

Impact Factor: 3.1 (UIF) DRJI Value: 5.9 (B+)

# Microbial mineralization of soil organic carbon influenced by long term fertilization in paddy soils under different temperature regimes

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#### Abstract:

The potential for CO<sub>2</sub> production from soil organic carbon (SOC) mineralization and its temperature dependence has been the subject of much research in the last decade. It is well known that SOC accumulation and decomposition are profoundly influenced by soil nutrient status. The extent to which fertilization practices impact upon SOC mineralization is a key issue for understanding the potential response of agricultural soils to global warming. Topsoil samples were collected from a long-term fertilization trial on a red earth rice paddy from Jiangxi Province, China and SOC mineralization was studied using aerobic incubation at 20 and 25 °C, respectively, SOC mineralization rates varied between 0.62 and 0.76 mgC  $g^{-1}OC d^{-1}$  at 20 °C, and between 0.65 and 0.97mgC g<sup>-1</sup>OC d<sup>-1</sup> at 25 °C, respectively. There was no significant correlation between the mineralization potential and SOC content for the samples under different treatments. However, a close correlation between total C mineralization and Kos, an indicator of the SOC portion resistant to chemical oxidation, was

found. This suggests a significant control of available C, rather than total SOC, on SOC mineralization. While no significant correlation was found between  $Q_{10}$  values and C/N, Corg, or Cmic/SOC, a potential dependence of warming could be indicated by a linear relationship between  $Q_{10}$  values and DCB-extracted Fe content of the samples under different treatments. Thus, the free oxyhydrates, rich in the red-earth derived paddy soil, and increased under the combined fertilization of manure and chemical fertilizers; tend to contribute to the chemical protection of SOC against enhanced mineralization that would be predicted by climate warming. Our results suggest that rational fertilization with combined chemical and manure fertilizers should be implemented to improve the chemical protection of SOC and, in turn, to reduce greenhouse gas emissions from red-earth-derived rice paddy.

Key words: Soil, Organic Matter, Carbon, Temperature, Fertilization

# Introduction

Global soil contains a huge stock of organic carbon (SOC) of about 1500 Pg and only small losses could cause changes in the atmosphere  $CO_2$  concentration (Kirschbaum, 1995). SOC mineralization and its temperature dependence have been widely debated when trying to assess the potential response of soils to global warming (IPCC, 2001; Valentini et al., 2000). Although Giardina and Ryan (2000) argued that soil warming may not affect SOC mineralization, more recent studies have suggested loss of SOC under climate warming (Fang et al., 2005; Knorr et al., 2005). Kirschbaum (2000), for example, analyzing available data from the literature and inferred that soil warming does affect soil C dynamics and soil-air CO<sub>2</sub> flux, since the rate of mineralization of SOC is widely found to correlate with soil temperature. Enhanced SOC mineralization under warming using laboratory incubation has been frequently reported for forest soils (Goulden et al., 1998; tundra

soils; Oechel et al., 1993, 1995), although this varies with soil type, pedogenic horizons, N availability and moisture regime (Bowden et al., 1998; Macdonald et al., 1999; Leirós et al., 1999). Many studies have shown that  $Q_{10}$  values of SOC mineralization are influenced mainly by soil temperature and moisture regime as well as N status (Gulledge et al., 2000; Xu et al., 2001; Qi et al., 2002). Little is known about how fertilization practice affects C mineralization or its temperature dependence in agricultural soils (Davidson et al., 2006).

Paddy soils are considered as a unique anthropogenic soil type formed under long-term hydroagric managements with seasonal submergence for rice growth in China (Gong, 1999). They play an important role in carbon sequestration (Pan et al., 2004; Pan et al., 2005a,b; Song et al., 2005) as well as in the food security of China, with an area of 30 Mha, and accounting for one guarter of the China's national cereals production (SSSSC, 1998). C enrichment has been observed in these paddy soils over the last two decades (Zhang et al., 2004; Li and Wu, 2006; Huang and Sun, 2006). Meanwhile, winter warming, as a result of global climatic change, has been observed recently in southern China, where 90% of the China paddy soils are distributed (Gal et al., 2003; Li, 1992). Red earth-derived paddy soils are a typical cropland soil in South China, and a consistent trend of SOC sequestration has been observed in recent decades. Chemical fertilizers have been increasingly applied, whilst organic manure application in the croplands of China has decreased over the past 20 years (FAOSTAT, 2005). However, little is known about whether different fertilization practices affect SOC sequestration and mineralization of the rice paddy soils in China.

