

RESEARCH

Open Access



Nitrogen addition mediates the response of foliar stoichiometry to phosphorus addition: a meta-analysis

Chengming You^{1,2}, Changhui Peng^{2,3}, Zhenfeng Xu¹, Yang Liu¹, Li Zhang¹, Rui Yin⁴, Lin Liu⁵, Han Li¹, Lixia Wang¹, Sining Liu¹, Bo Tan^{1*} and Paul Kardol⁶

Abstract

Background: Changes in foliar nitrogen (N) and phosphorus (P) stoichiometry play important roles in predicting the effects of global change on ecosystem structure and function. However, there is substantial debate on the effects of P addition on foliar N and P stoichiometry, particularly under different levels of N addition. Thus, we conducted a global meta-analysis to investigate how N addition alters the effects of P addition on foliar N and P stoichiometry across different rates and durations of P addition and plant growth types based on more than 1150 observations.

Results: We found that P addition without N addition increased foliar N concentrations, whereas P addition with N addition had no effect. The positive effects of P addition on foliar P concentrations were greater without N addition than with N addition. Additionally, the effects of P addition on foliar N, P and N:P ratios varied with the rate and duration of P addition. In particular, short-term or low-dose P addition with and without N addition increased foliar N concentration, and the positive effects of short-term or low-dose P addition on foliar P concentrations were greater without N addition than with N addition. The responses of foliar N and P stoichiometry of evergreen plants to P addition were greater without N addition than with N addition. Moreover, regardless of N addition, soil P availability was more effective than P resorption efficiency in predicting the changes in foliar N and P stoichiometry in response to P addition.

Conclusions: Our results highlight that increasing N deposition might alter the response of foliar N and P stoichiometry to P addition and demonstrate the important effect of the experimental environment on the results. These results advance our understanding of the response of plant nutrient use efficiency to P addition with increasing N deposition.

Keywords: Ecological stoichiometry, Plant nutrient regime, Nutrient resorption efficiency, Soil N and P availability, Global change, Global synthesis

*Correspondence: bobotan1984@163.com

¹ Forestry Ecological Engineering in the Upper Reaches of the Yangtze River Key Laboratory of Sichuan Province and National Forestry and Grassland Administration Key Laboratory of Forest Resources Conservation and Ecological Safety on the Upper Reaches of the Yangtze River and Rainy Area of West China Plantation Ecosystem Permanent Scientific Research Base, Institute of Ecology and Forestry, Sichuan Agricultural University, 211 Huimin Road, Wenjiang District, Chengdu 611130, Sichuan, People's Republic of China
Full list of author information is available at the end of the article

Introduction

Human activities, including the increased use of artificial fertilizers and fossil fuel combustion, have led to increased concentrations of atmospheric reactive nitrogen (N) (Liu et al. 2013; Peñuelas et al. 2013). In contrast, atmospheric phosphorus (P) deposition has remained relatively constant (Mahowald et al. 2008; Peñuelas et al. 2013). An imbalance in anthropogenic N and P inputs may shift plants from N-limited growth or N- and P-co-limited growth to P-limited growth (Jiang et al. 2019; Li et al. 2016; Peñuelas et al. 2013; Du et al. 2020). To improve our understanding of the P deficiency caused by human-induced imbalances in N and P inputs, many studies have investigated how P addition affects ecosystem processes and functions, such as foliar N and P stoichiometry (Mayor et al. 2014; Mo et al. 2019; Ostertag 2010; Yan et al. 2015). The ratio of foliar N to P indicates the ability of plants to maintain their internal chemical stoichiometry (Demars and Edwards 2007) and sheds light on the possible adaptation of plants to future environmental changes (Elser et al. 2010), but the effects of P addition on global foliar N and P stoichiometry remain unclear. It is well known that P addition can relieve P limitations in terrestrial ecosystems and profoundly alter the structure and function of ecosystems (Li et al. 2016; Mao et al. 2016) as well as influence the cycling of N (Rejmánková and Snyder 2008; Yuan and Chen 2015b). Therefore, comprehensive analysis of foliar N and P stoichiometry in response to P addition is important for understanding how plants will adapt to future environmental changes.

Many studies have focused on N and P stoichiometry because of its strong relationship with basic ecosystem functions, such as primary productivity and decomposition (e.g., Elser et al. 2010; Mayor et al. 2014; See et al. 2019). These studies have shown that foliar N and P stoichiometry is strongly mediated by biotic factors, e.g., plant species traits (Mao et al. 2016; Mayor et al. 2014) and growth type (Reich et al. 2009; Yuan and Chen 2015b), and by abiotic factors, e.g., geography (Yue et al. 2019) and climate (Chen et al. 2013; Sardans et al. 2016; Viciado et al. 2021). To date, numerous experimental studies have focused on how P addition affects foliar N and P stoichiometry, but their results are inconsistent (Gao et al. 2018; Iversen et al. 2010; Mayor et al. 2014; Ostertag 2010). For instance, the responses of foliar N concentration to P addition were found to be neutral (Gao et al. 2018; Mayor et al. 2014), negative (Ostertag 2010) and even positive (Iversen et al. 2010). These results indicate that various factors (including N addition and experimental design) and biochemical processes (including nutrient resorption) might also influence foliar N and P stoichiometry in response to P addition (Townsend and Asner 2013;

Yue et al. 2019). For example, foliar P concentrations increased in response to N addition due to alterations in phosphatase activity (Fujita et al. 2010; Marklein and Houlton 2012) and mycorrhizal activity (Rowe et al. 2008) and promoted the rate of organic matter release (Knorr et al. 2005). These findings suggest that foliar N and P stoichiometry may vary in response to P addition with and without N addition (Li et al. 2016). However, there is a lack of information on the global patterns of N addition that mediate the responses of foliar N and P stoichiometry to P addition. Furthermore, the change in foliar N and P stoichiometry in response to P addition may depend on the experimental design, including the rate and duration of P addition and plant growth type. For instance, P addition can promote foliar N concentrations by enhancing soil N availability (Reed et al. 2007; Rejmánková and Snyder 2008) or N resorption efficiency (NRE) (Huang et al. 2016; Yan et al. 2015), but which will be used for assimilation and thus diluted by biomass as P availability is continuously increased. Thus, a global analysis is needed to generalize the response patterns of foliar N and P stoichiometry to P addition with and without N addition among various experimental design parameters.

Because the N and P dynamics are tightly coupled in ecosystem processes (Ågren et al. 2012), understanding the mechanisms through which plants maintain the N and P balance in their leaves in response to P addition is challenging (Li et al. 2016; Yue et al. 2019). For example, in nutrient-poor soil, plants maintain their growth mainly through nutrient reabsorption, while in nutrient-rich soils, plants might take up new nutrients through their roots to maintain growth and development (Aerts and Chapin III 1999; Wright and Cannon 2001). P addition may allow plants to take up more P from soil, and thereby reduces their dependence on internal P recycling (Gao et al. 2018; Mao et al. 2015; Yan et al. 2015). Theoretically, as discussed above, to alleviate N deficiency caused by P addition, plants can take up more N from the soil, improve their NRE or use a combination of these two strategies (Huang et al. 2016; Reed et al. 2007; Rejmánková and Snyder 2008; Yan et al. 2015). Given the complex relationship between N and P cycling, how soil nutrient availability and nutrient resorption efficiency maintain the balance in the foliar N and P stoichiometry remains unclear, and predicting the changes in this stoichiometry after P addition is not easy.

In this study, we aimed to determine whether and how P addition with and without N addition affects foliar N, P concentrations, and N:P ratios. We hypothesized that (1) the response of foliar N and P stoichiometry to P addition will decrease with N addition and (2) the rate and duration of P addition will increase the effects

of P addition, and the effects of P addition will depend on plant growth type. To test these two hypotheses, we asked the following questions: (1) what are the global patterns of the responses of foliar N and P stoichiometry, nutrient resorption efficiency and soil N and P availabilities to P addition with and without N addition? (2) What factors regulate the effects of P addition on foliar N and P stoichiometry? (3) How do plants maintain the balance of foliar N and P in response to P addition?

Materials and methods

Dataset assembly

Google Scholar, Web of Science and China National Knowledge were used to search for peer-reviewed publications published before 2020. A database was established through literature collection, and this database included all research results on the effects of P addition on the foliar N and P stoichiometry, NRE, P resorption efficiency (PRE), and soil-available N and P concentrations. We used the following search string to identify the studies: “Phosphorus addition or Phosphorus deposition or Phosphorus supply or Phosphorus enrichment or Phosphorus fertilization or Nitrogen + Phosphorus addition or Nitrogen + Phosphorus deposition or Nitrogen + Phosphorus enrichment or Nitrogen + Phosphorus supply or Nitrogen + Phosphorus fertilization, foliar N and P stoichiometry, foliar N and P concentrations, foliar N:P ratios, N and P resorption efficiency”.

