

Soil organic matter fractions and aggregate distribution in response to functional composition of plant communities

Las fracciones de materia orgánica del suelo y la distribución de agregados en respuesta a composición funcional de comunidades de plantas

Fracionamento de matéria orgânica do solo e distribuição agregada em resposta a composição funcional das comunidades vegetais

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Abstract

Vegetation types are amongst the most relevant pedogenic factors controlling soil organic carbon. The impact of shrub encroachment (SE) on soil organic matter (SOM) of alpine grasslands has scarcely been investigated. We aimed to compare the effects of shrub invasion on soil carbon, nitrogen and organic matter in three representative communities. Three dominant plant communities representing low, intermediate and high SE were selected in alpine grassland habitats in north-western Iran. To compare the SOM parameters, soil samples from each community were collected from depths of 0-15 cm and 15-30 cm. The results showed that the spatial variation of SOM was probably affected by the shrub invasion. The highest values of total C and total N were observed in high SE (*Pteropyrum aucheri* community) and intermediate SE (*Astragalus microcephalus* community) as compared to low SE (*Prangus uloptera* community). Soil particulate organic matter for carbon and nitrogen were generally highest in the intermediate SE. There was a significantly smaller proportion of

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soil present as macro-aggregates and higher proportion of soil present as micro aggregates in the upper soil layer of intermediate SE as compared to the others, while no significant difference between micro- and macro-aggregates in the lower soil layer between plant communities were detected. Shrub functional groups in grassland communities should be considered in the interpretation of shrub encroachment influences on soil organic matter.

Key words: carbon sequestration, grasslands, shrub invasion, soil organic carbon, vegetation type.

Resumen

Los tipos de vegetación se encuentran entre los factores pedogénicos más relevantes que controlan el carbono orgánico del suelo. El impacto de la invasión de arbustos (IA) sobre la materia orgánica del suelo (MOS) de los pastizales alpinos apenas ha sido investigado. El objetivo fue comparar los efectos de la invasión de arbustos sobre el carbono, el nitrógeno y la materia orgánica del suelo en tres comunidades representativas. Tres comunidades de plantas dominantes que representaban SE bajo, intermedio y alto fueron seleccionadas en hábitats de pastizales alpinos en el noroeste de Irán. Para comparar las variables de MOS, se recogieron muestras de suelo de cada comunidad desde profundidades de 0-15 cm y 15-30 cm. Los resultados mostraron que la variación espacial de MOS probablemente se vio afectada por la invasión de arbustos. Los valores más altos de C total y N total se observaron en SE alto (comunidad de *Pteropyrum aucheri*) e SE intermedia (comunidad de *Astragalus microcephalus*) en comparación con SE bajo (comunidad de *Prangus uloptera*). La materia orgánica particulada del suelo para el carbono y el nitrógeno fue generalmente más alta en el SE intermedio. Hubo una proporción significativamente menor de suelo presente como macroagregados y una mayor proporción de suelo presente como microagregados en la capa superior del suelo del SE intermedio en comparación con los otros, mientras que no hubo diferencias significativas entre los micro y macroagregados en la capa inferior de suelo entre las comunidades de plantas. Se deben considerar los grupos funcionales de arbustos en las comunidades de pastizales en la interpretación de las influencias de invasión de arbustos sobre la materia orgánica del suelo

Palabras clave: secuestro de carbono, pastizales, invasión de arbustos, carbono orgánico del suelo, tipo de vegetación.

resumo

Los tipos de vegetação se encaixam entre os fatores pedogénicos mais relevantes que controlam o carbono orgânico do solo. O impacto da invasão de arbustos (IA) sobre a matéria orgânica do solo (MOS) dos alpes pastosos apenas ha sido investigado. O objetivo é comparar os efeitos da invasão de arbustos sobre o carbono, o nitrogênio e a matéria orgânica do solo em tres comunidades representativas. As comunidades de plantas dominantes que representam a

região, intermediário e alto nível selecionaram em habitats de pastizales alpinos no noroeste de Irã. Para comparar as variáveis de MOS, se recogieron muestras de suelo de cada comunidad de profundidades de 0-15 cm e 15-30 cm. Os resultados são mostrados na variação espacial de MOS e provavelmente são afetados pela invasão de arbustos. Os altos e máximos de C total e N total se verificando em SE alto (comunidad de *Pteropyrum aucheri*) e SE intermedia (comunidad de *Astragalus microcephalus*) en comparación con SE bajo (comunidad de *Prangus uloptera*). A matéria orgânica particulada do solo para o carbono e o nitrogênio é geralmente considerada alta na região intermediária. Hubo una proporción significativamente menor de suelo Presente Como macroagregados y una mayor proporción de suelo Presente Como microagregados en la capa superiores del suelo del SE intermedio en comparación con los otros, mientras que no hubo diferencias significativas entre los micro y macroagregados en la capa de inferior entre as comunidades de plantas. O enfraquecimento dos grupos funcionais de arbustos em comunidades de pastéis na interpretação das influências de invasão de arbustos sobre a matéria orgânica do solo

