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Soil carbon, nitrogen, and phosphorus stoichiometry of three dominant plant communities distributed along a small-scale elevation gradient in the East Dongting Lake

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ABSTRACT

Soil carbon (C), nitrogen (N), and phosphorus (P) stoichiometry greatly affects plant community succession and structure. However, few studies have examined the soil stoichiometric changes in different vegetation communities of freshwater wetland ecosystems along an elevation gradient distribution. In the present study, soil nutrient concentrations (C, N, and P), soil stoichiometry (C:N, C:P, and N:P ratios), and other soil physicochemical characteristics were measured and analyzed in 62 soil samples collected from three dominant plant communities (*Carex brevicuspis, Artemisia selengensis*, and *Miscanthus sacchariflorus*) in the East Dongting Lake wetlands. The concentration ranges of soil organic carbon (SOC), total soil nitrogen (TN), and total soil phosphorus (TP) were 9.42–45.97 g/kg, 1.09–5.50 g/kg, and 0.60–1.70 g/kg, respectively. SOC and TN concentrations were the highest in soil from the *C. brevicuspis* community (27.48 g/kg and 2.78 g/kg, respectively) and the lowest in soil from the *A. selengensis* community (17.97 g/kg and 1.71 g/kg, respectively). However, the highest and lowest TP concentrations were detected in soil from the *A. selengensis* (1.03 g/kg) and *M. sacchariflorus* (0.89 g/kg) communities, respectively, and the C:N ratios were the highest and lowest in soil from the *M. sacchariflorus* (12.72) and *A. selengensis* (12.01) communities, respectively. C:P and N:P ratios were the highest in soil from the *C. brevicuspis* community (72.77 and 6.46, respectively) and the lowest in soil from the *A. selengensis* community (45.52 and 3.76, respectively). Correlation analyses confirmed that SOC concentrations were positively correlated with TN and TP, and C:N and N:P ratios were positively correlated with C:P. These data indicated that soil C, N, and P stoichiometry differed significantly among different plant communities and that these differences might be accounted for by variations in the hydrological conditions of the three communities.

1. Introduction

Ecological stoichiometry integrates basic biological, chemical, and physical principles and examines energy and chemical (e.g., carbon, C; nitrogen, N; phosphorus, P) balances in biological systems. Knowledge of ecological stoichiometry can reveal control mechanisms in ecological systems and variation laws associated with element interactions and restrictions (Sterner and Elser, 2002; Michaels, 2003; Jeyasingh and Weider, 2007). C, N, and P are the fundamental elements associated with the chemical composition of living organisms (Michaels, 2003). For instance, soil organic carbon (SOC) directly affects the production capacity of ecological systems, and acts as an indicator of ecosystem responses to the environment (Xiao, 1999; Chapin, 2003). Soil N is the main source of plant N, which affects photosynthesis and plant processes associated with primary production (Mooney et al., 1987; Sun and Liu, 2008). In ecological systems, the composition of C, N, and P determines the main ecological system processes, including energy flow and material circulation (Cheng et al., 2010). The ratio of C, N, and P in soil directly reflects soil fertility conditions, indirectly indicates the nutritional status of plants (Wang and Yu, 2008), and in turn affects productivity and species composition of plant communities (Wassen et al., 2005; Mao et al., 2016).

Soil stoichiometry can vary due to differences in vegetation characteristics (Vinton and Burke, 1995; Sardans and Penuelas, 2012; Lawrence and Zedler, 2013). The concentration and distribution of nutrients can differ among diverse plant communities due to the different amounts of litter returned to the soil (Liu et al., 2012). In addition, dead roots can provide a rich source of soil nutrients (Chen et al., 2015b). In wetland ecosystems, various aspects of the hydrological regime, such as water level, submergence time and frequency, are primary factors that influence wetland structure and function and vegetation distribution. They also affect other ecological processes such as changing of soil C, N, and P concentrations and stoichiometry by

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altering the biogeochemical equilibrium (Ahmad et al., 1992; Bai et al., 2016; Bedford, 1999; Mitsch and Gosselink, 2007; Recha et al., 2013; Zhao et al., 2015a). For example, Xu et al. (2013) reported that multiple drying and wetting cycles promote the decomposition of SOC, while Wang et al. (2016) found that SOC and total N concentrations in wetlands are affected by flow-sediment regulation. Moreover, elevation changes in some floodplains and river-connected lakes affect the submergence duration and depth of wetlands. Although many studies have investigated the effects of elevation on plant distribution and growth (Chen et al., 2015a, 2015b), only few have examined the relationship of elevation and soil stoichiometry in wetlands.

