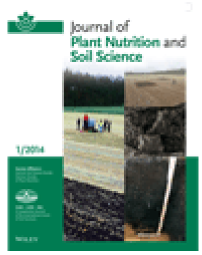

Intensive organic vegetable production increases soil organic carbon but with a lower carbon conversion efficiency than integrated management

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Intensive organic vegetable production increases soil organic carbon but with a lower carbon conversion efficiency than integrated management

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Abstract

Intensive vegetable production in greenhouses has rapidly expanded in China. We conducted an 11-year greenhouse vegetable production experiment from 2002 to 2013 to observe soil organic carbon (SOC) dynamics under three management systems, i.e., conventional (CON), integrated (ING), and intensive organic (ORG) farming. Soil samples (0–20 and 20–40 cm depth) were collected in 2002 and 2013 and separated into four particle-size fractions, i.e., coarse sand (> 250 μm), fine sand (250–53 μm), silt (53–2 μm), and clay (< 2 μm). The SOC content and $\delta^{13}C$ values of the whole soil and the four particle-size fractions were analyzed. After 11 years, ORG and ING significantly increased the SOC stocks (0–20 cm) to 4008±36.6 and 2880±365.1 kg C ha^{-1} y^{-1}, respectively, 8.1- and 5.8-times that in CON (494**±**42.6 kg C ha^{-1} y^{-1}). The SOC stock increase in ORG at 20–40 cm depth was 245±66.4 kg C ha^{-1} y^{-1}, significantly higher than in ING (65.5±13.4 kg C ha^{-1} y^{-1}) and CON (109±44.8 kg C ha^{-1} y^{-1}). Analyses of ^{13}C revealed a significant increase in newly produced SOC in both soil layers in ORG. However, the carbon conversion efficiency (CE) was lower in ORG (14.4%–21.7%) than in ING (18.3%–27.4%). Among the four particle-sizes in the 0–20 cm layer, the silt fraction showed the largest increase in SOC content (59.5% and 41.2% of the whole SOC increase in ORG and ING, respectively). A similar trend was detected in the 20–40 cm soil layer. Thus, ORG increased the SOC level, but its CE was lower than that of ING. The increased SOC content in the silt and clay fractions in ORG and ING suggested that organic fertilizer has an important role in stabilizing SOC.

Keywords: Greenhouse vegetable production; carbon conversion efficiency; organic carbon sequestration; particle-size fraction; $\delta^{13}C$; organic fertilizer

1. Introduction

Soil is the largest carbon (C) pool on earth, with organic carbon (OC) of 1550 Pg and inorganic carbon (IC) of 950 Pg (*Lal*, 2007). Therefore, a small change in soil C content can affect the global climate (*IPCC*, 2014; *Luo* et al., 2010). Land use/cover changes, especially agricultural activities, significantly affect ecosystem services including soil organic carbon (SOC) storage (*Gelaw* et al., 2014; *Wang* et al., 2016). It is important to understand the C dynamics within agro-ecosystems and identify appropriate farming practices to protect soil resources and provide adequate food and fiber for a growing population (*Stockmann* et al., 2013). Numerous studies have explored the impacts of irrigation (*Houlbrooke* et al., 2008; *Kelliher* et al., 2012), fertilization (*Lemke* et al., 2017; *Purakayastha* et al., 2008; *Yan* et al., 2017), tillage (*Franzluebbers* and *Steiner*, 2016; *West* and *Post*, 2002), and land use (*Aluri*, 2016; *Tan* et al., 2007; *Wiesmeier* et al., 2015) on SOC content and stocks in farmland soils. The findings so far have highlighted that there is great potential to increase C stocks in agricultural soil through various practices including organic farming (*Lal*, 2004; *Liao* et al., 2015; *Matsuura* et al., 2018). The results of other studies have suggested that the beneficial effects of organic farming on SOC are largely determined by disproportionately high applications of organic fertilizer compared with conventional farming (*Leifeld* and *Fuhrer*, 2010), and that the SOC increase due to organic fertilizer is not genuine carbon sequestration (*Powlson* et al., 2011).

In China, vegetable production in greenhouses has developed rapidly over the past two decades driven by economic development and increased consumer demands (*Guo* et al., 2012; *Yu*, 2011). Many farmers have shifted from conventional cereal cropping to greenhouse production systems (*Qiu* et al., 2010). China's total vegetable production reached 596.1 million Mg in 2014 (*FAO*, 2015), accounting for 51.0% of the world's production. In China, typical greenhouse vegetable production has two seasons per year. This production system achieves relatively high yields (up to 150 Mg ha^{-1}, average of 63 Mg ha^{-1} for tomatoes) through intensive farming methods. This approach includes high levels of organic and mineral fertilization (> 1000 kg N ha^{-1} per growing season from manure and fertilizer applications) (*Chen* et al., 2004; *He* et al., 2007; *Yuan* et al., 2015), frequent irrigation (8–11 irrigation events with total water input of 450–560 mm per growing season) (*Hou* et al., 2014; *Song* et al., 2012), and measures to maintain higher temperatures in greenhouses over winter (including thick back earth walls, plastic film, and cotton quilt coverings) (*Chen* et al., 2004; *Song* et al., 2012). In 2016, there were 61,000 ha of organic vegetable farmland in China (*Qiao* et al., 2018).

Compared with cereals, vegetables require more intensive management, including more frequent farming operations and more external nutrient inputs (*Power* and *Schepers*, 1989). However, some researchers have noted that the yield of vegetables does not increase linearly with the amount of fertilizer applied. Vegetable yields will decrease or at best level-off with excessive fertilizer (*Chang* et al., 2007). *Liang* et al. (2009) found that the application of large amounts of organic fertilizer alone did not increase vegetable yield compared with the application of combined organic and chemical fertilizers.

Intensive fertilizer inputs may influence the quality and sustainability of soil in greenhouses. In addition, different types of fertilizers have different effects on SOC. Studies have shown that organic manure can directly influence SOC by increasing C input. However, compared with inorganic fertilizer, organic fertilizer alone or in combination with inorganic fertilizers has been found to be more effective in increasing SOC and its fractions (*Blair* et al., 2006; *Rudrappa* et al., 2006; *Purakayastha* et al., 2008). Vegetables have been produced in greenhouses in China since the 1990s (*Chen* et al., 2017). However, the optimum amount of organic fertilizer (or the optimal ratio of organic fertilizer combination with inorganic fertilizer) in terms of vegetable yield and soil quality is still unknown. Furthermore, few studies have compared the long-term impacts of conventional and intensive organic vegetable production, particularly in terms of dynamic changes in SOC.

