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Stoichiometric shifts in surface soils over broad geographical scales: evidence from China's grasslands

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ABSTRACT

Aim The identification of stoichiometric flexibility is crucial for understanding carbon–nitrogen–phosphorus (C–N–P) interactions and ecosystem dynamics under a changing environment. However, current evidence of stoichiometric flexibility mainly comes from manipulation experiments, with little evidence from large-scale observations.

Location Alpine and temperate grasslands across northern China.

Methods Using soil profiles derived from a historical national soil inventory and a contemporary regional soil survey across China's grasslands, we examined temporal changes in topsoil C:N:P ratios over recent decades.

Results Topsoil C:N ratios of five major grassland types exhibited some flexibility but did not show significant changes over the sampling interval. Non-significant changes in topsoil C:N ratios were observed both in alpine grasslands on the Tibetan Plateau and in temperate grasslands on the Inner Mongolian Plateau. Consistent with the relatively stable C:N ratios, the slope of the soil C–N stoichiometric relationship did not differ significantly between the two sampling periods. Soil N:P ratios in the surface layer increased significantly over the sampling interval, however, with an overall increase of 0.60 (95% confidence interval 0.58–0.62). A larger increase in soil N:P ratio was found in temperate grasslands on the Inner Mongolian Plateau than in alpine grasslands on the Tibetan Plateau. Moreover, the slope of the soil N–P stoichiometric relationship in these grassland ecosystems became steeper over the sampling interval.

Main conclusions These results demonstrate the stability of topsoil C:N stoichiometry but variability in N:P stoichiometry over broad geographical scales, highlighting that soil C and N are tightly coupled, but N and P tend to be decoupled under a changing environment.

Keywords

Carbon–nitrogen–phosphorus interactions, carbon:nitrogen:phosphorus ratio, ecological stoichiometry, grassland ecosystems, soil inventory, stoichiometric flexibility.

INTRODUCTION

Rapid human population growth and increased industrial activity have led to profound environmental changes, such as elevated atmospheric carbon dioxide (CO_2) concentrations and enhanced nitrogen deposition (Forster *et al.*, 2007). These global environmental changes can exert strong effects on terrestrial biogeochemical cycles, including the carbon (C) (Ballantyne *et al.*, 2012), nitrogen (N) (Hietz *et al.*, 2011) and phosphorus (P) cycles (Marklein & Houlton, 2012). Altered biogeochemical cycles may lead to stoichiometric shifts in C:N:P ratios among various ecosystem components (Elser *et al.*, 2010; Yang *et al.*,

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2011; Sardans *et al.*, 2012). The stoichiometric flexibility of elemental ratios could in turn affect ecosystem structure and biogeochemical cycles (Elser *et al.*, 2009; Sistla & Schimel, 2012). For instance, the flexibility of the C:N ratio could regulate the terrestrial carbon cycle through its effects on decomposition processes (Sistla & Schimel, 2012). A decrease in the C:N ratio in plant litter or mineral soil may be favourable for microbial decomposition (Norby & Cotrufo, 1998) and could potentially amplify the positive feedback between the terrestrial carbon cycle and climate warming (Davidson & Janssens, 2006). Conversely, an increase in the C:N ratio in plant litter or mineral soil may inhibit microbial decomposition (Norby & Cotrufo, 1998) and so lead to progressive nitrogen limitation of terrestrial carbon sequestration (Luo *et al.*, 2004). Increased N:P ratios in leaf, litter or soil organic matter could lead to shifts in ecosystem nutrient limitation from nitrogen towards phosphorus, ultimately resulting in changes in community composition and ecosystem function (Wardle *et al.*, 2004; Elser *et al.*, 2009; Crowley *et al.*, 2012; Peñuelas *et al.*, 2012). Thus, the identification of stoichiometric flexibility could improve our understanding of C–N–P interactions and ecosystem dynamics under a changing environment (Elser *et al.*, 2010; Sistla & Schimel, 2012).

