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### Responses of terrestrial ecosystem phosphorus cycling to nitrogen addition: A meta-analysis

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Responding to reviewer and editor comments:

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#### EDITOR-IN-CHIEF'S COMMENTS TO AUTHORS

*We have now completed the review process on your manuscript 'Responses of terrestrial ecosystem phosphorus cycling to nitrogen additions: a meta-analysis' (Ref. GEB-2016-0359.R1). You will find the reports of the referees, and the comments of the handling editor, Mr. Xiaofeng Xu, appended below. As you will see, there are a number of points that require your attention, but these seem to constitute a minor revision. I invite you to prepare a modified version of your manuscript that addresses the referees' and the editor's concerns. In a cover letter, please explain how you have modified the manuscript in response to each of their comments, preferably point by point. On points where you disagree with the reviewers, or you feel that modification of the original text is unwarranted, please explain why. It is unlikely that we will need to refer the revised version of the manuscript to the referees again, provided that you adequately address the concerns that they raise in their reports. In addition, please make sure that your manuscript conforms with journal style and format, as described on our web site [http://onlinelibrary.wiley.com/journal/10.1111/\(ISSN\)1466-8238/](http://onlinelibrary.wiley.com/journal/10.1111/(ISSN)1466-8238/).*

**Response:** We thank the Editors-in-Chief for the consideration of our manuscript, and giving us another opportunity to improve the manuscript. In this revised version, we have addressed all reviewer's comments, and issues raised by the handling editor, Dr. Xu. We also double-checked to make sure that the manuscript conforms with journal style and format. We hope that the manuscript could be accepted for publication in Global Ecology and Biogeography.

#### EDITOR'S COMMENTS TO AUTHORS

Editor: Xu, Xiaofeng

Comments to the Author:

*Both reviewers appraised the work, and both agreed that the paper is publishable. And R2 had some minor comments on writing. I would like to see a little bit advanced discussion on the implication of this study to ongoing phosphorus modeling. The current writing with one sentence at the end of the manuscript is too vague; everyone is saying that. I would like to encourage the*

This is the author manuscript accepted for publication and has undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the [Version record](#). Please cite this article as [doi:10.1111/geb.12576](https://doi.org/10.1111/geb.12576).

*authors to take this opportunity to finalize the manuscript, fixing small pitfalls, improving writings, further raising the quality of this paper.*

**Response:** We thank Dr. Xiaofeng Xu for promptly handling our manuscript and for his thoughtful comments. We also appreciate the review comments from two anonymous reviewers. In this version, we expanded our discussion on P-modeling by adding more descriptions of P pools and processes, and linking some of our results to model simulations (Pages 16-17, Lines 438-463). We also fixed all minor errors and improved the writing (see details below). Hope these revisions are adequate. The following is our revised discussion on P-modeling:

“Given the importance of P on plant growth and productivity, inclusion of P cycling in ecosystem models could significantly improve our ability to better forecast ecosystem C sequestration in the future. Recently, P modules have been built into ecosystem models such as CASA-CNP (Wang *et al.*, 2010), JSBACH-CNP (Goll *et al.*, 2012), CLM-CNP (Yang *et al.*, 2014), and N-COM (Zhu *et al.*, 2016) to investigate nutrient limitations on C cycling and C-N-P interactions. In these models, P is usually stored in several pools including plant biomass, litter, plant available P forms in the soils, and unavailable organic P forms in the soils (Smil, 2000; Vitousek *et al.*, 2010). Major P processes include P uptake, mineralization, immobilization, dissolution, precipitation, occlusion, and leaching. P dynamics are often coupled with C and/or N cycling through stoichiometric relationship of C, N and P in plant tissues and soils (Wang *et al.*, 2010; Goll *et al.*, 2012; Yang *et al.*, 2014). The results of enhanced plant P uptake and P mineralization with N additions in this meta-analysis would disproportionately alter P in different pools in the models, and ultimately result in greater P limitation. Our results of stimulated plant biomass and P contents in plant tissues with N additions were also supported by some model simulations. For example, Yang *et al.* (2014) found that, using the CLM-CNP model, plant production is substantially increased with N addition at the N-limited site, resulting in higher P uptake and P contents in plant biomass, considering that C:P ratios for plant tissues are kept constant in the model. But the model showed no impact with N additions at P-limited site indicating the interaction of N and P could be complicated by field nutrient availability. The influences of N additions on the P pools revealed in this study should help us understand the roles that N and P play in controlling plant growth and internal nutrient accumulation and accurately parameterize these C-N-P models. However, more nutrient addition experiments will benefit model testing and calibration (Reed *et al.* 2015; Achat *et al.*, 2016a).”

We hope that this revision adequately addressed Dr/ Xu's concerns with P modeling.

#### REVIEWER COMMENTS TO AUTHOR

Referee: 1

Comments to the Author

*The authors have satisfactorily addressed the comments from last time.*

**Response:** We thank the reviewer for the positive comments.

Referee: 2

Comments to the Author

*This is my second review of the submitted study.*

*As a whole, authors have modified their manuscript following reviewers' comments. I find this second version much better than the former version, and I think it is now acceptable for publication.*

*I have only one problem to report (which could be fixed easily), a few minor corrections, and some non-mandatory suggestions.*

*My personal congratulations to the authors; I believe they produced a good job.*

**Response:** We thank the reviewer for the very positive comments and constructive suggestions.

*\* Problem:*

*The caption of Figure 2 contains information on three panels. But only two panels are in the document. This is probably an error.*

**Response:** Thanks for pointing this out. We removed one panel as it has a small sample size. We corrected this in the revised manuscript.

*\* Minor points:*

*- line 11, page 1: "Meta" and not "Mate".*

*- line 37: replace "8.4%" by "8%".*

*- line 170: put a comma after "NH4F" because there are four categories.*

*- line 191: please limit the number of references (1 or 2).*

- line 375: replace “may” by “likely”.
- line 427: “a major challenge for us, if N deposition continue to increase in the future”.
- Figure 6: indicate in the caption the pH values defining “alkaline”, “neutral”, and “acidic”.
- Appendix S2: You put  $\text{NH}_4\text{NO}_3$  with an exponent.

**Response:** We corrected all the above issues as suggested. Thanks.

\* *Suggestions:*

- lines 122-123: you can omit “with no restriction on publication year”.

**Response:** We deleted this part.

- line 324: I think it would be better to write “Our results demonstrated that external N can significantly alter terrestrial [etc.]”.

**Response:** We revised as suggested.

- line 438: A review paper has just been published on this topic (Achat et al. Future challenges in coupled C–N–P cycle models for terrestrial ecosystems under global change: a review. *Biogeochemistry, Online First*).

**Response:** We thank the reviewer for the reference. We added this reference here and cited it in other places in the manuscript.

- lines 440-449: I think that this paragraph could be placed between line 427 and line 428. It would enable finishing the manuscript by lines 436-439, which is a good “end message”.

**Response:** We moved this paragraph as suggested.

- Figures 3 and 5 could be moved to SI in order to give more importance to other figures (which are more interesting for most readers).

**Response:** We moved these two figures to SI, and also revised the abstract (Page 3, Lines 43-46) accordingly in order to demonstrate the core results for the readers. Thanks.

1 **Responses of terrestrial ecosystem phosphorus cycling**  
2 **to nitrogen additions: a meta-analysis**

3  
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5  
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10

11 **Type of paper:** [Meta-analysis](#)

12 **Key words:** Available phosphorus, meta-analysis, nitrogen additions, phosphatase  
13 activity, phosphorus limitation, total phosphorus.

14 **Running title:** Impact of N additions on P cycle

15

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19

20 **Number of words, table and figures:** 295 words in the Summary with 6 key words,  
21 5464 words in the main text (776 words in Introduction, 1718 words in Materials and  
22 methods, 863 words in Results, 2053 words in Discussion, and 54 words in  
23 Acknowledgments), 64 references, 6 figures in this manuscript, 4 tables and 4 figures  
24 in the Supporting Information.

25

26 **ABSTRACT**

27 **Aim** Anthropogenic nitrogen (N) additions are expected to drive terrestrial  
28 ecosystems toward greater phosphorus (P) limitation. However, a comprehensive  
29 understanding of how ecosystem P cycle responds to external N inputs remains  
30 elusive, making model predictions of the anthropogenic P limitation and its impacts  
31 largely uncertain.

32 **Location** Global.

33 **Methods** We conducted a meta-analysis with 288 independent study sites from 192  
34 articles to evaluate global patterns and controls of 10 variables associated ecosystem P  
35 cycling under N additions.

36 **Results** Overall, N additions increased biomass in plant (+34%) and litter (+15%),  
37 and plant P content (+17%), while decreasing plant and litter P concentrations (-8%  
38 and -11%). N additions did not change soil labile P and microbial P, but enhanced  
39 phosphatase activity (+24%). Effects of N additions on litter P pool and soil total P  
40 remained unclear due to significant publication biases. The response of P cycling to N  
41 additions in tropical forests was different from those in other ecosystem types. N  
42 additions did not change plant biomass or phosphatase activity in tropical forests, but  
43 significantly reduced plant P and soil labile P concentrations. Shift in plant P  
44 concentration under N additions was negatively correlated with N application rate or  
45 total N load. [N-induced change in soil labile P was strongly regulated by soil pH value](#)  
46 [at the control sites, with significant decrease of 14% only in acidic soils \(pH<5.5\).](#)

47 **Main conclusions** Our results suggest that, as anthropogenic N enhancement  
48 continues in the future, it could induce P limitation in terrestrial ecosystems while  
49 accelerating P cycling, particularly in the tropical forests. A quantitative framework  
50 generated based on this meta-analysis is useful for our understanding of ecosystem P  
51 cycling with N additions, and for incorporating the anthropogenic P limitation into  
52 ecosystem models used to analyze effects of future climate change.

53

54 **INTRODUCTION**

55 Globally, nitrogen (N) and phosphorus (P) are the most limited nutrients for plant  
56 growth (Elser *et al.*, 2007; Vitousek *et al.*, 2010), and their synergistic interactions are  
57 widespread as plants require elements in relatively constant proportions to catalyze  
58 metabolic reactions and synthesize essential compounds with specific ratios of N:P  
59 (Ågren, 2008; Harpole *et al.*, 2011; Yuan & Chen, 2015a). Terrestrial biogeochemical  
60 models have explicitly considered such interactions between the N and P cycles  
61 (Zhang *et al.*, 2011; Goll *et al.*, 2012; Zhu *et al.*, 2016), and suggested that elevated  
62 inputs of either N or P may have been implicated in massive shifts in nutrient cycle  
63 and balance, and in turn influence ecosystem productivity and functioning (Wang *et*  
64 *al.*, 2010; Goll *et al.*, 2012; Peñuelas *et al.*, 2013).

65 Due to fossil fuel combustion and intensive application of N-based fertilizers, the  
66 inputs of reactive N to the Earth's land surface have mostly been doubled during the  
67 past century (Gruber & Galloway, 2008; IPCC, 2013). Unlike N, P cycling is almost  
68 unidirectional moving from terrestrial ecosystems to rivers and streams with minor  
69 input back through dust and fly ash from wildfires and negligible gaseous P  
70 (phosphine, PH<sub>3</sub>) (Smil, 2000; Filippelli, 2002; Mahowald *et al.*, 2008; Peñuelas *et al.*,  
71 2012). The widespread enrichment in N inputs has considerably improved regional  
72 and global N availability and enhanced ecosystem productivity (e.g., Aber *et al.*, 1998;  
73 Elser *et al.*, 2007; Xia *et al.*, 2008; Lu *et al.*, 2011a). However, the human-induced  
74 imbalance of N and P inputs is also expected to drive terrestrial ecosystems toward  
75 greater P limitation of plant growth (Vitousek *et al.*, 2010; Peñuelas *et al.*, 2013). This  
76 'anthropogenic P limitation' has been acknowledged recently by a meta-analysis of  
77 impacts of P additions, either alone or with N additions, on plant biomass (Li *et al.*,  
78 2016).

79 However, our understanding of the response of ecosystem P cycling to N  
80 additions and the underlying mechanism is very limited. So far, only the impact of N  
81 additions on phosphatase activity is synthesized, which suggests that N inputs



82 enhance rather than reduce plant P acquisition due to stimulating both plant root and  
83 soil phosphatase activity (Marklein & Houlton, 2012). Ecosystem P cycling consists  
84 of multiple pools and processes that are interrelated and interdependent, jointly  
85 controlling soil P availability and plant P uptake (Filippelli, 2002; Vitousek *et al.*,  
86 2010). The limited synthesis in P process significantly limits our ability in  
87 incorporating the impact of anthropogenic P limitation into ecosystem models used to  
88 analyze effects of possible future climate change on global ecosystem productivity  
89 (Goll *et al.*, 2012; Peñuelas *et al.*, 2013; Zhu *et al.*, 2016). To improve our  
90 understanding of P cycling and develop robust ecosystem models with fully coupled  
91 C-N-P interactions, we urgently need a comprehensive synthesis of ecosystem P  
92 cycling in response to N additions.

93 Since N and P play important roles in plant growth and productivity, numerous  
94 studies have been conducted to examine the plant growth and ecosystem P cycling  
95 with N additions. But the conclusions remain controversial (Finzi, 2009; Braun *et al.*,  
96 2010; Tao & Hunter, 2012). For instance, Li *et al.* (2016) suggested that plant P  
97 concentration is the best indicator of P limitations based on a meta-analysis of P  
98 additions experiments. Yet the field experiments with N additions showed decreases  
99 (Tessier & Raynal, 2003), no change (Weand *et al.*, 2010), and even increases (Liu *et*  
100 *al.*, 2013) in plant P concentration. This is not surprising, as multiple factors and  
101 processes regulate ecosystem P cycling (Filippelli, 2002; Vitousek *et al.*, 2010). As an  
102 example, ecosystem P cycling may respond to N additions differently among different  
103 ecosystems, as it is well known that tropical forest is usually more P limited than  
104 other ecosystems (Elser *et al.*, 2007; Yang *et al.*, 2014). Therefore, a better  
105 understanding of ecosystem P cycling under N additions and the influencing factors is  
106 essentially required for enabling ecosystem models to accurately predict  
107 “anthropogenic P limitation” and its impacts.

