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Effects of soil tillage and planting grass on arbuscular mycorrhizal fungal propagules and soil properties in citrus orchards in southeast China

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ABSTRACT

Arbuscular mycorrhizal (AM) fungi play important roles in plant nutrition and soil conservation. AM fungal activity and amount of glomalin (a glycoprotein derived from AM fungi) are influenced by a series of abiotic and biotic factors associated with changes in soil management. However, the informations concerning AM fungal activity, amount of glomalin-related soil protein (GRSP), and its relationship with soil fertility and enzyme activities are scarce in citrus orchards under different soil management systems. Accordingly, this paper reports the effects of tilling soil and planting grass on AM colonization; AM fungal propagules; the amount of GRSP and its relationship with soil chemicals including organic carbon, available nitrogen, Olsen phosphorus, and available potassium; the activity levels of soil enzymes like catalase, invertase, urease, and acid phosphatase in citrus (*Citrus unshiu* Marc. grafted on *Poncirus trifoliata* L. Raf.) orchards in southeast China. Measurements were conducted in the fifth year of an ongoing tillage and grass planting experiment on an Acrisol soil under one of three treatments: natural grass cover, sod culture with white clover, and clean tillage. Soils and roots were sampled in March, July, and November 2013, during the vigorous growth period of citrus trees. AM colonization, AM fungal propagules, and GRSP were significantly higher in both no-tillage and grass-planted orchards than in clean tillage orchards. Soil organic carbon, available nitrogen, and soil enzyme activities showed a similar trend. Principal component analysis revealed that tillage and grass planting dramatically affected soil conditions in citrus orchards. However, sod culture promoted citrus mycorrhizal colonization and available nitrogen in soil more effectively than did grass cover, whereas natural grass cover enhanced AM fungal propagules, soil organic carbon, and soil enzyme activities more effectively than did sod culture. Soil organic carbon, available nitrogen, and soil enzymes activities were significantly ($P < 0.05$) correlated with AM fungal propagules or GRSP, suggesting that plentiful AM symbionts may help preserve soil quality, especially in grass-planted orchards.

1. Introduction

Arbuscular mycorrhizal (AM) fungi are very important microbial symbionts for most terrestrial plants (Smith and Read, 2008). In general, AM fungi colonize host plant roots and obtain photosynthetically fixed carbon from their hosts; in return, they benefit their host plants in several ways, including improving their uptake of mineral nutrients, especially phosphate (Toro et al., 1998), improving their resistance to biotic and abiotic stresses, such as drought, cold, salinity, and pathogens (Augé, 2004; Whipps, 2004; Wu, 2011). Of even more importance is the fact that AM fungi can contribute to soil quality by increasing the stability of soil aggregates, slowing the decomposition speed of soil organic carbon (SOC), and regulating the activity of soil enzymes (Rillig, 2004; Wu et al., 2015). Thus, AM fungi are an important component of ecosystems and are significant in sustainable agriculture.

Glomalin, a glycoprotein derived from AM fungi, may be measured in soils as glomalin-related soil protein (GRSP), which can form a lattice-like coating on the surface of aggregates (Rillig et al., 2003a). It increases soil stability against wind and water erosion and can store carbon, thereby improving soil quality

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(Nichols and Wright, 2004; Rillig et al., 2007). Hence, the amount of glomalin in soil is used to evaluate soil quality. Generally, GRSP is affected by multiple abiotic and biotic factors, such as habitat, tillage, herbicides, and so on in agricultural systems (Rillig et al., 2003a,b). Among these factors, soil management plays a particularly important role (Wright et al., 2007). For example, Wright et al. (1999) reported that the amount of glomalin was 1.5 times higher in no-tilled soils than in tilled soils.

In agricultural soils, management practices (e.g., crop rotation, mulching, tillage, and application of fertilizers and pesticides), and other environmental factors (e.g., pH, temperature, humidity) can influence the microbial community in the soil and thereby affect various enzyme activities in soils (Ajwaa et al., 1999; Giri et al., 2005). Among several different soil enzymes, catalase mediates protection of living cells in the presence of activated oxygen species like H_2O_2 (Weigand et al., 1995). The enzymes invertase, urease, and phosphatase are involved in the mineralization of important nutrient elements such as carbon (C), nitrogen (N), and phosphorus (P). These enzymes are highly correlated with levels of biological activity in soils (Brooks et al., 2013; Burne and Chen, 2000; Neumann and Lampen, 1967), and thereby directly mediate the biological catabolism of soil organic and mineral components, so they are sensitive indicators of soil quality alteration (Balota et al., 2004).

