Leaf nitrogen and phosphorus concentrations of woody plants differ in responses to climate, soil and plant growth form

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Leaf chemistry is important in predicting the functioning and dynamics of ecosystems. As two key traits, leaf nitrogen (N) and phosphorus (P) concentrations set the limits for plant growth, and leaf N:P ratios indicate the shift between N- and P-limitation. To understand the responses of leaf chemistry to their potential drivers, we measured leaf N and P concentrations of 386 woody species at 14 forest sites across eastern China, and explored the effects of climate, soil, and plant growth form on leaf N, P and N:P ratios. In general, leaf N and P were both negatively related to mean annual temperature and precipitation, and positively related to soil N and P concentrations. Leaf N:P ratios showed opposite trends. General linear models showed that variation in leaf N was mainly determined by a shift in plant growth form (from evergreen broadleaved to deciduous broadleaved to conifer species) along the latitudinal gradient, while variations in leaf P and N:P were driven by climate, plant growth form, and their interaction. These differences may reflect differences in nutrient cycling and physiological regulations of P and N. Our results should help understand the ecological patterns of leaf chemical traits and modeling ecosystem nutrient cycling.

Leaf nitrogen (N) and phosphorus (P) concentrations are two of the key leaf traits which play crucial roles in ecosystem function and dynamics (Chapin et al. 2002, Sterner and Elser 2002). As one of the major limiting elements for plant growth, leaf N concentration is tightly linked to many ecosystem processes, such as photosynthetic rate, plant productivity, and litter decomposition (Field and Mooney 1986, Vitousek 2004, LeBauer and Treseder 2008). As a vital element of genetic material, energy storage and cell structure, P is also limited in most environments. There is increasing evidence that leaf P concentration is probably more important than leaf N in limiting plant development and growth (Attiwill and Adams 1993, Zotz 2004, Reich et al. 2009). The ratio of leaf N to leaf P (N:P) is often suggested as an index to reflect the shift between N- and P-limitation (Koerselman and Meuleman 1996, Güsewell 2004).

Leaf N, P and N:P vary among species and sites, and P varies more than do N and N:P ratios (Güsewell and Koerselman 2002). However, the drivers of the greater variability in leaf P than in leaf N and N:P have not been systematically investigated, partly because of the challenge in differentiating between the effects of environmental variation and plant growth strategies on the variability of leaf chemistry (Ågren 2008, Elser et al. 2010).

Several factors have been proposed to explain the patterns of leaf chemical traits. Among these, climate, soil or plant growth form are thought to be the primary factors

influencing the leaf N and P. Likewise, several hypotheses have been put forward to explain geographical patterns of variability. For example, the plant physiology hypothesis assumes that all plant metabolic processes are temperature sensitive, and that increases in N and P concentration can compensate for decreases in metabolic rate at low temperature (Woods et al. 2003, Reich and Oleksyn 2004). However, experimental evidence at small scale and models indicate that the effects of temperature on leaf chemical traits are highly variable across a range of species and ecosystems, but our knowledge on the effects of temperature on leaf chemical traits at large scales is still limited (An et al. 2005, Rohrs-Richey and Mulder 2007, Lukac et al. 2010). The biogeochemical hypothesis suggests that soil nutrient availability, which is influenced by precipitation through leaching effects, is the main driver of leaf nutrient concentration (McGroddy et al. 2004, Reich and Oleksyn 2004). Soil P is mainly derived from rock weathering and its diffusion into the soil solution is considerably lower than that of N (Fitter et al. 1998, Lambers et al. 1998, Aerts and Chapin 2000), therefore leaf P may be more limited by low soil P content in highly leached soils (Han et al. 2005). The growth rate hypothesis states that leaf nutrient concentration is mainly determined by plant growth rate (Elser et al. 2003, Vrede et al. 2004); leaf N concentration [relative to carbon (C) content] increases linearly and P:C ratio follow a quadratic function of relative growth rate, and thus N:P responses are complicated to relate to growth rate (Ågren 2004).

The sensitive response in leaf P concentration to growth rate is also reflected in its responses to specific leaf area (SLA) and photosynthetic rate (Reich et al. 2010).

Numerous studies have tested these hypotheses at regional and global scales (Hedin 2004, Kerkhoff et al. 2005, He et al. 2006, Lovelock et al. 2007, Ordoñez et al. 2009). However, most studies have not examined the different responses of leaf N, P and N:P to environmental variables and plant growth form (but see Swenson et al.'s unpublished work, cited in Elser et al. 2010). In this paper we examine the responses of leaf N, P and N:P to climate, soil and plant growth form with data collected from 386 woody species at 14 forest sites across eastern China. The main objective is to understand the responses of leaf N, P and N:P to potential drivers (climate, soil and plant growth form). Specifically, we address the following questions: how do leaf nutrient concentrations vary along an environmental gradient? Are these environmental trends consistent across plant growth forms? Are there any differences in the responses of leaf N and P to their potential drivers?