Dongting Lake is the second largest freshwater lake in China, and it receives inflow from four rivers, namely, the Xiang, Zi, Yuan, and Li, in Hunan Province; it has four channels named Songzikou, Taipingkou, Ouchikou, and Tiaoxiankou that are connected to the Yangtze River. The wetlands are characterized by large seasonal fluctuations in water levels: they are usually completely flooded from May to October, and are drought-susceptible from November to April. Plants display zonation distribution patterns in this lake from the water's edge to the uplands (low elevation: *Phalaris arundinacea*; middle elevation: *Carex brevicuspis* and *Polygonum hydropiper*; high elevation *Miscanthus sacchariflorus*) (Xie and Chen, 2008; Chen et al., 2014). Therefore, the freshwater wetlands are an ideal area for the study of soil stoichiometry in varied plant communities along an elevation gradient.

In this study, we focused on soil C, N, and P stoichiometry in the three dominant plant communities along a small-scale elevation gradient in East Dongting Lake. The aims of our study were: 1) to investigate soil C, N, and P stoichiometry characteristics in the three dominant plant communities at different elevations; and 2) to investigate the relationships between soil stoichiometry and other environmental factors (e.g., elevation gradient, submergence time, soil bulk density, and soil moisture).

2. Methods and materials

2.1. Study area

The study area (28°59′8″- 29°30′56″ N, 112°41′51″ - 113°8′35″ E) was located in East Dongting Lake, which is in the northeast of Hunan Province, China (Fig. 1). This area has a continental subtropical monsoon humid climate, and the average annual rainfall is 1200–1300 mm; more than 60% of the rain falls between April and August (Chen et al., 2014). The annual fluctuation in water levels is approximately 12–14 m, with the maxima and minima occurring in August and January–February, respectively (Hu et al., 2015). The mean annual temperature is 16.4–17.0 °C, and the coldest and hottest months are January (3.9–4.5 °C) and July (28.6–29.1 °C), respectively. The annual frost-free period is about 285 days.

2.2. Sampling method

Soil samples were collected from East Dongting Lake in December 2013 (Fig. 1). Before sampling, the sites were selected on a map of East Dongting Lake using a systematic grid sampling method (Carter and Gregorich, 2006). The initial point on the grid was randomly selected using a 2-km interval, but a small portion of the sampling sites was omitted because of the presence of human activities. In addition, the amount of sampling varied among the three communities due to their different distribution areas. Sixty-two sampling sites were established during our field investigation; 21 sites were selected for *C. brevicuspis,* 11 for *A. selengensis*, and 30 for *M. sacchariflorus*. The co-

