Investigation on labile organic carbons in soils developed from limestone and sandstone in karst areas of southwest China

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Abstract. Although labile soil organic carbons (LSOC) represent only a small part of soil organic carbon (SOC), they are an important source of energy for the belowground portion of the ecosystem and play a crucial role regarding changes in the quality of soil organic matter (SOM). This study measured and compared the content of LSOC and SOC in limestone soil (developed from limestone) and sandstone soil (developed from sandstone) in the karstic area of southwest China to better understand the impacts of different soil types on LSOC (water-extractable and readily oxidizable organic carbon) and evaluate the existence of a possible relationship between LSOC and SOC in the soil. The results demonstrated that the amounts of SOC, soil readily oxidizable carbon (SROC), and soil water-extractable organic carbon (WEOC) (hot-water extractable organic carbon (hot-WEOC) and cold-water extractable organic carbon (cold-WEOC)) were the highest in the surface soil and decreased with increasing depth in both types of soil profiles. In addition, the content of SOC and LSOC were significantly higher in the limestone soil profiles than those in the sandstone soil profiles. This may be as a result of the occurrence of higher pH values and higher content of calcium, magnesium, and clay minerals in the limestone soil than in the sandstone soil. The proportions of WEOC in total SOC extracted at 25°C and 70°C in the sandstone soil profiles displayed higher average values of 0.040% and 0.527%, respectively, whereas the limestone soil profiles displayed lower average values of 0.015% and 0.255% for the WEOC extracted at 25°C and 70°C, respectively. Regarding the SROC content, both soil profiles showed higher values than WEOC content, ranging from 39.4%–55.2%. The results also showed that for the two types of soils significant correlations were found between samples of SROC, WEOC, and SOC (P < 0.01). Among the LSOC, the hot-WEOC showed clear differences between different soil types, and significant relationships were observed for not only SOC but also SROC. The correlation coefficients between hot-WEOC and SOC or SROC were higher than those between cold-WEOC and SOC or SROC. This study, therefore, suggests that hot-WEOC could be a useful indicator of changes in the quality of SOM in different soil types.

1. Introduction
Soil organic matter (SOM) is the most important factor in soil quality and fertility [1], therefore maintaining the quality and quantity of SOM is crucial in ensuring land sustainability and the restoration of degraded soils, especially in forest areas [2], [3]. However, SOM losses or gains in a
short or medium time scale are difficult to measure directly because of the occurrence of large amounts of organic matter in soils and their spatial variability [4]. In contrast, among the different forms of soil organic carbon (SOC), labile SOC (LSOC) with comparatively rapid turnover of SOM, such as microbial biomass carbon (MBC), soil readily oxidizable carbon (SROC), and soil water-extractable organic carbon (WEOC) can respond rapidly to changes in land management practices [5]. Therefore, such components have been suggested as sensitive indicators of the changes in SOM status [2]. In addition, LSOC account for only a small proportion of the SOC in the soil [6]. Nevertheless, they are key controlling factors of the soil biological activity [7], pollutant transport [8], mineral weathering [9], and soil formation [10].

SROC, which, mainly by virtue of its chemical oxidization of KMnO₄ isolates the readily oxidizable and unstable carbon in the soil, plays an equivalent role of soil microbial enzymes, which oxidizes SOC [11]. It has been shown that some types of labile organic carbons, such as soil particulate organic carbon and SROC, are rather responsive to various types of land use and soil management practices and can be used to characterize the early changes of SOM [12].

Soil WEOC according to the different temperature extractable method mainly includes the cold-WEOC (25°C) and hot-WEOC (70°C) [2]. It is concluded that the hot-WEOC has also been used in the past for quantifying the labile SOM in arable and pastoral soils [2], [13]. The hot-WEOC fraction contains microbial biomass, soluble soil carbohydrates, and other simple compounds, which may account for the labile fraction of SOM [2]. Therefore, many studies have shown that hot-WEOC strongly correlates with MBC and SOC in arable or pastoral soils [2], [3]. However, the application of this method has not been adequately studied in soils from the karstic areas. Chen and Xu [14] also found that the hot-WEOC content showed a better correlation with SOC than with MBC. Therefore, it may be worthwhile to compare hot-WEOC to SOC, SROC, and cold-WEOC as well as their ratios to SOC, and finally assess whether hot-WEOC could be a useful indicator of the quality of SOM in karstic areas.

