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Title: Biocrust reduced soil water retention and soil infiltration in the alpine *Kobresia* meadow

Running title: Biocrust reduced soil water retention and soil infiltration

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Abstract

Biocrust is a key component of ecosystems and plays a vital role in altering hydrological processes in terrestrial ecosystems. The role of biocrust on hydrological properties in arid and semi-arid ecosystems has been widely documented; however, the effects and mechanisms of biocrust on soil hydrological properties in alpine ecosystems are still poorly understood. In this study, we selected two meadow types from the northern Qinghai-Tibet Plateau: normal *Kobresia* meadow (NM) and biocrust meadow (BM). Both the soil hydrological and physicochemical properties were examined. We found that in the 0–30 cm soil layer, soil water retention and soil water content in NM were higher than those in BM, whereas the 30–40 cm layer's soil water retention and soil water content in NM were lower than those in BM. The topsoil infiltration rate in BM was lower than that in NM. Furthermore, the physicochemical properties were different between NM and BM. The 0–10 cm

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soil layer's clay content in BM was 9% higher than that in NM, whereas the 0–30 cm layer's soil capillary porosity in NM was higher than that in BM. In addition, the 0–20 cm layer's soil total nitrogen (TN) and soil organic matter (SOM) in NM were higher than those in BM, implying that the presence of biocrust did not favor the formation of soil nutrients owing to its lower soil microbial biomass carbon and microbial biomass nitrogen. Overall, soil water retention was determined by SOM by altering soil capillary porosity and bulk density. Our findings revealed that the establishment of biocrust did not improve soil water retention and infiltration, which may be more vulnerable to runoff generation and consequent soil erosion in biocrust meadows. These results provide a systematic and comprehensive understanding of the role of biocrust in the soil hydrology of alpine ecosystems.

Keywords: Alpine meadow; biocrust; soil-soil water retention; soil water infiltration; physicochemical properties

1 Introduction

Soil biocrust is the special soil structure in the terrestrial ecosystem, which is widely distributed in arid and semiarid regions throughout the world, and it plays a vital role in regulating biogeochemical processes, hydrology processes, and surface energy balance, such as improving soil aggregation and stability, increasing the soil fertility, reducing soil erosion and thus maintaining water availability (Li et al., 2016), which can serve as "ecological engineers" in systems. However, to our knowledge, the controlling mechanism of biocrust on soil hydrological processes is still unclear, and most previous studies were conducted in arid and semi-arid ecosystems, such as the Tengger Desert, Negev Deserts, and Loess Plateau, and display a positive effect on soil hydrological properties. Very few studies have focused on the role of biocrust on hydrological properties (i.e.,

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soil water content, soil water retention, and soil infiltration) in high-altitude alpine ecosystems, and the mechanisms are poorly understood. Thus, examining the impact of biocrust on hydrological properties could have substantial effects on water balance in alpine ecosystems.

The alpine meadow is an important ecosystem in the Qinghai-Tibet Plateau (QTP), which plays an important role in water retention (Dai et al., 2019), in preventing soil erosion (Qian et al., 2021) and in regulating energy exchange (Zhu et al., 2020) by altering soil surface features such as roughness, soil texture, porosity, and aggregation (Li et al., 2016), thereby modifying evaporation, soil water retention, and water infiltration processes. However, the formation of biocrust in alpine meadows is different from that in arid areas, where the biocrust is formed from intensive land use such as overgrazing, and the biocrust types vary with the succession stage of alpine meadows (Li et al., 2016b). For instance, as the degree of degradation increases, the moss-dominated crust is transformed into cyanobacteria-dominated crust, followed by lichen-dominated crust from *Graminoid*-dominated vegetation degradation to *Kobresia humilis* meadow (light degradation) and then to *K. pygmaea* meadow (moderate degradation) (Li et al., 2016). Thus, we suggest that the impact of biocrust on hydrologic processes in alpine meadows may differ from that in arid areas, and vice versa.

To date, the effects of biocrust on plant growth and seed germination in alpine meadows have been reported (Li et al., 2016b; Letendre et al., 2019), whereas the impact of biocrust on soil hydrology processes, such as soil water retention and soil infiltration, remains poorly understood. Although numerous studies have pointed out that biocrust has substantial effects on soil water retention and soil moisture infiltration processes by altering soil microenvironments, such as soil roughness, soil porosity, and aggregation, no consensus has been reached. For instance, some studies have found that biocrust could increase soil water infiltration and reduce runoff by increasing soil porosity and aggregate stability in cool desert ecosystems (Kidron and Benenson, 2014; Wei et al., 2015). In contrast, other studies found that soil water infiltration was significantly reduced in crusted areas compared with non-crusted areas in arid ecosystems (Li et al., 2010; Xiao and Hu, 2017). These discrepancies highlight the necessity to further explore the effects of biocrust on hydrological processes, such as exploring the specific hydraulic properties by conducting soil infiltration experiments and soil water retention curve measurements. Furthermore, most previous studies were mainly conducted in arid and semi-arid ecosystems, and very few studies have focused on the effects of biocrust on the soil's hydrological properties in high-altitude alpine ecosystems. Therefore, it is crucial to assess the role of biocrust in soil water retention and infiltration in alpine meadows.