Stoichiometric flexibility under global change is of great interest among the global change research community. Many manipulation experiments have demonstrated that increased CO_2 and additional nitrogen inputs might drive the dynamics of C:N ratios in opposite directions; i.e. elevated CO_2 usually increases C:N ratios (Cotrufo *et al.*, 1998; Norby *et al.*, 2001; Luo *et al.*, 2006; Yang *et al.*, 2011), whereas additional nitrogen inputs reduce C:N ratios in both plant and soils (Yang *et al.*, 2011; Sardans *et al.*, 2012). These experiments have also illustrated that elevated CO_2 does not usually exert significant effects on the N:P ratio, whereas additional nitrogen inputs increase N:P ratios in both plant and soils (Sardans *et al.*, 2012). These experiments have further suggested that warming and variability in precipitation could also lead to flexibility of C:N:P ratios in various ecosystem components (Sardans *et al.*, 2012). Taken together, these experimental results have greatly advanced our understanding of the dynamics of stoichiometric ratios under various global change scenarios, but it remains unknown if the predicted flexibility of C:N:P ratios is realistic in natural ecosystems that experience environmental changes. It is also unclear whether stoichiometric flexibility has occurred across broad geographical scales. To resolve these two issues, it is necessary to explore temporal changes in C:N:P ratios in natural ecosystems that have undergone significant environmental change over long periods and across large spatial scales. Most existing evidence of temporal changes in C:N:P ratios at large scales is, however, derived from lake ecosystems (Elser *et al.*, 2009; Crowley *et al.*, 2012), and little is known about stoichiometric changes in surface soils across temporal and spatial scales.

China's grasslands provide an ideal opportunity to evaluate the temporal dynamics of topsoil C:N:P ratios across a broad spatial scale. First of all, China's grasslands have experienced significant environmental changes, including elevated CO_2, N deposition, climate warming and precipitation variability (Editorial Committee of National Assessment Report of Climate Change, 2011). These environmental changes have been shown to exert significant effects on regional biogeochemical cycles (e.g. Tian *et al.*, 2011), which could then induce stoichiometric shifts of topsoil C:N:P ratios in these grassland ecosystems. Second, China's government launched a national soil inventory during the period 1979–1989, which generated a valuable historical dataset of soil C:N:P ratios (National Soil Survey Office, 1998). This dataset has been used to evaluate spatial patterns of soil C:N:P stoichiometry across the country (Tian *et al.*, 2010) and offers the possibility of quantifying stoichiometric shifts in topsoil C:N:P ratios by resampling across the grassland area. However, a repeated regional soil survey has not yet been conducted to examine the stoichiometric shifts in the surface soils across China's grasslands.

In this study, we evaluated temporal changes in topsoil C:N:P ratios of China's grasslands over recent decades. To this end, we conducted a regional soil survey during the period 2001–2005, and sampled 327 sites across the northern part of the country. We then estimated the spatial distributions of topsoil C:N:P ratios during the 1980s and the 2000s by interpolating site-level measurements at the regional scale. We further evaluated the stoichiometric changes over the sampling interval along a grassland transect, where kriging interpolation proved a reliable estimation during both sampling periods. Overall, this study aimed to answer the following questions: (1) Had topsoil C:N ratios and the slope of C–N stoichiometric relationship changed over the sampling interval? (2) Had topsoil N:P ratios and the slope of N–P stoichiometric relationship exhibited any flexibility during recent decades?

MATERIALS AND METHODS

Study area

The grassland area of China is mainly distributed across the northern part of the country, and located between the latitudes 29.25° N and 49.51° N and the longitudes 74.93° E and 120.06° E. It consists primarily of alpine grasslands on the Tibetan Plateau, temperate grasslands on the Inner Mongolian Plateau, and mountain meadow in the Xinjiang area. There are two types of alpine grasslands (alpine steppe and alpine meadow), three types of temperate grasslands (desert steppe, typical steppe and meadow steppe) and mountain meadow (Chinese Academy of Sciences, 2001). The mean annual temperature and mean annual precipitation of the study area range from –6.2 to 8.8 °C and from 92.3 to 694.0 mm, respectively. Related soil types of the grassland area are cambisols, haplic calcisols, kastanozems, chernozems and umbric leptisols/dystric cambisols (Yang *et al.*, 2007).

Soil inventory

Soil profiles derived from China's Second National Soil Inventory during 1979–1989 and from a regional soil survey during

2001–2005 were used to detect changes in topsoil C:N:P ratios across the study area. The national soil inventory characterized 275 soil profiles that were representative of the grasslands across the northern part of the country. Major soil physical and chemical properties (layer thickness, soil organic C, total N and total P concentration) were reported for all these soil profiles (Wu, 1991; Hou & Zeng, 1992; Feng & Wang, 1993; Zhao, 1993; Gong, 1994; Liu, 1995). The regional soil survey covered 327 sites (sampled at depths of 0–10 cm, with three replicates at each site) across the study area (Yang *et al.*, 2010, 2012a). Soil samples were air-dried, sieved through 2-mm mesh, hand-picked to remove plant detritus, and ground into fine powder using a ball mill (NM200, Retsch, Hann, Germany). Similar to the national soil inventory, soil organic carbon (SOC) content was determined using the Walkley–Black method (Nelson & Sommers, 1982), total nitrogen content (TN) was measured with the Kjeldahl method (Jackson, 1973), and phosphorus content (TP) was determined by the molybdate colorimetric test after perchloric acid digestion (Sommers & Nelson, 1972).

