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Moss-dominated biological soil crusts improve stability of soil organic carbon on the Loess Plateau, China

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Abstract: The succession of biological soil crust (biocrust) may alter soil organic carbon (SOC) stability by affecting SOC fractions in arid and semi-arid regions. In the study, the SOC fractions were measured including soil easily oxidizable carbon (SEOC), soil microbial biomass carbon (SMBC), soil water soluble carbon (SWSC), and soil mineralizable carbon (SMC) at the Loess Plateau of China by using four biocrusts. The results show that SOC fractions in the biocrust layer were consistently higher than that in the subsoil layers. The average SOC content of moss crust was approximately 1.3–2.0 fold that of three other biocrusts. Moss crusts contain the lowest ratio of SEOC to SOC compared with other biocrusts. The ratio of SMC to SOC was the highest in light cyanobacteria biocrust and the lowest in moss crust, but no difference was observed in SMBC to SOC and SWSC to SOC in biocrust layers among four studied biocrusts. The results show that the moss crusts increase the accumulation of organic carbon into soil and reduce the ratio of SEOC to SOC and SMC to SOC. Together, these findings indicate that moss crusts increase the SOC stability and have important implications that SOC fractions and mineralization amount are good indicators for assessing the SOC stability.

Keywords: biocrust type; soil layer; moss coverage; carbon fractions

Organic carbon stored in soils is the largest global reservoir of fixed carbon. As an important component of the global carbon cycle, soil organic carbon (SOC) has received widespread attention from environmental scientists and pedologists. A slight change in SOC could cause an obvious fluctuation in the concentration of atmospheric CO_2 which would directly influence climate change (Lal 2004). SOC stability is closely related to SOC fraction. Many studies have been conducted to elucidate the mechanisms of SOC stabilization (Semenov et al. 2008) which is important for predicting the effects of global climate change and controlling carbon emissions.

Biological soil crusts (biocrusts) consist of microscopic (cyanobacteria, algae, fungi and bacteria) and macroscopic (lichens, mosses) poikilohydric organisms that occur on or within the top few centimetres of the soil surface (Belnap and Büdel 2016). Biocrusts cover most dryland surface that are not occupied by stems of vascular plants, rocks or active disturbances, and account for 70% or more of dryland ecosystems' living cover (Belnap and Büdel 2016) which can alter the physicochemical characteristics of surface soils. Many studies also showed that biocrusts played an important role in carbon fixation and carbon emission in arid and semi-arid ecosystems which also greatly depends on the type of biocrust (Pietrasiak et al. 2013). However, few studies investigated the effect of biocrust types on SOC fractions.

In 1999, the Chinese government initiated the nationwide 'Grain for Green' project on the Loess Plateau region of China to improve the ecologi-

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cal environment by converting unsuitable sloping farmlands to forests or grasslands (Deng et al. 2017). Subsequently, extensive biocrusts were formed on open spaces of revegetated lands. The total biocrust coverage is estimated to be 60–70% on the Loess Plateau and the SOC content significantly increased with biocrust succession (Zhao et al. 2006). Some studies used alternative approaches to evaluate SOC stabilization (Liaudanskienė et al. 2013) and few data are available on the SOC concentrations in labile SOC fractions and the mechanisms that determine these concentrations. Therefore, it is unclear whether and to what degree biocrusts affect SOC stability. This situation makes it difficult to understand the role of biocrusts in the carbon cycle in arid and semiarid regions. In this study, the effects of four typical types of biocrusts on SOC and its stability on the Loess Plateau were investigated. The objective of our study was to assess SOC stability in four biocrusts types by determining their content of different SOC fractions and cumulative CO_2-C, clarify how different biocrust types improve SOC stability, and to analyse the internal mechanisms of SOC stability in biocrust soil.

