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LEAF STOICHIOMETRIC TRAIT AND SPECIFIC LEAF AREA OF DOMINANT SPECIES IN THE SECONDARY SUCCESSION OF THE LOESS PLATEAU

ABSTRACT: Ecological stoichiometry is the study of the balance of multiple chemical elements in ecological interactions. Terrestrial plants exhibit inter- and intra-specific differences in leaf C:N:P stoichiometry that has been used to study plant competition and succession, nutrient use efficiency, N and P nutrient limitation. However, little is known about leaf C:N ratio, N:P ratio in the Loess Plateau, particularly the pattern of leaf C:N:P stoichiometry during the secondary succession. Thus, leaf stoichiometric traits and specific leaf area (SLA) of dominant species at different secondary successional stages in the Loess Plateau were measured. The study was conducted at the Lianjiabian forest region of the Loess Plateau (35.03°–36.37°N and 108.10°–109.08°E), Gansu, China. Leaf C:N:P stoichiometry and specific leaf area of 18 dominant species (herb) community stage (*Stipa bungeana* Trin, *Bothriochloa ischaemum* (Linn.) Keng, *Carex lanceolata* Boott, *Artemisia sacrorum* Ledeb, *Pulsatilla chinensis* (Bunge) Regel, *Potentilla chinensis* Ser), shrub community stage (*Sophora viciifolia* Hance, *Hippophae rhamnoides* (Linn.), *Ostryopsis davidiana* Decne, *Rubus palmatus* Thunb, *Rosa xanthina* Lindl, *Acer ginnala* Maxim, *Spiraea pubescens* Turcz, *Ziziphus jujuba* var. *spinosa*), early forest community stage (*Populus davidiana* Dode, *Betula platyphylla* Suk, *Platycladus orientalis* (Linn.) Franco), climax forest community stage (*Quercus liaotungensis* Koidz)) at four secondary successional stages were determined in the Loess Plateau in early June of 2006.

The herb and shrub community stage had higher SLA than that in the early forest community stage and climax forest community stage. The Leaf C, N, C:N ratio and N:P ratio at different secondary successional stages were significantly different ($P < 0.05$), but the leaf P didn't differ. The N:P ratio of herb community stage were significant difference from those of the others stages (shrub community stage, early forest community stage and climax forest community stage), but there were not significant difference among the shrub community stage, early forest community stage and climax forest community stage. N:P ratio shifts of the species were consistent along the successional sere, although the N:P ratio of the different species at a successional stage varied considerably. On community level, the lowest N:P ratio (9.8) was found in herb community stage in the secondary succession, the N:P ratio increased to 12.9 in shrub community stage, declined to 12.4 in early forest community stage, and increased in climax forest community stage (e.g. 12.6 in the *Quercus liaotungensis* Koidz community). The results suggest that the vegetation productivity of the Loess Plateau was N-limited at each secondary successional stage.

KEY WORDS: leaf N:P ratio, secondary succession, specific leaf area, Loess Plateau

1. INTRODUCTION

The stoichiometric traits of leaves are closely associated with the structure and function of terrestrial ecosystems (Güsewell 2004). The most investigated relation is that of C:N:P because N and P commonly limit growth (Elser *et al.* 2007), and though C provides the structural basis, constituting a fairly stable 50% of a plant's dry mass, it also can act as a limiting element (Agren 2008). The variability of stoichiometry is determined in several factors, including variability between tissues, temporal variability and climatic variability (Agren 2008, Peñuelas *et al.* 2008). Specific leaf area (SLA) as a fundamental plant trait has been proven to be strongly linked to relative growth rate and resource use (Wilson *et al.* 1999). Species differences in specific leaf area and leaf nitrogen content have been associated with biogeographic, ecological and edaphic patterns (Reich *et al.* 1991). Nitrogen uptake and carbon assimilation by plant and leaf composition drive biogeochemical cycles (Chadwick *et al.* 1999, Sterner and Elser 2002, Wright *et al.* 2004b).