108 The objective of this study was to quantitatively evaluate N-induced changes in  
109 ecosystem P cycling and their potentially influencing factors. We compiled a large  
110 dataset of changes in ecosystem P cycling related to 10 variables at 288 independent

111 study sites from 192 articles, and conducted a comprehensive meta-analysis. The  
112 specific issues addressed here were as follows: 1) what ecosystem P variables were  
113 changed under N additions; 2) whether response of ecosystem P cycling to N  
114 additions would vary among different ecosystem types; 3) whether the responses were  
115 also affected significantly by other factors such as climate (temperature and  
116 precipitation), soil properties (soil pH) and N experimental manipulation (N sources,  
117 N application rate, experimental duration and total N load).

## 118 MATERIALS AND METHODS

### 119 Dataset assembly

120 We searched journal articles that reported ecosystem P dynamics in response to N  
121 additions using Google Scholar and ISI Web of Science. Searches included  
122 combinations of the terms “nitrogen addition”, “nitrogen fertilization” or “nitrogen  
123 input”, and “phosphorus” or “phosphatase”. We also screened previous meta-analysis  
124 on the similar topic such as Marklein & Houlton (2012) and Li *et al.* (2016). The  
125 former focused on phosphatase activity responses only, and the latter mainly assessed  
126 the impacts of P additions either alone or with N additions, rather than the direct N  
127 additions impacts. We systematically reviewed all results from the searched articles  
128 and included those studies that reported at least one of the selected P variables (A list  
129 of the data sources is given in Appendix 1 and the relevant information is shown in  
130 Appendix S1 in the Supporting Information) at both the N treatment and control sites.  
131 To avoid possible confounding factors caused by human disturbances, we only  
132 included studies from field manipulative experiments of N additions that were  
133 conducted in natural terrestrial ecosystems. For studies that included additional  
134 treatments such as elevated CO<sub>2</sub>, warming and rainfall changes, we only included the  
135 unmanipulated controls and the corresponding N treatment sites in order to avoid  
136 possible interactive effects. Finally, our searches yielded a total of 192 useful articles  
137 (A list of the data sources is given in Appendix 1), and the compiled database included  
138 10 variables related to ecosystems P cycling and other associated parameters

139 (Appendix S1 in the Supporting Information). Only 10% and 13% of these articles in  
140 this meta-analysis were reviewed in Marklein & Houlton (2012) and Li *et al.* (2016),  
141 respectively.

142 Most of the data were either obtained from tables or extracted from figures using  
143 the GetData Graph Digitizer (version 2.24, Russian Federation). Only a few plant and  
144 litter P contents were calculated using their biomass and P concentrations. For the  
145 plant biomass, different estimating methods (e.g. direct harvest for most herbaceous  
146 species and use of allometric relationships for some tree species) were accepted, but  
147 results from other proxy variables such as plant height, diameter or volume were not  
148 included in our biomass analysis. Plant tissues included foliar, above- (e.g., stem,  
149 shoot or above-ground), below- (coarse root, fine root or total root) and whole  
150 biomass. Phosphatase activity included plant root phosphatase activity and soil  
151 phosphatase activity. Soil P variables were used to represent both their concentrations  
152 and pool sizes, because very few studies reported soil bulk density at both the control  
153 and N treatment sites. It should be noted that N additions may not significantly alter  
154 soil bulk density, as showed in a previous meta-analysis (Lu *et al.*, 2011b).

155 We also collected other site specific information, including source of the data,  
156 location (latitude and longitude), ecosystem type, N source applied, N application rate,  
157 experimental duration, soil type, organic C, pH value, mean annual temperature  
158 (MAT), and mean annual precipitation (MAP) at each study site. Ecosystem types  
159 included tropical forest (36), temperate forest (52), boreal forest (12), grassland (88),  
160 wetland (87) and tundra (13). N sources were divided into Urea-N,  $\text{NH}_4\text{NO}_3\text{-N}$ ,  
161  $\text{NH}_4^+\text{-N}$  and  $\text{NO}_3^-\text{-N}$ . Soil pH values below 5.5 and above 7.5 could limit P  
162 availability to plants due to binding of phosphate ions with aluminum (Zn), iron (Fe),  
163 calcium (Ca) or magnesium (Mg) (Busman *et al.*, 2002; Devau *et al.*, 2009). Thus, we  
164 assigned alkaline (pH > 7.5), acidic (pH < 5.5), or neutral (pH 5.5-7.5) based on pH of  
165 the control site. We recorded soil depth and P extraction methods that were consistent  
166 between the control and treatment sites in all articles. The extraction methods of soil  
167 total P were divided into sequential (Hedley procedure),  $\text{HNO}_3$ ,  $\text{H}_2\text{SO}_4$ , and others

168 (e.g., fusion method). The extraction methods of soil labile P were divided into  
169 sequential, bicarbonate,  $\text{NH}_4\text{F}$ , and others (e.g., *Mehlich 1* and *Mehlich 3* methods).

170 In this study, we focused on the responses of P variables at a community level. To  
171 meet the statistical requirement of independence among observations (Koricheva &  
172 Gurevitch, 2014), we used one set of data that includes only one paired observations  
173 (control and treatment) for each of 10 variables at a given study site (Treseder, 2008).  
174 When plants were measured for multiple tissues at a study site, we used the data from  
175 the tissue that contains more observations in the corresponding variable. Similarly,  
176 when soils were sampled at multiple depths at a study site, we used the data from the  
177 top soil layer. For multiple measurements over time or year, or data presented in  
178 multiple publications at a study site, we selected the studies including the most P  
179 variables, otherwise the data from the latest sampling date were used. At sites when  
180 multiple N application rates were used, only the highest application rate was selected.  
181 It should be noted that measurements at different geographical locations, ecosystem  
182 types (including dominant vegetation), or N sources applied in a study were  
183 considered as independent observation. Finally, a total of 288 independent study sites  
184 were used in this meta-analysis. However, we included all data when the specific  
185 factor was the interest of the study, for example, all soil depths data at each  
186 independent study site were included when the effect of soil depth was examined.

### 187 **Statistical analysis**

188 We quantified the effects of N additions on the ecosystems P cycling by calculating  
189 the natural log of the response ratio (*RR*), a metric commonly used in meta-analyses  
190 (Hedges & Olkin 1985; Luo *et al.*, 2006):

$$191 \quad RR = \ln\left(\frac{\bar{X}_t}{\bar{X}_c}\right) = \ln(\bar{X}_t) - \ln(\bar{X}_c) \quad \text{Eqn 1}$$

192 where *RR* is the ratio of the mean value of the chosen variable in the N treatment  
193 group ( $\bar{X}_t$ ) to that in the control group ( $\bar{X}_c$ ), an index of the effect of the experimental

194 treatment on the target variable. To determine if N additions had a significant effect,  
195 we applied a random-effects model, and considered study site as the random effect  
196 factor (Hedges & Vevea, 1998; Borenstein *et al.*, 2010). Unlike the fixed-effect model  
197 with an assumption that all studies share a common *RR*, the random-effects model  
198 allows the studies to differ in their *RRs* (Borenstein *et al.*, 2010). Thus, there are two  
199 sources of variance in the analysis, including within-study variance ( $v$ ) and  
200 between-studies variance ( $\tau^2$ ) that both were used as the weighting factor ( $w =$   
201  $\frac{1}{v + \tau^2}$ ) to calculate the mean *RRs* and 95% confidence intervals (*CI*s). We firstly  
202 computed the mean  $v$  and *RR*, similar to the approach used in the fixed-effect model  
203 (Luo *et al.*, 2006; Deng *et al.*, 2015), and then used these mean values as an estimate  
204 of the  $\tau^2$ . When  $\tau^2$  is equal to zero, both fixed-effect and random-effects models are  
205 same. The computational details are given in Borenstein *et al.* (2010). The treatment  
206 effect of N additions was considered to be significant if the 95% *CI* of mean *RRs* did  
207 not overlap with zero (Luo *et al.*, 2006; Deng *et al.*, 2015). The mean *RRs* and 95%  
208 *CI*s were then transformed back (i.e. exponentially transformed) and converted to a  
209 percentage change.

210 We further evaluated if the mean *RRs* was affected by the potentially influencing  
211 factors that include categorical factors (ecosystem type, N sources applied, and soil  
212 pH value for each of the selected P variables, plant tissue for plant P variables, P  
213 extraction method and maximum soil depth for soil P variables) and continuous  
214 factors (experimental duration, N application rate, total N load, MAT, and MAP for  
215 each of the selected P variables). As data sets were not complete for all factors, the  
216 statistical significance of each factor was tested independently from the others. For the  
217 categorical factor, a separate mean *RR* and 95% *CI* for each category group were  
218 calculated based on the random-effects model described above. These mean *RRs* were  
219 then compared across groups for each categorical factor using an approach analogous  
220 to one-way weighted ANOVA, where total variability ( $Q_{\text{total}}$ ) was partitioned into  
221 within and between-group variability ( $Q_W$  and  $Q_B$ , respectively) (Paul *et al.*, 2005;  
222 Borenstein *et al.*, 2010). The statistical significances of the categorical factors were

223 evaluated based on a chi-square test of the between-group sum of squares and the  
224 degree of freedom (df). Accordingly, if the 95% *CI* of one group is not overlapped  
225 with another one within a categorical factor, there is a significant difference between  
226 these two groups (Luo *et al.*, 2006; Deng *et al.*, 2015). To avoid unreliable results,  
227 some subgroups without a sufficiently large dataset ( $n < 5$ ) were excluded in the  
228 categorical analysis. For the continuous factors ( $n > 20$  only), we used a weighed  
229 regression-type model to fit the *RRs* to each continuous factor. The weight-*w* was used  
230 as the regression weight in the analysis. The regression parameters (slope and  
231 intercept) were estimated using the method of moments, and their statistical  
232 significances were tested based on an F-test (Lipsey & Wilson, 2001; Paul *et al.*, 2005;  
233 SAS Institute Inc., 2015).

234 The publication bias was tested by the funnel plot method (Egger *et al.*, 1997).  
235 The funnel plot offers a visual sense of the relationship between effect size (*RR*) and  
236 precision (the reverse of standard error of the *RR*). The symmetrical funnel shape of  
237 the plot around the mean *RR* indicates the publication bias in the dataset is  
238 insignificant (Egger *et al.*, 1997). However, the interpretation of the funnel plot is  
239 largely subjective (Borenstein, 2005). Given a small dataset such as  $n < 5$ , it is difficult  
240 or almost impossible to determine if the funnel plot is symmetrical. Thus, in the case  
241 of which the mean *RR* had significant difference from zero, we further calculated the  
242 fail-safe number by a weighted method to estimate whether our conclusion is affected  
243 by the non-published data (Rosenberg, 2005). If the fail-safe number was over  $5n + 10$   
244 (*n* is the number of cases in the analysis), we made conclusion that our result was  
245 strong against publication bias. Otherwise, significant publication bias for the analysis  
246 was suggested. In addition, we assessed the sensitivity of our meta-analysis to the  
247 individual study site. We removed one study site from the database each time, and  
248 estimated the mean *RR* (Philibert *et al.*, 2012; Deng *et al.*, 2015).

249 All data were analyzed using SAS software (SAS Institute Inc., Cary, NC, USA),  
250 and the statistical results were considered to be significant at  $\alpha = 0.05$  level. The  
251 graphs were drawn with SigmaPlot software (SigmaPlot 12.5 for windows; Systat

252 Software Inc., San Jose, CA, USA).

## 253 RESULTS

254 Both plant and litter biomass were significantly increased under N additions (Fig. 1a,  
255 b). On average, plant biomass was increased by 34% (95% *CI*: +27%, +41%) across  
256 the study sites (Fig. 1a), with greater increases in above-ground (+40%; 95% *CI*:  
257 +31%, +48%) than below-ground (+11%; 95% *CI*: +0.6%, +24%) biomass (Fig. 2a).  
258 Litter biomass was increased by 15% (95% *CI*: +8%, +23%) across the study sites  
259 (Fig. 1b).

260 Plant and litter P contents were also significantly increased under N additions  
261 (Fig. 1a, b), but significant publication bias for the result of litter P content was  
262 suggested by both funnel plot and Rosenthal's fail-safe number (Appendix S2 and Fig.  
263 S1 in the Supporting Information). Plant P content was increased by 17% (95% *CI*:  
264 +11%, +23%) across study sites (Fig. 1a), with similar increases in different plant  
265 tissues (Appendix S3 in the Supporting Information).

266 For the plant and litter P concentrations, both were significantly decreased under  
267 N additions (Fig. 1a, b). Plant P concentration was decreased by 8% (95% *CI*: -10%,  
268 -6.6%) across study sites (Fig. 1a). N additions significantly decreased P  
269 concentration by 6.3% (95% *CI*: -8.4%, -4.1%) in foliar and by 9.0% (95% *CI*: -12%,  
270 -6.1%) in above-ground tissue (Fig. 2b), but not in below-ground tissue (-1.8%; 95%  
271 *CI*: -4.8%, +1.3%) (Fig 2c). For leaf/shoot litter, N addition significantly decreased its  
272 P concentration by an average of 8.9% (95% *CI*: -13%, -4.0%) across study sites (Fig.  
273 1b).

274 Phosphatase activity was significantly increased by 24 % (95% *CI*: +13%, +35%)  
275 under N additions (Fig. 1c), with greater increase in plant roots (+62%; 95% *CI*:  
276 +37%, +91%) than in the soil (+16%; 95% *CI*: +5.4%, +37%) (Fig. S3). Total soil P  
277 was slightly decreased under N additions by an average of only 5.1% (95% *CI*: -10%,  
278 -0.5%) across study sites, while both funnel plot and Rosenthal's fail-safe number

279 suggested a significant publication bias for this result (Appendix S2 and Fig. S1 in the  
280 Supporting Information). There was no significant change in soil labile P (-3.0%; 95%  
281 *CI*: -11%, +5.5%) and soil microbial P (+0.4%; 95% *CI*: -1.6%, +2.5%) under N  
282 additions (Fig. 1c).

