

Original Research

Assessing Nutrient Elements as Indicators for Soil Active Organic Carbon in Topsoil of Karst Areas

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Abstract

In karst areas, in order to ascertain the relationship between soil active organic carbon fractions and soil nutrient elements, topsoil samples (the sampling points were set at 80-m intervals) were collected for the analysis of soil physicochemical properties. In the sampling area, land use was divided into upland, paddy, and shrub land. The results showed that dissolved organic C (DOC) and microbial biomass C (MBC) contents in shrub land soil were higher than those values in upland and paddy soils ($p < 0.05$). The total nitrogen (TN) content in paddy was lower than in upland and shrub land ($p < 0.05$). The mean value of total phosphorus (TP) in upland and shrub land was approximately 1.5 times higher than that in paddy. Available nitrogen (AN) in shrub land soil was higher than in upland, whereas Olsen-P was lowest in shrub land in all soils ($p < 0.05$). The C:P and N:P ratios in upland and paddy were lower than in shrub land ($p < 0.05$). At plot scale, random forest analysis revealed that pH and soil organic carbon (SOC) were the most important variables determining DOC content in croplands (upland and paddy) and shrub land, respectively. The accumulation of SOC accelerates the growth of microbial biomass in upland. In shrub land, increases in SOC and total nitrogen were in favor of microbial growth. At the small-watershed scale, C:P and N:P ratios had a significant and positive effect on the content of DOC and MBC, respectively. The results implied that high C and N availability, especially in combination with low P availability, is helpful for increasing soil microbial biomass.

Keywords: ecological stoichiometry, soil nutrient, active organic carbon fractions, land use, karst area

Introduction

Soil nitrogen (N) and phosphorus (P) cycling are tightly coupled to carbon (C) cycling in a terrestrial ecosystem [1]. Dissolved organic carbon (DOC) and soil microbial biomass carbon (MBC) are the small part of the soil organic carbon (SOC) pool, whereas they play an important role in the soil carbon biogeochemical cycle [2]. Previous studies have suggested that the amount of soil active organic carbon depends on the relative ratio of C:N:P [3-4]. Nosrati et al. [5] found that the amount of DOC increased with increasing soil organic carbon content in a semi-arid drainage basin. At high C:N ratios, mineralization rate of the available DOC is limited and the DOC concentration increases [6]. In short, C:N ratio was a useful indicator of the content of DOC. MBC respond rapidly to land management practices and can be used as a sensitive indicator of changes in soil organic matter [7]. The soil microbial biomass acts as both sink and source of nutrients. The availability and limitation of essential nutrients can thus provide feedback on microbial biomass and SOC dynamics. On a global scale, soil microbial carbon pools were somewhat correlated with soil C:P and N:P ratios, although a weak general relationship between soil P and MBC could be described for pastures and forest soils [8]. Schimel and Weintraub [9] consider that N availability

directly constrains the size of microbial C pools across global soils. Sinsabaugh et al. [10] deem that microbial allocation to P uptake is considerably greater than that of C and N when compared to requirements for the growth of biomass. In general, the effects of availability of nutrients on biomass growth were quite different.

Land use types affect the amount of soil nutrients and SOC sequestration [11-12]. In karst areas, numerous studies were mainly focused on ecological restoration to improve fragile ecosystems, soil fertility, and SOC sequestration. Previous studies have indicated that soil physicochemical properties have been improved after restoration [13]. However, the relative dynamics and dependencies among C, N, and P in karst areas have not been well investigated. Notably, studies on the relationships between soil active organic carbon fractions and soil nutrients at different scales are lacking (such as plot scale and the watershed scale) [14-15]. Better knowledge on the contribution of soil C, N, and P ecological stoichiometries on soil active organic carbon fractions can provide the necessary information to develop effective measures for the rational utilization and management of land so as to enhance the potential capacity of SOC [16]. We proposed that the contribution of nutrient elements on active organic carbon fractions were discrepant with different land management, human disturbance, and scales. Thus, in this study,