Nitrogen and phosphorus play a vital role in plant functioning, and are generally considered the most limiting nutrients in terrestrial ecosystems (Chapin 1980, Reich *et al.* 1997). Nitrogen and phosphorus contents in plant biomass are determined in terms of the balance of nitrogen and phosphorus through turnover, leaching, exudation, herbivory and parasites (Aerts and Chapin 2000, Eckstein and Karlsson 2001). Several studies have revealed that plant nitrogen and phosphorus contents are associated with many biotic and abiotic factors, including habitat (Koerselman and Meuleman 1996, Thompson *et al.* 1997, Cunningham *et al.* 1999), growth stage (Thompson *et al.* 1997, Elser *et al.* 2000b) and plant functional group (Reich *et al.* 1998). Previous studies have also shown that N:P ratio of plant biomass can be an indicator of vegetation composition, functioning and nutrient limitation on community level (Koerselman and Meuleman 1996, Güsewell and Koerselman 2002, Güsewell 2004, Reich and Oleksyn 2004). Using N:P ratio of plant biomass as the indicator of N or P limitation, various studies have suggested that shifts be-

tween N and P limitation lead to changes in plant trait, vegetation composition and species diversity (Koerselman and Meuleman 1996, Verhoeven *et al.* 1996, Roem and Berendse 2000). Based on studies of European wetland plants, the threshold leaf N:P ratio is found <14 for N limitation, and >16 for P limitation (Güsewell and Koerselman 2002). Plant N:P ratio have proved useful to investigate the shifts between N to P limitation because leaf N and P are easily determined and compared across studies, and the relationships between N:P ratio and plant or vegetation properties have also been used to describe functional difference between naturally N- or P- limited plant communities and their responses to environmental change or human management.

Several recent studies have summarized the results of global-scale variation in leaf functional traits and multiple small-scale investigations to analyze regional and global patterns of leaf N, P, N:P ratio and SLA (Reich *et al.* 1997, Elser *et al.* 2000a, McGroddy *et al.* 2004, Reich and Oleksyn 2004, Wright *et al.* 2004b, Han *et al.* 2005, Luo *et al.* 2005). These studies have identified biogeographic patterns in terrestrial leaf stoichiometry and leaf traits across local, regional and global gradients (McGroddy *et al.* 2004, Reich and Oleksyn 2004), and examined global convergence in the relationships between leaf stoichiometry and other morphological characteristics (Reich *et al.* 1997, Wright *et al.* 2004b). The concentrations of leaf N ([N]) and P ([P]) often increase with increasing specific leaf area (SLA) (Wright *et al.* 2001, Niinemets *et al.* 2002). Wright *et al.* (2004a) studied leaf trait relationships of 258 Australian plant species, finding that SLA was positively correlated with leaf N and P concentrations. Such relationships may partly be explained by a larger fraction of support structures in thicker and denser leaves that possess a lower SLA, and accordingly by a reduced requirement for mineral nutrients for construction of foliage with lower SLA (Niinemets and Kull 1998, Niinemets 1999). These previous studies have greatly advanced the understanding of the variation and patterns of leaf C, N, P and SLA in terrestrial plants.

Specific leaf area variation is a strategy for plant to optimize carbon gain and adapt to specific temperature, moisture and nutrient regimes, and the species with lower SLA tend to have long average leaf life spans (LL) and low leaf nitrogen contents (Reich *et al.* 1997, Wright *et al.* 2004b). The relations among leaf traits generally exist within wide ranges of plant populations, communities and biomes, thus apparently reflecting convergent adaptation of plants to the given climate and/or other environmental constraints (Reich *et al.* 1999, Wright *et al.* 2002). The species with higher SLA maximize leaf nitrogen content and light-interception area, thus presenting higher photosynthetic rate (A_{mass}) (Reich *et al.* 1991, Reich *et al.* 1992). Conversely, the leaves of lower SLA species have thick epidermal wall and cuticle (Reich *et al.* 1998), abundant sclerification, high cell wall/cytoplasm ratio in tissues, and high crude fiber to protein ratio, thus forming thick and small leaves.

It is well known that vegetation succession can recover degraded soil properties. For this reason, understanding the secondary succession processes in the Loess Plateau is becoming increasingly important because of the vegetation cover tends to turn low and sparse in this area, and thus soil degradation ensues from extremely serious soil erosion. Therefore, more attention has been paid to the secondary succession in the Loess Plateau by Chinese scientists. A lot of studies focused on plant community sere and soil changes during the secondary succession in the Loess Plateau region (Zou *et al.* 2002, Li and Shao 2003, Jia *et al.* 2005, Fan *et al.* 2006), but few investigations on dynamics of leaf stoichiometric traits and specific leaf area at different stages of the secondary succession. In this study, an attempt was made to investigate the dynamics of the leaf stoichiometric traits and specific leaf area of dominant species along the secondary succession gradient of the Loess Plateau. The main objectives of the study were: (1) to determine the leaf C, N, P, C:N ratio, N:P ratio and specific leaf area of dominant species; (2) to analyze the difference of leaf C, N, P, C:N ratio, N:P ratio and specific leaf area of dominant species among different secondary successional stages.