Introduction

Grasslands are globally important for soil organic matter (SOM) and carbon sequestration, contributing more than 10% to the total biosphere carbon store Scurlock and Hall (1998). Altered management practices, land use change and climate changes not only strongly influence on grasslands structure, but may also potentially alter ecosystem function including carbon sequestration and total nitrogen (Guo and Gifford, 2002; Jones and Donnelly, 2004). The magnitude of SOM has been shown to be influenced by grassland vegetation types as well as by the specific plant (Turetsky et al., 2005). Also within the same plant communities, plant compositions can result in different SOM (Erfanzadeh et al., 2014). This might suggest that different communities' values were differently impacted upon by parameters like diversity and plant functional composition. The relation between diversity and soil carbon and nitrogen content has been well investigated in several studies (Fornara et al., 2009; Li et al., 2018). In this sense, Motamedi and Souri (2016) found that the numerical indexes of diversity used (heterogeneity including Shannon-

Wiener and Simpson, Pielou index of evenness, richness indices of Margalef and Menhinick) were not efficient to reflect the distribution of the species, and how these indexes they did not allow to explain the results in ecological terms, they recommended applying numerical and parametric indexes of biodiversity (diversity ranking curve, species abundance-ranking curve and species distribution models (broken stick, normal log, log series and geometric series).

The encroachment of woody plants into grasslands (shrub encroachment: SE), has been widely reported as a global phenomenon and is one of the major changes in plant functional composition (Maestre et al., 2009; Guido et al., 2017). Several factors including overgrazing, increases in atmospheric CO₂ and fire frequency could be considered as driver of shrub encroachment in grasslands (Archer, 2010). Shrub invasion can increase soil carbon and nitrogen of grasslands mainly in locations with higher precipitation (Eldridge et al., 2011). Furthermore, studies on the relationship between SOM and woody plants in natural habitats have mainly

in locations with higher precipitation (Eldridge et al., 2011). Furthermore, studies on the relationship between SOM and woody plants in natural habitats have mainly focused on total SOM and soil fertility (Hagen-Thorn et al., 2004). Shrubs can strongly influence habitat conditions. Therefore, we studied the impacts of shrub encroachment on soil organic matter of three plant communities with different levels of shrub invasion according to their shrub cover representing low, intermediate and high shrub encroachment. Plant functional composition of all three sites contained entries for different functional groups including grasses, forbs and shrubs. We aimed at an evaluation of changes in soil carbon and nitrogen, when plant compositions were varied between sites. We hypothesized that variation in SOM values assigned to certain grassland type are in part due to variable plant functional composition at different sampling sites.

Material and methods

Description of the study area

Sub-alpine dry grasslands are the main vegetation types occurring in the north-western Iran. This habitat contains mainly calcareous rich grasslands with high diversity, which has been used for domestic grazing since centuries. Dry (sub-) alpine calcareous rich grasslands occur throughout the Alborz and Zagros mountains including north-western Iran. Grazing, mainly by sheep, is the dominant management on this habitat. Along with disturbance it favoured either less palatable species or less competitive ones (Mahdavi et al., 2013; Noroozi et al., 2014). The study was conducted in Khanghah watershed between latitudes 37°46'18" N and 37°50'42" N and longitudes 44°57'04" E and 45°00'32" E.

Three main plant communities with similar topographical conditions were selected according to their SE levels in the study were including *Astragalus microcephalus*, *Pteropyrum aucheri* and *Prangus uloptera* as dominant species (table 1). Low, intermediate and high SE covered 12.4, 23.9 and 43.5% woody species, respectively. The communities were distinguished and named according to the dominant plant species (Heady and Child, 1994; Xu et al., 2018).

