



**Full Length Article**

## Variability of Soil Carbon Budget of the Reclaimed Lands with Different Straw Returning Modes and its Microbial Mechanism

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### Abstract

Many studies have focused on the effects of different improvement measures for reclaimed lands, but few have evaluated the effect of the different treatments on soil carbon budget in this study, the soil organic carbon (SOC), the plant biomass and soil respiration (SR) were analyzed to evaluate the effect of the different treatments on soil carbon budget. The soil microbial activities and characteristics were determined to further explore the effect mechanism on SR. Two treatments based on incorporating straw were used to improve reclaimed soil in Chongming Dongtan, China. Treatment 1 involved direct incorporation of straw into the soil through tilling, while in Treatment 2, the straw was first composted and then incorporated into the soil through tilling. The results show that after the SOC remediation with Treatment 1, there was an increase of 9.3% while Treatment 2 caused an increase of 9.4% compared with the control. Plant biomass with Treatments 1 and 2 was 3.44 and 1.67 times that of the control, respectively, which indicates a higher carbon input capacity, although soil respiration only increased by 23.5 and 46.4%, respectively. Microbial biomass and  $\beta$ -glucosidase activity was also higher at the treated sites compared to the control with a tendency of Treatment 2 > Treatment 1 > the control. The microbial community structure at the Treatment 2 site was different than Treatment 1 and the control site. These findings imply that the microbial activities with Treatment 2 were greater than those with Treatment 1, which means a higher rate of carbon metabolism, resulting in increased SR in Treatment 2. Both treatments decreased the soil salinity and bulk density and modified the soil physical structure thereby increasing the plant biomass. Moreover, Treatment 1 with a higher SOC input (plant biomass) and lower SR presented higher carbon sequestration potential. Composted straw incorporation biasing to soil microbial activities and changing the community structure might be an important cause of increased soil respiration in Treatment 2 compared to Treatment 1 (direct incorporation of the straw) and the control. © 2020 Friends Science Publishers

**Keywords:** Microbial activities; Plant biomass; Soil respiration; Soil organic carbon; Straw returning mode

### Introduction

Soil is considered a global carbon pool. The soil organic carbon (SOC) pool is usually affected by human disturbance of the terrestrial ecosystem (Lu *et al.* 2009), such as forest clearing and cropland cultivation. It is estimated that the carbon stored in the 30 cm surface soil of global cropland accounts for almost 10% of the global total SOC pool, exceeding 140 Pg C (Paustian *et al.* 2016; Zomer *et al.* 2017). These disturbances, especially traditional agricultural practices leading to inappropriate tillage and poor fertility management, cause soil degradation and decreased SOC (Varvel and Wilhelm 2011).

Crop straw has become the main source of exogenous carbon in the soil. The conversion and distribution of straw carbon in the soil directly affects the composition and content of soil organic carbon, thereby changing the soil

nutrient cycle (Zhang *et al.* 2016). Currently, straw management and tillage practices are attracting considerable research attention. Straw incorporation could increase the SOC content, improve the soil physical quality (soil salinity and soil bulk density) and enhance soil fertility if practiced on a long-term basis (Watanabe *et al.* 2009; Liu *et al.* 2014; Zhang *et al.* 2016; Dahri *et al.* 2018). Many researchers have reported changes in the soil carbon budget under different straw management practices (Liu *et al.* 2009; Badia *et al.* 2013). It has been shown that straw incorporation increases the source of carbon input into soils, thus stimulating soil microbial activity and soil respiration (Raubuch *et al.* 2010; Chen *et al.* 2017). Furthermore, small changes in the rate of soil respiration could have a large effect on the concentration of CO<sub>2</sub> in the atmosphere that can lead to increased greenhouse gas emissions (Schlesinger and Andrews 2000). As agriculture is considered one of the

major contributors to greenhouse gas emissions, it is important to develop a low-carbon agriculture system, with decreased soil respiration rates during straw incorporation, in order to reduce harmful greenhouse gas emissions.