2. STUDY AREA

The study was conducted at the Lianjiabi-an forest region of the Loess Plateau, Heshui County, Gansu, China (35.03°-36.37°N and 108.10°-109.08°E), at altitude is about 1500 m above sea level. The region belongs to the hilly and gully zone of the Loess Plateau. In the region, the average annual temperature is 7.4°C and the average rainfall is close to 587.6 mm of which 60–70% principally distributes in June, July, August and September (Zou *et al.* 2002, Wang *et al.* 2005); the aridity index is 0.72, and the atmospheric relative humidity is 63–68%. The soil of the study site was classified as calcareous cinnamon soil, which originated from 50–100 m deep primary or secondary loess and the thickness of laterite is 80–100 m below (Zou *et al.* 2002, Wang *et al.* 2005). Because of the influences of geographical and climatic factors, the vegetation distribution on sunny and shady hillsides was significantly different. Generally, there are the shrubs and herbs with few or no woods in sunny ridges and mounds, and sub-natural forests in shady hillside and valleys. The vertical forest vegetation distribution was indistinctive because of the little relative height variation. It is evident that in this area the vegetation belonged to northern warm temperate zone and deciduous *Quercus liaotungensis* Koidz sub-zone according to the characteristic of its vegetation type, distribution and succession (Zou *et al.* 2002).

The study area was situated in the transitional zone between forest-steppe and semi-arid steppe zones and most of its forests were sub-natural woods gradually recovered from frequent damages by human activities. Most vegetation are sub-natural woods that have gradually recovered on abandoned farmlands after 1866, and then large scale secondary forest landscape have formed and developed towards the climax forest community (*Quercus liaotungensis* Koidz) during the recovery. In the regions, there have relatively integrated vegetation sere: herb community (*Bothriochloa ischaemum* (Linn.) Keng, *Artemisia giraldii* Pamp, *Artemisia sacrorum* Ledeb, *Spodiopogon sibiricus* Trin) → shrub community (*Sophora viciifolia* Hance, *Hippophae rhamnoides* (Linn.), *Ostryopsis davidiana* Decne) → early forest

community (*Populus davidiana* Dode, *Betula platyphylla* Suk, *Platyclusus orientalis* (Linn.) Franco) → climax forest community (*Quercus liaotungensis* Koidz).

3. MATERIAL AND METHODS

Sampling and sample determination were conducted in early June of 2006. The different secondary successional stages were chosen to represent temporal series and the research sites were chosen as those places that suffered minimal grazing and other anthropogenic disturbances by visual vegetation examination. 18 species which represented four successional stages in the Loess Plateau (I, II, III and IV stands for herb community stage, shrub community stage, early forest community stage and climax forest community stage, respectively) were chosen:

I. Herb community stage (*Stipa bungeana* Trin, *Bothriochloa ischaemum* (Linn.) Keng, *Carex lanceolata* Boott, *Artemisia sacrorum* Ledeb, *Pulsatilla chinensis* (Bunge) Regel, *Potentilla chinensis* Ser),

II. Shrub community stage (*Sophora viciifolia* Hance, *Hippophae rhamnoides* (Linn.), *Ostryopsis davidiana* Decne, *Rubus palmatus* Thunb, *Rosa xanthina* Lindl, *Acer ginnala* Maxim, *Spiraea pubescens* Turcz, *Ziziphus jujuba* var. *spinosa*),

III. Early forest community stage (*Populus davidiana* Dode, *Betula platyphylla* Suk, *Platyclusus orientalis* (Linn.) Franco),

IV. Climax forest community stage (*Quercus liaotungensis* Koidz).

Nearly all the measurements or determinations were carried out at the flowering stage. The chosen trees were mainly distributed in the half-shady hillside or half sunny hillside, and the shrubs and herbs were distributed in the sunny hillside. The fully expanded leaves were collected from the middle part of a plant in the case of herbs and from the middle canopy in the case of shrubs and trees. Each sample consisted of 30–50 leaves from ten or twelve individual plants of each species, and the leaf numbers for each individual plant were approximately equal.