Material and methods

Site description

This study was conducted at 14 forest sites in eastern China (Fig. 1), spanning a range of 32 degrees in latitude from 18.7°N to 50.9°N and varying in altitude from 80 to 1857 m. Climatic data, such as mean annual temperature (MAT) and annual precipitation (AP) were documented from local reports (Supplementary material Table A1). MAT ranged from –5.7 to 25.3°C, and AP from 423 to 2031 mm across sites. Soil types also span a large variation in nutrient level, from brown soils with high organic content to tropical red soils with low organic content. Vegetation types changed north to south from temperate coniferous to temperate deciduous, subtropical evergreen, and tropical seasonal rain forests.

Sampling and measurement

Leaf samples were collected according to the protocol proposed by Cornelissen et al. (2003). Fully expanded sun leaves were collected from four or five individuals of each species at the same site during the growing season (July–August) from 2005 to 2007. In total, 386 woody plant species, belonging to 208 genera and 74 families, were sampled.

Samples were dried for 72 h at 60°C and then ground using a ball mill (NM200, Retsch, Haan, Germany) for the measurement of N and P concentrations. Leaf N concentration was measured using an elemental analyzer (2400 II CHNS/O Elemental Analyzer, Perkin-Elmer, USA) with a combustion temperature of 950°C and a reduction temperature of 640°C. Leaf P concentration was measured by a molybdate/ascorbic acid method (John 1970) after H_2SO_4-$HClO_4$ digestion, and the absorbance of each sample was measured at 700 nm after 20 min of adding molybdenum-stibium-ascorbic acid reagent.

In each site three soil samples were collected in three plots where the plant species were sampled. Soil samples (A horizon) were randomly collected across the plot and
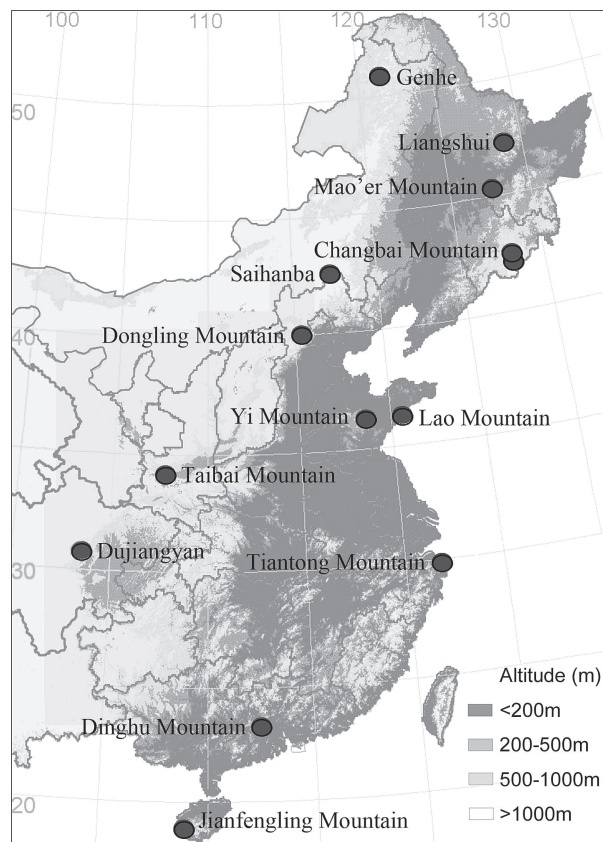

Figure 1. Locations of sampling sites, spanning 32 degrees in latitude across forests of eastern China.

thoroughly mixed. All soil samples were air-dried, sieved and roots removed by hand, then ground to pass through a 100-mesh sieve. Soil N concentration was measured using the elemental analyzer. Soil P concentration was measured by the molybdate/ascorbic acid method (John 1970). We took the average value of three samples at the same site as the soil N and P concentrations of the site.

Data analysis

The relationship between leaf chemical traits and environmental variables was explored at two levels: individual level and site × species level. For the former we used all original data of leaf N, P and N:P (mass ratio) from individual plants, and for the latter we averaged values by species within each site (Supplementary material Table A2). Because the results generated from the two datasets were quite similar, in this paper we present only the results from site × species dataset for the comparison with previous researches (Reich and Oleksyn 2004, He et al. 2006). The site × species dataset was statistically summarized to show the mean, coefficient of variance (CV) and sample size of leaf N, P and N:P. We divided all the species into three plant growth forms: deciduous broadleaved, evergreen broadleaved and coniferous plants. We used t-tests with Bonferroni corrections to compare the differences of leaf chemical traits among plant growth forms.

We first related leaf N, P and N:P to latitude and environmental variables using linear regression. Leaf N, P and

N:P were log-transformed to normalize distributions. Environmental variables included both climate data (MAT and AP) and soil nutrient levels (soil N and P concentrations). Furthermore, in order to examine the effects of plant growth form on leaf N, P and N:P, we related leaf N, P and N:P to environmental variables within each plant growth form.