At present, the most common practice when returning straw to land is the use of direct tillage technology, known as 'direct straw return remediation'. An alternative method is to compost the straw before incorporating the product into the soil, known as 'straw compost return remediation'. However, straw composts return remediation has rarely been compared to direct straw return remediation in terms of the impact on plant biomass and soil respiration, the primary path by which CO<sub>2</sub> fixed by land plant returns to the atmosphere.

Chongming Dongtan is located at the easternmost end of Chongming Island, Shanghai. Three large-scale land reclamation schemes have been undertaken in the past two decades to expand the land area available for agriculture. However, the soils on the reclaimed land have high salinity and form hard soil crusts, neither of which are conducive to good crop plant growth. As such, these soils need to be improved; however, improving measures usually disturb the soil conditions and lead to increased soil respiration. Therefore, it is necessary not only to improve soil structure and fertility, but also to limit the increase in soil respiration that may result from such measures.

As *Phragmites australis* is the most common plant in tidal flat areas and reclaimed land, as well as the most common straw resource on Chongming Island it is usually the alternative straw resource for soil amendment on Chongming Island. Although crop straw degradation has been previously studied (Liu *et al.* 2006; Shan *et al.* 2008), the organic matter retention of *P. australis* straw at soil improvement sites has seldom been reported not have the effects of the reed straw compost return and direct straw return methods been comprehensively evaluated in terms of the soil carbon budget and microbial activities.

In this paper, to investigate the effects of *P. australis* straw incorporation on plant biomass and the soil carbon budget, as well as determine the low carbon treatment for reclaimed land amendment, two treatments, based on *P. australis* straw incorporation, were used to improve reclaimed soil in Chongming Dongtan. Treatment 1 involved direct straw incorporation into the soil with tillage while in Treatment 2, composted straw was incorporated into the soil with tillage. SOC, plant biomass, and soil respiration rate were analyzed to assess the effects of the two treatments on the soil carbon budget capability. In addition, the soil microbial biomass and microbial communities were analyzed in order to ascertain the different soil respiration mechanisms in the sites with different straw returning modes.

## Materials and Methods

### Study area

Chongming Island is located in northeast Shanghai in the

estuary of the Yangtze River (Fig. 1). Our study period lasted from January 2015 to November 2015. The study area was in Chongming Dongtan (31°30'55.58"N, 121°57'05.28"E). Until soil improvement treatments were initiated in 2015, the area had not been artificially disturbed. The soil quality had degenerated since reclamation in 1998, and it was hindering the potential nutrient input brought by tides. Trial sites for remediation Treatments 1 and 2 were selected together with a control area located adjacent to the remediation treatment sites (Fig. 1).

### Experimental process

The treatments we compared were direct straw incorporation plus tillage (Treatment 1) and incorporation of composted straw plus tillage (Treatment 2). In Treatment 1, *P. australis* straw was cut into 25 cm lengths by a tree branch disintegrator (Trust TFS808Y, China), and directly incorporated into the soil by tillage. In Treatment 2, the straw was first pre-composted for two months with mixed microbial inoculums, which consisted of cellulolytic bacteria (Beijing Voto Sky & Land Biotech Co., Ltd.) and then incorporated into the soil by tillage. After the soil had been amended, both treatments and control areas were all seeded with *Sesbania cannabian*. Details of the soil improvement processes are listed in Table 1.

### Sample collection

Five sampling points were set up at each experimental site (Fig. 1). According to the standard sampling methods as outlined by Carter and Gregorich (1993), approximately 1 kg of surface soil sample (between -5 to -20 cm depth) was collected at each sampling point in January, July, September, and November 2015, taken to the lab, and stored at 4°C. Half of each sample was dried, ground and then sieved to < 0.25-mm particle size for SOC analysis. The other half of each soil sample was stored at 4°C for SR analysis.