The following criteria were used to minimize publication bias in the collected literature: (1) if only P was added, the experiment must have included a P addition treatment and a control treatment, whereas if the experiment included P addition combined with N addition, then the experiment must have included an N addition treatment and an N + P addition treatment. (2) The data had to include P addition effects on foliar N and P stoichiometry. (3) The data on foliar N and P stoichiometry in response to P addition must have been obtained at the community level or at the species level and derived from mature leaves. (4) The data must have been derived from measured data, i.e., empirical studies (Gao et al. 2018; Mao et al. 2016). (5) If the experimental data of the same site were published in multiple journals, we used the mean for statistical analysis. (6) If the original study included multiple P addition treatments or N + P addition treatments or multiple sampling occasions over time, we treated them as separate observations. (7) The data had to include the mean, sample size and standard deviation (SD) or standard error (SE). In addition, to understand whether and how factors influence the response of foliar N and P stoichiometry to P addition, we gathered other site-specific information from each selected study. For example, we collected data on experimental design

parameters (i.e., P addition rates, experiment duration, and plant growth types), geographic factors [i.e., altitude (m), longitude (°) and latitude (°)] and climatic factors [i.e., mean annual temperature (MAT, °C) and mean annual precipitation (MAP, mm)]. If needed, the GetData Graph Digitizer (version 2.24, Russian Federation) was used to extract data presented in graphs.

In total, data from 168 articles, i.e., 48 articles on P addition without N addition and 120 articles on P addition with N addition, were collected. The latitudes of the sample sites for the observations ranged from 80.00° S to 159.22° N; the longitude values ranged from 159.69° W to 144.72° E (Additional file 1: Fig. S1); the altitudes of the sample sites ranged from 2 to 3650 m; the MAT values were in the range of −10–28.6 °C; and the MAP values ranged from 130 to 4300 mm. This information indicates that our sample observations from experimental sites cover different biogeographical regions and ecosystems. To quantify the response of foliar N and P stoichiometry to P addition with and without N addition, we first grouped the data into a P addition without N addition group and a P addition with N addition group (sensu Li et al. 2016). The P addition with N addition group included N addition alone and combined N and P addition treatments; and the effects of P addition were then calculated based on the treatments consisting of only N addition (the control treatment) and the combination of N and P addition (the P addition treatment). Additionally, to understand how experimental design parameters affect the effect size of P addition with and without N addition, we divided the data into multiple subgroups. The P addition rates were divided into four classes (<5 g m⁻², 5–10 g m⁻², 10–15 g m⁻², and >15 g m⁻²), and experimental duration was divided into two subgroups (≤3 years vs. >3 years) based on a previous study (Li et al. 2016). Experimental test subjects were divided into the community and species levels, and plant growth types were further divided into evergreen plants, deciduous plants, coniferous plants, graminoids and forbs (Yuan and Chen 2015b).

Meta-analysis

We quantified the effects of P addition by calculating the natural log of the response ratio (RR) (Hedges et al. 1999). The RR was calculated as $\ln RR = \ln (X_t/X_c)$, indicating the responses of the target variable to the experimental treatment, in this equation, X_t represents the mean value of the treatment group, and X_c represents the mean of the control group. The variance of each RR was calculated from the SD and sample size of each independent target variable. To increase the credibility of the RR and quantify the significance of the RR, we calculated the weighted RR and the 95% confidence

interval (95% CI) using Meta-Win 2.1 software (Sinauer Associates, Inc. Sunderland, MA, USA). If the 95% CI of the weighted RR did not overlap with 1, the weighted RR of the P addition treatment relative to the control treatment was considered significantly negative or positive. Furthermore, to compare the effects of P addition with and without N addition on the target variables and to examine how experimental design parameters affect the effects of P addition, we assessed the differences between the subgroups based on the heterogeneity between (Q_{between}) and within (Q_{within}) subgroups (Hedges et al. 1999). According to the Chi-square test of the sum of squares between groups, if the 95% CIs of the subgroups did not overlap, the effects of P addition on the target variables were significantly different between subgroups (Deng et al. 2017).

Sensitivity analysis and publication bias

We conducted a sensitivity analysis to determine whether the independent individual experiments from previous studies affected the results of our study (Koricheva and Gurevitch 2014; Yue et al. 2017). First, we calculated the weight value of the target variables in response to P addition using a mixed model to minimize the occurrence of extreme variable effects. Second, we repeated the analysis of the results and determine the weighted RR by randomly sampling the case studies from each original study. The findings of these analyses indicated that reducing the number of observation samples did not affect the weighted RR results in our study (Additional file 1: Fig. S2). Furthermore, based on a previous study (Lu et al. 2011), a curve fitted by the Gaussian function was used to test the normal distribution of the main variable data, and the results of this analysis indicated that there is no publication bias in our results (Additional file 1: Fig. S3).

Statistical analysis

First, we performed a Pearson correlation analysis to assess the relationships between the RR of foliar N and P stoichiometry and soil nutrient availability (soil N and P availability) and nutrient resorption efficiency (NRE and PRE) and found that the RR of foliar N and P stoichiometry was mostly correlated with the soil P availability and PRE (Additional file 1: Table S2). A regression analysis was then performed to further quantify the relationships between these variables. Considering the relationships between variables, we performed partial least squares (PLS) regression in SIMCA 14.0 (Umetrics, Umeå, Sweden) to estimate the relative importance

of these factors to the RR of foliar N and P stoichiometry under P addition with and without N addition. We used the PLS standardized coefficients to determine the directions and magnitudes of the effects of these variables in explaining the response ratio of foliar N and P stoichiometry, and the relative importance of these factors was evaluated based on the variable importance values. The correlation analyses and regression analyses were conducted with SPSS software (SPSS 20.0 for Windows; SPSS Inc., Chicago, IL, USA), and the graphs were drawn using Origin (version 8.0).

Results

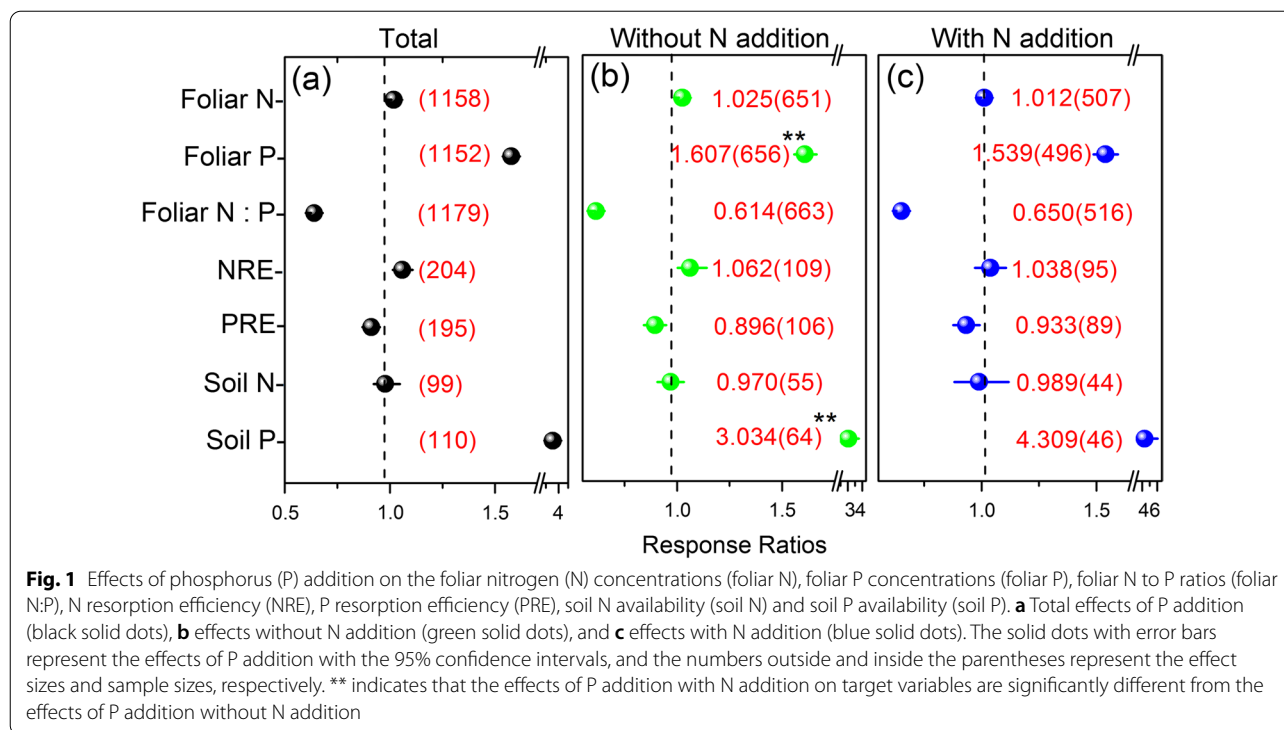
Comparison of the effects of P addition with and without N addition

On a global scale, P addition significantly enhanced foliar N concentrations by 1.9%, foliar P concentrations by 57.7%, NRE by 5.9%, and soil P availability by 251.4% but decreased the foliar N:P ratio by 36% and PRE by 8.8% (Fig. 1a). However, P addition without N addition promoted observable increases in the foliar N concentrations and NRE (Fig. 1b, c). Furthermore, the positive effects of P addition on foliar P concentrations were greater without N addition than with N addition, whereas the opposite pattern was found for the soil P availability (Fig. 1b, c, Additional file 1: Table S1). Moreover, P addition reduced the N:P ratio and PRE, but these effects were not regulated by N addition (Fig. 1bc, Additional file 1: Table S1).