In recent decades, many studies have been conducted on SOC [15] but only a few studies have focused on the components of labile SOM in the karst areas of Guizhou, southwest China. Two main types of soils are developed respectively from carbonate rocks and sandstone in the karstic area of Guizhou Province, southwest China. In this study, based on expectations arising from previous studies regarding the quality of SOM, we hypothesized that hot-WEOC can serve as an indicator of SOM quality in soils of karstic areas. Therefore, the labile SOM (LSOM) components including hot-WEOC, cold-WEOC, and SROC were selected as a case to describe SOM quality. This study characterized the vertical distribution patterns of LSOC and SOC in the limestone and sandstone soil profiles, explored the factors controlling the variations of LSOC and SOC content in karstic soils, and finally it assessed whether hot-WEOC can serve as an indicator of SOM quality in soils of karstic areas.

2. Materials and methods

2.1. Site description
The investigated sites are located at Baiyi, Wudang district and Wangjiazhai, Qingzhen city, about 15 km and 30 km respectively from Guiyang City (26°28’ 19” N, 106°49’ 18” E), in the central region of Guizhou Province, China (Figure 1), with an altitude of approximately 1300 m above sea level. The central region of Guizhou Province has a subtropical, monsoonal, and humid climate, with a mean annual temperature of 14.8°C and a mean annual precipitation of 1100 mm. Five soil profiles were sampled: three soil profiles were sampled in Wangjiazhai, Qingzhen city, which includes two soil profiles developed from limestone (profiles QZ-I and QZ-II) and one profile developed from sandstone (QZ-III); two soil profiles were sampled in Baiyi, Wudang district, which includes one soil profile developed from limestone (BY-II) and the other developed from sandstone (BY-I). According to the FAO-UNESCO soil classification system, soils developed from limestone are termed limestone soils, classified as calcic Cambisol, while the soils developed from sandstone are termed yellow sandstone soils, classified as Ferralsol. Limestone soils can be further classified into black limestone
soil (profiles QZ-I and QZ-II) and yellow limestone soil (soil profile BY-II) according to their color (or SOM content). The main forest types are Masson pine forest, made up of broad-leaved mixed forest and shrub. Detailed features of each soil profile during sampling (May 2009) are summarized in Table 1.

Figure 1. Map showing the Guiyang Prefecture (including the Guiyang urban area and its surrounding areas) and the sampling locations.

Table 1. Basic features of soil profiles studied.

| Study area          | Profiles | Soil type                  | Geographical coordinates | Altitude (m) | Landform     | Dominant species                                      |
|---------------------|----------|----------------------------|--------------------------|--------------|--------------|-------------------------------------------------------|
| BaiYi of WuDang     | BY-I     | Yellow sandstone soil      | 26°47' 3'' N, 107°01' 16'' E | 1214         | Middle mountains | Pinus massoniana Lam, Quercus alba L, Pterdium aquilinum var. ariusculum, Quercus glanca Thunb |
|         | BY-II    | Yellow limestone soil      | 26°48' 40'' N, 107°00' 16'' E | 1327         | Low land     | Pterdium aquilinum var. Latiasculum, Castanea seguinii Dode, Quercus alba L, Pyrakantha fortuneana |
| WangJiaZhai of QingZhen | QZ-I   | Black limestone soil       | 26°31' 25'' N, 106°20' 16'' E | 1285         | Middle mountains | Caesalpinia vernalis Champ, Loniceria Japanica, Herba Artemisiae Annuals, Pterdium aquilinum var. latiasculum |
|         | QZ-II    | Black limestone soil       | 26°31' 41'' N, 106°20' 18'' E | 1291         | Middle mountains | Pachyrhizus erosus, Pterdium aquilinum var. latiasculum |
2.2. Soil sampling and laboratory analysis

Litter horizons were removed before soil sampling. The soil samples were collected at the soil surface (0–5 cm depth), followed by sampling from the walls of soil pits from 5 cm depth to a depth of approximately 100 cm in the lower horizon. Visible roots and organic residues were removed at the time of sampling. Each sample was divided into two parts. One was immediately sieved through a 2 mm mesh and then stored at 4°C until analysis for the cold-WEOC within 3 d. The other part of the soil was also sieved through a 2 mm mesh and subsequently air-dried for determination of hot-WEOC, pH, and soil texture; but for the determination of carbon (C) and nitrogen (N), samples were sieved using 0.154 mm (100 mesh).

