



Functional soil organic matter fractions in response to long-term fertilization in upland and paddy systems in South China

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ABSTRACT

Soil organic matter (SOM) and its fractions play key roles in optimizing crop yield and improving soil quality. However, how functional SOM fractions responded to long-term fertilization and their relative importance for C sequestration were less addressed. In this study, we determined the effects of long-term fertilization on six functional SOM fractions (unprotected, physically protected, physico-biochemically protected, physico-chemically protected, chemically protected, and biochemically protected) based on two long-term fertilization experiments carried out in South China. The unprotected coarse particulate organic matter (cPOM), the biochemically and chemically protected silt-sized fractions (NH-dSilt and H-dSilt) were the primary C storage fractions under long-term fertilization, accounting for 23.6–46.2%, 15.7–19.4%, and 14.4–17.4% of the total soil organic carbon (SOC) content in upland soil and 19.5–29.3%, 9.9–15.5%, and 14.2–17.2% of the total SOC content in paddy soil, respectively. Compared with the control treatment (CK) in upland soil, the application of manure combined with mineral NPK (NPKM) resulted in an increase in the SOC content in the cPOM, pure physically protected fraction (iPOM), the physico-chemically protected (H- μ Silt), and the chemically protected (H-dSilt) fraction by 233%, 166%, 124%, and 58%, respectively. Besides, the SOC increase in upland soil expressed as SOC content per unit of total SOC for iPOM, H- μ Silt, cPOM and H-dSilt were the highest and as large as 283%, 248%, 194%, and 105% respectively. In paddy soil, the highest increase per unit of total SOC was H-dSilt (190%), followed by H-dClay (156%) and H- μ Silt (155%). These results suggested that the upland soil could stabilize more C through the pure physical, whereas the chemical protection mechanism played a more important role in paddy soil. Chemical protection mechanism within the microaggregates played important roles in sequestering C in both upland and paddy soils. Overall, the different responses of functional SOM fractions to long-term fertilization indicate different mechanisms for SOM cycling in terms of C sequestration under upland and paddy systems.

1. Introduction

Soil organic matter (SOM) is a key attribute for soil quality (Gregorich et al., 1994) and plays an important role in improving agricultural productivity (Smith et al., 2013). Furthermore, agricultural soils have the potential to mitigate global climate warming through C sequestration, counteracting increasing atmospheric CO₂

concentrations (Lal, 2004). Therefore, there is a great need to better understand the mechanisms of maintaining high SOM levels and to seek the optimal management practices to enhance C stock in agricultural soils.

The effects of long-term fertilization (mineral or manure) on the soil organic carbon (SOC) content have been extensively investigated. Previous studies have reported positive (Purakayastha et al., 2008;

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Gong et al., 2009) or no effects (Yang et al., 2007; Hai et al., 2010; Lou et al., 2011) of long-term mineral fertilization on SOC content. In addition, a number of studies showed that the SOC content tends to respond positively to manure amendments (Wang et al., 2015; Blanchet et al., 2016); however this is not always the case and the drivers behind the associated potential C storage still remains not clearly understood (Meersmans et al., 2012).

SOM is generally considered to be composed of several functional fractions differing in their intrinsic degradability and in factors controlling decomposition rates (Stevenson, 1994; von Lütow et al., 2007; Li et al., 2017; Tian et al., 2017). Given the strong heterogeneity of SOM, the labile SOM fractions are characterized by their rapid turnover and considered as more sensitive indicators of the effects of management practices as compared to total SOM (Haynes, 2005; von Lütow et al., 2007; He et al., 2015). According to stabilization mechanisms, four fractions are separated from the bulk SOM: the biochemically protected, chemically protected, physically protected, and unprotected fractions (Six et al., 2002). The unprotected fraction, which is not occluded within microaggregates, is labile and is an important nutrient source (Six et al., 2002). SOM is physically protected from decomposition through the formation of microaggregates, chemically protected by mineral (silt and clay) particles, and biochemically protected through the formation of recalcitrant SOM compounds (Six et al., 2002). Manure and mineral fertilization increased the SOC content within the free POM, occluded POM, and the OM associated with minerals (Sleutel et al., 2006). Based on a 35-year field experiment, Tian et al. (2017) showed that the physical, chemical, and biochemical protection mechanisms are important in maintaining high SOC levels after the addition of manure, but the relative importance of SOM stabilization mechanisms for C sequestration related to long-term fertilization remains unclear.

The changing trend of sequestered C varies with the increasing C input under long-term fertilization conditions. A linear relationship between C sequestration and C input in the soil has been reported in previous long-term fertilization field experiments (Sun et al., 2013; Fan et al., 2014; Wang et al., 2015). A few long-term agroecosystem experiments showed that the SOC stock exhibited little or no change with the varying C input (Stewart et al., 2007), indicating that there is a maximum capacity of C sequestration in the soil, known as soil C saturation (Six et al., 2002; Stewart et al., 2007). However, only a limited number of studies have assessed the response of differential functional SOM fractions to C input under long-term fertilization conditions.