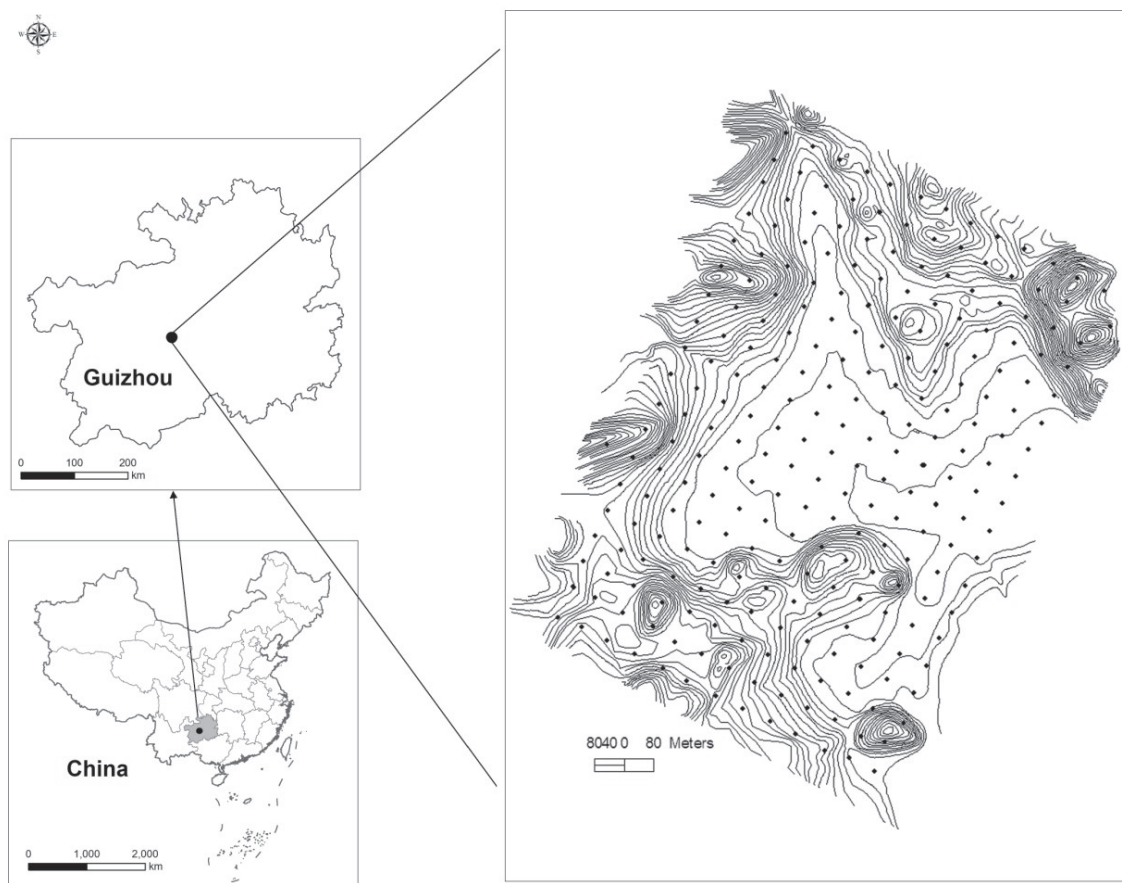


Fig. 1. Basic features of the sample plots.