Spatial interpolation

We conducted kriging interpolation to generate spatial patterns of the topsoil C:N ratios. The mean of the measured values of all sampling sites within the same area of a $0.05° \times 0.05°$ grid was calculated for the spatial interpolation (Yang *et al.*, 2012a,b). To explore spatial dependence in the underlying variable, semivariograms that described the relationship between the lag distance (x) and the semivariance of the underlying variable within the lag distance (y) were constructed using ArcGIS 10.0 (Esri, Redlands, CA, USA). Three types of semivariance models (Gaussian, exponential and spherical) were tested to construct the semivariograms (Yang *et al.*, 2012a,b), and an exponential model was selected because it provided the best fit to the data. Based on site-level measurements and exponential semivariograms, kriging interpolation was then conducted using idrisi32 (Clark Labs, Worcester, MA, USA).

Similarly, kriging interpolation was performed to estimate spatial distributions of soil N:P ratios in the surface layer. To create a spatial surface for kriging estimation, we averaged the measured values of all sampling sites within the same grid ($0.05° \times 0.05°$). Mean soil N:P ratios were then $\log_{10}$-transformed due to their log-normal distribution [Kolmogorov–Smirnov test for $\log_{10}$(N:P) ratios, $P > 0.05$]. An exponential model was used to construct the semivariograms for the topsoil N:P ratios. The interpolated $\log_{10}$(N:P) ratios were finally transformed back to the original scale based on the mean and variance of the kriging estimations (Cressie, 1993).

Transect-scale evaluation

We performed a transect-scale comparison to explore changes in stoichiometric ratios over recent decades. Based on the kriging results, we calculated differences in spatial distributions of topsoil C:N:P ratios between the 1980s and the 2000s, to quantify their temporal changes along a grassland transect that was sampled intensively with evenly distributed sites. We then overlaid the spatial distributions of stoichiometric ratios and their differences over the vegetation map of northern China (Chinese Academy of Sciences, 2001) to obtain the sizes and changes of stoichiometric ratios in five major grassland types within the grassland transect. The transect, which covers 8.94×10^5 km^2, extends from the central-southern Tibetan Plateau to the eastern Inner Mongolian Plateau and covers *c.* 45% of the total grassland of China (Appendix S4). The transect has been widely used in grassland carbon-cycling studies in China (e.g. Hu *et al.*, 2010; Shi *et al.*, 2012). In this study, the transect was selected because the sample size was comparable between the two sampling periods (112 sites during the 1980s versus 124 sites during the 2000s), and kriging interpolation provided reliable predictions within the transect for both sampling periods.

Error analysis

We determined the measurement error using repeated measurements on the same soil sample (for SOC) or repeated measurements on the standard material (for TN and TP). The relative error in SOC concentration was within 3%, and the relative error in TN and TP concentrations was within 2% (Appendix S1). The measurement error involved in this study was within the range expected for chemical analyses on soil samples (Bao, 2000). Moreover, the relative error was random, with a normal distribution and with the average error close to zero for all three elements, demonstrating that our laboratory measurements did not induce any systematic shifts.

We also quantified potential errors from the spatial interpolation and from the field survey by conducting a two-step Monte Carlo simulation (Yang *et al.*, 2012a,b). First, we examined kriging-based uncertainties within the selected grassland transect, and between the two sampling periods. Based on the mean and variance values derived from kriging interpolation, random sampling from the normal distribution of the topsoil C:N ratios (or log-normal distribution of soil N:P ratios) was performed for each grid to quantify errors in the spatial analysis. Random sampling was performed for both the 1980s and the 2000s, from which we obtained the difference between the two sampling periods. Second, we evaluated the sampling representativeness error in the field survey for the two sampling periods using the sample size as an indicator. A large sample size and evenly distributed sample sites reflect good sampling representation and vice versa. We randomly drew a certain number of grids based on the sampled values from the previous step and calculated the average C:N ratios (or N:P ratios) in the topsoil for both periods, and also for the difference between the two sampling periods. The sample size was equal to the number of sites investigated during the field soil survey. The process was repeated 10,000 times to determine the mean and 95% confidence interval. All statistical analyses were performed in R (R Development Core Team, 2012).