283 N-induced changes in plant biomass and P concentration varied significantly  
284 among different ecosystem types (Appendix S3 in the Supporting Information). Under  
285 N additions, plant biomass was increased by an average of 69% (95% *CI*: +45%,  
286 +98%) in temperate forest, by 33% (95% *CI*: +24%, +44%) in grassland and by 37%  
287 (95% *CI*: +28%, +48%) in wetland (Fig. 3a). For tropical forest, no significant change  
288 in plant biomass was estimated under N additions (Fig. 3a). Plant P concentration was  
289 decreased by 3.9% (95% *CI*: -4.7%, -1.2%) in tropical forest, by 11% (95% *CI*: -15%,  
290 -5.9%) in temperate forest, by 11% (95% *CI*: -14%, -8.4%) in grassland and by 8.5%  
291 (95% *CI*: -12%, -5.4%) in wetland (Fig. 3b). In tundra or boreal forest, plant P  
292 concentration did not significantly change under N additions (Fig. 3b). The response  
293 of phosphatase activity and soil labile P to N additions also significantly varied among  
294 the ecosystem types (Appendix S3 in the Supporting Information). Under N additions,  
295 phosphatase activity was increased by 22.3% (95% *CI*: +3.9%, +44%) in temperate  
296 forest, by 40% (95% *CI*: +24%, +57%) in grassland, and by 43% (95% *CI*: +8.7%,  
297 +88%) in wetland (Fig. 3c). Among the ecosystem types, only in tropical forest soil  
298 labile P was significantly reduced by 24% (95% *CI*: -33%, -13%) under N additions  
299 (Fig. 3d).

300 Different N sources had significant effects on P concentrations for both plant and  
301 litter in responses to N additions (Appendix S3 in the Supporting Information). N  
302 application with mono sources ( $\text{NH}_4^+\text{-N}$  and  $\text{NO}_3^-\text{-N}$ ) generally caused greater  
303 decreases in plant and litter P concentrations compared with N application with mixed  
304 sources (Urea-N and  $\text{NH}_4\text{NO}_3\text{-N}$ ) (Fig. S4). The response of soil labile P to N  
305 additions was likely regulated by soil pH value at the control site, with significant  
306 decrease of 14% (95% *CI*: -30%, -9.4%) only in acidic soils (Fig. 4a). Different P  
307 extraction methods had minor effect on soil total P in response to N addition (Table S3



308 in the Supporting Information), while the response of soil labile P significantly varied  
309 with P extraction methods (Appendix S3 in the Supporting Information). Among P  
310 extraction methods, only with the  $\text{NH}_4\text{F}$  method soil labile P was significantly  
311 reduced by 18% under N additions (Fig. 4b). Soil P variables did not significantly  
312 change with soil depth probably due to very few studies that sampled soil in multiple  
313 depths (Appendix S4 in the Supporting Information).

314 As the annual rate of N additions or total N load increased, the log response ratio  
315 of P concentration in plant and litter decreased across all plant tissues and ecosystems  
316 (Appendix S4 in the Supporting Information; Fig. 5a-d). However, the latter (N load)  
317 relationships might be partly caused by the fact that N application rate and N load  
318 were highly correlated to each other, as experiment duration had no effects on plant  
319 and litter P concentrations (Appendix S4 in the Supporting Information). For all  
320 variables, no significant effect was found in soil type, soil organic C, and climatic  
321 factors (MAT and MAP) (Appendix S4 in the Supporting Information).

## 322 **DISCUSSION**

323 [Our results demonstrated that external N inputs can significantly alter terrestrial](#)  
324 [ecosystem P cycling across multiple ecosystems](#). Overall, N additions stimulated P  
325 sequestration in both plant and litter biomass (Fig. 1). The net P accumulation in  
326 plants and litter may be primarily attributed to the increased biomass production, as  
327 more P is required to maintain faster plant growth under N additions (Ågren, 2008;  
328 Harpole *et al.*, 2011; Yuan & Chen, 2015a). Consistent with previous meta-analysis  
329 studies (e.g. Aber *et al.*, 1998; Xia & Wan, 2008; Liu & Greaver, 2010), N additions  
330 increased more biomass in the above-ground than below-ground tissues (Fig. 2a),  
331 while they stimulated P sequestration equally (Appendix S3 in in the Supporting  
332 Information). It appears that N additions did not alter P allocation pattern in plant  
333 tissues.

334 The greater uptake of P and consequently sequestration in biomass and litter

335 under N additions (Fig. 1a, b) may result in a significant decrease in soil P, as P inputs  
336 are generally very low in natural ecosystems (Smil, 2000; Ilg *et al.*, 2009; Vitousek *et*  
337 *al.*, 2010). In our meta-analysis, N additions only slightly decreased soil total P and  
338 this result suffered from significant publication bias (Fig. 1c, Appendix S2 and Fig.  
339 S1 in in the Supporting Information). This is probably due to the dearth of long-term  
340 experiments ( $\leq 11$  years) for soil total P in our dataset (Appendix S1 in in the  
341 Supporting Information). Given a much large P pool in soil than that in vegetation, it  
342 is difficult to detect a significant depletion in soil total P in a short-term (Vitousek *et*  
343 *al.*, 2010). Soil labile P fraction should be more sensitive to depletion than total P due  
344 to faster P uptake. However, our meta-analysis shows no change in soil labile P under  
345 N additions (Fig. 1c). This suggests that N additions likely promotes P mobilization,  
346 which is supported by the increase in phosphatase activity our and a previous  
347 meta-analysis showed (Fig. 1c; Marklein & Houlton, 2012). To meet greater P  
348 demands, trees may invest more C and other resources in root exudates and microbial  
349 symbioses that degrade clay minerals or organic P compounds (Chen *et al.*, 2008). In  
350 addition, the accumulation and recycling of P in litter may also account for the no  
351 change in soil labile P under N additions (Fig. 1b). P may be rapidly recycled  
352 internally through reabsorption and litter decomposition (Yuan & Chen, 2015b).

353 The decreased plant P concentration under N additions may not be simply related  
354 to decreasing P availability, since N additions increased phosphatase activity and did  
355 not change soil labile P in our meta-analysis (Fig. 1c). Alternatively, it may be due to  
356 the massive imbalance in soil availability of N and P with more N inputs, which  
357 stimulate plant to uptake more N but limit P uptake. We know that leaf photosynthesis  
358 rate is often positively correlated with foliar N and P concentrations (e.g., Reich *et al.*,  
359 2010). Thus, the decrease in foliar P concentration observed in this study (Fig. 2b)  
360 suggests that N additions might actually induce P limitation that would have negative  
361 feedback on plant growth in response to the increased foliar N concentration under N  
362 additions. Li *et al.* (2016) also demonstrated that N enrichment aggravated P  
363 limitation on biomass production, and suggested that plant P concentration is the best

364 indicator of these P limitations. P concentration in below-ground tissue was not  
365 changed under N additions, indicating that roots may be less affected by the reduced P  
366 availability compared to other plant tissues (Fig. 2b). However, the relatively smaller  
367 biomass gained in below-ground components may also contribute to this unchanged P  
368 concentration (Fig. 2a).

369 Although the very few study sites in certain ecosystems such as tundra or boreal  
370 forest limited us to fully evaluate the effect of ecosystem types on the response of P  
371 cycling to N additions (Appendix S3 in the Supporting Information), our  
372 meta-analysis clearly demonstrated that the responses in some P variables may be  
373 quite different between the tropical forest and other ecosystem types (Fig. 3). This  
374 suggests that the magnitudes of anthropogenic P limitation as well as the underlying  
375 mechanisms likely vary among these ecosystems. For example, we found that N  
376 additions significantly decreased plant P concentration and also soil labile P in the  
377 tropical forests (Fig. 3b, d). The combined decreases in plant P concentration and soil  
378 labile P indicates that N additions may induce greater P limitation to the tropical  
379 forests than other terrestrial ecosystems. In addition, plant growth in the tropical  
380 forests is less limited by N, due to that litter decomposition is fast and N availability is  
381 high (Cusack *et al.*, 2010). The greater P limitation likely offsets the slightly positive  
382 effect of N additions on plant growth in the tropical forests. As a result, our  
383 meta-analysis shows no significant response of plant biomass to N additions in the  
384 tropical forests (Fig. 3a).

385 Two processes may help explain the greater magnitudes of anthropogenic P  
386 limitation in the tropical forests. First, N additions can exacerbate soil acidification in  
387 the tropical forest (Tian & Niu, 2015). This exacerbated acidification may directly  
388 decrease soil P availability in the tropical forest by binding of phosphate ions with Al  
389 and Fe (Busman *et al.*, 2002; Devau *et al.*, 2009), as the tropical soils are often highly  
390 acidic (Vitousek *et al.*, 2010; Lu *et al.*, 2014). This was supported by that N additions  
391 significantly decreased soil labile P only in acidic soils (Fig. 5a). Second, the  
392 exacerbated acidification in the tropical forests may also restrain microbial activity

393 and hence phosphatase activity. In fact, we found that N additions did not stimulate  
394 phosphatase activity in the tropical forests (Fig. 3c).

395 Our analysis suggests a significant effect of P extraction methods on the response  
396 of soil labile P to N additions (Fig. 4b; Appendix S3 in the Supporting Information).  
397 The use of different P extraction methods usually depends on the soil types sampled.  
398 For example, bicarbonate extractions are commonly used in soils with higher pH,  
399 while  $\text{NH}_4\text{F}$  extractions are primarily used in acidic soils where soil labile P tends to  
400 decrease under N additions. There may also be a covariance with geography, as  $\text{NH}_4\text{F}$   
401 method is mainly used in North America and Oceania, but much less used in Europe  
402 or South America. Thus, the significant effect of the extraction method is likely due to  
403 a combination of methodological artifacts and covariance with physically significant  
404 soil properties.

405 There was a significant effect of N sources on the response of plant P  
406 concentration (Appendix S3 in the Supporting Information). Moreover, N additions  
407 with mono sources ( $\text{NH}_4^+\text{-N}$  and  $\text{NO}_3^-\text{-N}$ ) generally caused greater decreases in plant  
408 P concentration compared with mixed sources (Urea-N and  $\text{NH}_4\text{NO}_3\text{-N}$ ) (Fig. S4a).  
409 This suggests that other elements contained in  $\text{NH}_4^+\text{-N}$  or  $\text{NO}_3^-\text{-N}$  fertilizer regulate P  
410 availability or plant P uptake. For example, calcium nitrate additions may release  
411 calcium ion to bind with phosphate ions, in turn decreasing P availability in the soils  
412 (Devau *et al.*, 2009; Vitousek *et al.*, 2010). Thus, identity of any associated ions in N  
413 deposition in future studies is much important for accurate assessment of the impact  
414 of P limitation.

415 It is well understood that N application rate and total N load would have  
416 significant effects on the response of ecosystem P cycling as more N inputs would  
417 cause greater imbalance of N:P ratio. However, the quantification of their  
418 relationships remains challenging due to different background levels of soil N status  
419 and N deposition across study sites. For example, the tropic area generally  
420 experiences high N deposition in the past several decades, which may have

421 contributed to a high N loading in the tropical forests (Huang *et al.*, 2012a). In these  
422 tropical forests, P cycling is very sensitive to N additions and even small N inputs  
423 may cause massive alterations in P cycling (Vitousek *et al.*, 1993; Huang *et al.*,  
424 2012b). Despite such confounding factors, we did detect negative relationships of the  
425 change in plant P concentration with both N application rate and total N load (Fig. 5a,  
426 c). These findings indicate that the P limitation in terrestrial ecosystems could become  
427 a major challenge for us, if N deposition continues to increase in the future.

428 As with any meta-analysis, our results reflect uncertainty and assumptions in  
429 these case studies. For example, other soil properties such as soil texture, organic C,  
430 Al-Fe oxides and clay minerals may influence the dynamics of P availability (Yang &  
431 Post, 2011; Augusto *et al.*, 2013; Achat *et al.*, 2016b; Gerard, 2016). However, due to  
432 the limitation of data availability, we could not derive some clear relationships  
433 between them here (Appendix S3 and S4 in the Supporting Information). In addition,  
434 the dearth of long-term N manipulative experiments (90% studies of experimental  
435 duration are within 10 years) may limit us to obtain clearer trends of ecosystem P  
436 cycling in response to N additions. Thus, more long-term experiments are needed to  
437 observe P limitation with external N inputs.

438 Given the importance of P on plant growth and productivity, inclusion of P  
439 cycling in ecosystem models could significantly improve our ability to better forecast  
440 ecosystem C sequestration in the future. Recently, P modules have been built into  
441 ecosystem models such as CASA-CNP (Wang *et al.*, 2010), JSBACH-CNP (Goll *et al.*,  
442 2012), CLM-CNP (Yang *et al.*, 2014), and N-COM (Zhu *et al.*, 2016) to investigate  
443 nutrient limitations on C cycling and C-N-P interactions. In these models, P is  
444 usually stored in several pools including plant biomass, litter, plant available P forms  
445 in the soils, and unavailable organic P forms in the soils (Smil, 2000; Vitousek *et al.*,  
446 2010). Major P processes include P uptake, mineralization, immobilization,  
447 dissolution, precipitation, occlusion, and leaching. P dynamics are often coupled with  
448 C and/or N cycling through stoichiometric relationship of C, N and P in plant tissues  
449 and soils (Wang *et al.*, 2010; Goll *et al.*, 2012; Yang *et al.*, 2014). The results of

450 enhanced plant P uptake and P mineralization with N additions in this meta-analysis  
451 would disproportionately alter P in different pools in the models, and ultimately result  
452 in greater P limitation. Our results of stimulated plant biomass and P contents in plant  
453 tissues with N additions were also supported by some model simulations. For example,  
454 Yang et al. (2014) found that, using the CLM-CNP model, plant production is  
455 substantially increased with N addition at the N-limited site, resulting in higher P  
456 uptake and P contents in plant biomass, considering that C:P ratios for plant tissues  
457 are kept constant in the model. But the model showed no impact with N additions at  
458 P-limited site, indicating the interaction of N and P could be complicated by field  
459 nutrient availability. The influences of N additions on the P pools and processes  
460 revealed in this study could help us understand the roles that N and P play in  
461 controlling plant growth and internal nutrient accumulation and accurately  
462 parameterize these C-N-P models. However, more nutrient addition experiments will  
463 benefit model testing and calibration (Reed et al. 2015; Achat *et al.*, 2016a).