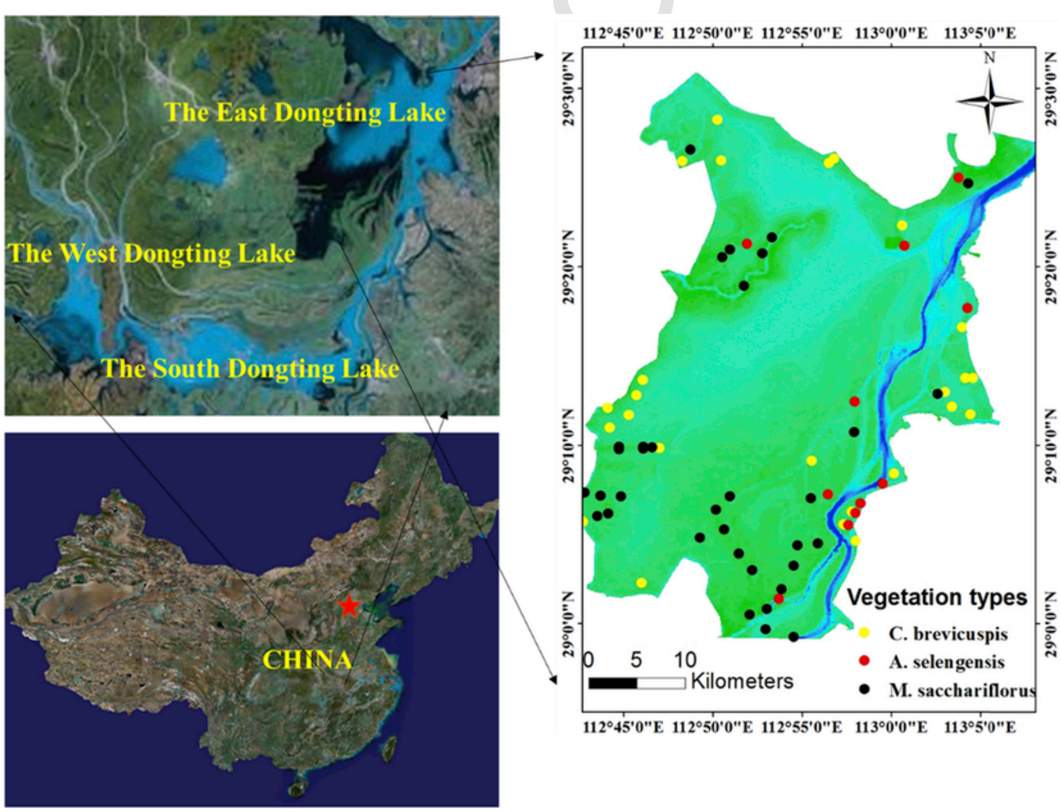

Fig. 1. Map of soil sample collection locations in the East Dongting Lake wetlands.

ordinates of the sampling sites were determined using a hand-held global positioning system (GPS). About 500 g soil was sampled from the 0–20 cm layer using a stainless steel drill; the samples were preserved in polyethylene sealed bag at each subplot. In addition, surface soil samples were collected using a stainless steel cutting ring with an inside diameter of 5 cm for soil bulk density (SBD) determinations. The cutting ring was sealed immediately and weighed in the laboratory to calculate the bulk density according soil moisture and volume of the cutting ring (Lu, 1999). All soil samples were stored at 4 °C and transported to the Key Laboratory of Agro-ecological Processes in the Subtropical Region Chinese Academy of Sciences within 24 h. The physicochemical characteristics of the soil samples were analyzed. Soil samples were air-dried at room temperature and sieved through a nylon mesh to remove coarse debris and stones (2 mm for alkali-hydrolyzable nitrogen (AN), available phosphorus (AP) and pH; 0.25 mm for SOC, total N concentration and total C concentration; 0.15 mm for total P concentration).

2.3. Chemical analysis

Soil samples associated with different vegetation types were collected, and soil stoichiometry and other physicochemical characteristics were analyzed. Each soil sample was oven dried at 105 °C for 24 h to determine soil moisture and soil bulk density. Samples were equilibrated with deionized water (soil:water = 1:2) for one week, and the pH values of the equilibrated samples were determined with a pH meter (pH/ion meter 225, Iwaki Glass, Japan). SOC concentrations were measured using the $K_2Cr_2O_7$-H_2SO_4 oxidation method (Zhang et al., 2009), and the total soil nitrogen (TN) and total soil carbon (TC) concentrations were measured using a Carlo Erba CNS Analyzer (Carlo Erba, Milan, Italy). Total soil phosphorus (TP) concentrations were determined using perchloric acid digestion and a UV-2450 spectrophotometer (Shimadzu Scientific Instruments, Japan; Duval, 2012). Alkali-hydrolyzable nitrogen (AN) was measured via titration with a diluted solution of H_2SO_4 after the samples were extracted with a mixture of $FeSO_4$ and NaOH. Samples were extracted with 0.5 M Na_2CO_3, and available phosphorus (AP) was measured using the molybdenum blue colorimetric method. Three replicates were performed for each soil sample.

2.4. Data source

The elevation of each plot was determined using a hand-held GPS and a digital elevation model (1:10000) of Dongting Lake that was produced in 1995 by the Changjiang Water Resources Commission (Ministry of Water Resources, China) with an accuracy of 0.1 m. The maximum submergence duration (days) was transformed into elevation data (m), using the water-level ranking method, from January to December of 2013 based on the daily water level at 8:00 a.m. at the Chenglingji Hydrological Gauging Station.

The SOC, TN, and TP concentrations (g/kg) were transformed to mol/kg. The C:N, C:P, and N:P ratios of each soil sample were then calculated as molar ratios (atomic ratios) using SOC:TN, SOC:TP, and TN:TP data.