MATERIAL AND METHODS

Study area. The research was conducted on revegetated grasslands in the Ansai county, located on a typically gully and hilly Loess Plateau landscape (36°51'N; 109°19'E) with mean altitude of approximately 1200 m a.s.l. in northern Shaanxi province, China. It has a typical continental monsoon climate with an average annual temperature of 8.8°C. The mean annual precipitation is 500 mm, and approximately 60% occurs from June to September. The typical loess soils at the site are classified as Calciustepts with a silty loam texture. Biocrusts developed naturally, and are mainly distributed in the open spaces of revegetated lands, especially on hills and gully slopes. Moss species include *Didymodon vinealis*, *D. tectorum*, *Crossidium squamiferum*, and *Bryum argenteum*. Cyanobacteria species, such as *Phorumidium calciola*, *P. tenue* and *Lyngbya allorgei*, typically dominate on sunny slopes.

Experimental design. Four biocrust types were randomly selected at four sampling sites, with each type approximately 1–8 km apart. Sampling sites were 60 × 60 m in size and were approximately 500 m apart from each other. Biocrusts were classified into four types based on the moss coverage. The biocrust types included: (1) light cyanobacteria biocrust; (2) cyanobacteria mixed with sparse moss; (3) moss mixed with sparse cyanobacteria; and (4) moss crust.

Soil sampling and laboratory analyses. Soil samples were collected in April 2016. Each biocrust type includes three sampling sites. At each sampling site, samples were collected from the following soil layers: (1) the 'biocrust-layer' (Table 1); (2) the 0–2 cm soil layer immediately beneath the biocrust; (3) the 2–5 cm soil layer, and (4) the 5–10 cm soil layer. Soil samples from each soil layer of the same plot were mixed thoroughly to obtain a composite sample.

Biocrust coverage was evaluated using the quadrate method (Belnap et al. 2001). Soil particle size distribution was performed using a laser-diffraction method (Mastersizer 2000, Malvern, UK). Biocrust bulk density was measured using the glue coating method, and the SOC content was measured according to the Walkey and Black's dichromate oxidation method. The soil easily oxidizable carbon (SEOC) content was determined using a modified potassium permanganate oxidation procedure (Mirsky et al. 2008). The soil microbial biomass carbon (SMBC)

Table 1. Site description of the four biocrusts

Biocrust type	Cyanobacteria coverage (%)	Moss coverage (%)	Biocrust thickness (mm)	Biocrust bulk density (g/cm^3)	Sand content (%)
LC	73	8	1.09 ± 0.06^{d}	1.30 ± 0.08^{a}	36.14 ± 2.47^{a}
CM	53	31	3.40 ± 0.26^{c}	1.23 ± 0.08^{ab}	34.55 ± 2.72^{ab}
MC	18	62	6.70 ± 0.11^{b}	1.15 ± 0.09^{b}	34.46 ± 0.74^{ab}
M	7	82	9.03 ± 0.12^{a}	0.99 ± 0.04^{c}	28.90 ± 6.09^{b}

LC – light cyanobacteria biocrust; CM – cyanobacteria mixed with sparse moss; MC – moss mixed with sparse cyanobacteria; M – moss crust. Values are in the form of mean ± standard deviation (n = 3). Different lower-case letters within a column indicate significant differences among different biocrusts at $P < 0.05$

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content was determined with the fumigation extraction method (Brookes et al. 1985). The SWSC content was measured with the extraction method described by Liang et al. (1997). The soil mineralizable carbon (SMC) was tested through a laboratory incubation experiment (Marumoto et al. 1982). The soil incubation was performed at constant humidity and temperature for 33 days. The soil moisture content corresponded to ~12% of the soil water content. All jars were kept overnight in the dark at 25°C.

Statistical analysis. Statistical analyses were performed using the SPSS software, ver. 18.0. Homogeneity of variance was ensured using the Levene's test. One-way ANOVA tests were performed to detect significant differences between the mean values of SOC fractions of different biocrust types, followed by the Fisher's *LSD* (least significant difference) post hoc test. Statistical significance was defined as $P < 0.05$. All figures were performed using Excel 2007.