To address these knowledge gaps, both soil and hydrological features were measured with the aim of exploring the role of biocrust in hydrological processes in alpine ecosystems. Specifically, the objectives of this study were to explore the effect of biocrust on soil-hydrological features in alpine ecosystems, to reveal how biocrust affects soil water retention by altering soil and vegetation properties, and provide insights into the management of biocrust in alpine meadows.

2 Materials and methods

2.1 Site description

The field test sites were located in the northeastern Qinghai-Tibet Plateau (101º 19′ E, 37º 37′ N), in Qinghai Province, China (Fig.1a). The area has a continental plateau climate with a mean air temperature of -1.7°C and a mean annual precipitation of approximately 562 mm (Dai et al., 2020). It should be noted that approximately 80% of the precipitation occurs during the growing season (between May and September), and the other 20% occurs during the non-growing season. The main

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vegetation type in this region is the *Kobresia* meadow, which is dominated by *Kobresia humilis* (Fig.1b). The soil type in the study area is silt loam according to the in the USDA soil taxonomy system of classification (Cusack and others 2018), with a soil thickness of approximately 60–80 cm. The pH and EC is 7.5 m s m^{-1} and 6.7 in the study area, respectively. (Li et al., 2016).

2.2 Experimental design and soil sampling

In August 2020, we choose two study sites on the northeastern Qinghai-Tibet Plateau to avoid pseudoreplication, and two types of soil surfaces were selected in each study site, i.e.a normal *Kobresia* meadow (NM, Fig. 1b) and a biocrust meadow (BM, Fig. 1c). To reduce the differences caused by spatial heterogeneity, the BM was selected adjacent to the NM. The vegetation cover in BMs is usually less than 20% with a thick turf but no litter layer in topsoil, and the BM type is dominated by moss crust (ca. 80%) (Li et al., 2016). In contrast, NM has a dense vegetation cover and is mainly dominated by *Kobresia pygmaea*, with average plant heights of 1–3 cm. Furthermore, a clear typical turf horizon and litter layer was observed within the topsoil in NM, that is, the Afe horizon. BM had a higher root biomass than that of NM, owing to its thick turf (Table 1).

We obtained the disturbed soil samples in

in NM and BM and four quadrats (1 × 1 m) were randomly selected for soil sampling with a depth of 10 cm in each treatment using an earth boring auger, and then brought back to the laboratory to measure and analyze soil organic matter (SOM), soil microbial biomass carbon (MBC), microbial biomass nitrogen (MBN), total carbon (TC), total nitrogen (TN), and soil particle size distribution (PSD). Undisturbed cylindrical ring samples were also obtained in each treatment to determine the soil bulk density (BD), soil porosity, and soil hydraulic properties (i.e., soil water retention and soil

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water supply capacity). The soil infiltration rates were measured using a double-ring infiltrometer for each treatment.

2.3 Laboratory measurements and analyses

First, the disturbed soil samples were sieved through 0.25 mm and 2-mm soil sieves to remove debris and roots for the analysis of soil properties; SOM was measured based on the Walkley & Black procedure (Nelson and Sommers, 1982), MBC and MBN were measured by the chloroform fumigation-direct extraction method (Vance et al., 1987), and TC and TN were measured using an element analyzer (Elementar Vario EL III, Hanau, Germany). PSD was determined using a Mastersizer 2000 (Malvern Instruments, UK). BD was measured as the ratio of the oven-dry soil mass to the core volume (100 cm^3). The soil total porosity, soil capillary porosity, and soil non-capillary were measured using the following equation (Dai et al., 2020):

$$\mathrm{TP} = (1 - \frac{BD}{d_s}) \times 100\% \quad (1),$$

$$\mathrm{CP} = CMC \times BD \quad (2),$$

$$\mathrm{NCP} = TP - CP \quad (3),$$

where TP, CP, and NCP represent soil total porosity (%), soil capillary porosity (%), and soil non-capillary porosity (%), respectively; CMC represents soil capillary water capacity; ds is the soil particle density, which was assumed to be 2.65 (g cm^{-3}).

The soil water retention curves (SWRCs) were measured using a pressure plate apparatus (1500 F1, Soil Moisture Equipment Corp., SEC, USA), and the relationship between soil water content and matric potential was fitted by the Gardner model. The formula of the Gardner model is as follows (Gardner et al., 1970):

$$h = A\theta^{-B},$$

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where h is the soil water content (%), θ is the matric potential (kPa), and A and B are the fitting parameters. Higher values of A*B and A indicate a higher soil water supply capacity and soil water retention capacity, respectively.