Factors influencing the effects of P addition with and without N addition

The changes in the foliar N concentrations in response to P addition with and without N addition were significantly affected by the experimental design parameters (Fig. 2, Additional file 1: Table S1). P addition rates less than 5 g m^{-2} , significantly increased the foliar N concentrations, and the effect showed an evident difference among different P addition rates with N addition (Fig. 2, Additional file 1: Table S1). Additionally, P addition clearly increased foliar N concentrations with and without N addition in the experiments with durations ≤ 3 years. The response of foliar N concentrations to P addition did not differ significantly between the plant community and species levels, and P addition without N addition markedly enhanced the foliar N concentration. However, P addition significantly increased the foliar N concentrations in evergreen plants and not in other plant growth types. Moreover, P addition affected foliar N concentration at the community level, and the foliar N concentration in evergreen plants was higher without N addition than with N addition (Fig. 2, Additional file 1: Table S1).

In general, P addition with and without N addition clearly enhanced foliar P concentrations under all



conditions, but significant differences were found among the experimental durations, P addition rates, and plant growth types (Fig. 3, Additional file 1: Table S1). For example, the increases in foliar P concentrations were significant for all P addition rates, and the greatest increase were obtained when P exceeded 15 g m⁻². However, P addition evidently promoted foliar P concentrations at the community and species levels, but no significant differences were found between the community level and the species level. Additionally, we found that the positive effects of short-term (≤ 3 years) or low-dose (< 5 g m⁻²) P addition on foliar P concentrations were greater without N addition than with N addition (marginally significant differences were found with the low-dose P addition). Moreover, the P-induced increases in foliar P concentrations at the species level and in evergreen plants were greater without N addition than with N addition (Fig. 3, Additional file 1: Table S1).

Overall, the foliar N:P ratios were clearly decreased by P addition with and without N addition, regardless of the experimental duration, P addition rate, and plant growth type (Fig. 4, Additional file 1: Table S1). In addition, the effects of P addition with and without N addition on the RRs differed significantly among different P addition rates and plant growth types. However, regardless of N addition, the effects of P addition were not significantly different between the community and species levels. Furthermore, the changes in the foliar N:P ratios of evergreen plants in

response to P addition were greater without N addition than with N addition (Fig. 4, Additional file 1: Table S1).

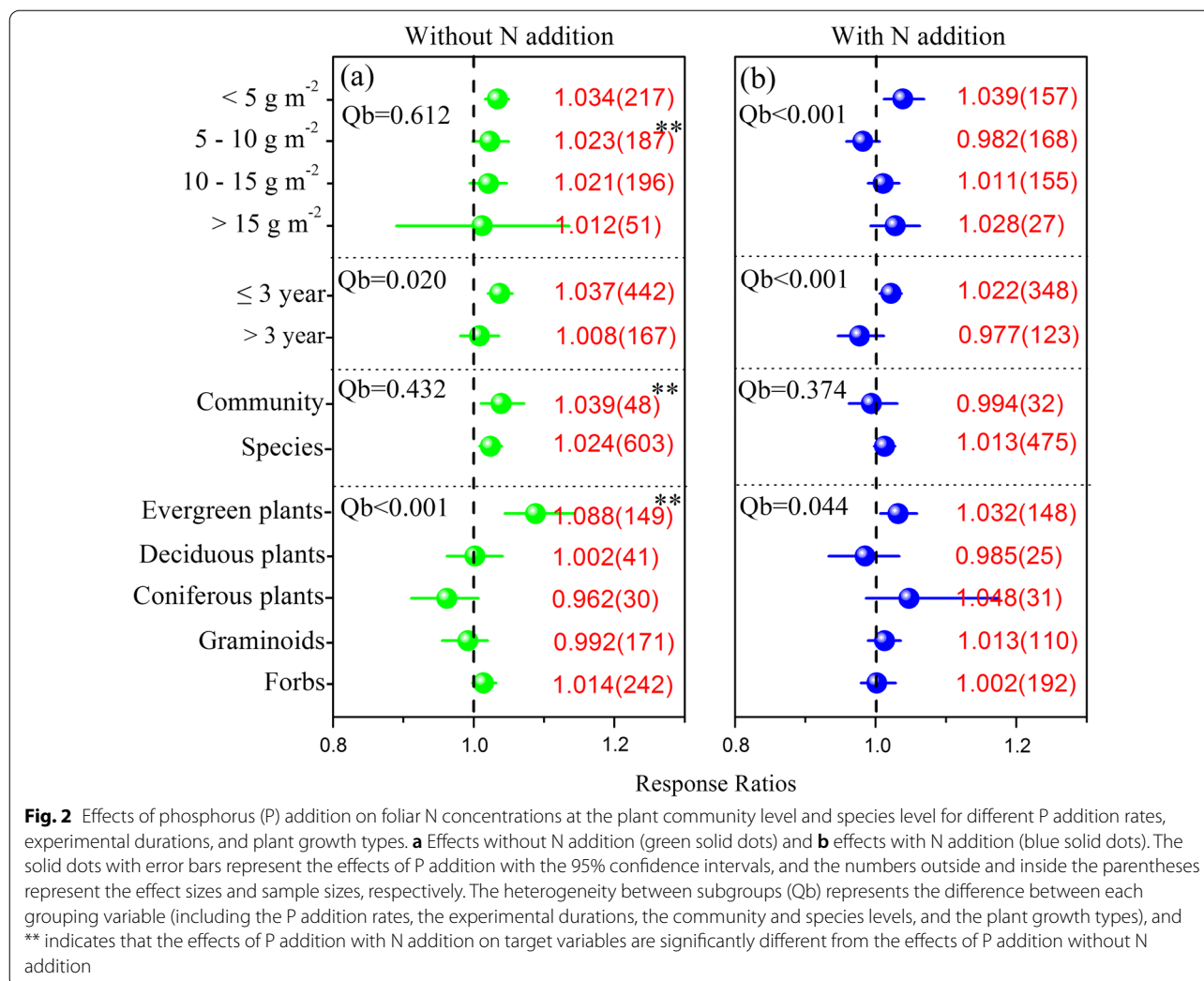
Relationships between foliar N and P stoichiometry and environmental factors

The correlation analysis showed that the RR of foliar N and P stoichiometry was mostly correlated with the soil P availability and PRE (Additional file 1: Table S2). The regression analysis revealed that under P addition with N addition, the RRs of the foliar P concentrations increased, but the foliar N:P ratios decreased with increasing PRE ($R^2=0.063$, 0.05; both $p<0.05$, Fig. 5b, c). Additionally, increases in the soil P availability induced increases in the foliar N (except N concentration under P addition with N addition, $R^2=0.02$) and P concentrations ($R^2=0.172$, 0.118), whereas the foliar N:P ratios ($R^2=0.198$, 0.141) decreased under P addition without and with N addition (all $p<0.05$, Fig. 5d–f). Furthermore, the PLS analysis showed that regardless of N addition, the effects of P addition on foliar N and P stoichiometry were strongly driven by the soil P availability and not by the PRE, because the soil P availability directly regulated the PRE (Fig. 6, Additional file 1: Table S2).

