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Higher Species Diversity Occurs in More Fertile Habitats Without Fertilizer Disturbance in an Alpine Natural Grassland Community

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Abstract: Are there some relationships among species diversity and soil chemical properties of high altitude natural grasslands? Plant community composition and chemical properties of soil samples were compared to investigate the relationship between soil and species diversity, and the richness in Tibetan alpine grasslands. Results showed that species diversity was significantly positively related to soil organic matter (SOM), total nitrogen (TN), available nitrogen (AN), total phosphorus (TP), available phosphorus (AP), and available potassium (AK) in the high alpine grasslands. Margalef's species richness index was also significantly positively related to SOM, TN, AN, and TP. Most soil chemical properties showed significantly positive correlation with species diversity and Margalef's richness index. Our results suggested that higher plant species richness index and diversity occurred in more fertile soil habitats in high altitude natural grassland community. In practice, fertilization management for the restoration of degraded grassland should be conducted with reference to the nutrient levels of natural grassland without the additional artificial fertilizer and with higher species-diversity and richness index.

Keywords: Species richness; Soil properties; Species diversity; Biomass

Introduction

Many ecological studies have focused on aboveground–belowground relationships and interactions in ecosystems (van der Putten et al. 2001; Bardgett et al. 2005; De Deyn and Van der Putten 2005). Plant species diversity has positive feedback on patterns of nutrient cycling in natural ecosystems, and plant–soil feedback depends upon plant species composition and site-specific differences in abiotic and biotic soil properties (Bezemer et al. 2006). Disturbance and productivity are often considered as major factors regulating species richness in plant communities (Huston 1994; Pollock et al. 1998). Studies have

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reported that productivity and disturbance (Grime 1979; Tilman 1982), environmental variation (Grace 1999), grazing (Grace et al. 2000), light penetration and soil properties can all affect the species pool and play an important role in determining species richness and diversity in plant communities (Grace and Pugesek 1997; Grace and Jutila 1999; Wu et al. 2009).

Further, soil properties may be considered as an indicator of the effects of plant abundance and species richness (Stohlgren et al. 1999; Wu et al. 2011). Richness is often, but not always, highest at intermediate disturbance levels when the biomass and abundance were also highest (Connell 1978; Grime 1979; Keddy 2005). Meanwhile, richness also can be regulated by soil heterogeneity (Tilman 1982; Grace et al. 2000; Fernandez-Lugo et al. 2009). Plant–soil feedback plays an important role in determining plant community structure and maintaining species coexistence (van der Putten et al. 2001; Bardgett et al. 2005; Haase et al. 2008). Soil properties can potentially affect species coexistence by influencing plant invasions (Callaway et al. 2004), species composition (Bever 1994), and community structure (Stohlgren et al. 1999; Bashkin et al. 2003; Jacquemyn et al. 2003; Kardol et al. 2006). Plant species richness index was significantly influenced by SOM and nitrogen content, and there was a variety of direct and indirect processes whereby mineral soil properties could influence richness and diversity (Stohlgren et al. 1999; Bashkin et al. 2003). Soil properties can affect directly and indirectly plant communities and they are crucial for understanding the consequences of above- and below- ground trophic interactions for ecosystem functioning (Ettema and Wardle 2002), e.g. Species diversity decreased significantly with additional artificial nitrogen supply rates in calcareous grassland of Belgium (Jacquemyn et al. 2003). Qiu et al. (2004) and Luo et al. (2004) found that species richness index increased at first and decreased subsequently along the additional artificial fertilizer gradient in an alpine meadow community. On the contrary, Steinbeiss et al. (2008) had reported that plant species richness index and certain plant functional traits accelerate the build-up of new carbon pools and higher plant diversity mitigated soil carbon losses in deeper horizons. So they suggested that higher biodiversity might lead to higher soil carbon sequestration in the long-term. The strength of above–below ground linkages may therefore depend on the availability of soil nutrients in ecosystems (Haase et al. 2008). In addition, variation of species composition can often affect key ecosystem processes (Wardle and Zackrisson 2005; Wu et al. 2011) in grasslands because change of plant species composition has significant effects on soil nutrients.

We hypothesized that soil chemical properties were positively related to species diversity and richness in natural grassland communities without the additional artificial fertilizer. The main objective of this study was to determine whether there is a positive relationship between soil chemical properties and species diversity and richness in typical natural alpine grassland in the hinterland of the Qinghai-Tibetan Plateau under the condition of without additional artificial fertilizer.