2.4 Statistical analysis

In this study, to compare the differences between BM and NM on soil water retention and soil properties, we conducted one-way analysis of variance (ANOVA) statistical tests to determine differences in plant and soil properties for the same soil layers between the crust and NM, and a least-significant-difference test (P<0.05) was conducted when significant differences were detected by ANOVA. To explore the relationship between soil properties and soil-soil water retention, and quantitative evaluation of the effects of soil properties on soil-soil water retention, Pearson's correlation and variance partition in the analysis were used by R software version 3.4.3 (R Development Core Team, 2006) with the "hier.part" and "corrplot" packages. Furthermore, structural equation modeling was used to examine the soil properties' direct and indirect effects on soil water retention.

3 Results

3.1 Soil particle size distribution among two surface soil types

Silt content dominated the soil particle size distribution in the 0–40 cm soil layer across the two surface soil types (mean 61.69%), followed by sand (mean 30.13%), and clay (mean 8.18%) (Fig. 2). Specifically, the 0–10 cm soil layer's clay content in BM was 9% higher than that in NM, whereas the 10–40 cm soil layer's clay content in BM was 16% lower than that in NM, especially

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for the 10–20 cm soil layer (P<0.001). In contrast, the 0–40 cm soil layer's silt content in BM was higher than that in NM, especially for the 20–30 cm soil layer (P<0.05). However, no clear pattern was observed for the sand content between BM and NM. Overall, in the 0-40 cm soil layer, clay content (8.62%) in NM was 11% higher than that in BM (7.69%), whereas in the 0-40 cm soil layer, silt content (61.24%) in NM was nearly equal to that in BM (62.13%).

3.2 Soil physicochemical properties among two surface soil types

There were no significant differences for 0–40 cm BD, 0–40 cm TP, 0–40 cm CP and 0–40 cm NCP (P>0.05) (Fig.3), but the 0–20 cm BD in NM was 13% lower than that of BM, and the TP and CP in NM were 7% and 5% higher than that of BM. No clear pattern was observed for NCP in NM and BM (Fig.3). Furthermore, the 0–20 cm TN and SOM in NM were much higher than those in BM and reached a significant level at 0–10 cm (P<0.05), whereas the 30–40 cm TN and SOM in NM were lower than those in BM (Fig.3). Similarly, the 0–10 cm soil layer's TC and C: N ratio in NM were significantly higher than those in BM, whereas the 30–40 cm soil layer's TC and C: N ratio in NM were lower than those in BM (Fig.3). Additionally, the 0–40 cm soil layer's MBC and MBN in NM were higher than those in BM and reached a significant level at 0–10 cm (P<0.05) (Fig. 4).

3.3 Soil hydrological properties among two surface soil types

The soil hydrological properties varied between crust BM and NM (Fig.5 and Table 1). Given that parameter A fitted by the Gardner model represents the soil water retention (a higher A value indicates higher soil water retention), the soil water content was reduced with decreasing matric potential and reduced sharply at high matric potential but remained stable at low matric potential (Fig. 5), the 0–30 cm layer's soil water content and soil water retention in NM were higher than

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those in BM, whereas the 30–40 cm layer's soil water content and soil water retention in NM were lower than those in BM (Table 1 and Fig. 6b). Similarly, the 0–10 and 20–30 cm layers' soil water supply capacity (i.e., A*B fitted by the Gardner model) in NM was higher than that in BM, while the 10–20 and 30–40 cm layers' soil water supply capacity in NM was lower than that in BM (Fig. 6a). Furthermore, the surface infiltration rate in the BM was significantly lower than that in the NM (Table 1).

3.4 Dominated factors affecting soil-soil water retention

Pearson correlation analysis showed that soil water retention was significantly negatively related to BD, but significantly positively related to TP, CP, and SOM (Fig.7a), whereas soil particle size distribution exerted weak soil water retention (Fig.7a). Furthermore, the variance partition showed that SOM explained the greatest variability in soil-soil water retention (24.40%), followed by CP (21.24%), BD (18.22), and TP (18.22%) (Fig. 8b), and structural equation modeling showed that the effect of SOM on soil water retention was achieved by altering CP and BD (Fig. 8).

4 Discussion

4.1 Effect of biocrust on soil properties

The effects of biocrust on soil properties have been widely explored in previous studies (Guo et al., 2008; Liu et al., 2019), and most studies conducted in arid regions have found that the presence of biocrust could improve soil aggregation and stability (Wu et al., 2020), increase soil fertility (Zhou et al., 2020), and reduce soil erosion (Chamizo et al., 2017). In this study, however, we found that the presence of biocrust could improve topsoil texture, but not that of deep soil. The 0–10 cm soil layer's clay content in BM was higher than that for NM, whereas the 10–40 cm soil layer's clay content in BM was lower than that for NM, which is in line with previous studies conducted in arid

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and semi-arid regions (Liu et al., 2016; Wu et al., 2020). The higher clay content in BN was attributed to the exudation and cohesiveness of the biocrust, which promoted clay and silt formation and reduced sand content (Wang et al., 2021). Furthermore, we found that the 0–20 cm soil layer's soil bulk density in NM was higher than that in BM, thereby leading to higher soil porosity and total capillary porosity in NM. The higher soil capillary porosity in NM was mainly attributed to its higher soil organic matter content, which was also confirmed by the significant positive relationship between soil organic matter and soil capillary porosity (Fig. 7). It has been well documented that a higher soil organic matter could improve soil aggregation and stability and subsequently increase soil capillary porosity (Cui et al., 2021).