Discussion

Effects of P addition on foliar N and P stoichiometry

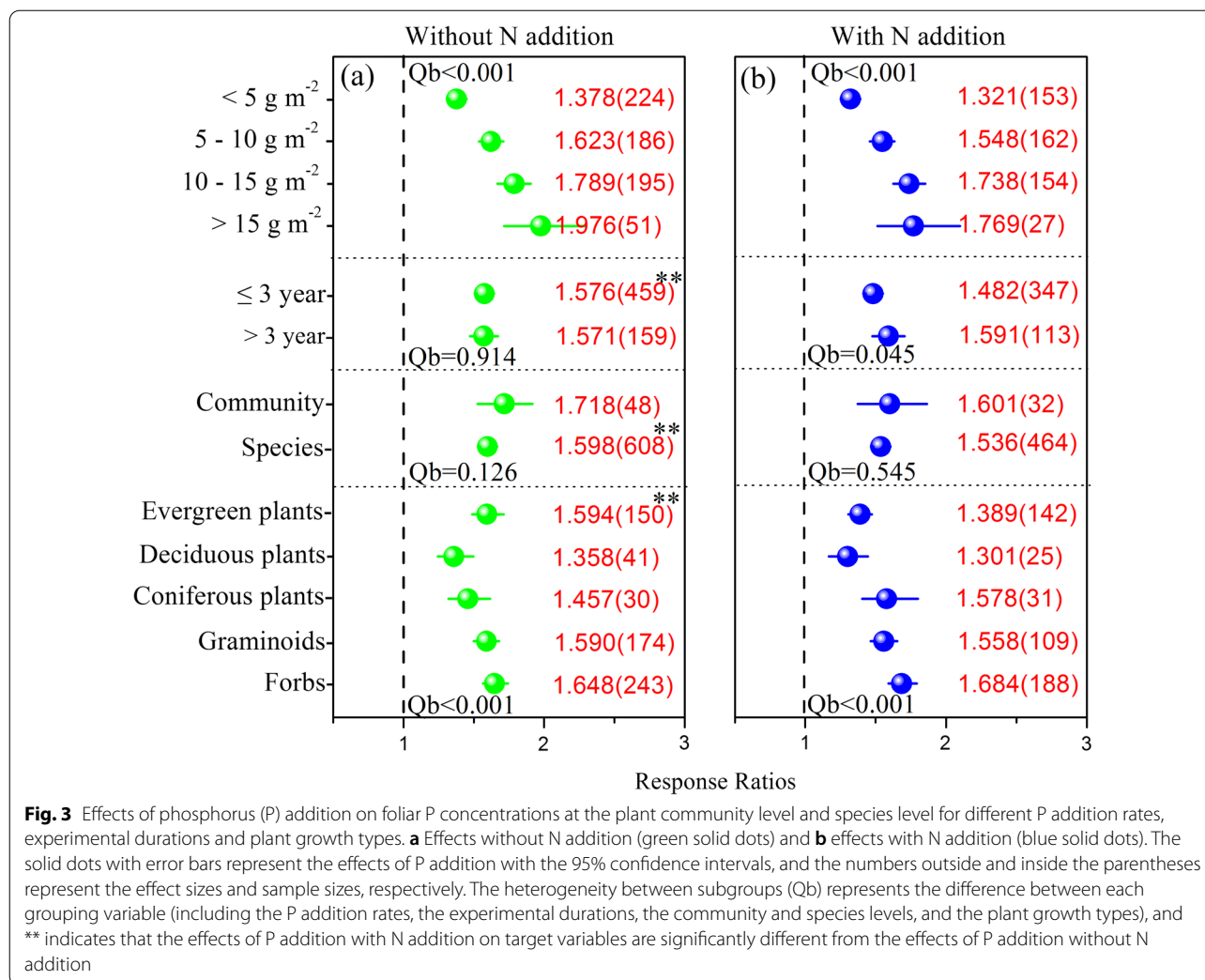
Previous studies showed that P addition evidently improved foliar P concentrations, but only slightly



affected N concentrations and thus distinctly decreased the N:P ratios (Li et al. 2016; Mayor et al. 2014; Yuan and Chen 2015b). However, we found that the positive effects of P addition on foliar N concentrations were weaker than those on foliar P concentrations, which yielded significant reductions in the foliar N:P ratios at the global scale (Fig. 1). These results can be explained by the finding that P addition significantly promoted soil P availability (Fig. 1), which alleviated P limitation but resulted in N deficiency in plants. To compensate for N deficiency and to maintain a balance between N and P in leaves, plants will alter their nutrient use efficiencies and nutrient resorption efficiency (He and Dijkstra 2015; Houlton et al. 2008; Iversen et al. 2010; van Huysen et al. 2016). For instance, our study also demonstrated that P addition promoted increases in the foliar P concentration by improving soil P availability but may have increased the foliar N concentration by promoting NRE (Figs. 1, 5). Additionally, previous studies have shown that P

enrichment can extensively improve soil P availability and therefore increase foliar P concentrations (Gao et al. 2018), whereas P addition can also clearly enhance the availability of soil N (Reed et al. 2007; Rejmánková and Snyder 2008) or NRE (Huang et al. 2016; Yan et al. 2015) to increase foliar N concentrations. Thus, these findings reveal that P addition not only changes the cycling rate or pathway of N and P but also induces plants to mediate their nutrient use efficiency to support the balance between foliar N and P.

However, when considering the effects of P addition with and without N addition on foliar N and P stoichiometry, we found two interesting phenomena. First, P addition without N addition evidently promoted increases in the foliar N concentration and NRE, but P addition with N addition did not affect these variables (Fig. 1). This is because combined N and P addition can alleviate the limitations of N and P on plant growth and development, but the potential increases in foliar N

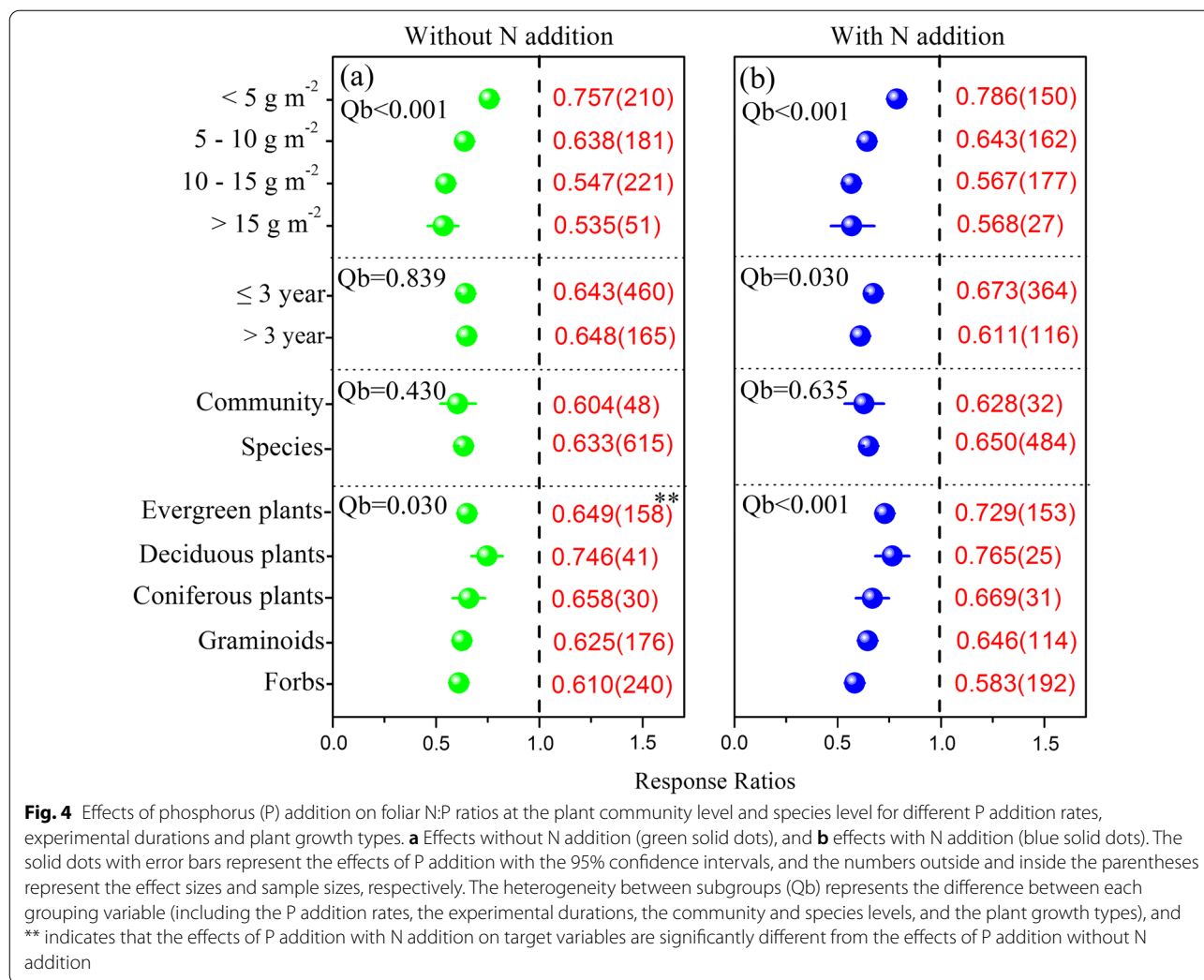


are insufficient to balance the foliar N used by plants, e.g., to increase plant biomass (Elser et al. 2007; Harpole et al. 2011; Li et al. 2016). A previous study also demonstrated that the positive effects of P addition on aboveground biomass were stronger with N addition than without N addition (Li et al. 2016). Second, the positive effect of P addition on soil P availability was greater with N addition than without N addition, whereas the opposite effect was found for foliar P concentrations. These results may be explained by the fact that N addition can increase the rate of release of organic matter (Knorr et al. 2005; Lü et al. 2013) and can alter phosphatase activity (Fujita et al. 2010) as well as mycorrhizal activity (Rowe et al. 2008) to increase soil P availability. However, the effects of P addition on P consumption and dilution in plants might be greater with N addition than without N addition (Sardans et al. 2016; Vitousek et al. 2010). These results suggest that P addition without N addition alleviates N limitation in

plants, but compared to P addition without N addition, P addition with increased N deposition levels might aggravate the limitation of P in plants. Furthermore, these findings reveal that the relationship between foliar N and P is subject to complex coupling constraints, which can explain why a change in one nutrient element might correspondingly alter the availability of another nutrient element.