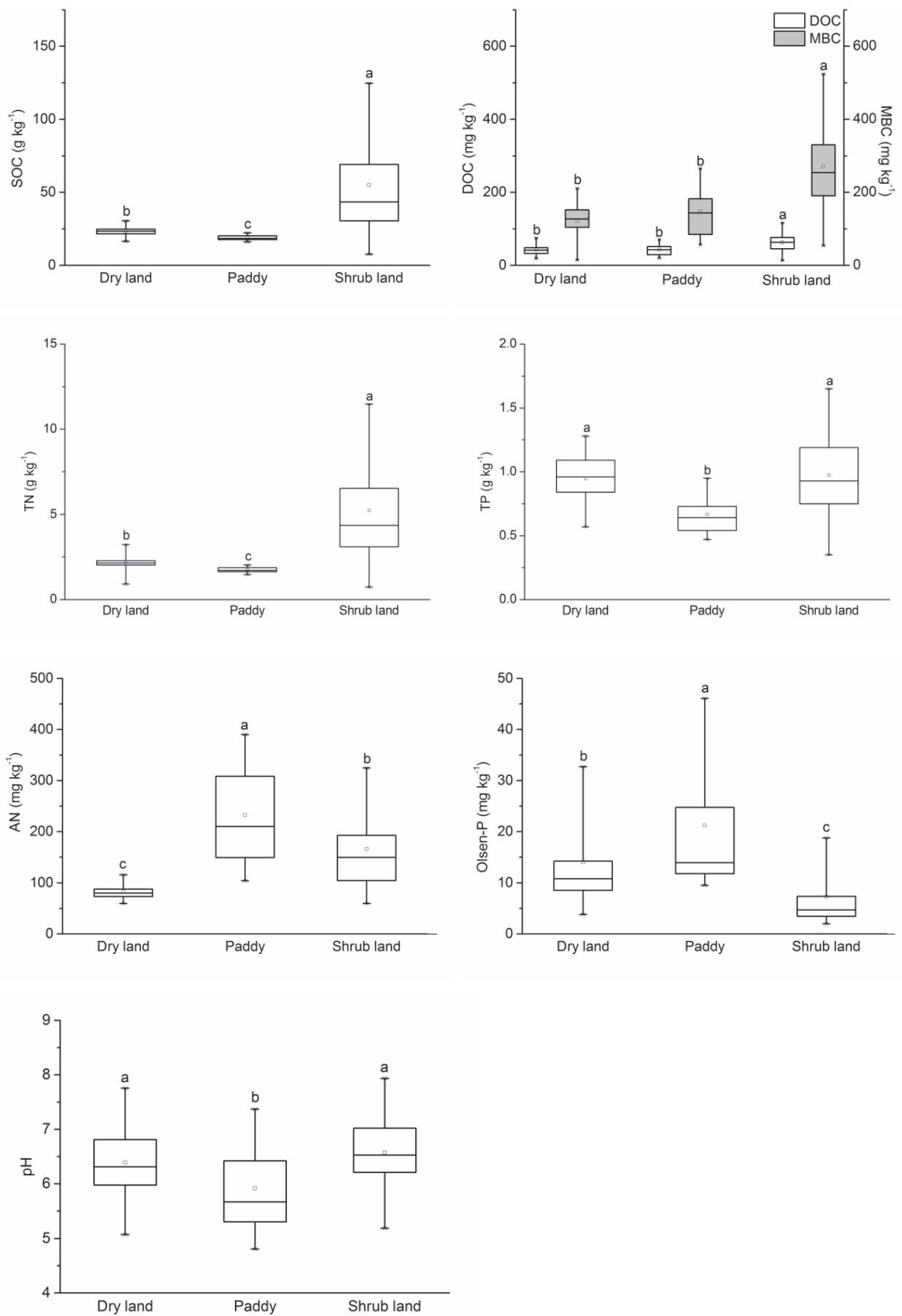


Fig. 2. Change of SOC (g kg^{-1}), DOC (mg kg^{-1}), MBC (mg kg^{-1}), TN (g kg^{-1}), TP (g kg^{-1}), AN (mg kg^{-1}), Olsen-P (mg kg^{-1}), and pH in topsoil under different land use types ($p < 0.05$; in dry land $n = 97$; in paddy $n = 19$; in shrub land $n = 132$).

we aimed to: 1) assess the changes of active organic carbon fractions and nutrient elements, 2) distinguish the effects of nutrient elements on active organic carbon fractions under different land uses, and 3) evaluate the relationships between soil C:N:P stoichiometry and active organic carbon fractions at watershed scale.

Material and Methods

Study Site

The experimental site is located in the small watershed of Wang (106°20'5"–106°21'8"E, 26°31'45"–26°30'27"N) situated in Boluo village, Qingzhen City, Guizhou Province, China. The area is a typical karst plateau with a peak cluster-depression ratio of approximately 1.65:1. The study region is characterized by a subtropical humid monsoon climate with an average annual precipitation of 1200 mm, mainly concentrated in 5-9 months. The altitude is 1271-1451 m, and the slope gradient ranges from 20° to 60°. In the depression, land is mainly used as agricultural land, whereas the slopes are covered with grass and shrub. The main species include *Zanthoxylum esquirolii*, *Rosa cymosa*, *Pyracantha fortuneana*, and *Broussonetia papyrifera*; *Rosa cymosa*, *Pyracantha fortuneana*, *Zanthoxylum armatum*, and *Sinopteris grevilleoides*; and *Themeda japonica*, *Carex lanceolata*, *Campylotropis macrocarpa*, and *Pyracantha fortuneana*.

