Griffiths, Halvor
Halvorson, et al.

► **To cite this version:**

David Costello, Scott Tiegs, Luz Boyero, Cristina Canhoto, Krista Capps, et al.. Global Patterns and Controls of Nutrient Immobilization on Decomposing Cellulose in Riverine Ecosystems. *Global Biogeochemical Cycles*, 2022, 36 (3), pp.e2021GB007163. 10.1029/2021GB007163 . hal-03613046

HAL Id: hal-03613046

<https://hal.science/hal-03613046v1>

Submitted on 20 Dec 2024

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

1
2 **Global Patterns and Controls of Nutrient Immobilization on Decomposing Cellulose**
3 **in Riverine Ecosystems**

4 **David M. Costello^{1,2*}, Scott D. Tiegs³, Luz Boyero^{4,5}, Cristina Canhoto⁶, Krista A. Capps^{7,8},**
5 **Michael Danger⁹, Paul C. Frost¹⁰, Mark O. Gessner^{11,12}, Natalie A. Griffiths¹³, Halvor M.**
6 **Halvorson¹⁴, Kevin A. Kuehn¹⁵, Amy M. Marcarelli¹⁶, Todd V. Royer¹⁷, Devan M. Mathie¹,**
7 **Ricardo J. Albariño¹⁸, Clay P. Arango¹⁹, Jukka Aroviita²⁰, Colden V. Baxter²¹, Brent J.**
8 **Bellinger²², Andreas Bruder²³, Francis J. Burdon²⁴, Marcos Callisto²⁵, Antonio Camacho²⁶,**
9 **Fanny Colas²⁷, Julien Cornut²⁸, Verónica Crespo-Pérez^{29,30}, Wyatt F. Cross³¹, Alison M.**
10 **Derry³², Michael M. Douglas³³, Arturo Elosegí⁴, Elvira de Eyto³⁴, Verónica Ferreira³⁵,**
11 **Carmen Ferriol³⁶, Tadeusz Fleituch³⁷, Jennifer J. Follstad Shah³⁸, André Frainer^{39,40}, Erica**
12 **A. Garcia⁴¹, Liliana García⁴², Pavel E. García^{43,44}, Darren P. Giling^{11,45,46}, R. Karina**
13 **Gonzales-Pomar⁴⁷, Manuel A. S. Graça³⁵, Hans-Peter Grossart^{11,48}, François Guérol⁹,**
14 **Luiz U. Hepp⁴⁹, Scott N. Higgins⁵⁰, Takuo Hishi⁵¹, Carlos Iñiguez-Armijos⁵², Tomoya**
15 **Iwata⁵³, Andrea E. Kirkwood⁵⁴, Aaron A. Koning⁵⁵, Sarian Kosten^{56,57}, Hjalmar Laudon⁵⁸,**
16 **Peter R. Leavitt⁵⁹, Aurea L. Lemes da Silva⁶⁰, Shawn J. Leroux⁶¹, Carri J. LeRoy⁶², Peter J.**
17 **Lisi⁶³, Frank O. Mase⁶⁴, Peter B. McIntyre⁶⁵, Brendan G. McKie⁶⁶, Adriana O.**
18 **Medeiros⁶⁷, Marko Miliša⁶⁸, Yo Miyake⁶⁹, Robert J. Mooney⁶³, Timo Muotka⁷⁰, Jorge**
19 **Nimptsch⁷¹, Riku Paavola⁷², Isabel Pardo⁴², Ivan Y. Parnikoza^{73,74}, Christopher J.**
20 **Patrick⁷⁵, Edwin T. H. M. Peeters⁷⁶, Jesus Pozo⁴, Brian Reid⁷⁷, John S. Richardson⁷⁸, José**
21 **Rincón⁷⁹, Geta Risnoveanu⁸⁰, Christopher T. Robinson²⁴, Anna C. Santamans²⁶, Gelas M.**
22 **Simiyu⁸¹, Agnija Skuja⁸², Jerzy Smykla³⁷, Ryan A. Sponseller⁸³, Franco Teixeira-de**
23 **Mello⁸⁴, Sirje Vilbaste⁸⁵, Veronica D. Villanueva¹⁸, Jackson R. Webster⁸⁶, Stefan Woelfl⁷¹,**
24 **Marguerite A. Xenopoulos¹⁰, Adam G. Yates^{87,88}, Catherine M. Yule⁸⁹, Yixin Zhang^{90,91}, and**
25 **Jacob A. Zwart⁹²**

26 ¹Department of Biological Sciences, Kent State University ²Environmental Science and Design
27 Research Institute, Kent State University ³Department of Biological Sciences, Oakland
28 University ⁴Department of Plant Biology and Ecology, University of the Basque Country ⁵
29 Ikerbasque, Bilbao, Spain ⁶Centre for Functional Ecology, Department of Life Sciences,
30 University of Coimbra ⁷Odum School of Ecology, University of Georgia ⁸Savannah River
31 Ecology Laboratory, University of Georgia ⁹LIEC-CNRS, University of Lorraine ¹⁰Department
32 of Biology, Trent University ¹¹Department of Experimental Limnology, Leibniz Institute of
33 Freshwater Ecology and Inland Fisheries (IGB) ¹²Department of Ecology, Berlin Institute of
34 Technology (TU Berlin) ¹³Climate Change Science Institute and Environmental Sciences
35 Division, Oak Ridge National Laboratory ¹⁴Biology Department, University of Central Arkansas
36 ¹⁵School of Biological, Environmental and Earth Sciences, University of Southern Mississippi
37 ¹⁶Department of Biological Sciences, Michigan Technological University ¹⁷O'Neill School of
38 Public and Environmental Affairs, Indiana University ¹⁸INIBIOMA, Universidad Nacional
39 Comahue (CONICET) ¹⁹Department of Biological Sciences, Central Washington University
40 ²⁰Freshwater Centre, Finnish Environment Institute (SYKE) ²¹Stream Ecology Center,
41 Department of Biological Sciences, Idaho State University ²²Watershed Protection Department,
42 City of Austin TX ²³Institute of Microbiology, University of Applied Sciences and Arts of
43 Southern Switzerland ²⁴Department of Aquatic Ecology, Eawag: Swiss Federal Institute of

44 Aquatic Science and Technology ²⁵Genetics, Ecology and Evolution Department of Institute of
 45 Biological Sciences, Universidade Federal de Minas Gerais ²⁶Cavanilles Institute for
 46 Biodiversity and Evolutionary Biology, University of Valencia ²⁷ENTPE, Université Claude
 47 Bernard Lyon 1 ²⁸Institute of Earth Sciences, University of Applied Sciences and Arts of
 48 Southern Switzerland ²⁹Limnology Laboratory, Zoology Museum QCAZ-I ³⁰School of
 49 Biological Sciences, Pontifical Catholic University of Ecuador ³¹Department of Ecology,
 50 Montana State University ³²Department of Biological Sciences, Université du Québec à Montréal
 51 ³³School of Biological Sciences, University of Western Australia ³⁴Marine Institute, Ireland
 52 ³⁵Marine and Environmental Sciences Centre (MARE), Department of Life Sciences, University
 53 of Coimbra ³⁶Department of Microbiology and Ecology, University of Valencia ³⁷Institute of
 54 Nature Conservation, Polish Academy of Sciences, Poland ³⁸Department of Geography,
 55 University of Utah ³⁹Department of Arctic and Marine Biology, UiT The Arctic University of
 56 Norway ⁴⁰Norwegian Institute for Nature Research (NINA), Tromsø, Norway ⁴¹Research
 57 Institute for the Environment and Livelihoods, Charles Darwin University ⁴²Department of
 58 Ecology and Animal Biology, University of Vigo ⁴³Escuela de Biología, Universidad de San
 59 Carlos de Guatemala ⁴⁴Ecology and Evolution Program, University of Montana ⁴⁵Centre for
 60 Applied Water Science, Institute for Applied Ecology, University of Canberra, Australia
 61 ⁴⁶CSIRO Land and Water, Canberra, Australia ⁴⁷Instituto de Ecología, Universidad Mayor de San
 62 Andrés ⁴⁸Institute for Biochemistry and Biology, Potsdam University ⁴⁹Universidade Federal de
 63 Mato Grosso do Sul, Campus Três Lagoas ⁵⁰International Institute for Sustainable Development,
 64 Canada ⁵¹Shiiba Research Forest, Kyushu University ⁵²Departamento de Ciencias Biológicas,
 65 Universidad Técnica Particular de Loja ⁵³Department of Environmental Sciences, University of
 66 Yamanashi ⁵⁴Faculty of Science, Ontario Tech University ⁵⁵Global Water Center, University of
 67 Nevada, Reno ⁵⁶Department of Aquatic Ecology and Environmental Biology, Radboud
 68 University ⁵⁷Institute for Water and Wetland Research, Radboud University ⁵⁸Forest Ecology
 69 and Management, Swedish University of Agricultural Sciences, Umeå ⁵⁹Department of Biology,
 70 University of Regina ⁶⁰Department of Ecology and Zoology, Federal University of Santa
 71 Catarina, Brazil ⁶¹Department of Biology, Memorial University of Newfoundland
 72 ⁶²Environmental Studies Program, The Evergreen State College ⁶³Center for Limnology,
 73 University of Wisconsin ⁶⁴Department of Fisheries & Aquatic Science, University of Eldoret,
 74 Kenya ⁶⁵Department of Natural Resources and the Environment, Cornell University
 75 ⁶⁶Department of Aquatic Sciences and Assessment, Swedish University of Agricultural Sciences
 76 ⁶⁷Institute of Biology, Federal University of Bahia (UFBA) ⁶⁸Department of Biology, Faculty of
 77 Science, University of Zagreb ⁶⁹Graduate School of Science and Engineering, Ehime University
 78 ⁷⁰Department of Ecology & Genetics, University of Oulu ⁷¹Instituto de Ciencias Marinas y
 79 Limnológicas, Universidad Austral de Chile ⁷²Oulanka Research Station, University of Oulu
 80 ⁷³National Antarctic Scientific Center of Ukraine ⁷⁴Institute of Molecular Biology and Genetics,
 81 National Institute of Science of Ukraine ⁷⁵Department of Biological Science, Virginia Institute of
 82 Marine Science, College of William and Mary ⁷⁶Aquatic Ecology and Water Quality
 83 Management Group, Wageningen University ⁷⁷Research Center on Ecosystems of Patagonia,
 84 Centro de Investigacion en Ecosistemas de la Patagonia ⁷⁸Department of Forest & Conservation
 85 Sciences, University of British Columbia ⁷⁹Departamento de Biología, Universidad del Zulia
 86 ⁸⁰Department of Systems Ecology and Sustainability, University of Bucharest ⁸¹School of
 87 Environmental Studies, University of Eldoret ⁸²Institute of Biology, University of Latvia
 88 ⁸³Department of Ecology and Environmental Science, Umeå University ⁸⁴Departamento de
 89 Ecología y Gestión Ambiental, Universidad de la República (CURE) ⁸⁵Institute of Agricultural

90 and Environmental Science, Estonian University of Life Sciences ⁸⁶Department of Biological
91 Sciences, Virginia Tech ⁸⁷Department of Biology, University of Waterloo ⁸⁸Canadian Rivers
92 Institute ⁸⁹School of Science, Technology and Engineering, University of the Sunshine Coast
93 ⁹⁰Department of Landscape Architecture, Soochow University ⁹¹Zhejiang Institute of Research
94 and Innovation, The University of Hong Kong ⁹² U.S. Geological Survey Integrated Information
95 Dissemination Division