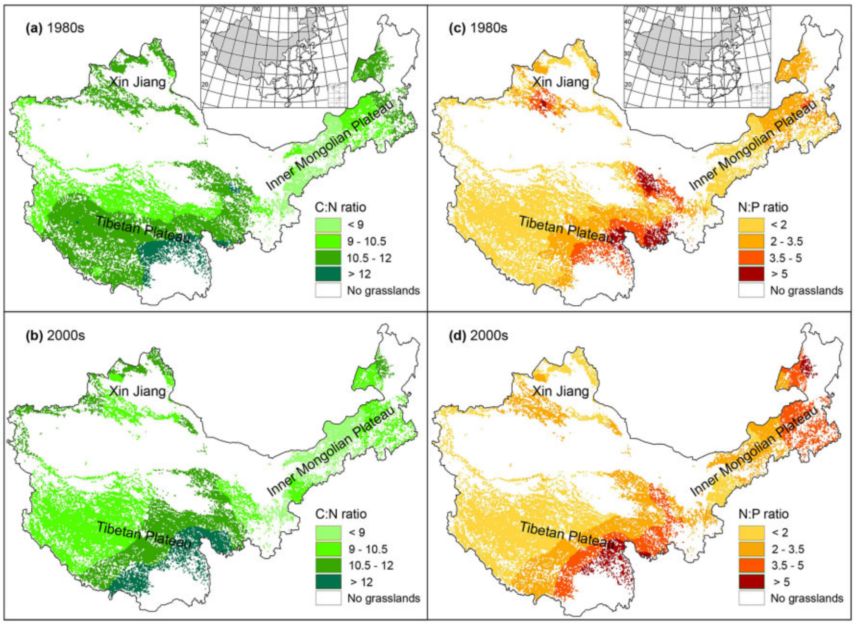

Figure 1 Spatial distributions of topsoil C:N ratios (a–b) and N:P ratios (c–d) across China's grasslands during the two sampling periods, generated using kriging interpolation of the site-level measurements. Inset: location of China's grassland ecosystems.

RESULTS

Spatial distributions of topsoil C:N:P ratios

Exponential models described the semivariograms of topsoil C:N ratios for both the sampling periods (Appendix S2a,b). The nugget: nugget + sill ratio was estimated at *c.* 1:2 for the 1980s and 1:3 for the 2000s, indicating relatively strong spatial dependence of the target variable across the study area. Likewise, exponential models provided the best fit between the semivariance and lag distance for the $\log_{10}$(N:P) ratios in the topsoil (Appendix S2c,d), with a nugget: nugget + sill ratio of *c.* 1:2 for the 1980s and 1:4 for the 2000s. The predicted soil C:N:P ratios accorded well with the measured values for both sampling periods (Appendix S3). The mean error was estimated to be –0.019 and –0.006 for the 1980s (Appendix S3a,c) and –0.015 and –0.001 for the 2000s (Appendix S3b,d), demonstrating that spatial patterns of topsoil C:N:P ratios could be reliably estimated through spatial interpolation. In particular, kriging interpolation provided robust estimation along a transect from the central-southern Tibetan Plateau to the eastern Inner Mongolian Plateau (Appendix S4). Topsoil C:N ratios decreased towards the north-west on the Tibetan Plateau and towards the south-west on the Inner Mongolian Plateau (Fig. 1a,b). There was, however, no clear spatial variation in topsoil C:N ratios in the Xinjiang area. In a similar manner, topsoil N:P ratios exhibited spatial heterogeneity across the study area (Fig. 1c,d), decreasing from the south-east to the north-west on the Tibetan Plateau and from the north-east to the south-west on the Inner Mongolian Plateau, but without any apparent patterns in the Xinjiang area.

