**RESEARCH ARTICLE** 

**OPEN ACCESS** 

## Effects of different regimes of fertilization on soil organic matter under conventional tillage

Zhibin Guo<sup>1, 2</sup>, Keke Hua<sup>1, 2</sup>, Jing Wang<sup>1, 2</sup>, Xisheng Guo<sup>1, 2</sup>, Chuanlong He<sup>1, 2</sup> and Daozhong Wang<sup>1, 2</sup>\*

<sup>1</sup> Soil and Fertilizer Research Institute. Anhui Academy of Agricultural Sciences. Hefei. Anhui, 230031 China. <sup>2</sup> Key Laboratory of Nutrient Cycling and Resources Environment of AnHui province. Hefei. Anhui, 230031 China

## Abstract

To explore the effects of different fertilization regimes on soil organic matter (SOM) sequestration in a wintersoybean/corn rotation, a long-term field experiment was conducted in Anhui, China, from 1982 to 2011. There were six treatments, as follows: (1) no fertilizer input (CK); (2) mineral fertilizers input (NPK); (3) mineral fertilizers + 3,750 kg ha<sup>-1</sup> wheat straw (WS/2-NPK); (4) mineral fertilizers + 7,500 kg ha<sup>-1</sup> wheat straw (WS-NPK); (5) mineral fertilizers + 15,000 kg ha<sup>-1</sup> composted farmyard manure (CNPK); and (6) mineral fertilizers + 30,000 kg ha<sup>-1</sup> composted farmyard manure (DNPK). Mineral fertilizer applications combined with organic amendments improved soil physical properties. For the WS/2-NPK, WS-NPK, CNPK and DNPK treatments, the soil bulk density decreased more than 10%, while the air porosity and field water content increased more than 90% and 15%, compared with the values at the start of the experiment in 1982. Our results indicate that about two decades are needed for SOM to reach its saturation point in all treatments. The SOM sequestration rate was related to the fertilization regime. The average SOM sequestration rate in 1982-2005 was 0.27 g kg<sup>-1</sup> yr<sup>-1</sup> with NPK, 0.45 g kg<sup>-1</sup> yr<sup>-1</sup> with WS/2-NPK, 0.56 g kg<sup>-1</sup> yr<sup>-1</sup> with WS-NPK, 0.60 g kg<sup>-1</sup> yr<sup>-1</sup> with CNPK and 1.02 g kg<sup>-1</sup> yr<sup>-1</sup> with DNPK. Therefore, both the quantity and the quality of the organic amendment determine the SOM sequestration rate and SOM saturation level.

Additional key words: soil organo-mineral complex; soil humic acid; soil fulvic acid.

## Introduction

Soil organic matter (SOM) is an important factor, as it determines the productivity and ultimately the sustainability of cropland. As the demand for food has increased rapidly during the last decades, agricultural production systems have become more and more intensive in China. The intensification of agricultural production has greatly increased grain, vegetable and fruit production, but also has led to a decline in SOM and to soil erosion (Zhao *et al.*, 2009). The decline in SOM is partly due to straw burning and the neglect of animal manure as a source of organic matter and nutrients during the last three decades (Shen *et al.*, 2007).

In response, series of agricultural managements have been adopted for improving SOM content, inclu-

ding fertilization, a ban on straw burning and return of crop residues to soil, and increased inputs of animal manures and compost (Melero-Sanchez *et al.*, 2008; Pan *et al.*, 2009; Zhao *et al.*, 2009; Zhang *et al.*, 2012). However, the sequestration of SOM in cropland is limited (Gulde *et al.*, 2008).

Previous studies have shown that the SOM content is dependent on environmental conditions, soil type and management, and the quantity and quality of the organic matter additions. Following the addition of organic matter, the SOM content generally increases until it reaches a specific point (Kool *et al.*, 2007; Gulde *et al.*, 2008). The specific point of SOM has been reported to be closely related to soil texture, which affects the mechanism of SOM sequestration. The sequestration of SOM is attributed to chemical

<sup>\*</sup> Corresponding author: wdzhong-3@163.com Received: 13-08-13. Accepted: 01-07-14.

Abbreviations used: FA (fulvic acid); HA (humic acid); HF (soil heavy fraction); HM (heavy fraction organic matter); HW (heavy fraction weight); PQ (humification degree); SOM (soil organic matter),

protection through sorption and association of processed organic matter with minerals, physical protection in aggregates, and biochemical protection through recalcitrance (von Lützow *et al.*, 2008). Physical and chemical mechanisms set a limit to the amount of SOM that can be protected (Baldock & Skjemstad, 2000; Six *et al.*, 2002). For example, Chung *et al.* (2010) found that the SOM pool in microaggregates and macroaggregates does not stabilize additional organic matter even at higher manure application rates.