We used general linear models (GLM) to quantify the effects of climate, soil and plant growth form on leaf chemical traits. Climate, soil and plant growth form were treated as fixed factors, and site was treated as a random factor to account for the non-independence of leaf chemical trait within site. F-tests were conducted by dividing the mean sum of squares (MS) of fixed effect by a MS of random factor. Given that all the climatic (MAT and AP) and soil (soil N and P concentrations) variables are highly correlated with each other (Supplementary material Table A3), only one of the climate and soil variables was included in each main-effect model to avoid multiple collinearity in GLM (Supplementary material Table A4). The environmental variable which had significant effect on leaf chemical traits, and interaction term between plant growth form and the environmental variable were included in the final model. If more than one environmental variable was significant, Akaike information criterion (AIC) was used to select competing models: model with the lower AIC value was chosen as the final model (Supplementary material Table A4). Statistical analyses were performed using R 2.12.0 (R Development Core Team 2010).

Results

Statistics of leaf N, P and N:P

Leaf N concentration ranged from 8.4 to 56.4 mg g^{-1}, leaf P from 0.27 to 5.58 mg g^{-1} and N:P from 3.8 to 53.2 (Table 1). Leaf N, P and N:P varied 6-20 fold across species. They differed significantly among plant growth forms. Both leaf N and P were higher in deciduous broadleaved species than in evergreen broadleaved and coniferous species, whereas N:P was higher in evergreen broadleaved than in deciduous broadleaved and coniferous species (Table 1). Among the three leaf chemical traits, leaf P showed the greatest variation with an overall mean CV of 0.53, followed by N:P (CV = 0.41) and N (CV = 0.31) (Table 1). Variation

Table 1. Statistics for leaf N, P and N:P of woody plants in forests in eastern China.

	N (mg g^{-1})			P (mg g^{-1})			N:P ratio		
	n	Mean	CV	n	Mean	CV	n	Mean	CV
Deciduous broadleaf	358	25.7[a]	0.25	357	1.85[a]	0.47	352	16.4[a]	0.39
Evergreen broadleaf	154	18.5[b]	0.36	153	1.01[b]	0.46	152	21.1[b]	0.37
Conifer	19	15.6[c]	0.27	21	1.40[b]	0.44	19	13.8[a]	0.54
All	531	23.2	0.31	531	1.59	0.53	523	17.6	0.41

Comparisons among plant growth forms are performed using t-tests with Bonferroni corrections. Different superscript letters (a, b and c) indicate significant differences (adjusted p < 0.05) in the mean values. n, sample size; CV, coefficient of variation.

in leaf P was also greater than in leaf N and N:P within deciduous and evergreen broadleaved plants.

Changes in leaf N, P and N:P along environmental gradients

For all species pooled together, leaf N, P and N:P were significantly related to latitude (p < 0.001, r^2 = 0.11, 0.32 and 0.21, respectively). Both leaf N and P increased but N:P decreased with increasing latitude (Fig. 2). Leaf P had the steepest slope against latitude with a 3.0-fold change across the 32° latitude-span, followed by N:P with a 2.0-fold change and N with a 1.5-fold change.

Leaf chemical traits were also significantly related to climate (MAT and AP) and soil N, P concentrations (with all p < 0.001, Fig. 3). In general, leaf N and P responded to environmental variables in the same direction: negatively to MAT and AP, and positively to soil N and P concentrations (Fig. 3a–h). The trends of leaf N:P along environmental gradients were exactly opposite to that of leaf N and P. Leaf N:P was negatively related to soil N and P, while positively related to MAT and AP (Fig. 3i–l). However, the relationships between leaf P and environmental variables were much stronger than those of leaf N and N:P (Fig. 3, r^2 = 0.33, 0.30, 0.15 and 0.30 for leaf P with MAT, AP, soil N and soil P, respectively; r^2 = 0.09, 0.14, 0.06 and 0.05 for leaf N; r^2 = 0.23, 0.16, 0.09 and 0.26 for leaf N:P).

For different plant growth forms, leaf N was not related to environmental variables for any of the three groups (Fig. 3a–d), while leaf P was significantly related to all the environmental variables for deciduous broadleaved plants, significantly related to MAT and AP for coniferous plants, and related to soil P concentration for evergreen broadleaved plants (Fig. 3e–h). Within each plant growth form, N:P again showed opposite relationships with environmental variables to that of leaf P (Fig. 3i–l).

Effects of climate, soil and plant growth form on leaf N, P and N:P

A GLM analysis showed that for leaf N, only plant growth form and site were included in the final model. Plant growth form explained 30.5% of the variation in leaf N, site accounted for an additional 8.0% of the variation (Table 2). For leaf P, the best model included plant growth form, MAT and their interaction as predictors; these variables explained 28.1, 12.2 and 0.7% of the variation in leaf P, respectively; site accounted for an additional 21.3% of the variation (Table 2). The best model of leaf N:P had the same terms as that of leaf P. Nevertheless plant growth form only explained 10.2% of the variation in N:P, and site accounted for 30.9% of the variation (Table 2).