RESULTS AND DISCUSSIONS

Effect of different biocrusts on SOC. In the study region, well-developed biocrusts contained a high cover of mosses, leading to an enrichment of SOC in the topsoil. This enrichment was more pronounced in more developed biocrusts (Gao et al. 2017). Biocrust types and soil depth had a significant impact on the SOC content. The SOC content of biocrust layers increased with their successional stage. The average SOC content of moss crusts was about 1.3-, 1.7-, and 2.0-fold higher than moss mixed with sparse cyanobacteria, cyanobacteria mixed with sparse moss, and light cyanobacteria biocrust, respectively. The SOC content decreased significantly with increasing soil depth. The SOC content in the biocrust layer was two to five times higher than that in the subsoil layers, but no significant difference in SOC content was observed among subsoil layers (Figure 1).

Among the four biocrusts, no difference in SOC content was found in the subsoil layer, but significant differences in the biocrust layer. The above phenomenon indicates that the variation in the SOC content in the biocrust layer is caused by biocrust type. Relative studies showed that the net photosynthetic rate of biocrusts is closely related to the dominant species of biocrusts (Li et al. 2012). Our results indicated that the SOC content of moss crust was significantly higher than that of cyanobacteria biocrust. This may be due to the fact that later-successional biocrusts experience a high level of photosynthesis and chlorophyll fluorescence (Lan et al. 2012) and cyanobacteria were mainly distributed in the 200–500 μm soil layer, which receives a low amount of sunlight (Brostoff et al. 2002).

Effect of different biocrusts on SOC fractions. Relevant studies have also indicated that the contents of both SEOC and SMBC are positively related to SOC stability (Hernandez-Soriano et al. 2016) and are important indicators of SOC accumulation or decomposition (Tarafdar et al. 2001). Carbon fraction contents decreased with increasing soil depth (Figure 2). The contents of all SOC fractions were consistently higher in the biocrust layer than in subsoil layers. The SEOC content of moss crust and moss mixed with sparse cyanobacteria was 1.4–1.7 fold that of light cyanobacteria biocrust (Figure 2a). The average SMBC content in moss mixed with sparse cyanobacteria and moss crust was 2.0–2.1 fold that of cyanobacteria mixed with sparse moss and light cyanobacteria biocrust (Figure 2b). The SWSC content of moss crust was significantly higher (1.2-fold) than that of light cyanobacteria biocrust (Figure 2c). Moss crusts contained the lowest ratio of SEOC to SOC compared with other biocrusts, and showed a significant difference compared with light cyanobacteria biocrust (Table 2).

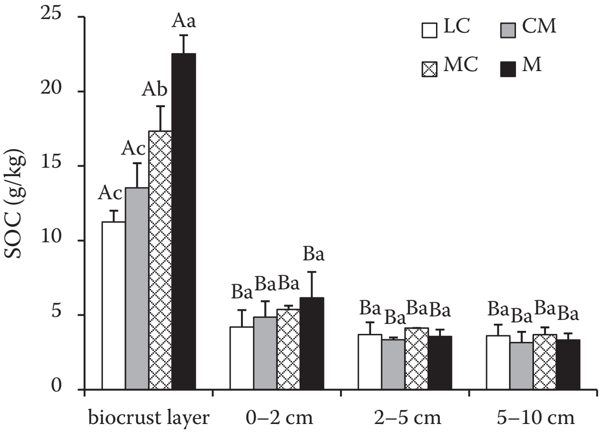

Figure 1. Soil organic carbon (SOC) content in four biocrusts at different soil layers. Values are in the form of mean ± standard deviation ($n = 3$). Different upper-case letters above the bars indicate significant differences between different soil layers of the same biocrust types. Different lower-case letters above the bars indicate significant differences between different biocrusts of the same soil layers ($P < 0.05$). LC – light cyanobacteria biocrust; CM – cyanobacteria mixed with sparse moss; MC – moss mixed with sparse cyanobacteria; M – moss crust