3.1. Specific leaf area measurement

About 30–50 fully expanded leaves were measured. The sample leaves were scanned

with digital scanner and the leaf surface areas were measured with digital image analysis software (Motic images advanced 3.0, China). All the measured leaves were carefully put into paper bags and the paper bags were labeled; then all the bags were oven dried at 70°C for 48 h to their constant weight weights and then the leaves were weighed with precision balance. The SLA was calculated as the ratio of leaf area to dry mass ($\text{cm}^2\cdot\text{g}^{-1}$).

3.2. Plant nutrient determination

All the leaves from the same species were mixed to form a single sample and the dried samples were ground to form a homogenous powder using plant-sample mill (1093 Sample Mill, Foss, Sweden) and then sieved with a 1-mm mesh screen before the sub-samples were prepared for element determination. The samples were then digested with modified Kjeldahl method (3h at 365°C in a mixture of concentrated sulphuric acid and peroxide), and total N were determined with an autoanalyser (Kjeltec 2300 Analyzer Unit, Foss, Sweden), and total P were determined by standard ammonium molybdate method with an autoanalyser (U-2800 spectrophotometer, China, Shanghai). The samples for C measurement were digested in $\text{K}_2\text{Cr}_2\text{O}_7$ - H_2SO_4 solution with the oil bath heating and their C concentration were determined by titration. Leaf C, N and P data were expressed on mass basis (C, $\text{mg}\cdot\text{g}^{-1}$), and C:N (quotient of [C] and [N]) and N:P (quotient of [N] and [P]) ratio were expressed on mass basis.

3.3. Data analysis

We compared the statistical differences in C, N, P, C:N ratio, N:P ratio and specific leaf area between different secondary successional stages by following three steps. First, the mean leaf C, N and P of each species were calculated, and its C:N ratio, N:P ratio were obtained. Second, the C, N, P, C:N ratio, N:P ratio and specific leaf area of each secondary successional stage were obtained as the means of all species at the successional stage. Finally, the C, N, P, C:N ratio, N:P ratio and specific leaf area of each successional stage were compared to the other secondary successional stages using one-way ANOVA, and LSD test was adopted to do

multiple comparison. All the statistical analyses were conducted using SPSS software (SPSS Inc., Chicago, USA).

4. RESULTS

The N:P ratio of herb community stage were significantly different from those of the others stages (shrub community stage, early forest community stage and climax forest community stage), but there were not significant differences among the shrub community stage, early forest community stage and climax forest community stage. N:P ratio shifts of the species were consistent along the successional sere, although the N:P ratio of the different species at a successional stage varied considerably. On community level, the lowest N:P ratio (9.8) was found in the herb community stage, which was usually considered

a primary stage of the secondary succession. Thereafter, the N:P ratio increased to 12.9 in the shrub community stage, declined to 12.4 in the early forest community stage, including coniferous-broadleaved mixed forest, and increased in the climax forest community stage (e.g. 12.6 in the *Quercus liaotungensis* Koidz community) (Table 1). The results suggest that the vegetation productivity in the Loess Plateau was N-limited at each successional stage.

The N:P ratio of the species at each successional stage varied considerably (Table 1), which means that different nutrient limitation for different species at the same successional stage. The N:P ratio primarily ranged within 7.6 (*Artemisia sacrorum* Ledeb) – 17.1 (*Carex lanceolata* Boott) in the herb community stage. Only the N:P ratio of *Carex lanceolata* Boott were higher than 16, and those of

Table 1. Leaf stoichiometric traits and specific leaf area of dominant species at different secondary successional stages in the Loess Plateau (means \pm SD).