2.5. Statistical analysis

The general linear model was used to detect significant differences in environmental factors (elevation, soil moisture, bulk density, submergence duration, pH), SOC, TN, TP, TC, AN, and AP concentrations, and C:N, C:P, and N:P ratios among the three plant communities. All data were checked for normality of distributions and homogeneity of variances before analysis, then multiple comparisons were performed using Tukey's test at the 0.05 significance level. Pearson correlation coefficients were used to detect the relationship between soil moisture and AN, AP, TC, SOC, TN, and TP concentrations as well as C:N, C:P, and N:P ratios. All statistical analyses were performed using SPSS 19.0 software (SPSS Inc., Chicago, IL, USA).

3. Results

3.1. Soil physicochemical properties in the three plant communities

The elevation and bulk density values were much higher in the *M. sacchariflorus* community than in the other two communities, but no significant differences were detected between the *C. brevicuspis* and *A. selengensis* communities. Soil moisture and submergence duration measurements were much lower in the *M. sacchariflorus* communities than in the other two communities (Table 1).

3.2. Soil C, N, and P stoichiometry in the three plant communities

AN and TC values were much higher in the *C. brevicuspis* community than in the other two communities, and there were no significant differences between the *A. selengensis* and *M. sacchariflorus* communities. The pH and AP measurements were not significantly different between the three communities (Table 2).

The concentrations of SOC (27.48 ± 2.35 g/kg) and TN (2.78 ± 0.25 g/kg) were significantly higher in the *C. brevicuspis* community than in the other two communities ($F = 4.656$, $p = 0.014$; $F = 7.981$, $p = 0.001$, respectively). The highest and lowest TP concentrations occurred in the *A. selengensis* (1.03 ± 0.06 g/kg) and the

Table 1
Soil elevation, submergence duration, moisture, bulk density, and pH in three plant communities.

	C. brevicuspis	*A. selengensis*	*M. sacchariflorus*
Elevation (m)	24.88 ± 0.22b	25.19 ± 0.4b	26.34 ± 0.25a
Submergence duration (Day)	162.48 ± 5.53a	153.73 ± 10.38a	126.73 ± 6.19b
Moisture (%)	45.93 ± 2.26a	41.04 ± 2.59ab	38.84 ± 0.86b
Bulk density (g/cm^3)	1.02 ± 0.04b	1.05 ± 0.06b	1.17 ± 0.02a
pH	6.74 ± 0.2a	6.86 ± 0.39a	7.37 ± 0.16a
Sample number	21	11	30

Note: the data are presented as mean ± SE. Different letters indicate significant differences based on Tukey tests ($p < 0.05$).

Table 2
C, N, and P characteristics in soils of three plant communities.

	C. brevicuspis	*A. selengensis*	*M. sacchariflorus*
AP (mg/kg)	11.17 ± 0.96a	11.33 ± 0.94a	9.23 ± 0.8a
AN (mg/kg)	269.34 ± 20.43a	169.8 ± 16.49b	192.85 ± 9.71b
TC (g/kg)	33.94 ± 3.71a	19.8 ± 1.78b	27.02 ± 1.38ab
SOC(g/kg)	27.48 ± 2.35a	17.97 ± 1.99b	23.28 ± 1.63ab
TN (g/kg)	2.78 ± 0.25a	1.71 ± 0.14b	2.11 ± 0.12b
TP (g/kg)	1 ± 0.05ab	1.03 ± 0.06a	0.89 ± 0.02b
C:N	12.12 ± 0.72a	12.01 ± 0.47a	12.72 ± 0.39a
C:P	72.77 ± 6.2a	45.52 ± 4.77b	67.28 ± 4.56a
N:P	6.46 ± 0.66a	3.76 ± 0.32b	5.23 ± 0.29a

Note: the data are presented as mean ± SE. Different letters indicate significant differences based on Tukey tests ($p < 0.05$). AN: alkali-hydrolyzable N; AP: available phosphorus; SOC: soil organic carbon; TC: total soil carbon; TP: total soil phosphorus.

M. sacchariflorus (0.89 ± 0.02 g/kg) communities, respectively, but with no significant differences among them ($F = 1.840$, $p = 0.167$) (Table 2).

