

# Decomposition of particulate organic matter is more sensitive to temperature than the mineral associated organic matter



D.K. Benbi\*, A.K. Boparai, Kiranvir Brar

Department of Soil Science, Punjab Agricultural University, Ludhiana 141004, India

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## ABSTRACT

Temperature sensitivity of soil organic matter decomposition is important in determining the role of soils in future climate change. We isolated coarse and fine particulate organic matter (cPOM and fPOM) and mineral associated organic matter (MinOM) to represent labile, relatively less labile and stable pools of soil organic matter (SOM), respectively and incubated each at four different temperatures to determine temperature sensitivity of decomposition. The coarse particulate organic C, which comprised the smallest pool of soil organic C (SOC) was most decomposable and the mineral associated organic C that accounted for more than half of the SOC was least decomposable. At all the temperatures, the C mineralization rate followed the order cPOM  $\geq$  fPOM > whole soil > MinOM. The disparity in the mineralization rates between cPOM and the other two SOM fractions and the whole soil widened with increase in temperature from 15° to 45 °C indicating that the labile pools of SOM were more sensitive to temperature than the stable pool. The Arrhenius, the Llyod and Taylor and the Gaussian models well-described the temperature dependence of organic matter decomposition, but the shape of the temperature response curve for different models varied considerably. Gaussian model yielded the highest decomposition  $Q_{10}$  and the Arrhenius model the lowest  $Q_{10}$  for different SOM fractions and whole soil. The decomposition temperature response of isolated SOM fractions mainly differed at temperatures below 25 °C beyond which the response tended to converge suggesting that the differential response of labile and stable pools to temperature will be foremost at temperatures below 25 °C beyond which the effect will be small and similar for SOM pools of different lability. The decomposition of cPOM fraction is likely to be influenced to the greatest extent and the MinOM at the least as a result of global warming.

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## 1. Introduction

In the terrestrial C cycle soil respiration represents the second largest flux ( $\sim 60$  Gt C yr<sup>-1</sup>) between ecosystems and the atmosphere and a small change in soil respiration could significantly intensify or mitigate atmospheric increase of CO<sub>2</sub> (Baveye, 2007; Reichstein and Beer, 2008). The decomposition of soil organic matter (SOM), which is a primary source of soil respiration, is significantly influenced by temperature (Kirschbaum, 1995; Zhang et al., 2006). It is, therefore, speculated that increase in temperature due to global warming can accelerate the decomposition of SOM and consequently increase the release of soil organic carbon (SOC) to the atmosphere (Davidson et al., 2000). These concerns have stimulated interest in understanding the temperature sensitivity of soil respiration and organic matter decomposition,

especially with regard to the factors that determine the temperature dependence of C mineralization (e.g., Lloyd and Taylor, 1994; Kätterer et al., 1998; Luo et al., 2001; Sandermann et al., 2003; Reichstein et al., 2005; Conant et al., 2011; Curtin et al., 2012; Wagai et al., 2013). Several studies have shown that C mineralization increases with increase in temperature and the relative increase depends on reference temperature (Field et al., 2007; Zheng et al., 2009). The relative increase in mineralization is greater at low reference temperature than at high temperature.  $Q_{10}$  values ranging from about 8 at 0 °C to 2.5 at 20 °C have been reported (Kirschbaum, 1995; Zhang et al., 2006). However, these studies described the decomposition temperature sensitivity of bulk SOM without recognizing the effect of organic matter quality.

The quality of SOM is the foremost factor that could influence the temperature response of organic matter decomposition. Questions have arisen, whether the temperature sensitivity of SOM decomposition varies with the composition of organic matter, that is, labile Vs stable soil C pools (Liski et al., 1999; Davidson et al., 2000; Giardina and Ryan, 2000). A number of models and methodologies have been used to express temperature sensitivity of the

\* Corresponding author. Tel.: +91 161 2401960x317, +91 9464035154 (mobile); fax: +91 161 2409257.