Discussion

Patterns of leaf N, P and N:P along environmental gradients

We found that leaf P was more variable and more tightly associated with climate and soil than was either leaf N or
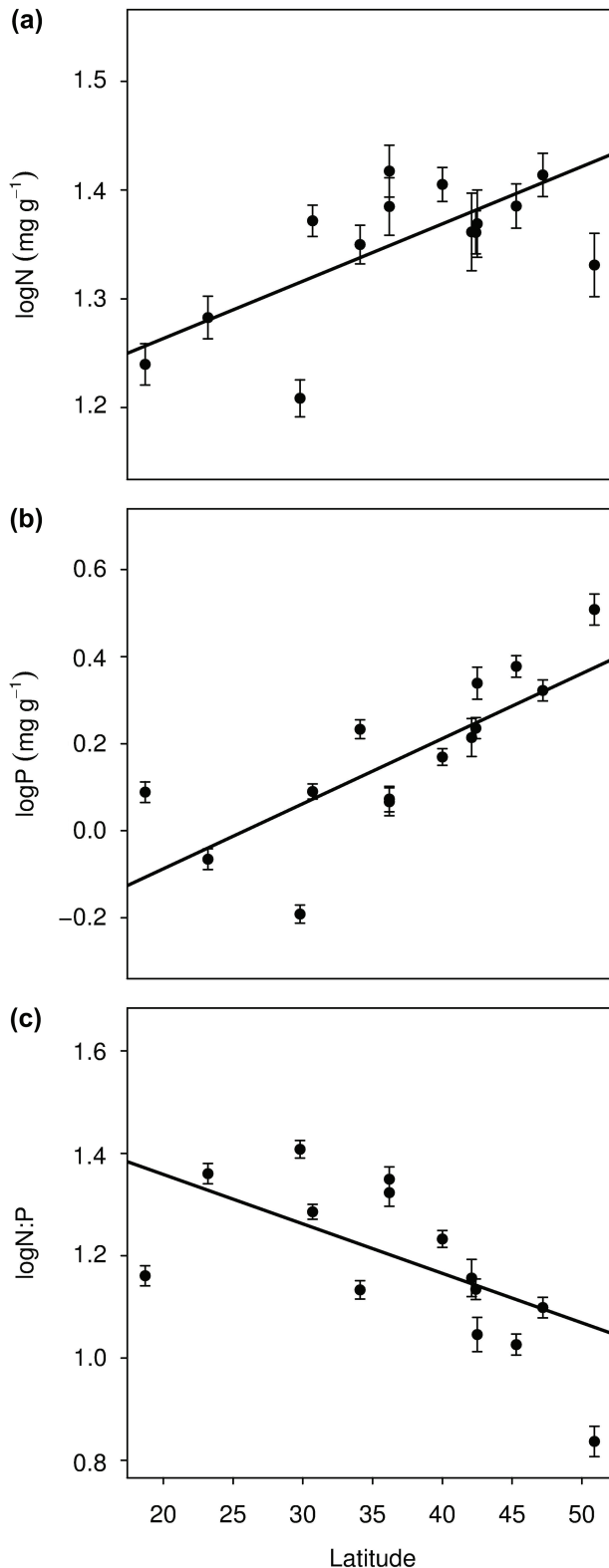
Figure 2. Leaf N, P and N:P in relation to latitude. Points and error bars show means and standard errors for each site, and regression lines are fit to the raw data.

N:P (Table 1, Fig. 3), in agreement with the previous studies conducted for the various plant groups and in different geographical regions (Güsewell and Koerselman 2002, Kerkhoff et al. 2005, Lovelock et al. 2007, Townsend et al.

2007, He et al. 2008, Ordoñez et al. 2009). Leaf N was mainly determined by shifts in plant growth form (deciduous broadleaved/evergreen broadleaved/coniferous plants) along the latitudinal gradient, while leaf P and N:P were determined by both climate and plant growth form, as well as their interaction (Table 2).

Influences of climate and soil on leaf N, P and N:P

The GLM analysis showed that climate and soil had no significant effects on leaf N when controlling plant growth form, while MAT and soil P concentration had significant effects on leaf P (Table 2). The stronger influence of climate and soil on leaf P than on leaf N can be explained by the stronger response in P cycling induced by climate (Swenson et al. unpublished work, cited in Elser et al. 2010). Contrasting to the adequate N supply in atmosphere and multiple mechanisms of N-fixation, soil P is mainly derived from rock weathering and its diffusivity in soil solution is considerably lower than that of N (Fitter et al. 1998, Lambers et al. 1998, Aerts and Chapin 2000). Therefore, soil P supply is strongly influenced by shifts in climate, and consequently affects leaf P content. The final general linear model of leaf N:P had the same explanatory terms as that of leaf P because leaf P is more variable than leaf N and leaf N:P is determined largely by leaf P (Güsewell and Koerselman 2002, Ordoñez et al. 2009).