### Soil physico-chemical analysis

SOC was measured by a SOC analyzer (TOC-VCPN Shimadzu, Japan) with an SSM-5000A solid burning device. Soil salinity was determined in a 1:5 soil: water slurry using a conductivity meter (HQ400d, Multi, HACH) and soil water content was determined after drying the sample at 105°C for 8h (Carter and Gregorich 1993). Soil bulk density was determined using the core cutter method. Soil respiration was tested by the LI-8100A Automated Soil Gas Flux System (USA) according to the method of Hu *et al.* (2014). Three polyvinyl chloride soil collars (10 cm in diameter) were installed at each sampling point and the gas was collected into a soil chamber connected to a soil respiration system. The SR was measured twice a day (at day and night) in each soil collar.

## Plant biomass analysis

Plant biomass was determined by measuring the above-ground dry weight biomass found one square meter quadrat. One quadrat was taken at each of the five sampling points. The whole sample was dried at 60°C until a constant weight was achieved.

## Soil microbial characteristics analysis

The soil microbial biomass was determined by measuring the content of adenosine triphosphate (ATP) (Yao and Huang 2006);  $\beta$  - glucosidase activity was determined by colorimetry (Tabatabai 1994).

Microbial diversity in the soil was measured in November using 16 S rDNA fingerprinting. Total soil DNA was extracted using the E.Z.N.A.® Soil DNA Kit (50) according to the instructions (the United States, Omega Bio-Tec, Inc.). The PCR-DGGE was determined following the method of Li *et al.* (2010).

## Data analysis

Statistical analyses were implemented using SPSS software (Version 16.0, IBM Inc., New York). Data were analyzed using the Analysis of Variance (ANOVA) for every five points at three sites sampled three times and collected per site. LSD was used for multiple comparisons.

The denaturing gradient gel electrophoresis (DGGE) profiles were analyzed using the UPGMA method (unweighted pair group method with arithmetic mean) using Quantity One software (Version 4.6.7, Bio-Rad Laboratories Inc., Hercules) to analyze the clustering similarity of the three sites. Soil microbial diversity was shown by the Shannon-Wiener Index through analyzing the DGGE profiles, and calculated by the following formula:

$$H = -\sum_i^S P_i \lg P_i \quad (1)$$

Where H is the Shannon-Wiener Index, S is the number of bands in the gel, and  $P_i$  is the relative abundance of the  $i$ th phenotype fraction.

## Results

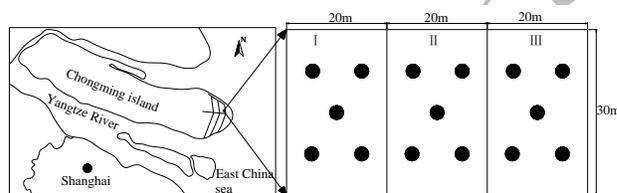
### Variability in soil organic carbon budget after improved with different straw returning modes

The results in Fig. 2 showed that both remediation treatments could increase SOC, indicating that the straw incorporation could favor SOC accumulation. The SOC content for Treatments 1 and 2 was 3.93 g kg<sup>-1</sup> and 3.94 g kg<sup>-1</sup>, respectively, which were 9.3 and 9.4% higher than that of the control at 3.60 g kg<sup>-1</sup>.