Soil pH was measured using a pH electrode (Orion) in a soil to de-ionized water ratio of 1:2.5 (m/v). The SOC and nitrogen were determined following the methods by Xiao [16]. Soil samples containing about 1–2 mg of SOC were put in 15 mL centrifuge tubes, while tubes without soil served as control: 10 mL of 2 mol L\(^{-1}\) KCl solution was added to the tubes and shaken for 1 h at 25°C and afterward centrifuged at 3000 r min\(^{-1}\) for 25 mins until the supernatant was drained, and then 10 mL of 0.1 mol L\(^{-1}\) HCl solution was added to the tubes and shaken for 24 h at 25°C and afterward centrifuged at 3000 r min\(^{-1}\) for 25 mins, washed until it attained a neutral pH using distilled water, centrifuged, dried at 60°C, pulverized, and kept for C and N content analysis. The organic carbon and nitrogen content were analyzed by combustion using an elemental analyzer (PE2400 II, USA) with an analytical precision of 0.1%. Two indices, SROC and WEOC including cold- and hot-WEOC, were used in the measurement of LSOC.

The SROC was determined by KMnO\(_4\) oxidation [17]. Soil samples containing approximately 15 mg of SOC were added to 50 mL centrifuge tubes and tubes without soil served as a control; 25 mL of 333 mmol L\(^{-1}\) KMnO\(_4\) solution was added to the tubes and shaken for 1 h at 25°C and then centrifuged at 2000 r min\(^{-1}\) for 5 min; the suspensions were diluted 250 times using deionized water and read on a spectrophotometer at 565 nm. The amount of readily oxidizable carbon in the soil was calculated according to the change in KMnO\(_4\) concentrations.

Hot-WEOC was determined following the methods by Sparling [13]. In brief, 4 g air-dried soil was extracted using 20 mL ultra-pure water in a capped test-tube at 70°C for 18 h. The tubes were shaken by hand to re-suspend the soil at the end of the incubation and then filtered through a 0.45 μm glass fiber filter. Hot-WEOC concentration in filtrate was quantified using the total organic carbon analyzer (O.I. 1030W, USA).

Cold-WEOC was extracted using the method described by Liang [3]. In brief, 20 g of fresh soil was extracted using 50 mL ultra-pure water in a centrifuge tube by shaking the mixture for 30 min at 25°C on a reciprocal shaker, and then centrifuging at 6000 rpm for 15 mins the supernatant was filtered through a 0.45 μm glass fiber filter. Cold-WEOC concentration in filtrate was quantified using the total organic carbon analyzer (O.I. 1030W, USA).

3. Results

3.1. Soil pH values

The yellow sandstone soils had significantly lower pH values (from 4.0–4.8) than the limestone soils, the pH values of which varied from 4.8–7.0 (Figure 2). Among the limestone soils, the yellow limestone soils (soil profile BY-II) showed the least pH values. For all of the limestone soil profiles, the soil pH values increased with increasing depth, while the soil pH values of the sandstone soil profiles varied differently: the pH values remained almost constant along the soil profile QZ-III, while the pH values in the soil profile BY-I initially decreased to a depth of 30 cm and then increased from pH 4.0 to pH 4.7 near the bottom of the profile.
3.2. SOC content and C/N ratio

The variations of SOC content with depth are presented in Figure 3. The amounts of SOC decreased with depth and were significantly higher in the limestone soils than those in the yellow sandstone soils. The SOC was unevenly distributed throughout the soil profiles, with the largest SOC pools occurring in the top 20 cm of the soil horizon. The surface soil of the yellow sandstone soil profile (QZ-III) showed the least SOC content (20.6 g kg\(^{-1}\)), while the surface soil of the black limestone soil profile (QZ-I) showed the highest SOC content (69.8 g kg\(^{-1}\)). For the limestone soils, SOC content was higher than 4.0 g kg\(^{-1}\), with a maximum value of 69.8 g kg\(^{-1}\) observed at the surface, while for the yellow sandstone soil, SOC content fluctuated between 2.0 g kg\(^{-1}\) and 51.2 g kg\(^{-1}\).
As shown in Figure 4, the mass ratios of SOC to nitrogen (C/N) for the surface soils ranged from 6–22, with the highest value observed in the surface soil of the yellow sandstone soil profile (BY-I) and the least value observed in the surface soil of the black limestone soil profile (QZ-II). Except for the surface soil of profile BY-I, the surface soils of the other soil profiles displayed a range of C/N mass ratios varying from 6–15, and a range of the C/N mass ratios varying from 2–10 at a depth of 60 cm, showing a general decrease from the top to the depth of the profile. The results also showed that there was no clear difference in the C/N mass ratios between the limestone and sandstone soils.