In the final GLM models, site explained 8.0, 21.3 and 30.9% of variance in leaf N, P and N:P, respectively (Table 2). These variance components represent the amongst-site variability which had not been captured by the climate and soil. There are also considerable unexplained variances for leaf N, P and N:P (61.5, 37.8 and 44.7%, respectively), which may be explained by various sources, such as unquantified micro-environments, species-specific variability, soil chronosequences and disturbance (Westoby and Wright 2006, Garnier et al. 2007, Ordoñez et al. 2009, Durán et al. 2010).

Influences of plant growth form on leaf N, P and N:P

Plant growth form explained 30.5, 28.1 and 10.2% of variance for leaf N, P and N:P, respectively (Table 2). According to the growth rate hypothesis (Sterner and Elser 2002, Elser et al. 2003, Ågren 2004, Vrede et al. 2004), deciduous broadleaved plants with short leaf lifespans have higher growth rate, therefore are rich in N and have higher photosynthetic rates than do evergreen broadleaved plants (Reich et al. 1992, Wright et al. 2004). Leaf P can also be determined by plant growth form because N and P are combined in organic chemicals and are both regulated by plant growth (Lambers et al. 1998, Aerts and Chapin 2000). Moreover, leaf P is present not only in organic forms (i.e. nuclear acid) but also in inorganic forms (i.e. orthophosphate). The inorganic P forms comprise a considerable proportion of leaf P (Chapin and Kedrowski 1983, Sterner and Elser 2002), which mainly exist as storage materials from luxury accumulation, and thus frequently reflect the soil P availability (Mulligan 1989, Sterner and Elser 2002, Oyarzabal and Oesterheld 2009). Therefore, leaf P is affected by both plant
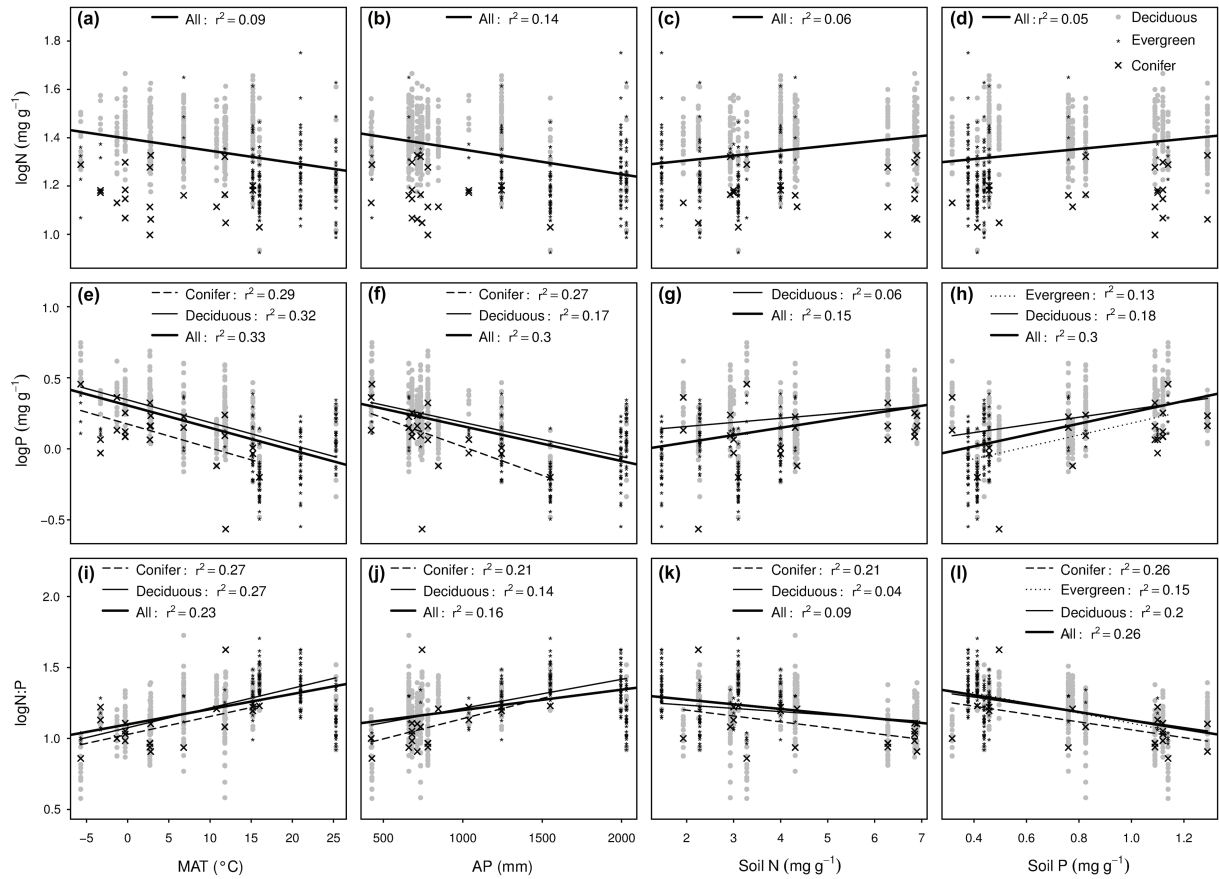
Figure 3. Leaf N, P and N:P in relation to environmental variables. (a–d) leaf N concentration; (e–h) leaf P concentration; (i–l) leaf N:P. Environmental variables include: (a, e, i) mean annual temperature (MAT); (b, f, j) annual precipitation (AP); (c, g, k) soil N concentration; and (d, h, l) soil P concentration. Lines were plotted for relationships with p < 0.05.

