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Large macroaggregates determine distribution of soil amidohydrolase activities at different landscape positions

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ABSTRACT

Microbial activities have been shown to be regulated by soil aggregation. However, it is unknown how landscape-induced changes in the soil structure shape microbial activity in the soil ecosystems. This study aims to analyze the spatial distribution of amidohydrolase activities within the soil aggregates obtained from different landscape positions. Soil samples were collected from five landscape positions (summit, shoulder, backslope, footslope, and toeslope) at two pasture sites with contrasting climatic condition. Soil organic C (SOC) content, total N (TN), urease (URS), L-glutaminase (LGL), and L-asparaginase (LAS) activities were measured in five sizes of soil aggregates (4–2, 2–1, 1–0.5, 0.5–0.25 and 0.25–0.05 mm). The results showed that the landscape position significantly influenced the mass distribution of aggregates. This was observed, in particular, for larger macroaggregates (4–2 mm). By increasing the SOC and TN contents along both the landscapes studied, the macroaggregate mass fractions (4–2; 2–1; 1–0.5 mm) increased. The results also showed that the SOC and TN contents as well as amidohydrolase activities were heterogeneously distributed within soil aggregates. Macroaggregates (generally 4–2 mm) exhibited greater SOC and TN contents as well as amidohydrolase activities compared to microaggregate, though the 4–2 mm aggregates constituted a small component of most soils. Our findings also revealed that proportional distribution of amidohydrolase activities within the soil aggregates was influenced by the landscape position. With increasing soil aggregation along the landscapes, the amidohydrolase activities tended to increase into macroaggregate fractions. Moreover, SOC, TN and, the enzyme activities in larger macroaggregates are more sensitive to landscape-induced changes than other size of aggregates.

1. Introduction

Soil biochemical properties, particularly soil enzymes, vary specially and are influenced indirectly by some landscape features, including topography, slope aspect and position, elevation, climate, parent material, and vegetation (Sidari et al., 2008; Nahidan et al., 2015). It is well known that topography influences local microclimates by changing the pattern of precipitation, temperature and relative humidity (Sidari et al., 2008) which, significantly affects the soil physiochemical properties, and influences microbial biomass as well as enzyme activity (Tajik et al., 2012; Nahidan et al., 2015). Understanding the variability of soil properties at a landscape scale will help refine land management practices and improve our understanding about factors controlling the distribution and activities of soil organisms.

Soil aggregates are a dynamic component of the soil fabric, mediated by soil organic matter (OM), biota, mineral particles and binding agents (Bronick and Lal, 2005). Complex and discontinuous pattern of pore spaces with varying sizes and shapes are mediated as a result of spatial arrangement of solid particles (Young and Ritz, 2000). The structural organization of soil particles can provide spatially heterogeneous habitat for microorganisms characterized by different substrates, nutrients, water content, and oxygen concentration. The location of microorganisms within the soil aggregates can influence their activity and functionality (Gupta and Germida, 2015). Previous studies revealed an uneven distribution of extracellular enzymes within the soil architecture. Enzyme activities were shown to be higher in macroaggregates (Hojati and Nourbakhsh, 2009), but also greater in microaggregates (Fansler et al., 2005). Differences in quantity and quality of substrate, moisture, and aeration status of soil aggregates can generally influence enzyme activities and microbial nutrient turnover, impacting soil nutrient release and plant uptake (Ladd et al., 1996). Aggregate scale investigations of enzymes in the soil should be utilized to strengthen our understanding about the microbial turnover of OM (Roldán et al., 2005).

Previous studies reported that various factors such as cultivation (Fansler et al., 2005), tillage (Muruganandam et al., 2009; Jiang et al.,

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2011) and manure amendment (Wang et al., 2017) induced changes in the soil aggregation regulating the microbial function in the soil ecosystem. It has also been reported that soil water-stable macroaggregates were more sensitive to soil management, and could help to evaluate the impacts of land management on the soil quality (Wang et al., 2017). Topography is another factor that modifies soil aggregation through alterations in temperature, precipitation, and soil properties (Bronick and Lal, 2005; Pierson and Mulla, 1990; Eneje and Adanma, 2007). However, the effect of topography on the variation in soil microbial activities in relation to soil aggregates is not recognized. More detailed information is required to understand how topography controls microbial functions in the soil ecosystem.

In this study, we considered amidohydrolase activities [urease (EC 3.5.1.5), L-glutaminase (EC 3.5.1.2) and L-asparaginase (EC 3.5.1.1)] within soil aggregates. The enzymes are central regulators of N cycle in microbial cells and produce inorganic N during the decomposition of aliphatic N compounds in the soil OM (Tabatabai, 1994). This study aims to analyze the distribution pattern of amidohydrolase activities within soil aggregates obtained from different landscape positions. We hypothesized that topography can influence soil enzyme activity through its effect on aggregate size distribution. Lacking information is available about the distribution pattern of amidohydrolase activities within the soil aggregate along a landscape. Such studies are important for identifying the microbial habitats with greater biochemical activity and sustaining soil management practices.

