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Wenjie Lu  
*China Agricultural University, China*

Yingjun Zhang  
*China Agricultural University, China*

Liu Nan  
*China Agricultural University, China*

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The 23rd International Grassland Congress (Sustainable use of Grassland Resources for Forage Production, Biodiversity and Environmental Protection) took place in New Delhi, India from November 20 through November 24, 2015.


Published by Range Management Society of India

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Changes in stable fraction of soil organic carbon and microbial structure in response of grazing intensity

LU Wenjie, Zhang Yingjun, Liu Nan
China Agricultural University, Beijing, China
*Corresponding author e-mail: zhangyingjun71@hotmail.com

Keywords: Carbon sequestration, Grazing intensity, Microbial structure

Introduction
Carbon sequestration is one of the most important ecological functions of the grassland. Soils contain more carbon than in vegetation in the form of soil organic matter (SOM). But grassland soils are vulnerable to disturbances caused by human activities. Grazing may result in various disturbances to both soil and vegetation under different grazing intensities (Bai et al., 2012). Grazing led to the shifts in amount and composition of soil organic matter. In previous studies, the chemical methods were used to separate soil into the different pools according to the stability of SOM (Carvalho Leite et al., 2004), but they cannot be convicive for their chemical destruction upon the SOM structure. In this study, we use more credible fractionating method to separate stable part of the SOM based on the dry sieving and winnowing (Kirkby et al., 2011).

Stable carbon fraction has two origins: microbial debris and recalcitrant plant materials (Lehmann et al., 2007). Evidences have showed that soil microbial community alters metabolic rates by changing its composition adjust to stoichiometry of substrates. We hypothesize that carbon content stable fraction SOM in the surface soils would decline as the intensity increasing, but the stoichiometry of carbon and nutrients (N and P) would remain constant. We also predict that the relative importance in carbon sequestration of microbial groups would change due to the soil environmental alteration. The objectives of the study is to evaluate the effects of grazing intensity (GI) on carbon in more stabilized SOM and on soil microbial community structure in steppe.

Materials and Methods
This study was conducted in the permanent fields in the Inner Mongolia Grassland Ecosystem Research Station (IMGERS), located in the Xilin River Basin, Inner Mongolia, China (43°38’ N, 116°42’E). The growing season is from early April to late September for perennial plants, whereas annual plants usually germinate in early July after rains. Mean annual temperature is 0.4°C, with the lowest -21.4°C in January and the highest 19.0°C in July. Mean annual precipitation is 336.9 mm yr⁻¹, about 80% occurring in the growing season (May–August). The dominant soil types are typical chestnut and dark chestnut. Plants in the plot of light intensity consist of tall perennial grasses such as Leymus chinensis, Stipa grandis and Achnatherum sibiricum, while ones in heavy intensity are dominated by species of less standing biomass such as Carex korshinskii, Cleistogenes squarrosa.

To evaluate the effects of sheep grazing intensity on a typical steppe, a grazing experiment, including 6 intensities from low (1.5 sheep/hm²) to high (6 sheep/hm²) to overgrazing (9 sheep/hm²), was designed based on the herbage allowance method in 2005. Soil samples were taken from the upper layer (0-5 cm) of each treatment in 2013, and were crumbled and sieved through 2 mm mesh sieves to remove plant material and stones. One part of the samples was air-dried at the room temperature for 2 weeks for chemical analysis, while the other was frozen in -80°C refrigerator for soil microbial community analysis.

The fractionating method was a modification of the procedure based on dry sieving and winnowing (Kirkby, 2011). The SOM was separated into two fractions of fine fraction (FF) and coarse fraction (CF). Hereafter in this study, we mainly focused on the carbon content of fine fraction (FFC) altered by grazing as well as its stoichiometry with nitrogen (FFN) and phosphorus (FFP). The soil microbial composition PLFA was analysed that the fatty acids i-15:0, a-15:0, i-16:0, 16:1w7c, 18:1w7c and cy-19:0w7c represented the total bacteria, and i-15:0, a-15:0 and i-16:0 gram-positive bacteria, and 16:1w7c, 18:1w7c and cy-19:0w7c gram-negative bacteria, and 18:2w6c and 18:1w9c fungi. Data analyses were done with SAS V8.0 and SigmaPlot 12.0.
Results and Discussion

The FFC of all soils decreased exponentially (Fig. 1) when the intensity increased from 1.5 sheep/hm$^2$ to 9 sheep/hm$^2$. The negative correlation between FFC and grazing intensity could be described as following equation ($n=90, F_{2,88}=75.56$):

$$y=1.384*\exp(0.9450/(x+0.0606))$$

$x$, $y$ stands for grazing intensity and FFC content, respectively.

![Fig. 1: The relationship between FFC and grazing intensity.](image)

Changes of nitrogen (N) and phosphorus (P) in fine fraction soils showed consistency with the result of FFC ($p<0.0001$), keeping homeostasis in carbon-nutrient stoichiometry, which indicated that our fractionating procedure run well. The mean FFC:N:P ratio was nearly 100 : 10.48 : 1.19 regardless of grazing intensity. Compared with the stoichiometric ratio in Australia soil (100:8.33:2) described by Kirkby et al., 2011), it contains more nitrogen and less phosphorus in grazed fields of grassland. Based on the stoichiometric ratio, the phosphorus limits carbon sequestration in this grazing steppe.

Grazing intensities give an impact on soil microbial groups as the PLFA profiles show (Fig. 2). Fungi and gram-positive PLFAs exhibit quadratic curve with increasing grazing intensity ($P<0.01$). Gram-negative decline linearly with increasing of GI ($P<0.01$). The molecular ratio of fungi and bacteria PLFAs also decrease ($P<0.01$), which mean that fungi own the higher rate of reduction. The relationship between GI and microbial PLFAs suggest that the soil environment was unfavorable to growth of fungi and bacteria, and the fungi were more sensitive. In previous studies, soil microbial communities dominated by fungi are linked to more slow-growing plants that allocate more photosynthetic products in roots, then hence sequestrate more carbon in soil (De Deyn et al., 2008). Slow-growing plants’ litters in the light grazing intensities are more recalcitrant to the soil microorganism. It was tested that the microbial debris and recalcitrant plant materials were main sources of more stable carbon in the soil, and were confirmed to be mediator for the changes in FFC in this study.
Fig. 2: Relationship between grazing intensity and microbial community
Conclusion
Increasing grazing intensity takes the negative effects on the accumulation of stable fraction carbon in soil, which was related to the alterations of vegetation and soil microbial structure. Therefore, the grazing intensity should be given attention for soil carbon sequestration management.

References


Acknowledgement
Research was supported by the earmarked fund for the 973 project (2014CB138805), and the Fundamental Research Funds for the Central Universities (2014XJ005).