growth form and environmental factors. Compared to the effects of plant growth form on leaf N and P, plant growth form only explained 10.2% of variance on leaf N:P, which may be caused by the positive relationship between leaf N and P (Niklas et al. 2005, Reich et al. 2010); both leaf N and P increased with increasing plant growth rate, thus differences of leaf N:P among plant growth forms are weakened by the change in leaf N and P (Ågren 2004).

Besides the main effects of plant growth form on leaf chemistry, it is noteworthy that the interaction between growth form and mean annual temperature significantly influenced leaf P and N:P (Table 2). None of the plant growth forms showed significant trends in leaf N along environmental gradients, while leaf P and N:P of the deciduous broadleaved plants were associated with climate and soil (Fig. 2). Previous experimental and field studies also found

Table 2. Summary of general linear models for the leaf N, P and N:P.

	logN				logP				logN:P			
	Main-effect model			Final model	Main-effect model			Final model	Main-effect model			Final model
Factor	DF	MS	F	SS%	DF	MS	F	SS%	DF	MS	F	SS%
GF	2	1.56	**127.66**	30.5%	2	3.83	**188.20**	28.1%	2	0.89	**56.83**	10.2%
Environmental variables												
MAT	1	0.06	0.91		1	3.32	**6.88**	12.2%	1	2.39	**5.30**	13.7%
AP	1	0.07	1.05		1	1.78	2.92		1	1.14	2.05	
Soil P	1	0.01	0.13		1	3.14	**6.32**		1	2.92	**7.17**	
Soil N	1	0.03	0.52		1	0.77	1.10		1	0.56	0.92	
Random factor												
Site*	12	0.06	**13.96**	8.0%	12	0.48	**24.06**	21.3%	12	0.45	**29.07**	30.9%
Interaction term												
GF:MAT	–	–	–		2	0.09	**4.61**	0.7%	2	0.05	**3.21**	0.6%

F values of environmental variables were corrected by the random effect (site). F values in bold indicate p < 0.05. Abbreviations: MAT, mean annual temperature; AP, annual precipitation; GF, plant growth form. *DF, MS and F values of site differ in the four main-effect models, values given here are calculated from the final model.

that deciduous plants were more sensitive to temperature than evergreen species, and the different responses between deciduous and evergreen plants were explained by their contrasting growth strategies (Way and Oren 2010). Evergreen plants are mainly distributed in sites with low nutrient availability (Givnish 2002), and thus their leaf traits might be adaptive to the limited nutrient supply, such as longer leaf lifespan, lower nutrient content and more allocation to structural components (Takashima et al. 2004, Wright et al. 2004, 2005). These economic strategies may have constrained evergreen species in their ability to respond to changing environmental conditions (Chapin et al. 1995, Valladares et al. 2000).

Linking leaf chemical traits with ecosystem functioning and nutrient cycling

Our results are helpful for understanding the link between leaf chemical traits and ecosystem functioning. We found that leaf N is mainly determined by plant growth requirements, while leaf P is regulated by the environment in addition to plant growth. These findings suggest that leaf N is more stable and has stronger stoichiometric homeostasis than leaf P. Similar evidence can be found in a recent study (Yu et al. 2010): at the species level, the homeostatic regulation coefficient of N (H_N) is higher than that of P (H_P), indicating that leaf N is more invariable relative to the external world (Sterner and Elser 2002). Moreover, H_N was much more tightly associated with key aspects of ecological performance than H_P (Yu et al. 2010). These evidences support our results that leaf N is more constrained by plant growth requirements than leaf P, and indicate leaf N is more tightly linked with ecosystem functioning.