Previous studies focused especially on how plant litter quality affects the sequestration or decomposition of SOM (e.g., Vanlauwe et al., 2005; Fonte et al., 2009). Relatively few studies examined the effects of long-term fertilization with different quantities and quality of fertilizers and organic amendments on SOM content. Therefore, the present study was designed to test the hypotheses that the sequestration rate of SOM in cropland is closely related to the quantity and quality of organic soil amendments, and that the SOM sequestration rate decreases over time. The main objectives of our study were (i) to find out how long it takes before SOM to reach its saturation level, and (ii) whether the saturation point of SOM was closely related to the quantity and quality of the organic soil amendment.

## Material and methods

### **Description of the experimental site**

A long-term field experiment was set-up in the northern region of Anhui Province, China (32°13'N, 116°37'E) in 1982. The mean annual temperature at the experimental site is 14.8°C, and the annual average potential evaporation is 1027 mm. During the experiment, the mean annual rainfall was 792 mm, ranging from 292 mm to 1469 mm (Fig. 1). The soil is classified as a vertisol; its pH is 6.0-8.6, and it consists of 33-50% coarse silt (0.05-0.01 mm), 43-45% fine silt (0.01-0.001 mm) and 25-26% fine clay (<0.001 mm) (Li et al., 2011). At the beginning of the experiment in 1982, in the 0-20 cm layer, soil organic matter was 10.40 g kg<sup>-1</sup>; total nitrogen, 0.96 g kg<sup>-1</sup>; available nitrogen, 84.5 mg kg<sup>-1</sup>; total phosphorus, 0.28 g kg<sup>-1</sup>; available phosphorus, 9.8 mg kg<sup>-1</sup>; available potassium, 111 mg kg<sup>-1</sup>; and pH, 7.20.



**Figure 1.** Annual precipitation (a) and average monthly precipitation (b) in the period 1986-2011 (except for 1991) at the experimental site. The wheat growing season ranges from November to May in the next year, and for corn and soybean from June to October.

The experiment included six treatments with four replicates: (1) no fertilizer (CK); (2) applied mineral fertilizers alone (NPK); (3) mineral fertilizers combined with 3750 kg ha<sup>-1</sup> wheat straw (WS/2-NPK); (4) mineral fertilizers combined with 7500 kg ha<sup>-1</sup> wheat straw (WS-NPK); (5) mineral fertilizers combined with 15,000 kg ha<sup>-1</sup> composted farmyard manure (CNPK); (6) mineral fertilizers combined with 30,000 kg ha<sup>-1</sup> composted farmyard manure (DNPK). All plots were arranged randomly, and the area of each plot was 70 m<sup>2</sup> (14.9 m  $\times$  4.7 m). The applied rates of mineral fertilizers were 180 kg ha<sup>-1</sup> N, 90 kg ha<sup>-1</sup> P, and 135 kg ha<sup>-1</sup> K. These amounts are commonly applied by local farmers. The mean composition of the wheat straw was 482.0 g C kg<sup>-1</sup>, 5.5 g N kg<sup>-1</sup>, 1.2 g P kg<sup>-1</sup> and 11.5 g K g kg<sup>-1</sup>; The composition of the compost was as follows: 70% moist, 367.0 g C kg<sup>-1</sup>, 7.9 g N kg<sup>-1</sup>, 4.2 g P kg<sup>-1</sup> and 3.9 g K kg<sup>-1</sup>. The annual input of organic C to the cropland was 1808 kg ha<sup>-1</sup> with WS/2-NPK, 3615 kg ha<sup>-1</sup> with WS-NPK, 1652 kg ha<sup>-1</sup> with CNPK and 3303 kg ha<sup>-1</sup> with DNPK, respectively. The organic amendments and mineral fertilizers were applied as base fertilizers prior to winter wheat seeding in autumn (October). The crop rotation was winter wheat-soybean, except in 1993 to 1997 when a winter

wheat-corn rotation was used. Wheat, soybean, and corn were planted at a seed rate of about 188, 75 and  $30 \text{ kg ha}^{-1}$ , respectively. After harvest, all crop residues were removed from the cropland.

#### Samplings and measurements

Soil samples (0 cm to 15 cm) were taken randomly each year in October before wheat seeding, in triplicate. Samples were air-dried before SOM analysis. Undisturbed soil samples for the determination of bulk density, and air porosity were collected from 0-15 cm with soil rings (100 cm<sup>3</sup>) in 2005.

