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Effects of long-term inorganic and organic fertilizations on the soil micro and macro structures of rice paddies

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The soil structure of paddy soil is very dynamic from the aggregate to the pedon scale because of intensive anthropogenic management strategies. In this study, we tested the hypothesis that long-term inorganic and organic fertilizations can affect soil structure at different scales. Microstructure assessed by soil aggregates (3–5 mm in diameter) and macrostructure assessed by small soil cores (CoreS) (5 cm in diameter, 5 cm in height) and large soil cores (CoreL) (10 cm in diameter, 10 cm in height) were sampled from three long-term fertilization treatments, including no fertilizer (CK), application of inorganic fertilizer (NPK), and a combination of inorganic fertilizer and organic manure (NPKOM), established in 1982. They were scanned at two scales with two types of micro-computed tomography (micro-CT) and quantified using image analysis. Results showed that relative to CK treatment, long-term NPKOM fertilization increased soil organic C (SOC) by 28% and available water content (AWC) by 20%, but decreased soil bulk density by 0.2 g cm−³ whereas NPK showed no difference. Soils under CK and NPK treatments exhibited an identical dense structure at both aggregate and core scales in which pores were mainly cracks resulting from shrink/swell processes, and showed no significant difference in porosity and size distribution of the CT-identified pores $(>3.7 \mu m)$. Compared with the CK treatment, the soil in the NPKOM treatment had greater intra- and inter-aggregate pores, and increased porosity by 58.3%, 144.9%, and 65.9% at aggregate, CoreS, and CoreL scales, respectively. These were attributed to the biopores formed from decayed roots, stubble, and organic manures as a result of increased yields and direct amendment of organic manure. Overall, this study demonstrates that organic fertilization can improve the physical qualities of paddy soils across different scales but inorganic fertilization in isolation does not.

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1. Introduction

Soil structure is a fundamental property of soil health because it impacts the storage and movement of water, gas and nutrients, root growth, and microbe activity [\(Bronick and Lal, 2005](#page--1-0)). Soil structure can be assessed over several orders of scales from mineral–organic complexes, aggregates, typically referred as soil microstructure, to peds and clods in the soil profile, usually considered soil macrostructure [\(Carter,](#page--1-0) [2004\)](#page--1-0). And the size of the corresponding pores ranges from μm to mm or even larger. [Tisdall and Oades \(1982\)](#page--1-0) proposed that the factors and processes controlling the formation of soil structure are different at contrasting scales in an aggregate hierarchy concept model. Management practices, e.g. tillage and fertilization, have been proven to impact each level of soil structure either directly or indirectly ([Bronick and](#page--1-0) [Lal, 2005\)](#page--1-0). [Kravchenko et al. \(2011\)](#page--1-0) showed large intra-aggregate

⁎ Corresponding author. E-mail addresses: zhouhu@issas.ac.cn (H. Zhou), xhpeng@issas.ac.cn (X. Peng). pores in no tillage and native succession vegetation treatments are more heterogeneous than those in conventional tillage treatment. Macropores $(>0.75$ mm) were more abundant in pastureland than under arable crops and provided pathways for preferential flow at the core scale [\(Luo et al., 2008](#page--1-0)). Despite the numerous evaluations of land use and management effects on soil structure, most studies have been limited to a specific scale and knowledge of the responses of soil structure at different scales is lacking.

Information about a soil's inner structure has usually been inferred from soil properties (e.g., hydraulic properties and gas permeability) [\(Hill et al., 1985](#page--1-0); [Marshall, 1958](#page--1-0); [Moldrup et al., 2001\)](#page--1-0). These calculations were based on assumptions of ideal pore shapes and typically could not provide information regarding the architecture of soil pore system. Therefore, a direct study of soil structure is necessary. Direct observation and quantification of the structure of soil were typically conducted on soil thin sections [\(Pagliai et al., 2004](#page--1-0); [Mooney et al.,](#page--1-0) [2007](#page--1-0)). However, soil thin section can only provide two-dimensional (2D) information of soil structure. And the preparation of thin sections is time consuming ([Murphy, 1986\)](#page--1-0). Computed tomography (CT) offers