Previous studies have used a stable isotope approach to explore soil C (*Kuzyakov* and *Bol*, 2006; *Schneckenberger* and *Kuzyakov*, 2007; *Shahbaz* et al., 2017). In both labeling experiments and natural isotope labeling, the natural ^{13}C range is useful to trace the mechanisms of SOC transformation and turnover. As the basic unit of soil structure (*Qian* et al., 2018; *Rabot* et al., 2018), soil particles are crucial for SOC stabilization (*Bol* et al., 2009). The SOC stocks in soil depend on the interactions between organic materials/matter and soil particles (*Mikha* et al., 2015; *Six* and *Paustian*, 2014). Particle-size fractionation is based on the concept that SOC is associated with different-sized particles with different structural and functional properties (*Bol* et al., 2009; *Christensen*, 1992). Thus, analyses of ^{13}C combined with soil particle-size fractionation can be used to assess the proportion of newly added C in different particle-size fractions and the SOC mean residence time (MRT) (*Six* et al., 2002b; *Beniston* et al., 2014). This method is useful for studying SOC dynamics and has been widely used in research on farmland ecosystems. For example, using ^{13}C and soil particle-size and density fractionation methods, *Meng* et al. (2014) found that incorporating whole straw combined with tillage retained more antecedent SOC than did the incorporation of pulverized straw. *Beniston* et al. (2014) reported that MRT of prairie-derived SOC 75 years after a land-use change to cropland were longer in fractions with smaller particle sizes.

In this study, we sampled greenhouse vegetable production soils under intensive organic (ORG), integrated (ING), and conventional (CON) farming management for 11 years. We mainly focused on the fertilizer-driven effects on SOC under these three management systems. The soil was separated into different particle-size fractions and the ^{12}C and ^{13}C contents were determined. The objectives were to: (1) trace SOC transformation in different sizes of soil particles; (2) compare the efficiency of ORG, ING and CON greenhouse production on SOC sequestration; and (3) calculate carbon conversion efficiency (CE: increased organic carbon in soil divided by input organic carbon) to analyze the efficacy of different management schemes to increase SOC in greenhouse production. The overall aim of our research is to identify optimal nutrient management practices for sustainable soil productivity and C sequestration in greenhouse vegetable production.

2. Material and methods

2.1 Experimental design

In 2002, a long-term experiment was initiated at the Quzhou Agricultural Experiment Station, China Agricultural University, Hebei Province (36°52'N, 115°100'E). The site is approximately 37 m above sea level. The region has a warm temperate climate, with a semi-humid continental monsoon and an average annual temperature of 13.2°C ranging from a maximum of 26.8 °C in July to a minimum of −2.9 °C in January. Mean annual precipitation is 543 mm, mainly occurring in July to September, and the average annual sunshine duration is 2330 h (*Bughio* et al., 2016). The parent material of the soil in the region is holocene Loess (10000 a B.P.) (*Wang* et al., 2002). The soil is classified as silt fluvo-aquic soil. When the experiment was set up, the soil properties (0–20 cm depth) were 7.6 pH ($CaCl_2$), 20 mg kg^{-1} available P (sodium bicarbonate method), 278 mg kg^{-1} available K (flame photometry), 1.24 g kg^{-1} total N (Kjeldahl method), and 16.9 g kg^{-1} soil organic matter (SOM).

The experiment consisted of three farming management systems: (1) conventional management (CON), i.e., organic fertilizer (39 Mg ha^{-1} y^{-1}), urea (115 kg ha^{-1} y^{-1}), calcium superphosphate (100 kg ha^{-1} y^{-1}), and potassium chloride (80 kg ha^{-1} y^{-1}); (2) integrated management (ING), i.e., organic fertilizer (72.6 Mg ha^{-1} y^{-1}), urea (57.5 kg ha^{-1} y^{-1}), calcium superphosphate (50 kg ha^{-1} y^{-1}), and potassium chloride (40 kg ha^{-1} y^{-1}). In CON and ING, insecticides (cyhalothrin, imidacloprid, and cartap) and fungicides (dimethomorph, carbendazim, cymoxanil, and mancozeb) were used to control insects and diseases according to the local management systems. (3) Intensive organic management (ORG), in which all nutrients were supplied by organic fertilizers (132.6 Mg ha^{-1} y^{-1}). Organic management was conducted without chemicals, so insects were controlled by manual and physical means, e.g. manual insect control and the use of insect-proof screens and yellow boards. Where needed, diseases were controlled using a biological fungicide (Wuyiencin) and sulfur fumigation.
Consistency of the organic fertilizer applied under the three treatments was ensured during the experiment. The organic fertilizer consisted of cow dung and chicken dung and was composted for about 1 month. The carbon content of organic fertilizer was 22.2% and its $\delta^{13}C$ was −14.52‰. The organic fertilizer containing 1.21% N, 0.60% P_2O_5, and 1.58% K_2O on a dry weight basis. All management systems were flood-irrigated 6–8 times with groundwater at the rate of 350–390 mm ha^{-1} y^{-1}. The tillage method under the three management systems was ploughing to a depth of 20–30 cm.

One greenhouse (52 m × 7 m, 0.04 ha) was established for each management system. Each greenhouse was separated from the neighboring greenhouse by distance of 2.5 m. The vegetable greenhouses had a semi round-arch structure. Within each greenhouse (management system), three subplots (9 m × 7 m) as pseudo-replicates were arranged with a distance of 4 m between subplots. The subplots were planted in rotation with tomato (*Lycopersicon esculentum* Mill), cucumber (*Cucumis sativus* Linn), celery (*Apium graveolens* L.), fennel (*Foeniculum vulgare* Mill), cauliflower (*Brassica oleracea* L.), and eggplant (*Solanum melongena* L.). Two (mainly tomato and cucumber) or three types of vegetables (tomato or cucumber with two types of leaf vegetables) were planted annually. About 100 m from the experimental greenhouse, a natural planting (NP) plot was chosen as a native undisturbed reference site, which had received no cropping or any other anthropogenic perturbation since 1974. The plot had been intentionally reserved since 1974 to observe the impacts of land use change and farming practices on soil properties (*Bughio* et al., 2016).