Moreover, an increasing number of studies have found that the presence of biocrust can also improve soil nutrient conditions in the process of mobile sand fixation (Belnap et al., 2004; Guo et al., 2008; Li et al., 2005a). In comparison, we found that the presence of biocrust reduces the 0–10 cm layer's soil total carbon, total nitrogen, and C: N ratio, which is in contrast to most previous studies conducted in arid and semi-arid regions, where soil nutrient conditions were improved under biocrust (Chamizo et al., 2012b; Zhao et al., 2010). A probable reason for these differences may be environmental differences. Considering that the formation of biocrust is a changing process from simple to complex in its morphology, the early cyanobacteria crust was formed only under favorable hydrothermal conditions such as temperature, soil water, solar radiation, and nutrient content (Belnap et al., 2004; Li et al., 2005b). For instance, biocrust is metabolically active when the external environment is wet, and its metabolically active environment is sensitive to temperature (Belnap et al., 2004; Li et al., 2005b), otherwise the biocrust may choose to enter the dormant stage when the external environment is under unfavorable conditions. Therefore, compared to the higher temperatures in arid and semi-arid lands, the biocrust in alpine ecosystems may be in a dormant stage owing to its lower temperature and less available nutrients. Moreover, the biocrust in our study was mostly dominated by cyanobacteria crust, which was vulnerable to external disturbances such as grazing activity; thus, the biocrust may choose dormancy when it is subjected to grazing pressure, which was confirmed by the significantly lower microbial soil carbon and microbial soil nitrogen content (Fig. 4).

4.2 Effect of biocrust on soil hydrology and their underlying mechanisms

In this study, we found that soil water infiltration was greatly reduced in BM compared with that in NM, which was consistent with the results of a previous study conducted in alpine meadows (Li et al., 2016b). However, it is in contrast to other studies conducted in cool desert ecosystems where biocrust increased soil water infiltration and reduced runoff by increasing soil porosity and aggregate stability (Kidron and Benenson, 2014; Wei et al., 2015). These discrepancies were associated with soil texture and biocrust developmental stage. In general, soil water infiltration in coarse-textured soils is higher than that in fine-textured soils owing to its large pores compared with the narrow pores in fine-textured soils, which reduces the movement of water into the soil (Belnap, 2006). However, we found that the establishment of biocrust increased clay content and subsequently reduced soil macropores, which hindered soil water infiltration. Therefore, we can conclude that the soil in the BM may be more vulnerable to runoff generation and consequent soil erosion, owing to its lower soil water infiltration and soil water retention capacity. On the other hand, biocrust can reduce available pore spaces for water to infiltrate by clogging the soil surface conductive pores owing to its higher water absorption and swelling of biocrust (Fischer et al., 2010), and consequently reduce soil infiltration. In addition, soil water infiltration was altered by the

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developmental stage of the biocrust in homogeneous soil. A previous study indicated that soil hydraulic parameters differed significantly between cyanobacterial biocrust and moss biocrust (Wang et al., 2017). For instance, Chamizo et al. (2012a) reported that the incipient-cyanobacterial crust had a lower soil infiltration rate than that of the cyanobacterial crust, whereas the dark-colored mosses' crust had higher surface soil infiltration capacity by increasing macroporosity and unsaturated hydraulic conductivity in the grasslands dominated by A. splendens (Jiang et al., 2018). In our study, the biocrust was dominated by incipient-cyanobacterial crust, which had low biological activity and low porosity owing to the predominance of vesicle pores, thereby leading to a lower soil infiltration rate.

Furthermore, the soil-soil water retention and soil water supply capacity varied significantly between the biocrust and normal meadows. We found that in the 0–10 cm soil layer, soil water retention and soil water supply capacity in NM was higher than that in BM, which was in contrast to the results of previous studies conducted in drylands in which biocrusts enhanced surface soil water retention capacity and water availability (Sun et al., 2022). We speculate that the lower soil water retention in the BM was due to lower soil organic matter; this was verified by the presence of lower microbial biomass carbon (Fig. 4a). The structural equation model indicated that the effect of soil organic matter on water retention was mainly achieved by altering soil bulk density and soil porosity (Fig. 8) because higher soil organic matter could reduce soil bulk density and increase soil porosity (Liu et al., 2019), leading to higher soil water retention, which also confirmed a significant positive relationship between soil organic matter and soil water retention (Fig. 7). Soil organic matter was derived from vegetation litter and root biomass, whereas the vegetation litter in BM was lower than that in NM owing to its lower aboveground biomass and vegetation coverage, ultimately

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resulting in lower soil organic matter in BM.