In this study, we use laboratory incubation at 20 and 25 centigrade of paddy topsoil samples from a long-term fertilization trial from Jiangxi, China, to understand the variation of SOC mineralization potential and its temperature

dependence. From this, we suggest possible measures for GHG mitigation in China's rice agriculture.

# **Materials and Methods**

### Soil and the site of the long-term experiment

Soil samples were collected from a long-term field trial with different fertilization treatments, run by the Red Earth Institute, Jiangxi Academy of Agricultural Sciences in Jinxian Country, Jiangxi Province, China. The site is located at 28°15′N and 116°20′E at an elevation of 26m above sea level. Being derived from red earth in the downs of northeast Jiangxi, the soil is classified as a typical Hapludults according to Chinese Soil Taxonomy (Gong, 1999). A subtropical monsoon climate has been prevalent in the local area with mean annual temperature of 17.7 centigrade and mean annual precipitation of 1400mm between 1994 and 2004. The local mean double rice yield reached 9000 kg ha<sup>-1</sup> yr<sup>-1</sup> for the last 5 years.

# **Fertilization treatments**

Five different fertilization treatments were consistently used in the trial since 1981: no fertilization (NF), inorganic nitrogen fertilizer only (N), compound fertilizer of inorganic N and P (NP), and plus K (NPK), and compound fertilizer of inorganic N, P, K plus pig manure (NPKM). N is applied as urea at 150 kg ha<sup>-1</sup>, P<sub>2</sub>O<sub>5</sub> as calcium super-phosphate at 560 kg ha<sup>-1</sup>, and K<sub>2</sub>O as potassium chloride at 150 kg ha<sup>-1</sup> with early rice and at 180 kg ha<sup>-1</sup>, 560 kg ha<sup>-1</sup> and 150 kg ha<sup>-1</sup> with later rice, respectively. Pig manure is applied at 22500 kg ha<sup>-1</sup> (fresh weight) as basal fertilizer before rice seedling transplantation. A completely randomized block design with three replications was used. The size of each plot was  $45m^2$  (9m x 5m). The trial plots have been rotated with double rice since its initiation in 1981. The soil is submerged during early rice growth before ripening, and at the seedling stage of later rice within each year.

### Soil sampling and treatment

A composite soil sample was collected from topsoil at a depth of 0 to 15cm from each treatment plot after the rice harvest in July of 2005. After shipping to the laboratory, the crop detritus was removed and the soil mass divided into two portions. One portion was air-dried at room temperature and ground to pass a 0.25mm sieve for chemical analysis. Another portion was directly sieved to pass a 2mm-mesh sieve for the incubation experiment.

# Soil basic properties determination

Basic properties were determined using the conventional laboratory methods described by Lu (2000). Soil pH was measured using a Metter-Toledo pH meter with a soil:water ratio of 1:2.5, cation exchange capacity(CEC) with ammonium acetate (1 mol  $1^{-1}$ ; pH 7) leaching method, and free iron oxyhydrates (DCB-Fe) measured by dithionate-citrate-bicarbonate extraction, and determination of Fe by atomic adsorption spectro-photoscopy (AAS). Available P was extracted with 0.03M NH<sub>4</sub>F-0.025M HCl and determined with colorimetery of vanadomolybdo-phosphate. The moisture content of the samples was determined by oven-drying at 105 °C for 6h. Basic soil properties are listed in Table 1.

		-				
Treat- ments	pH (H <sub>2</sub> O)	TN (g·kg-1)	Olsen- P (mg·kg·1)	CEC (Cmol (+) kg <sup>.</sup> <sup>1</sup> )	DCB- Fe (g·kg <sup>.</sup> 1)	WHC (g·kg·1)
CK	5.50	2.27	4.00	9.13	19.09	488.7
Ν	5.32	2.30	4.09	8.68	18.48	507.1
NP	5.44	2.37	16.95	8.76	19.95	566.9
NPK	5.34	2.32	17.83	10.48	19.35	521.8
NPKM	5.54	2.68	54.43	13.47	17.44	554.7

Table 1 Basic physical and chemical properties of soil under different treatments sampled and measured in 2005

Soil water holding capacity (WHC) measurement

WHC of the soil for incubation was determined following the procedure described by Cai and Mosier (1999). Soil mass, ground to pass a 20 mesh sieve, was placed on the funnel with a cotton stopper and submerged for 2 h and the funnel top was then covered. The cotton stopper was removed, and the water of the submerged soil was allowed to fall freely for 12h. A portion of the soil was sampled to determine the moisture content and expressed as the WHC on the basis of dry soil mass.