Species	C (mg·g ⁻¹)	N (mg·g ⁻¹)	P (mg·g ⁻¹)	C:N ratio	N:P ratio	Specific leaf area (cm ² ·g ⁻¹)
Herb community stage						
<i>Stipa bungeana</i> Trin	360.6 \pm 60.0	8.9 \pm 0.01	1.1 \pm 0.11	40.6 \pm 6.3	8.2 \pm 0.9	172.6 \pm 8.5
<i>Bothriochloa ischaemum</i> (Linn.) Keng	397.2 \pm 30.3	13.4 \pm 0.15	1.7 \pm 0.12	29.6 \pm 2.2	7.7 \pm 0.5	234.4 \pm 4.5
<i>Carex lanceolata</i> Boott	377.8 \pm 8.5	11.8 \pm 0.11	0.7 \pm 0.08	32.1 \pm 0.9	17.1 \pm 2.0	209.5 \pm 1.7
<i>Artemisia sacrorum</i> Ledeb	428.0 \pm 16.8	18.3 \pm 0.08	2.4 \pm 0.02	23.4 \pm 1.0	7.6 \pm 0.1	210.8 \pm 0.2
<i>Pulsatilla chinensis</i> (Bunge) Regel	380.8 \pm 2.1	12.3 \pm 0.06	1.4 \pm 0.36	30.9 \pm 0.2	9.1 \pm 2.0	107.7 \pm 0.9
<i>Potentilla chinensis</i> Ser	406.1 \pm 18.9	18.5 \pm 0.08	2.0 \pm 0.32	21.9 \pm 0.9	9.4 \pm 1.4	150.8 \pm 4.5
Average	391.8 \pm 23.8	13.9 \pm 3.8	1.6 \pm 0.6	29.8 \pm 6.7	9.8 \pm 3.6	181.0 \pm 46.7
Shrub community stage						
<i>Hippophae rhamnoides</i> (Linn.)	404.9 \pm 25.6	26.9 \pm 0.36	1.9 \pm 0.11	15.0 \pm 0.8	14.1 \pm 0.6	142.2 \pm 2.0
<i>Ostryopsis davidiana</i> Decne	399.5 \pm 29.1	14.4 \pm 0.05	1.2 \pm 0.04	27.7 \pm 2.1	11.8 \pm 0.3	153.4 \pm 2.5
<i>Rubus palmatus</i> Thunb	338.9 \pm 78.1	15.7 \pm 0.07	1.7 \pm 0.13	21.6 \pm 5.1	9.0 \pm 0.7	200.3 \pm 1.1
<i>Ziziphus jujuba</i> var. <i>spinosa</i>	404.0 \pm 15.5	24.5 \pm 0.13	1.4 \pm 0.11	16.5 \pm 0.7	17.2 \pm 1.2	147.1 \pm 3.4
<i>Spiraea pubescens</i> Turcz	437.4 \pm 52.3	13.1 \pm 0.10	1.0 \pm 0.04	33.3 \pm 4.0	12.9 \pm 0.5	165.4 \pm 4.5
<i>Sophora viciifolia</i> Hance	423.5 \pm 18.4	30.3 \pm 0.32	1.8 \pm 0.14	14.0 \pm 0.5	17.1 \pm 1.6	135.8 \pm 1.8
<i>Acer ginnala</i> Maxim	430.0 \pm 19.4	10.0 \pm 0.27	1.1 \pm 0.21	42.8 \pm 1.0	9.3 \pm 1.8	153.1 \pm 2.3
<i>Rosa xanthina</i> Lindl.	470.1 \pm 81.4	14.1 \pm 0.01	1.2 \pm 0.06	33.4 \pm 5.8	11.9 \pm 0.6	149.7 \pm 2.3
Average	413.6 \pm 37.9	18.6 \pm 7.5	1.4 \pm 0.3	25.5 \pm 10.5	12.9 \pm 3.1	155.9 \pm 19.9
Early forest community stage						
<i>Populus davidiana</i> Dode	457.6 \pm 10.7	17.2 \pm 0.23	1.1 \pm 0.09	26.6 \pm 0.9	15.8 \pm 1.6	152.5 \pm 0.4
<i>Betula platyphylla</i> Suk	507.2 \pm 55.1	17.4 \pm 0.12	1.6 \pm 0.25	29.2 \pm 3.1	11.2 \pm 1.7	176.1 \pm 1.2
<i>Platycladus orientalis</i> (Linn.) Franco	517.2 \pm 7.1	7.5 \pm 0.21	0.7 \pm 0.04	69.1 \pm 2.9	10.1 \pm 0.4	46.4 \pm 0.3
Average	494.0 \pm 31.9	14.0 \pm 5.7	1.1 \pm 0.4	41.7 \pm 23.8	12.4 \pm 3.0	125.0 \pm 69.1
Climax forest community stage						
<i>Quercus liaotungensis</i> Koidz	383.6 \pm 49.5	16.9 \pm 0.09	1.6 \pm 0.20	22.7 \pm 2.9	12.6 \pm 1.8	145.9 \pm 0.6
Significance test	$P < 0.05$	$P < 0.05$	NS	$P < 0.05$	$P < 0.05$	$P < 0.01$