Field Investigation, Soil Sampling, and Laboratory Analysis

As shown in Fig. 1, the sampling points were set at 80-m intervals and located by GPS (Trimble GeoXT3000). Environmental factors of every sampling point, including parent material, vegetation types and coverage, altitude, slope position, slope steepness and aspect, soil depth, and bare-rock ratio, were recorded. The bare-rock ratio was estimated by using frames (1 × 1 m) containing grids (10 × 10 cm). Ten measurements of soil depth around the sampling point were taken by pressing an iron stick marked at 10 cm intervals. At each sampling site, 5-8 soil cores from the surface soil (0-15 cm) were collected and bulked. A total of 248 representative soil samples were collected (with 97 samples in upland, 19 samples in paddy, and 132 samples in shrub land).

All the soil samples were sieved through a 2-mm mesh and divided into two parts. One part was air-dried for analysis of physicochemical properties. The other part was immediately stored at 4°C and subsequently incubated at 25°C for 2 weeks at 45% water-holding capacity before measuring the MBC. The SOC was assayed according to the Walkley-Black dichromate oxidation procedure [17]. Total nitrogen (TN) was determined according to the Kjeldahl method using a

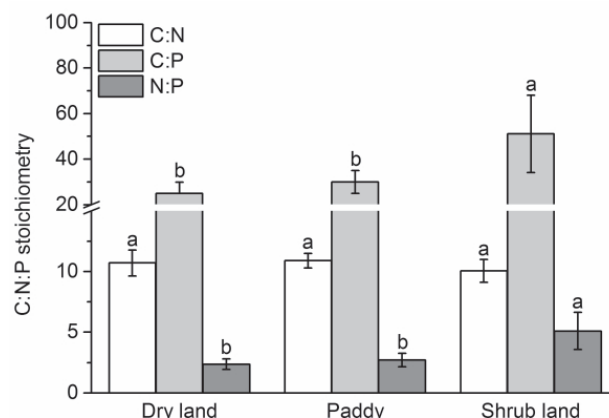


Fig. 3. Change of C:N, C:P, and N:P ratios under different land use types; values are means \pm standard deviations, $p < 0.05$ (in dry land $n = 97$, in paddy $n = 19$, in shrub land $n = 132$).

flow-injection auto-analyzer (FIA Star 5000 Analyzer; Foss Tecator, Höganäs, Sweden) [18]. Total phosphorus (TP) was measured using NaOH digestion and the molybdenum blue colorimetric method [19]. Available nitrogen (AN) was determined using the micro-Kjeldahl method [20]. Available phosphorus (Olsen-P) was extracted using the sodium bicarbonate Olsen method [21]. DOC in the soil was extracted in duplicate with 0.5 M K_2SO_4 , and the filtrate was analyzed using a TOC analyzer (TOC-VWP; Shimadzu Corp., Tokyo, Japan) for DOC. MBC of the soil was determined according to the fumigation-extraction method [22] using a TOC analyzer (TOC-VWP, Shimadzu Corporation, Japan).

Statistical Analysis

The means and standard errors of the data were calculated in Excel 2010 (Microsoft Corp., Redmond, WA, USA). Statistical analyses were performed using SPSS 11.5 (IBM, Chicago, IL, USA). Differences between C:N, C:P, and N:P ratios were evaluated using one-way analysis of variance (ANOVA), followed by the Bonferroni test. Pearson correlation coefficients were calculated to quantify the correlation between soil active organic carbon fractionS and nutrients. The classification of random forests analysis was conducted to assess the most important drivers of nutrient elements and pH on the content of soil active organic carbon fractionS. These analyses were conducted using the RANDOMFOREST package [23] for the R statistical software, version 3.4.1.

Results

Changes in C, N, and P

The mean SOC content in shrub land was 2-3 times higher than that in upland and paddy soils ($p < 0.05$).

Similarly, the DOC and MBC in shrub land were higher than in the other two land use types ($p < 0.05$; Fig. 2). The TN content in paddy was lower than in upland and shrub land ($p < 0.05$). The mean value of TP in upland and shrub land was approximately 1.5 times higher than that in paddy. AN and Olsen-P in paddy were higher than in upland and shrub land. AN in shrub land was higher than in upland, whereas Olsen-P was lower in shrub land than in upland and paddy.