2.2 Soil sampling and analysis

After the tomato harvest in September 2013, composite soil samples were collected using a soil auger (diameter 3.0 cm) from two depths; 0–20 cm and 20–40 cm. Three cores of soil samples from each replicate (three replicates from each management system/greenhouse and three pseudo-replicates of NP) were mixed into composite samples. Archived soil samples (0–20 and 20–40 cm) collected in 2002 using the same sampling methods as in 2013 were also available for analysis. Soil samples were air-dried, crushed, and passed through a 2.0 mm sieve. Subsamples were ground and passed through a 0.15 mm sieve before SOC content and isotopic analyses.

Soil samples were prepared for SOC analysis following a procedure adapted from *Bughio* et al. (2016). Briefly, soil samples were soaked in 0.5 M HCl for 12 h to remove carbonates, then washed with de-ionized water and centrifuged. This procedure was repeated until the pH of the solution was neutral to ensure all excess acid had been removed. The soil was oven-dried at 60 °C. Then, the SOC contents were analyzed with a Finnigan Delta-Plus XP mass spectrometer at the Chinese Academy of Agricultural Sciences.

2.3 Particle-size fractionation of soil

The soil was fractionated into different particle-size fractions as described by *Cambardella and Elliott* (1992) and *Beniston* et al. (2014) with some modifications. Soil samples were separated into four particle-size fractions, i.e., coarse sand (> 250 µm), fine sand (250–53 µm), silt (53–2 µm), and clay (< 2 µm). Briefly, each 20 g soil sample was dispersed in 60 ml 5 g L^{-1} sodium hexametaphosphate with shaking for 16 h. The soil slurry was then passed through a 25-µm sieve and then a 53-µm sieve. The residual finer fractions were separated into silt and clay fractions by repeated centrifugation. All fractions were washed into pre-weighed glass beakers and oven dried at 45°C. After drying, soil was ball-milled and carbonates in the soil were removed by soaking in 0.5 M HCl for 12 h using the same method as that used in the determination of total SOC described above and simultaneously measure $\delta^{13}C$ values. The C contents were corrected for the loss of soil weight during acidification.

The SOC content in each particle-size fraction was calculated using Eq. (1) (*Sollins* et al., 1999). The SOC content in each particle-size fraction was multiplied by the mass of that fraction, as follows:

$$C_{(fr)} = C_{fr} \times W_{fr} \quad (1)$$

where, $C_{(fr)}$ is SOC (g C kg soil^{-1}) in each fraction, C_{fr} is total SOC in that fraction (g C kg^{-1} fraction), and W_{fr} is the dry mass of that fraction (g fraction^{-1} g soil^{-1}).

2.4 SOC stocks and carbon conversion efficiency

We used two methods to calculate SOC stocks. (1) The soil bulk density (BD) was determined by the ring cutter method during sampling, and then the SOC stocks were calculated by multiplying the carbon content in soil BD by the soil volume. The soil BD values in the 0–20 cm soil layer in 2002 were CON (1.15 g cm^{-3}), ING (1.12 g cm^{-3}), and ORG (1.10 g cm^{-3}); and in 2013 were CON (1.12 g cm^{-3}), ING (1.09 g cm^{-3}), and ORG (1.05 g cm^{-3}). The soil BD values in the 20–40 cm layer in 2002 were CON (1.30 g cm^{-3}), ING (1.28 g cm^{-3}), and ORG (1.26 g cm^{-3}); and in 2013 were CON (1.28 g cm^{-3}), ING (1.26 g cm^{-3}), and ORG (1.24 g cm^{-3}). (2) The minimum equivalent soil mass was used to evaluate SOC stocks, using the calculations reported by *Juhwan* et al. (2009).

We used Eq. 2 to calculate the carbon conversion efficiency (CE) to evaluate the efficacy of different fertilization schemes to increase SOC under greenhouse production:

$$CE = \frac{\Delta SOC}{F * C\%} \quad (2)$$

where, CE= conversion efficiency of carbon, F= amount of organic fertilizer applied each year, C%= carbon content in applied organic fertilizer, and ΔSOC=the increase in SOC in each year.

2.5 Calculation of SOC turnover

Only vegetables using the C3 pathway (with $\delta^{13}C$ ca. −26‰) were grown in the greenhouse from 2002 to 2013. The applied organic fertilizer consisted of a mixture of cow dung and chicken manure. The cattle and chicken feed was mainly (C4) corn and corn straw. The fertilizer $\delta^{13}C$ was −14.52‰ and its SOC content was 22.2%. Hence, organic fertilizer was the main C4 input source, and it increased the $\delta^{13}C$ values of SOC over the experimental period. Eq. (3) was used to estimate the proportion of original SOC and newly input SOC from organic fertilizer in the SOC pool:

$$f = \frac{(\delta - \delta_0)}{(\delta_1 - \delta_0)} \quad (3)$$

where, f = the fraction of newly input SOC derived from organic fertilizer, δ = $\delta^{13}C$ value of SOC in 2013, δ_1 = $\delta^{13}C$ value of organic fertilizer, and δ_0 = $\delta^{13}C$ value of SOC in 2002.

2.6 Statistical analysis

Statistical analyses were performed with SAS (SAS Institute Inc., Cary, NC, USA). Data were tested for normality using the Shapiro-Wilk test and then analyzed by two-way analysis of variance (ANOVA) with "management system (T)" and "year (Y)" as fixed effects (in Fig.1–Fig.4). Differences between the means greater than the least significant difference (LSD) were considered statistically significant ($P < 0.05$). T-tests were conducted to assess statistical differences between samples collected in 2002 and 2013, and significant differences are indicated by asterisks: * ($P < 0.05$) or ** ($P < 0.01$). Reported data are mean ± standard error (SE) (n=3). In addition, data were analyzed by one-way ANOVA in Table 2, Table 3 and Table 5.

3. Results

3.1 SOC content in different particle-size fractions

After 11 years (from 2002 to 2013) of continuous greenhouse vegetable production, SOC contents in the 0–20 cm soil layer were significantly increased in all particle-size fractions in ORG and ING. The relative increases in SOC content (0–20 cm) during the 11-year experiment were 24% (coarse sand) for CON, 13% (coarse sand), 199% (fine sand), 234% (silt), and 153% (clay) for ING; and 57% (coarse sand), 179% (fine sand), 236% (silt), and 186% (clay) in ORG. In the 20–40 cm soil layer, the SOC increases were significant only for the coarse sand fraction, i.e., 41% (CON), 124% (ING), and 142% (ORG).