E-mail address: [dkbenbi@yahoo.com](mailto:dkbenbi@yahoo.com) (D.K. Benbi).

decomposition of labile and stable SOM pools with contradictory results (Powlson, 2005; Kirschbaum, 2006; von Lützow and Kögel-Knabner, 2009). Knorr et al. (2005) postulated that the dominant slow pools of organic carbon are more sensitive to temperature than the faster pools causing a larger positive feedback in response to global warming. On the contrary, Reichstein et al. (2005) showed that this conclusion is equivocal and argued that it is premature to conclude that stable soil C is more sensitive to temperature than labile C and there could be very similar responses of labile and stable SOM decomposition to temperature (Fang et al., 2005). The conflicting results may partially be attributed to the range of methods used to estimate SOM decomposition temperature sensitivity, and the inability to consistently define and quantify labile and stable SOM (Kirschbaum, 2006; von Lützow and Kögel-Knabner, 2009).

A variety of physical and chemical fractionation methods have been developed to characterize SOM pools with distinct physico-chemical properties, functions, and turnover times (Hassink et al., 1997; Christensen, 2001; Six et al., 2001). Recent findings suggest that C turnover in soils is only weakly controlled by the molecular structure (Marschner et al., 2008). Similarly, it has also been suggested that decomposition of SOM is independent of microbial biomass size, community structure or specific activity, but is governed by abiological processes (Kemmitt et al., 2008). Therefore, studies that described the decomposition temperature sensitivity of chemically or kinetically defined SOM fractions were unable to precisely characterize labile and stable SOM pools and yielded conflicting results (e.g., Conant et al., 2008b; Plante et al., 2010). In contrast, physical stabilization mechanisms such as adsorption on mineral surfaces and occlusion in aggregates have been suggested to effectively stabilize soil C and thus influence turnover rates (Torn et al., 1997; Marschner et al., 2008). Studies published in the last three decades have shown that physical fractionation of SOM according to size provides a useful tool for the study of its functions and turnover in soil (Cambardella and Elliott, 1992; von Lützow et al., 2007). Soil organic matter is generally fractionated into coarse particulate organic matter (cPOM; size >250  $\mu\text{m}$ ), fine particulate organic matter (fPOM; size 53–250  $\mu\text{m}$ ) and mineral associated organic matter (MinOM; size <53  $\mu\text{m}$ ). Measures of POM have been tied to microbial growth and nutrient supply and suggest that it is closely related to biologically mediated C and N, and aggregation (Buyanovsky et al., 1994; Hassink et al., 1997; Gregorich and Beare, 2007). Accordingly, POM is commonly used as an index of the labile SOM pool and cPOM and fPOM are considered to represent active or labile and slow or relatively less labile pools of SOM, respectively (Benbi et al., 2012). The mineral associated organic carbon (MinOC) that includes physically and chemically stabilized organic carbon with low bioavailability and long turnover times is resistant to decomposition and is considered to represent passive pool of SOC (Parton et al., 1988; Six et al., 2002; Krull and Skjemstad, 2003; Benbi et al., 2012). Although cPOM, fPOM and MinOM have been suggested as SOM pools with differential turnover times, little information exists about the actual decomposability and temperature sensitivity of isolated SOM fractions (Swanston et al., 2002; Leifeld and Fuhrer, 2005; Mutuo et al., 2006; Plante et al., 2010). Knowledge about temperature dependence of mineralization of SOC fractions will help in understanding the mechanism of C-turnover in soils. Focus on POM, instead of other measures of labile SOM, is warranted mainly because this fraction typically has a higher proportional response to management than do other measures of labile SOM (Conteh et al., 1998; Alvarez and Alvarez, 2000; Carter, 2002). Recalcitrant SOM fractions that are equated with the passive or resistant pools have greater relevance for long-term C sequestration, sorption, and cation exchange capacity. Therefore, in order to predict the influence of global