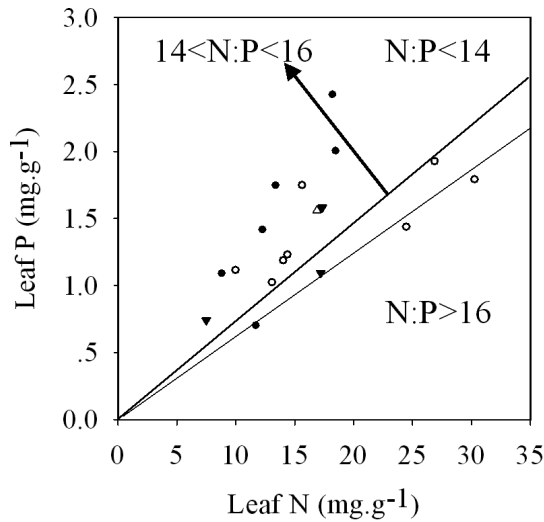


Fig. 1. Relationships between the N and P concentrations and N:P ratio of all species. Symbols show means per species. Close circle symbols stands for the herb community stage and open circle symbols stands for the shrub community stage; close triangle symbols stands for the early forest community stage and open triangle symbols stands for the climax forest community stage

the other species were lower than 14, which indicated that the vegetation productivity in the herb community stage was limited by N, except that *Carex lanceolata* Boott which was limited by P. The N:P ratio of *Sophora viciifolia* Hance and *Ziziphus jujuba* var. *spinosa* were higher than 16, and the N:P ratio of *Hippophae rhamnoides* (Linn.) was between 14 to 16, and the N:P ratio of the other species were lower than 14. Nutrient limitation in shrub community stage were similar to those in the herb community stage. At the early forest community stage and climax forest community stage, the N:P ratio of *Populus davidiana* Dode was between 14 to 16, and the N:P ratios of the other species were lower than 14. Therefore, some P-limited (or N-limited) species emerged under the N-limited (or P-limited) plant community at each successional stage.

The leaf C, N, C:N ratio and N:P ratio were significantly different among the different secondary successional stages ($P < 0.05$, Fig. 2A, Fig. 2D–F), but the leaf P didn't differ ($P > 0.05$, Fig. 2C). The leaf N and N:P ratio were higher in the shrub community stage than in the other three successional stages

(Fig. 2A, D), and this was probably because there were two symbiotic N-fixers (*Sophora viciifolia* Hance and *Hippophae rhamnoides* (Linn.)) in the shrub community stage. The Leaf C and C:N ratios were higher in the early forest community stage than in the other three successional stages (Fig. 2E, F). Conifer species (*Platycladus orientalis* (Linn.) Franco) exhibited the highest C:N ratio in the early forest community stage (Table 1), and the C:N ratio of *Platycladus orientalis* (Linn.) Franco was 2.6 and 2.4 times higher than those of *Populus davidiana* Dode and *Betula platyphylla* Suk.

The specific leaf area at different successional stages differed ($P < 0.01$, Fig. 2B). The variation of specific leaf area showed a high→low→high pattern along secondary succession gradient, and the lowest specific leaf area was found in the early forest community stage, and increased in the climax forest community stage (Table 1). The herb and shrub community stage had higher specific leaf area than the early forest community stage and climax forest community stage. Conifer species (*Platycladus orientalis* (Linn.) Franco) had the lower specific leaf area than other dominant species.

5. DISCUSSION

Leaf N:P ratio in plant biomass is used as the indicator of relative N or P limitation (the N:P ratio < 14 often indicates N limitation and the N:P ratio > 16 often indicates P limitation) (Koerselman and Meuleman 1996, Güsewell and Koerselman 2002). In terrestrial ecosystems, tropical soil generally are N-rich but P-poor (owing to the prevalence of symbiotic N-fixers and highly weathered soils) and temperate forests are N-poor and P-rich (owing to lack of symbiotic N-fixer and weakly weathered soils) (Sollins 1998, Chadwick *et al.* 1999, Perakis and Hedin 2002, Hedin *et al.* 2003, McGroddy *et al.* 2004). The N:P ratio in herb community stage were significantly different from those of the other stages (shrub community stage, early forest community stage and climax forest community stage), but there were not significant differences among the shrub community stage, early forest community stage and climax forest community stage. N:P ra-