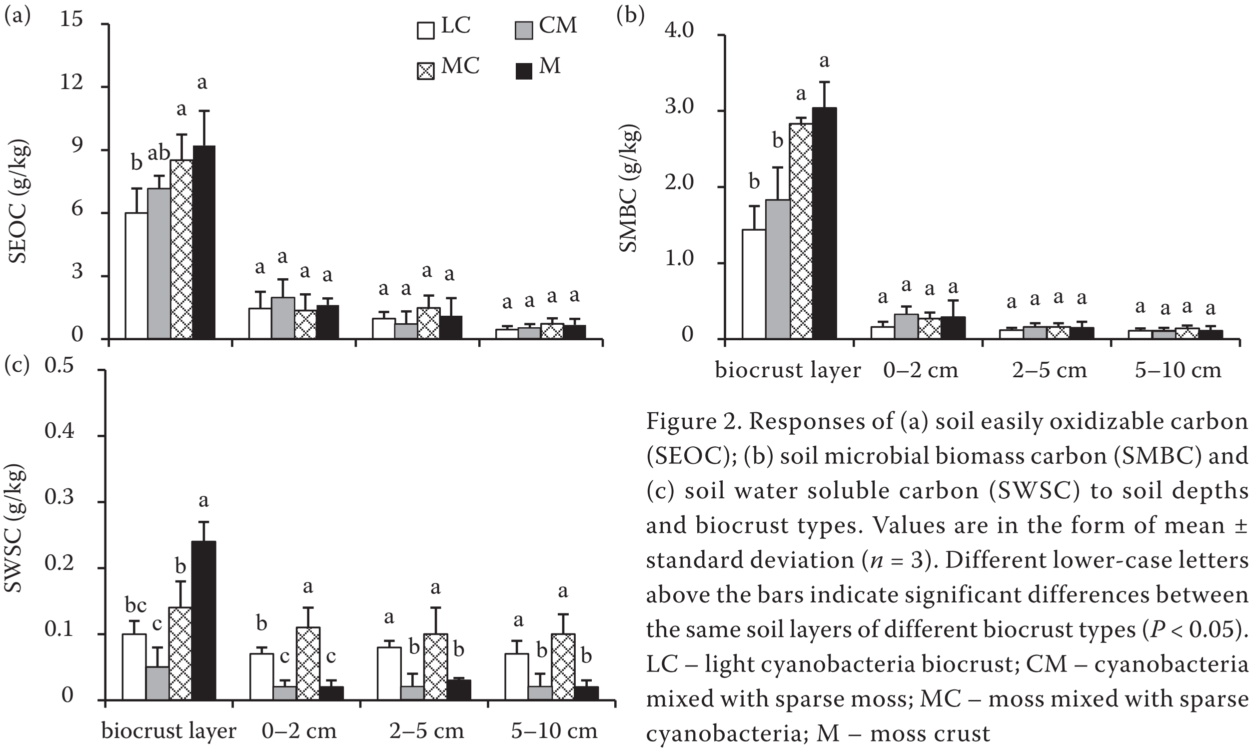

Figure 2. Responses of (a) soil easily oxidizable carbon (SEOC); (b) soil microbial biomass carbon (SMBC) and (c) soil water soluble carbon (SWSC) to soil depths and biocrust types. Values are in the form of mean ± standard deviation ($n = 3$). Different lower-case letters above the bars indicate significant differences between the same soil layers of different biocrust types ($P < 0.05$). LC – light cyanobacteria biocrust; CM – cyanobacteria mixed with sparse moss; MC – moss mixed with sparse cyanobacteria; M – moss crust

Previous research has shown that SOC decomposition rates are strongly associated with the chemical composition of SOC (Grandy and Neff 2008). This observation indicates that biocrust types can influence SOC chemical composition (Chamizo et al. 2012). This may be due to different biocrusts inputting