96 Corresponding author: David Costello (dcostel3@kent.edu)

97 **Key Points:**

- 98 • Nitrogen and phosphorus immobilization was measured on organic matter (cotton) in 100
99 rivers and riparian zones representing 11 biomes
- 100 • Elevated temperature in riparian zones and phosphate in rivers increased immobilization,
101 and consequently accelerated decomposition
- 102 • Nitrogen and phosphorus immobilization was strongly linked by microbial stoichiometry
103 despite widely varied surface-water nutrient ratios
104

105 **Abstract**

106 Microbes play a critical role in plant litter decomposition and influence the fate of carbon in
107 rivers and riparian zones. When decomposing low-nutrient plant litter, microbes acquire nitrogen
108 (N) and phosphorus (P) from the environment (i.e., nutrient immobilization), and this process is
109 potentially sensitive to nutrient loading and changing climate. Nonetheless, environmental
110 controls on immobilization are poorly understood because rates are also influenced by plant litter
111 chemistry, which is coupled to the same environmental factors. Here we used a standardized,
112 low-nutrient organic matter substrate (cotton strips) to quantify nutrient immobilization at 100
113 paired stream and riparian sites representing 11 biomes worldwide. Immobilization rates varied
114 by 3 orders of magnitude, were greater in rivers than riparian zones, and were strongly correlated
115 to decomposition rates. In rivers, P immobilization rates were controlled by surface water
116 phosphate concentrations, but N immobilization rates were not related to inorganic nitrogen. The
117 N:P of immobilized nutrients was tightly constrained to a molar ratio of 10:1 despite wide
118 variation in surface water N:P. Immobilization rates were temperature-dependent in riparian
119 zones but not related to temperature in rivers. However, in rivers nutrient supply ultimately
120 controlled whether microbes could achieve the maximum expected decomposition at a given
121 temperature. Collectively, we demonstrated that exogenous nutrient supply and immobilization
122 are critical control points for decomposition of organic matter.

123 **Plain Language Summary**

124 Bacteria and fungi contribute to the breakdown of leaf litter in rivers and floodplains. To break
125 down leaf litter, these microbes need the nutrients nitrogen and phosphorus, and microbes can
126 get nutrients either from the leaf litter itself or from the environment. Most leaf litter has low
127 nutrient content and microbes must rely on the environment to supply nutrients. We studied

128 microbial nutrient uptake from the environment during litter breakdown to determine whether it
129 varies predictably across the globe and how it is influenced by changing climate and nutrient
130 pollution. In 100 rivers and floodplains in 11 of Earth's major biomes we placed small strips of
131 cotton as stand-ins for leaf litter. Nutrient uptake was consistently greater on cotton strips that
132 were submerged in the river compared to cotton on the floodplain. For microbes in the river,
133 nutrient uptake was faster in instances where there was more phosphorus in the water. For
134 microbes in the floodplain, nutrient uptake was faster where temperatures were warmer. Faster
135 nutrient uptake by microbes was linked with faster cotton breakdown in rivers and floodplains.
136 Our study shows that climate change and nutrient pollution can alter the activity of microbes in
137 rivers and floodplains.

138

139 **1 Introduction**

140 The uptake, storage, transformation, and release of nutrients by microorganisms are among the
141 most important contributions that riverine ecosystems make to global biogeochemical cycles.
142 However, microbially mediated nutrient uptake and transformation are influenced by
143 environmental conditions, which makes these processes particularly sensitive to global change
144 drivers including rising temperature, nutrient loading, and chemical contaminants (Boyero et al.,
145 2011; Burdon et al., 2020; Woodward et al., 2012). Detrital carbon (C) from terrestrial plants is
146 abundant in riparian zones and rivers (Sutfin et al., 2016; Tank et al., 2010) and this resource
147 fuels microbial communities and associated nutrient uptake and transformation across the
148 terrestrial–aquatic boundary. Detrital C is used by microbial decomposers most efficiently when
149 nitrogen (N) and phosphorus (P) are readily available; however, pools of N and P can differ
150 between riparian and river habitats and are increasing worldwide, with potential implications for

151 rates of organic-matter mineralization. In both habitats, the dynamics of N and P on
152 decomposing plant litter have shown characteristic patterns (Manzoni et al., 2010; Webster &
153 Benfield, 1986). Early in decomposition of carbon-rich materials, nutrients are acquired by
154 microbes from the environment, resulting in a net increase in microbial pools of N and P above
155 that supplied from the litter itself (i.e., nutrient immobilization), whereas in later stages, there is a
156 net release of nutrient mass from microbial pools (i.e., nutrient mineralization) as substrates
157 degrade. Nutrient mineralization has received more attention than nutrient immobilization, and
158 litter chemistry (e.g., C:N, lignin content) appears to be the dominant driver of mineralization
159 rates (Manzoni et al., 2010; Parton et al., 2007).

160 Current understanding of the controls on nutrient uptake and release during plant litter
161 decomposition of plant matter begins with litter chemistry, which varies widely among plant
162 species (Boyero et al., 2017; Parton et al., 2007). Exogenous factors such as climate and soil
163 nutrient concentrations are predicted to indirectly influence microbial nutrient dynamics through
164 changes in litter quality (Classen et al., 2015; Manzoni et al., 2008). However, comparative
165 studies of nutrient dynamics during decomposition at broad spatial scales have mostly used
166 natural litter, potentially confounding the chemistry of plant litter with the environmental factors
167 that produced it (Boyero et al., 2017; Pastor et al., 2014). Standardized organic-matter substrates
168 allow for stronger inferences about the role of exogenous factors in governing nutrient
169 immobilization by the microbes that drive decomposition. However, the use of standardized
170 organic matter substrates is commonly used at small spatial scales along narrow nutrient
171 gradients (e.g., Cheever et al., 2013; Pastor et al., 2014) and the only large-scale study using
172 standardized substrates did not directly measure microbial uptake (Woodward et al., 2012). Few
173 studies have quantified both N and P immobilization within the same decomposing substrate (but

174 see Robbins et al., 2019) thus we have limited evidence about the stoichiometry of
175 immobilization. Bacteria (inclusive of all decomposer prokaryotes) and fungi are the primary
176 decomposers of plant litter, and both taxa can produce biomass with an N:P reflective of the
177 nutrient supply (Danger & Chauvet, 2013; Godwin & Cotner, 2015). Thus, we may expect
178 substantial flexibility in the N:P of new decomposer biomass, resulting in N:P of immobilization
179 that is related to the exogenous nutrient supply. Furthermore, microbial uptake of nutrients, like
180 other metabolic processes, may be constrained by temperature (e.g., Brown et al., 2004, Follstad
181 Shah et al. 2017) and affected by moisture availability (Boyero et al., 2011). Therefore, studies
182 of how homogenous substrates decompose along broad climatic gradients and outside the wetted
183 channel are needed to gain understanding about a more complete set of exogenous drivers.

184 To isolate how exogenous drivers influence nutrient dynamics during litter decomposition, we
185 conducted a field experiment in which we deployed a standard organic substrate (cotton) in 100
186 rivers and their riparian zones distributed across Earth's major biomes and measured nutrient
187 immobilization potential. Cotton is composed almost entirely of cellulose (Tiegs et al., 2013),
188 and natural sources of plant litter also have substantial cellulose content (e.g., 40% for common
189 oak species in Europe; Fioretto et al., 2005). This nutrient-poor organic-matter substrate
190 obligates microbes to immobilize N and P in order to respire the cellulose. The rate of
191 immobilization, and thus indirectly the rate of cotton decomposition, is limited by the availability
192 of nutrients in the environment. Nutrient availability differs along the river–riparian boundary,
193 and we hypothesized that immobilization in riparian zones would be less than in adjacent rivers
194 due to intermittent fluxes of water and nutrients. We hypothesized that immobilization rates
195 would be governed by ambient temperature in accordance with metabolic scaling (Brown et al.,
196 2004). Thus, we predicted that global patterns of nutrient immobilization rates and ratios could

197 be predicted from latitude and terrestrial biome classification as proxies for climate (Dodds et al.
198 2019). Finally, we hypothesized that coupling between N and P immobilization would be weak
199 as the bacteria and fungi that colonize and decompose cotton (Burdon et al., 2020; Colas et al.,
200 2019) are stoichiometrically flexible.

201 **2 Materials and Methods**

202 2.1 Field sites

203 A standardized decomposition assay using cotton strips was implemented in more than 500
204 riverine ecosystems by the Cellulose Decomposition Experiment (CELLDEX) (Tiegs et al.,
205 2019). Partners completed the decomposition assay in rivers and their adjacent riparian zone
206 during peak litterfall. When possible, they logged ambient temperature (hourly) in both habitats
207 (details in Tiegs et al. 2019) and when deploying cotton strips recorded specific conductance,
208 ammonium (NH_4^+), nitrate (NO_3^-), phosphate (PO_4^{3-}), and dissolved organic carbon (DOC)
209 concentrations. Surface water PO_4^{3-} and NO_3^- spanned three orders of magnitude, which was
210 similar to the gradient in nutrient concentrations observed in the only other large scale study of
211 nutrient effects on decomposition (Woodward et al., 2012). Measurements of nutrient
212 immobilization were made for a subset of the CELLDEX sites (99 rivers and 100 riparian zones),
213 with priority given to sites with supporting data on river temperature and nutrient concentrations
214 (50–71% of sites, depending on analyte, Text S1). If >10 sites within a biome had supporting
215 data, 10 of them were randomly selected. The resulting subset represented 11 biomes with at
216 least two rivers in each biome (with the exception of deserts) that had NO_3^- and PO_4^{3-}

217 measurements (Figure S1). Paired rivers and riparian zones were included in the subsample with
218 the exception of a river in a tropical wet forest where high flows caused loss of the strips.

219 2.2 Cotton deployment and analyses

220 Cotton strips were used as a low-nutrient analogue for leaf litter. The standardized material was
221 composed of 95% cellulose with 7180 $\mu\text{g N g}^{-1}$ dry mass (dm) and 64 $\mu\text{g P g}^{-1}$ dm. The molar
222 stoichiometry of cotton (C:N = 275, C:P = 17,000) suggests large nutrient deficiencies (relative
223 to C) near the maxima observed in natural leaf litter (Manzoni et al., 2010; McGroddy et al.,
224 2004). However, the N:P ratio of 62:1 was similar to that observed for some tropical leaf litter
225 (Boyero et al., 2017; McGroddy et al., 2004). We expected that the cotton strips would be
226 colonized by heterotrophic bacteria and fungi, and growth of autotrophs was limited by shading
227 from riparian vegetation in most of our rivers (channel width <5 m at 80% of sites). Strips were
228 prepared according to published methods (Tiegs et al., 2013, 2019) and 4 replicate strips per
229 river channel and 4 per riparian zone were deployed. In the river, strips were attached to nylon
230 rope with cable ties and secured to a stake. In the riparian zone, strips were placed in contact
231 with the soil or surface organic matter. Deployment lasted approximately 3-4 weeks (range 12-57
232 d), which is a sufficient period to detect microbial degradation (as loss of tensile strength) in both
233 habitats (Tiegs et al., 2019). Upon retrieval, strips were placed in ethanol (<1 min) to arrest
234 microbial activity and dried (40 °C) before analysis. The brief submergence in ethanol had no
235 effect on nutrient concentrations (Figure S2).