4.3 Implications for the role of biocrust in alpine meadows

Grassland ecosystems cover more than 60% of the QTP and provide important ecosystem services, such as biodiversity conservation, carbon storage, and water conservation (Dong et al. 2020). However, in recent decades, grasslands in the QTP have suffered from serious degradation due to increasing human activity (Cao et al. 2019). Biocrust is an important surface feature of the degraded alpine meadows. It is acknowledged that biocrust has a positive effect on soil nutrient and soil water content retention in arid regions. In contrast, we found that the presence of biocrust decreased soil water retention and soil infiltration rate, which did not improve water conservation in alpine meadows. Therefore, the soil in the biocrust region may be more vulnerable to runoff generation and consequent soil erosion. Moreover, soil nutrients, such as SOM, TC, and TN, were reduced significantly in the biocrust meadow, suggesting that the growth of vegetation in the biocrust region may be limited by soil nutrients. Considering the negative effects of biocrust on alpine meadows, some steps should be taken to reduce the formation of biocrust in degraded alpine meadows, such as reducing grazing intensity.

5 Conclusions

Soil hydrological properties were significantly affected by the establishment of biocrust, and we found that the biocrust could retain topsoil water and infiltrate topsoil, which suggested that the establishment of biocrust did not favor soil hydrological properties in alpine meadows, and the soil in the BM might be more vulnerable to runoff generation when a heavy rainfall event occurs. Furthermore, the presence of biocrust increased topsoil clay content, while the 0–30 cm layer's soil capillary porosity in NM was higher than that in BM, indicating that the presence of biocrust reduced

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soil porosity and thereby reduced topsoil water infiltration. We thus concluded that the discrepancies in soil water retention and topsoil infiltration were close to physicochemical properties, and that SOM plays a role in soil water retention by affecting CP and BD. Our study provides insight into the role of biocrust in soil hydrological processes in alpine ecosystems.

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Author contribution

Licong Dai: Investigation, Data curation, Writing – original draft, Formal analysis. Ruiyu Fu : Investigation, Data curation, Writing – original draft, Formal analysis, Visualization. Xiaowei Guo and Zhongmin Hu: Investigation, Data curation, Project administration, Supervision. Yangong Du: Writing – original draft, review & editing. Guangmin Cao: Conceptualization, Methodology, Funding acquisition, Supervision.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper

Data availability statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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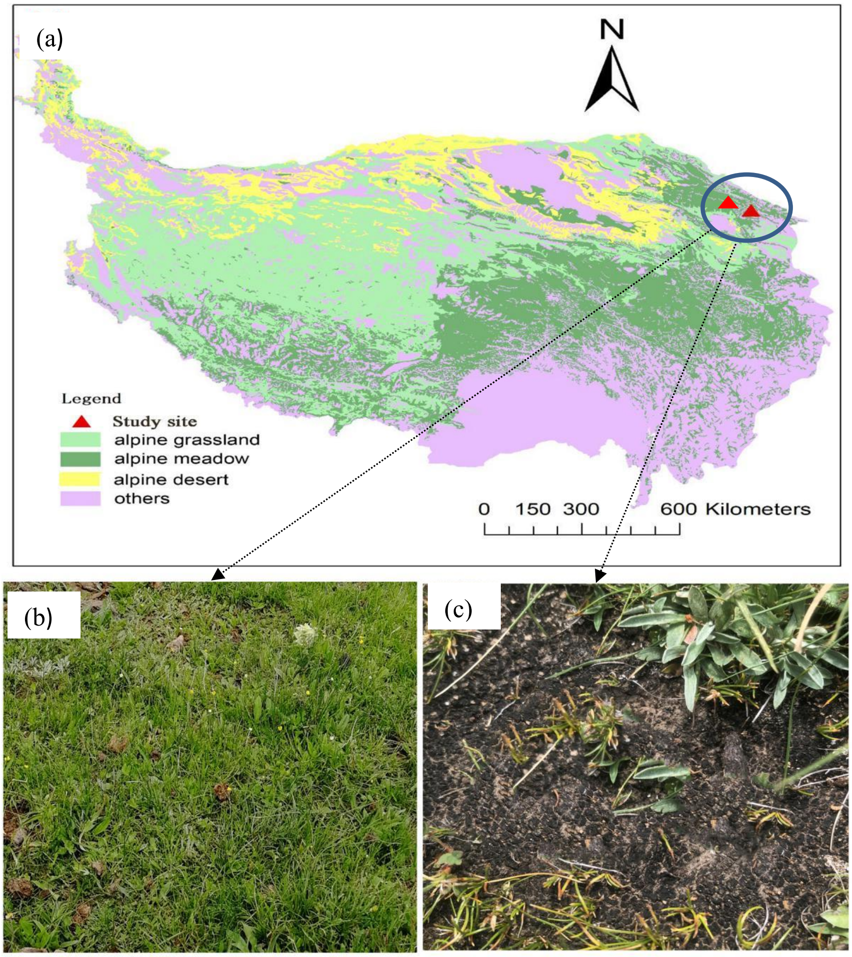