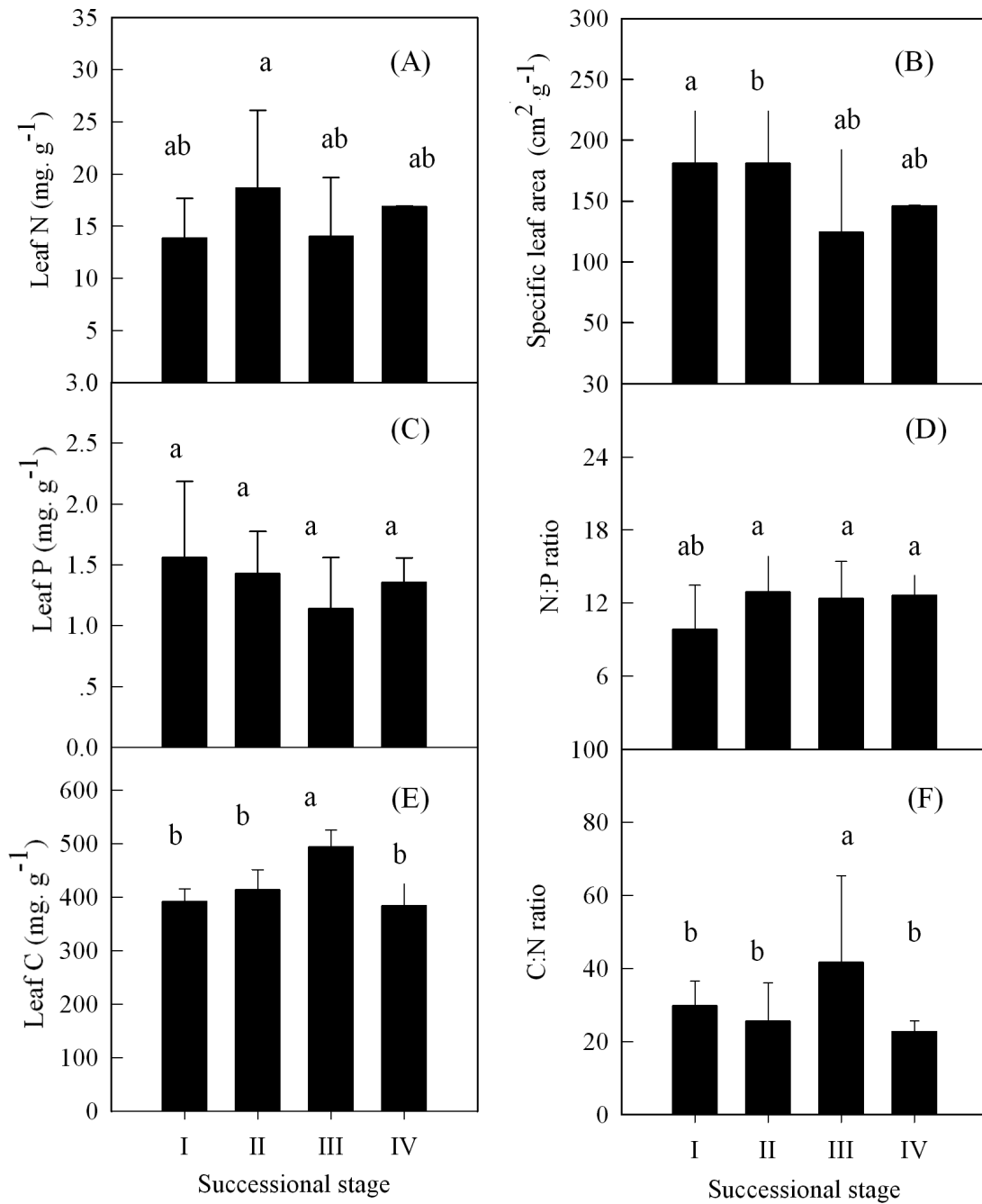


Fig. 2. Leaf stoichiometric traits and specific leaf area at the different successional stages in the Loess Plateau. Means and SD are presented. Bars with different letters mean significant differences at $P < 0.05$. In the horizontal axis, I – stands for the herb community stage (n = 6 species); II – stands for the shrub community stage (n=8 species); III – stands for the early forest community stage (n=3 species); IV – stands for the climax forest community stage (*Quercus liaotungensis* Koidz community)

tio shifts of the species were consistent along the successional sere, although the N:P ratio of the different species at a successional stage (given community) varied considerably, and this means that nutrient limitation of differ-

ent species differed in a given community. Some P-limited species emerged in N-limited plant communities, which is the niche differentiation maintaining the mechanism of species coexistence in plant community