# Soil C pool analysis

(1) Soil total organic carbon (Corg) and total nitrogen (TN) was measured with a CNS Macro Elemental Analyzer (Elementar Analysen Systeme GmbH, Germany, 2003).

(2) The Liable organic carbon pool (LOC) was determined with 0.333 M KMnO<sub>4</sub> following a procedure recommended by Blair et al. (1995). A portion of a soil sample containing about 15 mg C was weighed into a 50 ml centrifuge tube with a plastic screw top and 25 ml of 0.333 M KMnO<sub>4</sub> was added. The centrifuge tube was sealed tightly, and tumbled at 12 rpm for 1 h on a tumbler with a radius of 15 cm, and centrifuged for 5 minutes at 2000 rpm (corresponding to a relative centrifugal force of 815 g). The aliquot was diluted to 1:250 with deionized water. The absorbencies of the diluted samples and standards were read on a split beam spectrophotometer at 565 nm. The range for the standards was chosen to adequately cover the sample range, normally 300 to 333 mM. The quantity of consumed KMnO<sub>4</sub> was used to calculate the amount of carbon oxidized, assuming that 1mM KMnO<sub>4</sub> is consumed in the oxidation of 0.75 mM, or 9 mg, of carbon. A blank sample, containing no soil, and a reference sample were inserted for each determination run. The results were expressed as g C kg<sup>-1</sup> soil.

The organic carbon stability index (CSI) was calculated with the following equation:

CSI= (Corg-LOC)/Corg=1-LOC/Corg

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(3) Soil microbial biomass carbon (C*mic*) was determined using the chloroform fumigation-extraction method described by Jenkinson and Powlson (1976) and modified by Voroney et al. (1993) as follows: 25 g of fresh soil was fumigated at 25 °C for 24h prior to extraction with K<sub>2</sub>SO<sub>4</sub>. Fumigated and unfumigated samples were extracted with 100 ml of 0.5 M K<sub>2</sub>SO<sub>4</sub> for 1 h. The organic C in the extracts was determined with a TOC analyzer (Jena MultiN N/C 2100, 2005), and the C*mic* was expressed as the difference in OC between the un-fumigated and fumigated samples following Voroney et al. (1993).

# Laboratory incubation of SOC mineralization

In this study, SOC mineralization of the soils was studied with laboratory incubation experiment using a device shown in Fig. 1. The procedure was as follows: 20g of dry weight equivalent fine soil (2mm mesh-sieved) was put into a 125ml incubation bottle. The required quantity of distilled water was added to simulate the field condition of approximate 60%WHC (Schinner et al., 1993). The bottle was then sealed by a silica-rubber stopper, mounted with 2 turflon tubes for gas sampling and air filling (Meng et al., 2005). The bottle was filled with standard air (Commercial purified air) for 2min at a flow rate of 200ml min<sup>-1</sup>. The soil moisture within the bottle was maintained by weight balance after each CO<sub>2</sub> evolution sampling, avoiding any significant disturbance. The incubation was conducted at 20 °C and 25 °C for 58 days in a temperature- and humidity-constant (LRH-250-S. Medicine incubator Machinerv Co. Ltd.. Guangdong, China, 2002), respectively. The experiment was conducted in triplicate. A blank without soil was used as the control of gas concentration in the bottle. Gas samples were collected on 0.5, 1, 1.5, 2, 2.5, 3.5, 4.5, 6, 8, 10.5, 13.5, 16.5, 20, 23.5, 27.5, 31.5, 35.5, 39.5, 43.5, 47.5, 52.5, 58 days after initiation of the incubation.  $CO_2$  concentration of the collected gas samples were determined by Gas Chromatography (Agilent 4890D) equipped with a stainless steel column (Porapak Q)

(80/100 mesh) and flame-ionization detector (FID). Column, injector and detector temperature were 35, 130 and 250 °C, respectively. Nitrogen gas (carrier gas), FID hydrogen and FID airflow were set at the rate of 30, 45 and 400ml min<sup>-1</sup>, respectively. The C mineralization rate in any given time interval was calculated from the quantity of  $CO_2$  produced, and normalized on the basis of the initial Corg contents of the sample.