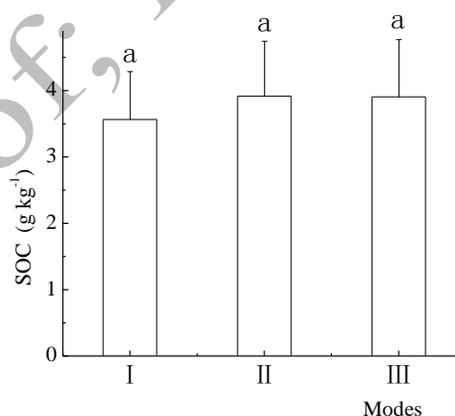
The results shown in Fig. 3 indicate that the plant biomass (*S. cannabian*) of Treatments 1 and 2 was 3.44 and

**Table 1:** Experiment process

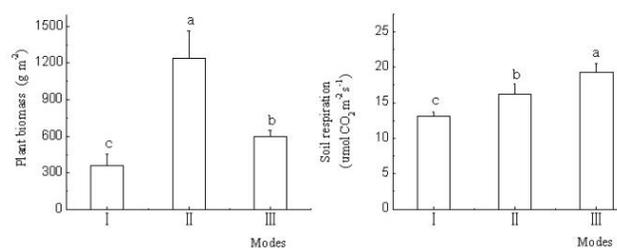
Treatment	Time	The control	Treatment 1	Treatment 2
Cut <i>P. australis</i> straw (kg/600 m <sup>2</sup> )	18 <sup>th</sup> , March	/	150	150
Pre-decomposed	29 <sup>th</sup> , March	/	/	Compost at tank
Return	13 <sup>th</sup> , June	/	Done	Done
Tillage	14 <sup>th</sup> , June	-20 cm surface	-20 cm surface	-20 cm surface
Plant	14 <sup>th</sup> , June	<i>S. cannabiana</i>	<i>S. cannabiana</i>	<i>S. cannabiana</i>
Plant harvest	09 <sup>th</sup> , November	Done	Done	Done
Area (m <sup>2</sup> )		600	600	600



**Fig. 1:** Sketch map of improved areas. I: The control; II: Treatment 1; III: Treatment 2



**Fig. 2:** SOC contents at three different modes. I: The control; II: Treatment 1; III: Treatment 2

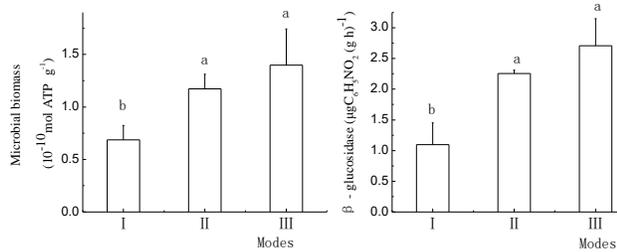


**Fig. 3:** Difference of plant biomass and soil respiration at three different modes. I: The control; II: Treatment 1; III: Treatment 2

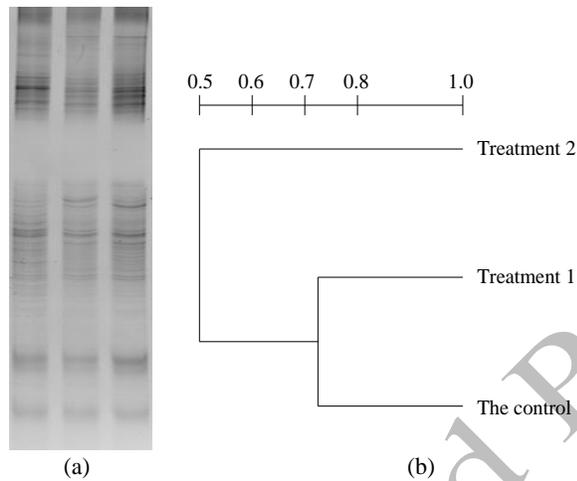
1.67 times that of the control, respectively. However, the soil respiration rate in Treatments 1 and 2 only increased by 23.5 and 46.4% over the control, respectively. The SR of Treatment 1 was lower than of Treatment 2. Soil respiration rates at the control site were the lowest, but the plant biomass weight was also significantly reduced. This indicated that the