(Drenovsky and Richards 2004, Güsewell 2004, Güsewell 2005). The leaf N:P ratio were higher in the shrub community stage than in the other three successional stages. The lowest N:P ratio (9.8) was found in the herb community stage, and thus the N:P ratio increased to 12.9 in the shrub community stage and declined to 12.4 in the early forest community stage, and increased at the end successional stages (e.g. 12.6 in the *Quercus liaotungensis* Koidz community). Phenotypic plasticity in leaf C:N:P ratio is explained in part by environmental nutrient availability, considering that increased nutrient availability leads to higher leaf nutrient contents (Aerts and Chapin 2000). As the succession go on, soil nitrogen and carbon tend to increase, while soil phosphorus tend to decrease (He *et al.* 2007). There were several reasons for the leaf N:P ratio higher in shrub community stage than in other three successional stages. Firstly, shrubs have higher productivity and higher growth rate, thereby requiring more nutrient during the growth period. Since soil N has a faster turnover rate than soil P, and most of soil P is not available to plants, so that shrub community stage have higher leaf N:P ratio than other successional stages. Secondly, in the early successional stages, soil nutrient availability is relatively poor due to large biomass removal by clear cutting and high nutrient leach from exposed soil. In the shrub community stage, the vegetation coverage increased and hydrologic losses of soil N decreased, so that soil N increased significantly. Furthermore, higher N concentration and higher N:P ratio in the shrub community stage than those in the other three successional stages were probably attributed to the presence of legumes (e.g. *Hippophae rhamnoides* (Linn.) and *Sophora viciifolia* Hance). Because legumes are associated with N-fixing bacteria such as *Rhizobium* spp, which can utilize atmospheric N₂, thus the N:P ratio of legumes were higher than those of other plant groups.

The C:N ratio in the early forest community stage significantly differed from those in the others stages (herb community stage, shrub community stage and climax forest community stage), but there were not significant difference among the herb community stage, shrub community stage and climax

forest community stage. The mean C:N ratio in the herb community stage of the secondary succession was 29.8, higher than that of grasslands in China (C:N ratio 17.9) (He *et al.* 2006). The C:N ratio in the shrub community stage (25.5) and in the climax forest community stage (22.7) were lower than that of global forests (C:N ratio 37.1) (McGroddy *et al.* 2004), but the C:N ratio in the early forest community stage (41.7) were higher than that of global forests. He *et al.* (2006) found that life form and genus identity explained more than 70% of the total variations in C:N ratio. Higher C:N ratio in the early forest community stage than in the other three successional stages was also probably attributed to the presence of conifer species (*Platyclusus orientalis* (Linn.) Franco). Because conifer forests showed higher C:N ratio compared with both temperate broadleaf and tropical forests, such leaf N use efficiency is thought to be a general nutrient use strategy of conifers (Aerts 1996).

Specific leaf area is a plant trait that appears extremely important in the regulation and control of plant functions such as carbon assimilation and carbon allocation. Species with low specific leaf area tend to have long average leaf life span (LL), low nutrient concentrations and lower maximum photosynthetic and dark respiration rates (McGroddy *et al.* 2004, Takashima *et al.* 2004, Warren and Adams 2004). Together, these traits form a spectrum of 'leaf economics' (Wright *et al.* 2004b), running from species with potential for quick returns on investments of nutrients and dry mass in leaves (e.g. species with high specific leaf area, such as many herbs, deciduous shrub and trees) to species with slower potential rate of return in terms of carbon acquisition (e.g. many evergreen shrubs and trees with low specific leaf area). Specific leaf area in the early forest community stage was lower than those in the other successional stages; it was probably attributed to the presence of evergreen tree (*Platyclusus orientalis* (Linn.) Franco). Herb and shrub community stage had higher specific leaf area than early forest community stage and climax forest community stage, and this is in accordance with that fast-growing species which have higher specific leaf area than slow-growing species, as was previously identified (Poorter and Bergkotte 1992, Wright

and Westoby 2000). Slow-growing species with lower specific leaf area was accompanied by long leaf lifespan (Wright *et al.* 2004b), but fast-growing species are expected to have relatively higher specific leaf area.

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