a rapid and non-destructive way to study soil structure over a range of scales [\(Taina et al., 2008](#page--1-0); [Wildenschild et al., 2002](#page--1-0); [Helliwell et al.,](#page--1-0) [2014](#page--1-0)). High-resolution CT can show the detailed organization of soil aggregates and has been used to study aggregation processes [\(Atkinson](#page--1-0) [et al., 2009](#page--1-0); [Zhou et al., 2013\)](#page--1-0), soil microstructure ([Peth et al., 2008](#page--1-0)), and soil biophysical interactions [\(Martin et al., 2012](#page--1-0); [Vos et al., 2013](#page--1-0)) at the aggregate scale. CT with a low resolution, on the other hand, can scan large samples and is frequently used to study macropores and their relationship with soil hydraulic properties ([Luo et al., 2008](#page--1-0)) at the soil core scale. The study of the micro and macro scale soil structure is possible by using a combination of CT systems with different resolution capabilities. [Schlüter et al. \(2011\)](#page--1-0) studied soil structure development at two different scales and proposed a method to combine soil pore size distribution (PSD) acquired at the different scales. [Dal](#page--1-0) [Ferro et al. \(2013\)](#page--1-0) found that both the micro- and macro-scale soil structures were affected by fertilization from the scanning of soil aggregates and soil cores using micro-CT.

Rice is the most important staple food in China and the cultivation area of rice is 25 million ha, accounting for 25% of the national arable land area [\(Li, 1992\)](#page--1-0). Long-term traditional cultivation of rice, specifically flooding during most of the growing season, drastically changed soil physical, chemical, and biological properties and resulted in a special anthropogenic paddy soil [\(Gong, 1986\)](#page--1-0). The structure of paddy soil is more dynamic at the aggregate to soil core scales compared with those of upland soils. The plow layer of the paddy soil is homogenized before each growing season to prepare the seedbed, which destroys surface soil structure considerably [\(Eickhorst and Tippkötter,](#page--1-0) [2009](#page--1-0); [Kirchhof et al., 2000;](#page--1-0) [Sharma and Datta, 1986\)](#page--1-0). Moreover, paddy soil experiences frequent swell-shrink cycles caused by periodic flooding and drying management [\(Zhang et al., 2013](#page--1-0)). These processes are accompanied by the creation and closing of cracks which has critical importance for the evolution of the structure of paddy soil [\(Liu et al.,](#page--1-0) [2003;](#page--1-0) [Sander and Gerke, 2007](#page--1-0)). At the micro-scale, aggregation of paddy soil is greatly influenced by the oxidation-reduction conditions caused by flooding and drainage cycles [\(Kögel-Knabner et al., 2010](#page--1-0)). For example, Fe oxides are important binding agents of soil aggregates, but their effects vary among different Fe species [\(Duiker et al., 2003](#page--1-0)). Poorly crystalline Fe has a larger and more reactive surface area and therefore is more effective in soil aggregation than crystalline Fe [\(Duiker et al., 2003;](#page--1-0) [Yan et al., 2013\)](#page--1-0). Repeated flooding and drainage cycles have been shown to increase $Fe₀$ oxides while reducing Fe_d oxides; therefore, these processes are beneficial to soil aggregation [\(Zhang et al.,](#page--1-0) [2003](#page--1-0)).

The application of organic or inorganic fertilizers can directly or indirectly introduce different ions and organic matter to the soil, which may cause soil disaggregation or aggregation ([Haynes and Naidu, 1998](#page--1-0)). In the past few years, research regarding the effects of fertilization on paddy soil has mostly focused on SOC sequestration ([Anders et al.,](#page--1-0) [2012](#page--1-0); [Brar et al., 2013;](#page--1-0) [Das et al., 2014\)](#page--1-0), greenhouse gas emissions [\(Yagi and Minami, 1990](#page--1-0)), and microbial and geochemical processes [\(Zhong and Cai, 2007\)](#page--1-0) due to environmental and ecological concerns. These processes are closely linked with soil structure, which determines the transport of water, gas and solutes and provides a habitat for soil microorganisms [\(Young and Crawford, 2004](#page--1-0)). Although the change in aggregate stability under fertilization in paddy soils has been evaluated [\(Li and Zhang, 2007](#page--1-0); [Huang et al., 2010;](#page--1-0) [Yan et al., 2013](#page--1-0)), the effect of fertilization on the formation and dynamics of the structure of paddy soil is still unclear.