236 Decomposition during incubation in the field was assessed as loss of tensile strength, resulting
237 from the microbial degradation of cellulose (Tiegs et al., 2013), as reported by Tiegs and
238 colleagues (2019). Strips were subsequently stored in a desiccator until reaching a stable mass
239 and then subsampled with a paper punch which removed small disks from the centerline of the

240 strips. Cotton disks were acid digested to measure P concentration, and C and N concentration
241 was measured with an elemental analyzer. Detailed methods, recovery of standard reference
242 materials, and procedural reproducibility are described in the Supporting Information (Text S1,
243 Table S1).

244 2.3 Data analysis

245 N and P immobilization was calculated based on the assumption that C:N and C:P of
246 decomposing litter decreases linearly as a function of mass loss (Aber & Melillo, 1982; Manzoni
247 et al., 2010). We assumed that any increase in nutrient concentration was a result of assimilation
248 by heterotrophic microbes, but we acknowledge that other processes may also contribute to
249 changes in cotton nutrient content (e.g., algal colonization, P precipitation). Algal assimilation is
250 a relatively minor contribution to net nutrient uptake on detritus except under high light and high
251 nutrient conditions (Elosegi et al. 2018; Halvorson et al. 2019). If litter has a high initial
252 C:nutrient ratio, linear changes in litter stoichiometry result in curvilinear trajectories in total N
253 and P mass as a function of mass remaining, where N and P are initially net immobilized (i.e.,
254 nutrient mass increases) until reaching a peak when net nutrient mineralization starts (Aber &
255 Melillo, 1982; Berendse et al., 1987; Manzoni et al., 2008). We used the initial and final C
256 content of disks punched from the cotton strips to estimate mass loss and the C:N and C:P ratios
257 to calculate the maximum nutrient mass immobilized (N and P factors), the rate at which N and P
258 were immobilized (N_{IMM} and P_{IMM} , respectively), and the length of time that cotton immobilized
259 nutrients (T_{IMM}). See Supporting Information for details of the calculations (Text S1). Nutrient
260 immobilization rates, factors, and T_{IMM} were log-normally distributed with a positive skew, and
261 thus all rates, factors, ratios, and times were summarized as geometric means (Isles, 2020). It was
262 not possible to calculate N_{IMM} and P_{IMM} when the substrate had no detectable carbon loss, and

263 thus some strips could not generate estimates of immobilization (7 and 15% of the river and
264 riparian strips). Additionally, estimates of T_{IMM} and nutrient factors (but not N_{IMM} and P_{IMM}) for
265 strips showing minimal, although detectable, carbon mass loss were insufficiently precise and
266 hence were also excluded from all analyses (Text S1, Figures S3–4).

267 Nutrient immobilization factors and rates were calculated for individual cotton strips and
268 compared among biomes and latitude using linear mixed models with site as a random effect. For
269 strips that immobilized both N and P (68 rivers and 57 riparian zones), we calculated N:P of
270 immobilization on individual strips and tested for differences among biomes and latitude using
271 linear mixed models. A single decomposition rate (rate of tensile-strength loss, k) was estimated
272 at each river and riparian site; thus, k was correlated with mean immobilization rate from all
273 strips in a river or riparian zone. Mean immobilization rates in rivers were related to measures of
274 water quality using linear regression. Arrhenius plots were used to examine temperature
275 sensitivity of nutrient immobilization rates in riparian and river sites (separate mixed effects
276 models) with directly measured mean daily temperature during the incubation period.
277 Immobilization rates were also regressed against the deviations (i.e., residuals) from the mean
278 rate of tensile-strength loss (Brown et al., 2004), which were calculated from Arrhenius plot
279 best-fit lines using the complete dataset of Tiegs and colleagues (2019). All statistical analyses
280 were completed in R version 4.0.2 (R Core Team, 2020) and mixed models were fit using the
281 lme4 package.

282 **3 Results**

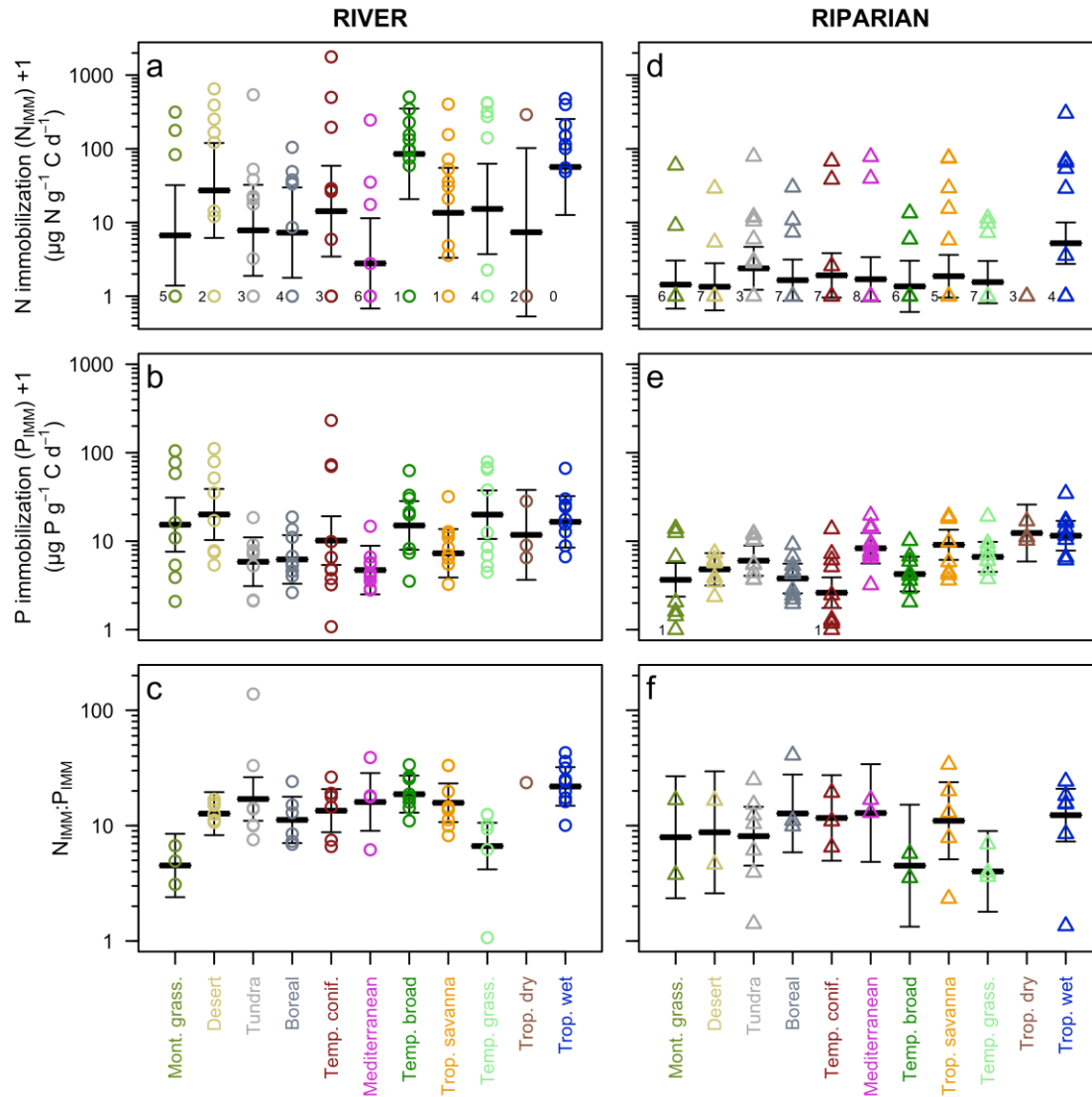
283 In all 99 rivers, the decomposer community immobilized P while breaking down cotton. In
284 contrast, N immobilization was detectable in only 68 rivers. Of the 98 sites where strip mass was
285 lost in riparian zones, each site immobilized P during breakdown, but N was immobilized at only

286 35 sites. Globally, nutrient immobilization was faster in rivers than in riparian zones with the
287 geometric mean (and interquartile ranges (IQR)) of 9.3 (4.4–19.7) and 4.8 (3.1–9.4) $\mu\text{g P g}^{-1} \text{C d}^{-1}$
288 ¹, respectively. N immobilization showed an even larger discrepancy between river (72 $\mu\text{g N g}^{-1}$
289 C d^{-1} ; IQR 32–277) and riparian habitats (16 $\mu\text{g N g}^{-1} \text{C d}^{-1}$; IQR 17–74). On average,
290 decomposers were projected to immobilize P in rivers and riparian zones for 78 and 82 days (i.e.,
291 T_{IMM}), respectively, before net mineralization started. In contrast, N was predicted to be
292 immobilized in rivers and riparian zones for just 58 and 46 days, respectively.

293 3.1 Biome as a predictor of nutrient immobilization

294 Nutrient immobilization among rivers was highly variable, but this variation was only minimally
295 explained by the biome classification. Mean N_{IMM} was greatest in rivers of temperate broadleaf
296 forests (85 $\mu\text{g N g}^{-1} \text{C d}^{-1}$) and least in rivers in mediterranean regions (1.8 $\mu\text{g N g}^{-1} \text{C d}^{-1}$)
297 (Figure 1a), yet biome explained only 13% of the variation in N_{IMM} across all cotton strips
298 (Figure S5). The N factor was similar among biomes, with slightly more variance explained by
299 biome classification (19%, Figure S5). Biome offered slightly more explanatory power for P_{IMM}
300 (19%) than N_{IMM} , with the slowest rates in mediterranean rivers (3.7 $\mu\text{g P g}^{-1} \text{C d}^{-1}$) and
301 significantly faster rates in rivers in desert, temperate grassland, and tropical wet forest biomes
302 (19, 19, 16 $\mu\text{g P g}^{-1} \text{C d}^{-1}$, respectively, Figure 1b). In rivers, P factor exhibited similar biome
303 patterns to P_{IMM} (18% variance explained).

304



305

306 **Figure 1.** Nitrogen and phosphorus immobilization rates (N_{IMM} and P_{IMM} , respectively) and the
 307 molar ratio of the two rates from rivers (a-c) and riparian zones (d-f) in 11 biomes. Black bars
 308 indicate means for biomes $\pm 95\%$ confidence intervals. Rates are shown on a log+1 axis to
 309 include sites where nutrients were not immobilized during cotton decomposition (count of sites
 310 with N_{IMM} or P_{IMM} = 0 are given near x-axis). Biomes are ordered from slowest to fastest mean
 311 decomposition rate in riparian strips (Tiegs et al. 2019).