The ratios of C:P (45.52 ± 4.77) and N:P (3.76 ± 0.32) were much lower in the *A. selengensis* community than in the other two communities (Fig. 2), and they showed significant differences ($F = 5.730$, $p = 0.005$; $F = 7.875$, $p = 0.001$, respectively), but the C:N ratio did not significant differences between the three communities ($F = 0.703$, $p = 0.499$) (Table 2).

3.3. Correlations among C, N, and P concentrations and stoichiometry

The SOC concentration was positively correlated with TN ($R^2 = 0.5$, $p < 0.01$), TP ($R^2 = 0.08$, $p < 0.05$), and N:P ($R^2 = 0.3$, $p < 0.01$) ratios. However, the TN concentration was not correlated with TP, but was positively correlated with the C:P ratio ($R^2 = 0.44$, $p < 0.01$). The C:P ratio was positively correlated with C:N ($R^2 = 0.17$, $p < 0.01$) and N:P ($R^2 = 0.49$, $p < 0.01$) ratios, and the C:N ratio was negatively correlated with the N:P ratio ($R^2 = 0.07$, $p < 0.05$) (Table 3).

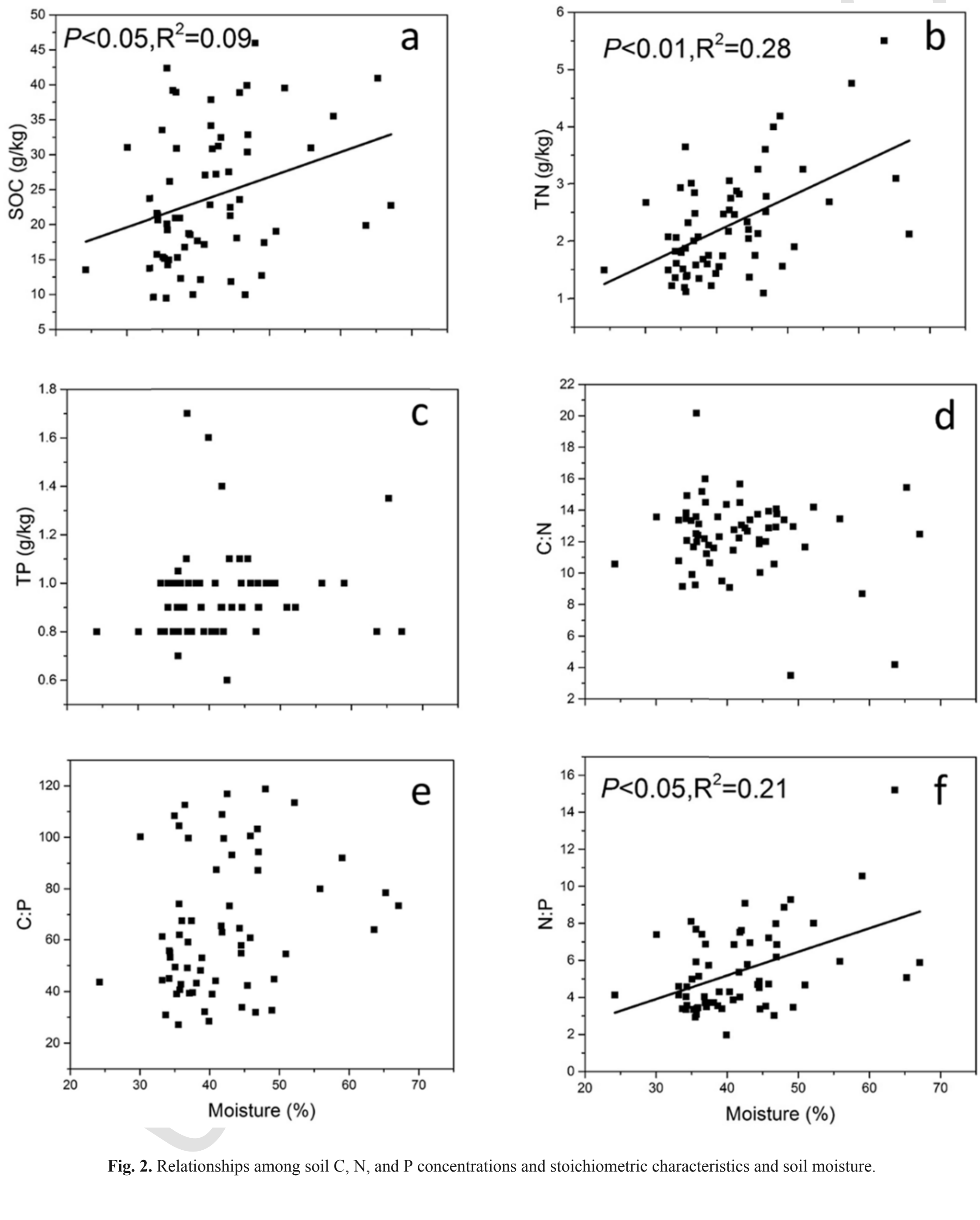

Fig. 2. Relationships among soil C, N, and P concentrations and stoichiometric characteristics and soil moisture.

Table 3
Correlation coefficients of soil properties and soil C, N, and P stoichiometry in East Dongting Lake.

	Elevation(m)	Submergence duration (day)	Moisture (%)	Bulk density (g/cm^3)	pH	AP (mg/kg)	AN (mg/kg)	TC (g/kg)	SOC (g/kg)	TN (g/kg)	TP (g/kg)	C:N	C:P	N:P
Elevation(m)	1													
Submergence duration (day)	-0.985**	1												
Moisture (%)	-0.177	0.174	1											
Bulk density (g/cm^3)	0.240	-0.211	-0.766**	1										
pH	0.048	-0.062	-0.195	0.309*	1									
AP (mg/kg)	-0.129	0.136	0.078	-0.281*	-0.229	1								