warming on SOM decomposition, it is crucial to understand the decomposition temperature sensitivity of physically separated labile and recalcitrant SOM fractions. We hypothesized that labile SOM fraction reacts faster to temperature changes than the stable pools because it has shorter turnover times, easily available to microorganisms and rapidly decomposable. Conversely, the stable SOM fraction that is bonded onto the surface of minerals such as clay and silt and might be biochemically recalcitrant will react slowly to temperature changes. To test this hypothesis, we studied the decomposition temperature sensitivity of physically isolated labile and stable SOM fractions. The results of the study will help in understanding the feedback of SOM to global warming. If labile SOM pools exhibit greater temperature sensitivity than the stable SOM pools, the response to  $\text{CO}_2$  evolution would be relatively small in magnitude and short-lived and vice versa. We used different temperature response models (see Appendix A) for upscaling experimental results and to predict the influence of warming on SOM decomposition.

## 2. Materials and methods

### 2.1. Site characteristics and soil sampling

Soil samples were collected from the surface layer (0–15 cm) of four selected treatments of two on-going field experiments on rice-wheat system at Punjab Agricultural University research farm Ludhiana (30° 56' N, 75° 52' E and 247 m above sea level), India. The aim was to select soils with variable input of organic matter so that these differed in organic C content. The selected soils had received one of the following treatments for 6-years preceding sampling: i) RS—an annual input of rice straw at 10  $\text{Mg ha}^{-1}$  and fertilizer NPK at 90 kg N, 30 kg  $\text{P}_2\text{O}_5$  and 30 kg  $\text{K}_2\text{O ha}^{-1}$  ii) IN-application of fertilizer NPK as per (i) above, iii) FYM—an annual input of FYM at 40  $\text{Mg ha}^{-1}$  on dry weight basis, and iv) CONT—no input of organic and inorganic sources. Samples were collected after wheat harvest (end of crop cycle) in May, 2012 from three replicates of each treatment. The soils from both the experiments were sandy loam in texture (Typic Ustorthents; USDA, 1999), non-saline and neutral in reaction ( $\text{pH} = 7.03$ ).

The experimental area is characterized as semiarid sub-tropical with monsoonal climate. Annual rainfall ranges between 700 and 800 mm and more than 70% of it occurs during the monsoon months of July to September. The mean monthly minimum and maximum air temperature averages 18° and 35 °C during rice growth season (June–October) and 7° and 23 °C during wheat season (November–April), respectively. The daily maximum temperature during May and June frequently exceeds 43 °C (Kaur and Hundal, 2008).

### 2.2. Size fractionation of soil organic matter

The samples were air-dried and passed through 2 mm sieve. The plant residues retained on the sieve were discarded. Each soil sample was divided into two parts. One portion of the sample was separated into three SOM fractions viz. cPOM, fPOM and MinOM and the other portion was kept as whole soil for subsequent incubation studies. Particulate organic matter (POM) was separated by dispersing soil in 0.5% sodium hexametaphosphate solution and shaking for 15 h on a reciprocal shaker (Cambardella and Elliott, 1992; Benbi et al., 2012). The dispersed soil sample was passed through a set of 250 and 53  $\mu\text{m}$  sieves. The material retained on the 250  $\mu\text{m}$  sieve consisted of cPOM and coarse sand (250–2000  $\mu\text{m}$  size). The material retained on the 53  $\mu\text{m}$  sieve comprised fPOM and fine sand (53–250  $\mu\text{m}$  size). After rinsing several times with water, the material retained on the sieves was dried at 40 °C in a

hot-air oven. The slurry that passed through 53  $\mu\text{m}$  sieve comprising silt and clay particles and MinOM was centrifuged and solution was decanted and the mineral matter was dried at 40 °C.

Total C (TC) in the whole soil samples and isolated size fractions was determined by dry combustion using CHN analyzer (Elementar model Vario EL III). Inorganic C in the samples, determined by HCl method (Jackson, 1967), was absent; therefore total organic C (TOC) was similar to TC.