464 To concluded, this study, to the best of our knowledge, is the first comprehensive  
465 evaluation of global response of the ecosystem P cycling to the external N inputs (Fig.  
466 6). Previous synthesis has indicated that N inputs can accelerate terrestrial ecosystem  
467 P cycling due to stimulating phosphatase activity (Marklein & Houlton, 2012). Our  
468 results demonstrate that N additions accelerated P cycling but still decreased plant P  
469 concentration, and might eventually lead to P limitation in terrestrial ecosystems,  
470 particularly in tropical forests where both plant P concentration and soil labile P were  
471 reduced. In addition to the effect of faster plant growth and greater P demand,  
472 alterations in soil environment such as soil acidification and chemical reaction  
473 between phosphate ions and other associated ions in N fertilizers could also be a  
474 major cause of shift toward P limitation under N additions. Moreover, the P limitation  
475 could become more profound as total N load increases. The effects of continuous  
476 anthropogenic N enrichment on ecosystem structure and function will likely be  
477 constrained by the P availability in the future. To improve the prediction of the  
478 potential impacts of anthropogenic P limitation in ecosystem modeling, ecosystem

479 type, soil properties, previous and future N loads, as well as other associated ions in N  
480 deposition must be considered (Fig. 6).

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681

682 **APPENDIX 1: DATA SOURCES**

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198

## 199 SUPPORTING INFORMATION

200 Additional Supporting Information may be found in the online version of this article:

201 **Appendix S1.** Studies included in the meta-analysis.

202 **Appendix S2.** Rosenthal's fail-safe number used to test the publication bias.

203 **Appendix S3.** Statistical significances of effects of categorical factors.

204 **Appendix S4.** Statistical significances of relationships between the response ratio and  
205 continuous factors.

206 **Fig. S1.** Funnel plots of the effect size plotted against the precision for each P  
207 variable.

208 **Fig. S2.** Sensitivity analysis for each P variable.

209 **Fig. S3.** Percentage change ( $\times 100$ ) of phosphatase activity in plant root and soil under  
210 N additions.

211 **Fig. S4.** Percentage change ( $\times 100$ ) of plant P concentration (a) and litter P  
212 concentration (b) to N additions with different N sources.

213

214 **FIGURE LEGENDS**

215 **Fig. 1.** Percentage change ( $\times 100$ ) of ecosystem P cycling to N additions. Error bars  
216 represent 95% bootstrap confidence intervals. The sample size for each  
217 variable is shown next to the error bar. \*: significant publication bias for the  
218 analysis suggested by both funnel plot and Rosenthal's fail-safe number.

219 **Fig. 2.** Percentage change ( $\times 100$ ) of **plant biomass (a)** and **plant P concentration (b)** in  
220 different plant tissues. Error bars represent 95% confidence intervals. The  
221 sample size for each variable is shown next to the error bar. Statistical  
222 significances of effects of plant tissue were evaluated based on a chi-square  
223 test (also see Appendix S3 in the Supporting Information).

224 **Fig. 3.** Percentage change ( $\times 100$ ) of plant biomass (a), plant P concentration (b),  
225 phosphatase activity (c), and soil labile P (d) to N additions in different  
226 ecosystem types. Error bars represent 95% confidence intervals. The sample  
227 size for each variable is shown next to the error bar. Statistical significances of  
228 effects of ecosystem type were evaluated based on a chi-square test (also see  
229 Appendix S3 in the Supporting Information).

230 **Fig. 4.** Percentage change ( $\times 100$ ) of soil labile P with different soil pH value (a) and  
231 with different P extraction methods (b). Error bars represent 95% confidence  
232 intervals. The sample size for each variable is shown next to the error bar.  
233 **Alkaline: pH > 7.5; acidic: pH < 5.5; and neutral: pH = 5.5-7.5.** Statistical  
234 significances of effects of P extraction method were evaluated based on a  
235 chi-square test (also see Appendix S3 in the Supporting Information).

236 **Fig. 5.** Log response ratios of plant P concentration (a, c) and litter P concentration (b,  
237 d) as a function of N application rate and total N load, and Log response ratios  
238 of soil labile P as a function of control soil pH value. Statistical significances  
239 of relationships were evaluated based on an F-test (also see Appendix S4 in the



240 Supporting Information).

241 **Fig. 6.** A quantitative framework for the responses of ecosystem P cycling to N  
242 additions. +: positive; -: negative; n.s.: non-significant; #: data for soil microbe  
243 and pH from Treseder (2008) and Tian & Niu (2015), respectively. \*:  
244 significant publication bias for the analysis suggested by both funnel plot and  
245 Rosenthal's fail-safe number.

Accepted Article

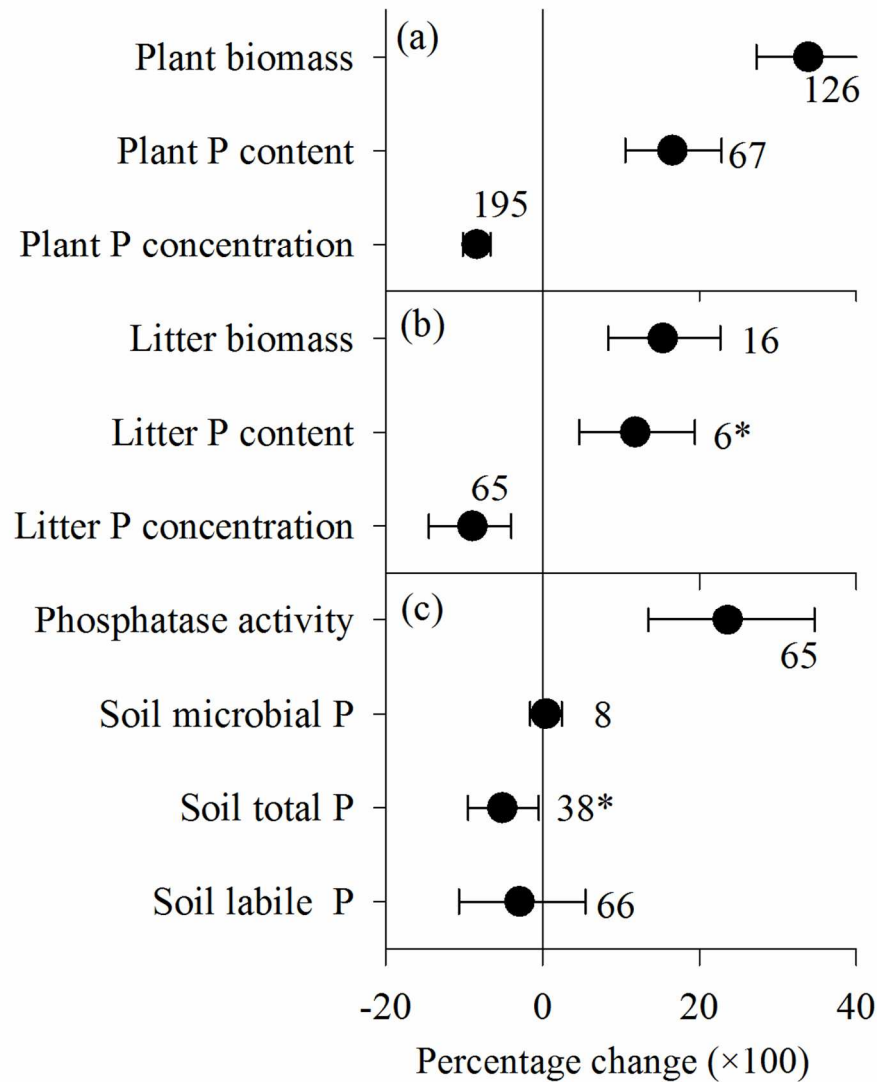


Fig 1. Percentage change ( $\times 100$ ) of ecosystem P cycling to N additions. Error bars represent 95% bootstrap confidence intervals. The sample size for each variable is shown next to the error bar. \*: significant publication bias for the analysis suggested by both funnel plot and Rosenthal's fail-safe number.

Fig. 1

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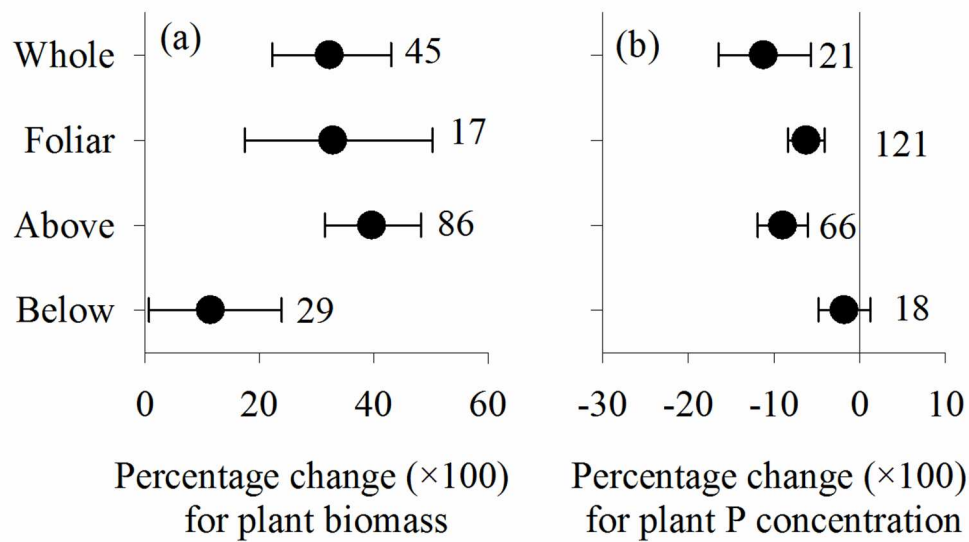


Fig. 2. Percentage change ( $\times 100$ ) of plant biomass (a) and plant P concentration (b) in different plant tissues. Error bars represent 95% confidence intervals. The sample size for each variable is shown next to the error bar. Statistical significances of effects of plant tissue were evaluated based on a chi-square test (also see Appendix S3 in the Supporting Information).

Fig. 2

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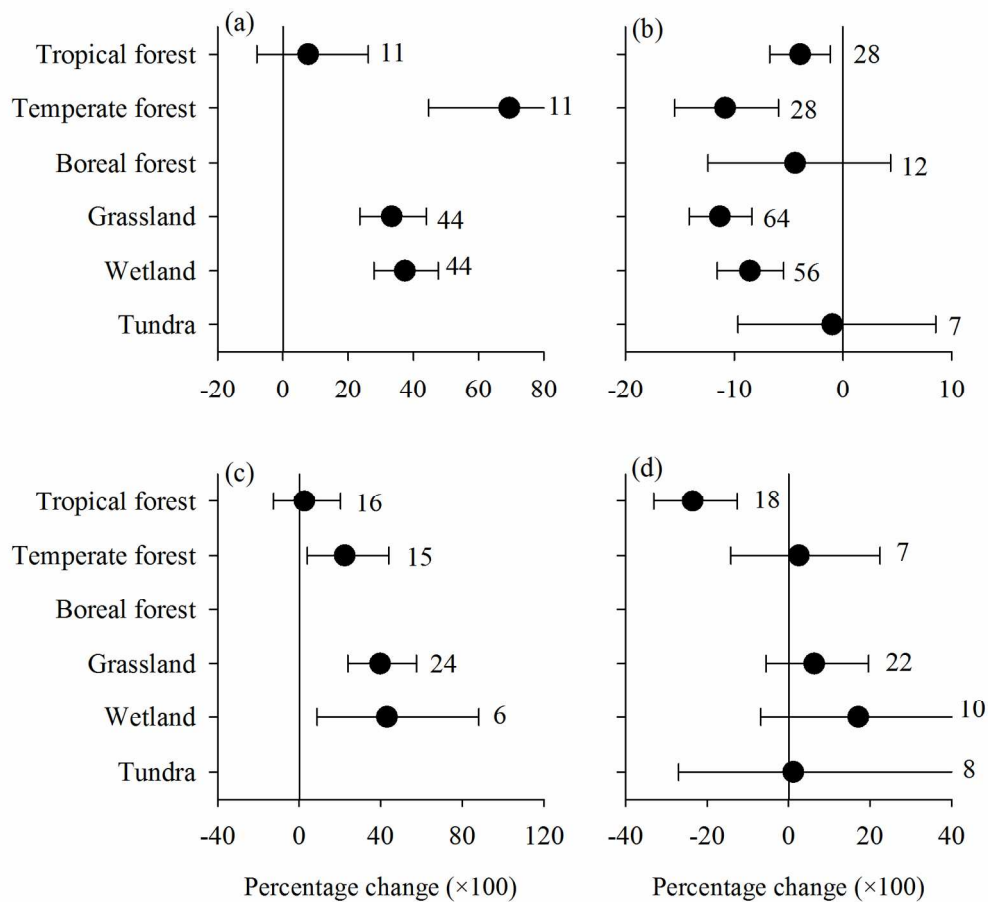


Fig. 3. Percentage change ( $\times 100$ ) of plant biomass (a), plant P concentration (b), phosphatase activity (c), and soil labile P (d) to N additions in different ecosystem types. Error bars represent 95% confidence intervals. The sample size for each variable is shown next to the error bar. Statistical significances of effects of ecosystem type were evaluated based on a chi-square test (also see Appendix S3 in the Supporting Information).