312

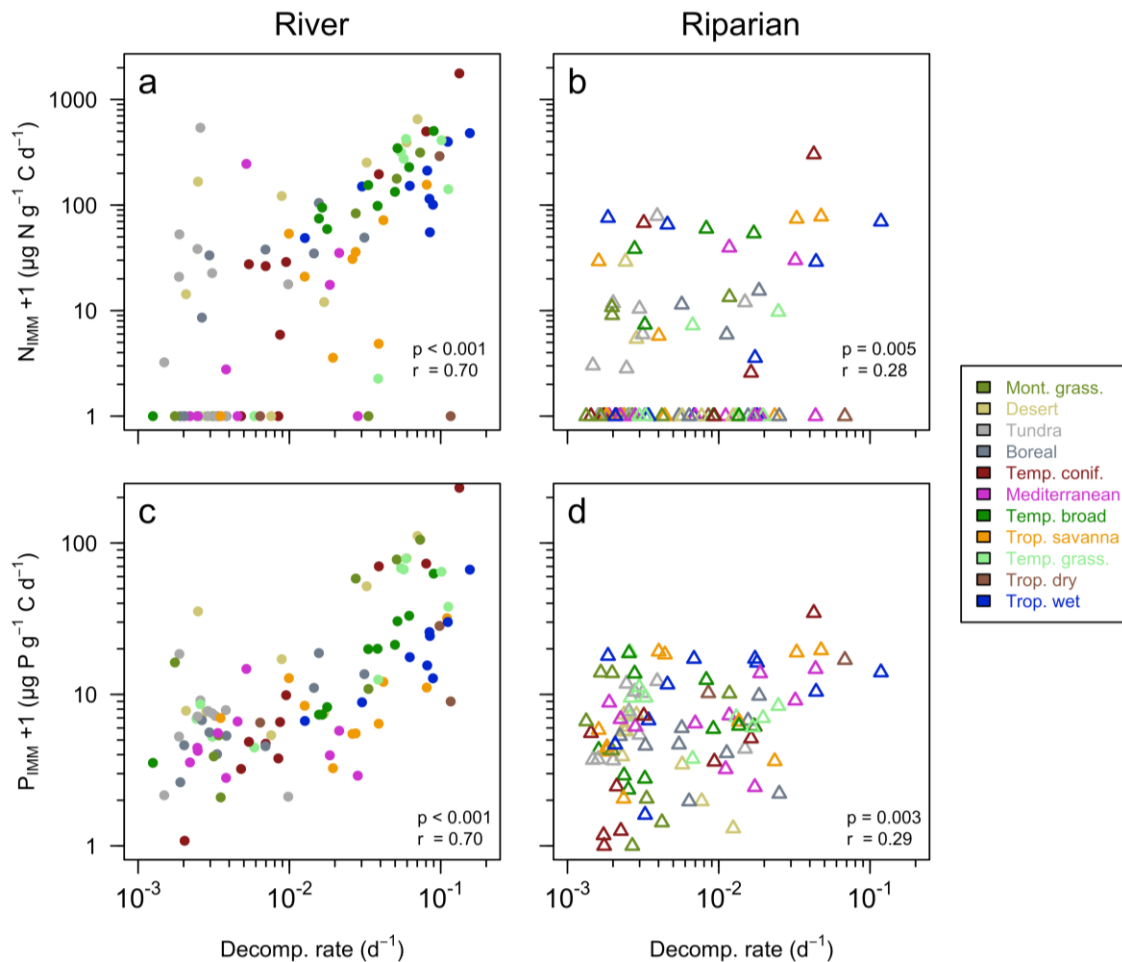
313 In rivers where both N and P were immobilized ($n = 68$), the N:P ratio of immobilization (i.e.,
314 $N_{\text{IMM}}:P_{\text{IMM}}$) was always >1 , with 31% of the variation explained by biome type (Figure 1c,
315 Figure S5). Rivers in montane and temperate grasslands had the lowest $N_{\text{IMM}}:P_{\text{IMM}}$ (5 and 7,
316 respectively), whereas tropical dry, tropical wet, and temperate broadleaf forests had the highest
317 $N_{\text{IMM}}:P_{\text{IMM}}$ (24, 22, and 19 respectively). The N factor:P factor did not differ substantially among
318 biomes (Figure S5), but the relationship between N factor and P factor in log-log space showed a
319 proportional relationship (slope = 1).

320 Nitrogen was not immobilized ($N_{\text{IMM}} = 0$) in 63% of the riparian sites with a null value observed
321 in at least one site from each biome (Figure 1d). Biome type did not predict riparian N_{IMM} , likely
322 due to high within-site variation (Figure S5). On average, the rate of N_{IMM} in riparian zones was
323 $0.9 \mu\text{g N g}^{-1} \text{C d}^{-1}$. The N factor was greatest in tropical wet forests and similar among all other
324 biomes. P immobilization was measured in all but two riparian sites that had detectable mass
325 loss, and 30 and 23% of the variation in P_{IMM} and P factor was explained by biome, respectively.
326 In general, tropical riparian zones had faster rates of P_{IMM} than temperate zones (Figure 1e).
327 Tropical dry and tropical wet forest riparian zones had the fastest rates of P_{IMM} (both $11 \mu\text{g P g}^{-1}$
328 C d^{-1}) and temperate coniferous forests had the slowest ($1.6 \mu\text{g P g}^{-1} \text{C d}^{-1}$). P_{IMM} was only
329 greater than N_{IMM} in the 61 sites where N_{IMM} was 0, but for the riparian zones from sites where
330 N_{IMM} and P_{IMM} were both detectable, rates of N_{IMM} always exceeded P_{IMM} (Figure 1f). Biome
331 was not predictive of $N_{\text{IMM}}:P_{\text{IMM}}$, which averaged 9:1 globally.

332 3.2 Immobilization and cellulose decomposition

333 There was a strong positive association between rates of nutrient immobilization and k in both
334 habitats but the relationship was much stronger in rivers than riparian zones (Figure 2). In rivers
335 and riparian zones, sites with slow decomposition more frequently did not immobilize any N

336 (i.e., $N_{\text{IMM}} = 0$) (Figure 2a & b). In riparian zones, k was weakly associated with N_{IMM} (Figure
 337 2b) but there was a stronger correlation in rivers (Figure 2a). Decomposition was correlated
 338 positively to P_{IMM} , but similar to N , relationships were stronger in rivers (Figure 2c & d).



339

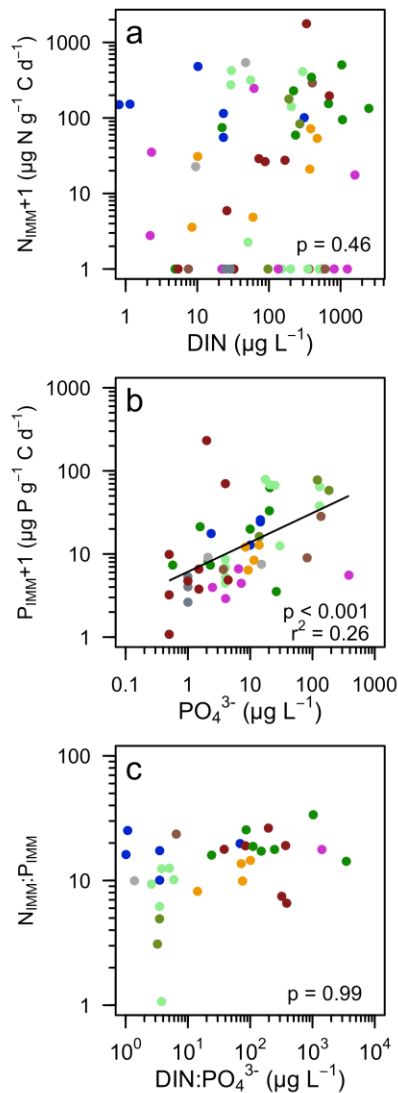
340 **Figure 2.** Decomposition rates are positively associated with nitrogen (a & b) and phosphorus
 341 (c & d) immobilization rates in rivers (a & c) and riparian zones (b & d). Immobilization rates
 342 are on a log+1 axis to include sites where nutrients were not immobilized during
 343 decomposition. Immobilization rates plotted are means of 2-4 cotton strips at each site.
 344 Decomposition rates (loss of tensile strength) were calculated at the site scale with 2-4 cotton
 345 strips (see Tiegs et al. 2019). Symbol colors correspond to different biomes.

346

347 3.3 Water quality and immobilization

348 Some water quality parameters exhibited broad geographic patterns (Text S2) – most notably a
349 negative correlation between PO_4^{3-} and absolute latitude ($p = 0.001$) and greater concentrations
350 of NH_4^+ in temperate grasslands ($p = 0.02$). However, NO_3^- , dissolved inorganic N (DIN), and
351 $\text{DIN}:\text{PO}_4^{3-}$ did not show any geographic patterns. River water PO_4^{3-} was the only variable
352 significantly related to k ($p = 0.002$). There was also a strong relationship between P_{IMM} and
353 PO_4^{3-} concentrations (Figure 3b). In contrast, neither k ($p = 0.27$) nor N_{IMM} ($p = 0.46$) were
354 correlated with DIN (Figure 3a). For instance, we documented rivers with relatively high DIN
355 (i.e., $>100 \mu\text{g L}^{-1}$) that did not immobilize N and rivers with relatively low DIN (i.e., $<10 \mu\text{g L}^{-1}$)
356 where N_{IMM} was in excess of $100 \mu\text{g N g}^{-1} \text{C d}^{-1}$. The lack of a relationship between surface
357 water N and N_{IMM} was also reflected in poor relationships between NH_4^+ and N_{IMM} ($p = 0.68$),
358 surface water $\text{DIN}:\text{PO}_4^{3-}$ and $N_{\text{IMM}}:P_{\text{IMM}}$ (Figure 3c), and $\text{NH}_4^+:\text{PO}_4^{3-}$ and $N_{\text{IMM}}:P_{\text{IMM}}$ ($p = 0.75$).
359 Interestingly, there was a weak positive relationship between N_{IMM} and PO_4^{3-} concentrations ($p =$
360 0.06). Notably, $N_{\text{IMM}}:P_{\text{IMM}}$ were constrained to a relatively narrow range (50% of $N_{\text{IMM}}:P_{\text{IMM}}$
361 between 10 and 20), whereas surface water N:P varied by 3 orders of magnitude (50% of
362 $\text{DIN}:\text{PO}_4^{3-}$ between 4 and 194).

363



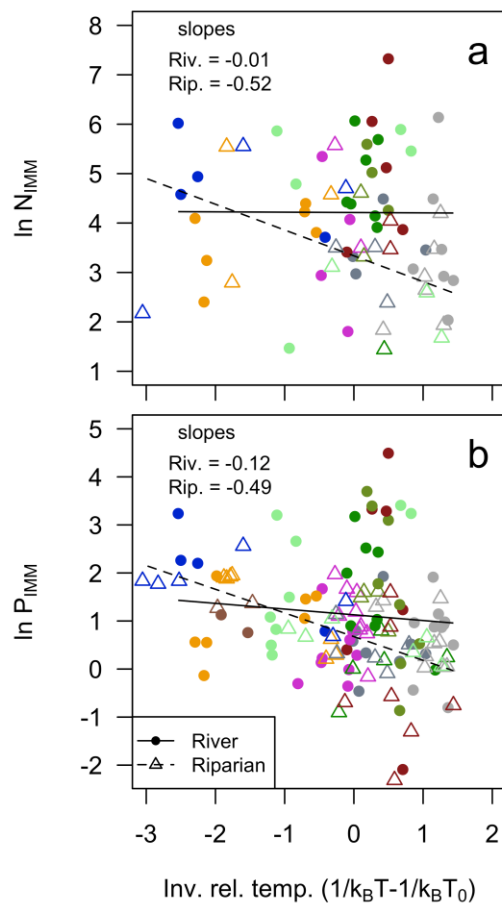
364

365 **Figure 3.** Relationships between ambient surface water nutrients and nutrient immobilization
 366 rates by the microbial community on cotton incubated in rivers. DIN is the sum of ammonium-
 367 N and nitrate-N concentrations. N:P for surface water and immobilization are molar ratios.
 368 Colors denote rivers from different biomes (see legend in Fig. 2).

369 3.4 Temperature and immobilization

370 Arrhenius plots of the entire dataset of all sites ($n = 415$ river and $n = 533$ riparian sites)
 371 indicated strong temperature dependence of k in rivers, but only a weak relationship in riparian
 372 zones (Tiegs et al., 2019). The sites used in the current analysis reflected the same relationships
 373 between k and temperature as the larger dataset (Tiegs et al., 2019, Figure S7), which suggests

374 little bias in our site selection. Nutrient immobilization exhibited strong temperature dependence
 375 in riparian sites (slopes: $N_{\text{IMM}} = -0.52$, $P_{\text{IMM}} = -0.49$), but not in rivers (slopes: $N_{\text{IMM}} = -0.01$,
 376 $P_{\text{IMM}} = -0.12$) (Figure 4). The contrasting temperature effects on riparian and river habitats were
 377 reflected in latitudinal patterns in immobilization (Text S2, Figure S8).