Factors influencing the effects of P addition

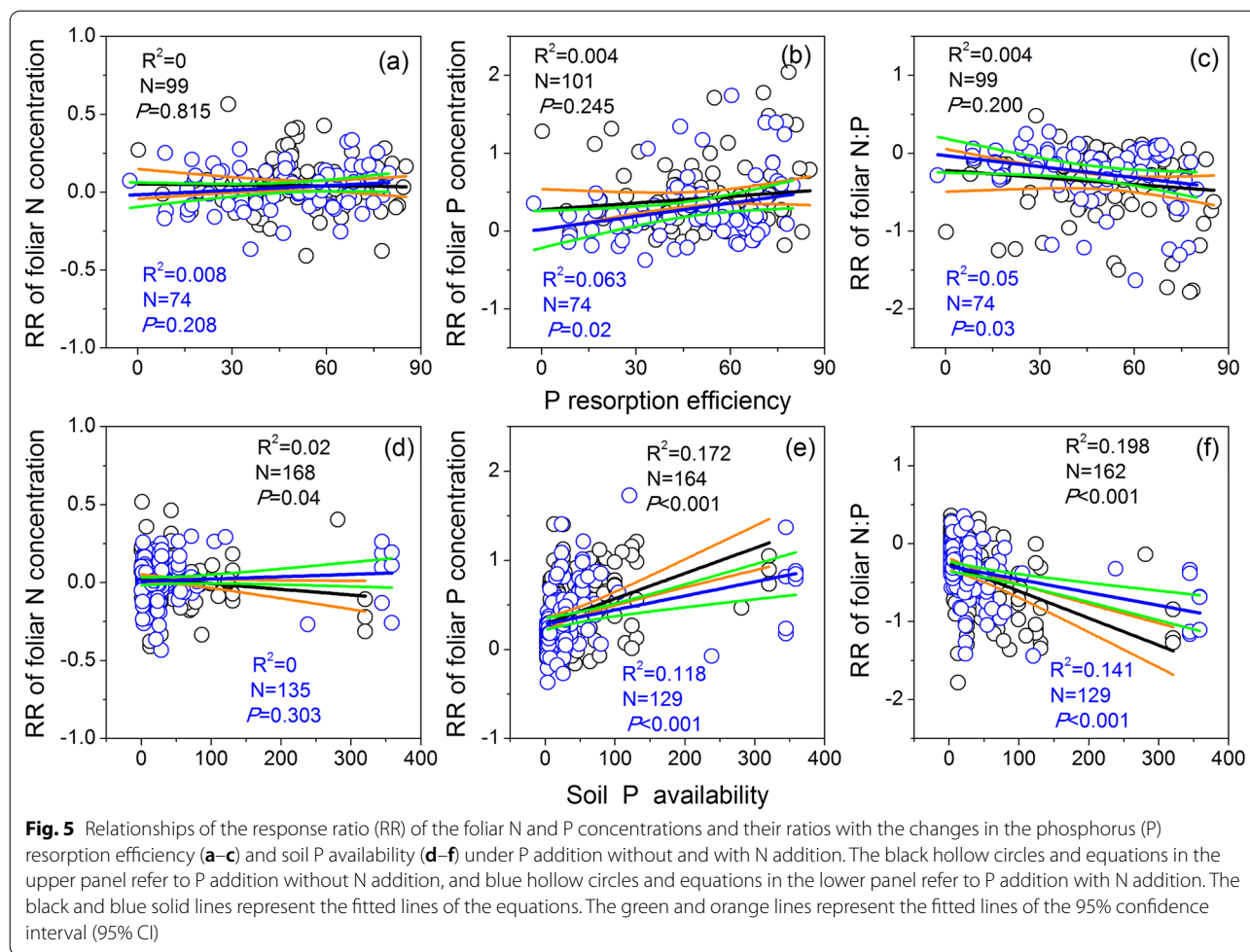
We found that low-dose or short-term P addition without and with N addition significantly increased the foliar N concentration (Fig. 2). This phenomenon reflects the fact that P addition can alleviate P limitation in ecosystems (Li et al. 2016) and promote the N cycle (Yan et al. 2015), but as the P addition rates and experimental durations increase, the ecosystem can gradually shift the nutrient limitation status of plants from P or N and P to N (Burson et al. 2016; Mischler et al. 2014). In addition,



the positive effects of low-dose or short-term P additions on foliar P concentrations were greater without N addition than with N addition (Fig. 3). These results can be attributed to P addition enhancing plant assimilation, and long-term or high-dose P addition is sufficient to compensate for P deficiency in plants due to N addition (Cleveland et al. 2011; Li et al. 2016). These results might explain why the addition of one nutrient element does not immediately lead to the lack of another nutrient element in plants (Finzi 2009), and further indicate that the biochemical processes in ecosystems can be altered by nutrient addition, which results in the relative limitation of other nutrients in plants (Marklein and Houlton 2012).

Additionally, we found that P addition without and with N addition affected the foliar N and P stoichiometry at the community and species levels, but the effects showed significant differences among plant growth types (Figs. 2, 3, 4). This finding is supported by the following findings: first, changes in foliar N and

P stoichiometry at the community level are mainly driven by the N and P stoichiometry of the dominant species (Grime 1998; Tessier and Raynal 2003). Second, the effects of P addition were significantly different among plant growth types, which is most likely due to their unique ecological and biological properties (e.g., nutrient use strategies, adaptabilities and nutrient-limited conditions) (Tian et al. 2019; Townsend and Asner 2013; You et al. 2018). However, different from the effects on other plant growth types, P addition significantly enhanced the foliar N concentration in evergreen plants, and the effects of P addition on the foliar N and P stoichiometry in evergreen plants were greater without N addition than with N addition (Figs. 2, 3, 4). Two mechanisms explain this phenomenon. First, in comparison with other plant growth types (particularly deciduous species), evergreen plants utilize conservative nutrient use strategies and have lower leaf N concentrations and photosynthetic N use efficiencies