**Table 2:** Analysis results of DGGE fingerprinting

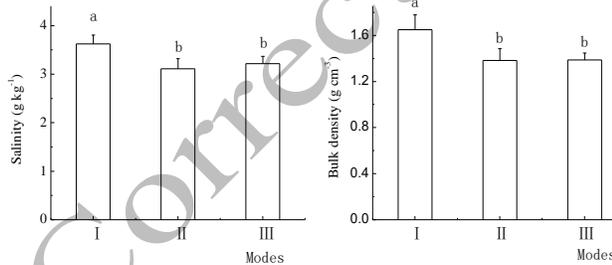
Study sites	Bands number of DGGE	Shannon- Wiener Index
The control	34	2.92
Treatment 1	32	2.16
Treatment 2	38	4.15



**Fig. 4:** Soil microbial biomass and  $\beta$ -glucosidase activity at three different modes. **I:** The control; **II:** Treatment 1; **III:** Treatment 2



**Fig. 5:** **a.** PCR-DGGE fingerprinting; **b.** UPGMA analysis of DGGE banding profiles at three different modes



**Fig. 6:** Soil salinity and bulk density at three different modes. **I,** The control; **II,** Treatment 1; **III,** Treatment 2

CO<sub>2</sub> fixed by photosynthesis had also decreased, as well as did the input of organic carbon from plant debris, which and caused a lower percentage of SOC at the control. According to the plant biomass and SR, the soil with Treatment 1 showed higher organic C input and lower organic C output, thus a higher organic carbon budget.

### Soil microbial characteristics after amendment with different returning modes and its relationship to SR

Further study of the soil microbial activities showed that the soil microbial biomass and  $\beta$ -glucosidase activity was higher at the treated sites compared to the control (Fig. 4). The tendency was Treatment 2 > Treatment 1 > the control. The microbial biomass in Treatments 1 and 2 was not significantly different, but the soil respiration rate for the two treatments was significantly different. This suggests that the microbial biomass might not be the only factor affecting soil respiration, and that microbial communities could play an important role. To clarify the cause of the different soil microbial respiration rates between Treatments 1 and 2, microbial diversity and bacterial clustering were compared between the treatments and the control.

Different microbial communities are indicated by different bands in DGGE fingerprinting (Fig. 5a). UPGMA clustering analysis showed that the similarity coefficient of the microbial community between Treatment 1 and the control was 72.5%, which was higher than that of Treatment 2 vs. Treatment 1 and the control at 50.5% (Fig. 5b). This indicates that the microbial community found in Treatment 2 soil was different from Treatment 1 soil and the control. Furthermore, the microbial diversity accounted for by the Shannon-Wiener Index was also significantly higher with Treatment 2 soil than Treatment 1 soil or the control (Table 2). One possibility could be that a more heterotrophic microorganism community was created by the incorporation of composted straw compared to direct straw incorporation or the control, enhanced microbial activity leading to a significant increase of soil respiration.

### Soil physical properties after amendment with treatments 1 and 2

The results showed that both remediation treatments could significantly decrease the soil salinity and bulk density ( $P = 0.001, 0.002 < 0.05$ ) (Fig. 6). Compared to the control, soil salinity was 15.6% lower with Treatment 1 and 12.8% lower with Treatment 2. Soil bulk density values were 15.1 and 15.8% less than that of the control, respectively (Fig. 6). Lower soil salinity and bulk density indicated that straw incorporation could increase the soil porosity and decrease the soil bulk density, which is conducive to the leaching of soil mineral salts, thereby promoting plant growth.

### Discussion

It has been widely reported that straw retention can increase soil respiration (Liu *et al.* 2014). Soil respiration could increase in remediated land because the conditions for organic matter decomposition, soil aeration and moisture content are often improved when soils were disturbed. In addition, SR has been positively correlated with microbial biomass under agroforestry systems (Lee and Jose 2003).

In this study, both treatments increased the microbial biomass, which resulted in higher soil respiration at the two treatment sites than at the control site (Fig. 3).