Fig.1 The study site (a) and two type meadows in this study: normal *Kobresia* meadow (b) and biocrust meadow (c)

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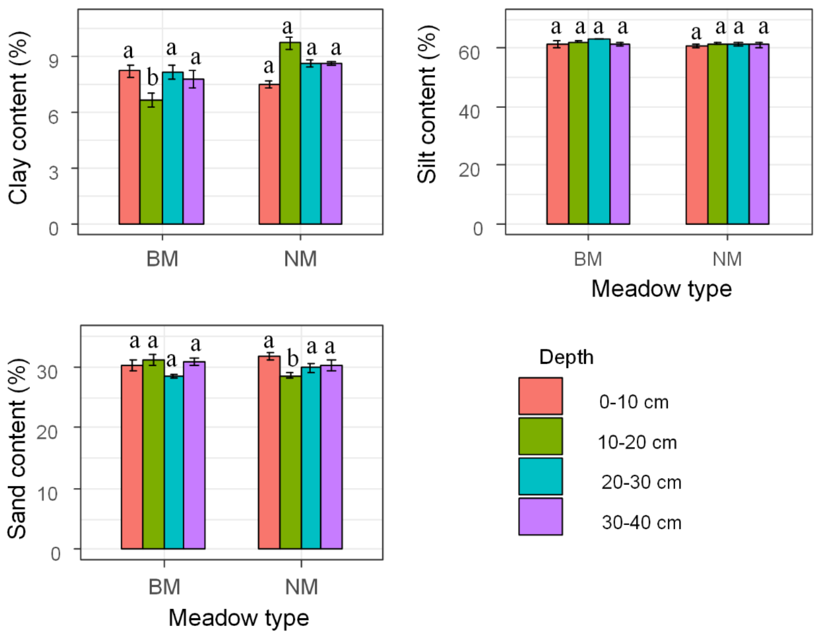

Fig.2 Soil particle size distribution among two surface soil types. Note: NM, normal *Kobresia* meadow; BM, biocrusts meadow, the different letters mean significant differences (P<0.05) between normal *Kobresia* meadow and crust meadow at the same soil layer

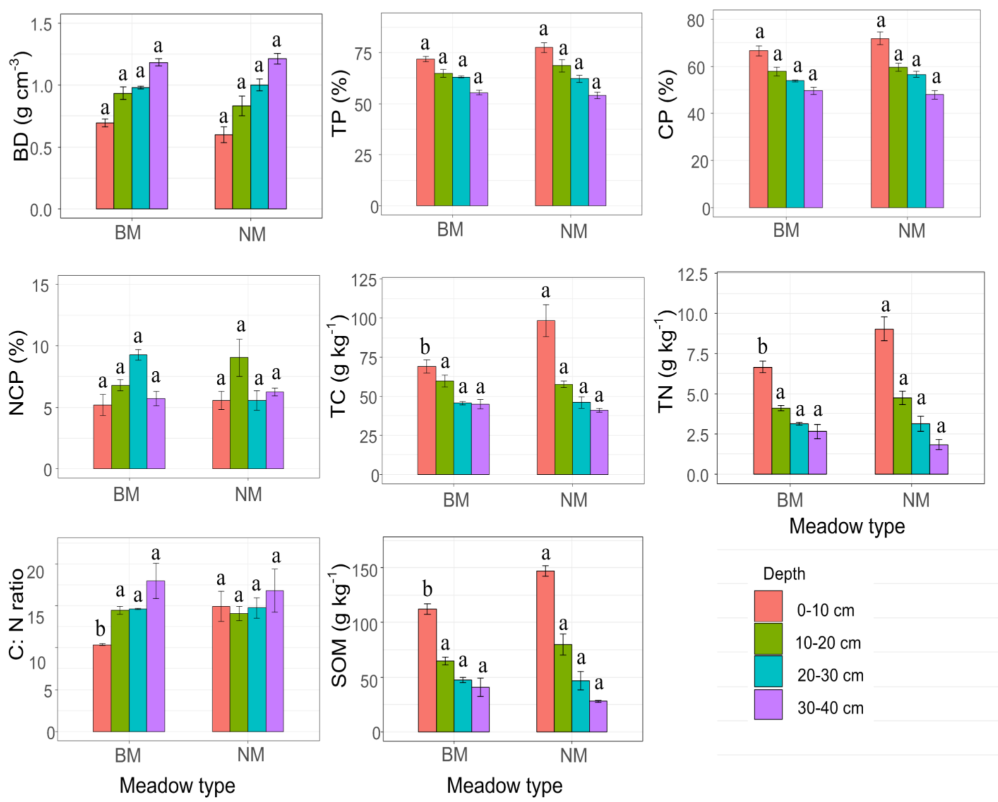

Fig.3 The soil physicochemical among two surface soil types, BD: soil bulk density, TP: soil total porosity, CP: soil capillary porosity, NCP: non-capillary porosity, TN: soil total nitrogen, TC: soil total carbon, C:N: soil C: N ratio, SOM: soil organic matter, the different letters mean significant differences (P<0.05) between normal *Kobresia* meadow and crust meadow at the same soil layer