Soil bulk density was measured by drying soil samples in an oven at 105°C for a minimum of 8 hours. Soil air porosity and capillary porosity were measured using the air pycnometer method (Wooldridge, 1968). Soil water-holding capacity was measured with a handheld tensiometer (Soil Moisture Equipment Corp., Santa Barbara, CA, USA), and pH in a 1:1 soil solution ratio. Soil organic carbon was measured using the Walkley-Black dichromate oxidation method (Nelson & Sommers, 1982). Soil total nitrogen (TN) was determined using the semimicroKjeldahl digestion method (Kieltec Foss 2200, Denmark) (Bremner & Mulvaney, 1982), and available nitrogen (AVN), including ammonium, nitrate and easily decomposable and hydrolysable organic nitrogen, were analyzed by alkali distillation. Soil total phosphorus (TP) was measured spectrophotometrically after digestion using perchloric and sulfuric acid (Murphy & Riley, 1962), and available P (AVP) was measured by extracting soil with 0.5 M NaHCO<sub>3</sub> at pH 8.5 (Olsen & Sommers, 1982). Soil available potassium (AVK) was measured via atomic absorption spectrophotometry after extracting soil with 1 M NH<sub>4</sub>-OAc at pH 7 (Richards & Batess, 1989).

Soil samples collected in October of 2005 and 2007 were used for fractionation of OM and humic fractions analysis. To separate light and heavy OM fractions, the centrifuge method was adapted (Strickland & Sollins, 1987). Firstly, 50 mL of NaI with a density of 1.7 g cm<sup>-3</sup> was added to 25 g of air-dried soil, followed by shaking for 45 min to disperse soil samples. Then, the heavy fraction was separated by centrifugation at 4068 × g for 10 min, and the light fraction was aspirated from the surface of each sample into a vacuum flask. After washing with 200 mL of deionized water and 150 mL of 0.01 M CaCl<sub>2</sub>, the light fraction was collected. Both light and heavy fractions were dried at 105°C for 48 h, weighed, and analyzed for total C using Multi N/C 2100 (Analytic Jena, German). Humic and fulvic acids were extracted using the method described by Spaccini et al. (2002). Briefly, 10 g of air-dried soil combined with 100 mL of 1 MNaOH + 1 M  $Na_4P_2O_7$ (50/50 v/v) was shaken for 16 hours at room temperature. Then, the mixture was centrifuged at 3000 g for 30 min to collect the supernatant containing humic extracts (HA and FA). The soil residue in the centrifuge was extracted again and the addition solution was collected. The two solutions were combined and acidified to pH 1.0-1.5 with concentrated HCl. After 24 hours, HA was collected and purified from inorganic impurities through repeated dissolution, precipitation, dialysis and freeze-drying. Soluble FA was collected by bringing the solution to pH 5, dialysis and freeze-drying.

### **Calculation of SOM accumulation**

The sequestration rate of SOM (SOMSR,  $g kg^{-1} yr^{-1}$ ) was calculated from:

$$SOMSR = (SOM_n - SOM_{n0}) / n$$

where SOM<sub>n</sub> and SOM<sub>n0</sub> represent the SOM in the treated and initial plots, respectively, and n indicates the year of experimentation. The SOM sequestration rates under different fertilization regimes were calculated by deducting the rate pertaining to control treatment from other treatments (Tong *et al.*, 2014).

Soil primary organo-mineral complexes (OOC), the degree of primary organo-mineral complexes (OOD), secondary organo-mineral complexes (AOC), and the degree of secondary organo-mineral complexes (AOD) were calculated as follows:

$$OOC = HM \times HW / SW;$$
  
 $OOD = OOC \times 100\% / SM;$   
 $AOC = MQC-OOC;$   
 $AOD = AOC \times 100\% / (MC-SM);$ 

where HM (g kg<sup>-1</sup>) represents the SOM in heavy fraction (HF), HW (g) is the mass of heavy fraction, SW (g) is the mass of original soil, SM (g kg<sup>-1</sup>) represents the organic matter of the original soil, and MQC and MC represent the organo-mineral complex and organic matter in the fertilization treatments, respectively.

Treatments	BD (g cm <sup>-3</sup> )	SAP (%)	CP (%)	FWC (%)	рН
СК	1.38±0.01ª*	6.47±0.24 <sup>d</sup>	41.45±0.68ª	29.35±0.35 <sup>b</sup>	7.17±0.04 <sup>b</sup>
NPK	1.36±0.01ª	8.42±0.21°	40.26±0.34ª	29.79±0.22 <sup>b</sup>	$5.71 \pm 0.05^{d}$
WS/2-NPK	1.24±0.01 <sup>b</sup>	12.70±0.41 <sup>b</sup>	$40.51 \pm 0.17^{a}$	$32.49{\pm}0.37^{a}$	$5.62 {\pm} 0.02^{d}$
WS-NPK	1.22±0.01 <sup>b</sup>	$14.26{\pm}0.44^{a}$	$40.46{\pm}0.54^{a}$	33.40±0.42ª	$5.56 {\pm} 0.04^{d}$
CNPK DNPK	1.23±0.01 <sup>b</sup> 1.10±0.01 <sup>c</sup>	13.10±0.35ª 14.75±0.45ª	40.48±0.37ª 42.98±0.25ª	$\begin{array}{c} 34.47{\pm}0.33^{a} \\ 34.63{\pm}0.42^{a} \end{array}$	6.72±0.07° 7.62±0.02ª

**Table 1.** Soil bulk density (BD), soil air porosity (SAP), capillary porosity (CP), field waterholding capacity (FWC) and pH in the six treatments (means  $\pm$  SE) (n = 3)

\* Values followed by the same letter within a column are not significantly different at  $p \le 0.05$ .

