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## Effects of soil management practices on soil microbial communities and development of southern blight in vegetable production



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#### ABSTRACT

Soil microbial communities and their relationship with major soil factors and disease development under tillage, no-tillage and organic farming practices were studied. Real-time quantitative-PCR (Q-PCR) was performed to quantify soil fungal and bacterial abundance. Length heterogeneity polymerase chain reaction (LH-PCR) combined with cloning and sequencing was used to assess microbial communities. Soil chemical characteristics were significantly different under different management practices. Organic agricultural practice significantly increased soil bacterial and fungal abundances, but not the diversity when compared to tillage and no-tillage management practices. Tillage farming agricultural practice yielded a significant higher bacterial abundance than no-tillage agricultural practice. No difference in fungal abundance was observed between tillage and no-tillage. Bacterial communities in no-tillage and organic cultivation were relatively similar, but significantly different from those in tillage cultivation. Differences of bacterial communities were correlated to soil organic matter ( $R^2 = 0.558$ ), cation exchange capacity ( $R^2$  = 0.583) and percentage of Mg ( $R^2$  = 0.590). Fungal communities in soil under organic cultivation were significantly different from relatively similar fungal community structure in no-tillage and tillage cultivation treatments. Differences of fungal communities were correlated to soil pH  $(R^2 = 0.421)$  and soil organic matter  $(R^2 = 0.401)$ . In greenhouse assays, severity of southern blight on tomato plants grown in soils collected from the no-tillage was significantly lower than in soils from the tillage field. Different soil management practices have played an important role in altering soil microbial communities and potentially reducing disease incidence of southern blight on tomato.

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#### 1. Introduction

Soil microorganisms play important role in regulation of terrestrial ecosystem functions including nutrient cycling and soil-borne disease suppression (Bever, 2003; King et al., 2004; Went and Stark, 1968). Soil management practices have greatest effects on soil physical and chemical characteristics, and especially on soil microbial diversity and community structure (Frey et al., 1999; Hirsch et al., 2010; Tiedje et al., 1999; Wardle et al., 1995). Because of the importance of soil microorganisms in sustainable agricultural production, exploring of the diversity and community structure of soil microorganisms induced by soil management practices can contribute to further an understanding of ecosystem-level process and thus, the development of sustainable agricultural systems (Anderson and Cairney, 2004; Stromberger et al., 2005).

Organic agricultural practices increase microbial diversity and enhance nutrient cycling because of a higher level of genes involved in carbon, nitrogen, phosphorus, and sulfur cycles in organic than in conventional practices (Reeve et al., 2010; Xue et al., 2013). Organically managed fields also maintain a more stable microbial C-to-N ratio than conventionally managed fields do (Gunapala and Scow, 1998). Microbial biomass was nearly doubled when fertilization was based on organic manure in comparison to chemical fertilizers (Qiu et al., 2012; Zhang et al., 2012). The plant pathogenic fungus Sclerotium rolfsii, causal agent of southern blight, progressed faster and had a higher level of incidence on tomato plants grown in conventional soils than in organic soils (Liu et al., 2008). Total numbers of Actinobacteria isolated from tomato rhizosphere soils were significantly higher in the organic than in conventional soil (Drinkwater et al., 1995).

No tillage managements increase soil organic matter (SOM) contents, reduce erosion, and lower energy consumption and production costs (Carter et al., 1994). The rate of soil organic matter decomposition and nutrient availability are affected by no tillage

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managements due to the shifting of soil microbial relative abundance (Frey et al., 1999). Different agricultural tillage practices can strongly influence the abundance and biomass of soil microorganisms. Fungal biomass increased in surface soil of reduced tillage, while bacterial biomass was not strongly affected by tillage (Frey et al., 1999). Reduced tillage changes soil environment and affects the disease incidence or severity, depending on the cropping system and disease (Bockus and Shroyer, 1998; Paulitz et al., 2002; Rothrock, 1992; Sumner et al., 1981). The shifting of microbial community and abundance of soil pathogen under reduced tillage practices may directly affect the incidence of soil-borne diseases (Page et al., 2013). The abundances of bacteria and fungi in no-tillage and conventional tillage soils were traditionally quantified using plate counts (Doran, 1980; Linn and Doran, 1984; Norstadt and Mccalla, 1969). However, these data do not allow quantitative comparison of the relative amounts of bacterial and fungal biomass, a critical requirement when linking community composition to nutrient dynamics. Soil bacterial and fungal diversity was increased under conservation tillage systems, and a great abundance of fungi relative to bacteria was reported at the surface of conservation tillage systems (Page et al., 2013).