Given that leaf chemistry is a critical component in nutrient cycling, the responses of leaf chemical traits to climate, soil and plant growth form will help us to understand the influence of future climate change on nutrient cycling (Chapin 2003, Díaz et al. 2004). Our analyses highlight that leaf P is more flexible than leaf N in responses to changing environment. Indeed, such a characteristic is also found in other reservoirs of P and N elements. For example, litter P responds more obviously to nutrient addition than litter N (Vitousek 2004); concentration of soil P is much more variable than that of N in China's biota (CV = 0.95 vs 1.99 for N vs P) (National Soil Survey Office of China 1997, 1998). These further support our findings that N cycling is mainly controlled by biotic factors, while P cycling is jointly controlled by biotic and abiotic factors. In this way, our findings help to improve our understanding of N and P cycling, and modeling ecosystem nutrient cycling under the global change.

References

Aerts, R. and Chapin, F. S. 2000. The mineral nutrition of wild plants revisited: a re-evaluation of processes and patterns. – Adv. Ecol. Res. 30: 1–67.

Ågren, G. I. 2004. The C:N:P stoichiometry of autotrophs – theory and observations. – Ecol. Lett. 7: 185–191.

Ågren, G. I. 2008. Stoichiometry and nutrition of plant growth in natural communities. – Annu. Rev. Ecol. Evol. Syst. 39: 153–170.

An, Y. A. et al. 2005. Plant nitrogen concentration, use efficiency, and contents in a tallgrass prairie ecosystem under experimental warming. – Global Change Biol. 11: 1733–1744.

Attiwill, P. M. and Adams, M. A. 1993. Nutrient cycling in forests. – New Phytol. 124: 561–582.

Chapin, F. S. 2003. Effects of plant traits on ecosystem and regional processes: a conceptual framework for predicting the consequences of global change. – Ann. Bot. 91: 455–463.

Chapin, F. S. and Kedrowski, R. A. 1983. Seasonal-changes in nitrogen and phosphorus fractions and autumn retranslocation in evergreen and deciduous taiga trees. – Ecology 64: 376–391.

Chapin, F. S. et al. 1995. Responses of arctic tundra to experimental and observed changes in climate. – Ecology 76: 694–711.

Chapin, F. S. et al. 2002. Principles of terrestrial ecosystem ecology. – Springer.

Cornelissen, J. H. C. et al. 2003. A handbook of protocols for standardised and easy measurement of plant functional traits worldwide. – Aust. J. Bot. 51: 335–380.

Díaz, S. et al. 2004. The plant traits that drive ecosystems: evidence from three continents. – J. Veg. Sci. 15: 295–304.

Durán, J. et al. 2010. Changes in leaf nutrient traits in a wildfire chronosequence. – Plant Soil 331: 69–77.

Elser, J. J. et al. 2003. Growth rate-stoichiometry couplings in diverse biota. – Ecol. Lett. 6: 936–943.

Elser, J. J. et al. 2010. Biological stoichiometry of plant production: metabolism, scaling and ecological response to global change. – New Phytol. 186: 593–608.

Field, C. and Mooney, H. A. 1986. The photosynthesis–nitrogen relationship in wild plants. – In: Givnish, T. J. (ed.), On the economy of plant form and function. Cambridge Univ. Press, pp. 25–55.

Fitter, A. H. et al. 1998. The phosphorus nutrition of wild plants and paradox of arsenate tolerance: does leaf phosphorus concentration control flowering? – Penn State Univ.

Garnier, E. et al. 2007. Assessing the effects of land-use change on plant traits, communities and ecosystem functioning in grasslands: a standardized methodology and lessons from an application to 11 European sites. – Ann. Bot. 99: 967–985.

Givnish, T. J. 2002. Adaptive significance of evergreen vs. deciduous leaves: solving the triple paradox. – Silva Fenn. 36: 703–743.

Güsewell, S. 2004. N:P ratios in terrestrial plants: variation and functional significance. – New Phytol. 164: 243–266.

Güsewell, S. and Koerselman, M. 2002. Variation in nitrogen and phosphorus concentrations of wetland plants. – Perspect. Plant Ecol. Evol. Syst. 5: 37–61.

Han, W. X. et al. 2005. Leaf nitrogen and phosphorus stoichiometry across 753 terrestrial plant species in China. – New Phytol. 168: 377–385.

He, J. S. et al. 2006. Stoichiometry and large-scale patterns of leaf carbon and nitrogen in the grassland biomes of China. – Oecologia 149: 115–122.

He, J. S. et al. 2008. Leaf nitrogen: phosphorus stoichiometry across Chinese grassland biomes. – Oecologia 155: 301–310.

Hedin, L. O. 2004. Global organization of terrestrial plant–nutrient interactions. – Proc. Natl Acad. Sci. USA 101: 10849–10850.

John, M. K. 1970. Colorimetric determination of phosphorus in soil and plant materials with ascorbic acid. – Soil Sci. 109: 214–220.

Kerkhoff, A. J. et al. 2005. Plant allometry, stoichiometry and the temperature-dependence of primary productivity. – Global Ecol. Biogeogr. 14: 585–598.