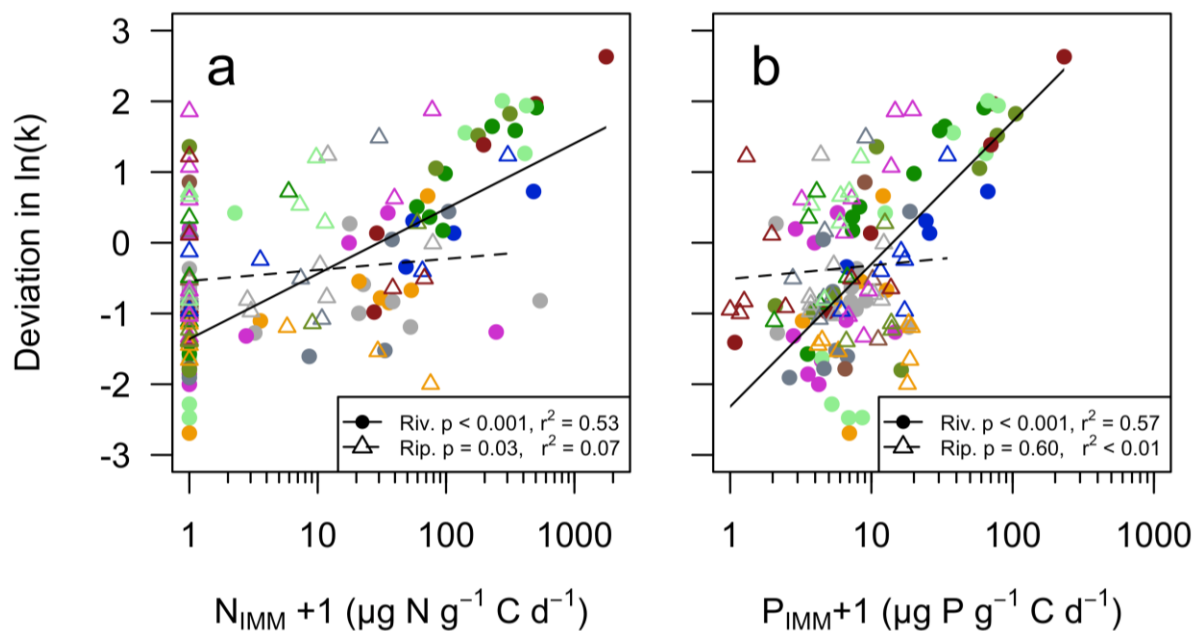


378

379 **Figure 4.** Temperature dependence of nitrogen (a) and phosphorus (b) immobilization on
 380 cotton in rivers and riparian zones. Immobilization in riparian zones increased with increasing
 381 temperature ($p < 0.02$) but immobilization in rivers was not related to temperature ($p > 0.40$).
 382 Colors denote rivers and riparian zones from different biomes (see legend in Fig. 2).

383

384 Collectively, our data indicate that k was sensitive to temperature, especially in rivers, but there
 385 was substantial variation among individual sites with similar temperatures (Figure S7, Tiegs et
 386 al., 2019). Deviations in $\ln(k)$ from the expected value (based on Tiegs et al., 2019) were
 387 strongly related to immobilization rates in rivers, but not in riparian zones (Figure 5). In rivers,
 388 sites with positive deviations from the temperature relationship (i.e., faster k than expected) were
 389 characterized by faster rates of N_{IMM} and P_{IMM} , whereas the opposite was true for sites with
 390 slower k than expected (Figure 5). In contrast, N_{IMM} only weakly explained deviations in $\ln(k)$ in
 391 riparian zones (Figure 5a), particularly when we excluded all sites with $N_{\text{IMM}} = 0$ ($p = 0.24$, $r^2 <$
 392 0.06). Similarly, riparian P_{IMM} did not significantly explain the deviations in $\ln(k)$ from expected
 393 values (Figure 5b).



394

395 **Figure 5.** Relationship between nitrogen (a) and phosphorus (b) immobilization rates on cotton and deviations in decomposition rate from global trends in temperature sensitivity (Tiegs et al.
 396 2019). In rivers, faster rates of nutrient immobilization are related to positive deviation (i.e.,
 397 decomposition rates that are faster than the global average at that temperature). Nutrient
 398 immobilization on cotton in riparian soils was not predictive of deviations in decomposition.
 399

400 **4 Discussion**

401 The microbes on our experimental cotton substrates immobilized N and P during decomposition
402 at rates within the range measured for natural leaf litter. In riparian zones, N_{IMM} on cotton was
403 similar to rates measured on six leaf litter species ($20\text{--}33 \mu\text{g N g}^{-1} \text{C d}^{-1}$; Aber & Melillo, 1982)
404 but lower than those for crop residues in agricultural soils ($462 \mu\text{g N g}^{-1} \text{C d}^{-1}$; Recous et al.,
405 1995). In rivers, N_{IMM} on cotton was within the range of immobilization on natural litter
406 measured either by N accumulation in residual litter (Robbins et al., 2019) or isotopic dilution in
407 microbial pools (Cheever et al., 2013; Pastor et al., 2014). P immobilization on decomposing
408 litter is rarely measured, but our rates of P_{IMM} on cotton were similar to those recorded for three
409 litter types in an artificial stream ($0\text{--}12 \mu\text{g P g}^{-1} \text{C d}^{-1}$; Robbins et al., 2019). Our approach, like
410 that used by most previous studies of nutrient immobilization, could not distinguish assimilation
411 by autotrophs on cotton from immobilization by decomposers, but natural shading of the stream
412 channel likely limited algal contributions to net nutrient uptake (Elosegi et al. 2018; Halvorson et
413 al. 2019).

414 Nutrient immobilization exhibited broad spatial patterns for cellulose decomposing in riparian
415 zones, whereas in rivers, rates were more strongly controlled by local factors. Terrestrial biome,
416 which is a proxy for climate, offered limited explanatory power of immobilization for rivers (13–
417 19%), but explained slightly more variance for immobilization in riparian zones (23–30%).

418 Similarly, latitude was a poor predictor of immobilization with the exception of P_{IMM} in riparian
419 zones, which was slower at high latitudes. We observed an unbalanced influence of temperature
420 and moisture across the riparian–river boundary. Annual precipitation is understandably less of a
421 driver in rivers given that the sites studied were perennial. Conversely, moisture likely influences
422 microbial decomposition and immobilization in riparian zones given their drier environment

423 (Sutfin et al., 2016). Temperature also had greater influence on immobilization in riparian zones
424 than rivers, likely due to differences in the accessibility of exogenous nutrients between habitats.
425 Rivers provide a continuous supply of inorganic nutrients that are accessible to microbial
426 decomposers, and the supply rates influence immobilization. However, riparian zones receive
427 more irregular supply of nutrients, and microbes are reliant on biologically mediated processes
428 (e.g., N fixation, extracellular enzymes) (Allison & Vitousek, 2005; Vitousek & Hobbie, 2000).
429 Conductivity, pH, DOC, and ammonium differed in rivers within some biomes, but none of these
430 variables corresponded with immobilization rates. Although there was a strong latitudinal
431 gradient in PO_4^{3-} in our study sites, there was only a weak, but directionally consistent, negative
432 relationship between latitude and P_{IMM} ($r^2 = 0.03$). Collectively, these results indicate that
433 controls of nutrient immobilization on decomposing litter can differ profoundly between riparian
434 zones and adjacent rivers.

435 We observed strong relationships between rates of cellulose degradation and P_{IMM} , which
436 suggests that P availability is a fundamental control on microbial decomposition of low-nutrient
437 plant litter. Although we did not directly manipulate nutrients, our findings are consistent with
438 experiments that have demonstrated faster decomposition under nutrient enrichment in many
439 biomes (Ferreira et al., 2015). Correlations between immobilization and decomposition were
440 stronger for P than N in both rivers and riparian zones. Enrichment of N and P have both been
441 shown to stimulate microbial decomposers (Ferreira et al., 2015; Rosemond et al., 2015;
442 Woodward et al., 2012), but in some rivers only P enrichment has stimulated decomposition
443 (Burdon et al., 2020; Elwood et al., 1981; Newbold et al., 1983). The substrate used in this study
444 was low in both N and P, but the relative abundance of these two elements (N:P = 62:1)
445 indicated a greater P imbalance relative to microbial decomposers. This relative N enrichment is

446 common for litter; global analyses indicated average litter N:P of 46:1 for all senesced leaves
447 (McGroddy et al., 2004) and 58:1 for riparian litter (Boyero et al., 2017). Therefore, P-limited
448 litter may be a common substrate for microbial decomposers regardless of streamwater nutrient
449 concentrations, and immobilization of exogenous P may be a critical limiting reaction for
450 decomposition. Human-dominated catchments often receive excessive inputs of P from
451 fertilizers or sewage (Birk et al., 2020; Carpenter et al., 1998), and our study provides further
452 evidence that nutrient loading may increase organic matter decomposition in addition to its
453 widely recognized enhancement of plant growth.

454 The relationships we observed between immobilization rates and water quality demonstrate the
455 importance of exogenous sources of N and P for microbial decomposers in rivers globally
456 (Woodward et al., 2012). The strong positive relationship between PO_4^{3-} and P_{IMM} is reasonable
457 given the P-limited substrate. However, future work would be beneficial to understand the
458 relative contribution of heterotrophic assimilation, autotrophic assimilation, and abiotic
459 precipitation of P (potentially stimulated by microbial activity) to our calculated immobilization
460 rates. The lack of relationship between dissolved N and N_{IMM} may be explained by the microbial
461 decomposers being P limited or due to alternative sources of N. We observed river sites with
462 relatively high DIN ($>200 \mu\text{g L}^{-1}$) that immobilized no N during decomposition. In these rivers,
463 P concentrations were low ($\text{PO}_4^{3-} < 5 \mu\text{g L}^{-1}$) and the substrate may have supplied sufficient N to
464 support microbial activity. Alternatively, some rivers had relatively low DIN concentrations (<10
465 $\mu\text{g L}^{-1}$) with high rates of N_{IMM} , which may have been sustained by organic sources of N and/or
466 atmospheric N_2 through N fixation. Finally, the cotton substrate is a potential carbon source for
467 denitrifying bacteria, whose dissimilatory N products (i.e., N gasses) would not be included in
468 N_{IMM} . However, the thin woven structure of the cotton strips and the deployment locations (i.e.,

469 in the water column and at the soil surface) would be unlikely to generate the anoxic sites needed
470 for denitrification.