C, N, and P Stoichiometry

As shown in Fig. 3, the C:N ratio of soil was similar under all land use types (from 10 to 11), and all C:P ratios were below 55. The C:P ratio in shrub land soil was approximately 1.5-2 times higher than that in upland and paddy ($p < 0.05$), but no difference was observed between upland and paddy. The N:P ratio was lower in the upland and paddy sites than in the shrub-land site, with mean values of 2.4, 2.7, and 5.1, respectively.

Contribution of Soil Nutrients and pH on Soil Active Organic Carbon Fractions

As shown in Fig. 4, random forest analysis revealed that pH was the most important variable affecting the amount of DOC in upland and paddy soils. In shrub land, SOC was the important factor determining the content of DOC, whereas SOC and TN affected the content of MBC. In the sample area, the C:P ratio affected the content of DOC, and N:P ratio affected the content of MBC (Fig. 5, $p < 0.05$).

Discussion

Change in Soil C, N, and P under Different Land Uses

Land use has been shown to have significant impacts on the cycling of soil C, N, and P, and on their

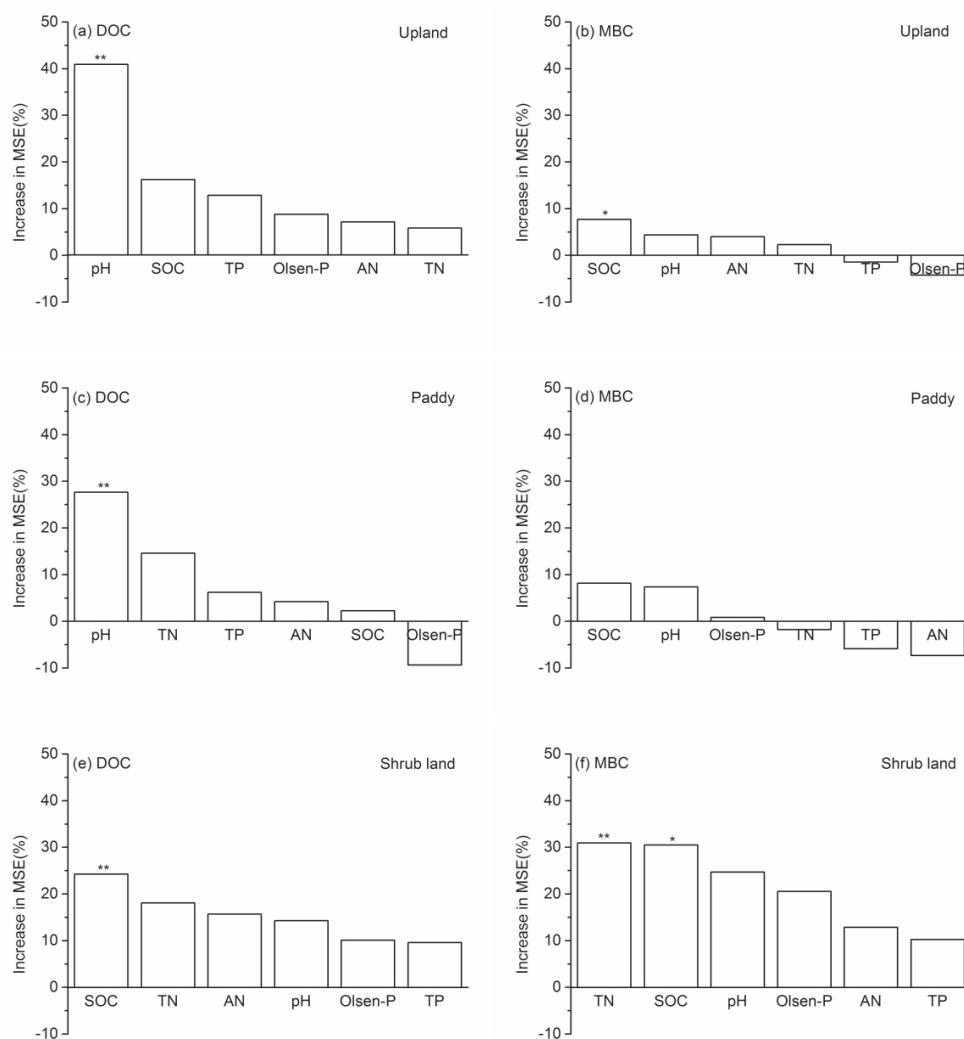


Fig. 4. Random forest mean predictor importance (percentage of increase of mean square error) of soil physical and chemical properties as drivers of the different active organic carbon fractions based on land use (significance levels are as follows: * $p < 0.05$ and ** $p < 0.01$; in dry land $n = 97$; in paddy $n = 19$; in shrub land $n = 132$).