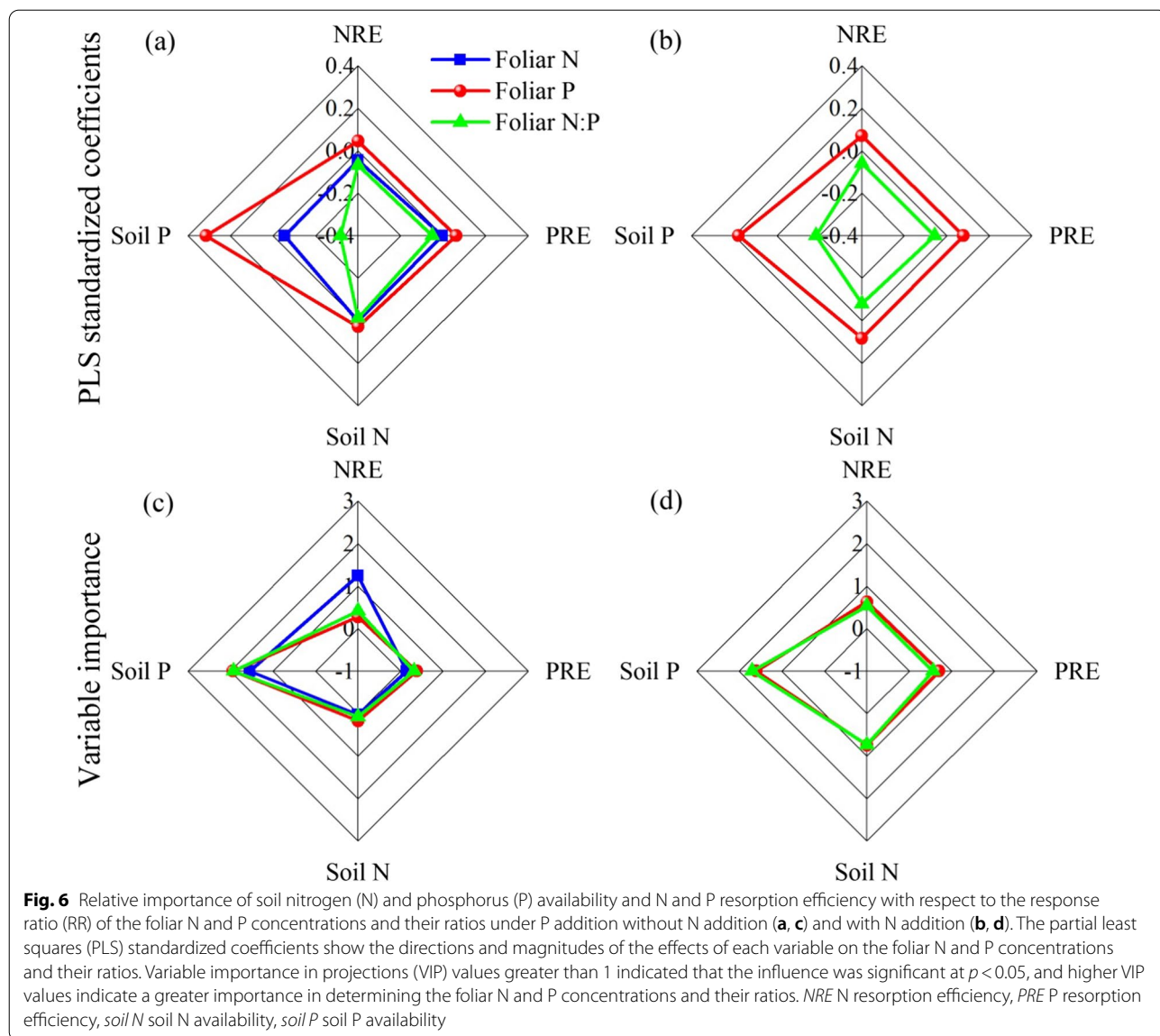


(Chapin et al. 1980; Takashima et al. 2004); thus, they can grow in barren and high-stress environments (Chapin et al. 1980; Zamin et al. 2014) and are more sensitive to nutrient changes (Chapin et al. 1995). Second, P addition with N addition can alleviate the relative limitation of elements (Elser et al. 2007), and can better enhance the assimilation of plants and the effects of biomass dilution on the foliar N and P concentrations (Vitousek et al. 2010).

How plants maintain the balance of foliar N and P in response to P addition

We found that the foliar N:P ratios under P addition with and without N addition were not significantly different (Fig. 4). This result was obtained mainly because the N:P ratios changed by P addition with and without N addition reached the minimal value, as supported by the homeostatic mechanism (Sterner and Elser 2002; You et al. 2018). These results are supported by the finding that the foliar N:P ratios did not increase with increasing P addition rates (Additional file 1: Fig. S4). In our study, the

RR of the foliar N and P stoichiometry was mostly correlated with the soil P availability and PRE (Fig. 5, Additional file 1: Table S2). The results indicated that plants can regulate their uptake of soil-available nutrients and nutrient reabsorption to maintain the balance of foliar N and P stoichiometry in response to P addition (Fig. 5), which is consistent with the findings from previous studies (Li et al. 2016; Rejmánková and Snyder 2008; Yan et al. 2015). Additionally, we also found that the relative contributions of the PRE to maintain the balance of N and P in response to P addition were smaller than those of the soil P availability. These results might be explained as follows: P addition improved the soil P availability and then regulated the PRE (Additional file 1: Table S2). For example, previous studies have found that P addition can induce plants to take up more P from soil, which reduces their dependence on the PRE (Mao et al. 2015; Yan et al. 2015). Our PLS analysis also showed that regardless of N addition, the effects of P addition on foliar N and P stoichiometry were strongly driven by the soil P availability and not by the PRE. Therefore, these results indicate that



the soil P availability was more effective than the PRE in predicting the changes in foliar N and P stoichiometry in response to P addition.

Conclusions

Our study demonstrated that (1) P addition without N addition enhanced foliar N concentrations and NRE, but P addition with N addition did not affect these variables. The positive effects of P addition on foliar P concentrations were greater without N addition than with N addition, whereas the opposite effect was observed for the availability of soil P. These findings reveal that N enrichment can alter the foliar N and P stoichiometry in response to P addition, which suggests that we should consider the effects of experimental background

conditions on the experiment to better understand how global change affects plants. (2) Short-term or low-dose P addition with and without N addition can increase foliar N concentrations, and short-term or low-dose P addition induces plants to use P more efficiently with N addition than without N addition. These results suggest that the rates and durations of P addition regulate the effects of P addition on the foliar N and P stoichiometry. We conclude that experiments involving short-term or low-dose P addition might not provide convincing evidence that can be used to predict how long-term or high-dose P addition affects ecosystem processes, and more attention should be paid to the effects of long-term and high-dose P addition in future experiments. (3) The effects of P addition on foliar N and P stoichiometry were significantly

influenced by geographic and climatic factors (Additional file 1: Table S3) and plant growth type, suggesting that uniform standards for large-scale experiments involving nutrient addition along a rainfall or temperature gradient are needed to improve the predictions of how ecosystem processes will respond to global change. Together, these results indicate that increasing N deposition might drive the responses of the foliar N and P stoichiometry to P addition and demonstrate the important effects of the experimental environment on the results.

Abbreviations

P: Phosphorus; N: Nitrogen; Foliar P: Foliar P concentration; Foliar N:P: Foliar N to P ratio; NRE: N resorption efficiency; PRE: P resorption efficiency; Soil N: Soil N availability; Soil P: Soil P availability; MAP: Mean annual precipitation; MAT: Mean annual temperature; Qb: Heterogeneity between subgroups; RR: Response ratio; PLS: Partial least squares; VIP: Variable importance in projections.

Supplementary Information

The online version contains supplementary material available at <https://doi.org/10.1186/s13717-021-00329-x>.

Additional file 1: Figure S1. Global distribution of the study sites included in the meta-analysis. **Figure S2.** Repeat analysis of the effects of phosphorus (P) addition on the foliar nitrogen (N) concentrations (a), foliar P concentrations (b), and foliar N to P ratios (c) for different sample sizes. **Figure S3.** Frequency distribution of the response ratio (RR) of phosphorus (P) addition on the foliar nitrogen (N) concentrations (a), foliar P concentrations (b), and foliar N to P ratios (c). The curves were fitted by a Gaussian function. **Figure S4.** Response ratios of the foliar nitrogen (N): phosphorus (P) ratios to the P addition rates (a) without N addition (b) with N addition. Different letters indicate significant differences between the P addition rates at the 0.05 level. **Table S1.** Comparison of the effects of phosphorus (P) addition with or without N addition on the foliar nitrogen (N) concentrations (Foliar N), foliar P concentrations (Foliar P), foliar N to P ratios (Foliar N:P), N resorption efficiency (NRE), P resorption efficiency (PRE), soil N availability (Soil N) and soil P availability (Soil P). The effect sizes indicate the mean of the effect of P addition on the variables, and 95% CI represents the 95% confidence interval. The heterogeneity between subgroups (Qb, QB) is significant at $p < 0.05$; Qb represents the difference between each grouping variable (including the P addition rates, the experimental durations, the community and species levels and the plant growth types), and QB indicates that the effects of P addition with N addition on the target variables are different from those without N addition. **Table S2.** Correlation analysis of the relationship between the response ratio (RR) of the foliar nitrogen concentration (N), phosphorus (P), N to P ratios (N:P) and N and P resorption efficiencies (NRE and PRE, respectively), and soil N and P availabilities (Soil N and Soil P, respectively). The numbers in black in the upper right refer to P addition without N addition, and the numbers in blue in the lower left refer to P addition with N addition. The numbers inside and outside the parentheses represent the sample sizes and correlation coefficients, respectively. * and ** indicate the relationship is significant at the $p < 0.05$ and $p < 0.01$ level, respectively. **Table S3.** F-values from the regression analysis of the relationship between the response ratios (RRs) of foliar nitrogen (N) concentrations (Foliar N), foliar phosphorus (P) concentrations (Foliar P), foliar N to P ratios (Foliar N:P ratios), N and P resorption efficiencies (NRE and PRE, respectively) and soil N and P availabilities (Soil N and Soil P, respectively) to P addition with and without N addition and geographical [including latitude ($^{\circ}$), longitude ($^{\circ}$) and altitude (m)] and climatic factors [i.e., mean annual precipitation (MAP, mm) and the mean annual temperature (MAT, $^{\circ}$ C)]. **Appendix S1.** List of 168 articles from which data were extracted for the meta-analysis.