3.2 Whole-soil based SOC content

After the 11-year experiment, the proportion of sand was almost unchanged in the three management systems. The proportion of silt had decreased and the proportion of clay had increased (Table 1). At 0–20 cm depth, silt accounted for the largest proportion of total soil (81.5%–85.3%), followed by coarse sand (0.3%–4.2%), fine sand (4.8%–8.0%), and clay (6.3%–10.6%, Table 1). The whole-soil based particle SOC content was highest in the silt fraction, followed by sand and then clay (Fig. 2b–e). At 20–40 cm depth, however, the order was silt > clay > fine sand > coarse sand (Fig. 2b–e).

At the 0–20 cm depth, the SOC contents were significantly increased after 11 years of greenhouse vegetable production in ING (8.5 to 23.3 g C kg^{-1} soil, 173%) and ORG (9.3 to 31.8 g C kg^{-1} soil, 241%), but not in CON (Fig. 2a). The whole-soil based SOC content in the four particle-size fractions (particularly in the fine sand and silt fractions) also showed significant increases in both ING and ORG (Fig. 2b–e). We calculated the contribution of each particle-size fraction to the whole-soil SOC content after 11 years of the experiment. In ING and ORG, silt contributed the highest proportion (55.4%–57.8%), followed by fine sand (20.6%–20.8%), coarse sand (13.8%–14.8%), and clay (7.8%–8.9%, Table 2). In CON, fine sand made the largest contribution (64%), followed by clay (29.1%), coarse sand (4.2%), and silt (2.7%, Table 2).

For the 20–40 cm layer, the increase in whole-soil OC content in the 11-year experiment was only significant in ORG (4.2 to 5.4 g C kg^{-1} soil, 27%). Among the soil fractions, the largest increase in SOC was in the coarse sand fraction (212%), followed by the clay fraction (47.4%; Fig. 2b–e). In ORG, the contributions to the increase in whole-soil SOC content were similar for the silt (36.9%), clay (30.0%), and coarse sand (23.4%) fractions, and lowest for the fine sand fraction (9.7%) (Table 2). In ING, coarse sand made the largest contribution (58.2%), followed by silt and clay (29.6% and 26.0%). In CON, clay made the largest contribution (98.8%), followed by silt (25.0%) (Fig. 2e). The SOC content in the sand fraction decreased in CON, so this fraction negatively contributed to the whole-soil SOC increase (Fig. 2b–c).

After the 11-year experiment, compared with CON, the ORG and ING management systems increased the whole-soil OC content by 108.3% and 52.6% (0–20 cm), and 50.5% and 16.1% (20–40 cm), respectively (Fig. 3). In the 0–20 cm soil layer, the silt fraction contributed 41.2% (ING) and 59.5% (ORG) of the total SOC increase, followed by coarse sand, fine sand, and clay. In the 20–40 cm soil layer, the silt fractions made the largest contributions to the increase in whole-soil OC content (67.8% for ING and 66.5% for ORG), followed by coarse sand and fine sand. The clay fraction did not contribute to the whole-soil OC increase because its proportion of total soil mass did not differ among the three management systems (Table 1, Fig. 2e).

3.3 SOC stocks and carbon conversion efficiency

From 2002 to 2013, SOC stocks increased by 5.4, 31.7, and 44.1 Mg C·ha^{-1} in CON, ING, and ORG (Table 3). The corresponding SOC stock increase rates in ING and ORG were 2880±365 and 4008±36.6 kg C·ha^{-1} y^{-1}, respectively, or 8.1-fold and 5.8-fold that in CON (494±42.6 kg C·ha^{-1} y^{-1}) ($P < 0.01$). Compared with NP, the CON, ING, and ORG management systems showed SOC stock increase rates of 510±83.5, 2017±355, and 3470±67.4 kg C ha^{-1} y^{-1}, respectively.

Similarly, for the 20–40 cm depth, the SOC stock increases after 11 years of CON, ING, and ORG were 1.2, 0.7, and 2.7 Mg ha^{-1}, respectively, and the corresponding SOC stock increase rates were 109±44.8 (CON), 66±13.4 (ING), and 245±66.4 kg C·ha^{-1} y^{-1} (ORG). Compared with NP, ORG had a SOC stock increase of 254±20.0 kg C ha^{-1} y^{-1}, significantly higher than those of ING and CON (55–92 kg C ha^{-1} y^{-1}). To eliminate the impacts of soil bulk density during the calculation of SOC stocks, we also calculated the minimum equivalent soil mass to quantify the SOC stocks and C increase rates. The results obtained using those calculations were similar (Table 3).

Because the tillage depth in this experiment was 20–30 cm, we calculated CE for the two extreme cases, that is, plough depths of 20 cm and 30 cm (Table 4). In either case, the treatments were ranked, from highest total CE to lowest, as follows: ING (18.3%–27.4%) > ORG (14.4%–21.7%) > CON (7.0%–10.5%).

3.4 $\delta^{13}C$ values of SOC in whole-soil and particle-size fractions

At 0–20 cm depth, the $\delta^{13}C$ values of SOC in the coarse sand and silt fractions differed significantly among the three management systems, i.e., ORG > ING > CON (Fig. 4). For the fine sand and clay fractions, the differences among the three management systems were not significant. After 11 years of fertilization, the $\delta^{13}C$ values were significantly increased in all particle-size fractions in three management systems ($P < 0.05$).

Similarly, at 20–40 cm depth in 2013, the $\delta^{13}C$ values of all fractions were in the order of ORG > ING > CON, although differences were significant only in the silt and clay fractions (Fig. 4). The $\delta^{13}C$ values were also significantly increased by 11 years of fertilization in all management systems and all fractions, except for the clay fraction in ING.

The $\delta^{13}C$ values of SOC were higher in the 0–20 cm layer than in the 20–40 cm layer after 11 years of organic fertilization. This was due to the input of exogenous organic materials. At the start of the experiment, the greenhouse was only planted with C3 crops ($\delta^{13}C$ of −26 ‰). As the organic C input from vegetable roots (all vegetables and their roots were harvested and removed from the greenhouse) was negligible, the $\delta^{13}C$ of SOC was mainly affected by exogenous organic materials ($\delta^{13}C$ of −14.5‰).

3.5 Contribution of fertilization to SOC turnover

The contributions of fertilization to new SOC in particle-size fractions from 2002 to 2013 were calculated using Eq. (3) and are shown in Table 5. In the 0–20 cm soil layer, fertilization most strongly affected the silt and sand (coarse and fine sand) fractions and the proportion of new SOC was > 40% (compared with 25.5% – 35.8% in the clay fraction). In the 20 – 40 cm soil layer, there was much less new SOC from fertilizer than in the 0–20 cm layer, and the largest proportion of new SOC was in the sand fraction (23.5%–50.6%), followed by the silt and clay fractions.