3.3. LSOC content

The cold-WEOC content for the surface soils ranged between 0.0022–0.0132 g kg\(^{-1}\), with the highest content found in the surface soil of the yellow sandstone soil profiles (BY-I) and the least content found in the surface soil of the yellow limestone soil profiles (BY-II) (Figure 5a). For the limestone soils, the cold-WEOC content was greater than 0.0009 g kg\(^{-1}\), with a maximum value of 0.0055 g kg\(^{-1}\), while for the yellow sandstone soil, cold-WEOC content fluctuated between 0.0010–0.0132 g kg\(^{-1}\). Except for soil profile BY-I from the surface soils to a depth of 20 cm, the amount of cold-WEOC was significantly higher in the limestone soil than in the yellow sandstone soil. In addition, the cold-WEOC content showed a large fluctuation and varied significantly from the surface soils to a depth of 40 cm. From 40 cm depth downwards, the cold-WEOC content remained almost constant with increasing depth between the yellow sandstone and limestone soils.

The hot-WEOC content, like the SOC content, decreased in depth, was highest in the surface soils (Figure 5b) and was higher in the limestone soils than in the sandstone soils. Samples from the two black limestone soil profiles showed distinctively high hot-WEOC content, especially for the soils above the 40 cm depth. The hot-WEOC and cold-WEOC were unevenly distributed throughout the soil profiles, with the largest WEOC pools occurring in the top 20 cm of the soil horizon. The surface soils of profile QZ-III (yellow sandstone soil) showed the least hot-WEOC content (0.0597 g kg\(^{-1}\)), while the surface soil of profile BY-I (yellow sandstone soil) showed the highest hot-WEOC content (0.2633 g kg\(^{-1}\)). For the limestone soils, the hot-WEOC content was higher than 0.0107 g kg\(^{-1}\), with a maximum value of 0.2527 g kg\(^{-1}\) observed at the surface, while for the yellow sandstone soil, hot-WEOC content fluctuated between 0.0107–0.2633 g kg\(^{-1}\).

![Figure 5](image_url)  
**Figure 5.** Changes in cold-WEOC (a) and hot-WEOC (b) of the five soil profiles with depth.
The depth profiles of SROC content for the soil samples are shown in Figure 6. In all profiles, the SROC content for the surface soils ranged between 9.8–46.6 g kg\(^{-1}\), with the highest content found in the surface soil of the yellow sandstone soil profiles (BY-I) and the least content found in the surface soil of the yellow sandstone soil profiles (QZ-III). The SROC content, similar to the SOC content, was the highest in the surface soils and the least in the soils at a certain depth (Figure 6) and was higher in the limestone soil than in the sandstone soil. The soils from the two black limestone soil profiles showed distinctively high SROC content, especially for soils above the depth of 60 cm. The yellow limestone soil and two yellow sandstone soil profiles showed an undifferentiated down-depth varying style of SROC, first decreasing from the topsoil to a depth of approximately 40 cm and then displaying SROC content, which remained constant with increasing depth.

**Figure 6.** Depth profiles of SROC content in the soils studied

3.4. Correlation between LSOC and SOC

Table 2. Variations in average labile soil organic carbon (LSOC) and soil organic carbon (SOC), the average percentages of the LSOC in total SOC and the relationship between LSOC and SOC in soils.

| Profile | Soil type          | Cold-WEOC (g kg\(^{-1}\)) | Hot-WEOC (g kg\(^{-1}\)) | SROC (g kg\(^{-1}\)) | SOC (g kg\(^{-1}\)) | Cold-WEOC/SOC (%) | Hot-WEOC/SOC (%) | SROC/SOC (%) | Correlation between SOC and SROC | Correlation between SOC and hot-WEOC | Correlation between SROC and cold-WEOC | Correlation between SROC and hot-WEOC | Correlation between SROC and cold-WEOC |
|---------|--------------------|-----------------------------|---------------------------|----------------------|---------------------|-------------------|------------------|--------------|-----------------------------------|------------------------------------|----------------------------------------|----------------------------------------|----------------------------------------|
| QZ-I    | Black limestone soil | 0.0032                      | 0.0779                    | 16.4                 | 26.6                | 0.016             | 0.255            | 53.4         | 0.978**                           | 0.844**                            | 0.975**                                | 0.771**                                | 0.989**                                |
| QZ-II   | Black limestone soil | 0.0025                      | 0.0446                    | 9.3                  | 16.8                | 0.015             | 0.262            | 47.9         | 0.926**                           | 0.890**                            | 0.866**                                | 0.829**                                | 0.965**                                |
| QZ-III  | Yellow sandstone soil | 0.0015                     | 0.0260                    | 3.8                  | 9.2                 | 0.020             | 0.289            | 39.4         | 0.991**                           | 0.877**                            | 0.969**                                | 0.904**                                | 0.989**                                |
| BY-I    | Yellow sandstone soil | 0.0031                     | 0.0495                    | 5.9                  | 8.9                 | 0.040             | 0.527            | 55.4         | 0.988**                           | 0.912**                            | 0.968**                                | 0.867**                                | 0.935**                                |
| BY-II   | Yellow limestone soil | 0.0014                     | 0.0250                    | 4.0                  | 7.0                 | 0.030             | 0.335            | 42.0         | 0.988**                           | 0.322                              | 0.991**                                | 0.326                                  | 0.997**                                |