Fig. 3

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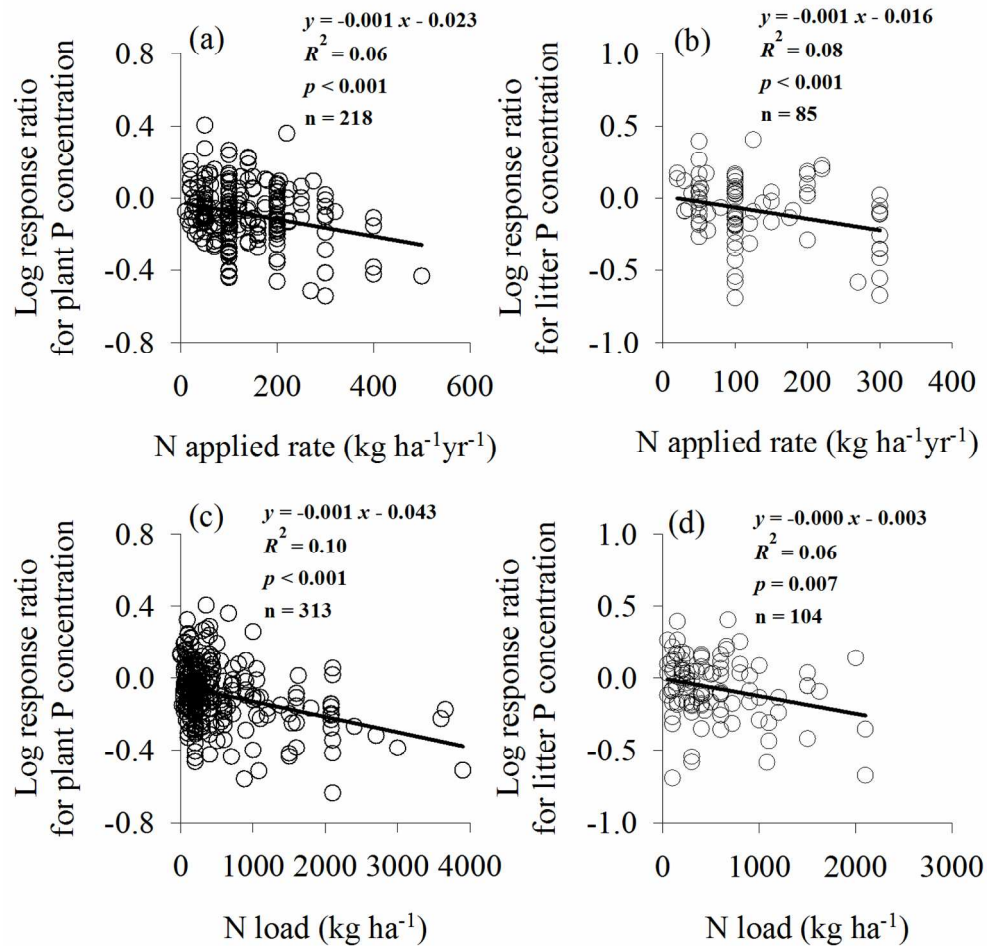


Fig. 4. Percentage change ( $\times 100$ ) of soil labile P with different soil pH value (a) and with different P extraction methods (b). Error bars represent 95% confidence intervals. The sample size for each variable is shown next to the error bar. Alkaline: pH > 7.5; acidic: pH < 5.5; and neutral: pH = 5.5-7.5. Statistical significances of effects of P extraction method were evaluated based on a chi-square test (also see Appendix S3 in the Supporting Information).

Fig. 4

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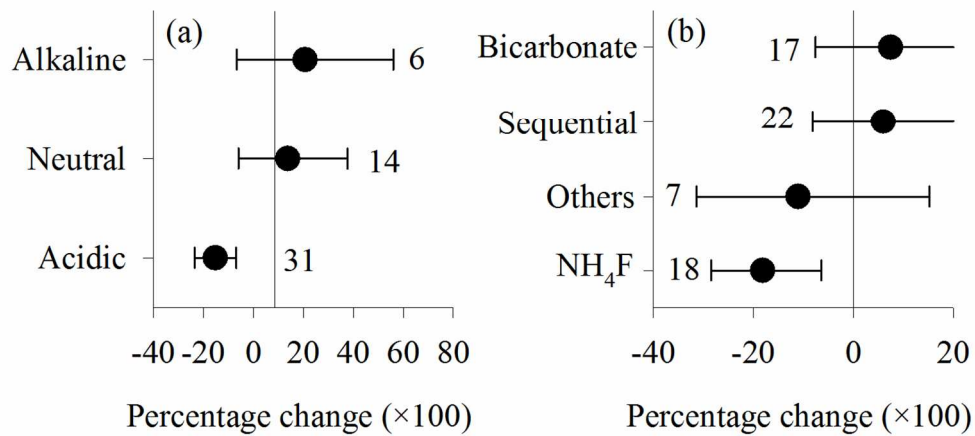


Fig. 5. Log response ratios of plant P concentration (a, c) and litter P concentration (b, d) as a function of N application rate and total N load, and Log response ratios of soil labile P as a function of control soil pH value. Statistical significances of relationships were evaluated based on an F-test (also see Appendix S4 in the Supporting Information).

Fig. 5

127x67mm (300 x 300 DPI)

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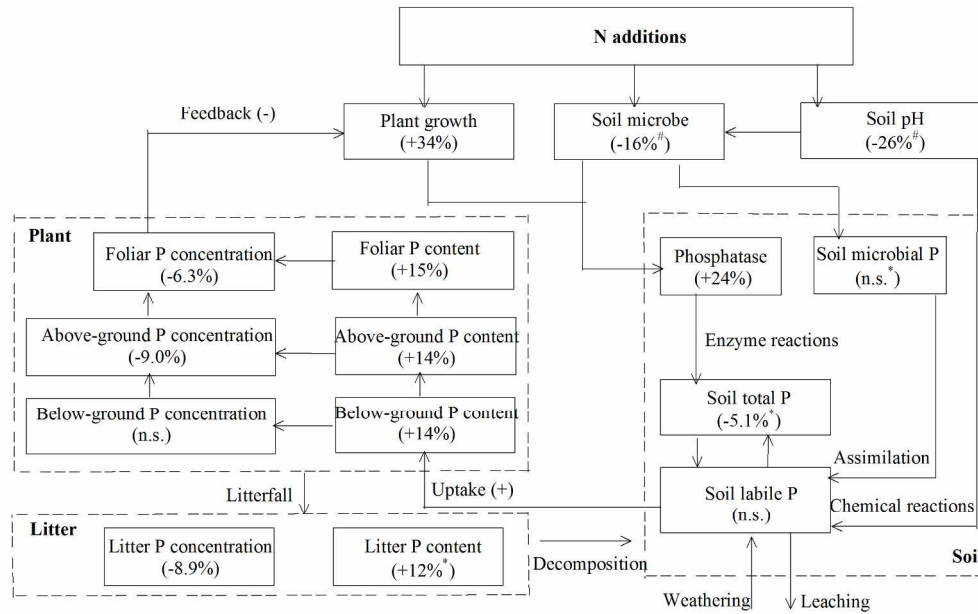


Fig. 6. A quantitative framework for the responses of ecosystem P cycling to N additions. +: positive; -: negative; n.s.: non-significant; #: data for soil microbe and pH from Treseder (2008) and Tian & Niu (2015), respectively. \*: significant publication bias for the analysis suggested by both funnel plot and Rosenthal's fail-safe number.

Fig. 6

224x136mm (300 x 300 DPI)

Accept

## Responses of terrestrial ecosystem phosphorus cycling to nitrogen additions: a meta-analysis

Qi Deng, Dafeng Hui, Sam Dennis, & K. Chandra Reddy

### Supporting Information

**Appendix S1.** Studies included in the meta-analysis.

**Appendix S2.** Rosenthal's fail-safe number used to test the publication bias.

**Appendix S3.** Statistical significances of effects of categorical factors on the response ratios were evaluated based on a chi-square test.

**Appendix S4.** Statistical significances of relationships between the response ratio and continuous factors were evaluated based on a F-test.

**Fig. S1.** Funnel plots of the effect size (log response ratio) plotted against the precision (the reverse of standard error of the response ratio) for each P variable (a, plant biomass; b, plant P content; c, plant P concentration; d, litter biomass; e, litter P content; f, litter P concentration; g, phosphatase activity; h, soil microbial P; i, soil total P; j, soil labile P). The blue line represents the mean log response ratio ( $RR$ ) of N additions on ecosystem P cycling.



**Fig. S2.** Sensitivity analysis for each P variable (a, plant biomass; b, plant P content; c, plant P concentration; d, litter biomass; e, litter P content; f, litter P concentration; g, phosphatase activity; h, soil microbial P; i, soil total P; j, soil labile P). The blue line represents the mean log response ratio (*RR*) of N additions on ecosystem P cycling using the dataset from all study site. Circles show estimates of mean *RR* when one study site is removed.

**Fig. S3.** Percentage change ( $\times 100$ ) of phosphatase activity in plant root and soil under N additions. Error bars represent 95% confidence intervals. The sample size for each variable is shown next to the error bar. Statistical significances of response ratios between plant root phosphatase activity and soil phosphatase activity were evaluated based on a *t*-test.

**Fig. 4.** Percentage change ( $\times 100$ ) of plant P concentration (a) and litter P concentration (b) to N additions with different N sources. Error bars represent 95% confidence intervals. The sample size for each variable is shown next to the error bar. Statistical significances of effects of N source applied were evaluated based on a chi-square test (also see Appendix S3 in the Supporting Information).

**Appendix S1** Experimental sites included in meta-analysis. The selected 10 variables including plant biomass, P content and P concentration, litter biomass, P content and P concentration, phosphatase activity, soil microbial P, soil total P and labile P.

Number	Reference	Ecosystem	N source	N applied rate (kg N ha <sup>-1</sup> yr <sup>-1</sup> )	Duration (yr)	Total N load (kg N ha <sup>-1</sup> )	P variable
1	Adamek, 2009	Tropical Forest	Urea	125	1.4	250	Plant biomass, P content and P concentration, litter biomass and soil total P.
2	Adegbidi et al., 2003	Temperate Forest	Urea	300	3.4	900	Plant biomass and soil total P.
3	Aerts & Berendse, 1988	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	200	2.4	600	Plant biomass, P content and P concentration.
4	Aerts et al., 2001	Wetland	NH <sub>4</sub> NO <sub>3</sub>	100	3.4	400	Plant biomass, P content and P concentration.
5	Aerts et al., 2003a	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	11	1100	Plant biomass, P content and P concentration, soil total and labile P.
6	Aerts et al., 2003b	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	11	1100	Litter P concentration.
7	Aerts et al., 2006	Wetland	NH <sub>4</sub> <sup>+</sup>	100	4	400	Litter P concentration.
8	Aerts, 2009	Wetland	NH <sub>4</sub> <sup>+</sup>	50	4	200	Litter P concentration.
9	Akinola et al., 2010	Grassland	Urea	23	1.5	46	Plant biomass, P content and P concentration.
10	Allen et al., 2010	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	100	7	700	Plant P concentration and soil labile P.
11	An et al., 2011	Grassland	Urea	138	0.3	138	Plant P concentration and litter P concentration
12	Andersen et al., 2010	Tropical Forest	Urea	125	1.4	250	Plant biomass, P content and P concentration, and soil labile P.
13	Arroniz-Crespo et al., 2008	Grassland	NH <sub>4</sub> NO <sub>3</sub>	140	11.2	1400	Plant P concentration and phosphatase activity.
14	Barger et al.,	Grassland	Urea	200	0.4	200	Plant biomass and P concentration, and soil

	2002						labile P.
	Barger et al., 2003	Grassland	Urea	200	0.4	200	Plant biomass and P concentration, and soil labile P.
	Bassin et al., 2012	Grassland	NH <sub>4</sub> NO <sub>3</sub>	50	2	100	Plant biomass and P concentration.
	Bennett & Adams, 2001	Grassland	Urea	50	2	100	Plant biomass, P content and P concentration, litter biomass, P content and P concentration.
	Birk & Vitousek, 1986	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	450	0.5	450	Plant biomass and soil labile P.
	Blanke et al., 2011	Grassland	NH <sub>4</sub> NO <sub>3</sub>	85	4	340	Plant biomass, P content and P concentration, and soil labile P.
	Blanke et al., 2012	Grassland	NH <sub>4</sub> NO <sub>3</sub>	50	2	100	Plant biomass, P content and P concentration.
	Bobbink et al., 1991	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	2	200	Plant biomass, P content and P concentration
	Bowman et al., 1993	Tundra	Urea	250	1.3	500	Plant biomass and P concentration, and soil labile P.
	Bowman, 1994	Tundra	Urea	250	1.3	500	Plant biomass, P content and P concentration.
	Braun et al., 2010	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	160	14	2240	Plant P concentration and phosphatase activity.
	Britton et al., 2008	Grassland	NH <sub>4</sub> NO <sub>3</sub>	50	3	150	Plant P concentration.
	Bubier et al., 2011	Wetland	NH <sub>4</sub> NO <sub>3</sub>	200	8	1600	Plant P concentration.
	Bucci et al., 2006	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	1.7	200	Plant P concentration.
	Campo & Dirzo, 2003	Tropical Forest	Urea	220	3.9	880	Plant P concentration.
	Campo et al., 2007	Tropical Forest	Urea	220	3.9	880	Litter biomass, P content and P concentration.

Chiang et al., 2000	Wetland	NH <sub>4</sub> <sup>+</sup>	140	4	640	Plant biomass and P concentration.
Clarholm, 1993	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	60	21	1260	Plant P concentration, phosphatase activity and soil microbial P
Corbin et al., 2003	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	97	3	291	Plant biomass, P content and P concentration, and soil labile P.
Craft et al., 1995	Wetland	NH <sub>4</sub> <sup>+</sup>	224	2	448	Plant biomass, P content and P concentration, soil total and labile P.
Craine et al., 2008	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	1.6	200	Plant biomass, P content and P concentration.
Cuevas-Reyes et al., 2011	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	1.7	200	Plant P concentration.
Cui et al., 2010	Grassland	NH <sub>4</sub> NO <sub>3</sub>	205	2	410	Plant P concentration and soil labile P.
Cusack et al., 2010	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	50	5.7	300	Phosphatase activity.
D'Antonio & Mack, 2006	Grassland	Urea	100	0.4	100	Plant and litter P concentration
Davidson et al., 2004	Tropical Forest	Urea	100	0.5	100	Plant biomass and P concentration, litter P concentration, soil total and labile P.
Davison et al., 1997	Grassland	Urea	300	7	2100	Plant and litter P concentration, soil total and labile P.
DeForest et al., 2004	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	30	1	30	Phosphatase activity.
Drenovsky & Richards, 2004	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	210 <sup>§</sup>	2	420 <sup>§</sup>	Plant P concentration.
Drenovsky & Richards, 2005	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	233.6 <sup>§</sup>	1	233.6 <sup>§</sup>	Plant P concentration.
Du & Fang, 2014	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	100	2	200	Plant biomass and P concentration.

