



Effects of endophyte-infected and non-infected tall fescue residues on aggregate stability in four texturally different soils

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ARTICLE INFO

Article history:

Received 31 July 2016

Received in revised form 26 September 2016

Accepted 1 October 2016

Available online 13 October 2016

Keywords:

Epichloë coenophiala

High energy moisture characteristic

Stability ratio

Soil organic carbon

Basal microbial respiration

ABSTRACT

Fungal endophyte (*Epichloë coenophiala*) usually infects cool-season perennial forage grasses and enhances host plant resistance to biotic and abiotic stresses and, by potentially altering the litter quality and decomposition rate, can affect soil properties. Aggregate stability, as an important determinant of plant root growth and soil quality, may be affected by the endophyte status of plant residues. In this study, the effects of endophyte-infected (E+) and endophyte-free (E−) tall fescue residues (0%, 1% and 2%) on soil organic carbon (SOC), basal soil respiration (BSR) and aggregate stability were investigated in four texturally different soils in the laboratory. The aggregate stability indices were determined by the high energy moisture characteristic (HEMC) method. Moist soil samples were thoroughly mixed with either E+ or E− tall fescue residues before being incubated at 25 °C. During the 2-month incubation period, the amended soil samples were subjected to ten wetting and drying cycles, after which soil properties were measured. The results indicated that SOC and aggregate stability were higher and that BSR was lower in the finer-textured soils. Furthermore, an increase in the application rate of plant residues significantly increased SOC, BSR and aggregate stability. Treating the soils with E+ tall fescue residues increased SOC and aggregate stability (i.e., stability ratio, SR, a dimensionless index defined as the ratio of the structural index for the fast wetting to the structural index for the slow wetting), and significantly decreased BSR due to the toxic effects of phenolic compounds on soil microbial communities and changes in the litter chemical quality. The interactive effect of soil type and endophyte status on the SOC and BSR values was significant but no clear trend was observed. Our findings indicated that tall fescue residues (especially E+ ones) can improve soil physical quality due to increased SOC storage and greater aggregate stability. Therefore, these plants have great potential for use in grassland reclamation and soil conservation plans in semi-arid pasture and agricultural lands.

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1. Introduction

Incorporating plant residues into soil is an important conservation management practice that can restore and maintain the soil quality of degraded lands (Abiven et al., 2009). Decomposition of fresh organic matter components in the soil is an essential and crucial natural process that affects soil organic carbon (SOC) storage (Leifheit et al., 2015) and, hence, soil physical properties. Decomposing organic residues may release some hydrophobic compounds (e.g., humic acids, aliphatic substances, and organic polymers) that increase the hydrophobicity of aggregates and induce moderate (sub-critical) water repellency in the soil (Blanco-Canqui and Benjamin, 2013). Recent studies indicate that development of sub-critical water repellency may be essential to the stabilization of soil aggregates because it reduces their breakdown due

to a lower rate of air pressure build-up inside the soil pores (Bottinelli et al., 2010; Blanco-Canqui, 2011; Hosseini et al., 2015a).

Endophytic fungi of the genus *Epichloë* are systemic and nonpathogenic symbionts of temperate grasses, colonizing the leaves and other aboveground tissues by growing between the plant cells (Clay, 1988). The presence of endophytes in the aerial parts of their host grasses induces physiological and biochemical changes that ultimately increase plant tolerance against biotic and abiotic stresses (Malinowski and Belesky, 2000; Hosseini et al., 2016; Hume et al., 2016). Furthermore, it may affect soil physical and biological conditions through changes induced in the quality and quantity of root exudation (Van Hecke et al., 2005; Hosseini et al., 2015a, 2015b). Hosseini et al. (2015a, 2015b) found that endophyte-infected (E+) tall fescue associated soils had greater SOC and lower basal soil respiration (BSR) rates than endophyte-free (E−) plants, which resulted in greater hydrophobicity and aggregate stability in the rhizosphere soil. Similarly, Franzluebbers and Stuedemann (2005) reported that E+ tall fescue pastures in central Georgia had higher SOC and N storages, lower microbial biomass and reduced BSR rates than adjacent E− pastures.

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Endophyte has a significant effect on decomposition rates of plant litter in the soil that is mainly due to changes in litter quality because of the modification of plant chemical constitutions and alkaloid contents, increases in litter quantity caused by enhancing plant growth, and the stimulation or inhibition of soil microbial activity (Bernard et al., 1997; Omacini et al., 2004). Lemons et al. (2005) and Siegrist et al. (2010) reported that E+ tall fescue plant residues decomposed more slowly than E− ones.

Soil structure controls numerous ecological functions, and has a strong impact on plant root growth, retention and movement of water and oxygen diffusion. Usually, a well-structured soil has stable macro-aggregates with proportions of micro- and macro-aggregates that are beneficial (Dexter, 1988). Soil structure and pore size distribution can be affected by several factors including soil texture, SOC content, tillage practices and land use, as well as by plant root exudations, microbial activity and the presence of fungi hyphae (An et al., 2010; Sleutel et al., 2012). Aggregate stability, i.e., the ability of aggregate bonds to resist external destructive forces, is an important soil physical property that determines its resistance to erosion and degradation (Six et al., 2000). Arshad and Cohen (1992) defined structural stability as an indicator of soil physical quality. Hortensius and Welling (1996) incorporated aggregate stability in the international standardization of soil quality measurements. Therefore, determination of soil aggregate stability can provide an indication of soil conditions (Rohošková and Valla, 2004).