We hypothesized that (1) long-term fertilization will affect the functional SOM fractions, thus will influence the relative importance of different protection mechanisms in controlling C sequestration; and (2) the functional SOM fractions vary with the increasing C input under different fertilization practices. To test our hypotheses, we investigated the effects of long-term manure and mineral fertilization on various functional SOM fractions based on two long-term field experiments in upland and paddy soils, and quantified the response of functional SOM fractions to the C input under long-term fertilization conditions.

2. Materials and methods

2.1. Study sites

The two experimental sites (adjacent upland and paddy field) and located at the Institute of Red Soil, Jiangxi Province, China (28°21'N, 116°10'E). This region is characterized by a subtropical climate with a mean annual temperature of 18.1 °C and a mean annual precipitation of 1537 mm. In the upland field, an early maize (*Zea mays* L.) - late maize - winter fallow rotation experiment has been conducted since 1986, whereas in the paddy field, an early rice (*Oryza sativa* L.) - late rice - winter fallow rotation experiment has been conducted since 1981. The upland soil had no irrigation measure and that for paddy soil was furrow irrigation. The upland and paddy soils are classified as red soil

according to the Chinese soil classification system and developed from quaternary red clay parental materials. At the beginning of field experiments, the basic soil characteristics of plough horizon were 9.39 g kg⁻¹ SOC, 0.98 g kg⁻¹ total N, 60.3 mg kg⁻¹ available N, and pH 6.0 for the upland field, and were 16.3 g kg⁻¹ SOC, 1.49 g kg⁻¹ total N, 144 mg kg⁻¹ available N, and pH 6.9 for the paddy field. All aboveground crop residues were removed from the field following harvest.

2.2. Experimental design

Both experiments were carried out in a randomized complete block design with three replicates per treatment. Each plot was 22.2 m² in size in the upland field and 46.7 m² in the paddy field. Four treatments were selected for this study, including: (1) control (CK, no fertilizer application); (2) N fertilizer application (N); (3) N, P, and K fertilizer application (NPK); and (4) combined N, P, and K fertilizer and manure application (NPKM). In both field experiments, urea, calcium-magnesium phosphate, and potassium chloride were used as N, P, and K fertilizers, respectively. The pig manure at a dose of 1500 kg ha⁻¹ (fresh weight) was applied for upland soil in each season. Manures for paddy soil were Chinese milkvetch (*Astragalus sinicus*) in the early rice season and pig manure in the late rice season and both of them were applied at 2250 kg ha⁻¹ (fresh weight) in each season. The water contents for pig manure and Chinese milkvetch were 85.5% and 70.6% respectively. The dry matter of pig manure contained 340 g kg⁻¹ C, 6.0 g kg⁻¹ total N, 4.5 g kg⁻¹ total P and 5.0 g kg⁻¹ total K, and that of Chinese milkvetch contained 467 g kg⁻¹ C, 4.0 g kg⁻¹ total N, 1.1 g kg⁻¹ total P and 3.5 g kg⁻¹ total K.

In upland soil, the fertilizer rates were 60 kg N ha⁻¹, 30 kg P₂O₅ ha⁻¹, and 60 kg K₂O ha⁻¹ in each season. The P and K fertilizers and the pig manure were applied before maize seeding. The N fertilizer was applied 70% as basal fertilizer and 30% as topdressing fertilizer. While in paddy soil, the fertilizer rates were 90 kg N ha⁻¹, 45 kg P₂O₅ ha⁻¹, and 75 kg K₂O ha⁻¹ for each rice season. P and organic fertilizers were applied prior to rice seedling transplantation. Both N and K fertilizers were applied as topdressing fertilizers. The N fertilizer was divided into two parts and applied twice, whereas the K fertilizer was applied once.

2.3. Soil sampling and analysis

All soil samples were collected from the 0–20 cm deep layer in May 2015. In each plot, eight cores were randomly collected and pooled together as one composite sample. Subsequently, the composite samples were air-dried at room temperature and passed through a 2-mm sieve for further analysis. The C contents were measured by a Vario EL III Elemental Analyzer (Elementar, Germany).

2.4. Soil SOM fractionation

Functional SOM fractions were separated using a combined physical, chemical, and density fractionation method as described by Stewart et al. (2008, 2009) (Fig. 1). In the first step, three size fractions were obtained using physical fractionation and partial dispersion. They consisted of > 250 μm coarse unprotected particulate organic matter (cPOM), 53–250 μm microaggregate fraction (μagg), and < 53 μm easily dispersed silt and clay (dSilt and dClay). All the obtained fractions were oven-dried at 60 °C and weighed.

In the second step, the microaggregate fraction isolated in the first step was further fractionated. Density flotation was used to isolate the fine unprotected POM fraction (fPOM) with 1.85 g cm⁻³ sodium polytungstate. After fPOM was removed, dispersion was conducted for the heavy fraction to separate the > 53-μm microaggregate-protected POM fraction (iPOM) and the microaggregate-derived silt- and clay-sized fractions (μSilt and μClay).

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