** Significant at 0.01 level

The correlations between LSOC and SOC are listed in Table 2. The hot-WEOC was significantly correlated with SOC and SROC in yellow sandstone soils and limestone soil profiles. Except for the yellow limestone soil profiles (BY-II), significant correlations between cold-WEOC, SROC, and SOC were also observed in both soil types. These correlation coefficients between hot-WEOC and SOC or SROC were higher than those between cold-WEOC, SROC, or SOC.
3.5. Ratio of LSOC
The percentages of the cold-WEOC in total SOC showed different depth profiles from those of the hot-WEOC in the soils: the former showed a general increase from the topsoil to the bottom, ranging between 0.006%–0.091% (Figure 7a), while the latter showed a general decrease from the topsoil to the bottom, ranging between 0.152%–0.932% (Figure 7b). The average percentages of the WEOC in total SOC were the highest in the sandstone soil profile, being 0.040% (25°C) and 0.527% (70°C), while the average percentages of the WEOC in total SOC in limestone soil were the lowest, being 0.015% (25°C) and 0.255% (70°C) (Table 2). The soils from the sandstone soil profiles (BY-I) showed distinctively high ratios of WEOC/SOC, especially for soils over 60 cm depth. The results showed that the percentages of the SROC in total SOC also presented a decreasing trend with increasing soil depth (Figure 8). The average percentages of the SROC in the total SOC were much higher than those of the WEOC, and were variable in different soil types, ranging between 39.4%–55.4% (Table 2). As stated above, unlike the ratios of WEOC/SOC, there was no clear difference between the SROC/SOC ratios of the limestone and sandstone soils.

Figure 7. Ratios of cold-WEOC to SOC (a) and hot-WEOC to SOC (b) content in different soil profiles.

Figure 8. Ratios of SROC to SOC content in different soil profiles.
4. Discussion

4.1. Vertical distribution of SOC

The changes in SOC content with depth can be divided into three soil layers in each of the five soil profiles (Figure 3). From the maximal value of the topsoil, SOC content first decreased quickly with depth, then decreased slightly below a certain depth, and finally kept a nearly constant value down to the bottom. However, the decreasing rate and thickness of the surface soil layer were different at each sampling site. C/N ratios were commonly used as an indicator of SOM decomposition and tended to decrease with increased decomposition. Figure 4 shows this regular change, but because of the difference in vegetation forms on the five profiles with different soil types, C/N ratios of litter added to the soil were different.

The content of SOC is determined by the balance between plant material input and losses of organic carbon caused mainly by heterotrophic decomposition [18]. Under the subtropical, monsoonal, and humid climate, the high temperature and abundant rainfall environment are propitious to vegetation growth. There is a considerable annual input of plant debris into the soil, which usually maintains a high level of SOC content (> 20 g kg$^{-1}$) in the topsoil [19]. In addition, the distribution of SOC with increasing depth is closely related to the evolution of soil profiles. The source of SOC decreases gradually from the topsoil to the bottom, but the losses due to decomposition increase continuously. As a result, SOC content reduces with increasing depth [20].

Heckman [21] recently conducted a systematic study on the geological controls of soil carbon cycling and microbial dynamics in temperate conifer forests, and demonstrated that carbon stabilization mechanisms vary in different soils developed from different parent rocks. They concluded that the variation in the mechanism of C stabilization may be directly related to the parent material control over the soil pH and mineral assemblage. There were distinct differences in SOC content between the yellow sandstone and limestone soils and their amounts were higher in limestone soils than in yellow sandstone soils. As shown in Figure 9d, the black limestone soils have high amounts of SOC and high pH values, while the yellow sandstone soils have low amounts of SOC and low pH values, which indicated a possible pH control on the SOM content in the soils.