Fig.1. A sketch diagram of the incubation device

# Statistical analysis

Statistical differences were tested with the analysis of variance procedure (ANOVA) using the SPSS11.0 statistical package. Statistical significance was determined at the 95% or 99% confidence level.

# **Results and Discussion**

# The C pool of soils under different fertilization treatments

As shown in Table2, the Corg and C*mic* ranged from 19.71g kg<sup>-1</sup> and 1123.2 mg kg<sup>-1</sup> under NF, to 24.24 g kg<sup>-1</sup> and 1852.6 mg kg<sup>-1</sup>under NPKM, respectively. The contents of LOC followed a similar trend. This suggests that fertilization using compound

inorganic and organic fertilizers greatly enhances SOC accumulation in the paddy soil. Greater variation among fertilization treatments was found for *Cmic* and LOC than for Corg. Of the three treatments with inorganic fertilizations, Corg was significantly higher under in plots treated with P fertilization, than those without P fertilizers. However, Cmic and LOC were much higher under NPKM than under any of the inorganic fertilization treatments. While no significant correlations of Cmic and LOC was found with Corg, a strong positive correlation was observed between the microbial quotient (Cmic/Corg) and total N in the soil, with a weaker correlation with soil Olsen-P. (Fig.2). These data suggests that fertilization practices have greatly affected the portioning of OC pools, rather than the size of the pools, through the of soil nutrient availability for modification microbial utilization.

Table 2 Pools of SOC of the studied soil under different fertilization treatments

<b>m</b> , , ,	$\mathrm{C}_{\mathrm{org}}$	$\mathrm{C}_{\mathrm{mic}}$	LOC
Treatments	$(g \cdot kg^{-1})$	$(mg \cdot kg^{-1})$	$(\mathbf{g} \cdot \mathbf{kg}^{-1})$
CK	$19.71 \pm 0.24c$	$1123.2\pm35.2c$	6.24±0.19d
Ν	$19.74 \pm 0.28c$	$1174.1 \pm 15.6c$	$7.25 \pm 0.22c$
NP	$20.45 \pm 0.12 b$	1383.4±41.7b	$7.99{\pm}0.24b$
NPK	$20.41 \pm 0.22 b$	1273.5±34.1c	$7.49\pm0.22c$
NPKM	24.24±0.30a	$1852.6 \pm 50.5a$	11.48±0.31a



Fig.2 Correlation of the microbial quotient with total N (A) and available P (B) of soil

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### Dynamics of C mineralization and total C mineralized

The dynamics of C mineralization among the samples is shown in Fig.3. Three stages of SOC mineralization dynamics could be recognized: a brief period of rapidly increasing C mineralization in the first 2days, followed by a period of high and rapidly decreasing mineralization during days 2.5 to 25, and finally a period of low and stable mineralization in the last 27 days. The mean SOC mineralization rate ranged from 0.30 mgC g<sup>-1</sup> Corg d<sup>-1</sup> to 1.01 mgC ·g<sup>-1</sup>·Corg d<sup>-1</sup> in the first stage, from 1.25 mgC ·g<sup>-1</sup> <sup>1</sup>·Corg d<sup>-1</sup> to 1.85 mgC g<sup>-1</sup>·Corg d<sup>-1</sup> in the second stage and from 0.23 mgC ·g<sup>-1</sup> ·Corg d<sup>-1</sup> to 0.31 mgC ·g<sup>-1</sup> ·Corg d<sup>-1</sup> for the final stage. These three phases could represent three sub-pools of mineralizable C in the soils: an active, a slow and a passive mineralizable C pool, as described by SOM models such as Century (Parton et al., 1987) or RothC (Jenkinson, 1990). As shown in Table 3, the contribution of each stage to the total mineralized C was 1%-5%, 66%-73% and 24%-30% for the first, second, and third stage, respectively, demonstrating that the first 3.5 weeks is the key period for C mineralization under incubation. The fact that C mineralization rate varied with incubation time could indicate different microbial activity and C pool bioavailability, which varied throughout the incubation.