### 2.3. Carbon mineralization

Carbon mineralization of isolated SOM fractions and whole soil drawn from the four treatments in the field was studied by conducting aerobic incubations in the laboratory under controlled conditions. Whole soil and physical fractions of SOM ranging from 5 to 50 g, depending on availability of material, were taken in 250 ml conical flasks along with vials containing 0.1 M NaOH to trap evolved  $\text{CO}_2$  and water to maintain humidity. Each sample was moistened to 50 per cent of water holding capacity and incubated for 54 days at four different temperatures viz. 15, 25, 35 and 45 °C. Alkali traps were replaced daily during the first week and every 2nd or 3rd day thereafter depending on the amount of  $\text{CO}_2$  evolved. The quantity of  $\text{CO}_2\text{--C}$  evolved was determined by titrating alkali in the traps with 0.1 M HCl using phenolphthalein as indicator. Temperature of 45 °C was kept as the soils in the study area are frequently exposed to such a high temperature during summer.

Microbial biomass carbon (MBC) was determined at the end of incubation at day 54 following the Chloroform Fumigation Extraction (CFE) method (Vance et al., 1987) using a recovery factor ( $K_{\text{EC}}$ ) of 0.41. The results on MBC have been presented as Supplementary information.

### 2.4. Modeling C mineralization kinetics

First-order single compartment model (Benbi and Richter, 2002) was fitted to the cumulative C mineralization ( $C_t$ ,  $\text{mg kg}^{-1}$ ) Vs time ( $t$ , days) data (Equation (1)) for each size fraction and whole soil.

$$C_t = C_0 e^{-kt} \quad (1)$$

Where  $C_0$  is the C mineralization potential ( $\text{mg kg}^{-1}$ ) and  $k$  is the mineralization rate coefficient ( $\text{d}^{-1}$ ).  $C_0$  was fixed equal to the quantity of TOC in a given sample at the start of incubation. Therefore,  $k$  was the only parameter estimated. For the sake of clarity, the rate coefficients estimated for the C mineralization data averaged across field treatments are presented.

### 2.5. Temperature sensitivity of C mineralization

The influence of temperature on mineralization of isolated SOM fractions and whole soil was evaluated by fitting mineralization rate coefficients estimated at different temperatures to five different models viz. the  $Q_{10}$  function, van't Hoff's equation (van't Hoff, 1898), Arrhenius equation (Arrhenius, 1889), the Lloyd and Taylor (1994) equation, and the Gaussian equation (O'Connell, 1990). The description of the models is given in Appendix A.

### 2.6. Statistical analyses

The first-order kinetic model and the temperature response models were fitted by non-linear least squares technique using SPSS software. The differences in means for C concentration and stocks and mineralization rate coefficients for SOM fractions and

**Table 1**  
Carbon concentration and stocks in various soil organic matter fractions in differently fertilized soils. Numbers in parenthesis indicate standard error.