1 Study Sites

The study was conducted in alpine grassland of the northern Tibet, China, which is located in the hinterland of the Qinghai-Tibetan Plateau. The location, altitude and dominant species of three sampling sites are shown in Table 1. The local mean annual temperature is below 0°C, ranging from -18°C to -10°C in winter and 7°C to 12°C in summer. Mean annual precipitation is 100 - 200 mm, with the main rainy period during the short, and cool summer. The vegetation is typical alpine grassland dominated by clonal graminoids. The soil types were classed as felty soils.

Table 1 Sampling location, altitude (m), main dominant species, and grassland type of the three studied alpine grassland communities

Sites	Location	Altitude	Dominant species
1	N 30°29′ E 90°40′	4318	*Carex moorcroftii* Falc ex Boot, *Artemisia frigida* Willd
2	N 30°85′ E 91°62′	4692	*Carex moorcroftii* Falc ex Boot, *Saussurea* spp.
3	N32°15′ E 91°40′	4780	*Carex moorcroftii* Falc ex Boot, *Stipa purpurea* Griseb.

2 Methods

2.1 Sampling

We selected three alpine meadow community blocks dominated by *Carex moorcroftii* (Table 1), which were located at different sites but had undergone similar moderate grazing management by a nomadic grazing from a medium density of Tibetan sheep and yaks (the approximate proportion of sheep vs yaks was about 2:1) by Tibetan herders. The stocking density was about 1.6 head hm^{-2}, which were decided by livestock carrying density (livestock carrying density = livestock (yak) numbers/pasture area). Sampling blocks (about 10 ha) were excluded from livestock grazing with fences during the growing season from April to October and were lightly grazed by Tibetan sheep and yaks only during the hay-stage in winter. Three repeated plots (30 m × 30 m) were arranged randomly in each sampling block. Then in each plot five quadrats (50 cm × 50 cm) were randomly selected for measuring species composition and collecting soil samples. All quadrats were sampled when peak biomass was reached in mid-August of 2009. The cover, height, density, and frequency of each species in quadrat were measured separately by the methods of Wu et al. (2013). The Shannon-Wiener diversity index (*H*) and Margalef's richness index (*R*) for each quadrats were calculated as:

Margalef's richness index (*R*):

$$R = (S - 1) / \ln N$$

Shannon-Wiener diversity index (*H*):

$$H = -\sum \mathrm{Pi}(\ln \mathrm{Pi})$$

where *S* is the total species numbers of each quadrats, *N* is the total numbers of individual for all species in a quadrat, and *Pi* is the abundance proportion of *i* species of each quadrats.

Five soil cores to the depth of 0-20 cm by diagonal pattern and then mixed to form one composite sample in each quadrat. The 45 composite soil samples were used to analyze soil properties. All soil samples were air-dried because of long-distance transportation from the study site. In the laboratory measures should be taken to prevent ammonium from being converted into ammonia and then volatilizing if soils were not kept cool and extracted for analysis as soon as possible after sampling in the field. All air-dried soils were passed through a 2 mm sieve. SOM was measured using the $K_2Cr_2O_7$ method; TN, AN, TP and AP were measured using air-dried samples according to standard procedures, TK was determined by a Perkin–Elmer analyst 300 atomic absorption spectrophotometer; AK were measured by ICP–AES with 3% $(NH_4)_2CO_3$ solution extract (Agriculture Chemistry Council, Soil Science Society of China 1983). The content of each nutrient properties was calculated by the proportions of SOM, TN, AN, TP, AP, TK and AK in dry weight of soil.

2.2 Data analysis

Differences in the Shannon-Wiener diversity index (*H*), Margalef's richness index (*R*), SOM, TN, AN, TP, AP, TK, and AK were tested by ANOVAs with a Tukey test for differences ($p < 0.05$) among the three wetland blocks. Correlations between soil chemical properties and diversity/richness indices (*H* and *R*) were calculated using the data from all sample quadrats by a linear model, after values were log-transformed. All calculations were done with SPSS 13.0 (SPSS Inc., Chicago, IL, USA) software.

3 Results and Discussions

The differences of plant community characteristics, aboveground biomass, cover, height and species numbers per square meter in the three study sites is shown in Table 2. The overall ANOVA results showed significant differences in community

Table 2 Description of plant community characteristics in the three study sites. Values (±S.D.) are means of 15 squares. Different letters indicate significant differences at P<0.05.