The difference of mineralization rate also existed between treatments at different stages under same temperature. In the first stage, the mean mineralization rate from different treatments follows the order: N and NPKM >NP CK, NPK at 20 °C, and NPKM and N >NP, CK> NPK at 25 °C. However, for the second stage, the rate follows the order: NPKM > N and NPK at 20 °C, and NPKM > NP, N and NPK at 25 °C, respectively, with the rate under CK being the lowest for both temperature regimes. In the last stage, there were no treatment differences in mean mineralization rate at either temperature, though rates were higher at 25 °C than at 20 °C.



Fig.3 Dynamics of SOC mineralization and  $CO_2$  production of the samples under different fertilization treatments during incubation at 20 °C and 25 °C respectively. ( $\diamond$ :CK;  $\bullet$ :N;  $\bullet$ : NP;  $\diamond$ : NPK;  $\diamond$ : NPKM)

stages during incubation					
Treatm	Tem	Total C mineralized (mgCO <sub>2</sub> -C)			
ents	pera	0-2.0 days	2.0-23.5 days	23.5-58.0 days	
	ture				
CK	20°C	14.61±0.40 (2.08)Cb	497.8±15.9 (71.0)Ab	189.0±17.6 26.9)Ba	
	25°C	31.98±0.29 (4.33)Ca	528.9±17.6 (75.6)Aa	178.2±9.1 (24.1)Ba	
Ν	20°C	17.18±0.92 (2.27)Cb	585.7±23.2 (75.2)Aa	181.5±4.9 (23.3)Ba	
	25°C	38.66±1.35 (4.54)Ca	606.7±32.8 (75.6)Aa	173.0±13.0 21.5)Ba	

658.3±30.5 (73.5)Aa

669.3±30.9 (74.8)Aa

573.3±18.3 (75.8)Ab

621.8±20.6 (73.1)Aa

780.5±17.6 (77.0)Ab

1034.7±31.2 (76.2)Aa

223.3±18.7 24.9)Ba

192.8±8.0 (21.5)Bb

165.7±17.2 21.9)Bb

190.4±3.4 (22.4)Ba

213.2±9.1 (21.0)Bb

274.7±4.0 (20.2)Ba

NP

NPK

NPKM

20°C

25°C

20°C

25°C

20°C

25°C

14.50±0.55 (1.62)Cb

33.28±0.88(3.72)Ca

12.05±0.58 (1.55)Cb

23.15±0.92 (2.88)Ca

19.3±0.3 (1.9)Cb

48.9±2.3 (3.6)Ca

Table 3 Amount of C mineralized of the samples from the in different stages during incubation

Note: 1.Different capital letters and lower letters indicated significant difference in C mineralization amount between different stages and between the temperature regimes (p < 0.05). 2, The value in parenthesis was the mean portion (%) of C mineralized in a given stage to the total in the whole duration.

# Variation of SOC mineralization with temperature

The total C mineralized, and the mean mineralization rate as calculated from  $CO_2$  evolved from the samples under different treatments, is presented in Table 4. SOC mineralization rate and its variation among treatments in the first stage at 25 °C were much higher than at 20 °C. While a significant difference in mean mineralization rate between the two temperature regimes could be detected in all samples during the first stage, the significance of the difference in the later two stages varied with the fertilization treatments, suggesting a change in C bioavailability under warming among the different fertilization treatments. For the first stage, the C mineralization rate of all the samples was significantly higher under 25 °C ranging from 0.56 mgC ·g<sup>-1</sup>Corg ·d<sup>-1</sup> to 1.01mgC ·g<sup>-1</sup>Corg ·d<sup>-1</sup>, than that under 20 °C, ranging from 0.30 mgC ·g<sup>-1</sup>Corg ·d<sup>-1</sup> to 0.44mg C ·g<sup>-1</sup>Corg ·d<sup>-1</sup>. However, differences in the C mineralization rate for the later two stages of the incubation between the two temperature regimes varied with the different fertilization treatments.