**Table 2.** Organic matter contents (g kg<sup>-1</sup>) of the top soil (0 to 15 cm) of the six treatments in the years 1987 to 2011 (means  $\pm$  SE) (n = 3)

Treatments	1987	1991	1997	2001	2003	2005	2007	2009	2011
СК	11.81±0.40 <sup>a</sup> *	10.67±0.41 <sup>ab</sup>	11.50±0.41ª	10.02±0.51 <sup>ab</sup>	10.71±0.21 <sup>ab</sup>	10.29±0.11 <sup>ab</sup>	9.52±0.30 <sup>b</sup>	9.70±0.20 <sup>b</sup>	9.81±0.20 <sup>b</sup>
NPK	12.09±0.41 <sup>b</sup>	12.11±0.21 <sup>b</sup>	$13.48{\pm}0.50^{ab}$	15.90±0.50ª	16.21±0.30ª	16.50±0.32ª	14.33±0.30ª	15.81±0.42ª	15.46±0.51ª
WS/2-NPK	13.32±0.30°	13.75±0.22°	$15.31{\pm}0.40^{ab}$	18.30±0.42ª	19.29±0.50ª	20.79±0.50ª	19.67±0.30ª	18.90±0.50ª	20.30±0.41ª
WS-NPK	11.21±0.20°	13.64±0.31°	16.02±0.41bc	18.51±0.21 <sup>b</sup>	$20.21{\pm}0.33^{ab}$	23.21±0.51ª	22.10±0.53ª	20.20±0.20ª	22.91±0.40ª
CNPK	12.53±0.30°	14.51±0.50°	17.81±0.40 <sup>b</sup>	22.49±0.60ª	24.03±0.50ª	24.10±0.63ª	24.21±0.20ª	23.50±0.33ª	24.56±0.30ª
DNPK	$15.70{\pm}0.52^{d}$	$24.33{\pm}0.70^{\text{bc}}$	$29.83{\pm}0.52^{ab}$	35.70±0.60ª	35.20±0.42ª	33.81±0.51ª	32.89±0.42ª	33.20±0.20ª	35.40±0.33ª

\* Values followed by the same letter within a column among different treatments are not significantly different at  $p \le 0.05$ .

## Statistical methods

Statistical analyses were performed using SAS 9.1.3 (SAS Institute Inc., Cary, NC, USA). Data for SOM, bulk density, air porosity, capillary porosity, field water-holding capacity and pH were analyzed through standard analysis of variance (ANOVA), and the differences among treatments were considered significant when p < 0.05. The graphs were plotted using Sigma Plot 10.0 (Systat Software Inc., San Jose, CA, USA).

## Results

#### Soil physical properties

Fertilization improved soil physical properties (Table 1). At the beginning of the experiment in 1982, soil bulk density, air porosity, capillary porosity and field water-holding capacity were 1.43 g cm<sup>-3</sup>, 6.42%, 39.61%, and 27.69%, respectively. In 2005, the soil bulk density had decreased in all treatments, as follows: by 4.90% in NPK, 13.29% in WS/2-NPK, 14.69% in WS-NPK, 13.99% in CNPK, and by 23.07% in DNPK. As a consequence, the air porosity and the field water content increased. Soil pH decreased significantly in treatments with mineral fertilization and wheat straw additions, compared to the CK treatment and the treatments with manure additions (Table 1).

### Soil organic matter and its fractions

Fertilization had also a strong impact on SOM and its fractions. SOM did not increase continuously in the fertilization treatments. From 2005, SOM more or less stabilized in all treatments. The average SOM content in the period ranging from 2005 to 2011 was 9.85 g kg<sup>-1</sup> in CK, 15.53 g kg<sup>-1</sup> in NPK, 19.92 g kg<sup>-1</sup> in WS/2-NPK, 22.85 g kg<sup>-1</sup> in WS-NPK, 24.10 g kg<sup>-1</sup> in CNPK and 34.08 g kg<sup>-1</sup> in DNPK, respectively (Table 2). Compared to the CK treatment, the light fraction SOM was 61.70% higher in NPK, 431.91% higher in WS/2-NPK, 489.36% higher in DNPK treatments. Similarly, the heavy fraction SOM had also increased relative to the CK treatment, by 64.20% in NPK, 84.81% in WS/2-NPK, 103.25% in WS-NPK, 123.33% in