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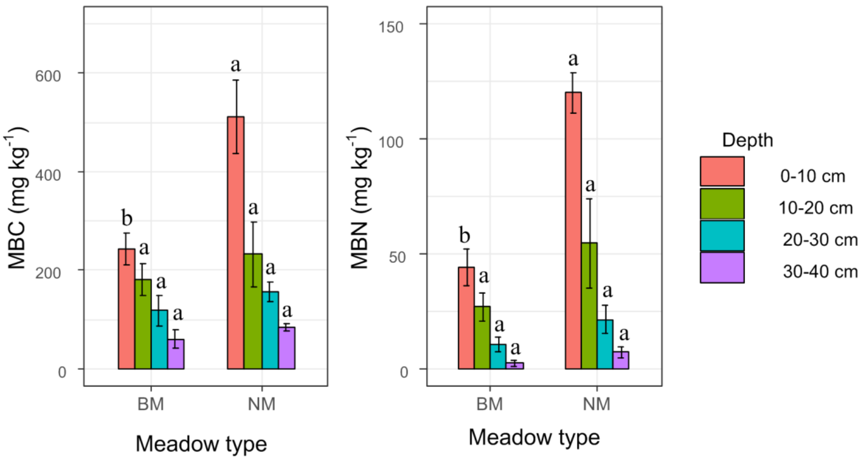

Fig. 4 Soil microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN) across two type meadow, the different letters mean significant differences (P<0.05) between normal *Kobresia* meadow and crust meadow at the same soil layer

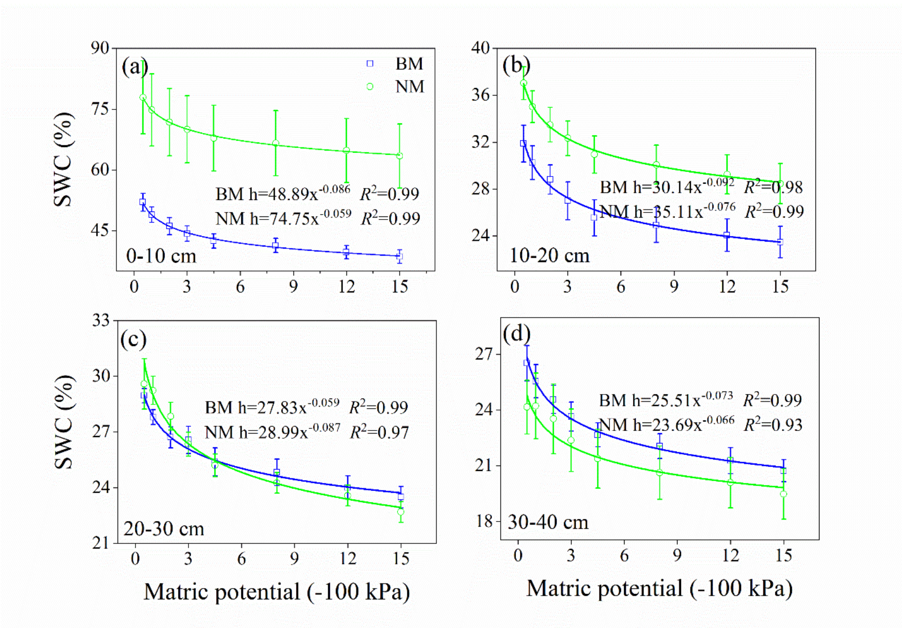

Fig.5 Soil water retention curve of different soil layer (a: 0-10 cm, b: 10-20 cm, c: 20-30 cm, d: 30-40 cm) across two type meadow between soil water content (SWC) and matric potential. Note: NM, normal Kobresia meadow; BM, biocrusts meadow, the soil water retention curve was fitted by

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Gardner model (i.e. h = Aθ^{-B}), A and B are the fitting parameters; a higher value of A indicated a higher soil water-holding capacity.