AN (mg/kg)	-0.145	0.130	0.351**	-0.211	-0.195	0.119	1							
TC (g/kg)	-0.046	0.019	0.503**	-0.297*	0.095	-0.067	0.723**	1						
SOC(g/kg)	-0.046	0.044	0.303*	-0.282*	-0.061	0.111	0.440**	0.627**	1					
TN (g/kg)	-0.047	0.028	0.530**	-0.363**	-0.060	0.012	0.754**	0.967**	0.708**	1				
TP (g/kg)	-0.179	0.177	0.118	-0.146	-0.054	0.263*	0.126	0.072	0.290*	0.118	1			
C:N	-0.101	0.130	-0.165	0.062	0.048	0.190	-0.227	-0.212	0.516**	-0.158	0.301*	1		
C:P	0.004	-0.007	0.236	-0.184	-0.069	-0.002	0.410**	0.592**	0.886**	0.660**	-0.152	0.416**	1	
N:P	-0.013	-0.005	0.457**	-0.257*	-0.064	-0.091	0.712**	0.898**	0.551**	0.911**	-0.262*	-0.263*	0.698**	1

Note: **Correlation is significant at the 0.01 level (2-tailed); * Correlation is significant at the 0.05 level (2-tailed).

Table 4
Comparison of C:N, C:P, and N:P in China and in Chinese wetlands.

	C:N	C:P	N:P	Reference
China (Overall)	11.9 ± 0.4	61 ± 0.9	5.2 ± 0.1	(Tian et al., 2010)
Costal wetland (Sanjiang Plain, China)	12.80 ± 1.45	161.96 ± 38.21	12.75 ± 3.08	(Zhang et al., 2012)
Estuarine wetland (Shuangtaizi, China)	16.15 ± 6.08	90.66 ± 107.31	5.07 ± 4.51	(Zhang et al., 2013)
River-connected lakes wetland (Dongting Lake, China)	12.39 ± 0.32	65.27 ± 3.35	5.39 ± 0.29	In this paper

Note: the data are presented as mean ± SE.

3.4. Relationships between soil C:N:P stoichiometry and environmental factors

Table 3 indicated that soil moisture was the main factor affecting soil C, N, and P characteristics in this lake (Table 3). AN, TC, TN, and SOC concentrations and the N:P ratio increased significantly with increasing soil moisture (Fig. 2).