Sites	Biomass	Cover(%)	Height(cm)	Richness
1	341.10 ±10.20b	78±3a	7.5±2.5b	13±2a
2	182.17 ±10.15c	58±2b	4.5±1.8b	7±1b
3	396.43 ±9.94a	80±5a	12.4±3.2a	12±2a

Notes: Biomass = Aboveground biomass (g m^{-2}); Richness= Species richness (sps m^{-2})

Table 3 **Mean values (±S.D.) of soil organic matter (SOM), soil total nitrogen (TN), available nitrogen (AN), total phosphorus (TP), available phosphorus (AP), total potassium (TK) and available potassium (AK) in the three study sites (N = 15). Different letters indicate significant differences at P<0.05.**

Sites	SOM (%)	TN (%)	AN (mg/kg)	TP (%)	AP (mg/kg)	TK (%)	AK (mg/kg)
1	5.96±0.12b	33.12±1.06b	106.72±3.06a	0.061±0.003b	5.64±0.27a	1.65±0.04a	31.30±0.48a
2	5.71±0.30b	31.35±1.15b	106.79±3.18a	0.063±0.004ab	5.82±0.06a	1.67±0.02a	31.50±0.35a
3	6.21±0.02a	35.37±0.14a	110.19±2.09a	0.068±0.002a	5.84±0.05a	1.70±0.02a	21.12±0.09b

Table 4 **Results of ANOVA tests for the effects of sampling sites on Shannon-Wiener diversity index (*H*), Richness index (*R*), soil organic matter (SOM), total nitrogen (TN), available nitrogen (AN), total phosphorus (TP), available phosphorus (AP), total potassium (TK), and available potassium (AK) in this study. S.S. =Sum of Squares, M.S. = Mean Square.**

Index	S.S.	M.S.	F
H	1.507	0.753	27.079**
R	7.795	3.897	19.026**
SOM	1.297	0.649	13.913**
TN	0.749	0.374	39.552**
AN	60.856	30.428	3.361*
TP	0.000	0.000	9.022**
AP	0.370	0.185	5.581*
TK	0.010	0.005	5.011*
AK	3.018	1.509	9.783**

Notes: *df* = 2, * show correlation is significant at the 0.05 level, ** show correlation is significant at the 0.001 level.

diversity and richness (Table 3), and in soil nutrient contents among the three sites (Table 4).

Species diversity was significantly positively correlated with SOM, TN, AN, TP, AP, and AK in our alpine grassland communities (Figure 1). Species richness index was also significantly positively correlated with SOM, TN, AN, and TP (Figure 2). Most soil chemical properties showed significantly positive correlation with species diversity and Margalef's richness index in this study.

Consistent with our hypothesis, we found that plant species richness index and diversity were significantly positively related to SOM, TN, AN and TP in alpine grasslands. All these suggested that plant species richness index and diversity were related to soil properties. Plant composition and diversity of chemical compounds in litter are potentially important functional traits affecting decomposition, simple metrics like plant species richness index may fail to capture variation in these traits (Meier and Bowman 2008; Wu et al. 2011). Species composition, litter compounds and all aboveground community traits that influence soil C and N cycling have potential effects on below-ground ecosystem function (Wu et al. 2013). Soil properties can impose direct and indirect constraints on local species diversity, however, decomposition of plant leaf and root litter is the most important process supplying nitrogen and phosphorus to plants, and litter chemical composition and species diversity are significantly related to functional traits of soil C and N cycling (Hattenschwiler et al. 2005; Meier and Bowman 2008; Wu et al. 2013). Soil properties also may be influenced by the identity (and litter quality) of plant species (Meier and Bowman 2008). Studies suggest that mechanisms of plant–soil feedback depend on plant species, plant taxonomic (or functional) groups and site-specific differences in abiotic and biotic soil properties (Bezemer et al. 2006; Wu et al. 2011). Our study indicated that higher species diversity and richness may be used as an indicator of soil nutrient properties in similar natural grasslands which had not undergone serious disturbance and the additional artificial fertilizer.

Based on our results, it can be advanced that higher nutrient concentration soil have positive effects on plant species richness index and diversity because higher nutrient availability enables the sharing of resources among many species within a plant community. Meanwhile, exogenous fertilization with diammonium phosphate ($(NH_4)_2HPO_4$) significantly decreased species richness index and diversity in alpine grassland (Luo et al. 2004), because palatable graminoids out-competed other species under fertilization (Qiu and Luo 2004) and more fertile soil habitats (Wu et al. 2009). So, we suggest that there will be a trade-off between soil nutrient availability and species diversity: too high or too low soil nutrients can lead to the reduction of species diversity and richness in plant communities (De Deyn et al. 2005). This pattern suggests that optimal fertilization management for restoration of degraded grasslands should be taken into account given the close relationship between soil nutrient properties and plant diversity and richnesss during the restoration management.