secondary organo-inneral complex ( $XOD$ ) in the six experiments (means $\pm$ 5D) ( $n = 5$					
Treatments	OOC (g kg <sup>-1</sup> )	OOD (%)	AOC (g kg <sup>-1</sup> )	AOD (%)	
СК	9.48±0.34e*	92.13±2.75ª			
NPK	$15.19 \pm 0.84^{d}$	92.06±3.04ª	$5.71 \pm 0.13^{d}$	93.57±3.67ª	
WS/2-NPK	$16.48 \pm 0.81^{cd}$	79.27±1.46 <sup>b</sup>	7.00±0.11°	66.99±2.11 <sup>b</sup>	
WS-NPK	$18.43 \pm 0.38^{bc}$	79.41±1.17 <sup>b</sup>	8.95±0.19°	$69.57 {\pm} 2.08^{b}$	
CNPK DNPK	19.90±0.40 <sup>b</sup> 26.64±0.33 <sup>a</sup>	$78.03{\pm}0.8^{\rm b} \\ 78.79{\pm}1.51^{\rm b}$	10.41±0.10 <sup>b</sup> 17.15±0.14 <sup>a</sup>	$\begin{array}{c} 68.56{\pm}2.57^{\rm b} \\ 73.23{\pm}2.08^{\rm b} \end{array}$	

**Table 3.** Soil primary organo-mineral complex (OOC), the degree of primary organomineral complex (OOD), secondary organo-mineral complex (AOC) and the degree of secondary organo-mineral complex (AOD) in the six experiments (means  $\pm$  SE) (n = 3)

\* Values followed by the same letter within a column among different treatments are not significantly different at  $p \le 0.05$ .

**Table 4.** Humic acid (HA) and fulvic acid (FA) contents of the top soil (0-15 cm) of CK, NPK, WS/2-NPK, WS-NPK, CNPK and DNPK treatments (means  $\pm$  SE) (n = 3)

Treatments	HA (g kg <sup>-1</sup> )	FA (g kg <sup>-1</sup> )	PQ* (%)	HA/FA	
СК	0.81±0.04e**	1.09±0.05°	$42.75 \pm 0.99^{d}$	0.75±0.03°	
NPK	$1.53{\pm}0.03^{d}$	$2.24{\pm}0.04^{d}$	40.55±0.17 <sup>d</sup>	$0.68{\pm}0.01^{d}$	
WS/2-NPK	2.28±0.06°	2.64±0.03°	46.37±0.20b	$0.87 {\pm} 0.01^{b}$	
WS-NPK	2.60±0.05°	2.95±0.03°	46.91±0.15 <sup>b</sup>	$0.88 {\pm} 0.01^{b}$	
CNPK	$3.03{\pm}0.06^{b}$	$3.33{\pm}0.05^{b}$	47.66±0.39b	$0.91 \pm 0.02^{b}$	
DNPK	$4.11 \pm 0.07^{a}$	$3.75{\pm}0.06^{a}$	52.32±0.34ª	$1.10{\pm}0.02^{a}$	

\* PQ represents the degree of humification and is calculated by the ratio between soil humic acid (HA) and humic acids (HA + FA). \*\* Values followed by the same letter within a column among different treatments are not significantly different at  $p \le 0.05$ .

CNPK, and by 195.66% in the DNPK treatment. However, there were no significant differences among treatments with organic matter additions in the percentages of light fraction SOM. The percentage of light fraction SOM was 7.84% in CK, 8.08% in NPK, 22.16% in WS/2-NPK, 23.14% in WS-NPK, 23.35% in CNPK, and 20.46% in the DNPK treatment.

Table 3 shows the changes in soil organo-mineral complexes for the various treatments. Primary organomineral complex was 8.51% higher in WS/2-NPK, 21.34% higher in WS-NPK, 30.99% higher in CNPK, and 75.37% higher in the DNPK treatment in comparison with the NPK treatment. Similarly, the secondary organo-mineral complex had increased significantly in the treatments with organic additions in comparison to the NPK treatment. Moreover, the degree of humification [PQ, which is calculated by the ratio of soil humic acid (HA) to humic acids (HA+FA)] was significantly lower in the mineral fertilizer treatment NPK than in the WS/2-NPK, WS-NPK, CNPK, and DNPK treatments (Table 4).

## Discussion

### Soil organic matter and fertilization

SOM content is influenced by agricultural management practices. Long-term cultivation lowers the SOM content, whereas fertilization, input of organic materials, and fallow commonly enhance the SOM content (Vanlauwe et al., 2001; Huang & Sun, 2006; Bationo et al., 2007). The results of our experiment indicate that changes in SOM occurred rapidly during the first 20 to 25 years, while changes were small thereafter (Table 2). This suggests that a period of about two decades is needed for establishing a new equilibrium (saturation point) following a change in management practices. This suggestion is consistent with the findings of Rui & Zhang (2010), who reported that the enhancing effects of rice residue and farmyard manure addition on SOM content could last for 20 to 40 years. Thereafter, SOM content remained

stable because it had reached a steady state called saturation.