Table 2. Ratios of soil organic carbon (SOC) fractions to SOC in different soil depths and biocrust types

Biocrust type		SEOC/SOC	SMBC/SOC	SWSC/SOC
Biocrust layer	LC	0.53 ± 0.07^{a}	0.13 ± 0.02^{a}	0.01 ± 0.00^{a}
	CM	0.50 ± 0.11^{ab}	0.16 ± 0.01^{a}	0.01 ± 0.00^{a}
	MC	0.50 ± 0.11^{ab}	0.16 ± 0.01^{a}	0.01 ± 0.00^{a}
	M	0.41 ± 0.06^{b}	0.14 ± 0.01^{a}	0.01 ± 0.00^{a}
0–2 cm	LC	0.33 ± 0.12^{a}	0.04 ± 0.01^{a}	0.02 ± 0.00^{a}
	CM	0.36 ± 0.15^{a}	0.06 ± 0.02^{a}	0.00 ± 0.00^{a}
	MC	0.38 ± 0.21^{a}	0.06 ± 0.02^{a}	0.00 ± 0.00^{a}
	M	0.27 ± 0.08^{a}	0.04 ± 0.02^{a}	0.00 ± 0.00^{a}
2–5 cm	LC	0.27 ± 0.08^{a}	0.03 ± 0.00^{a}	0.02 ± 0.00^{a}
	CM	0.21 ± 0.18^{a}	0.05 ± 0.01^{a}	0.01 ± 0.00^{a}
	MC	0.28 ± 0.14^{a}	0.04 ± 0.01^{a}	0.04 ± 0.01^{a}
	M	0.30 ± 0.20^{a}	0.04 ± 0.02^{a}	0.01 ± 0.00^{a}
5–10 cm	LC	0.12 ± 0.03^{a}	0.03 ± 0.01^{a}	0.02 ± 0.01^{a}
	CM	0.16 ± 0.03^{a}	0.03 ± 0.01^{a}	0.01 ± 0.00^{a}
	MC	0.19 ± 0.05^{a}	0.04 ± 0.01^{a}	0.06 ± 0.02^{a}
	M	0.20 ± 0.07^{a}	0.03 ± 0.01^{a}	0.00 ± 0.00^{a}

SEOC – soil easily oxidizable carbon; SMBC – soil microbial biomass carbon; SWSC – soil water soluble carbon; LC – light cyanobacteria biocrust; CM – cyanobacteria mixed with sparse moss; MC – moss mixed with sparse cyanobacteria; M – moss crust. Values are in the form of mean ± standard deviation ($n = 3$). Different lower-case letters within a column indicate significant differences between different biocrusts of the same soil layers at $P < 0.05$

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Table 3. Cumulative mineralizable soil organic carbon content (g/kg) in different soil layers under four biocrusts

Biocrust type	Biocrust layer	0–2 cm	2–5 cm	5–10 cm
Light cyanobacteria biocrust	0.78 ± 0.08^{b}	0.09 ± 0.02^{a}	0.07 ± 0.01^{a}	0.06 ± 0.00^{a}
Cyanobacteria mixed with sparse moss	0.88 ± 0.12^{b}	0.13 ± 0.04^{a}	0.07 ± 0.01^{a}	0.06 ± 0.01^{a}
Moss mixed with sparse cyanobacteria	1.21 ± 0.03^{a}	0.11 ± 0.02^{a}	0.07 ± 0.00^{a}	0.06 ± 0.00^{a}
Moss crust	1.09 ± 0.06^{a}	0.11 ± 0.02^{a}	0.07 ± 0.01^{a}	0.05 ± 0.01^{a}

Different lower-case letters within a column indicate significant differences between different biocrusts of the same soil layers ($P < 0.05$)

different amounts of SOC into the soil (Chamizo et al. 2012). However, the effects of biocrust type on SOC fractions were limited to the biocrust layer, mainly owing to decreasing SOC input with depth.