Koerselman, W. and Meuleman, A. F. M. 1996. The vegetation N:P ratio: a new tool to detect the nature of nutrient limitation. – J. Appl. Ecol. 33: 1441–1450.

Lambers, H. et al. 1998. Plant physiological ecology. – Springer.

LeBauer, D. S. and Treseder, K. K. 2008. Nitrogen limitation of net primary productivity in terrestrial ecosystems is globally distributed. – Ecology 89: 371–379.

Lovelock, C. E. et al. 2007. Testing the growth rate vs. geochemical hypothesis for latitudinal variation in plant nutrients. – Ecol. Lett. 10: 1154–1163.

Lukac, M. et al. 2010. Global climate change and tree nutrition: effects of elevated CO_2 and temperature. – Tree Physiol. 30: 1209–1220.

McGroddy, M. E. et al. 2004. Scaling of C:N:P stoichiometry in forests worldwide: implications of terrestrial redfield-type ratios. – Ecology 85: 2390–2401.

Mulligan, D. R. 1989. Leaf phosphorus and nitrogen concentrations and net photosynthesis in eucalyptus seedlings. – Tree Physiol. 5: 149–157.

National Soil Survey Office of China 1997. Survey of soils in China. – Chinese Agriculture Press.

National Soil Survey Office of China 1998. Soils of China. – Chinese Agriculture Press.

Niklas, K. J. et al. 2005. Nitrogen/phosphorus leaf stoichiometry and the scaling of plant growth. – Ecol. Lett. 8: 636–642.

Ordoñez, J. C. et al. 2009. A global study of relationships between leaf traits, climate and soil measures of nutrient fertility. – Global Ecol. Biogeogr. 18: 137–149.

Oyarzabal, M. and Oesterheld, M. 2009. Phosphorus reserves increase grass regrowth after defoliation. – Oecologia 159: 717–724.

Reich, P. B. and Oleksyn, J. 2004. Global patterns of plant leaf N and P in relation to temperature and latitude. – Proc. Natl Acad. Sci. USA 101: 11001–11006.

Reich, P. B. et al. 1992. Leaf life-span in relation to leaf, plant, and stand characteristics among diverse ecosystems. – Ecol. Monogr. 62: 365–392.

Reich, P. B. et al. 2009. Leaf phosphorus influences the photosynthesis–nitrogen relation: a cross-biome analysis of 314 species. – Oecologia 160: 207–212.

Reich, P. B. et al. 2010. Evidence of a general 2/3-power law of scaling leaf nitrogen to phosphorus among major plant groups and biomes. – Proc. R. Soc. B 277: 877–883.

Rohrs-Richey, J. K. and Mulder, C. P. H. 2007. Effects of local changes in active layer and soil climate on seasonal foliar nitrogen concentrations of three boreal forest shrubs. – Can. J. For. Res. 37: 383–394.

Sterner, R. W. and Elser, J. J. 2002. Ecological stoichiometry: the biology of elements from molecules to the biosphere. – Princeton Univ. Press.

Takashima, T. et al. 2004. Photosynthesis or persistence: nitrogen allocation in leaves of evergreen and deciduous *Quercus* species. – Plant Cell Environ. 27: 1047–1054.

Townsend, A. R. et al. 2007. Controls over foliar N:P ratios in tropical rain forests. – Ecology 88: 107–118.

Valladares, F. et al. 2000. Low leaf-level response to light and nutrients in Mediterranean evergreen oaks: a conservative resource-use strategy? – New Phytol. 148: 79–91.

Vitousek, P. M. 2004. Nutrient cycling and limitiation: Hawai'i as a model system. – Princeton Univ. Press.

Vrede, T. et al. 2004. Fundamental connections among organism C:N:P stoichiometry, macromolecular composition, and growth. – Ecology 85: 1217–1229.

Way, D. A. and Oren, R. 2010. Differential responses to changes in growth temperature between trees from different functional groups and biomes: a review and synthesis of data. – Tree Physiol. 30: 669–688.

Westoby, M. and Wright, I. J. 2006. Land-plant ecology on the basis of functional traits. – Trends Ecol. Evol. 21: 261–268.

Woods, H. A. et al. 2003. Temperature and the chemical composition of poikilothermic organisms. – Funct. Ecol. 17: 237–245.

Wright, I. J. et al. 2004. The worldwide leaf economics spectrum. – Nature 428: 821–827.

Wright, I. J. et al. 2005. Modulation of leaf economic traits and trait relationships by climate. – Global Ecol. Biogeogr. 14: 411–421.

Yu, Q. A. et al. 2010. Linking stoichiometric homeostasis with ecosystem structure, functioning and stability. – Ecol. Lett. 13: 1390–1399.

Zotz, G. 2004. The resorption of phosphorus is greater than that of nitrogen in senescing leaves of vascular epiphytes from lowland Panama. – J. Trop. Ecol. 20: 693–696.

Supplementary material (Appendix E6833 at <www.oikosoffice.lu.se/appendix>). Appendix 1.