The SOM saturation level varied with the regime of fertilization. Mineral fertilizer inputs (NPK) increased the SOM content only slightly relative to the CK treatment. Straw additions and especially manure additions strongly increased the SOM content in the WS/2-NPK, WS-NPK, CNPK, and DNPK treatments. The different saturation levels under various fertilization regimes might be attributed to the different sequestration rates of SOM.

# SOM sequestration rate and fertilization regime

The results presented in Table 2 indicate that the average annual sequestration rate of SOM during the period 1982-2005 was 0.27 g kg<sup>-1</sup> yr<sup>-1</sup> with NPK, 0.45 g kg<sup>-1</sup> yr<sup>-1</sup> with WS/2-NPK, 0.56 g kg<sup>-1</sup> yr<sup>-1</sup> with WS-NPK, 0.60 g kg<sup>-1</sup> yr<sup>-1</sup> with CNPK and 1.02 g kg<sup>-1</sup> yr<sup>-1</sup> with DNPK. Interestingly, a lower annual sequestration rate of SOM was observed in WS/2-NPK, which received a larger amount of organic C than the CNPK treatment. A similar phenomenon was noticed for the WS-NPK and DNPK treatments. The different sequestration rates of SOM between wheat straw and composted manure treatments might be attributed to the availability of soil nutrients (Averill et al., 2014). For the most stable component of SOM such as fine fraction SOM, it not only contains carbon, hydrogen and oxygen, but also nitrogen, phosphorus and sulphur in approximately constant ratios, and a limitation of one or more of these essential nutrients may set a ceiling on the absolute quantity of SOM that can be synthesised (Stevenson, 1994; Kirkby et al., 2013). Limited availability of nutrients after organic carbon input can limit humus formation (Li et al., 2010; Kirkby et al., 2013, 2014). In our experiment, the composted manure had higher contents of (micro) nutrients than wheat straw, which helps improve the humification efficiency of new organic matter input in cropland. Higher organo-mineral complexes and PQ in CNPK and DNPK treatments compared to the wheat straw treatments strongly supports this suggestion.

The sequestration rate of SOM was about 0.40 g  $kg^{-1}$  yr<sup>-1</sup> per 1652 kg ha<sup>-1</sup> of organic C input in the CNPK and DNPK treatments. In contrast, the sequestration rate of SOM was 0.19 g kg<sup>-1</sup> yr<sup>-1</sup> per 1808 kg ha<sup>-1</sup> of organic C input in the WS/2-NPK treatment

and only 0.11 g kg<sup>-1</sup> yr<sup>-1</sup> per 1808 kg ha<sup>-1</sup> of organic C inputin the WS-NPK treatment. This latter result is consistent with that of Gude *et al.*(2012), who found that the C storage efficiency decreased from 51% in the low input treatment to only 20% in the high C input treatment.

For wheat straw treatments, the decreasing sequestration rate of SOM with a larger wheat straw input might be due to three reasons. Firstly, by the priming effect, which is defined as a short-term change in the turnover of SOM caused by the addition of organic carbon into soil (Kuzyzkov et al., 2000). Previous research surveyed that soil microbes played an important role in regulating SOM (Courtier-Murias et al., 2013). Following a high input of wheat straw combined with mineral fertilizers, a larger microbial population is found because of the priming effect. Consequently, more C is respired by soil microbes (Courtier-Murias et al., 2013). Secondly, by a lower soil pH; in the wheat straw treatments, the pH was significantly lower compared to the CK, CNPK and DNPK treatments. We cannot exclude the possibility that organic matter is less stabilized and less bound to soil mineral surfaces if pH is relatively low (Liu et al., 2014). Also, soil pH has a positive effect on soil microbial biomass, which is positively related to the stabilization and accrual of SOM, because microbial biomass and microbial byproducts resulting from microbial growth and degradation processes are major sources for SOM (Heinze et al., 2010; Miltner et al., 2012; Courtier-Murias et al., 2013; Wallenstein et al., 2013). Thirdly, low soil nutrients availability. In the terrestrial C cycle, the competition between plants and decomposers for soil nutrients play a fundamental control (Averill et al., 2014). For the most stable components of SOM, the carbon, nitrogen, phosphorus and sulphur ratio is approximately constant (C:N:P:S = 10,000:833:200:143). This ratio for plant material incorporated into the soil is often much higher, *i.e.*, contains less N, P and S per molecule of C (Kirkby et al., 2013). As a consequence, organic material with high C-to-nutrient ratios may require additional inputs of mineral fertilizers to allow the formation of new fine fraction SOM and to improve the humification efficiency of SOM (Kirkby et al., 2013, 2014). However, there were no differences in the amounts of mineral fertilizers applied between WS/2-NPK and WS-NPK treatments. Thus, no significant differences in soil primary organo-mineral complex and secondary organo-mineral complex were observed between WS/2-NPK and WS-NPK treatments.