Temporal changes in topsoil C:N:P ratios

Topsoil C:N ratios of the five major grassland types did not show significant changes between the two sampling periods ($P > 0.05$; Fig. 2a). Although not statistically significant, the dynamics of topsoil C:N ratios was different among various grassland types, with slight increases in alpine grasslands (i.e. alpine steppe and alpine meadow) on the Tibetan Plateau, but slight decreases in temperate grasslands (i.e. desert steppe, typical steppe and meadow steppe) on the Inner Mongolian Plateau (Fig. 2a; Table 1). SOC across these grassland types had a linear relationship with the TN content during both sampling periods ($y = 13.49x - 0.46$, $r^2 = 0.93$, $P < 0.001$ for the 1980s; $y = 12.02x - 0.13$, $r^2 = 0.89$, $P < 0.001$ for the 2000s; Fig. 3a). The linear relationship between SOC and TN demonstrates a strong stoichiometric link between C and N in mineral soil. Interestingly, the slope of the linear relationship between SOC and TN in these grassland ecosystems did not differ significantly between the two sampling periods (13.49 for the 1980s; 12.02 for the 2000s; $P > 0.05$; Fig. 3a). This is logically consistent with the non-significant changes in topsoil C:N ratios (Fig. 2a), providing further evidence that the kriging analysis successfully captured the direction of the stoichiometric flexibility over recent decades.

Soil N:P ratios in the surface layer had increased significantly from 2.9 during the 1980s to 3.5 during the 2000s ($P < 0.05$), with an average increase of 0.60 and a 95% confidence interval of 0.58–0.62. We observed a similar direction in the dynamics of topsoil N:P ratios, but the magnitude of those changes varied among the grassland types (Fig. 2b). Soil N:P ratios in temperate grasslands on the Inner Mongolian Plateau showed larger increases than those in alpine grasslands on the Tibetan Plateau. More specifically, the largest increase of the soil N:P ratios (1.12; 95% confidence of 0.82–1.41) was observed in meadow steppe on the Inner Mongolian Plateau, while the lowest (0.14; 95% confidence of 0.10–0.17) was found in alpine meadow on the Tibetan Plateau (Table 1). Consistent with the elevated N:P ratios, the slope of the positive soil N–P relationship in these grassland ecosystems increased from 0.57 during the 1980s to 1.39 during the 2000s ($P < 0.05$, Fig. 3b).

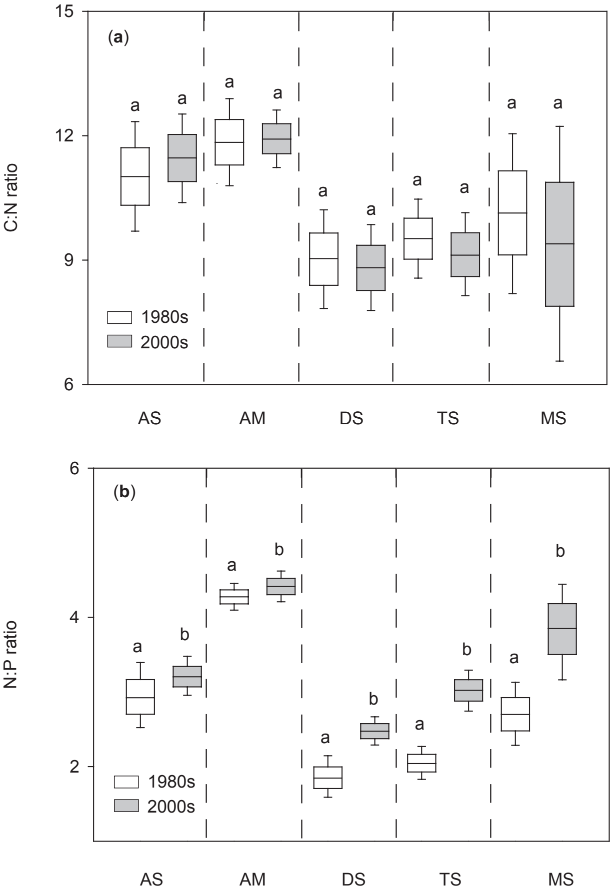

Figure 2 Box-and-whisker plots showing changes in (a) topsoil C:N ratios and (b) topsoil N:P ratios among various grassland types within a transect from the 1980s to the 2000s. Different letters indicate significant differences between the two sampling periods (Tukey's test, $P < 0.05$). AS, alpine steppe; AM, alpine meadow; DS, desert steppe; TS, typical steppe; MS, meadow steppe.