4. Discussion

Both the soil C:N (12.39) and C:P (65.27) ratios in the study area were higher than the mean values reported previously for Chinese soil (11.9 and 61), while the ratios were lower than the mean values found in wetlands soils of Sanjiang Plain (12.8 and 161.96) and Shuangtaizi (16.15 and 90.66). The N:P (5.39) ratio was similar to the mean value of Chinese soil (5.2) (Table 4) (Tian et al., 2010; Zhang et al., 2012, 2013). The different soil stoichiometries in different regions might be result from variations in soil properties, vegetation types, microorganism activities, and sediment deposition (Jackson et al., 1997; Pan et al., 2012; Lawrence and Zedler, 2013). Since Dongting Lake is a river-connected lake wetland, the soil stoichiometry is influenced by complex factors such as hydrological regime, flow-sediment regulation, and soil parent material (Zhao et al., 2015b). In addition, the ratios of C:N, C:P, and N:P in the study area had high variation coefficients of 20.02%, 40.43% and 42.65%, respectively. The high variation coefficients might be due to different vegetation composition, soil parent materials, and hydrothermal conditions (Zhang et al., 2012).

The concentrations of SOC, TN, and TP were significantly higher in the *C. brevicuspis* community than in the *M. sacchariflorus* community (Table 2). Moreover, the SOC and TN concentrations in the *A. selengensis* community were the lowest among the three communities, while the TP concentration was the highest. Soil nutrients are influenced by soil parent materials (Zeng and Chen, 2005), sedimentation (Pan et al., 2016), the amount of aboveground plant residues returned to the soil (Li, 2010), and the decay of large numbers of dead roots (Chen et al., 2015b). The *C. brevicuspis* community at the lowest elevation exhibited the longest inundation time and highest sediment deposition. Thus, high concentrations of soil C, N, and P were found in the *C. brevicuspis* community. In Dongting Lake, *M. sacchariflorus* is important for paper making, and its harvest might reduce the organic matter that is contributed to the soil, because of relatively low amounts of litter and decomposing matter (Liu, 2013). Therefore, the soil C, N, and P concentrations in the *M. sacchariflorus* community were lower than those observed in the *C. brevicuspis* community.

The ratios of C:P and N:P were significantly higher in the *C. brevicuspis* community than in the other two communities because of its high C and N concentrations and low P concentration. The downward leaching of P in the *C. brevicuspis* community was disseminated into deeper soil or underground water via the higher soil moisture concentrations under the prevailing hydrological conditions, because the *C. brevicuspis* community existed at the lowest elevation of the three communities, resulting in high surface soil ratios of C:P and N:P (Zhang et al., 2012). In contrast, lower surface soil moisture reduces soil P leaching and results in relatively lower C:P and N:P ratios, and this was observed in the *M. sacchariflorus* community, which had the highest elevation of the three communities (Liu et al., 2006). Zhang et al. (2013) found a similar result in estuarine wetlands.

Our results indicated that soil moisture was positively correlated with AN, TC, SOC, and TN concentrations and the ratio of N:P. These results show that soil moisture plays an important role in determining soil stoichiometry. This conclusion is consistent with previous studies (Bonetto et al., 1994; Wang et al., 2016). Under conditions of high soil moisture content, anoxic decomposition of organic matter is inhibited, resulting in organic carbon accumulation (Mitsch and Gosselink, 2007). In the *C. brevicuspis* community, the higher content of soil moisture was unfavorable for decomposition of litter, which led to a higher concentration of SOC. In addition, SOC and TN were significantly correlated as the main N source was derived from litter, animal residue, biological nitrogen fixation, and organic matter (Bai et al., 2001). Similar effects have been described in previous studies (Lu et al., 2007; Wang et al., 2016). Moreover, soil moisture also affects soil N in wetlands as a high soil moisture concentration limits soil microbial activities and is unfavorable to the mineralization and decomposition of organic nitrogen (Bai et al., 2006; Zhao et al., 2015b). Thus, high soil moisture might lead to the relatively high TN concentrations observed in the *C. brevicuspis* community as compared to the other communities. The factors that influence the patterns of soil P concentration are usually quite complex, and the hydrological regime and soil parent material both play important roles in determining soil P concentration (Smeck, 1985; Cross and Schlesinger, 1995; Zhang et al., 2005; Gao et al., 2016).

In conclusion, our results confirmed that soil C, N, and P stoichiometric characteristics changed significantly among different plant communities distributed along an elevation gradient in the East Dongting Lake wetland. The results of our study also showed that plant communities and hydrological regime were the key factors affecting soil C, N, and P stoichiometry in this wetland. However, these results are based on a preliminary field investigation, and further controlled incubation experiments and field experiments are needed to confirm the mechanisms by which the hydrological regime influences soil C, N, and P stoichiometry.

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