To better understand the sustainability of paddy soil to continuously received inorganic and organic fertilizers, this study aimed to evaluate the soil micro and macro structure of a long-term fertilization experiment. The specific objectives were: (1) to evaluate the effects of fertilization on aggregate- and core- scale structure using synchrotron based micro-CT and industrial micro-CT and (2) to investigate the mechanisms of the structure evolution of paddy soil.

2. Materials and methods

2.1. Experimental site

Soil was taken from long-term experiment established in 1982 at the Jiangxi Institute of Red Soil, Jinxian County, Jiangxi Province, China (116°10′ E, 28°21′ N). The experiment site lies in a flat area of the hilly region of Southern China. The experiment site has a subtropical climate and a mean annual temperature and precipitation of 17.7 °C and 1706 mm, respectively. The paddy soil (Typic Stagnic Anthrosols, Chinese Soil Taxonomic Classification, 2002) is clay loam (20% sand, 48% silt, and 32% clay) for the plow layer (0- to 15 cm). Before the long-term experiment, the paddy soil had organic C 16.3 g kg⁻¹, total N 1.49 g kg^{-1} , and pH 6.9 in the plow layer. The site had been cultivated with rice for more than 100 years prior to the experiment. The cropping system is early rice–late rice from April to October and fallow in the winter.

The experiment was designed as a randomized complete block with three replicates. Three fertilization treatments were studied: (1) no fertilization as a control (CK), (2) a combination of inorganic fertilizers (NPK), including 90 kg N ha⁻¹, 20 kg P ha⁻¹, and 62 kg K ha⁻¹ for each season; and (3) organic manure and the inorganic fertilizers (NPKOM) together, including the same amount of inorganic fertilizer as the NPK treatment plus 22.5 t ha^{-1} pig manure. Each plot had an area of 46.67 m². A detailed description of the management of the field experiment can be found in [Yan et al. \(2013\).](#page--1-0)

2.2. Sampling

Sampling was conducted in September 2012 just before the harvest of late rice. Two sizes of undisturbed soil cores, a large size (diameter 10 cm, height 10 cm, CoreL) and a small size (diameter 5 cm, height 5 cm, CoreS), were randomly collected using PVC tubes with triplicates in each plot. The tubes were gently pushed into the topsoil and were excavated using a spade. Soil cores were wrapped with plastic film to prevent water evaporation and stored in the refrigerator at 4 °C. A total of 27 CoreL and 27 CoreS were sampled. Bulk soil was also sampled with a spade from the 0–10 cm depth. In each plot five samples were randomly collected and they were then mixed together to form one bulk sample. The bulk soil samples were manually broken to small parts $(< 8$ mm) and air-dried at room temperature. Care was taken to prevent compression during sampling and breaking.

2.3. CT scanning and image reconstruction

Both CoreL ($n = 27$) and CoreS ($n = 27$) were scanned at field moisture (0.30–0.35 $\text{cm}^3 \text{ cm}^{-3}$) using an industrial Phoenix Nanotom X-ray μ-CT (GE, Sensing and Inspection Technologies, GmbH, Wunstorf, Germany). Detailed scan parameters are listed in [Table 1.](#page--1-0) The voltage and current were higher for CoreL than for CoreS because more energy was needed to penetrate larger samples. A 0.2 mm Cu filter was used to reduce the beam hardening effect. The distance between the source and the sample and between the sample and the detector was 30 cm and 20 cm, respectively. At this distance, the detector could fully capture the signal of CoreS but detector shift was needed to acquire the full image of the CoreL samples. Reconstruction was performed using the Datos \times 2.0 software using the filtered back-projection algorithm. This generated slices of 4000 \times 4000 and 2000 \times 2000 voxels for CoreL and CoreS, respectively, with each voxel representing a volume of 30 \times 30 \times 30 μ m³. The slices were stored in 8-bit format, which means that each voxel had a value between 0 and 255 representing the attenuation coefficient of the corresponding material.

The scanning of aggregates from the bulk samples was conducted with a synchrotron-based μ-CT at beam line BL13W1 of the Shanghai Synchrotron Radiation facility (SSRF). Air-dried aggregates (3–5 mm) were first collected by sieving the bulk samples and then randomly

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