DISCUSSION

Relatively stable topsoil C:N ratios across Chinese grasslands

Topsoil C:N ratios across the grassland area of China did not show statistically significant changes over the sampling interval. The relatively stable soil C:N ratios observed in this study accord with observations derived from long-term chronosequences, where C:N ratios in litter, forest floor and mineral soil had been reported to remain constant over the age sequence (Yang & Luo, 2011). These two lines of evidence highlight that carbon and nitrogen are closely coupled in natural ecosystems that experience gradual changes in environmental conditions (Schimel *et al.*, 1997; Cleveland & Liptzin, 2007; Xu *et al.*, 2013). This raises the question of what the potential mechanisms responsible for soil C–N coupling across China's grasslands are. Relatively constant soil C:N ratios may be driven by the similar direction and proportional magnitude in soil carbon and nitrogen changes over recent decades (Yang & Luo, 2011). To test this hypothesis, we examined changes in soil carbon and nitrogen concentrations between two sampling periods, using data from the historical soil inventory during the 1980s and a soil survey during the 2000s. We also explored associations between changes in soil C:N ratios and soil carbon dynamics and between changes in soil C:N ratios and soil nitrogen dynamics. Our analyses indicated that neither soil carbon (Appendix S5a) nor nitrogen concentrations (Appendix S5b) showed significant changes from the 1980s to the 2000s. Moreover, changes in soil C:N ratios were positively correlated with changes in the soil carbon concentration ($r^2 = 0.25$, $P < 0.05$; Fig. 4a) but did not exhibit any significant association with soil nitrogen dynamics ($P = 0.26$; Fig. 4b), indicating that the changing direction and magnitude of the soil C:N ratios largely depended on soil carbon changes. Taken together, these analyses suggest that relatively stable soil carbon and nitrogen concentrations between the two sampling periods could be responsible for non-significant changes in topsoil C:N ratios across the study area. In addition, relatively stable soil C:N ratios may also be attributed to the relatively short time period of this study, although similar analyses have revealed that topsoil N:P ratios significantly increased in these grassland ecosystems over the same period (Fig. 2b), suggesting that the study period may not be the main factor driving the lack of significant changes in soil C:N ratios over the sampling interval.

Widespread increase in topsoil N:P ratios across China's grasslands

Topsoil N:P ratios across the grassland area exhibited a widespread increase from the 1980s to the 2000s. The increased topsoil N:P ratios observed in this study, together with reported increases in N:P ratios in both litter and soil during long-term ecosystem development (Wardle *et al.*, 2004), suggest that nitrogen and phosphorus tend to be decoupled under changing environments (Cleveland & Liptzin, 2007). It has been proposed that an increase in N:P ratios over time could be induced by reduced phosphorus content, because phosphorus is often subjected to leaching and occlusion in strongly weathered soils, whereas nitrogen is usually replenished through biological nitrogen fixation and atmospheric nitrogen deposition (Wardle *et al.*, 2004; Vitousek *et al.*, 2010). Consistent with this hypothesis, soil nitrogen concentrations remained relatively stable across the study area (Appendix S5b), whereas soil phosphorus concentrations decreased significantly during the study period (Appendix S5c). Moreover, changes in soil N:P ratios were positively correlated with changes in soil nitrogen concentrations ($r^2 = 0.73$, $P < 0.05$; Fig. 4c) but negatively associated with changes in soil phosphorus concentrations ($r^2 = 0.62$, $P < 0.05$; Fig. 4d), indicating that the changing direction and magnitude of the topsoil N:P ratios depended on both soil nitrogen and phosphorus dynamics. Taken together, these analyses suggest that the elevated topsoil N:P ratios across the study area were largely due to the decline of soil phosphorus concentrations. Interestingly, the spatial

Table 1 Soil C:N:P ratios in the surface layer and theirs changes among five major grassland types across northern China from the 1980s to the 2000s.

Grassland type	C:N ratio 1980s	C:N ratio 2000s	Change in C:N ratio	N:P ratio 1980s	N:P ratio 2000s	Change in N:P ratio
AS	11.02 (10.55, 11.48)	11.46 (11.08, 11.85)	0.44 (−0.19, 1.05)	2.95 (2.79, 3.10)	3.21 (3.12, 3.30)	0.27 (0.14, 0.40)
AM	11.84 (11.56, 12.14)	11.92 (11.77, 12.08)	0.07 (−0.22, 0.36)	4.28 (4.25, 4.30)	4.41 (4.37, 4.46)	0.14 (0.10, 0.17)
DS	9.02 (8.65, 9.39)	8.81 (8.45, 9.17)	−0.22 (−0.73, 0.30)	1.86 (1.77, 1.95)	2.48 (2.41, 2.54)	0.61 (0.53, 0.68)
TS	9.52 (9.28, 9.75)	9.13 (8.82, 9.44)	−0.38 (−0.77, 0.02)	2.05 (1.99, 2.10)	3.02 (2.93, 3.11)	0.97 (0.91, 1.03)
MS	10.13 (9.21, 11.06)	9.37 (6.88, 11.84)	−0.72 (−2.72, 1.26)	2.71 (2.50, 2.92)	3.83 (3.25, 4.37)	1.12 (0.82, 1.41)