In summary, fertilizer application and straw and manure additions clearly increased SOM contents relative to the CK treatment. About two decades were needed for SOM to reach its saturated point. The saturation level for SOM depended on the quantity and quality of the organic amendment. The SOM sequestration rates were dependent on the regimes of fertilization. The SOM sequestration was highest with composted farmyard manure applied in combination with mineral fertilizer. Combinations of wheat straw and mineral fertilizer were less effective in enhancing SOM sequestrations than combinations of composted manure and mineral fertilizers.

## Acknowledgements

This research was supported by grants from Stategic Priority Research Program of Chinese Academy of Sciences (XDB15010103), the Special Fund for Agroscientific Research in the Public Interest of China (201203030-07-01), AnHui Province technical program (1206c0805033 and 1406c085025) and President Youth Innovation Fund (No. 13B1043, No. 14B1007 and 14B1008) of Anhui Academic of Agricultural Science. We also thank Pro OeneOenema for his technical help and two anonymous reviewers whose insight comments and suggestions greatly improved the quality of the manuscript.

## References

- Averill C, Turner BL, Finzi AC, 2014. Mycorrhiza-mediated competition between plants and decomposers drives soil carbon storage. Nature 505: 543-545.
- Baldock JA, Skjemstad JO, 2000. Role of the soil matrix and minerals in protecting natural organic materials against biological attack. Org Geochem 31: 697-710.
- Bationo A, Kihara J, Vanlauwe B, Waswa B, Kimetu J, 2007. Soil organic carbon dynamics, functions and management in West African agro-ecosystems. Agric Syst 94: 13-25.
- Bremner JM, Mulvaney CS, 1982. Nitrogen-Total. In: Methods of soil analysis, Part2, Chemical and microbiological properties. American Society of Agronomy, Madison, WI, USA.
- Chung H, Ngo KJ, Plante A, Six J, 2010. Evidence for carbon saturation in a highly structured and organic-matter-rich soil. Soil Sci Soc Am J 74: 130-138.
- Courtier-Murias D, Simpson AJ, Marzadori C, Baldoni G, Ciavatta C, Fernadez JM, Lopez-de-Sa EG, Plaza C, 2013. Unraveling the long-term stabilization mechanisms of organic materials in soils by physical fractionation and NMR spectroscopy. Agr Ecosyst Environ 171: 9-18.

- Fonte SJ, Yeboah E, Ofori P, Quansah GW, Vanlauwe B, Six J, 2009. Fertilizer and residue quality effects on organic matter stabilization in soil aggregates. Soil Sci Soc Am J 73: 961-966.
- Gude A, Kandeler E, Gleixner G, 2012. Input related microbial carbon dynamic of soil organic matter in particle size fractions. Soil Biol Biochem 47: 209-219.
- Gulde S, Chung H, Amelung W, Chang C, Six J, 2008. Soil carbon saturation controls labile and stable carbon pool dynamics. Soil Sci Soc Am J 72: 605-612.
- Heinze S, Raupp J, Joergensen RG, 2010. Effects of fertilizer and spatial heterogeneity in soil pH on microbial biomass indices in a long-term field trial of organic agriculture. Plant Soil 328: 203-215.
- Huang Y, Sun WJ, 2006. Changes in topsoil organic carbon of croplands in mainland China over the last two decades. Chinese Sci Bull 51: 1785-1803.
- Kirkby CA, Richardson AE, Wade LJ, Batten GD, Blanchard C, Kirkegaard JA, 2013. Carbon-nutrient stoichiometry to increase soil carbon sequestration. Soil Biol Biochem 60: 77-86.
- Kirkby CA, Richardson AE, Wade LJ, Passioura JB, Batten GD, Blanchard C, Kirkegaard JA, 2014. Nutrient availability limits carbon sequestration in arable soils. Soil Biol Biochem 68: 402-409.
- Kool DM, Chung H, Tate KR, Ross DJ, Newton PCD, Six J, 2007. Hierarchical saturation of soil carbon pools near a natural CO<sub>2</sub> spring. Global Change Biol 13: 1282-1293.
- Kuzyakov Y, Friedel JK, Stahr K, 2000. Review of mechanisms and quantification of priming effects. Soil Biol Biochem 32: 1485-1498.
- Li DC, Zhang GL, Gong ZT, 2011. On taxonomy of Shajiang black soils in China. Soils 43: 623-629. [In Chinese].
- Li Z, Liu M, Wu X, Han F, Zhang T, 2010. Effects of longterm chemical fertilization and organic amendments on dynamics of soil organic C and total N in paddy soil derived from barren land in subtropical China. Soil Till Res 106: 268-274.
- Liu M, Zhang Z, He Q, Wang H, Li X, Schoer J, 2014. Exogenous phosphorus inputs alter complexity of soil-dissolved organic carbon in agricultural riparian wetlands. Chemosphere 95: 572-580.
- Melero-Sanchez S, Madejon E, Herencia JF, Ruiz-Porras JC, 2008. Long-term study of properties of a Xerofluvent and Guadalquivir River Valley under organic fertilization. Agron J 100: 611-618.
- Miltner A, Bombach P, Schmidt-Brücken B, Kastner M, 2012. SOM genesis: microbial biomass as a significant source. Biogeochemistry 111: 41-55.
- Murphy J, Riley JP, 1962. A modified single solution method for the determination of phosphate in natural waters. Anal Chem Acta 27: 31-36.
- Nelson DW, Sommers LE, 1982. Total carbon, organic carbon and organic matter. In: Methods of soil analysis, Part 2. Agron. Monogr. No. 9. ASA-SSSA, Madison, WI, USA.
- Olsen SR, Sommers LE, 1982. Phosphorus. In: Methods of soil analysis (Page AL,ed), Agron No.9, Part 2: Chemical