There was no great increase in the rate under any given treatment, and almost no change in the variability between the treatments at 25 °C compared to that at 20 °C.

Table 4 Mean SOC mineralization rates and the variation in different
stages during the incubation at different temperature regime

Treatmonts	Mean C mineralization rate (mgC g <sup>-1</sup> OC d <sup>-1</sup> )					
Treatments	0-2.0 days	2.0-23.5 days	23.5-58.0 days	Whole period		
20 °C						
CK	0.36±0.014Bb	1.17±0.038Da	$0.28 \pm 0.008 Bc$	$0.62 \pm 0.014 \text{ C}$		
Ν	0.44±0.012Ab	1.38±0.040Ba	$0.27{\pm}0.025\mathrm{Bc}$	$0.67 \pm 0.015 B$		
NP	0.35±0.014Bb	1.50±0.038Aa	0.32±0.030Ab	$0.76 \pm 0.022 A$		
NPK	$0.30{\pm}0.017{\rm Cb}$	1.30±0.030Ca	$0.25 \pm 0.006 Bc$	$0.66 \pm 0.017 B$		
NPKM	0.40±0.031Ab	1.43±0.062ABa	$0.24 \pm 0.006 Bc$	0.73±0.021A		
Average	0.37	1.36	0.27	0.69		
CV(%)	14.30	9.37	11.45	8.18		
25 °C						
CK	0.81±0.032Bb	$1.25 \pm 0.030 \text{Da}$	$0.26 \pm 0.014 Bc$	$0.65 \pm 0.019 \mathrm{C}$		
Ν	0.98±0.023Ab	1.43±0.024Ca	$0.25{\pm}0.005\mathrm{BCc}$	$0.75 \pm 0.033 B$		
NP	0.81±0.033Bb	$1.52 \pm 0.027 Ba$	$0.24 \pm 0.011 BCc$	$0.76 \pm 0.027 B$		
NPK	$0.56 \pm 0.039 C$	1.28±0.024D	0.23±0.011C	$0.68 \pm 0.019 C$		
NPKM	$1.01 \pm 0.051 \text{Ab}$	1.85±0.020Aa	$0.31 \pm 0.014 Ac$	$0.97 \pm 0.030 aA$		
Average	0.83	1.47	0.26	0.76		
CV(%)	21.49	16.46	12.07	16.43		

Note: 1.Different lower case letter indicated significant difference in C mineralization rate between different stages (p < 0.05). 2. Different capital letters indicated the significant difference in C mineralization rate between treatments under a single temperature (p < 0.05).

The temperature dependence of SOC mineralization was examined by calculating the  $Q_{10}$  value. Very small  $Q_{10}$  values were obtained for C mineralization over the whole period under all treatments except for MNPK treatment with fresh, bioavailable OM (Table 5). In the first stage, lasting only 2.5days however, much higher  $Q_{10}$  values of up to 5~6 under 25 °C were detected under all treatments, despite their minor contribution to total mineralization. The warming effect on SOC mineralization in the second and last stages was small, except for the NPKM treatment. When considering all

treatments together, an overall effect of warming on SOC mineralization was not observed.. While C mineralization rate was significantly higher at 25 °C than that at 20 °C for NPK and NPKM treatments in the third stage, the effect of warming on the C mineralization was not significant under CK and N treatments, where the C mineralization rate was even lower at 25 °C than at 20 °C in the third stage.