Soil/Treatment <sup>a</sup>	Fraction weight ( $\text{g kg}^{-1}$ soil)			C Concentration ( $\text{g kg}^{-1}$ fraction)			C Stocks ( $\text{g kg}^{-1}$ soil)		
	cPOM	fPOM	MinOM	cPOM	fPOM	MinOM	cPOM	fPOM	MinOM
RS	48.7 ( $\pm 0.7$ )	592.1 ( $\pm 5.3$ )	347.4 ( $\pm 7.4$ )	19.9 ( $\pm 0.9$ )	5.5 ( $\pm 0.35$ )	11.8 ( $\pm 0.71$ )	0.97 ( $\pm 0.04$ )	3.27 ( $\pm 0.20$ )	4.10 ( $\pm 0.26$ )
IN	45.7 ( $\pm 1.7$ )	590.7 ( $\pm 9.0$ )	366.3 ( $\pm 13.1$ )	9.2 ( $\pm 0.3$ )	2.2 ( $\pm 0.03$ )	8.9 ( $\pm 0.15$ )	0.47 ( $\pm 0.01$ )	1.30 ( $\pm 0.03$ )	3.25 ( $\pm 0.12$ )
FYM	40.9 ( $\pm 0.4$ )	627.1 ( $\pm 10.4$ )	327.0 ( $\pm 0.6$ )	36.3 ( $\pm 6.8$ )	7.7 ( $\pm 0.75$ )	15.9 ( $\pm 0.69$ )	1.48 ( $\pm 0.26$ )	4.83 ( $\pm 0.50$ )	5.21 ( $\pm 0.22$ )
CONT	41.7 ( $\pm 1.2$ )	625.2 ( $\pm 6.9$ )	332.5 ( $\pm 5.3$ )	9.2 ( $\pm 0.8$ )	2.7 ( $\pm 0.13$ )	14.0 ( $\pm 0.11$ )	0.38 ( $\pm 0.03$ )	1.71 ( $\pm 0.08$ )	4.66 ( $\pm 0.04$ )
Mean	44.3 ( $\pm 1.1$ )	609.0 ( $\pm 7.7$ )	343.5 ( $\pm 5.7$ )	18.6 ( $\pm 3.7$ )	4.5 ( $\pm 0.70$ )	12.6 ( $\pm 0.80$ )	0.82 ( $\pm 0.15$ )	2.78 ( $\pm 0.45$ )	4.35 ( $\pm 0.23$ )
LSD	3.6	26.6	26.0	11.2	1.4	1.6	0.43	0.88	0.58
						3.0			0.30

<sup>a</sup> The treatment acronyms RS, IN, FYM and CONT, respectively represent annual input of rice straw at 10  $\text{Mg ha}^{-1}$  along with fertilizer NPK (RS), application of fertilizer NPK only (IN), annual input of FYM at 40  $\text{Mg ha}^{-1}$  (FYM), and no input of organic and inorganic sources (IN).

whole soil were tested for significance by analysis of variance (ANOVA) technique in completely randomized design using locally developed software (Cheema and Singh, 1990). The least significant difference (LSD) values were tested at 5% ( $P < 0.05$ ) level of probability.

### 3. Results

#### 3.1. Physical fractions of soil organic matter

Coarse POM (cPOM) comprised the smallest proportion of whole soil and its quantity ranged between 40.9 and 48.7 g kg<sup>-1</sup> soil under different field treatments (Table 1). The fPOM, which ranged between 590.7 and 627.1 g kg<sup>-1</sup> soil comprised the largest proportion of whole soil. The field treatments resulted in a wide range of C concentrations and C stocks in different SOM fractions. The C concentration of the three SOM fractions ranged (g kg<sup>-1</sup> fraction) between 9.2 and 36.3 for cPOM, 2.7 and 7.7 for fPOM, and 8.9 and 15.9 for MinOM. The C concentration of all the SOM fractions was significantly ( $P < 0.05$ ) higher in FYM-amended soils than the other treatments. The fPOM and MinOM fractions in RS amended soils were characterized by significantly ( $P < 0.05$ ) higher C concentration than the IN and CONT soils. Averaged across treatments cPOM exhibited the highest C concentration (18.6 g kg<sup>-1</sup>) and fPOM had the minimum C concentration (4.5 g kg<sup>-1</sup>); the MinOM was in-between. Similar to C concentration, the C stocks (g kg<sup>-1</sup> soil) in the three SOM fractions were highest under FYM treatment. Irrespective of the treatment the greatest C stocks occurred as MinOC and the least as cPOC. Under all the treatments the C/N ratio was greater for cPOM than the other two fractions and followed the order (except for CONT treatment) cPOM > fPOM > MinOM (Table 2). The cPOM and fPOM in RS and FYM amended soils generally exhibited higher C/N ratios than the IN and CONT treatments.