471 Our data indicate that the stoichiometry of microbial immobilization is influenced by microbial
472 biomass N:P and not availability in exogenous pools or substrate imbalance. When microbes
473 were immobilizing both nutrients, more N was immobilized than P (all $N_{\text{IMM}}:P_{\text{IMM}} > 1$), and the
474 ratio of immobilized nutrients was constrained to a relatively narrow range (mean 10:1) among
475 biomes and between rivers and riparian zones. On average, $N_{\text{IMM}}:P_{\text{IMM}}$ was similar to the global
476 mean N:P of microbial biomass in soil (7:1, Cleveland & Liptzin, 2007) and litter-associated
477 fungal biomass (9:1, Gulis et al., 2017), suggesting the stoichiometry of heterotrophic
478 immobilization is connected to the stoichiometry of microbial biomass. The mean $N_{\text{IMM}}:P_{\text{IMM}}$
479 matched the global average for microbial biomass despite the fact that the litter was more
480 deficient in P than N and many rivers provisioned nutrients at a low N:P (a third of rivers had
481 $\text{DIN}:\text{PO}_4^{3-} < 10:1$). Exogenous N:P in rivers varied widely ($\text{DIN}:\text{PO}_4^{3-}$ between 1:1 and 1000:1),
482 but $N_{\text{IMM}}:P_{\text{IMM}}$ covered a much narrower range. Although bacterial and fungal taxa can be non-
483 homeostatic (Danger & Chauvet, 2013; Godwin & Cotner, 2015) the communities decomposing
484 cotton immobilized nutrients at a relatively fixed N:P across our globally distributed sites. This
485 invariance in N:P at the ecosystem scale is consistent with observations of “ecosystem
486 homeostasis” in bacterial-dominated heterotrophic rivers (Schade et al., 2011) and fixed ratios of
487 exoenzymes in heterotrophic microbes (Hill et al., 2012; Sinsabaugh et al., 2009). Furthermore,
488 the observed $N_{\text{IMM}}:P_{\text{IMM}}$ may represent the averaging of the N:P of the microbial communities
489 that can decompose this cellulose substrate (sensu Klausmeier et al., 2004). Consequently, if

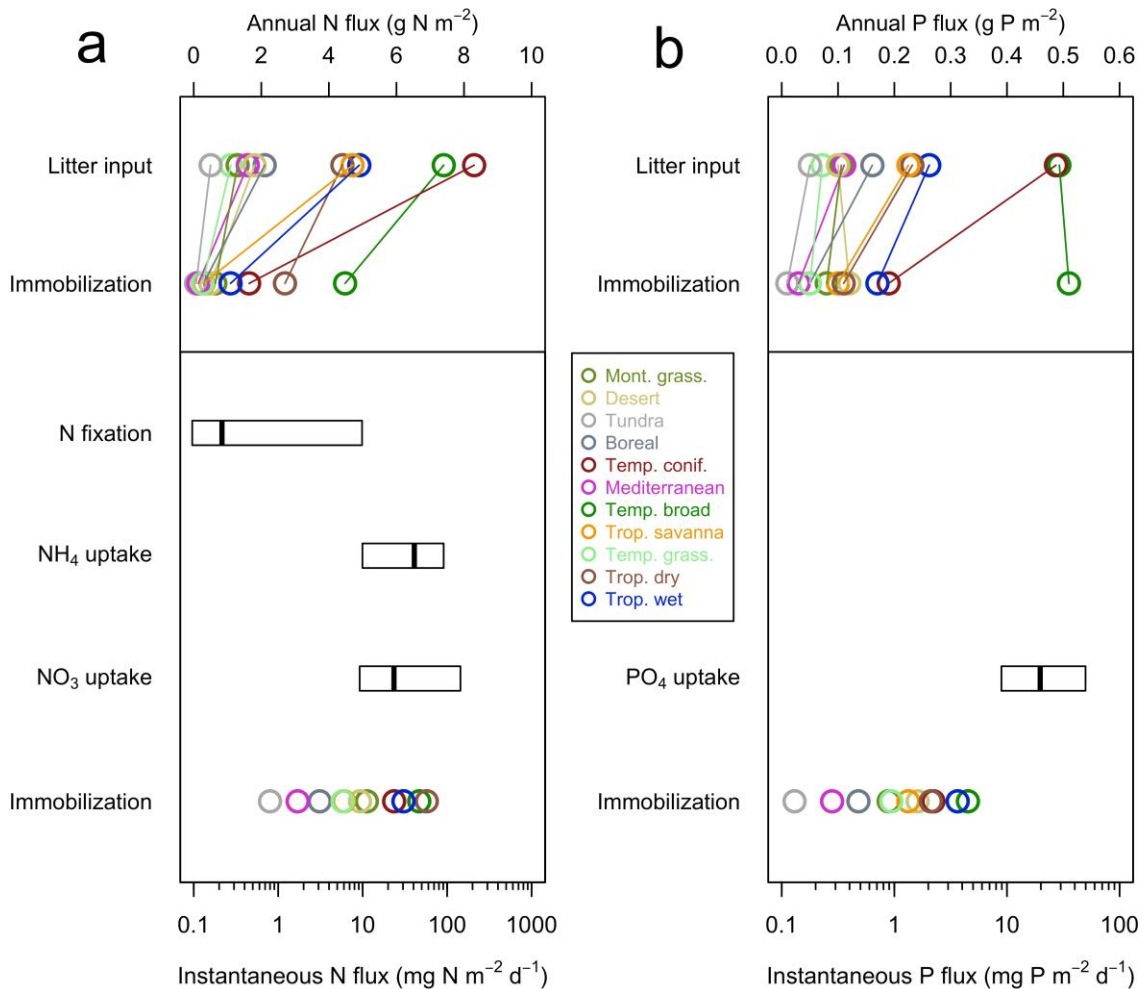
490 microbes are P-limited, increasing PO_4^{3-} through nutrient loading is expected to increase rates of
491 P_{IMM} and N_{IMM} , as microbes immobilize nutrients at a fixed ratio.

492 Although immobilization in rivers was not directly influenced by surface water temperatures,
493 about half of the previously observed residual variation in the temperature dependence of
494 decomposition (Tiegs et al., 2019) was explained by N_{IMM} and P_{IMM} . Thus, while water
495 temperature sets the potential rate of decomposition, the external nutrient supply appears to
496 determine whether microbes achieve the maximum potential rate. In contrast, riparian nutrient
497 immobilization was directly controlled by temperature and thus co-varied with decomposition.
498 Together these observations suggest that warmer temperatures in the future may have contrasting
499 effects on decomposition and immobilization in riparian zones versus rivers. Although terrestrial
500 decomposition is expected to increase with warming, our data indicate that future changes in
501 precipitation may have more of an influence on riparian immobilization and decomposition than
502 warming (Tiegs et al., 2019; Yin et al., 2019). Decomposition was temperature-sensitive in
503 rivers, which supports predictions of faster decomposition under warming conditions (e.g.,
504 Boyero et al., 2011; Follstad Shah et al., 2017; Tiegs et al., 2019), but immobilization did not
505 show strong relationships with temperature. Therefore, warming may not increase decomposition
506 rates if surface water nutrient supply is stable or declining. The interactive effects of warming
507 and eutrophication are well-recognized in autotrophic ecosystems (e.g., Binzer et al., 2016), and
508 here we identified pathways by which these two global stressors may interact in heterotrophic
509 ecosystems.

510 In rivers, leaf litter delivers an important subsidy of C to fuel microbial decomposers, but the N
511 and P in microbial biomass is derived from both the plant litter itself and the environment. To
512 understand the relative contributions of these two processes we made a hypothetical comparison

513 of N and P fluxes that were sourced exclusively from plant litter or solely from the environment
514 (i.e., litter only provides C). For representative ecosystems within each biome (Table S2), we
515 compared expected annual nutrient flux from leaf litter tissue (using models from Boyero et al.,
516 2017) to the flux from nutrient immobilization (using mean P_{IMM} , N_{IMM} , and T_{IMM} for each
517 biome). Across all biomes, leaf litter contributed more N to rivers than microbial immobilization
518 (Figure 6a). At most, immobilized N flux was 60% of the flux expected from leaf litter (in
519 tropical dry and temperate broadleaf forests). For P, it is possible for immobilization to provide a
520 similar or larger annual flux of P to microbial pools than leaf litter (Figure 6b), with larger
521 immobilization fluxes than litter flux possible in deserts and temperate broadleaf forests.
522 Microbial N and P on natural litter is a mix of litter-derived and immobilized nutrients, but this
523 hypothetical analysis demonstrates that the two sources may be comparable, especially for P.
524 However, nutrients supplied through litter may be in relatively recalcitrant organic molecules,
525 whereas immobilized N and P are likely to be in more labile forms. The eventual fate of
526 endogenous and exogenous nutrients involved in litter breakdown has consequences for the long-
527 term fate of nutrients, whether those detrital nutrients fuel riverine communities or are
528 transported downstream.

529



530

531 **Figure 6.** Nitrogen (a) and phosphorus (b) immobilization areal flux in rivers compared to
 532 other major fluxes. Immobilization fluxes are the product of geometric mean rates for a biome
 533 and measured litter fluxes in representative rivers within those biomes (see Table S2 for site
 534 details). “Litter input” shows N and P flux from endogenous nutrients in leaf litter; estimates
 535 are the product of litter flux in representative rivers and N and P content, predicted from mean
 536 annual precipitation (N) and mean annual temperature (P) (Boyero et al. 2017). Instantaneous
 537 N and P flux assume a concentrated pulse of leaf litter (i.e., time of litter fall \ll time of
 538 immobilization) and rates of litter input from representative rivers. NO₃⁻, NH₄⁺, and PO₄³⁻
 539 uptake are whole-stream uptake (U) summarized by boxplots of median (vertical line) and
 540 interquartile range (IQR). Measurement of NH₄⁺ and PO₄³⁻ uptake are from the review by
 541 Ensign and Doyle (2006), and measurement of NO₃⁻ uptake is from Ensign and Doyle (2006)
 542 and the LINX-II study (Hall et al. 2009). N fixation fluxes are summarized as a median and
 543 IQR from Marcarelli et al. (2008).

544 We also examined whether immobilization can account for a large portion of nutrient flux at the
545 ecosystem scale by comparing microbial immobilization fluxes to whole-stream nutrient fluxes.
546 The flux of nutrients due to immobilization only occurs during the seasons of peak litter fall (i.e.,
547 autumn in temperate zones and the dry season in the tropics) and instantaneous rates of
548 immobilization were comparable to whole-stream uptake for N but not P. Instantaneous N
549 immobilization was highest in forest biomes and was similar or higher than whole-stream uptake
550 of NH_4^+ and NO_3^- in many biomes (Figure 6a). Instantaneous N immobilization was higher than
551 N fixation by 1–2 orders of magnitude (Figure 6a). Thus, during peak litterfall, immobilization
552 by microbes may be the dominant flux of N from the water column. Although our estimates
553 showed that P flux from immobilization could be similar to P flux from litter, instantaneous P
554 immobilization was 1–2 orders of magnitude lower than estimated whole-stream PO_4^{3-} uptake
555 (Figure 6b). Riverbeds remove P through both biotic assimilation and abiotic sorption, and
556 microbial immobilization only approaches this flux at very high rates of litter input (e.g.,
557 temperate broadleaf and tropical wet forests). For streams where both N and P uptake were
558 measured, the average N:P ratio of whole-stream uptake ($\text{NH}_4^+:\text{PO}_4^{3-} = 4:1$, $\text{NO}_3^-:\text{PO}_4^{3-} = 3:1$,
559 Ensign & Doyle, 2006) was lower than the average N:P of immobilization (10:1). Furthermore,
560 whole stream uptake $\text{NO}_3^-:\text{PO}_4^{3-}$ was <1 in 25% of streams ($n = 65$), but $\text{N}_{\text{IMM}}:\text{P}_{\text{IMM}} <1$ was
561 never observed in our study. These comparative approaches indicate that the total flux of P from
562 immobilization may be comparable to leaf litter inputs and N immobilization may be an
563 important component of whole-stream uptake during peak litterfall.