Acknowledgements

We are grateful to all scientists whose research was used to design and perform this meta-analysis. We also appreciate the contributions from Dr. Yinzhao Liu to this manuscript.

Authors' contributions

PK, PC and TB conceived the idea for the study. CY, XZ, TB and LY designed the structure of the article. CY, YR, LH, WL, and LS collected the data. CY, LL, and ZL analyzed the data. All of the authors contributed to the interpretation of the results and the writing of the article. All authors read and approved the final manuscript.

Funding

This study was supported by the National Natural Science Foundation of China (Grant Nos. 31870602, 31901295, and 32071745), the Program of Sichuan Excellent Youth Sci-Tech Foundation (20JCQN0145), the Chinese Postdoctoral Science Foundation (2020M673278), and the CAS "Light of West China" Program (Y8C2021). Chengming You also acknowledges the China Scholarship Council for supporting a Ph.D. programme Grant (201806910030).

Availability of data and materials

The datasets used and/or analyzed in the current study are available from the corresponding author on reasonable request or the related researches in the Additional files.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Author details

¹Forestry Ecological Engineering in the Upper Reaches of the Yangtze River Key Laboratory of Sichuan Province and National Forestry and Grassland Administration Key Laboratory of Forest Resources Conservation and Ecological Safety on the Upper Reaches of the Yangtze River and Rainy Area of West China Plantation Ecosystem Permanent Scientific Research Base, Institute of Ecology and Forestry, Sichuan Agricultural University, 211 Huimin Road, Wenjiang District, Chengdu 611130, Sichuan, People's Republic of China. ²Department of Biological Science, Institute of Environmental Sciences, University of Quebec at Montreal, Case Postale 8888, Succursale Centre-Ville, Montreal, QC H3C 3P8, Canada. ³Laboratory for Ecological Forecasting and Global Change, College of Forestry, Northwest A & F University, No. 3 Taicheng Road, Yangling 712100, People's Republic of China. ⁴Department of Community Ecology, Helmholtz-Centre for Environmental Research-UFZ, Theodor-Lieser-Strasse 4, 06110 Halle, Saale, Germany. ⁵College of Animal Science and Technology, Sichuan Agricultural University, Chengdu 611130, People's Republic of China. ⁶Department of Forest Ecology and Management, Swedish University of Agricultural Science, 90183 Umeå, Sweden.

Received: 13 May 2021 Accepted: 17 August 2021

Published online: 24 August 2021

References

- Aerts R, Chapin III FS (1999) The mineral nutrition of wild plants revisited: a re-evaluation of processes and patterns. *Adv Ecol Res* 30:1–67
- Ågren GI, Wetterstedt JM, Billberger MF (2012) Nutrient limitation on terrestrial plant growth - modeling the interaction between nitrogen and phosphorus. *New Phytol* 194:953–960
- Burson A, Stomp M, Akil L, Brussaard CP, Huisman J (2016) Unbalanced reduction of nutrient loads has created an offshore gradient from phosphorus to nitrogen limitation in the North Sea. *Limnol Oceanogr* 61:869–888

- Chapin III FS, Johnson DA, Mckendrick JD (1980) Seasonal movement of nutrients in plants of differing growth form in an Alaskan tundra ecosystem: implications for herbivory. *J Ecol* 189–209
- Chapin FS III, Shaver GR, Giblin AE, Nadelhoffer KJ, Laundre JA (1995) Responses of arctic tundra to experimental and observed changes in climate. *Ecology* 76:694–711
- Chen Y, Han W, Tang L, Tang Z, Fang J (2013) Leaf nitrogen and phosphorus concentrations of woody plants differ in responses to climate, soil and plant growth form. *Ecography* 36:178–184
- Cleveland CC, Townsend AR, Taylor P, Alvarez-Clare S, Bustamante MMC, Chuyong G, Dobrowski SZ, Grierson P, Harms KE, Houlton BZ (2011) Relationships among net primary productivity, nutrients and climate in tropical rain forest: a pan-tropical analysis. *Ecol Lett* 14:939–947
- Demars BO, Edwards A (2007) Tissue nutrient concentrations in freshwater aquatic macrophytes: high inter-taxon differences and low phenotypic response to nutrient supply. *Freshwater Biol* 52:2073–2086
- Deng Q, Hui D, Dennis S, Reddy KC (2017) Responses of terrestrial ecosystem phosphorus cycling to nitrogen addition: a meta-analysis. *Global Ecol Biogeogr* 26:713–728
- Deng M, Liu L, Jiang L, Liu W, Li S, Yang S, Wang B (2018) Ecosystem scale trade-off in nitrogen acquisition pathways. *Nat Ecol Evol* 2:1724–1734
- Dijkstra FA, Pendall E, Morgan JA, Blumenthal DM, Carrillo Y, Lecaín DR, Follett RF, Williams DG (2012) Climate change alters stoichiometry of phosphorus and nitrogen in a semiarid grassland. *New Phytol* 196:807–815
- Du E, Terrer C, Pellegrini AF, Ahlström A, van Lissa CJ, Zhao X, Jackson RB (2020) Global patterns of terrestrial nitrogen and phosphorus limitation. *Nat Geosci* 13:221–226
- Elser JJ, Bracken ME, Cleland EE, Gruner DS, Hillebrand H, Ngai JT, Seabloom EW, Shurin JB, Smith JE (2007) Global analysis of nitrogen and phosphorus limitation of primary producers in freshwater, marine and terrestrial ecosystems. *Ecol Lett* 10:1135–1142
- Elser JJ, Fagan W, Kerkhoff A, Swenson N, Enquist B (2010) Biological stoichiometry of plant production: metabolism, scaling and ecological response to global change. *New Phytol* 186:593–608
- Finzi AC (2009) Decades of atmospheric deposition have not resulted in widespread phosphorus limitation or saturation of tree demand for nitrogen in southern New England. *Biogeochemistry* 92:217–229
- Fujita Y, Robroek BJ, De Ruiter PC, Heil GW, Wassen MJ (2010) Increased N affects P uptake of eight grassland species: the role of root surface phosphatase activity. *Oikos* 119:1665–1673
- Gao Y, Cooper DJ, Zeng X (2018) Nitrogen, not phosphorus, enrichment controls biomass production in alpine wetlands on the Tibetan Plateau, China. *Ecol Eng* 116:31–34
- Grime J (1998) Benefits of plant diversity to ecosystems: immediate, filter and founder effects. *J Ecol* 86:902–910
- Harpole WS, Ngai JT, Cleland EE, Seabloom EW, Borer ET, Bracken ME, Elser JJ, Gruner DS, Hillebrand H, Shurin JB, Smith JE (2011) Nutrient co-limitation of primary producer communities. *Ecol Lett* 14:852–862
- He M, Dijkstra FA (2014) Drought effect on plant nitrogen and phosphorus: a meta-analysis. *New Phytol* 204:924–931
- He M, Dijkstra FA (2015) Phosphorus addition enhances loss of nitrogen in a phosphorus-poor soil. *Soil Biol Biochem* 82:99–106
- Hedges LV, Gurevitch J, Curtis PS (1999) The meta-analysis of response ratios in experimental ecology. *Ecology* 80:1150–1156
- Houlton BZ, Wang YP, Vitousek PM, Field CB (2008) A unifying framework for dinitrogen fixation in the terrestrial biosphere. *Nature* 454:327–330
- Huang J, Yu H, Lin H, Zhang Y, Searle EB, Yuan Z (2016) Phosphorus amendment mitigates nitrogen addition-induced phosphorus limitation in two plant species in a desert steppe, China. *Plant Soil* 399:221–232
- Iversen CM, Bridgman SD, Kellogg LE (2010) Scaling plant nitrogen use and uptake efficiencies in response to nutrient addition in peatlands. *Ecology* 91:693–707
- Jiang J, Wang YP, Yang YH, Yu XM, Wang C, Yan JH (2019) Interactive effects of nitrogen and phosphorus additions on plant growth vary with ecosystem type. *Plant Soil* 440:523–537
- Knorr M, Frey S, Curtis P (2005) Nitrogen additions and litter decomposition: a meta-analysis. *Ecology* 86:3252–3257
- Koricheva J, Gurevitch J (2014) Uses and misuses of meta-analysis in plant ecology. *J Ecol* 102:828–844
- Li Y, Niu S, Yu G (2016) Aggravated phosphorus limitation on biomass production under increasing nitrogen loading: a meta-analysis. *Global Change Biol* 22:934–943
- Li Z, Tian D, Wang B, Wang J, Wang S, Chen HY, Xu X, Wang C, He N, Niu S (2018) Microbes drive global soil nitrogen mineralization and availability. *Global Change Biol* 25:1078–1088
- Liu X, Zhang Y, Han W, Tang A, Shen J, Cui Z, Vitousek P, Erisman JW, Keith GD, Christie P, Fangmeier A, Zhang F (2013) Enhanced nitrogen deposition over China. *Nature* 494:459–462
- Lu M, Yang Y, Luo Y, Fang C, Zhou X, Chen J, Yang X, Li B (2011) Responses of ecosystem nitrogen cycle to nitrogen addition: a meta-analysis. *New Phytol* 189:1040–1050
- Lü XT, Reed S, Yu Q, He NP, Wang ZW, Han XG (2013) Convergent responses of nitrogen and phosphorus resorption to nitrogen inputs in a semiarid grassland. *Global Change Biol* 19:2775–2784
- Mahowald N, Jickells TD, Baker AR, Artaxo P, Benitez-Nelson CR, Bergametti G, Bond TC, Chen Y, Cohen DD, Herut B, Kubilay N, Losno R, Luo C, Maenhaut W, McGee KA, Okin GS, Siefert RL, Tsukuda S (2008) Global distribution of atmospheric phosphorus sources, concentrations and deposition rates, and anthropogenic impacts. *Global Biogeochem Cycles* 22:GB4026
- Mao R, Zeng DH, Zhang XH, Song CC (2015) Responses of plant nutrient resorption to phosphorus addition in freshwater marsh of Northeast China. *Sci Rep* 5:8097
- Mao R, Chen HM, Zhang XH, Shi FX, Song CC (2016) Effects of P addition on plant C:N:P stoichiometry in an N-limited temperate wetland of Northeast China. *Sci Total Environ* 559:1–6
- Marklein AR, Houlton BZ (2012) Nitrogen inputs accelerate phosphorus cycling rates across a wide variety of terrestrial ecosystems. *New Phytol* 193:696–704
- Mayor JR, Wright SJ, Turner BL (2014) Species-specific responses of foliar nutrients to long-term nitrogen and phosphorus additions in a lowland tropical forest. *J Ecol* 102:36–44
- Mischler JA, Taylor PG, Townsend AR (2014) Nitrogen limitation of pond ecosystems on the plains of eastern Colorado. *PLoS ONE* 9:e95757
- Mo Q, Li ZA, Sayer EJ, Lambers H, Li Y, Zhou B, Tang J, Heskell M, Ding Y, Wang F (2019) Foliar phosphorus fractions reveal how tropical plants maintain photosynthetic rates despite low soil phosphorus availability. *Funct Ecol* 33:503–513
- Niu S, Classen AT, Dukes JS, Kardol P, Liu L, Luo Y, Rustad L, Sun J, Tang J, Templer PH (2016) Global patterns and substrate-based mechanisms of the terrestrial nitrogen cycle. *Ecol Lett* 19:697–709
- Ostertag R (2010) Foliar nitrogen and phosphorus accumulation responses after fertilization: an example from nutrient-limited Hawaiian forests. *Plant Soil* 334:85–98
- Peñuelas J, Poulter B, Sardans J, Ciais P, Velde MVD, Bopp L, Boucher O, Godderis Y, Hinsinger P, Llusia J (2013) Human-induced nitrogen-phosphorus imbalances alter natural and managed ecosystems across the globe. *Nat Commun* 4:2934
- Reed SC, Seastedt TR, Mann CM, Suding KN, Townsend AR, Cherwin KL (2007) Phosphorus fertilization stimulates nitrogen fixation and increases inorganic nitrogen concentrations in a restored prairie. *Appl Soil Ecol* 36:238–242
- Reich PB, Oleksyn J, Wright IJ, Niklas KJ, Hedin L, Elser JJ (2009) Evidence of a general 2/3-power law of scaling leaf nitrogen to phosphorus among major plant groups and biomes. *Proc Biol Sci* 277:877–883
- Rejmánková E, Snyder JM (2008) Emergent macrophytes in phosphorus limited marshes: do phosphorus usage strategies change after nutrient addition? *Plant Soil* 313:141–153
- Rowe EC, Smart SM, Kennedy VH, Emmett BA, Evans CD (2008) Nitrogen deposition increases the acquisition of phosphorus and potassium by heather *Calluna vulgaris*. *Environ Pollut* 155:201–207
- Sardans J, Alonso R, Janssens IA, Carnicer J, Vereseglou S, Rillig MC, Fernández-Martínez M, Sanders TGM, Peñuelas J (2016) Foliar and soil concentrations and stoichiometry of nitrogen and phosphorus across European *Pinus sylvestris* forests: relationships with climate, N deposition and tree growth. *Funct Ecol* 30:676–689
- See CR, McCormack ML, Hobbie SE, Flores-Moreno H, Silver WL, Kennedy PG (2019) Global patterns in fine root decomposition: climate, chemistry, mycorrhizal association and woodiness. *Ecol Lett* 22:946–953