Linkage between microbial community structure and soilborne disease is not completely understood, but more and more research supports that crop management practices influence ecological processes that affect microbial communities involved in the suppression of soil-borne disease development and incidence (Burton et al., 2010; Chellemi et al., 2012; Vanbruggen, 1995). Proper manipulation of the microbial community structure can alter the population of antagonistic microorganisms and decrease the amounts of soil-borne pathogens. The mechanisms may include creating competition for space, nutrients, and metabolic competition (Harrier and Watson, 2004; Mazzola, 2004). The application of organic manure including the antagonistic microorganisms Bacillus subtilis, Paenibacillus polymyxa, and Trichoderma harzianum has shown a suppression of Fusarium wilt by 83% in cucumber cultivation (Qiu et al., 2012), and a suppression of 58-73% of southern blight disease in tomatoes (De Curtis et al., 2010). Compost amendments increase soil pH and are one of the key factors in suppression of some soil-borne diseases (Alabouvette, 1999; Noble, 2011). Till now, limited studies determined the contribution of soil management practices on soil chemical and physical factors, their effects on belowground biodiversity, and especially the linkage between soil microbial community structure and soil-borne diseases in vegetable production. The objectivities of this study were: (1) to investigate the effects of long-term organic and no-tillage management practices in contrast to conventional tillage management practices on microbial diversity and community structure in agricultural soils for vegetable production; (2) to examine the effects of different agricultural management practices on soil physical and chemical characteristics and their correlation with microbial community structure: and (3) to explore the relationship of microbial communities and their suppression of southern blight disease caused by S. rolfsii on tomato. We hypothesize that agricultural management practices determine soil physical and chemical characteristics, which further affect soil microbial communities and suppression of soil-borne diseases.

#### 2. Materials and methods

#### 2.1. Study site

The study was conducted at the Horticulture Farm, Tifton Campus, University of Georgia, USA. The soil was a Tifton Sandy Loam (a fine loamy-siliceous, thermic Plinthic Kandiudults). Different management practices were applied to the fields in

the past years: (a) conventional tillage, (b) organic, and (c) notillage, in a complete random block design with five replicates.

#### 2.1.1. Conventional tillage field

Field was used to grow vegetables in the past decade. Tomato (Solanum lycopersicum) or bell peppers (Capsicum annum L.) was grown in the spring and fall and cereal rye (Secale cereal L.) was grown as a cover crop in winter each year. Most recently, bell pepper was grown in the spring of 2011, squash (Cucurbita sp.) in the fall of 2011, and cereal rye as a winter cover crop in 2011–2012. In the spring of 2012, cereal rye was terminated at blooming stage with a rototiller three weeks before planting the tomato crop. The residue was incorporated into the soil. Before laying mulch with a mulch-laying machine, the soil was fertilized with N, P, and K at 67, 30, and 56 kg ha<sup>-1</sup> respectively, using 10 N-4.4 P-8.3 K granular fertilizer. At the same time, plastic film mulch [silver on black, low density polyethylene with a slick surface texture, 1.52 m wide and 25 mm thick (RepelGro; ReflecTek Foils, Inc., IL, USA)] was laid, and drip irrigation tape [20.3-cm emitter spacing and a 8.3-mLmin<sup>-1</sup> emitter flow (Ro-Drip; Roberts Irrigation Products, Inc. CA, USA)] was placed 5 cm deep in the center of the bed. Tomato plants were established on individual raised beds  $(6 \text{ m} \times 0.76 \text{ m})$  on 1.8-m centers). Starting three weeks after planting, calcium nitrate and potassium thiosulfate were applied weekly via the drip tape. Total fertilization for the season was 180 N, 30 P, and 149 K.

#### 2.1.2. Organic field

Field was converted to organic in 2003. Tomato was grown in the spring and sunnhemp (Crotalaria juncea L.) was grown in summer/fall each year. Cereal rve or sunnhemp was grown in the winter. Field was planted with cereal rye cover crop in the winter of 2010–2011 and sunnhemp in winter of 2012. Cover crop was terminated and incorporated into the soil as in the conventional field. Tomato plants were established on individual raised beds  $(6 \text{ m} \times 0.76 \text{ m} \text{ on } 1.8\text{-m} \text{ centers})$ . Before shaping the beds, soil was fertilized with a total of 6.7 t  $ha^{-1}$  of organic fertilizer (4 N:  $2P_2O_5$ : 3 K<sub>2</sub>O; MicroSTART 60, Perdue AgriRecycle, LLC, Delamarva, Delaware) one week prior to transplanting the tomato plants. Plastic film mulch and drip tape were as in the conventional. Organic fertilizer was applied at 6tha<sup>-1</sup> (4N: 2P<sub>2</sub>O<sub>5</sub>: 3K<sub>2</sub>O; MicroSTART 60) before laying plastic mulch, one week before planting tomato crop. Starting three weeks after transplanting, plants were fertilized weekly through the drip tape with fish emulsion. Total fertilization for the season was 180 N, 30 P, and 149 K.

#### 2.1.3. No-Tillage field

Field has been in no-tillage since 1990. Cereal rye and crimson clover (*Trifolium incarnatum* L.) were used as winter cover crops and were terminated with a rototiller, as in the conventional field. Cover crop residue was left on the soil surface as mulch. Tomato or peppers were grown in spring and fall. Plants were drip-irrigated and no plastic film mulch was used. Fertilization was as in the organic field.

#### 2.2. Sample collection and DNA extraction

Soil samples were collected in August 2012 from plots in the same field with different soil management practices: (a) conventional tillage, (b) organic, and (c) no-tillage. Five replicates of the samples were collected from 0–20 cm depth in each of the plots under three soil management practices. A total of 15 samples were obtained and analyzed individually. The lowest temperature of the day was 20.6 °C and highest temperature was 36.7 °C with an average relative humidity of 65%. Soil physical and chemical characteristics including soil pH, organic matter content, P, K, Mg,

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