For the four particle-size fractions at 0–20 cm depth, the proportion of new SOC was highest in ING (except silt, which was the highest in ORG), followed by ORG and CON. The differences between ORG and CON were significant except in the clay fraction. However, at 20–40 cm, the proportion of new SOC was always highest in ORG, followed by ING, but the differences between ING and CON were not significant except in the fine sand fraction.

4. Discussion

4.1 Influence of different fertilization management systems on SOC in greenhouse vegetable production

Compared with chemical fertilizers, organic fertilizers can significantly increase soil C content (*Brar* et al., 2013; *Jens* et al., 2009; *Manna* et al., 2007). *Lou* et al. (2011) found that organic manure alone (M) and NPK fertilizer combined with manure

(MNPK) significantly increased SOC storage and SOC content, consistent with our results. In fact, in this experiment, the fertilization treatments in CON and ING were mixtures of organic fertilizer and NKP at different ratios. However, the SOC stock was significantly higher in ING than in CON. A previous study also found that vegetable yield was significantly lower in CON than in the other two management systems (*Liang* et al., 2009). Thus, from the viewpoints of SOC sequestration and vegetable yield, CON is not necessarily the best option. Compared with CON, ING and ORG had more positive effects on SOC sequestration and vegetable production, but there was no significant difference in vegetable yield (*Liang* et al., 2009). However, comparing CE among treatments, ING was more advantageous than ORG. The reason why CE was lower in ORG than in ING may be because of carbon saturation in soil. *Six* et al. (2002a) suggested that soils can become carbon saturated as a result of physiochemical processes that stabilize or protect organic compounds. Some studies found that high-C soils showed little or no increase in soil C content, even with two- to three-fold increases in C inputs (*Campbell* et al., 1991; *Solberg et al*., 1998). On the basis of the results of this study and a previous study on vegetable yield (*Liang* et al., 2009) we suggest that when taking yield and SOC sequestration into account, ING is the most appropriate greenhouse vegetable management system for use in northern China.

In recent years, many developing countries have increased their organic production (*Gomiero* et al., 2011; *Reganold* and *Wachter*, 2016). However, excess fertilization in organic agriculture is still a serious problem. In China, large quantities of organic fertilizers are available, and farmers generally attach more importance to the application of organic fertilizers in greenhouse vegetable systems than in farmland (*Lou* et al., 2011). This high nutrient supply has the potential to pollute air, water, and other ecosystem components (*Meng* et al., 2016; *Tuomisto* et al., 2012). Therefore, with the expansion of greenhouse vegetable production, more attention should be paid to its potential adverse effects on the environment due to excess fertilization, and mitigation measures should be explored and implemented.

4.2 SOC dynamics within different particle-size fractions

The natural ^{13}C isotopic difference between C3 and C4 plants allows new carbon derived from each pathway to be traced in the SOC (*Balesdent* et al., 1990; *Bol* et al., 2004; *Krull* et al., 2007). In this study, we used the natural abundance ^{13}C tracer approach to estimate old and new SOC in particle-size fractions as influenced by fertilization. During the experiment, after harvesting each vegetable, the plants and their roots were removed from the greenhouse before establishing the new crop. During the growth of vegetables, some (C3) plant rhizosphere exudates or associated materials would have entered the soil. However, this C3 carbon source would very small compared with the large amount of C4-based organic fertilizer applied during the experiment. Therefore, we assumed in our new-C calculations that the only the applied (C4) organic fertilizer significantly affected the proportions of new SOC in the whole-soil and particle-size fractions.

The texture of soil affects how easy it is to work and also its fertility. The soil texture tended to be uniform among the three management systems. The proportion of clay increased, which increased the retention of soil water and fertilizer and also stabilized SOC (*Barthès* et al., 2008; *Baldock*, 2002). Many studies have shown that SOC content tends to be positively correlated with soil clay content among sites (*Zinn* et al. 2005; *McLauchlan*, 2006; *Plante* et al. 2006). In addition, clay+silt content or clay content were identified as the best parameters in models for predicting SOC stocks (*Gonçalves* et al., 2017). We used the ^{13}C tracer approach to estimate old and new SOC in particle-size fractions, and found that the turnover rate of SOC was significantly higher in the sand fraction than in the silt and clay fractions, consistent with many other studies (e.g. *Six* et al., 2002b; *Grandy and Neff*, 2008; *Von Lutzow* et al. 2007; *Bol* et al., 2009). For SOM bound to minerals and particulate organic matter (POM), the turnover rates generally decrease as particle size decreases, i.e. coarse sand<fine sand<silt<clay (*Derrien* et al., 2006; *Glaser*, 2006). With other factors being equal, the longer turnover time of OC associated with silt and clay than OC associated with sand is because sand-sized particles of OC are generally derived from recently deposited, undecomposed plant residues, and reflect the molecular composition of their source materials (*Amelung* et al., 1999; *Von Lutzow* et al. 2007). Conversely, OC associated with silt and clay is composed of simpler, microbially processed C compounds that form more stable organo-mineral associations (*Bock* et al., 2007; *Derrien* et al., 2006; *Grandy* and *Neff*, 2008). *Bol* et al. (2009) confirmed

that sand-associated OC was dominated by pyrolysis products from lignins and carbohydrates, but silt and clay fractions had much higher protein contents. The increased SOC contents in the silt and clay fractions in ING and ORG suggested that organic fertilizer has an important role in stabilizing SOC.

Comparing our study with previous studies, we found that on an equal area basis, the proportion of new SOC in each fraction was much higher in greenhouse production than in farmland (*Meng* et al., 2014). There are two possible explanations for this: (1) Climatic factors constrained both the production and decomposition of SOM (*Six* et al., 2002b; *Amelung* et al., 1998). Compared with farmland, greenhouse soil has more stable and suitable temperature and moisture conditions. This is beneficial for the survival and reproduction of soil microorganisms, thereby increasing the turnover rate of all four particle-size fractions. (2) The amount of organic fertilizer applied was much larger in (Chinese) greenhouse production than in farmland. High application rates of organic fertilizer affect soil texture, soil microbes, and other factors (*Rasmussen* et al., 1991; *Sharma* et al., 1995; *Sharma* et al., 2001), which in turn enhances the SOC turnover rate. This suggests that in the same region, the soil carbon pool is more ‘active’ in greenhouses than in farmland. In 2016, there were 61,000 ha under organic vegetable production in China (*Qiao* et al., 2018), and this area has continued to increase. This implies that soil carbon pools and their turnover dynamics in greenhouse systems will have an increasing impact on SOC sequestration and greenhouse gas emissions in China. Therefore, it is very important to improve the monitoring and management of soil carbon pools in greenhouse production in China.