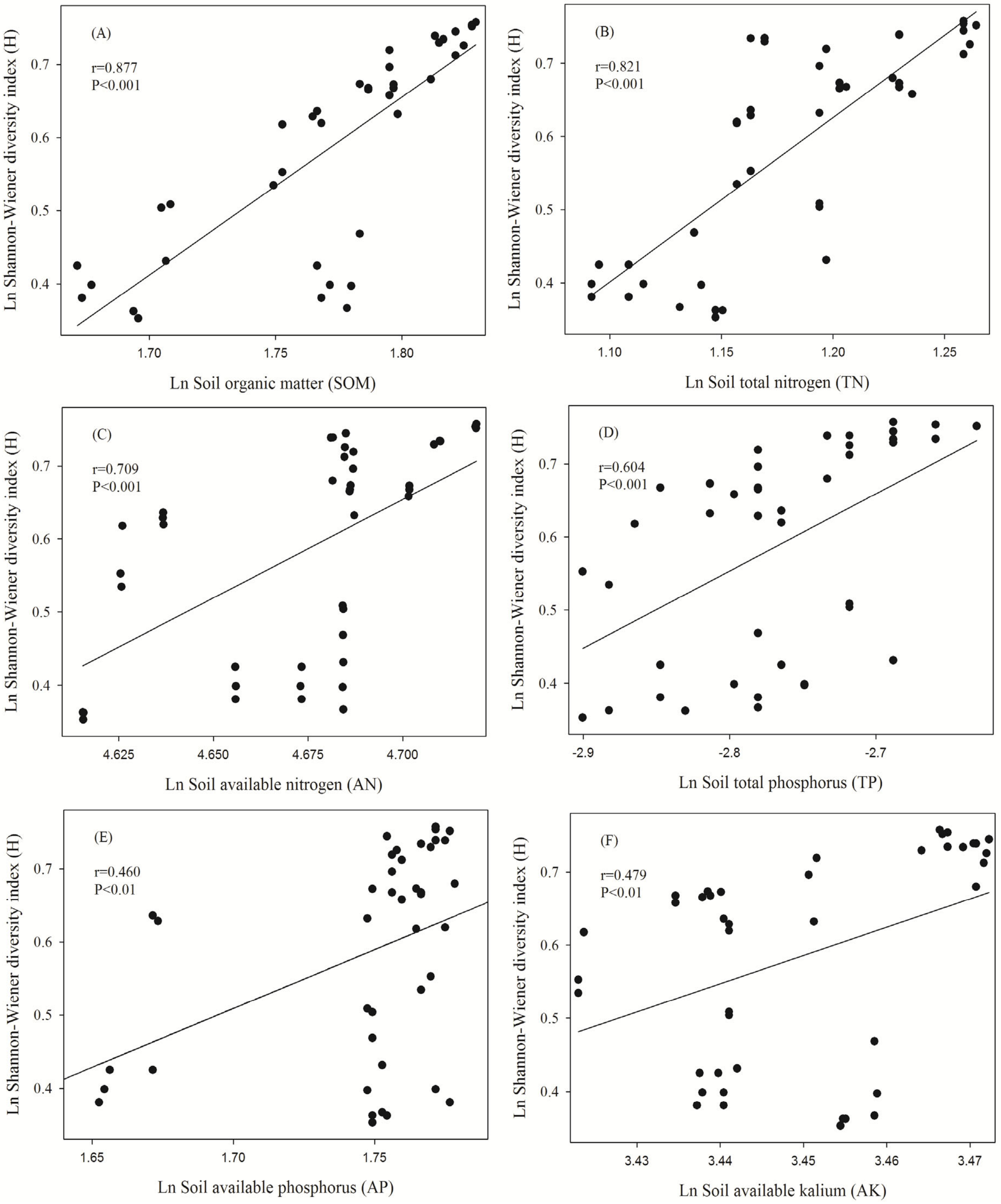

Figure 1 Relationships of Shannon-Wiener diversity index (*H*) and soil organic matter (A, SOM, %), total nitrogen (B, TN, g kg^{-1}), available nitrogen (C, AN, mg kg^{-1}), total phosphorus (D, TP, %), available phosphorus (E, AP, mg kg^{-1}), and available potassium (F, AK, mg kg^{-1}) in studied alpine wetlands.