Soil pH can have an important overarching control of SOC dynamics, although pH and SOC content are not directly correlated [21]. Soil pH can play an important role in affecting SOM by controlling mineral and microbial variables [22] as well as mineral dissolution and metal speciation [23]. The high pH values of black limestone soils favor the conservation of mobile cations and trace metals through mineral sorption, which may stabilize the SOC by calcium-humus complexation and sorption of C [21]. Our study data indicate that the yellow sandstone soils have pH values below 5, and hence more calcium and magnesium are lost by leaching from soils, and weak calcium enrichment by organisms leads to lower SOM content than those in the limestone soils. This may be an important reason to explain why the limestone soils maintain higher SOC content than the yellow sandstone soils.

In all soil types, the SOC content showed a similar trend, i.e., it decreased quantitatively with increasing depth, which agreed with earlier studies [24]. However, the extent of this decrease in SOC concentration varied between the different soil profiles (Figure 3). In comparison with SOC in the 0–20 cm soil horizon, the amount of SOC from the 20 cm depth to the bottom depth sampled was, on average, reduced by 7.1% in the limestone soil profiles and reduced by 31.5% in the yellow sandstone soil profiles. The SOC content was mainly concentrated between the depths of 0–20 cm in the yellow limestone and sandstone soils, but the SOC content decreased from the surface soils to depths of 60 cm in the two black limestone soil profiles. These results indicated that the extent of this decrease in SOC concentration with increasing depth was lower in limestone soils than in yellow sandstone soils from topsoil to the bottom. It is well-known that there is a positive correlation between SOC content and soil clay content [15]. According to Zhu and Liu [25], the limestone soils have high clay content and show obvious clay enrichment from the topsoil to the bottom. Clay minerals have a high specific surface area and carry a charge, enabling them to bind, and thereby chemically stabilize the organic matter. Clay aggregates also provide micropores for the physical protection of SOM [26]. These two
effects may be another important reason to explain why the limestone soils maintain higher SOC content than the yellow sandstone soils.

Figure 9. Variations of cold-WEOC (a), hot-WEOC (b), SROC (c), and SOC (d) content with pH values in the soils.

4.2. Vertical distribution of WEOC and SROC

Water-extractable organic matter (WEOM) accounts for only a small proportion of SOM in soils [6]. Nevertheless, it plays a crucial role in the changes in the quality of SOM. The WEOM is composed of an array of molecules, which reflects the composition of total SOM since the soluble phase tends to be in equilibrium with the solid phase of SOM [27]. A detailed description of compounds that can be found in the soil solution is beyond the scope of this study, but the subject has been reviewed by Stevenson [28]. Numerous biotic and abiotic factors control the temporal and spatial dynamics of WEOM [29]. Briefly, soil properties such as pH and ionic strength of the water phase determine the solubility of organic matter, whereas aluminum (Al) and iron (Fe) oxides and clay minerals determine the sorption/desorption equilibrium between the dissolved phase and the solid phase of SOM. The production and consumption of WEOM are mainly dependent on microbial activities and the equilibrium with the solid phase of SOM.

In all soil types, both variations of cold-WEOC and hot-WEOC content displayed the same trend and decreased in the following order: QZ-I > BY-I > QZ-II > QZ-III > BY-II (Table 2). The difference in the content of WEOC (25°C and 70°C) in soils under different vegetation cover and soil types might have resulted from the ability of soil adsorption and soil pH value. WEOC concentration in soil minerals is related primarily to the ability of soil horizons to adsorb WEOC [30]. In addition, the percentages of the WEOC in total SOC showed different amounts between the yellow sandstone and
limestone soils, with the highest percentages of the cold- and hot-WEOC in total SOC found in the yellow sandstone soil profiles (BY-I) and the least ratios of the cold-WEOC/SOC and hot-WEOC/SOC respectively found in the black limestone soil profiles (QZ-II) and profiles (QZ-I) (Table 2). The WEOC values, like the SOC content decreased significantly with increasing depth and showed remarkable changes between the different soil types. In particular, the amounts of WEOC were often higher in the topsoil than the values in the deepest ones (Figure 5). The decrease in the WEOC concentration in deep soil horizons may be attributed to adsorptive interaction of WEOM with reactive surfaces of the mineral matrix (minerals and oxides) [29]. Earlier studies suggested that the readily mineralizable SOC and easily available pool of organic nitrogen could be determined using the hot-water extraction method [31]. Therefore, depletion in hot-WEOC or total N could also give an early indication of the decline or deterioration in SOC and total N, which agrees with the results of the earlier study [2].