5. Conclusion

We analyzed soils in Chinese greenhouses producing vegetable crops under intensive organic (ORG), integrated (ING) and conventional (CON) farming management systems for 11 years. The results showed that: (1) ING optimized carbon sequestration and vegetable yield, and (2) the SOC increases in ORG and ING were mainly in the silt and clay fractions, suggesting that organic fertilizer improved soil structure and promoted SOC stabilization.

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Table 1 Mass proportion of four particle-size fractions in total soil (*n*=3). CON: conventional management, ING: integrated management, ORG: intensive organic management.

		CON		ING		ORG	
		2002 (%)	2013 (%)	2002 (%)	2013 (%)	2002 (%)	2013 (%)
0-20 cm	Coarse sand	2.3±0.1	1.9±0.2	2.1±0.1	3.1±0.3	3.4±0.4	4.2±0.5
	Fine sand	3.6±0.4	7.1±2.2	4.7±0.2	8.0±2.0	5.4±0.1	7.5±1.6
	Silt	89.5±1.4	82.0±2.7	86.7±0.5	81.5±2.0	83.7±0.7	82.0±1.7
	Clay	4.6±0.2	9.0±0.6	6.5±0.3	7.3±0.2	7.5±0.3	6.3±0.7
20-40 cm	Coarse sand	0.5±0.3	0.3±0.1	0.3±0.1	0.4±0.1	0.3±0.1	0.4±0.1
	Fine sand	4.7±0.1	5.3±1.7	5.0±0.1	5.6±0.5	4.6±0.4	4.8±1.6
	Silt	88.3±2.6	83.8±0.7	88.4±0.8	84.2±0.5	88.0±1.1	85.3±1.7
	Clay	6.5±1.7	10.6±1.3	6.3±0.6	9.8±0.2	7.2±0.1	9.5±0.7

Table 2 Contribution of particle-size fractions to whole soil organic carbon content increase after 11-year experiment (*n*=3). CON: conventional management, ING: integrated management, ORG: intensive organic management. Different lower-case letters indicate significant differences ($P < 0.05$) among particle-size fractions (*n*=3).

		CON (%)	ING (%)	ORG (%)
0-20 cm	Coarse sand	4.2±3.0c	14.9±0.8c	13.8±0.2c
	Fine sand	64.0±0.2a	20.8±0.1b	20.6±1.0b
	Silt	2.7±0.1d	55.4±3.6a	57.8±0.4a
	Clay	29.1±2.2b	8.9±0.4d	7.8±1.2d
20-40 cm	Coarse sand	-12.0±7.1c	58.2±1.0a	23.4±0.5c
	Fine sand	-11.8±1.3c	-13.7±4.6c	9.7±1.7d
	Silt	25.0±5.8b	29.5±8.7b	36.9±1.0a
	Clay	98.8±7.0a	26.0±4.0b	30.0±1.1b

Table 3 Soil organic carbon (SOC) stocks affected by different fertilization management systems from 2002 to 2013. Different lower-case letters within the same column indicate significant differences ($P < 0.05$) among management systems (n=3). CON: conventional management, ING: integrated management, ORG: intensive organic management.

		Original equivalent soil mass			Minimum equivalent soil mass		
		SOC stock in 2002 (Mg C·ha^{-1})	SOC stock in 2013 (Mg·ha^{-1})	ΔSOC (kg·ha^{-1} yr^{-1})	SOC stock in 2002 (Mg·ha^{-1})	SOC stock in 2013 (Mg·ha^{-1})	ΔSOC (kg·ha^{-1} yr^{-1})
0-20 cm	NP	/*	28.6±0.7d	/	/	26.1±0.6d	/
	CON	28.7±0.6a	34.2±0.5c	494±42.6c	27.5±0.6a	32.0±2.2c	414±40.7c
	ING	19.1±1.0b	50.8±3.2b	2880±365b	18.7±0.9b	48.9±3.1b	2741±353b
	ORG	22.6±0.3b	66.7±0.1a	4008±36.6a	22.6±0.3b	66.7±0.1a	4008±36.6a
20-40 cm	NP	/	10.5±0.30b	/	/	10.0±0.29b	/
	CON	7.9±0.1b	9.1±0.5c	109±44.8b	7.8±0.1b	8.9±0.5c	92±43.3b
	ING	9.7±0.2a	10.4±0.3b	66±13.4b	9.7±0.2a	10.3±0.3b	55±12.9b
	ORG	10.6±0.6a	13.3±0.3a	245±66.4a	10.6±0.6a	13.3±0.3a	245±66.4a

* not applicable.

Table 4 Carbon conversion efficiency affected by different fertilization management systems from 2002 to 2013. CON: conventional management, ING: integrated management, ORG: intensive organic management, a: ploughing to 20 cm, b: ploughing to 30 cm, c: -0.1 is means gain off 0.1.

		Yearly input ($Mg\ C\cdot ha^{-1}$)		Yearly increase	Yearly loss ($Mg\ C\cdot ha^{-1}$)		Carbon conversion efficiency (%)	
		P20[a]	P30[b]	($Mg\ C\cdot ha^{-1}$)	P20[a]	P30[b]	P20[a]	P30[b]
0-20 cm	CON	8.7	5.8	0.5	8.2	5.3	5.7	8.6
	ING	16.1	10.8	2.9	13.3	7.9	17.8	26.8
	ORG	29.5	19.7	4.0	25.5	15.7	13.6	20.4
20-40 cm	CON	0	2.9	0.1	-0.1[c]	2.8	1.3	1.9
	ING	0	5.4	0.1	-0.1[c]	5.3	0.4	0.7
	ORG	0	9.8	0.2	-0.2[c]	9.6	0.8	1.3

Table 5 Old and new soil organic carbon in particle-size fractions under different fertilization management systems. Different lower-case letters within the same row indicate significant differences ($P < 0.05$) among management systems (n=3). Different upper-case letters within the same column indicate significant differences ($P < 0.05$) among particle-size fractions (n=3). CON: conventional management, ING: integrated management, ORG: intensive organic management.