564 Local variation in ecosystem characteristics can be as important as global scale factors when
565 considering drivers of decomposition (Bradford et al., 2015, 2017). Most studies emphasize
566 local-scale variation in litter quality as the primary control of decomposition (Bradford et al.,

567 2015; Follstad Shah et al., 2017; LeRoy et al., 2020); yet, we observed substantial local-scale
568 variation in decomposition of a uniform substrate that we linked to differences in nutrient supply
569 and temperature. Moreover, we observed substantial patch-scale differences in the controls and
570 rates of microbial decomposition across river–riparian boundaries within ecosystems. By
571 exploiting natural gradients in nutrient concentrations and climate at a global scale, we identified
572 the direct and indirect pathways by which exogenous nutrient supply and temperature modify
573 microbial processes. Although degradation of cotton strips surely differs from decomposition of
574 plant litter, the broad mechanisms and patterns identified here would presumably apply to any
575 organic substrate with low nutrient content. In particular, the stoichiometric constraints on
576 immobilization in rivers across a broad gradient in nutrient supply and the temperature
577 invariance of immobilization rates among globally distributed rivers are new insights about how
578 nutrient loading and elevated temperatures from climate change may directly influence riverine
579 decomposition.

580 **Acknowledgements**

581 We thank a large number of assistants who helped field work, G. Moeen and M. Moeen for
582 preparing cotton strips, and N. Johnson, A. Minerovic, and C. Blackwood for help with C:N
583 analysis. This research was supported by awards to S.D.T. from the Ecuadorian Ministry of
584 Science (Secretaría de Educación Superior Ciencia, Tecnología e Innovación, SENESCYT)
585 through the PROMETEO scholar-exchange program, the Oakland University Research
586 Development Grant program, and a Huron Mountain Wildlife Foundation research grant. N.A.G.
587 was supported by the U.S. Department of Energy’s Office of Science, Biological and
588 Environmental Research. Oak Ridge National Laboratory is managed by UT-Battelle, LLC, for
589 the U.S. Department of Energy under contract DE-AC05-00OR22725. PRL was supported by

590 NSERC and Canada Research Chair programs. Any use of trade, firm, or product names is for
 591 descriptive purposes only and does not imply endorsement by the U.S. Government.

592 **Data Availability Statement**

593 All data and code for analyses and figures are available on GitHub which can be accessed from
 594 the persistent DOI <https://doi.org/10.5281/zenodo.5764917>.

595 **References**

- 596 Aber, J. D., & Melillo, J. M. (1982). Nitrogen immobilization in decaying hardwood leaf litter as
 597 a function of initial nitrogen and lignin content. *Canadian Journal of Botany*, *60*(11), 2263–
 598 2269. <https://doi.org/10.1139/b82-277>
- 599 Allison, S. D., & Vitousek, P. M. (2005). Responses of extracellular enzymes to simple and
 600 complex nutrient inputs. *Soil Biology and Biochemistry*, *37*(5), 937–944.
 601 <https://doi.org/10.1016/j.soilbio.2004.09.014>
- 602 Berendse, F., Berg, B., & Bosatta, E. (1987). The effect of lignin and nitrogen on the
 603 decomposition of litter in nutrient-poor ecosystems: a theoretical approach. *Canadian*
 604 *Journal of Botany*, *65*(6), 1116–1120. <https://doi.org/10.1139/b87-155>
- 605 Binzer, A., Guill, C., Rall, B. C., & Brose, U. (2016). Interactive effects of warming,
 606 eutrophication and size structure: impacts on biodiversity and food-web structure. *Global*
 607 *Change Biology*, *22*(1), 220–227. <https://doi.org/10.1111/GCB.13086>
- 608 Birk, S., Chapman, D., Carvalho, L., Spears, B. M., Andersen, H. E., Argillier, C., et al. (2020).
 609 Impacts of multiple stressors on freshwater biota across spatial scales and ecosystems.
 610 *Nature Ecology & Evolution 2020 4:8*, *4*(8), 1060–1068. <https://doi.org/10.1038/s41559-020-1216-4>
- 612 Boyero, L., Pearson, R. G., Gessner, M. O., Barmuta, L. A., Ferreira, V., Graça, M. A. S., et al.
 613 (2011). A global experiment suggests climate warming will not accelerate litter
 614 decomposition in streams but might reduce carbon sequestration. *Ecology Letters*, *14*(3),
 615 289–294. <https://doi.org/10.1111/j.1461-0248.2010.01578.x>
- 616 Boyero, L., Graça, M. A. S., Tonin, A. M., Pérez, J., Swafford, A. J., Ferreira, V., et al. (2017).
 617 Riparian plant litter quality increases with latitude. *Scientific Reports*, *7*(1), 1–10.
 618 <https://doi.org/10.1038/s41598-017-10640-3>
- 619 Bradford, M. A., Berg, B., Maynard, D. S., Wieder, W. R., & Wood, S. A. (2015).
 620 Understanding the dominant controls on litter decomposition. *Journal of Ecology*, *104*,
 621 229–238. <https://doi.org/10.1111/1365-2745.12507>

- 622 Bradford, M. A., Veen, G. F., Bonis, A., Bradford, E. M., Classen, A. T., Hans Cornelissen, J.
623 C., et al. (2017). A test of the hierarchical model of litter decomposition. *Nature Ecology &*
624 *Evolution*, 1, 1836. <https://doi.org/10.1038/s41559-017-0367-4>
- 625 Brown, J. H., Gillooly, J. F., Allen, A. P., Savage, V. M., & West, G. B. (2004). Toward a
626 metabolic theory of ecology. *Ecology*, 85(7), 1771–1789.
- 627 Burdon, F. J., Bai, Y., Reyes, M., Tamminen, M., Staudacher, P., Mangold, S., et al. (2020).
628 Stream microbial communities and ecosystem functioning show complex responses to
629 multiple stressors in wastewater. *Global Change Biology*, 26(11), 6363–6382.
630 <https://doi.org/10.1111/gcb.15302>
- 631 Carpenter, S. R., Caraco, N. F., Correll, D. L., Howarth, R. W., Sharpley, A. N., & Smith, V. H.
632 (1998). Nonpoint pollution of surface waters with phosphorus and nitrogen. *Ecological*
633 *Applications*, 8(3), 559–568. [https://doi.org/10.1890/1051-](https://doi.org/10.1890/1051-0761(1998)008[0559:NPOSWW]2.0.CO;2)
634 [0761\(1998\)008\[0559:NPOSWW\]2.0.CO;2](https://doi.org/10.1890/1051-0761(1998)008[0559:NPOSWW]2.0.CO;2)
- 635 Cheever, B. M., Webster, J. R., Bilger, E. E., & Thomas, S. A. (2013). The relative importance
636 of exogenous and substrate-derived nitrogen for microbial growth during leaf
637 decomposition. *Ecology*, 94(7), 1614–1625. <https://doi.org/10.1890/12-1339.1>
- 638 Classen, A. T., Sundqvist, M. K., Henning, J. A., Newman, G. S., Moore, J. A. M., Cregger, M.
639 A., et al. (2015). Direct and indirect effects of climate change on soil microbial and soil
640 microbial-plant interactions: What lies ahead? *Ecosphere*, 6(8).
641 <https://doi.org/10.1890/ES15-00217.1>
- 642 Cleveland, C. C., & Liptzin, D. (2007). C:N:P stoichiometry in soil: Is there a “Redfield ratio”
643 for the microbial biomass? *Biogeochemistry*, 85(3), 235–252.
644 <https://doi.org/10.1007/s10533-007-9132-0>
- 645 Colas, F., Woodward, G., Burdon, F. J., Guérold, F., Chauvet, E., Cornut, J., et al. (2019).
646 Towards a simple global-standard bioassay for a key ecosystem process: organic-matter
647 decomposition using cotton strips. *Ecological Indicators*, 106, 105466.
648 <https://doi.org/10.1016/j.ecolind.2019.105466>
- 649 Danger, M., & Chauvet, E. (2013). Elemental composition and degree of homeostasis of fungi:
650 Are aquatic hyphomycetes more like metazoans, bacteria or plants? *Fungal Ecology*, 6(5),
651 453–457. <https://doi.org/10.1016/j.funeco.2013.05.007>
- 652 Dodds, W. K., Bruckerhoff, L., Batzer, D., Schechner, A., Pennock, C., Renner, E., et al. (2019).
653 The freshwater biome gradient framework: predicting macroscale properties based on
654 latitude, altitude, and precipitation. *Ecosphere*, 10(7), e02786.
655 <https://doi.org/10.1002/ECS2.2786>
- 656 Eloegi, A., Nicolás, A., & Richardson, J. S. (2018). Priming of leaf litter decomposition by
657 algae seems of minor importance in natural streams during autumn. *PLoS ONE*, 13(9), 1–
658 13. <https://doi.org/10.1371/journal.pone.0200180>

- 659 Elwood, J. W., Newbold, J. D., Trimble, A. F., & Stark, R. W. (1981). The limiting role of
660 phosphorus in a woodland stream ecosystem: Effects of P enrichment on leaf decomposition
661 and primary producers. *Ecology*, *62*(1), 146–158. <https://doi.org/10.2307/1936678>
- 662 Ensign, S. H., & Doyle, M. W. (2006). Nutrient spiraling in streams and river networks. *Journal*
663 *of Geophysical Research: Biogeosciences*, *111*(4), 1–13.
664 <https://doi.org/10.1029/2005JG000114>
- 665 Ferreira, V., Castagnyrol, B., Koricheva, J., Gulis, V., Chauvet, E., & Graça, M. A. S. (2015). A
666 meta-analysis of the effects of nutrient enrichment on litter decomposition in streams.
667 *Biological Reviews*, *90*(3), 669–688. <https://doi.org/10.1111/brv.12125>
- 668 Fioretto, A., Di Nardo, C., Papa, S., & Fuggi, A. (2005). Lignin and cellulose degradation and
669 nitrogen dynamics during decomposition of three leaf litter species in a Mediterranean
670 ecosystem. *Soil Biology and Biochemistry*, *37*(6), 1083–1091.
671 <https://doi.org/10.1016/j.soilbio.2004.11.007>
- 672 Follstad Shah, J. J., Kominoski, J. S., Ardon, M., Dodds, W. K., Gessner, M. O., Griffiths, N. A.,
673 et al. (2017). Global synthesis of the temperature sensitivity of leaf litter breakdown in
674 streams and rivers. *Global Change Biology*, *23*(8), 1–12. <https://doi.org/10.1111/gcb.13609>
- 675 Godwin, C. M., & Cotner, J. B. (2015). Aquatic heterotrophic bacteria have highly flexible
676 phosphorus content and biomass stoichiometry. *ISME Journal*, *9*(10), 2324–2327.
677 <https://doi.org/10.1038/ismej.2015.34>
- 678 Gulis, V., Kuehn, K. A., Schoettle, L. N., Leach, D., Benstead, J. P., & Rosemond, A. D. (2017).
679 Changes in nutrient stoichiometry, elemental homeostasis and growth rate of aquatic litter-
680 associated fungi in response to inorganic nutrient supply. *The ISME Journal*, *11*, 2729–
681 2739. <https://doi.org/10.1038/ismej.2017.123>
- 682 Hall, R. O., Tank, J. L., Sobota, D. J., Mulholland, P. J., O'Brien, J. M., Dodds, W. K., et al.
683 (2009). Nitrate removal in stream ecosystems measured by ¹⁵N addition experiments: Total
684 uptake. *Limnology and Oceanography*, *54*(3), 653–665.
685 <https://doi.org/10.4319/LO.2009.54.3.0653>
- 686 Halvorson, H. M., Francoeur, S. N., Findlay, R. H., & Kuehn, K. A. (2019). Algal-mediated
687 priming effects on the ecological stoichiometry of leaf litter decomposition: A meta-
688 analysis. *Frontiers in Earth Science*, *7*(April), 1–12.
689 <https://doi.org/10.3389/feart.2019.00076>
- 690 Hill, B. H., Elonen, C. M., Seifert, L. R., May, A. A., & Tarquinio, E. (2012). Microbial enzyme
691 stoichiometry and nutrient limitation in US streams and rivers. *Ecological Indicators*,
692 *18*(July), 540–551. <https://doi.org/10.1016/j.ecolind.2012.01.007>
- 693 Isles, P. D. F. (2020). The misuse of ratios in ecological stoichiometry. *Ecology*, *101*(11), 1–7.
694 <https://doi.org/10.1002/ecy.3153>