2. Materials and methods

2.1. Site description and soil sampling

The landscapes studied are located at two pasture sites in west central Iran. Site 1 is located in Fereydan area which is west of the Isfahan province, central Iran (50° 11′E, 32° 45′N). The mean annual precipitation and temperature are 600 mm and 5 °C, respectively. The landscape studied was 100 m long and 40 m wide, and elevation ranged from 2583 to 2630 m. The soils along the landscape were classified as Haplic Calcisols (WRB, 2014). The dominant types of vegetation at the landscape were *Cousinia bachtiarica*, *Eryngium bilardierie*, *Astragalus verus and Astragalus sousianus*. Site 2 is located in Chelgerd area, west central Iran (50° 22′E, 32° 21′N). The mean annual precipitation and temperature are 1414 mm and 9.8 °C, respectively. The landscape studied was 140 m long and 40 m wide, and elevation ranged from 2454 to 2560 m. The soils were classified as Chromic Vertisols (WRB, 2014) and the dominant types of vegetation were *Daphne mucronata*, *Gypsophila* spp., *Astragalus* spp. *and Ciricium bracteusum* along the landscape.

Soil samples were collected along the landscapes based on the landform features including summit, shoulder, backslope, footslope and toeslope. At site 2, the summit was omitted from soil sampling because a real summit was not observed in that hillslope. At each landscape position, three equally-spaced points were selected as three replicates, at a distance of 10 m. In each point, ten soil cores of 0–10 cm depth were taken within a 2-m radius and composited. The soil samples were broken along natural points of weakness and passed through a 4-mm mesh before aggregate size fractionation. Particles that did not pass through the 4-mm sieve contained mostly stone and plant fragments and were discarded.

2.2. Aggregate size fractionation

Wet sieving method was used for aggregate separation (Elliott, 1986). The sieve sizes were 2, 1, 0.5, 0.25, and 0.05 mm; therefore the aggregates were separated into five size ranges (4–2, 2–1, 1–0.5, 0.5–0.25 and 0.25–0.05 mm). The soil (100 g dry weight) was wet-sieved in a water bucket for 2 min with a vertical stroke of 1.3 cm and a speed of 50 times minute^{-1}. After sieving, the soils on the sieves remained in the water bucket undisturbed for 5 min to allow the fine particles to settle. Then, the sieves were gently pulled out and the soil aggregates remaining on each sieve were collected. The collected aggregates were kept at 4 °C before analysis. To determine the proportion of each aggregate in the whole soil mass, the subsamples of each aggregate size were oven dried at 105 °C and then weighed.

2.3. Soil analysis

Soil particle size distribution was measured using the pipette method. Soil pH was determined in 1:2 (soil: water) suspension. Ca–carbonate equivalent (CCE) was determined by the titrimetric method. SOC was assayed by potassium dichromate oxidation. TN was determined by the Kjeldhal digestion method (Burt, 2004).

URS, LGL, and LAS activities were measured in accordance with the protocols described by Tabatabai (1994). These methods involve the determination of ammonia released when a soil sample is incubated with toluene, THAM buffer at the optimal pH, and an enzyme specific substrate solution at 37 °C for 2 h. Urea, L-glutamine, and L-asparagine were used as substrates for URS, LGL, and LAS, respectively.

SOC, TN, and amidohydrolase activities were determined either in bulk soils (non-fractionated soils) or in the separated aggregate size fractions. The relative contribution of each aggregate size fraction (R) to total measured parameters represented as a percentage of bulk soil was calculated as follows:

$$R = (A_i \times C_i)/B,$$

where A_i is absolute value of SOC and TN (g kg^{-1}) or enzyme activities (mg NH_4^+–N kg^{-1} h^{-1}) in each aggregate size (i), C_i is the contribution of each aggregate to whole soil mass (%), and B is the values of measured parameters in the bulk soil.

2.4. Statistical analysis

A randomized complete block design was used with three replicates. The data were checked for normality and homogeneity of variance and then analyzed by using one–way ANOVA procedure of SAS (SAS Institute, 1990). Mean comparisons (LSD, $P < 0.05$) were accomplished for each site, separately. Pearson correlations were performed using the software SAS (SAS Institute, 1990).

3. Results

3.1. Physico-chemical properties and amidohydrolase activities of bulk soil

Soil properties such as particle size distribution, CCE, SOC, and TN were significantly different among landscape positions (Table 1). The pH and EC did not vary much along the landscapes studied. Landscape position considerably affected URE, LGL and LAS activity. At site 1, the activity of URS, LGL and LAS ranged from 16.5 to 40.1, 14.0 to 69.8 and 1.67 to 7.83 mg NH_4^+–N kg^{-1} h^{-1}, respectively. At site 2, URS, LGL, and LAS activities varied from 41.1 to 64.5, 23.2 to 167.9 and 3.71 to 17.1 mg NH_4^+–N kg^{-1} h^{-1} along the landscape, respectively (Table 1).

3.2. Aggregate size distribution

At site 1, the distribution of water-stable aggregates was skewed toward microaggregates (0.25–0.05 mm) and did differ among landscape positions (Table 2). Macroaggregates (4–0.25 mm) contributed 41.7% to the whole soil at the summit position, while the percentage decreased to 30.2, 23.4, 23.5 and 34.8% at the shoulder, backslope, footslope and toeslope, respectively. The effect of the landscape position on the aggregate size distribution was most pronounced for larger macroaggregates (4–2 mm). The proportion of larger macroaggregates (4–2 mm) at the summit was 4.8% which is 6.00, 6.86, 6.86 and 2.67 times greater than that found at the shoulder, backslope, footslope and toeslope, respectively (Table 2). The amount of macroaggregates (4–2,