		CON		ING		ORG	
		Old (%)	New (%)	Old (%)	New (%)	Old (%)	New (%)
0-20 cm	Coarse sand	53.4±1.1	46.6±1.1 Ab	36.5±1.6	63.5±1.6 Ba	35.5±3.4	64.5±3.4 Aa
	Fine sand	55.9±1.2	44.1±1.2 Bc	33.5±0.1	66.5±0.1 Aa	46.5±2.1	53.5±2.1 Cb
	Silt	59.2±0.7	40.8±0.7 Cb	58.9±2.9	41.1±2.9 Cb	44.2±3.6	55.8±3.6 Ba
	Clay	74.5±2.6	25.5±2.6 Db	64.2±3.2	35.8±3.2 Da	71.9±1.5	28.1±1.5 Db
20-40 cm	Coarse sand	74.2±0.6	25.8±0.6 Ab	54.8±2.8	45.2±1.2 Ab	53.0±1.3	50.6±1.3 Aa
	Fine sand	76.5±3.4	23.5±3.4 Ac	65.5±0.7	34.5±0.7 Bb	52.8±2.5	47.2±2.5 Aa
	Silt	75.9±1.2	24.1±1.2 Ab	70.4±3.5	29.6±3.5 Cb	63.3±4.3	36.7±4.3 Ba
	Clay	92.3±2.5	7.7±2.5 Bb	90.1±3.1	9.9±3.1 Db	66.7±3.0	33.3±3.0 Ca

Figure captions

Figure 1 Changes in soil organic carbon (SOC) contents in four particle-size fractions after 11 years under different fertilization management systems: (a) coarse sand, (b) fine sand, (c) silt, and (d) clay fractions. Different lower-case letters indicate significant differences among management systems in the same experimental year ($P < 0.05$). * ($P < 0.05$) or ** ($P < 0.01$) indicates statistical differences between 2002 and 2013. Bars show standard error (n=3). T refers to management system, Y refers to experiment year, and T*Y refers to interactive effect of management system and year. Upper: 0–20 cm soil layer, below: 20–40 cm soil layer. CON: conventional management, ING: integrated management, ORG: intensive organic management.

Figure 2 Changes in SOC contents in whole-soil and different particle-size fractions after 11 years under different fertilization management systems: (a) whole soil, (b) coarse sand, (c) fine sand, (d) silt, and (e) clay fractions. Different lower-case letters indicate significant differences among management systems in the same experimental year ($P < 0.05$). * ($P < 0.05$) or ** ($P < 0.01$) indicates statistical differences between 2002 and 2013. Bars show standard error (n=3). T refers to management system, Y refers to experiment year, and T*Y refers to interactive effects of management system and year. Upper: 0–20 cm soil layer, below: 20–40 cm soil layer. CON: conventional management, ING: integrated management, ORG: intensive organic management.

Figure 3 Contribution of soil organic carbon (SOC) increase in four particle-size fractions to whole SOC increase in intensive organic management (ORG) and integrated management (ING) management systems compared with conventional management (CON) management system. CS: coarse sand, FS: fine sand, S: silt, and C: clay. Different lower-case letters indicate significant differences among four particle-size fractions in same management system ($P < 0.05$). Bars show standard error (n=3). Upper: 0–20 cm soil layer, below: 20–40 cm soil layer.

Figure 4 δ^{13}C values of soil organic carbon (SOC) in four particle-size fractions under different fertilization management systems: (a) Coarse sand, (b) fine sand, (c) silt, and (d) clay fractions. Different lower-case letters indicate significant differences among management systems in same experimental year ($P < 0.05$). * ($P < 0.05$) or ** ($P < 0.01$) indicates statistical differences between 2002 and 2013. Bars show standard error (n=3). T refers to management system, Y refers to experiment year, and T*Y refers to interactive effects of management system and year. Upper: 0–20 cm soil layer, below: 20–40 cm soil layer. CON: conventional management, ING: integrated management, ORG: intensive organic management.