Litterfall represents the most important source of C inputs to the forest floor [32]. The annual terrestrial total litter production almost equals the net terrestrial primary production of approximately 60 PgC yr$^{-1}$ [33]. It is concluded that WEOM originates primarily from recent litter decomposition, humus, and root exudates, which releases a large number of WEOC when soaked in the rain [29]. The quality of litter is determined largely by the dominant vegetation, which, thus, plays an essential role in controlling WEOM concentrations in soil solutions [34]. The soil solutions from mixed and coniferous stands often contain significantly more WEOC than those from hardwood stands [35]. Cronan [36] also pointed out that WEOC export from coniferous forests was 50% higher than those from hardwood stands. As shown in Figure 5, the WEOC content was higher in BY-I yellow sandstone soils than those in the BY-II and QZ-III soil profiles from the topsoil to a depth of approximately 20 cm. This result may be ascribed to the abundant litter material (e.g., leaf fall) in the surface soils and more active microbial action in this layer forming a higher WEOC than those in the BY-II and QZ-III soil profiles.

Similarly, the SROC content and the ratios of SROC/SOC are sensitive indicators, which can reflect the stability of organic carbon in soils: the larger the ratio of SROC/SOC, the greater the activity of soil carbon [37]. The results showed that the LSOC/SOC ratios differed between the limestone and yellow sandstone soil profiles and the average percentages of the SROC in the total SOC were larger than those of WEOC and were variable in different soil types, ranging between 39.4%–55.2% (Figure 7 and Table 2). Similar to the total SOC, the SROC content tended to decrease with increasing depth and showed different amounts in different soil types, with the content of SROC in limestone soil significantly higher than in the yellow sandstone soil. This result illustrated that the soil carbon pool activity was higher in limestone soils than in yellow sandstone soils and was easily transformed and lost mainly due to changes in land use. However, the amounts of SOC, WEOC, and SROC were lower in BY-II yellow limestone soil than in black limestone soil and even in the yellow sandstone soil profile. This may be attributed to the dominant vegetation (i.e., shrubs) and the thicker, higher humidity, and looser soil in this sampling site.

In this study, there were distinct differences in WEOC and SROC content between yellow sandstone and limestone soils and their amounts were higher in limestone soils than in yellow sandstone soils. As shown in Figure 9, similar to SOC, the black limestone soils have high amounts of LSOC and high pH values, while the yellow sandstone soils have low amounts of LSOC and pH values. These results indicate that the pH exerts control over the LSOC in the soils. As observed by Kalbitz [29], soil pH can affect the dynamics of LSOM via physico-chemical processes. Similarly, laboratory studies also show that LSOC released from organic soil horizons is positively correlated with pH [38]. Furthermore, Tipping and Woof [39] calculated that an increase in soil pH of 0.5 units would lead to increase of approximately 50% in the amount of LSOM. Therefore, the solubility of dissolved organic matter is diminished by the high degree of protonation resulting from low pH [40]. The yellow sandstone soils have pH values generally below 5.0, and hence more calcium and magnesium are lost by leaching from soils and weak calcium enrichment by organisms leads to lower LSOM content than that in the limestone soils.
4.3. Relationship of hot-WEÖC to other soil parameters

All data collected from limestone soils and sandstone soil profiles were pooled together to examine the correlations between hot-WEÖC and SOC or SROC and between cold-WEÖC and SOC or SROC. All correlations between hot-WEÖC and these variables were significant at \( P < 0.01 \). SOC was also strongly correlated with the hot-WEÖC content in soils (Table 2). This suggests that, as expected, total SOM content was a major determinant of the amount of LSOM present. Sparling [13] also reported a strong positive correlation between hot-WEÖC and soil MBC or SOC. The amounts of hot-WEÖC extracted from soils were 10 times higher than the cold-WEÖC; the former varied in the range of 0.0107–0.2633 g kg\(^{-1}\) and the latter varied in the range of 0.0009–0.0132 g kg\(^{-1}\). It is considered that the higher amounts of C in the hot-WEÖC were because it would have extracted not only the MBC but also root exudates, soluble carbohydrates, and amino acids.

Ghani [2] demonstrated that a strong positive correlation was found between hot-WEÖC and anaerobically mineralizable N. This is another indication that hot-WEÖC can extract a mineralizable pool of organic matter from soils. There have been suggestions that the WEÖC, especially the hot-WEÖC, being part of the highly labile pool of C, may also be sensitive to perturbation and stress in the soil–plant ecosystems [41] and therefore could be used as a sensitive indicator of the quality of SOM. However, WEÖC is usually considered to be smaller than other labile pools. There is a positive correlation between hot-WEÖC and other labile SOM fractions present in this study. This is in agreement with the results of earlier studies [42]. The correlation between the total SOC and hot-WEÖC was often better than those observed for SROC. Changes in total organic carbon in soils also reflected the hot-WEÖC pools and changes in hot-WEÖC levels also often reflected the organic carbon measurements.