Effect of different biocrusts on SOC stability. After 33 days of incubation, significant differences between the cumulative SMC contents of different biocrust layers were observed, but no significant difference was observed in other soil layers. The SMC content was highest in moss with sparse cyanobacteria and lowest in light cyanobacteria biocrust (Table 3). The lowest SMC to SOC ratio (4.7%) was measured in moss crust and the highest (6.1%) was measured in light cyanobacteria biocrust. The SMC to SOC ratio significantly varied between four biocrusts ($P < 0.05$), especially among biocrust layers (Figure 3). Related studies have shown that biocrusts play an important role in reducing SOC loss, and this effect is more noticeable in more developed biocrusts (Chamizo et al. 2017). The loss of SOC may be up to nine times higher under early cyanobacteria biocrusts relative to later-successional biocrusts (Barger et al. 2006). Previous studies have frequently reported potentially mineralizable carbon as an indicator of the SOC stability (Tian et al. 2016), which confirms our findings that soils associated with moss crusts showed the highest SOC stability. A portion of the SOC accumulated by cyanobacteria may be decomposed under conditions favouring strong oxidation. Consistent with our hypothesis, as biocrust succession advances, moss crust increases SOC fractions, reduce the ratio of SEOC to SOC and the ratio of SMC to SOC, and improves the SOC stability.

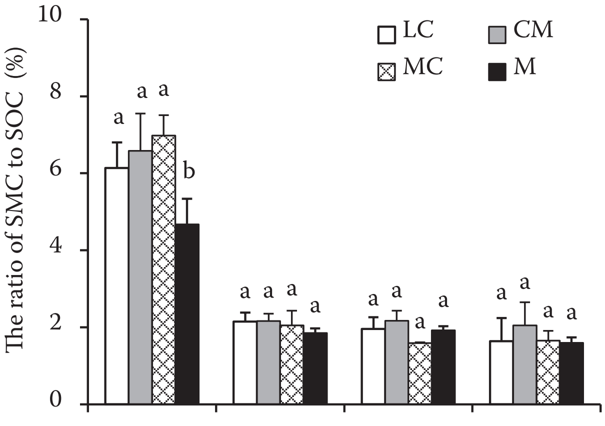

Figure 3. The ratio of soil mineralizable carbon (SMC) to soil organic carbon (SOC) in different biocrusts. LC – light cyanobacteria biocrust; CM – cyanobacteria mixed with sparse moss; MC – moss mixed with sparse cyanobacteria; M – moss crust. Different lower-case letters above the bars indicate significant differences between different biocrusts of the same soil layers ($P < 0.05$)

From a physiological standpoint, the internal components of biocrusts affect the SOC stability. Moss crusts contain phenolic compounds similar to lignin which inhibit corrosion and predation and are resistant to microbial decomposition (Xu et al. 2003). Cyanobacteria excreted exopolysaccharides, which provide the 'glue' that enables filaments to adhere to soil particles and other organisms (Belnap and Büdel 2016). In addition, soil bulk density decreased with biocrusts succession. With the succession of biocrusts, total phototrophic biomass and fine particles accumulate, both of which lead to a gradual increase in biocrust thickness (Lan et al. 2012). In our study, with the succession of biocrusts, biocrust thickness showed a significantly increasing trend (Table 1). Related researches also suggest that the formation of microaggregates protects SOC from physical decomposition (Six et al. 2002). Our study also revealed that a large proportion of carbon accumulated in light cyanobacteria may be lost under conditions favouring strong mineralization. Overall, SOC mineralization was controlled by many complex factors and a further study of the SOC stability is needed.

In conclusion, moss crusts significantly increase the SOC content in soils of the Loess Plateau, reducing the ratios of SEOC to SOC and SMC to SOC compared with other three biocrust types. Therefore, moss crusts facilitated carbon accumulation and were not highly vulnerable to oxidation or mineralization. These results highlight the importance of moss crust in improving the SOC stability, which may play an important role in controlling the greenhouse effect.

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