Vegetation and soil sampling

Two key areas were selected within each plant community. The key area can be considered to be representative of the entire community in that location (Heady and Child, 1994). Soil samples from each key area were collected from depths of 0 to 15 cm and 15 to 30 cm. These sampling depths are in accordance with the highest presence of root biomass in temperate grasslands (Reeder et al., 2001). In each key area, six transects were established, three parallel

and three perpendicular to the slope. Along each transect, three 4 × 4 meter quadrats were established at the two ends and in the middle of each transect. The percentage cover of all species was recorded visually. In addition, with an auger (diameter: 5 cm), 10 soil cores were collected at random to a depth of 30 cm in the quadrats. Each core was divided into two sub-cores (0-15 and 15-30 cm) and sub-cores were then pooled per depth for each core. All soil samples were immediately transferred to the cooled, insulated container for transport to the laboratory and were stored at 4 °C until processing. The samples were sieved, the roots and coarse gravel were removed, and the <5 mm soil was used to examine the effects of vegetation composition on soil parameters.

Soil and data analysis

Organic C was determined by Loss on Ignition in 600 °C, 4 h (Lal et al., 2001) and total soil N was assessed using the Kjeldahl method (Zagal et al., 2009). Particulate organic matter (POM) was determined by physical fractionation. Twenty-five grams of air-dried soil were dispersed with 100 mL of sodium hexametaphosphate. The soil solution mixture was shaken and poured over a 0.053 mm sieve with several deionised water rinses. The soil remaining on the sieve was back washed into a pre-weighed aluminium dish, ground and analyzed for C and N (Handayani et al., 2009). Aggregate size distribution was determined using wet sieving with screen diameters of 0.25 and 0.50 mm. Soils were submersed in water on the largest screen before sieving under water by gently moving the sieve 3 cm vertically 50 times over period of 2 min through water contained in a shallow pan. Material remaining on the sieve was transferred to an aluminium container and dried at 60 °C, then weighed and measured for C (Elliott and Cambardella, 1991). General linear model and post-hoc tests were used to compare the soil parameters between communities and between depths. Each soil parameter was introduced as a dependent variable while depth categories (0-15 and 15-30 cm) and SE categories were introduced as fixed factors.

Results and discussion

Plant functional compositions

Our grasslands contained different plant functional composition within different SE levels. Low SE contains mainly perennial forbs and grasses and also dwarfs half shrubs. In intermediate SE, legumes half shrubs and perennial forbs and grasses were increased as compared to the low SE. In high SE, cover of shrubs strongly increased compare to the low and intermediate SE. Total shrub cover increased about

twofold and threefold in the intermediate and high SE as compared to the low SE, respectively (table 1). The studied rangeland was characterized with four plant communities, the vegetation cover of *P. aucheri* and *A. microcephalus* was 55% and for *P. aucheri* and *P. uloptera* it reached 57% of plant cover (Motamedi and Souri, 2016). The reduction of light interception by the canopy of the bushes affected mainly the shrubs species due to its lower height and different leaf area, which does not represent any advantage for the caespitose grasses (Guido et al., 2017).

Total organic carbon and nitrogen content in soils

Total C in both 0-15 and 15-30 cm soil depths and also in 0-30 cm as pooled depth was highest in the high SE and lowest in the low SE (figure 1A and table 2). Total N in all layers including pooled, lower and upper depths were highest in the intermediate SE and lowest in the low SE (figure 1B and table 2).

Labile SOM fractions

Soil POM-C and POM-N were generally highest in the intermediate SE (figures 2A and 2B). Particulate organic matter-C (POM-C) contents in the intermediate, high and low SE were 2.9, 1.8 and 1.6 g·kg⁻¹, respectively, in the upper soil layer. Particulate organic matter-C (POM-C) content in the intermediate SE was 1.9 g·kg⁻¹ in the deeper soil layer and significantly higher than in low and high SE. Particulate organic matter-N (POM-N) contents in both depths of the intermediate and high SE were significantly higher than the low SE (table 2).

Aggregate distribution and carbon associated with aggregate size classes
There was a significantly smaller proportion of soil present as macro-aggregates in the intermediate SE compared to high SE and low SE in the upper soil layer. There was no

significant difference between the proportions of macro-aggregates of the communities in the lower soil layer (table 2). There was a significantly higher proportion of soil present as micro-aggregates in both intermediate SE and low SE as compared to high SE in the upper soil layer and no significant difference between the proportions of micro-aggregates of the different communities in the lower soil layer (table 2). Carbon content was significantly greater for each type of aggregate in the intermediate and high SE than low SE in the upper soil layer (table 2). Carbon content was significantly highest for both types of aggregate in the intermediate SE and lowest in low SE in the deeper soil layer (table 2). The two-way ANOVA results revealed that main factors ('SE levels', 'soil depth') and their interactions had significant effects on SOM parameters (table 1). According to their F-values, the factor most strongly influencing SOM was 'SE level'. Although 'soil depth' had a significant effect on SOM, too, its effect was less pronounced than that of SE levels.