4.4. Hot-WEÖC as an integrated indicator of SOM quality

Hot-WEÖC extracts not only labile C but also other labile nutrients [43]. Earlier work by Keeney and Bremner [31] suggested that the easily mineralizable pool of organic N could be determined by a simple hot-water extraction method. Therefore, if hot-WEÖC declines either because of cultivation, excessive N fertilization, or overgrazing of soil, the pool of other labile nutrients would also decrease. From a production viewpoint, this would mean a comparatively greater dependency on chemical fertilizers and thus a greater environmental risk through leaching or run-off of nutrients to waterways. Hence, the maintenance of a threshold amount of hot-WEÖC would be regarded as an environmentally desirable feature for sustainable production and environmental protection. Depletion in hot-WEÖC could also give an early indication of the deterioration or decline of SOC.

Ghani [2] reported that approximately 40.0%–50.0% of the C in the hot-water extracts was present as carbohydrates and the hot-water extract in the forest soils is also rich in carbohydrates. Furthermore, Haynes and Francis [44] confirmed the results of Ghani [2]. These researchers have also reported a better correlation between aggregate stability and hot-water extractable carbohydrates than the total soil carbohydrates measured by acid hydrolysis. This might indicate that in these soils the proportion of carbohydrate is consistent regardless of the origin of SOM. Feller [45] confirmed that hot-WEÖC fraction of organic matter is rich in amorphous polysaccharides, which are similar to those originating from microbial and plant exudates. Further, hot-WEÖC is also regarded as one of the key labile components of organic matter responsible for soil micro-aggregation [46] which is an important soil physical parameter to consider in terms of soil quality. Given among the LSOC, the hot-WEÖC showed clear differences between different soils, and the hot-WEÖC was significantly correlated with the SOC and SROC in the yellow sandstone soils and limestone soil profiles, and the correlation coefficients were higher than those between the cold-WEÖC and SROC or SOC (Table 2). Therefore, the hot-WEÖC could be used as a surrogate measure to estimate these soil parameters. Measurement of any of these soil properties individually is costly and time consuming.

To the time of reporting, this is the first study, which has evaluated the usefulness of hot-WEÖC as an indicator of SOM quality in Guizhou karst areas of southwest China. It can detect subtle changes caused in the SOM due to land use management. Several soil parameters such as SOM, MBC, SROC,
CO₂ respiration rate, mineralizable N, enzyme activities etc. have been suggested by many researchers as indicators of SOM quality which can be tracked over time to gauge the performance of certain management practices on soil [41]. However, some of these measurements are either too insensitive in this temperate climate (e.g. organic matter) or oversensitive (e.g., enzyme activities, microbial biomass, CO₂ respiration etc.) to be useful as an indicator of SOM quality. Our results demonstrate that hot-WEOC having good correlation with some of these measurements has considerable potential for being a useful indicator of SOM quality. Measurement of hot-WEOC in soils also provides an opportunity to keep a closer eye on the impacts of soil–plant–animal interactions on the wider labile pool of soil carbon than other soil parameters.

5. Conclusions
The amounts of SOC, SROC, and WEOC, including hot- and cold-WEOC, mainly concentrated in the surface soil, decreased with increasing depths, and showed different amounts in different soil types. The contents of SOC and LSOC in limestone soils were significantly higher than those in the yellow sandstone soils. The higher SOC and LSOC content in the limestone soils can be ascribed to soil mineralogy, higher pH values, and more calcium ion than in the yellow sandstone soils.

The percentages of the LSOC in total SOC in different soil profiles were distinctly different, and the ratios of SROC/SOC were much higher than that of WEOC. The content of SROC was also much higher than the WEOC and its average content accounted for 39.4%–55.2% of SOC in the soil profiles under study. The trends of a decrease in SROC and WEOC with increasing depth were similar to those in terms of the amount of SOC. These results suggest that they are interrelated properties and the change in the amount of SOC dominates that of LSOC in the study area.

All the LSOC, including cold- and hot-WEOC as well as SROC vary with SOC in a positive relationship. Among the LSOC, the hot-WEOC showed clear differences between the different soil types. Its content was 10 times higher than the cold-WEOC, and significant relationships existed not only with SOC but also SROC. The correlation coefficients between the hot-WEOC and SOC or SROC were higher than those between cold-WEOC and SROC or SOC. Therefore, hot-WEOC can be used as a sensitive indicator of changes in the quality of SOM in the soils of the karstic areas.

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