AS, alpine steppe; AM, alpine meadow; DS, desert steppe; TS, typical steppe; MS, meadow steppe. Values in parentheses denote the 95% confidence intervals of the mean; they were estimated using a Monte Carlo approach with 10,000 iterations.

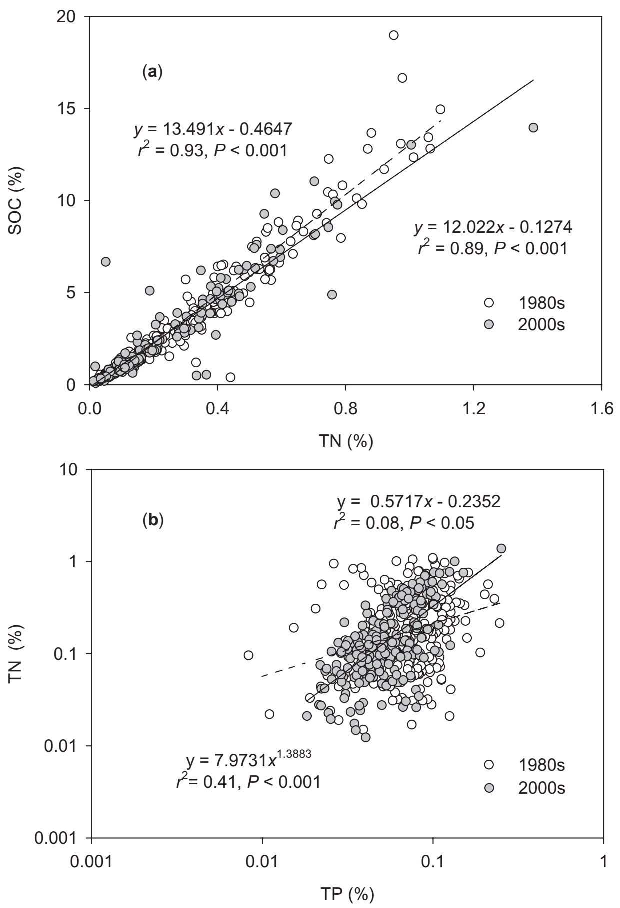

Figure 3 Stoichiometric associations (a) between topsoil organic carbon (SOC) and total nitrogen content (TN), and (b) between total nitrogen and total phosphorus (TP) content across China's grasslands during the two sampling periods.

variation in foliar N:P ratios across China's grasslands was also primarily determined by leaf phosphorus content (He *et al.*, 2008).

The increase in topsoil N:P ratios could also be driven by an imbalance between nitrogen and phosphorus inputs from the atmosphere into the biosphere (Peñuelas *et al.*, 2012). It has been reported that atmospheric phosphorus deposition was very limited across the study area (Mahowald *et al.*, 2008). In contrast, increased anthropogenic nitrogen emissions due to rapid economic development in China have induced significant nitrogen deposition across the northern part of the country, even on the remote Tibetan Plateau where there is relatively little local human disturbance (Lü & Tian, 2007; Liu *et al.*, 2013). Along the grassland transect selected in this study, the rate of atmospheric nitrogen deposition varied from 5.2 to 18.7 kg N ha^{-1} yr^{-1} and had accelerated during the past decade (Lü & Tian, 2007). Moreover, considering other components of atmospheric nitrogen deposition, such as particulate NH_4^+ in dry deposition and dissolved organic nitrogen in wet deposition, the rate of nitrogen deposition may be higher than previously reported across the study area (Liu *et al.*, 2011). Given the low proportion of inorganic nitrogen in the total nitrogen pool in the soil, atmospheric nitrogen deposition could not directly alter soil N:P stoichiometry through its effects on total nitrogen content in the soil (Appendix S5b). Atmospheric nitrogen deposition may, however, indirectly alter soil N:P stoichiometry through its effects on plant N:P stoichiometry. A recent manipulation experiment demonstrated that atmospheric nitrogen deposition could lead to significant increases of N:P ratios in both green and senesced leaves in a semi-arid grassland on the Inner Mongolian Plateau (Lü *et al.*, 2012). Given that plant detritus is the major source of soil organic matter (Davidson & Janssens, 2006), atmospheric nitrogen deposition could be indirectly responsible for increased topsoil N:P ratios, as frequently observed in site-level nitrogen addition experiments (Sardans *et al.*, 2012).