- Sterner RW, Elser JJ (2002) Ecological stoichiometry: the biology of elements from molecules to the biosphere. Princeton, New Jersey: Princeton University Press
- Sundqvist MK, Liu Z, Giesler R, Wardle DA (2014) Plant and microbial responses to nitrogen and phosphorus addition across an elevational gradient in subarctic tundra. *Ecology* 95:1819–1835
- Takashima T, Hikosaka K, Hirose T (2004) Photosynthesis or persistence: nitrogen allocation in leaves of evergreen and deciduous *Quercus* species. *Plant Cell Environ* 27:1047–1054
- Tessier JT, Raynal DJ (2003) Use of nitrogen to phosphorus ratios in plant tissue as an indicator of nutrient limitation and nitrogen saturation. *J Appl Ecol* 40:523–534
- Tian D, Reich PB, Chen HY, Xiang Y, Luo Y, Shen Y, Cheng M, Han W, Niu S (2019) Global changes alter plant multi-element stoichiometric coupling. *New Phytol* 221:807–817
- Townsend AR, Asner GP (2013) Multiple dimensions of resource limitation in tropical forests. *Proc Natl Acad Sci USA* 110:4864–4865
- Van Huysen TL, Perakis SS, Harmon ME (2016) Decomposition drives convergence of forest litter nutrient stoichiometry following phosphorus addition. *Plant Soil* 406:1–14
- Viciedo DO, Prado RM, Martinez CA, Habermann E, Piccolo MC, Hurtado AC, Barreto RF, Calzada KP (2021) Changes in soil water availability and air-temperature impact biomass allocation and C:N:P stoichiometry in different organs of *Stylosanthes capitata* Vogel. *J Environ Manage* 278:111540
- Vitousek PM, Porder S, Houlton BZ, Chadwick OA (2010) Terrestrial phosphorus limitation: mechanisms, implications, and nitrogen-phosphorus interactions. *Ecol Appl* 20:5–15
- Wright I, Cannon K (2001) Relationships between leaf lifespan and structural defences in a low-nutrient, sclerophyll flora. *Funct Ecol* 15:351–359
- Yan Z, Kim N, Han W, Guo Y, Han T, Du E, Fang J (2015) Effects of nitrogen and phosphorus supply on growth rate, leaf stoichiometry, and nutrient resorption of *Arabidopsis thaliana*. *Plant Soil* 388:147–155
- You C, Wu F, Yang W, Xu Z, Tan B, Kai Y, Ni X (2018) Nutrient-limited conditions determine the responses of foliar nitrogen and phosphorus stoichiometry to nitrogen addition: a global meta-analysis. *Environ Pollut* 241:740–749
- Yuan Z, Chen HY (2015a) Decoupling of nitrogen and phosphorus in terrestrial plants associated with global changes. *Nat Clim Change* 5:465–469
- Yuan Z, Chen HY (2015b) Negative effects of fertilization on plant nutrient resorption. *Ecology* 96:373–380
- Yue K, Fornara DA, Yang W, Peng Y, Li Z, Wu F, Peng C (2017) Effects of three global change drivers on terrestrial C:N:P stoichiometry: a global synthesis. *Global Change Biol* 23:2450–2463
- Yue K, Peng Y, Fornara DA, Van Meerbeek K, Vesterdal L, Yang W, Peng C, Tan B, Zhou W, Xu Z, Ni X, Zhang L, Wu F (2019) Responses of nitrogen concentrations and pools to multiple environmental change drivers: a meta-analysis across terrestrial ecosystems. *Global Ecol Biogeogr* 28:690–724
- Zamin TJ, Bret-Harte MS, Grogan P (2014) Evergreen shrubs dominate responses to experimental summer warming and fertilization in Canadian mesic low arctic tundra. *J Ecol* 102:749–766

Publisher's Note

Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Submit your manuscript to a SpringerOpen[®] journal and benefit from:

- Convenient online submission
- Rigorous peer review
- Open access: articles freely available online
- High visibility within the field
- Retaining the copyright to your article

Submit your next manuscript at ► [springeropen.com](https://www.springeropen.com)