The aim of this study was to explore the impact of different plant functional compositions on SOM of three different communities. Here, we found that not shrub encroachment itself but its functional groups and also the presence of other functional groups determine the total C and N in sub-alpine grasslands. Vegetation types generally have effects on soil C and N. Influence of management and cultivation has been considered in several studies in cultivated lands (Luan et al., 2010; Poirier et al., 2018). However, in natural and non-cultivated grasslands mainly vegetation types and soil properties play the major role (Oueslati et al., 2013). In the current study, results showed that the spatial variation of SOM in these natural habitats was affected by the plant community as well. The SOM generally increased with developing shrubs and perennial

grasses in the study area. This confirms that under natural conditions not only management attributes but also vegetation factors define soil SOM.

Shrub enhancement or shrub functional compositions

We generally know that shrub encroachment increase total C and N (Eldridge et al., 2011, Li et al., 2018). We also expected that communities might have higher C and N along shrub encroachment. However, our results showed that higher shrub invasion does not lead to higher N content. Therefore, we tried to explain the results according to the shrubs functional groups. In the low SE, dwarf half shrubs with high participation of perennial forbs were

observed. In the intermediate SE, legumes half shrubs and cushion half shrubs was dominated compare to the high SE with mainly open canopy shrubs and half shrubs (table 1). These functional compositions lead to different values of SOM. Firstly, high values of intermediate SE show that their dominant functional compositions including legumes and cushions half shrubs can accumulate more N and high C-levels in the soil. These results are consistent with studies that show legumes and cushion plants to better accumulate soil organic matter (Fornara and Tilman 2008; McClaran et al., 2008). Cushion species produce small sized spiny stems, which tend to die back almost to the base each winter. This, together with legume-shrub associated rhizobia fixing N₂ may increase total N in soils of these communities. Litter accumulation in the form of fertile 'islands' beneath woody plants in grasslands is a common phenomenon and provides opportunities for carbon and nitrogen sequestration in arid and semi-arid regions (Jackson et al., 2002).

In addition, perennial grasses also occurred more frequently in the intermediate SE as compared to other levels. These groups also increase soil carbon and nitrogen by producing high amount of fine root (Li et al., 2010, Yang et al., 2018). Furthermore, functional groups occurring in the high SE have lower N and non-significantly higher C than intermediate SE. Open canopy shrubs and dwarf half shrubs as a main group of this site have lower ability to accumulate nitrogen. This result is consistent with studies that show open canopy shrubs to have low potential for resources capture and nutrient cycling (Toranjzar et al., 2010, Yang et al., 2018). In addition, open canopy shrubs have deep root systems with high participation of macro aggregate, which leads to high available carbon in the soil. Total organic carbon and carbon in micro- and macro-aggregates and labile fractions of SOM confirmed our hypothesis of functional groups impacting on soil C- and N-relations. The highest value of micro-aggregates was observed in intermediate SE, indicating the spiny shrubs and fine root of perennial grasses increase total carbon and nitrogen. In addition, high values of POM-C and POM-N in intermediate SE also confirmed the role of plant functional composition. The change in vegetation from low to intermediate SE increased POM-C in topsoil (0-15 cm depth) by 81.3%. At the same time, total C in the upper layer was increased by 49.0%. By contrast, the variation range of total N was higher than POM-N after a change in the vegetation from low SE to intermediate SE. Thus, significantly different levels of

POM-C and POM-N between the three communities in this study suggest differences in root biomass.

Conclusion

In current study, the picture of total communities was considered and we tried to explain variation between grassland communities including different SE levels using plant functional compositions. However, the participation of each functional group in carbon and nitrogen were not considered. We tried to show that how this variation should be considered in content of shrub encroachment. Classification of woody plants to different plant functional groups according to their canopy shape, which affect litter and resources accumulation, root structure and aggregate conditions for future interpretation of natural habitat, is promising.

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Table 1. Cover percentage of plant functional groups in the three SE (shrub encroachment) levels.

| Functional groups/SE levels | Low SE | Intermediate SE | High SE | |
|-----------------------------|---------------------|-----------------|---------|-------|
| Arbustals | 7.96 | 8.63 | 8.10 | |
| Potential forbs | 23.08 | 4.00 | 3.23 | |
| Potential grasses | 6.99 | 2.41 | 1.77 | |
| Shrub functional groups | Shrubs | 0.08 | 1.66 | 37.49 |
| | Cushion half shrubs | 0.59 | 6.46 | 0.65 |
| | Dwarf half shrubs | 4.75 | 4.19 | 3.87 |
| | Legumes half shrubs | 7.04 | 11.53 | 1.48 |