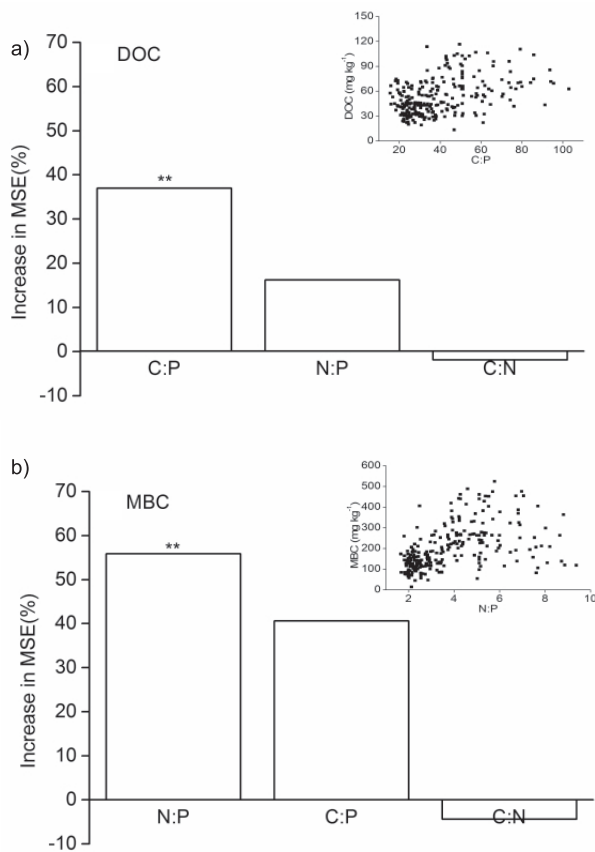


Fig. 5. Contributions of soil nutrient stoichiometries to the content of soil active organic carbon fractions at the small watershed scale (significance levels are as follows: * $p < 0.05$ and ** $p < 0.01$. $n = 248$).

associative storage capacities [24-25]. In the study area, the SOC content was significantly affected by land use. The mean SOC content in shrub land was 2-3 times higher than that in upland and paddy soils (Fig. 2). In shrub-land, there were relatively higher amounts of aboveground biomass and roots, resulting in relatively greater organic litter input and less soil erosion than in cropland (upland and paddy) [26]. In the cropland soils, SOC is less physically protected than in shrub land. Periodical tillage breaks up soil macro-aggregates and exposes previously protected organic matter within such aggregates [27], which increases the SOC decomposition rate and weakens organic carbon sequestration ability. Moreover, in a paddy field, the flooded-drainage circulation accelerates SOC mineralization rate because of the change of microbial community structure, biomass, and metabolic process [28-29]. The TN content in paddy was lower than in upland and shrub land (Fig. 2). Oelmann et al. [30] considered that soil management and plant species influenced N availability and accumulation. In shrub land, organic matter input (litter, dead roots, mycorrhizae, and exudates) leads to an increase of TN [31]. During the rice-growing season, the flooded-drainage cycle promoted the loss of nitrogen compared with upland soils. The amount of P in paddy

soil was shown to be lower than that in upland and shrub land soils (Fig. 2). The result may be due to the reason that flooded and dry-wet conditions improved the content of Olsen-P, but accelerate the loss of P. The continuous accumulation of soil organic C might have occluded P in its organic form, thereby decreasing soil P availability [32].

Soil Physical and Chemical Properties Influencing Soil Active Organic Carbon

In the present study, DOC was higher in shrub-land soil than in cropland soils, which may be attributable to the limited return of straw to upland and paddy soils (Fig. 2). Furthermore, frequent disturbance of cultivated soil was considered to accelerate the mineralization and leaching loss of DOC [33-34].