and microbiological properties, 2<sup>nd</sup>edn. Am SocAgron, Madison, WI, USA. pp: 403-430.

- Pan G, Smith P, Pan W, 2009. The role of soil organic matter in maintaining the productivity and yield stability of cereals in China. Agr Ecosyst Environ 129: 344-348.
- Richards JE, Bates TE, 1989. Studies on the potassiumsupplying capacities of southern Ontario soils. III. Measurement of available K. Can J Soil Sci 69: 597-610.
- Rui W, Zhang W, 2010. Effect size and duration of recommended management practices on carbon sequestration in paddy field in Yangtze Delta Plain of China: a metaanalysis. Agr Ecosyst Environ 135: 199-205.
- Shen MX, Yang L Zh, Yao YM, Wu DD, Wang JG, Guo RL, Yin SX, 2007. Long-term effects of fertilizer managements on crop yields and organic carbon storage of a typical rice-wheat agroecosystem of China. Biol Fertil Soils 44: 187-200.
- Six J, Conant RT, Paul EA, Paustian K, 2002. Stabilization mechanisms of soil organic matter: implications for C-saturation of soils. Plant Soil 241: 155-176.
- Spaccini R, Piccolo A, Conte P, Haberhauer G, Gerzabek MH, 2002. Increased soil organic carbon sequestration through hydrophobic protection by humic substances. Soil Biol Biochem 34: 1839-1851.
- Stevenson FJ, 1994. Humus chemistry: genesis, composition, reactions, 2<sup>nd</sup> ed. John Wiley & Sons, NY, USA.
- Strickland TC, Sollins P, 1987. Improved method for separating light- and heavy-fraction organic material from soil. Soil Sci Soc Am J 51: 1390-1393.
- Tong X, Xu M, Wang X, Bhattacharyya R, Zhang W, Cong R, 2014. Long-term fertilization effects on organic

carbon fractions in a red soil of China. Catena 113: 251-259.

- Vanlauwe B, Wendt J, Diels J, 2001. Combined application of organic matter and fertilizer. In: Sustaining soil fertility in West Africa. SSSA Spec Publ 58. SSSA and ASA, Madison, WI, USA.
- Vanlauwe B, Gachengo C, Shepherd K, Barrios E, Cadisch G, Palm CA, 2005. Laboratory validation of a resource quality-based conceptual framework for organic matter management. Soil Sci Soc Am J 69: 1135-1145.
- von Lützow M, Kögel-Knabner I, Ludwig B, Matzner E, Flessa H, Ekschmitt K, Guggenberger G, Marschner B, Kalbitz K, 2008. Stabilization mechanisms of organic matter in four temperate soils: development and application of a conceptual model. J Plant Nutr Soil Sci 171: 111-124.
- Wallenstein MD, Haddix ML, Ayres E, 2013. Litter chemistry changes more rapidly when decomposed at home but converges during decomposition-transformation. Soil Biol Biochem 57: 311-319.
- Wooldridge DB, 1968. An air-pycnometer for forest and range soils. Pacific NW Forest & Range Exp. Stn. Portland, OR, USA.
- Zhang WJ, Xu MG, Wang XJ, Huang QH, Nie J, Li ZZ, Li SL, Hwang SW, Lee KB, 2012. Effects of organic amendments on soil carbon sequestration in paddy fields of subtropical China. J Soils Sediments 12: 457-470.
- Zhao Y, Wang P, Li J, Chen Y, Ying X, Liu S, 2009. The effect of two organic manures on soil properties and crop yields on a temperate calcareous soil under a wheat-maize cropping system. Eur J Agron 31: 36-42.