CONCLUSIONS

This study provides the first large-scale evidence of temporal changes in topsoil C:N:P ratios in China's grasslands over recent decades. Our results indicate that the topsoil C:N ratios did not change significantly between the two sampling periods, suggesting that carbon and nitrogen were closely coupled under a changing environment. The relatively stable soil C:N ratios observed in this study, together with the relatively constant soil organic carbon stock reported in our earlier analyses (Yang

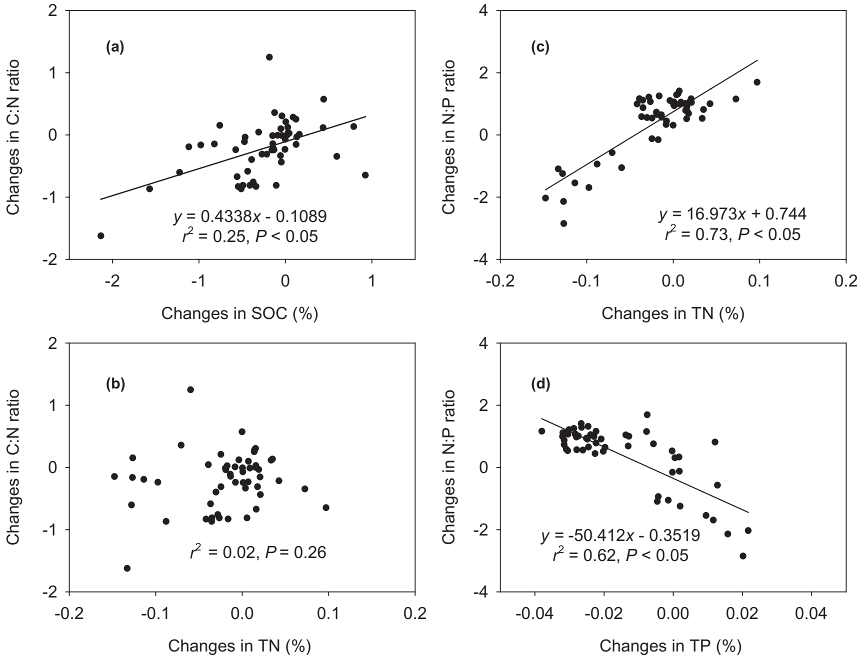

Figure 4 Relationships between changes in topsoil C:N:P ratios and changes in soil organic carbon (SOC), total nitrogen (TN) and total phosphorus (TP) concentrations across 53 community types within the grassland transect across northern China.

et al., 2010), highlight that neither the quality nor the quantity of soil organic matter across China's grasslands has been reduced from the 1980s to the 2000s. Such stability is favourable for maintaining ecosystem function under a changing environment. Our results also reveal that topsoil N:P ratios showed significant increases between the two sampling periods, suggesting that nitrogen and phosphorus may be decoupled under a changing environment. The N–P decoupling may lead to increased P limitation in these globally important ecosystems. Additional research is required to gain a deeper understanding of the implications of such changes in grassland soil biogeochemistry.

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SUPPORTING INFORMATION

Additional Supporting Information may be found in the online version of this article at the publisher's web-site.

Appendix S1 Measurement error of elemental concentrations in grassland soils across northern China.
Appendix S2 Semivariance models used to interpolate the spatial distributions of topsoil C : N : P ratios across China's grasslands.
Appendix S3 Frequency distributions of the difference between interpolated and measured topsoil C : N : P ratios across China's grasslands.
Appendix S4 Spatial patterns of kriging variance in topsoil C : N : P ratios across China's grasslands.
Appendix S5 Changes in topsoil organic content (SOC), total nitrogen (TN) and total phosphorus (TP) concentrations across China's grasslands over the sampling interval.

BIOSKETCH

Yuanhe Yang is interested in exploring spatial patterns and temporal dynamics of biogeochemical cycles in terrestrial ecosystems.

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