Random forest revealed that an important impact factor for the content of DOC was pH in cropland, whereas it was SOC in shrub land. Soil pH affects selective preservation or metabolic modification of specific components (e.g., lignin-cellulose, lipids) during decomposition and therefore also has a more direct impact on soil organic matter composition. Low soil pH has the potential to retard organic matter decomposition [35]. Compared with shrub land, pH in upland and paddy was low, which was one reason to explain the obvious contribution of pH on DOC in croplands to some extent. In shrub land, it was easy to understand that DOC is mainly due to the decomposition of litter and humus [6].

SOM aase of f M cause of input MBC, as an active fraction of SOC, has been suggested to be a sensitive indicator of changes in total SOM, and it readily responds to land use [36]. Abundant exogenous carbon from litter and root exudates in shrub land may supply a greater amount of organic carbon for microbial growth [2]. So in this study, MBC content in shrub-land soil was approximately two times larger than that in upland and paddy soils (Fig. 2). Usually, nutrients and soil physical conditions affect soil microbial biomass [24]. In the study area, the ratio of C:N was from 10 to 11 and the growth of microorganisms is limited by carbon availability (Fig. 3). In cropland, especially in upland soil, SOC was the most important variable determining the content of MBC. In shrub land, both SOC and TN regulated the content of MBC. In shrub land, the nutrient elements were mainly from vegetation and litter fall [37]. Soil microbial biomass pools appeared constrained primarily by soil nitrogen because of the competition for nitrogen between plants and microorganisms [8]. Therefore, TN plays an important role in regulating MBC content.

Relationship between of C, N, and P Stoichiometries and Soil Active Organic Carbon

It is well established that the cycling of soil N and P is tightly coupled to C cycling in ecosystems.

Table 1. Correlation coefficients (Spearman's ρ) between soil nutrients and soil active organic carbon fractions (* $p < 0.05$; ** $p < 0.01$).

	SOC	DOC	MBC	TN	AN	TP	Olsen-P
SOC	1	0.506**	0.391**	0.892**	0.117	0.555**	-0.131*
DOC		1	0.411**	0.492**	0.183**	0.301**	-0.213**
MBC			1	0.337**	0.177**	0.150*	-0.401**
TN				1	0.116	0.603**	-0.176**
AN					1	-0.061	-0.085
TP						1	0.021
Olsen-P							1

The effects of nutrient elements on active organic carbon fractions varied among different land use depending on the specific situations encountered [1, 38]. In the present study, the C:N ratio of soil was similar under all land use types (from 10 to 11), which illustrated that the biomass of soil microorganisms is N enriched. As shown (Fig. 3). At the watershed scale, random forest revealed that C:P and N:P ratios had a significant effect on the content of DOC and MBC, respectively. The positive relations between C:P ratio and DOC indicated that the accumulation of SOC accelerated the production of DOC [39]. Combined with the result of positive relations between MBC and AN (Table 1), the positive relationship between N:P ratio and MBC implies that improvement on the content of TN, especially nitrogen availability, was helpful for the growth of soil microbial biomass [4,8]. From Table 1, Olsen-P was negatively correlated with MBC, which means the growth of soil microbial biomass did not seem to be impaired by low P availability. Hu et al. [40] considered that microbial growth due to organic input is mainly dependent on the increased availability of C in the soil. In the study area, MBC was positively correlated with DOC (Table 1). In general, we deduced that, in karst area, high C and N availability (especially in combination with low P availability) is helpful to increase the soil microbial biomass. In short, SOC accumulation accelerates the increase of DOC content. Improvement of carbon and nitrogen availability was in favor of the growth of microbes.

Conclusions

In the study area it was possible that contributions of physical and chemical properties to the content of soil active organic carbon fractions was discrepant under different scales. pH and SOC were the most important variables determining the content of DOC in croplands (upland and paddy) and shrub land, respectively. SOC accumulation accelerates the growth of microbial biomass in upland and shrub land. In watershed scale, the accumulation of SOC promoted the increase of DOC. Although the content of Olsen-P in the shrub land

appeared to be lower than that in croplands, the growth of soil microbial biomass did not seem to be impaired by P availability.

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Conflict of Interest

The authors declare no conflict of interest.

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