Treatments	Temperature.	0-2.0d	2.0-23.5d	23.5-58.0d	Whole period
СК	20°C	0.36±0.014 b	1.17±0.038 b	0.28±0.008 a	0.62±0.014 b
	25°C	0.81±0.032 a	1.25±0.030 a	0.26±0.014 a	0.65±0.019 a
	$Q_{10}$	$5.06 \pm 0.137 C$	1.14±0.019B	$086\pm0.044B$	$1.11 \pm 0.016C$
	20°C	0.44±0.012 b	138±0.040 a	$0.27 \pm 0.025$ a	0.67±0.015 b
Ν	25°C	098±0.023 a	1.43±0.024 a	0.25±0.005 a	0.75±0.033 a
	$Q_{10}$	496±0.040C	$107 \pm 0.026$ C	$0.86 \pm 0.127 B$	1.27±0.073B
	20°C	0.35±0.014 b	150±0.038 a	0.32±0.030 a	0.76±0.022 a
NP	25°C	0.81±0.033 a	152±0.027 a	0.24±0.011 b	0.76±0.027 a
	$Q_{10}$	536±.008B	$1.03\pm0.016C$	$0.56 \pm 0.055 C$	$1.00\pm 0.013E$
	20°C	0.30±0.017 b	130±0.030 a	0.25±0.006 a	0.66±0.017 a
NPK	25°C	0.56±0.039 a	128±0.024 a	0.23±0.011 b	0.68±0.019 a
	$Q_{10}$	$3.48 \pm 0.091 D$	0.97±0.008D	$0.85 \pm 0.040 B$	$1.06\pm 0.005 D$
NPKM	20°C	0.40±0.031 b	1.43±0.062 b	0.24±0.006 b	0.73±0.021 b
	25°C	1.01±0.051 a	$1.85\pm0.020$ a	0.31±0.014 a	0.97±0.030 a
	$Q_{10}$	6.38±0.338A	1.67±0.109A	$1.67 \pm 0.067 A$	1.79±0.08A

Table 5 Mean SOC mineralization rate and  $Q_{10}$  value in different stages during the whole incubation

Note: 1. Different lower case letters indicate significant difference at p < 0.05 in same treatment under different temperature (p < 0.05). 2. Different capital letter indicated the significant difference  $Q_{10}$  value between treatments (p < 0.05).

# 4 Discussions

# C mineralization and soil C pools

There is a significant relationship under the different treatments under both temperature regimes between the mineralized C pool and the pool size of Cmic and LOC respectively in each treatment, but not the Corg content. This suggests that C mineralization and  $CO_2$  production potential from this red earth-derived paddy under different fertilization

management regimes, was not dependent on total SOC content. Zhang et al. (2006), reported independence of aerobic C mineralization on the SOC pool of topsoils from three geologically different paddy soils. In a previous study, using anaerobic laboratory incubations, SOC mineralization and  $CO_2$ and  $CH_4$  production potential were found to be independent in topsoils from a rice paddy fertilizer trial from the Tai lake region, Jiangsu, China (Zheng et al., 2007). Nevertheless, negative correlations were observed here between SOC mineralized and the OC stability index CSI and the ratio of (Corg-LOC) to Corg, but not with the size of a single carbon pool (Fig.4).

Although the CSI values of the studied soils under fertilization treatments are similar to those reported for paddy soils from the Tai Lake Region of China (CSI ranges from 0.63 to 0.67; Li et al., 2000), great variation in CSI was found among the different long-term fertilization practices. It appears that fertilization practices have affected SOC different mineralization intensity through changing SOC stability, possibly via different binding strength of SOC to the iron oxyhydrates, which are richer in red-earth-derived soils (Zhou et al., 2007). Song (2005) observed a low ratio of liable OC to SOC, in correlation with the high portion of OC bound to DCBextracted oxy-hydrates for the treated soils. Pan et al (2003) argued that protection and stabilization of OC by iron oxyhydrates could be a more important mechanism for chemical protection in China's paddy soils than is clay content. The negative correlation of mean C mineralization rate with the stability index could help to explain the rapid OC enhancement in the paddy soils observed in the red soil region over the last two decades (Pan et al., 2003; Huang and Sun, 2006; Zhang et al., 2004; Li Zhong-pei et al., 2006). Hence, chemical protection of SOC plays an important role in the production of  $CO_2$  upon controlled mineralization through affecting the microbial utilization.

Microbial mineralization of soil organic carbon influenced by long term fertilization in paddy soils under different temperature regimes 1400
y = -2090.8x + 2119.4
y = -2090.8x + 2119.4
x<sup>2</sup> = 0.9267

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Fig. 4.Correlation of mineralization as calculated from CO₂ production and SOC stability coefficient (A: 20 ℃; B: 25 ℃)