Table 2. Main soil organic matter fraction and aggregate distribution (mean ± SE) in the 0-5 and 15-30 cm soil layers of the *Astragalus microcephalus* community compared to the *Pteridium aucheri* and the *Prangus uloptera* communities in the grasslands, north-western, Iran.

| Depth (cm) | SOC (g/kg) | TN (g/kg) | POM-C (g/kg) | POM-N (g/kg) | Macro-aggregates (%) | Micro-aggregates (%) | CMC-C (g/kg) | CMC-N (g/kg) | |
|-------------------------------------|------------|------------|--------------|--------------|----------------------|----------------------|--------------|--------------|-------------|
| 0-5 | High SE | 1.20(0.04) | 2.60(0.17) | 0.30(0.02) | 20.60(1.70) | 28.80(1.70) | 14.20(1.70) | 16.20(0.80) | |
| SE | 9.60(3.80) | 0.90(0.09) | 1.60(0.09) | 0.30(0.04) | 34.60(2.10) | 20.36(1.90) | 11.61(1.43) | 17.60(1.70) | |
| Low SE | 6.50(0.60) | 0.80(0.09) | 1.60(0.09) | 0.20(0.05) | 33.20(2.10) | 23.90(2.30) | 6.90(1.00) | 12.60(0.40) | |
| Intermediate | 8.30(0.50) | 1.20(0.05) | 3.80(0.06) | 0.30(0.05) | 33.20(0.80) | 23.70(0.80) | 11.60(0.50) | 13.60(0.80) | |
| 15-30 | High SE | 9.90(0.20) | 0.90(0.07) | 1.00(0.10) | 0.20(0.09) | 35.67(1.09) | 17.67(0.89) | 7.70(0.60) | 13.70(0.63) |
| Low SE | 3.60(0.30) | 0.70(0.06) | 1.10(0.12) | 0.10(0.03) | 35.61(1.21) | 21.11(0.74) | 3.81(0.26) | 10.70(0.25) | |
| Two-way ANOVA results (mean square) | | | | | | | | | |
| Community | 1.87(1) | 0.02(5) | 0.139(3) | 0.003(9) | 842.780(1) | 342.850(1) | 23.75(1) | 2.387(3) | |
| (C) | | | | | | | | | |
| Depth (D) | 0.27(9) | 0.00(2) | 0.176(3) | 0.003(9) | 438.612(1) | 548.303(1) | 14.32(1) | 1.177(3) | |
| C × D | 0.089(3) | 0.00(3) | 0.007(3) | 6.445(9) | 425.812(1) | 70.014(1) | 0.084(3) | 0.263(3) | |

Abbreviations: SOC= soil organic carbon; TN= total nitrogen; POM-C= particulate organic matter carbon; POM-N= particulate organic nitrogen; CMC= carbon in macro-aggregates; CMI= carbon in micro-aggregates; ns= not significant; *P<0.05; **P<0.01; within a column mean values with different letters a, b and c, in each soil depth are significantly different between communities at P<0.05 (Duncan's test at α=0.05).

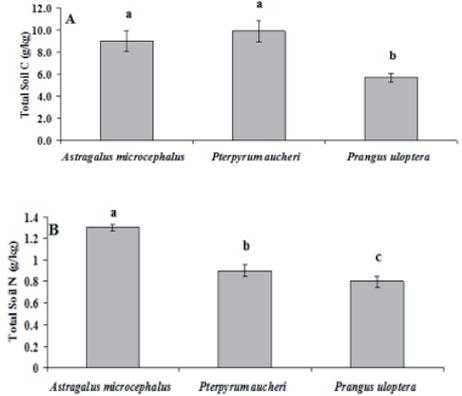


Figure 1. Total C (A) and total N (B) in the 0-30 cm depth in three communities. Different successive letters indicate significant differences (at α= 0.05) between the three communities.

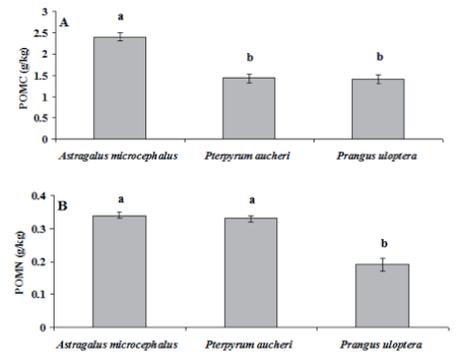


Figure 2. Particulate organic matter-carbon (A) and particulate organic matter-nitrogen (B) in 0-30 cm depth in three communities. Different successive letters indicate significant differences (at P<0.05) between the three communities.