The practice of sowing into no-tilled soils was developed to reduce soil degradation and costs of agricultural production (Tebrügge and Düring, 1999). No tillage positively influences soil characteristics, including the contents of SOC, available N (AN), Olsen P, and increases P acquisition by crops (Borie et al., 2006). Apart from soil fertility properties, the impact of soil disturbance is commonly on biological properties, including both free and symbiotic fungal populations. Kabir (2005) reported that conventional tillage could decrease AM fungi hyphal survival and proliferation, thus reducing the benefits of the symbiosis to associated plants and soils. Curaqueo et al. (2011) reported that no-tillage management increased the density of AM fungal propagules compared with conventional tillage.

Citrus are among the most economically valuable fruit trees in China, and indeed, the world. In the field, citrus trees are fairly dependent on AM symbiosis for the uptake of water and nutrients for growth and fruit production (Wu et al., 2008). Moreover, AM fungi have been proposed as a potent biofertilizer in organic agriculture in China. However, the effects of different soil management practices on AM symbiosis development, the amount of GRSP in soil, and soil C and enzymes in citrus orchards remain largely unknown, although AM associations play a vital role in the maintenance of agroecosystem sustainability. Accordingly, the aim of the present study was to evaluate the influences of natural grass cover, sod culture with white clover (*Trifolium repens* L.), and clean tillage soil management on AM colonization in citrus roots, AM fungal propagules in citrus rhizospheres, and GRSP content, and to relate these measures with soil chemical and biochemical characteristics in citrus orchards in southeast China.

2. Material and methods

2.1. Experimental field and design

The experiment was conducted in hillside citrus orchards planted with roughly 10-year-old Satsuma mandarin trees (*C. unshiu* Marc. grafted on *P. trifoliata* L. Raf.) and located at Taizhou (120°56′–121°39′E, 28°27′–29°04′N). This area has a semi-tropical monsoon climate with annual sunlight of 1800–2037 h, a frost-free period of about 235–322 days, mean precipitation of 1632 mm, annual mean temperature of 16.6–17.5 °C, July mean temperature of 26.6–28.5 °C and January mean temperature of 5.0–6.9 °C. Prior to the present study, traditional clean tillage (frequently tilling soil to remove all weeds) had been always applied in these citrus orchards.

During the experiment, the orchards were subjected to three different types of soil management strategies. Under sod culture (SC), white clover was planted across the entire orchard. Under natural grass cover (NC), natural grass (dominant species of weeds are listed in the Table 1) was planted across the entire orchard, competitive weeds were eliminated, and grass was cut several times to inhibit its vigorous growth and covered under the canopy of citrus trees. Under clean-tillage (CT), the control group, the soil was frequently tilled to remove as many weeds as possible. The SC and NC treatments were continuously employed for 5 years prior to the beginning of the study and were arranged in a randomized complete-block design with three replicates. The orchard soil was classified as yellow sandy clay soil (Acrisol in FAO Taxonomy) and organic fertilizer (7% N, 4% P_2O_5, 4% K_2O, and 25% organic matter; 2500 kg per 667 m^2) was used as the base fertilizer in all experimental orchards after fruit was harvested.

2.2. Sample collection

In each soil management type orchard, three randomly replicated experimental plots were chosen, and in each of these, five uniform citrus trees with similar growing potential were sampled in March, July, and November 2013 during the vigorous growth period of citrus trees. Soils and fine roots from each citrus tree were collected from east, south, west, and north sides from soil layer depths of 0 to 30 cm after removing the upper vegetation within the dripping line (the outermost circumference of the tree canopy) by using a small digging shovel. Rhizospheric soil was collected according to the method described by Scott and Condron

Table 1
Species of the dominant weeds in four seasons in natural grass cover orchards.

Family	Species	Season			
		Spring	Summer	Autumn	Winter
Gramineae	*Digitaria sanguinalis* (L.) Scop.	+	+	+	
	Eleusine indica(L.) Gaertn.	+	+	+	
	Alopecurus aequalis Sobol.		+	+	+
	Echinochloa hispidula	+	+	+	
	Poa annua L.	+			+
Asteraceae	*Conyza canadensis* (L.) Cronq.	+	+	+	+
Scrophulariaceae	*Veronica didyma* Tenore	+	+		+
Caryophyllaceae	*Stellaria media* (L.) Cyr.	+	+		+
Euphorbiaceae	*Acalypha australis* L.	+			+
Rubiaceae	*Galium aparine* L.		+	+	

"+" indicated the weed distributed in this season.