# Temperature dependence of SOC mineralization in response to different long-term fertilization treatments

It has been widely demonstrated that warming promotes  $CO_2$ production and flux from the soil (Schlesinger et al., 2000, Kirschbaum, 1995, Macdonald et al., 1999). The sensitivity of SOC mineralisation to temperature is often expressed as a  $Q_{10}$ value. Bowden et al. (1998) reported the Q<sub>10</sub> value was around 2 under laboratory conditions from a temperate zone soil. Leirós et al. (1999) showed that under laboratory incubation conditions, the Q<sup>10</sup> value of woodland soils from the temperate zone ranged from 2.5 to 4.0. Smith et al. (2003) suggested that the Q<sub>10</sub> values of representative soils ranged from 1.8 to 2.5, and varied greatly with soil type. Comparatively, the Q<sub>10</sub> values of the paddy soils studied here were significantly lower ranging from 0.99 to 1.79, showing a weaker temperature dependence for aerobic SOC mineralization. Zhang et al. (2006) reported Q<sub>10</sub> values ranging from 1.0 to 2.4 under laboratory aerobic incubations for three typical pedogenetic types of paddy soils from China.

Total SOC mineralized was found not to be well correlated with the Corg pool at either temperature (Fig. 5), but was correlated well with the Cmic pool well at 20 °C, and weakly at 25 °C (Fig.6). The decomposition of SOC is widely attributed to microbial activity in soils. Temperature dependence of OC decomposition of paddy soils may be influenced by both the availability of the OC pool, the pool of microbial biomass and its structure (Zhou et al., 2003; Zhang et al., 2005). Zhou et al. (2003) attributed enhanced SOC mineralization under warming of some typical paddy soils from the Tai Lake region China, to the increased microbial mass, which in turn, allowed enhanced C utilization. The results shown in Fig.6 and 7 suggest that the microbial attack on SOC by indigenous soil microbial mass becomes less significant under warming. In other words, under chemical fertilization treatments without fresh OM inputs the soil, microbial attack did not respond to warming. This may explain why warming does not significantly enhance SOC mineralization, due to unavailability of SOC to microbial attack.



Fig.5. Relationship between mineralization and the Corg pool (A: 20 ℃; B: 25 ℃)

Zhang et al. (2006) reported a warming effect on SOC mineralization controlled by soil C/N ratio in an aerobic

incubation using three geologically different types of paddy soils. In this study, neither of SOC nor microbial quotient, nor C/N was found to be correlated with the calculated  $Q_{10}$  values (Fig.8). Nevertheless, a close relationship was observed between  $Q_{10}$  and DCB-Fe content of the samples under different fertilization treatments (Fig.9). This supports the previous assertion, that C availability was not determined by indigenous soil microbial biomass, and that lack of accessibility prevents enhanced C utilization under warming. The link between Q<sub>10</sub> values and free iron oxyhydrates in paddy soils provides evidence for an important role of chemical protection of SOC through binding to pedogenetical components. Pan et al. (2003) have previously suggested that free iron oxyhydrates may play an important role in the chemical protection of OC in China's paddy soils. Oxyhydrates, rich in these paddy soils, may help to protect C as well as to enhance C sequestration in these soils. also conferring low C mineralization rates and low sensitivity to global warming in these soils.



Fig.6. Correlation of SOC mineralized with the microbial quotient of the samples (A: 20  $^\circ\!\!C$  ; B: 25  $^\circ\!\!C$ )



Fig. 7 Relationship between the mean  $Q_{10}$  values and  $C_{\text{org}}$ ,  $q_{\text{mic}}$ , C/N of the samples incubated



Fig. 8 Correlation of  $Q_{10}$  values with content of DCB-Fe contents of the samples incubated.

#### Conclusion

The Kos of paddy soils is obviously higher than that of natural or cropland soils suggesting that OC in paddy soils is more stable than in non-paddy soils. The combined application of mineral and organic fertilizer has had a markedly positive

effect on SOC sequestration, mainly through modified the partitioning of the OC pool rather than through changing the size of the OC pools, via amendment of soil nutrients and enhanced availability for microbial utilization. The С mineralization rate was not promoted significantly by temperature under balance fertilization, which indicated soil warming may be not lead to greater losses of OC under balanced fertilization treatments. There was ล close relationship between Q<sub>10</sub> and DCB-Fe content, but not between Q10 and SOC, Cmic/SOC or C/N, which implies that the protection of OC by DCB-Fe is the main mechanism for SOC stabilisation in red earth-derived paddy soils, also affecting the sensitivity of SOC mineralization to temperature.

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