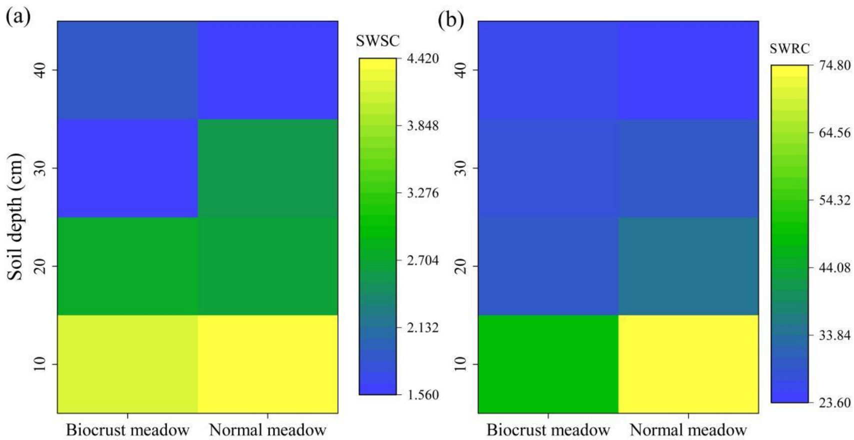

Fig.6 Soil water supply capacity (SWSC) (a) and soil water retention capacity (SWRC) of different soil layer across two type meadows, the SWSC was represent the A*B from Gardner model, the SWRC represent the A from Gardner model, a higher value of A*B and A indicated a higher soil water supply capacity and soil water retention capacity, respectively.

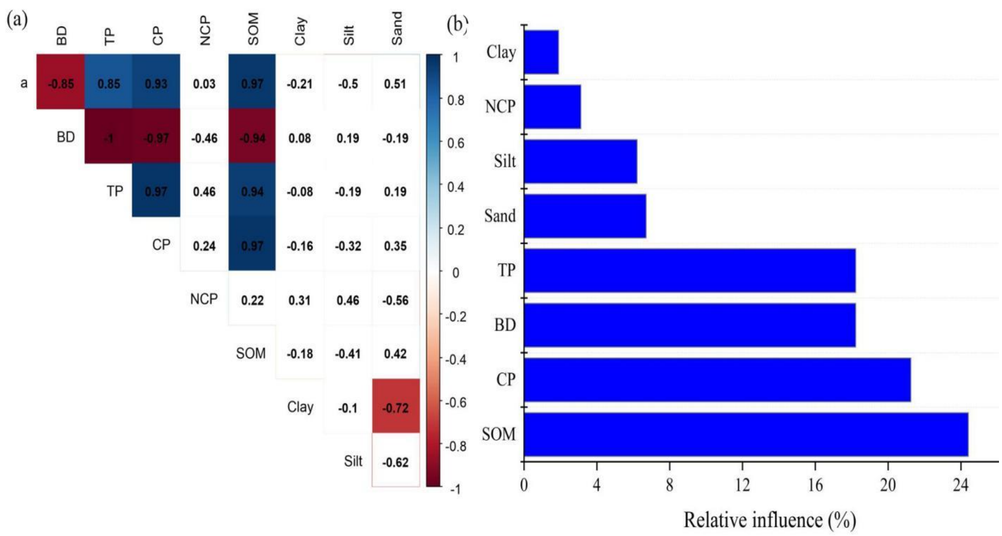

Fig. 7 Pearson correlation between soil water retention and soil properties (a), and the relative

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influence of soil properties on soil water retention. Note: "*", "**"and "***" indicated significant at 0.05, 0.01 and 0.001 level, respectively. Note: a: the parameter fitted by Gardner model, BD: soil bulk density, TP: soil total porosity, CP: capillary porosity, NCP: non-capillary porosity, SOM: soil organic matter.

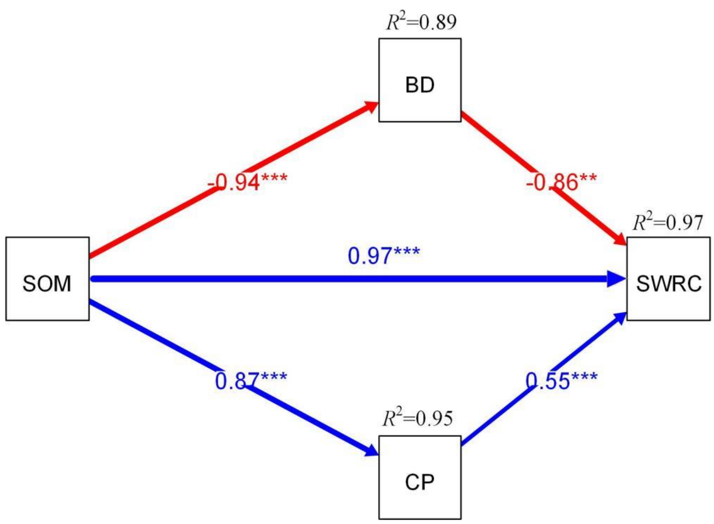

Fig. 8 Structural equation modeling of the direct and indirect effects of soil properties on soil water retention capacity (SWRC). Standardized path coefficients, adjacent to arrows, are analogous to partial correlation coefficients, and indicative of the effect size of the relationship. Continuous blue and red lines represent positive and negative correlations, respectively. Model fit: Fisher.C=5.48, *df*=2, *P*-value=0.065.