Elvir et al., 2006	Temperate Forest	NH <sub>4</sub> <sup>+</sup>	25.2	14	352.8	Plant biomass and P concentration.
Falk et al., 2010	Grassland	NH <sub>4</sub> NO <sub>3</sub>	50	2	100	Plant biomass, P content and P concentration, and soil labile P.
Fang et al., 2012	Grassland	Urea	320	7	2240	Plant biomass, P content and P concentration, soil total and labile P.
Feller et al., 1999	Wetland	Urea	150 <sup>§</sup>	5	750 <sup>§</sup>	Plant biomass, P content and P concentration.
Feller et al., 2003a	Wetland	Urea	150 <sup>§</sup>	2	300 <sup>§</sup>	Plant and litter P concentration.
Feller et al., 2003b	Wetland	Urea	150 <sup>§</sup>	2	300 <sup>§</sup>	Plant and litter P concentration.
Feller et al., 2007	Wetland	Urea	150 <sup>§</sup>	4	600 <sup>§</sup>	Plant and litter P concentration, and soil labile P.
Feller et al., 2009	Wetland	Urea	150 <sup>§</sup>	4	600 <sup>§</sup>	Litter P concentration.
Feller, 1995	Wetland	Urea	150 <sup>§</sup>	2	300 <sup>§</sup>	Plant P concentration and soil labile P.
Ferdie & Fourqurean, 2004	Wetland	Urea	77	1.2	77	Plant biomass and P concentration.
Fetcher et al., 1996	Tropical Forest	Urea	102	0.7	102	Plant biomass and P concentration.
Frost et al., 2009	Wetland	NH <sub>4</sub> <sup>+</sup>	500	2.4	1000	Plant biomass and P concentration.
Gerdol et al., 2007	Wetland	NH <sub>4</sub> NO <sub>3</sub>	30	3	90	Plant biomass, P content and P concentration.
Gordon et al., 2001	Tundra	NH <sub>4</sub> NO <sub>3</sub>	50	7	350	Plant P concentration.
Grogan & Chapin III, 2000	Grassland	NH <sub>4</sub> NO <sub>3</sub>	20	1	20	Plant biomass, P content and P concentration.
Gundersen, 1998	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	35	4	140	Plant P concentration, litter biomass, P content and P concentration.
Güsewell et al., 2002	Wetland	NH <sub>4</sub> NO <sub>3</sub>	200	2	400	Plant biomass, P content and P concentration, and soil labile P.

Güsewell et al., 2003	Wetland	NH <sub>4</sub> NO <sub>3</sub>	200	2	400	Plant biomass, P content and P concentration.
Gutknecht et al., 2010	Grassland	NO <sub>3</sub> <sup>-</sup>	70	6	420	Phosphatase activity.
Haines et al., 2015	Grassland	NH <sub>4</sub> NO <sub>3</sub>	135	2.6	405	Plant biomass, P content and P concentration, and soil labile P.
Han et al., 2014	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	3.8	400	Plant biomass, P content and P concentration.
Heijmans et al., 2001	Wetland	NH <sub>4</sub> NO <sub>3</sub>	50	2.3	150	Plant biomass, P content and P concentration.
Heijmans et al., 2002a	Wetland	NH <sub>4</sub> NO <sub>3</sub>	50	1.4	100	Plant biomass, P content and P concentration.
Heijmans et al., 2002b	Wetland	NH <sub>4</sub> NO <sub>3</sub>	50	1.4	100	Plant biomass and P concentration, litter biomass, P content and P concentration.
Henry et al., 2006	Grassland	NO <sub>3</sub> <sup>-</sup>	70	6.5	490	Plant biomass, P content and P concentration.
Herbert, 1995	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	100	1.6	200	Plant biomass and P concentration, and litter biomass.
Herbert & Fownes, 1995	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	100	1.6	200	Plant biomass and P concentration, and litter biomass.
Herbert et al., 1999	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	100	1.6	200	Plant biomass and P concentration, and litter biomass.
Hobbie et al., 2000	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	300	13	3900	Litter P concentration.
Hogan et al., 2010	Wetland	NO <sub>3</sub> <sup>-</sup> ; NH <sub>4</sub> <sup>+</sup> ; NH <sub>4</sub> NO <sub>3</sub>	56	3.6	224	Phosphatase activity.
Högberg et al., 2006	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	118	30	3540	Plant P concentration.
Holub & Tuma, 2010	Wetland	NH <sub>4</sub> NO <sub>3</sub>	50	2	100	Plant biomass, P content and P concentration.

Holub & Záhora, 2008	Grassland	NH <sub>4</sub> NO <sub>3</sub>	50	2	100	Plant biomass, P content and P concentration, and litter P concentration.
Holub et al., 2010	Grassland	NH <sub>4</sub> NO <sub>3</sub>	50	2	100	Plant biomass, P content and P concentration, and litter P concentration.
Hoosbeek et al., 2002	Wetland	NH <sub>4</sub> NO <sub>3</sub>	30	3	90	Litter P concentration.
Hu et al., 2010	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	100	6.3	700	Phosphatase activity and soil total P.
Huang et al., 2012a	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	100	4.5	500	Plant P concentration.
Huang et al., 2012b	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	100	6	600	Phosphatase activity and soil total P.
Huang et al., 2015	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	100	4.5	500	Plant biomass, P content and P concentration.
Huber et al., 2004	Temperate Forest	NH <sub>4</sub> <sup>+</sup> ; NH <sub>4</sub> NO <sub>3</sub>	62	7	434	Plant and litter P concentration.
Huenneke et al., 1990	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	2	200	Plant biomass and soil labile P.
Jacobson & Pettersson, 2001	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	150	7	1050	Plant P concentration.
Joergensen & Scheu, 1999	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	1080	1.3	1320	Soil microbial P.
Johnson et al., 1997	Temperate Forest	NH <sub>4</sub> <sup>+</sup>	200	3.5	800	Plant biomass, P content and P concentration, soil total and labile P.
Johnson et al., 1998	Grassland	NH <sub>4</sub> NO <sub>3</sub>	120	7	840	Phosphatase activity.
Johnson et al., 1999	Grassland	NH <sub>4</sub> NO <sub>3</sub>	140	6	840	Plant P concentration and phosphatase activity.
Judd et al., 1996	Temperate Urea 400 4 1600 Plan					t biomass and P concentration.

		Forest					
	Keeler et al., 2009	Grassland; Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	100	6	600	Phosphatase activity.
	Ket et al., 2011	Wetland	NH <sub>4</sub> <sup>+</sup>	50	4	200	Plant biomass and P concentration.
	Kishchuk et al., 2002	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	150	14	2100	Plant P concentration.
	Koehler et al., 2009	Tropical forest	Urea	125	8	1000	Soil total P.
	Kozovits et al., 2007	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	1.7	200	Plant and litter P concentration.
	Li et al., 2010	Grassland	NH <sub>4</sub> NO <sub>3</sub>	200	5	1000	Plant biomass and P concentration.
	Li et al., 2012	Grassland	NH <sub>4</sub> NO <sub>3</sub>	200	5	1000	Litter P concentration, soil total and labile P.
	Li J et al., 2015	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	100	3.1	100	Soil labile P.
	Li X et al., 2015	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	3.3	400	Plant and litter P concentration.
	Limpens et al., 2004	Wetland	NH <sub>4</sub> NO <sub>3</sub>	40	3	120	Soil labile P.
	Liu et al., 2010	Grassland	Urea	640	2	1280	Litter P concentration.
	Liu et al., 2012	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	100	4.5	500	Plant P concentration.
	Liu et al., 2013	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	100	4.5	500	Plant P concentration.
	Liu et al., 2015	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	100	4.5	500	Litter P concentration.
	Lovelock et al., 2004	Wetland	Urea	300 <sup>§</sup>	2.5	900 <sup>§</sup>	Plant P concentration.
	Lovelock et al., 2007	Wetland	Urea	300 <sup>§</sup>	4	1200 <sup>§</sup>	Litter P concentration.



8	Lu et al., 2012	Tropical forest	$\text{NH}_4\text{NO}_3$	100	6	600	Soil labile P.
9	Ludwig et al., 2001	Grassland	$\text{NO}_3^-$	200	2.5	600	Plant biomass and P concentration.
0	Lv et al., 2011	Grassland	$\text{NH}_4\text{NO}_3$	175	2.8	525	Plant P concentration.
1	Lv et al., 2012	Grassland	$\text{NH}_4\text{NO}_3$	175	2.8	525	Plant and litter P concentration, and soil labile P.
2	Mao et al., 2013	Wetland NH	$\text{NH}_4^+$	120	6.5	840	Plant biomass and P concentration, litter biomass, P content and P concentration.
3	Mao et al., 2014	Wetland NH	$\text{NH}_4\text{NO}_3$	120	6	720	Plant biomass and P concentration.
4	May et al., 2005	Temperate Forest	$\text{NH}_4^+$	35	12	420	Plant P concentration.
5	Mayor et al., 2014a	Tropical forest	Urea	125	12.7	1625	Soil labile P.
6	Mayor et al., 2014b	Tropical forest	Urea	125	12.7	1625	Plant and litter P concentration.
7	McDowell & Koopmans, 2006	Grassland	Urea	400	9	3600	Phosphatase activity, soil total and labile P.
8	McKenzie & Jacobs, 2002	Grassland	Urea	60	1	60	Plant P concentration.
9	Menge, 2003	Grassland NO	$\text{NO}_3^-$	270	4	1080	Plant and litter P concentration, and phosphatase activity.
0	Mirmanto et al., 1999	Tropical forest	Urea	225	3.2	675	Litter biomass, P content and P concentration.
1	Mo et al., 2008	Tropical forest	$\text{NH}_4\text{NO}_3$	300	1.1	300	Plant biomass and P concentration, and soil labile P.
2	Morecroft et al., 1994	Grassland	$\text{NH}_4\text{NO}_3$	140	3	420	Plant P concentration.
3	Morse et al., 2004	Wetland	Urea	225	1	225	Plant biomass and soil total P.
4	Niinemets &	Grassland	$\text{NH}_4\text{NO}_3$	40	1	40	Plant biomass, P content and P concentration.

	Kull, 2005						
5	Ochoa-Hueso & Stevens, 2013a	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	50	3	150	Soil total and labile P.
6	Ochoa-Hueso & Stevens, 2013b	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	50	3	150	Phosphatase activity.
7	Ochoa-Hueso & Stevens, 2015	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	50	3	150	Plant biomass and soil labile P.
8	Øien, 2004	Wetland	Urea	120	2	240	Plant biomass, P content and P concentration.
9	Ostertag, 2010	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	300	13	3900	Plant P concentration.
0	Otto et al., 2007	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	80	3	240	Plant P concentration.
1	Pakeman & Lee, 1991	Grassland	NH <sub>4</sub> NO <sub>3</sub>	175	0.5	175	Plant biomass and P concentration.
2	Pasquini & Santiago, 2011	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	150	7.5	1200	Plant P concentration.
3	Persson & Ahlstrom, 2002	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	200	4	800	Plant biomass, P content and P concentration.
4	Pilkington et al., 2005	Wetland	NH <sub>4</sub> NO <sub>3</sub>	50	1.2	50	Phosphatase activity.
5	Plassmann et al., 2009	Grassland	NO <sub>3</sub> <sup>-</sup>	50	2.1	100	Plant biomass, P content and P concentration.
6	Raich et al., 1996	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	100	2	200	Plant biomass and P concentration.
7	Rainey et al., 1999	Temperate Forest	NH <sub>4</sub> NO <sub>3</sub>	150	7.5	1200	Plant biomass, P content and P concentration.
8	Rejmánková & Snyder, 2008	Wetland	NH <sub>4</sub> NO <sub>3</sub>	200	4	800	Plant and litter P concentration, and phosphatase activity.
9	Robinson et al., 2005	Tundra	NH <sub>4</sub> NO <sub>3</sub>	50	1.2	50	Soil total and labile P.

	2004						
0	Robroek et al., 2009	Grassland	NH <sub>4</sub> NO <sub>3</sub>	20	2.1	40	Plant biomass, P content and P concentration, phosphatase activity, soil total and labile P.
1	Roem et al., 2002	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	5	500	Plant biomass and P concentration.
2	Rueth et al., 2003	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	25	3	75	Plant P concentration.
3	Sarmiento et al., 2006	Grassland	Urea	100	0.5	100	Plant P concentration.
4	Schaberg et al., 1997	Temperate Forest	NH <sub>4</sub> <sup>+</sup>	31.4	7.2	219.8	Plant P concentration.
5	Schils & Snijders, 2004	Grassland	NO <sub>3</sub> <sup>-</sup>	400	4	1600	Plant P pool and soil total P.
6	Simms, 1987	Temperate Forest	Urea	100	1	100	Plant P concentration.
7	Sinsabaugh et al., 2005	Temperate Forest	Urea	80	2	160	Phosphatase activity.
8	Son, 2002	Temperate Forest	NH <sub>4</sub> <sup>+</sup>	400	0.5	400	Plant P concentration.
9	Soudzilovskaia et al., 2005	Tundra	Urea	90	3.3	360	Plant biomass, P content and P concentration.
0	Soudzilovskaia et al., 2007	Tundra	Urea	90	3.3	360	Litter biomass, P content and P concentration.
1	Sparrius et al., 2013	Grassland	NH <sub>4</sub> NO <sub>3</sub>	42.9	3.5	171.6	Plant biomass, P content and P concentration.
2	Stursova et al., 2006	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	4	400	Phosphatase activity.
3	Suding et al., 2008	Tundra	Urea	160	5	900	Plant biomass and phosphatase activity.
4	Sundqvist et al.,	Tundra	NH <sub>4</sub> NO <sub>3</sub>	100	2	200	Soil labile P.

	2014						
5	Tanner et al., 1990	Tropical forest	Urea	100	3	300	Plant P concentration.
6	Tanner et al., 1992	Tropical forest	Urea	150	3	450	Litter P concentration.
7	Thayer et al., 2008	Grassland	NO <sub>3</sub> <sup>-</sup>	70	7	490	Plant biomass and P concentration.
8	Tomassen et al., 2004	Wetland	NH <sub>4</sub> NO <sub>3</sub>	80	3	240	Plant biomass, P content and P concentration, and soil labile P.
9	Treseder & Vitousek, 2001	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	300	13	3900	Plant and litter P concentration.
0	Tu et al., 2014	Temperate forest	NH <sub>4</sub> NO <sub>3</sub>	300	2	600	Phosphatase activity and soil labile P.
1	Turner et al., 2013	Tropical forest	Urea	125	9	1125	Phosphatase activity.
2	Turner & Wright, 2013	Tropical forest	Urea	125	9	1125	Phosphatase activity.
3	Tyler et al., 1992	Boreal Forest	NH <sub>4</sub> NO <sub>3</sub>	180	5	900	Plant and litter P concentration.
4	van der Hoek et al., 2004	Grassland	NH <sub>4</sub> NO <sub>3</sub>	200	3	600	Plant biomass, P content and P concentration.
5	van der Waal et al., 2011	Grassland	NH <sub>4</sub> NO <sub>3</sub>	785 <sup>§</sup>	2	1570 <sup>§</sup>	Plant P concentration.
6	van Dijk et al., 2012	Wetland	NH <sub>4</sub> NO <sub>3</sub>	29.5	3	88.8	Plant P concentration and phosphatase activity.
7	van Duren et al., 1997	Wetland	Urea	200	1	200	Plant biomass, P content and P concentration.
8	van Heerwaarden et al., 2003	Wetland	NH <sub>4</sub> <sup>+</sup>	100	3	300	Plant and litter P concentration.