- 695 Klausmeier, C. A., Litchman, E., Daufresne, T., & Levin, S. A. (2004). Optimal nitrogen-to-
 696 phosphorus stoichiometry of phytoplankton. *Nature*, *429*(6988), 171–174.
 697 <https://doi.org/10.1038/nature02454>
- 698 LeRoy, C. J., Hipp, A. L., Lueders, K., Follstad Shah, J. J., Kominoski, J. S., Ardón, M., et al.
 699 (2020). Plant phylogenetic history explains in-stream decomposition at a global scale.
 700 *Journal of Ecology*, *108*(1), 17–35. <https://doi.org/10.1111/1365-2745.13262>
- 701 Manzoni, S., Jackson, R. B., Trofymow, J. A., & Porporato, A. (2008). The global stoichiometry
 702 of litter nitrogen mineralization. *Science*, *321*(5889), 684–686.
 703 <https://doi.org/10.1126/science.1159792>
- 704 Manzoni, S., Trofymow, J. A., Jackson, R. B., & Porporato, A. (2010). Stoichiometric controls
 705 on carbon, nitrogen, and phosphorus dynamics in decomposing litter. *Ecological*
 706 *Monographs*, *80*(1), 89–106. <https://doi.org/10.1890/09-0179.1>
- 707 Marcarelli, A. M., Baker, M. A., & Wurtsbaugh, W. A. (2008). Is in-stream N₂ fixation an
 708 important N source for benthic communities and stream ecosystems? *Journal of the North*
 709 *American Benthological Society*, *27*(1), 186–211. <https://doi.org/10.1899/07-027.1>
- 710 McGroddy, M. E., Daufresne, T., & Hedin, L. O. (2004). Scaling of C:N:P stoichiometry in
 711 forests worldwide: Implications of terrestrial redfield-type ratios. *Ecology*, *85*(9), 2390–
 712 2401. <https://doi.org/10.1890/03-0351>
- 713 Newbold, J. D., Elwood, J. W., Schulze, M. S., Stark, R. W., & Barmeier, J. C. (1983).
 714 Continuous ammonium enrichment of a woodland stream: uptake kinetics, leaf
 715 decomposition, and nitrification. *Freshwater Biology*, *13*(2), 193–204.
 716 <https://doi.org/10.1111/j.1365-2427.1983.tb00671.x>
- 717 Parton, W., Silver, W. L., Burke, I. C., Grassens, L., Harmon, M. E., Currie, W. S., et al. (2007).
 718 Global-scale similarities in nitrogen release patterns during long-term decomposition.
 719 *Science*, *315*, 361–364.
- 720 Pastor, A., Compson, Z. G., Dijkstra, P., Riera, J. L., Martí, E., Sabater, F., et al. (2014). Stream
 721 carbon and nitrogen supplements during leaf litter decomposition: Contrasting patterns for
 722 two foundation species. *Oecologia*, *176*(4), 1111–1121. <https://doi.org/10.1007/s00442-014-3063-y>
- 724 R Core Team. (2020). R: A language and environment for statistical computing. Vienna, Austria:
 725 R Foundation for Statistical Computing. Retrieved from <http://www.r-project.org>
- 726 Recous, S., Robin, D., Darwis, D., & Mary, B. (1995). Soil inorganic N availability: Effect on
 727 maize residue decomposition. *Soil Biology and Biochemistry*, *27*(12), 1529–1538.
 728 [https://doi.org/10.1016/0038-0717\(95\)00096-W](https://doi.org/10.1016/0038-0717(95)00096-W)
- 729 Robbins, C. J., Matthaeus, W. J., Cook, S. C., Housley, L. M., Robison, S. E., Garbarino, M. A.,
 730 et al. (2019). Leaf litter identity alters the timing of lotic nutrient dynamics. *Freshwater*
 731 *Biology*, *64*(12), 2247–2259. <https://doi.org/10.1111/fwb.13410>

- 732 Rosemond, A. D., Benstead, J. P., Bumpers, P. M., Gulis, V., Kominoski, J. S., Manning, D. W.
733 P., et al. (2015). Experimental nutrient additions accelerate terrestrial carbon loss from
734 stream ecosystems. *Science*, *347*(6226), 1142–1145.
735 <https://doi.org/10.1126/science.aaa1958>
- 736 Schade, J. D., Macneill, K., Thomas, S. A., Camille Mcneely, F., Welter, J. R., Hood, J., et al.
737 (2011). The stoichiometry of nitrogen and phosphorus spiralling in heterotrophic and
738 autotrophic streams. *Freshwater Biology*, *56*(3), 424–436. [https://doi.org/10.1111/j.1365-
739 2427.2010.02509.x](https://doi.org/10.1111/j.1365-2427.2010.02509.x)
- 740 Sinsabaugh, R. L., Hill, B. H., & Follstad Shah, J. J. (2009). Ecoenzymatic stoichiometry of
741 microbial organic nutrient acquisition in soil and sediment. *Nature*, *462*(7274), 795–798.
742 <https://doi.org/10.1038/nature08632>
- 743 Sutfin, N. A., Wohl, E. E., & Dwire, K. A. (2016). Banking carbon: A review of organic carbon
744 storage and physical factors influencing retention in floodplains and riparian ecosystems.
745 *Earth Surface Processes and Landforms*, *41*(1), 38–60. <https://doi.org/10.1002/esp.3857>
- 746 Tank, J. L., Rosi-Marshall, E. J., Griffiths, N. A., Entekin, S. A., & Stephen, M. L. (2010). A
747 review of allochthonous organic matter dynamics and metabolism in streams. *Journal of the
748 North American Benthological Society*, *29*(1), 118–146. <https://doi.org/10.1899/08-170.1>
- 749 Tiegs, S. D., Clapcott, J. E., Griffiths, N. A., & Boulton, A. J. (2013). A standardized cotton-strip
750 assay for measuring organic-matter decomposition in streams. *Ecological Indicators*, *32*,
751 131–139. <https://doi.org/10.1016/j.ecolind.2013.03.013>
- 752 Tiegs, S. D., Costello, D. M., Isken, M. W., Woodward, G., McIntyre, P. B., Gessner, M. O., et
753 al. (2019). Global patterns and drivers of ecosystem functioning in rivers and riparian zones.
754 *Science Advances*, *5*(1), eaav0486. <https://doi.org/10.1126/sciadv.aav0486>
- 755 Vitousek, P. M., & Hobbie, S. (2000). Heterotrophic nitrogen fixation in decomposing litter:
756 Patterns and regulation. *Ecology*, *81*(9), 2366–2376. [https://doi.org/10.1890/0012-
757 9658\(2000\)081\[2366:HNFDL\]2.0.CO;2](https://doi.org/10.1890/0012-9658(2000)081[2366:HNFDL]2.0.CO;2)
- 758 Webster, J. R., & Benfield, E. F. (1986). Vascular plant breakdown in freshwater ecosystems.
759 *Annual Review of Ecology and Systematics*, *17*, 567–594.
- 760 Woodward, G., Gessner, M. O., Giller, P. S., Gulis, V., Hladyz, S., Lecerf, A., et al. (2012).
761 Continental-scale effects of nutrient pollution on stream ecosystem functioning. *Science*,
762 *336*(6087), 1438–1440. <https://doi.org/10.1126/science.1219534>
- 763 Yin, R., Eisenhauer, N., Auge, H., Purahong, W., Schmidt, A., & Schädler, M. (2019). Additive
764 effects of experimental climate change and land use on faunal contribution to litter
765 decomposition. *Soil Biology and Biochemistry*, *131*, 141–148.
766 <https://doi.org/10.1016/j.soilbio.2019.01.009>

767 **References from the Supporting Information**

- 768 Benfield, E. F. (1997). Comparison of litterfall input to streams. *Journal of the North American*
769 *Benthological Society*, 16(1), 104–108.
- 770 Mahowald, N., Jickells, T. D., Baker, A. R., Artaxo, P., Benitez-Nelson, C. R., Bergametti, G., et
771 al. (2008). Global distribution of atmospheric phosphorus sources, concentrations and
772 deposition rates, and anthropogenic impacts. *Global Biogeochemical Cycles*, 22(4), 1–19.
773 <https://doi.org/10.1029/2008GB003240>
- 774 Martínez-Yrizar, A., Núñez, S., Miranda, H., & Búrquez, A. (1999). Temporal and spatial
775 variation of litter production in Sonoran Desert communities. *Plant Ecology*, 145(1), 37–48.
776 <https://doi.org/10.1023/A:1009896201047>
- 777 McDowell, R. W., Noble, A., Pletnyakov, P., Haggard, B. E., & Mosley, L. M. (2020). Global
778 mapping of freshwater nutrient enrichment and periphyton growth potential. *Scientific*
779 *Reports*, 10(1), 1–13. <https://doi.org/10.1038/s41598-020-60279-w>
- 780 Olson, D. M., Dinerstein, E., Wikramanayake, E. D., Burgess, N. D., Powell, G. V. N.,
781 Underwood, E. C., et al. (2001). Terrestrial ecoregions of the world: A new map of life on
782 earth. *BioScience*, 51(11), 933–938.
- 783 Pozo, J., González, E., Díez, J. R., Molinero, J., & Elósegui, A. (1997). Inputs of particulate
784 organic matter to streams with different riparian vegetation. *Journal of the North American*
785 *Benthological Society*, 16(3), 602–611. <https://doi.org/10.2307/1468147>
- 786 Rotundo, J. L., & Aguiar, M. R. (2005). Litter effects on plant regeneration in arid lands: a
787 complex balance between seed retention, seed longevity and soil-seed contact. *Journal of*
788 *Ecology*, 93(4), 829–838. <https://doi.org/10.1111/j.1365-2745.2005.01022.x>
- 789 Tonin, A. M., Boyero, L., Bambi, P., Pearson, R. G., Correa-Araneda, F., & Gonçalves Jr., J. F.
790 (2020). High within-stream replication is needed to predict litter fluxes in wet–dry tropical
791 streams. *Freshwater Biology*, 65(4), 688–697. <https://doi.org/10.1111/fwb.13459>
- 792 Velez-Ruiz, A. M., Nevescanin-Moreno, L., Vargas-Terminel, M. L., Flores-Espinoza, A. R.,
793 Álvarez-Yépiz, J. C., & Yépez, E. A. (2020). Data on litterfall production and meteorology
794 at an old-growth tropical dry forest in northwestern Mexico. *Data in Brief*, 31, 105723.
795 <https://doi.org/10.1016/j.dib.2020.105723>
- 796 Vitousek, P. M., Porder, S., Houlton, B. Z., & Chadwick, O. A. (2010). Terrestrial phosphorus
797 limitation: Mechanisms, implications, and nitrogen-phosphorus interactions. *Ecological*
798 *Applications*, 20(1), 5–15. <https://doi.org/10.1890/08-0127.1>
- 799