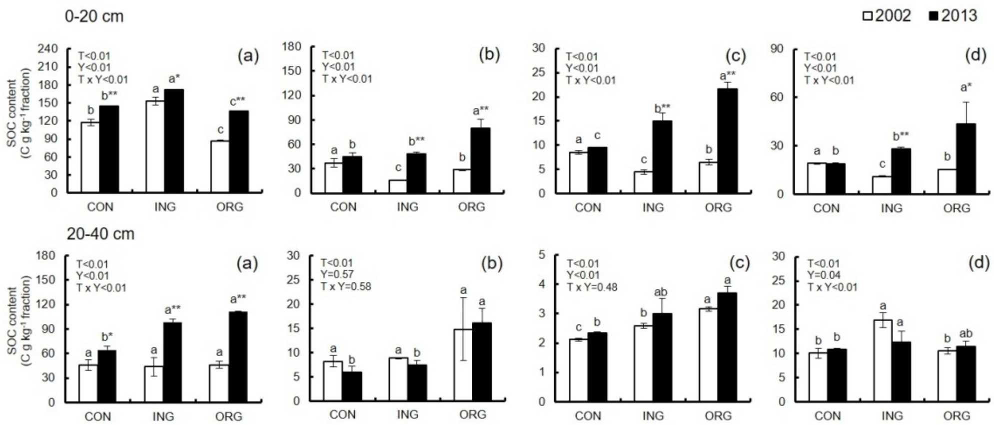

Fig.1

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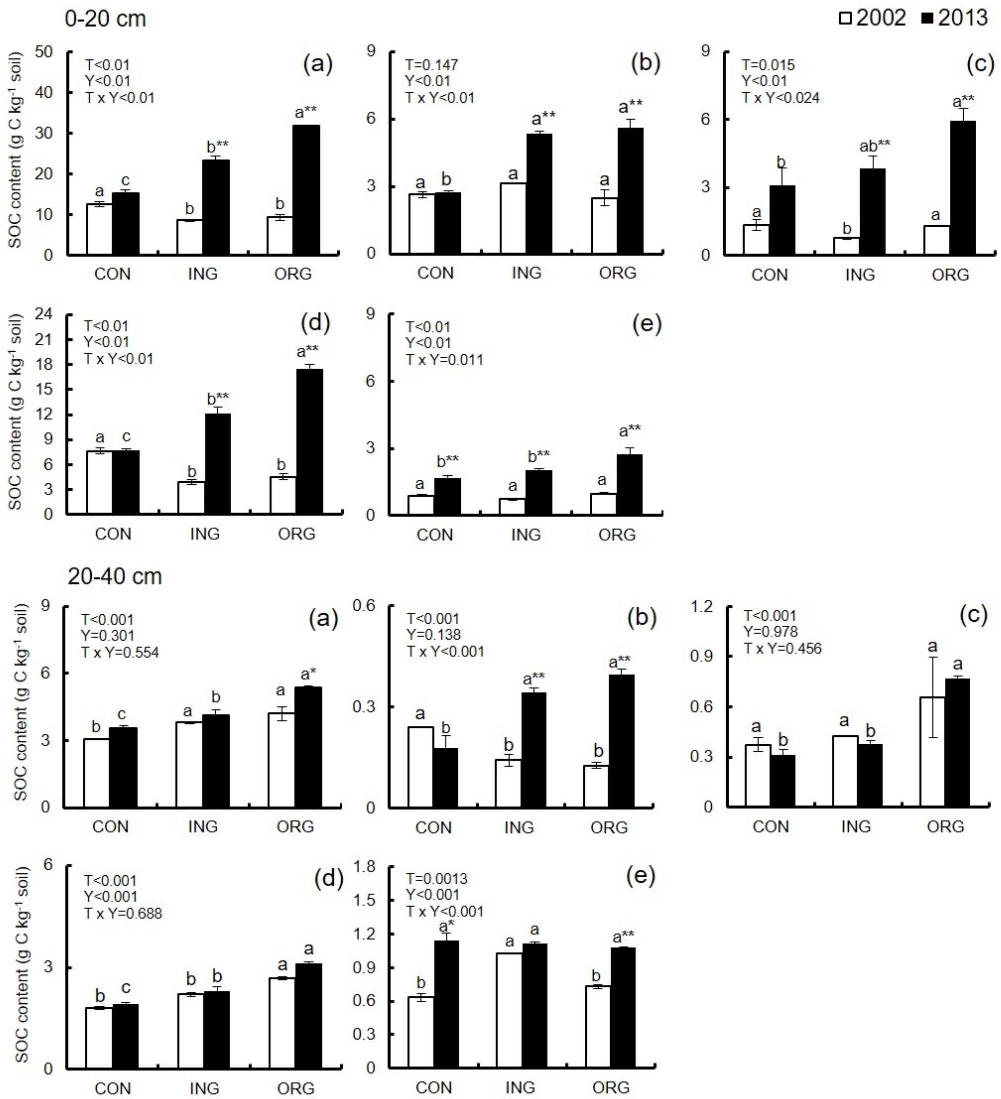

Fig.2

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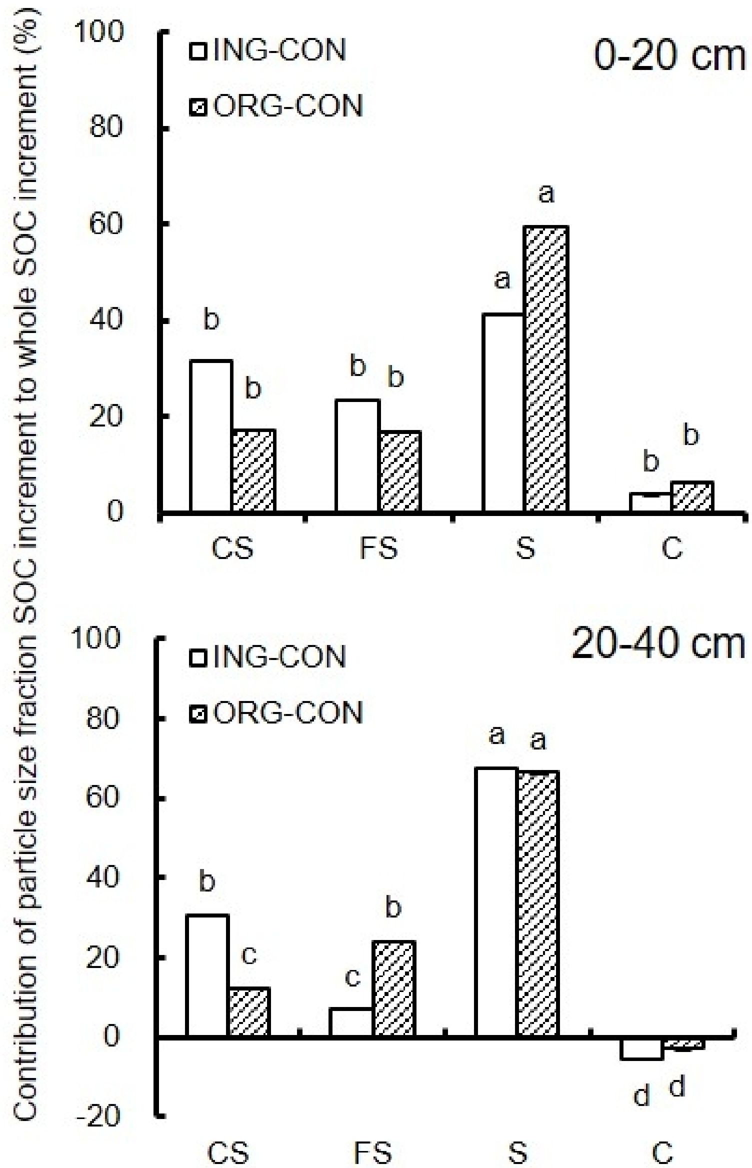

Fig.3

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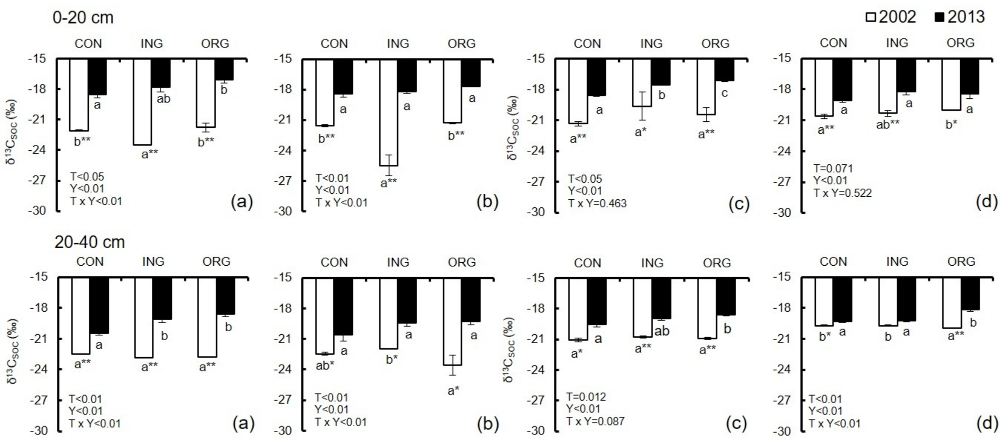

Fig.4

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