9	Venterink et al., 2001	Grassland	NH <sub>4</sub> NO <sub>3</sub>	200	0.2	200	Plant biomass, P content and P concentration.
0	Verhoeven & Schmitz, 1991	Wetland	NH <sub>4</sub> NO <sub>3</sub>	100	0.4	100	Plant biomass, P content and P concentration.
1	Verhoeven et al., 2011	Wetland	NO <sub>3</sub> <sup>-</sup> ; NH <sub>4</sub> <sup>+</sup>	70	4	280	Plant biomass, P content and P concentration.
2	Vitousek & Farrington, 1997	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	100	2	200	Plant P concentration.
3	Vitousek & Hobbie, 2000	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	100	2	200	Litter P concentration.
4	Vitousek et al., 1993	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	100	2	200	Plant P concentration and soil labile P.
5	Vitousek, 1998	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	100	10	1000	Plant and litter P concentration.
6	von Oheimb et al., 2010	Grassland	NH <sub>4</sub> NO <sub>3</sub>	50	4	200	Plant P concentration and soil labile P.
7	Wang et al., 2008	Temperate Forest	Urea	100	0.4	100	Soil microbial P, phosphatase activity, soil total and labile P.
8	Wang et al., 2010	Grassland	Urea	200	3	600	Plant biomass and phosphatase activity.
9	Wang et al., 2015	Grassland	Urea	150	8	1200	Phosphatase activity.
0	Wang FM et al., 2014	Tropical forest	NH <sub>4</sub> NO <sub>3</sub>	100	3.1	300	Soil labile P.
1	Wang JS et al., 2014	Grassland	Urea	100	2	200	Soil labile P.
2	Wang XG et al., 2014	Grassland	NH <sub>4</sub> NO <sub>3</sub>	52.5	2	105	Litter P concentration.
3	Warren et al., 2005	Temperate Forest	Urea	275	2	550	Plant P concentration.
4	Weand et al.,	Tropical NH	4NO <sub>3</sub>	50	5	250	Plant P concentration, soil microbial P,

	2010	Forest					phosphatase activity, soil total and labile P.
185	Willems et al., 1993	Grassland	NH <sub>4</sub> NO <sub>3</sub>	10	4	40	Plant biomass, P content and P concentration.
186	Zeglin et al., 2007	Grassland	NH <sub>4</sub> NO <sub>3</sub>	142	55	7810	Phosphatase activity and soil total P.
187	Zhang et al., 2011	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	150	2	300	Soil total and labile P.
188	Zhang et al., 2014	Grassland	Urea	100	3	300	Soil total and labile P.
189	Zhang GN et al., 2013	Grassland	Urea	560	2	1120	Phosphatase activity, soil total and labile P.
190	Zhang NY et al., 2013	Grassland	NH <sub>4</sub> NO <sub>3</sub>	100	4	400	Soil total and labile P.
191	Zheng et al., 2015	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	150	5	750	Phosphatase activity, soil total and labile P.
192	Zhu et al., 2013	Tropical Forest	NH <sub>4</sub> NO <sub>3</sub>	150	5	750	Plant biomass, P content and P concentration, phosphatase activity and soil labile P.

<sup>§</sup>The unit of N applied rate and total N load are g N tree<sup>-1</sup> yr<sup>-1</sup> and g N tree<sup>-1</sup>, respectively.

**Appendix S2.** Rosenthal's fail-safe number used to test the publication bias. Fonts in bold indicate that the mean response ratio (*RR*) had significant difference from zero. Asterisk indicates a significant publication bias for the analysis by the Rosenthal's fail-safe number.

Treatment	Plant biomass				Plant P content				Plant P concentration			
	Mean <i>RR</i>	95% CI	n	Fail-safe number	Mean <i>RR</i>	95% CI	n	Fail-safe number	Mean <i>RR</i>	95% CI	n	Fail-safe number
Overall	<b>0.29</b>	<b>0.24~0.34</b>	<b>126</b>	<b>65236</b>	<b>0.15</b>	<b>0.10~0.21</b>	<b>67</b>	<b>6976</b>	<b>-0.08</b>	<b>-0.11~-0.06</b>	<b>195</b>	<b>17560</b>
Tropical forest	0.07	-0.08~0.23	11		0.24	-0.15~0.64	1		<b>-0.04</b>	<b>-0.07~-0.01</b>	<b>28</b>	<b>175</b>
Temperate forest	<b>0.53</b>	<b>0.37~0.68</b>	<b>11</b>	<b>807</b>	0.23	-0.1~0.47	3		<b>-0.11</b>	<b>-0.17~-0.06</b>	<b>28</b>	<b>372</b>
Boreal forest	0.17	-0.22~0.57	2		0.04	-0.29~0.38	2		-0.04	-0.13~0.04	12	
Grassland	<b>0.29</b>	<b>0.21~0.36</b>	<b>44</b>	<b>7944</b>	<b>0.13</b>	<b>0.06~0.20</b>	<b>36</b>	<b>1235</b>	<b>-0.12</b>	<b>-0.15~-0.09</b>	<b>64</b>	<b>3352</b>
Wetland	<b>0.32</b>	<b>0.25~0.39</b>	<b>44</b>	<b>12674</b>	<b>0.20</b>	<b>0.11~0.28</b>	<b>22</b>	<b>1231</b>	<b>-0.09</b>	<b>-0.12~-0.06</b>	<b>56</b>	<b>1348</b>
Tundra	0.03	-0.21~0.27	4		0.05	-0.21~0.31	3		-0.01	-0.10~0.08	7	
NO <sub>3</sub> <sup>-</sup> -N	0.20	-0.08~0.49	5		<b>0.43</b>	<b>0.14~0.71</b>	<b>3</b>	<b>50</b>	<b>-0.20</b>	<b>-0.31~-0.11</b>	<b>6</b>	<b>44</b>
NH <sub>4</sub> <sup>+</sup> -N	<b>0.34</b>	<b>0.15~0.53</b>	<b>11</b>	<b>139</b>	<b>0.26</b>	<b>0.06~0.45</b>	<b>4</b>	<b>26*</b>	<b>-0.12</b>	<b>-0.17~-0.07</b>	<b>25</b>	<b>438</b>
NH <sub>4</sub> NO <sub>3</sub>	<b>0.30</b>	<b>0.24~0.36</b>	<b>84</b>	<b>34360</b>	<b>0.13</b>	<b>0.08~0.18</b>	<b>51</b>	<b>3147</b>	<b>-0.08</b>	<b>-0.11~-0.06</b>	<b>119</b>	<b>6236</b>
Urea	<b>0.27</b>	<b>0.16~0.38</b>	<b>24</b>	<b>2079</b>	0.07	-0.08~0.22	7		<b>-0.06</b>	<b>-0.10~-0.03</b>	<b>43</b>	<b>527</b>

Note: If the fail-safe number was over  $5n+10$  ( $n$  is the number of cases in the analysis), we make conclusion that our result was strong against publication bias. Otherwise, a significant publication bias for the analysis was suggested by the Rosenthal's fail-safe number.

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Treatment	Litter biomass				Litter P content				Litter P concentration			
	Mean <i>RR</i>	95% CI	n	Fail-safe number	Mean <i>RR</i>	95% CI	n	Fail-safe number	Mean <i>RR</i>	95% CI	n	Fail-safe number
Overall	<b>0.14</b>	<b>0.08~0.20</b>	<b>16</b>	<b>140</b>	<b>0.11</b>	<b>0.05~0.18</b>	<b>6</b>	<b>30*</b>	<b>-0.09</b>	<b>-0.17~-0.04</b>	<b>65</b>	<b>828</b>
Tropical forest	0.10	-0.06~0.24	8		0.08	-0.03~0.18	1		-0.00	-0.08~0.08	20	
Temperate forest			0				0		-0.17	-0.44~0.09	2	
Boreal forest	0.05	-0.19~0.28	1		-0.09	-0.31~0.11	1		-0.08	-0.32~0.16	2	
Grassland	0.17	-0.05~0.38	1		<b>0.19</b>	<b>0.03~0.33</b>	<b>1</b>	<b>2*</b>	<b>-0.23</b>	<b>-0.33~-0.13</b>	<b>14</b>	<b>293</b>
Wetland	<b>0.16</b>	<b>0.06~0.26</b>	<b>5</b>	<b>3*</b>	<b>0.10</b>	<b>0.02~0.18</b>	<b>3</b>	<b>3*</b>	<b>-0.13</b>	<b>-0.21~-0.05</b>	<b>27</b>	<b>146</b>
Tundra	0.06	-0.17~0.29	1				0				0	
NO <sub>3</sub> <sup>-</sup> -N			0				0		<b>-0.25</b>	<b>-0.38~-0.08</b>	<b>18</b>	<b>2721</b>
NH <sub>4</sub> <sup>+</sup> -N	<b>0.28</b>	<b>0.13~0.42</b>	<b>8</b>	<b>10*</b>			0		<b>-0.11</b>	<b>-0.17~-0.05</b>	<b>34</b>	<b>972</b>
NH <sub>4</sub> NO <sub>3</sub>	<b>0.20</b>	<b>0.07~0.30</b>	<b>6</b>	<b>9*</b>	0.12	-0.04~0.27	5		<b>-0.04</b>	<b>-0.07~-0.01</b>	<b>10</b>	<b>50*</b>
Urea	<b>0.10</b>	<b>0.03~0.16</b>	<b>2</b>	<b>3*</b>	<b>0.12</b>	<b>0.07~0.17</b>	<b>1</b>	<b>2*</b>	-0.04	-0.09~0.00	1	

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Treatment	Phosphatase activity				Soil microbial P			
	Mean <i>RR</i>	95% CI	n	Fail-safe number	Mean <i>RR</i>	95% CI	n	Fail-safe number
Overall	<b>0.21</b>	<b>0.13~0.30</b>	<b>65</b>	<b>9487</b>	0.01	-0.02~0.02	8	
Tropical forest	0.02	-0.14~0.18	16		-0.00	-0.02~0.02	5	
Temperate forest	<b>0.20</b>	<b>0.04~0.36</b>	<b>15</b>	<b>806</b>	-0.18	-0.48~0.12	1	
Boreal forest	0.13	-0.26~0.52	3		0.04	-0.01~0.10	2	
Grassland	<b>0.33</b>	<b>0.21~0.45</b>	<b>24</b>	<b>1895</b>				
Wetland	<b>0.36</b>	<b>0.08~0.63</b>	<b>6</b>	<b>47</b>				
Tundra	-0.14	-0.82~0.54	1					
NO <sub>3</sub> <sup>-</sup> -N	0.10	-0.23~0.43	1					
NH <sub>4</sub> <sup>+</sup> -N	0.44	-0.27~1.15	4					
NH <sub>4</sub> NO <sub>3</sub>	<b>0.20</b>	<b>0.11~0.30</b>	<b>49</b>	<b>4932</b>	0.00	-0.02~0.03	7	
Urea	<b>0.27</b>	<b>0.06~0.48</b>	<b>11</b>	<b>152</b>	-0.18	-0.48~0.12	1	

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Treatment	Soil total P				Soil labile P			
	Mean <i>RR</i>	95% CI	n	Fail-safe number	Mean <i>RR</i>	95% CI	n	Fail-safe number
Overall	<b>-0.05</b>	<b>-0.10~-0.01</b>	<b>38</b>	<b>150*</b>	-0.03	-0.11~0.05	66	
Tropical forest	0.00	-0.09~0.09	12		<b>-0.27</b>	<b>-0.40~-0.13</b>	<b>18</b>	<b>483</b>
Temperate forest	<b>-0.18</b>	<b>-0.30~-0.06</b>	<b>4</b>	<b>13*</b>	0.03	-0.15~0.20	7	
Boreal forest	-0.09	-0.26~0.07	2		0.03	-0.58~0.63	1	
Grassland	-0.02	-0.09~0.05	14		0.06	-0.06~0.18	22	
Wetland	-0.11	-0.25~0.02	5		0.16	-0.07~0.39	10	
Tundra	-0.02	-0.30~0.26	1		0.01	-0.31~0.34	8	
NO <sub>3</sub> <sup>-</sup> -N	-0.04	-0.21~0.12	12		-0.04	-0.91~0.82	15	
NH <sub>4</sub> <sup>+</sup> -N	<b>-0.20</b>	<b>-0.34~-0.05</b>	<b>20</b>	<b>139</b>	-0.10	-0.52~0.33	46	
NH <sub>4</sub> NO <sub>3</sub>	-0.03	-0.09~0.03	4		-0.09	-0.21~0.01	4	
Urea	-0.05	-0.12~0.03	2		0.13	-0.08~0.31	1	

**Appendix S3.** Statistical significances of effects of categorical factors on the response ratios were evaluated based on a chi-square test. Significant factors are indicated in bold. In order to avoid gaining unreliable conclusions, some subgroups without a sufficiently large dataset ( $n < 5$ ) were excluded in the analysis. ‘-’ indicates that there was no at least two subgroups with sufficiently large dataset ( $n > 5$ ).