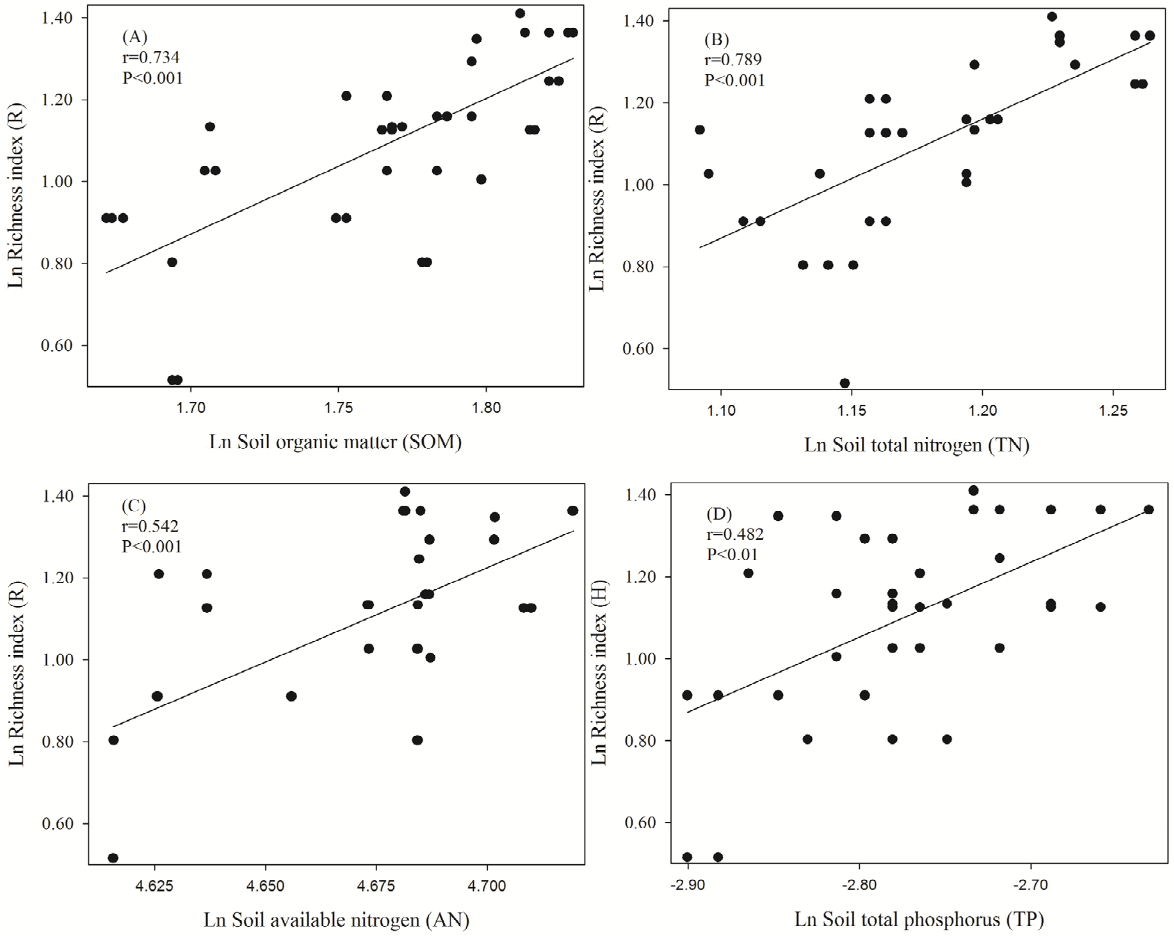

Figure 2 Relationships of Margalef's richness index (*R*) and soil organic matter (A, SOM, %), total nitrogen (B, TN, g kg^{-1}), available nitrogen (C, AN, mg·kg^{-1}), and total phosphorus (D, TP, %) in studied alpine wetlands.

4 Conclusion

The relationships of species diversity and richness in plant communities and soil chemical properties in this study may have potential contribution to understanding of plant-grazer-soil feedbacks in Tibetan alpine grasslands. However, the sample time is also important for change of soil properties and plant diversity and understanding of the processes and mechanisms involved in these interactions is still very incomplete, and should be the focus of subsequent study. Our results suggested that higher plant species richness index and diversity occurred in more fertile soil habitats in high altitude natural grassland community. In practice, fertilization management for the restoration of degraded grassland should be conducted with reference to the nutrient levels of natural grassland without the additional artificial fertilizer and with higher species-diversity and richness index.

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