There are various approaches to determine soil aggregate stability (Amezketta, 1999; Pulido Moncada et al., 2015) among which the high energy moisture characteristic (HEMC) method is identified as a practical and useful way for determining the structural stability of various soil types; the HEMC method can even detect small changes in the structural stability of different soils from arid and humid regions, and its indices can be related to management practices (Pierson and Mulla, 1989; Levy and Mamedov, 2002; Mamedov and Levy, 2013). The method identifies the sensitivity of soil aggregates to slaking and destructive forces of entrapped air by measuring differences in the water retention curves of fast- and slow-wetted aggregates at high matric potentials (e.g., matric suctions of 0 to 50 hPa). This information allows a stability ratio (SR) to be calculated that can be used as an index to evaluate soil structure stability (see Section 2.4) where higher values of SR indicate greater soil aggregate stabilities ($0 < SR < 1$).

Mamedov et al. (2014) used the HEMC method to evaluate the effects of amending soils with two organic wastes (composted manure and activated sludge), and with orthophosphate, phytic acid and humic acid on the structural stability of three arable soils before and after exposure to rain storms. The results showed that the amendments enhanced soil aggregate stability relative to the control prior to the rain storms. However, only activated sludge and humic acid treatments enhanced the soil structural stability during and after the rain storms. They concluded that the effects of these soil amendments on aggregate stability were inconsistent and would depend on the soil type and the amendment chemical composition. Furthermore, addition of the activated sludge stabilized fine- and medium-textured soils and decreased their vulnerability to sediment loss due to the buffering capacity of the amended soils and the maintaining of a high humic acid concentration in the soil solution (Mamedov et al., 2014).

The influence of plant residue applications on soil physical properties such as aggregate stability, compactibility and friability, and on soil-water relations have been extensively reported (e.g., Soane, 1990; Blanco-Canqui and Benjamin, 2013). For instance, Gupta et al. (1987) indicated that adding 3.4% w/w of maize residue decreased the bulk density of a wet clay loam under a confined axial stress of 100 kPa from 1.2 to 0.9 Mg m^{−3}. Barzegar et al. (2002) indicated that applications of organic materials including wheat straw, composted sugarcane bagasse residue and farmyard manure significantly increased the aggregate stability, infiltration rate, and soil water retention at matric suctions lower than 1000 hPa, and decreased soil bulk density. Tejada et al. (2009) examined the effects of composted plant residues on plant

cover and soil physical (i.e., structural stability and bulk density), chemical, and biological properties during a period of 4 years in a semiarid Mediterranean agro-ecosystem. Their results indicated that the soils amended with composted plant residues (with higher humic acid concentrations) had greater aggregate stabilities and lower bulk densities at the end of the experimental period.

There is a wide variety of cool-season grasses and of their endophytic fungi symbionts in native pastures of Iran (Karimi et al., 2012). These plants and their residues have the potential to be used in soil conservation and rangeland rehabilitation strategies. However, reports regarding the effect of E+ and E− plant residues on the physical quality and properties of soils varying in texture are scarce. We hypothesized that the presence of *Epichloë coenophiala* in the residues of tall fescue (*Festuca arundinacea*) could affect SOC storage and microbial activity of a soil by changing the litter quality, and that hence, soil aggregate stability might be affected. The aim of this study is to assess the influence of endophyte-infected and non-infected tall fescue residues on (1) soil organic carbon and basal microbial respiration, and (2) aggregate stability indices (determined by the HEMC method) in four texturally different soils in a laboratory experiment.

2. Material and methods

2.1. Soil sampling and plant material preparation

Soil samples were collected in the fall 2012 from the surface layer (0–30 cm) of four arable lands around Isfahan City. All of the fields had been previously under wheat cultivation with conventional management practices. However, when the soil samples were collected, agricultural crop was harvested, and there was no crop on the land. After air-drying and passing through a 2-mm sieve, a portion of soil samples was transferred to the laboratory for determination of basic soil properties. The following methods were used: particle size distribution by the pipette method (Soil Conservation Service, 1984), soil organic carbon (SOC) content using wet-oxidation (Walkley and Black, 1934), calcium carbonate equivalent (CCE) by back-titration with NaOH (Sims, 1996), and pH and electrical conductivity (EC) of a saturated soil extract by pH and EC meters. These properties of the studied soils are presented in Table 1.

Plant residues of tall fescue (*F. arundinacea*) were collected from the Research Farm of Isfahan University of Technology. Prior to collecting the plant residues, the presence or absence of the endophytic fungus in E+ and E− clones, respectively, was confirmed by staining plant leaf tissues with rose bengal as described by Saha et al. (1988). Plant residues from E+ and E− tall fescue shoots were harvested in April 2013. Tall fescue produced a lot of new tillers in the spring; we cut young and well-developed leaves to achieve plant residues for the experiment. Therefore, plant materials were harvested approximately 2 months after leaf generation and development. The collected plant residues were oven-dried at 40 °C, ground with an electric mill (Moulinex-model AR1066Q, China) and sieved through a 1-mm mesh. Chemical characteristics of the plant residues were measured: organic C content by the wet-oxidation method (Walkley and Black, 1934), total Kjeldahl nitrogen (Bremner, 1996), C/N ratio and water-soluble phenolic compounds (Table 2).

Water-soluble phenolic compounds were extracted using the method of Kong et al. (2008). The powdered residues were suspended in de-ionized water (1 g:8 ml) and shaken for 24 h at 100 rpm on a shaker at room temperature. The plant extract solution was filtered through filter paper and retained for further analysis. The concentration of water-soluble phenolic compounds in the prepared extract solution was determined using Folin-Ciocalteu reagent as described by Beta et al. (2005). One milliliter of plant extract was mixed with 5 ml of Folin-Ciocalteu reagent diluted with water (1:10) and 4 ml of a saturated sodium carbonate solution. After 15 min, the absorbance at 760 nm was measured by an UV/vis spectrophotometer (Jenway 6505 UV/vis, Bibby Scientific

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