Variable	Plant tissue			Ecosystem			N source		
	$\chi^2$	df	<i>P</i>	$\chi^2$	df	<i>P</i>	$\chi^2$	df	<i>P</i>
Plant biomass	<b>13.14</b>	<b>3</b>	<b>0.004</b>	<b>15.35</b>	<b>3</b>	<b>0.002</b>	0.90	3	0.825
Plant P content	2.27	3	0.518	1.28	1	0.258	0.90	1	0.343
Plant P concentration	<b>27.51</b>	<b>3</b>	<b>&lt;0.001</b>	<b>15.43</b>	<b>5</b>	<b>0.009</b>	<b>8.20</b>	<b>3</b>	<b>0.042</b>
Litter biomass				0.05	1	0.831	0.00	1	0.998
Litter P content				-	-	-	-	-	-
Litter P concentration				<b>12.41</b>	<b>2</b>	<b>0.002</b>	<b>10.85</b>	<b>2</b>	<b>0.014</b>
Phosphatase activity				<b>8.85</b>	<b>3</b>	<b>0.031</b>	0.24	1	0.622
Soil microbial P				-	-	-	-	-	-
Soil total P				2.18	3	0.337	0.17	1	0.676
Soil labile P				<b>17.32</b>	<b>4</b>	<b>0.002</b>	0.06	1	0.840

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Variable	P extraction method			Soil pH			Soil type		
	$\chi^2$	df	<i>P</i>	$\chi^2$	df	<i>P</i>	$\chi^2$	df	<i>P</i>
Plant biomass				0.22	2	0.894	0.79	2	0.674
Plant P content				4.46	1	0.108	-	-	-
Plant P concentration				3.16	2	0.175	0.23	2	0.890
Litter biomass				2.47	1	0.291	-	-	-
Litter P content				-	-	-	-	-	-
Litter P concentration				2.71	2	0.272	-	-	-
Phosphatase activity				1.79	1	0.408	0.30	1	0.581
Soil microbial P				-	-	-	-	-	-
Soil total P	2.58	3	0.461	0.76	1	0.685	0.33	2	0.850
Soil labile P	<b>9.79</b>	<b>3</b>	<b>0.021</b>	<b>8.74</b>	<b>2</b>	<b>0.013</b>	1.23	1	0.267

**Appendix S4.** Statistical significances of relationships between the response ratio and continuous factors (n>20 only) were evaluated based on an F-test. Significant factors are indicated in bold. ‘-‘ indicate insufficiently large dataset (n<20) for the analysis.

Variable	Experimental duration			N applied rate			Total N load			SOC		
	F	n	P	F	n	P	F	n	P	F	n	P
Plant biomass	0.02	178	0.749	0.85	182	0.235	1.80	215	0.094	0.11	25	0.723
Plant P content	2.07	81	0.079	1.30	85	0.215	0.28	97	0.497	-	-	-
Plant P concentration	0.09	193	0.661	<b>15.62</b>	<b>218</b>	<b>&lt;0.001</b>	<b>26.61</b>	<b>313</b>	<b>&lt;0.001</b>	0.10	37	0.795
Litter biomass	2.49	21	0.114	-	-	-	-	-	-	-	-	-
Litter P content	-	-	-	-	-	-	-	-	-	-	-	-
Litter P concentration	1.99	89	0.166	<b>8.27</b>	<b>85</b>	<b>&lt;0.001</b>	<b>7.71</b>	<b>105</b>	<b>0.007</b>	-	-	-
Phosphatase activity	0.68	81	0.308	1.43	85	0.131	1.79	98	0.108	3.01	28	0.083
Soil microbial P	-	-	-	-	-	-	1.64	8	0.201	-	-	-
Soil total P	0.43	60	0.312	1.29	55	0.156	0.23	74	0.429	-	-	-
Soil labile P	1.69	99	0.143	0.57	85	0.351	0.06	123	0.610	-	-	-

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Variable	Soil pH			MAT			MAP			Soil depth		
	F	n	<i>P</i>	F	n	<i>P</i>	F	n	<i>P</i>	F	n	<i>P</i>
Plant biomass	0.33	74	0.566	1.36	48	0.244	2.83	67	0.093			
Plant P content	0.23	36	0.629	0.39	23	0.532	1.33	31	0.250			
Plant P concentration	1.45	114	0.206	1.40	99	0.236	1.02	126	0.205			
Litter biomass	-	-	-	-	-	-	-	-	-			
Litter P content	-	-	-	-	-	-	-	-	-			
Litter P concentration	1.01	49	0.316	1.29	56	0.255	2.16	57	0.141			
Phosphatase activity	0.87	45	0.351	1.04	34	0.307	0.77	41	0.379	0.01	53	0.990
Soil microbial P	-	-	-	-	-	-	-	-	-	-	-	-
Soil total P	0.84	28	0.358	0.01	23	0.927	1.40	26	0.237	2.81	70	0.932
Soil labile P	<b>12.85</b>	<b>51</b>	<b>&lt;0.001</b>	1.53	43	0.216	3.22	56	0.078	0.79	128	0.376

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Variable	Experimental duration			N applied rate			Total N load		
	F	n	<i>P</i>	F	n	<i>P</i>	F	n	<i>P</i>
Plant biomass	0.02	178	0.749	0.85	182	0.235	1.80	215	0.094
Plant P content	2.07	81	0.079	1.30	85	0.215	0.28	97	0.497
Plant P concentration	0.09	193	0.661	<b>15.62</b>	<b>218</b>	<b>&lt;0.001</b>	<b>26.61</b>	<b>313</b>	<b>&lt;0.001</b>
Litter biomass	2.49	21	0.114	1.50	17	0.220	1.07	18	0.284
Litter P content	-	-	-	-	-	-	-	-	-
Litter P concentration	1.99	89	0.166	<b>8.27</b>	<b>85</b>	<b>&lt;0.001</b>	<b>7.71</b>	<b>105</b>	<b>0.007</b>
Phosphatase activity	0.68	81	0.308	1.43	85	0.131	1.79	98	0.108
Soil microbial P	-	-	-	-	-	-	-	-	-
Soil total P	0.43	60	0.312	1.29	55	0.156	0.23	74	0.429
Soil labile P	1.69	99	0.143	0.57	85	0.351	0.06	123	0.610

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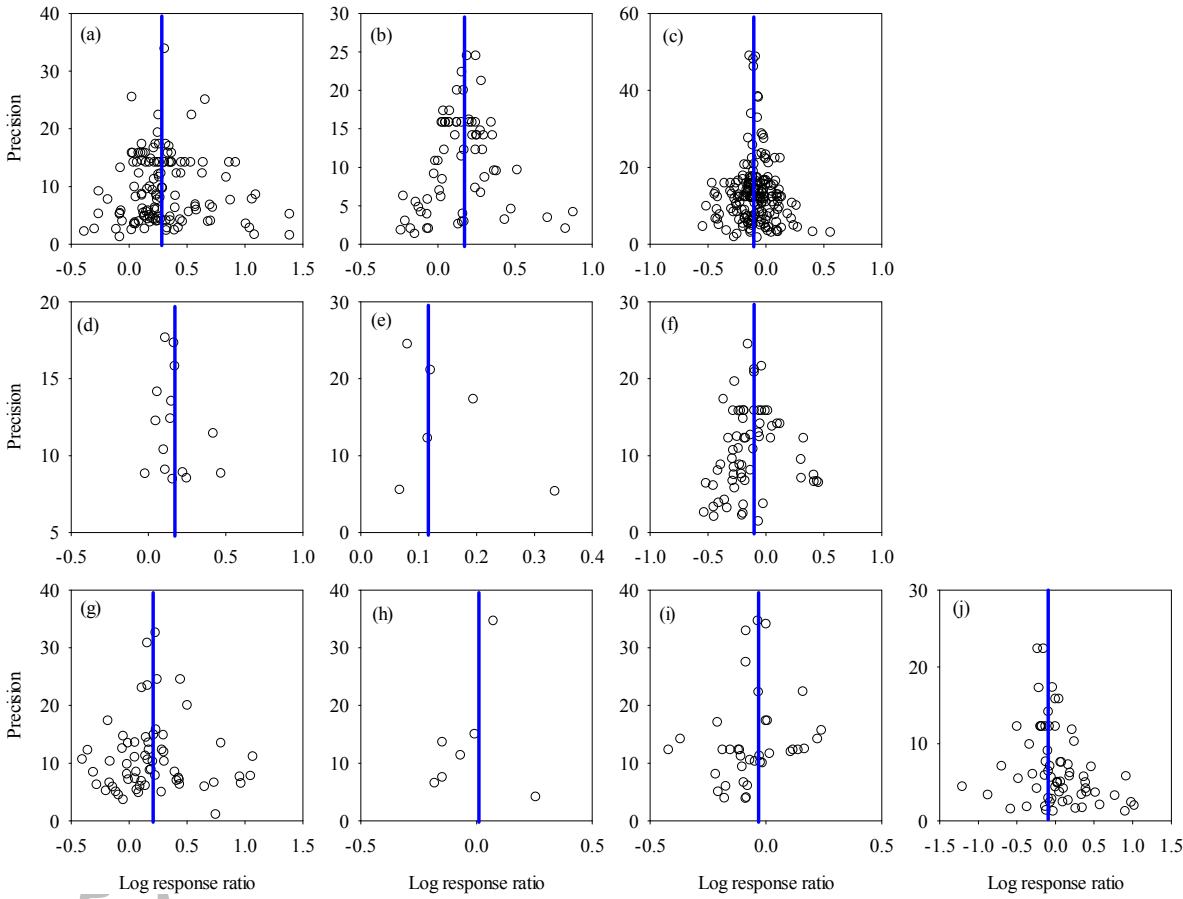


Fig. S1

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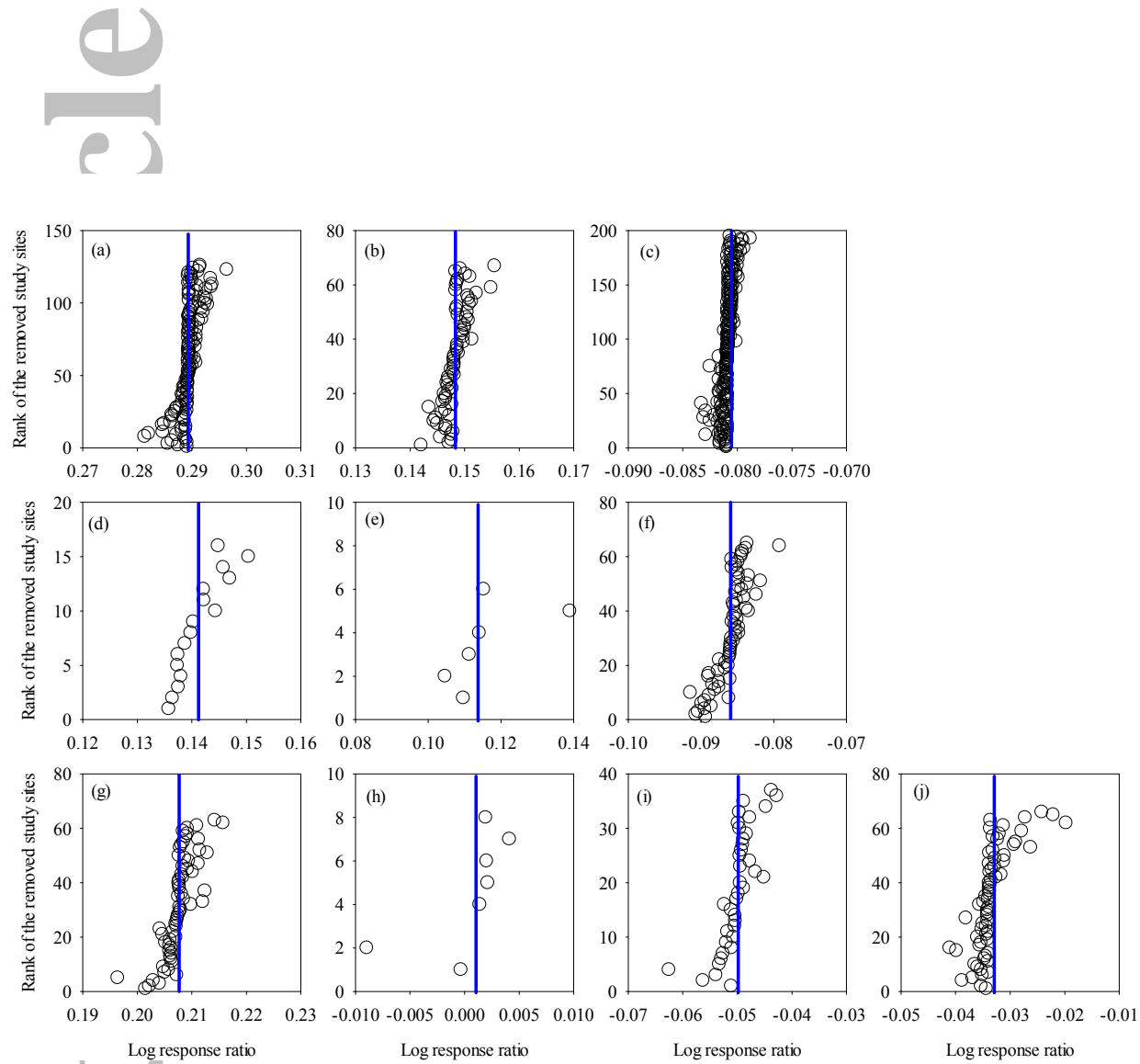


Fig. S2

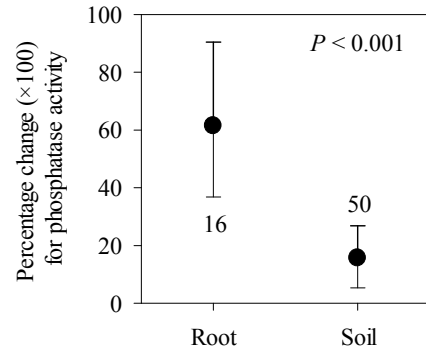


Fig. S3

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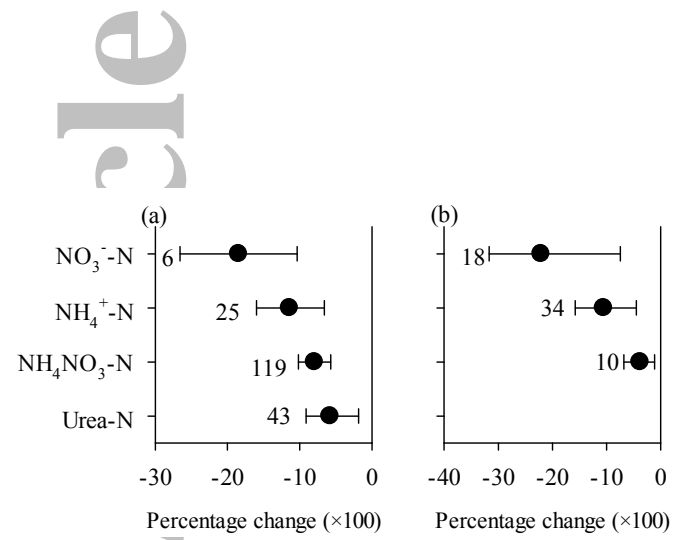


Fig. S4