Returning straw leads to differences in microbial species communities (Guo *et al.* 2017; Su *et al.* 2020). In addition to the microbial biomass, differences in the soil microbial community could affect the ratio of carbon converted to carbon dioxide or to SOC (Rice 2002). Bastian *et al.* (2009) reported that residue decomposition induced significant changes in bacterial community dynamics with the magnitude in change between the different soil zones ordered as follows: residue > bulk soil. The microbial community at the Treatment 2 site was significantly different from that of Treatment 1 site and the control (Fig. 5a, 5b), as was the biodiversity (Table 2). Bacteria were the primary colonizers of plant detritus (Benner *et al.* 1986), straw compost improves the growth and reproduction of soil microbial, and active bacteria were propagated when the straw was composted, which explains why the soil  $\beta$ -glucosidase activity and soil respiration at the Treatment 2 site were significantly higher than at the Treatment 1 site. Furthermore, the practice of crop rotation and the differences in straw retention treatments could affect the soil respiration, temperature, and water content in the field (Kong *et al.* 2019). In addition, the microbial species differed in metabolic capacity and therefore decomposed a given compound at different rates (Killham 1994), which might also lead to different soil respiration rates. Therefore, the different microbial communities at the Treatment 1 and Treatment 2 sites could play an important role in affecting soil respiration rates. Analysis of the detail of microbial community composition continues to be ongoing.

SOC in the global carbon cycle depends on the stability of soil carbon and its availability to soil microorganisms (Zhang *et al.* 2009). The decomposition rates were negatively correlated with litter C: N ratio, cellulose, and lignin content (Chimney and Pietro 2006; Wang *et al.* 2020). Tu *et al.* (2006) found that composted cotton gin trash was more effective in enhancing microbial biomass and microbial activity than conventional synthetic fertilizer due to its favorable C:N ratios. In this study, the C/N ratio in the directly incorporated straw was 37.8 and 26.8 in the composted straw, which meant that carbon and nitrogen in the composted straw (Treatment 2) were more available to microorganisms and resulted in the observed increase in microbial activities and respiration.

It is reported that straw management could increase soil porosity and improve near-surface hydraulic properties (Blanco-Canqui and Lal 2007; Chen *et al.* 2016). It was found the soil salinity and soil bulk density after both treatments were significantly lower than the control (Fig. 6), because the straw incorporation by tillage could increase the porosity and decrease salinity through improved soil flushing thereby washing out excess dissolved mineral salts. It could be deduced that the incorporation of straw physically improved the soil quality and hence favored the

growth of *S. cannabiana*. Lou *et al.* (2011) proved that topsoil (0–20 cm) C storage significantly increased due to the increased residue C input. The plant biomass increased significantly in Treatments 1 and 2, and the SOC also improved. Although the SOC content after the two treatments was almost the same, the organic C input (from the death of plants) on the Treatment 1 site was significantly higher than on the Treatment 2 site and the organic C output (from soil respiration) was lower, which implies that the Treatment 1 sites had a higher soil carbon sequestration potential in the long term, and thus may be a low carbon improvement mode.

## Conclusion

In this study, we conclude that both direct straw incorporation and straw incorporation after compost could increase the SOC (9.3 and 9.4%, respectively) and plant biomass (3.44 and 1.67 times respectively) compared to the control. Both direct straw incorporation and straw incorporation after composting also enhanced SR compared to the control, but the enhanced degree by direct straw incorporation was significantly lower than that by straw incorporation after compost. According to the plant biomass and SR, direct straw incorporation is a low-carbon improvement mode for reclaimed lands compared to straw incorporation after composting. Composted straw incorporation biasing to soil microbial activities and changing the community structure could be an important cause of increased soil respiration compared to direct straw incorporation.

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## Author Contributions

QiuxiaoYin performed the experiment, analyzed the data and wrote the paper. Yanli Li contributed reagents/materials/analysis tools and performed the experiment, and Lei Wang conceived and designed the experiments.

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