#### 3.2. Carbon mineralization

Temporal trends of CO<sub>2</sub>-C respired from the SOM fractions and the whole soil, averaged across field treatments, showed that the C mineralization was faster during initial days of incubation and decreased gradually as the incubation progressed (Fig. 1). Carbon mineralization generally plateaued towards the end of incubation. Irrespective of temperature, the greatest quantity of C was mineralized from cPOM and the least from MinOM. The amount of cPOC mineralized at different temperatures ranged between 1674 and 8337 mg C kg<sup>-1</sup>, which was almost 4.4 to 5.4 times the quantity mineralized from fPOC, 4.8 to 8.0 times from MinOC and 5.6 to 9.1 times from whole soil C. The proportion of total C mineralized from isolated SOM fractions and the whole soil increased with increase

in temperature; the increase being greater between 15 and 35 °C compared with 35–45 °C. At the end of incubation 10–49 percent of cPOC and 10–41 per cent of fPOC were mineralized at different temperatures compared with 3–8 percent of MinOC and 4–12 percent of the whole soil C.

At all the temperatures, the first-order rate coefficients for C mineralization followed the order cPOM ≥ fPOM > whole soil > MinOM (Fig. 2). The disparity in rate coefficients between cPOM and the other two SOM fractions and the whole soil widened with increase in temperature from 15 to 45 °C. While at 15 and 25 °C, the rate coefficients for cPOM and fPOM were similar but at 35 and 45 °C the rate coefficient for cPOM was 1.3–1.4 times that for fPOM. Similarly, the ratio of rate coefficients for cPOM to MinOM increased from 3.1 at 15 °C to 6.3 at 45 °C. Compared with whole soil, the mineralization rate coefficient for cPOM was higher by 2.3 and 5.1 times at 15 and 45 °C, respectively. The MinOM had the lowest rates of C mineralization and these were even lower than the whole soil (Fig. 2). Estimates of mean residence time (MRT) showed that cPOM was most decomposable with MRT ranging between 490 days at 15 °C and 81 days at 45 °C. The MinOM was least decomposable with MRT's of 1534 and 508 days at 15 °C and 45 °C, respectively (Table 3).

#### 3.3. Temperature dependence of C mineralization

Parameter estimates for various models fitted to mineralization rates as a function of temperature are presented in Table 4. All the four models provided a good fit to the temperature response data as shown by high  $R^2$  between the modeled and the measured respiration rates. The  $Q_{10}$  values estimated using  $Q_{10}$  function were generally highest at 15 °C, except for the cPOM, and decreased linearly with increase in temperature (Fig. 3). However, for cPOM the  $Q_{10}$  was slightly higher at 25 °C (2.13) than at 15 °C (1.99). At all the temperatures, MinOM showed the lowest temperatures sensitivity with  $Q_{10}$  ranging between 1.45 and 1.60 at different temperatures. van't Hoff's model also yielded highest temperature sensitivity for cPOM ( $Q_{10} = 1.87$ ) followed by fPOM ( $Q_{10} = 1.67$ ), MinOM ( $Q_{10} = 1.45$ ) and the least for whole soil ( $Q_{10} = 1.40$ ). For the Arrhenius model the  $Q_{10}$  at 15 °C ranged between 1.95 for cPOM and 1.48 and 1.44 for MinOM and whole soil, respectively. The  $Q_{10}$  gradually declined to 1.8 for cPOM and 1.4 for MinOM and whole soil as the temperature increased to 45 °C. The Lloyd and Taylor (1994) model predicted relatively higher  $Q_{10}$  for cPOM (2.2–1.8) and fPOM (2.0–1.7) than the Arrhenius model especially at temperatures below 25 °C. The model also showed that the temperature sensitivity of MinOM was slightly higher than the whole soil. The Gaussian model provided the highest  $Q_{10}$  (2.9) for cPOM followed by fPOM (2.4) at 15 °C, which decreased with increase in temperature. However, the reduction in temperature response with increase in temperature was steeper for Gaussian model than the other models (Fig. 3).

We used Arrhenius, the Lloyd and Taylor and the Gaussian models to estimate the effect of unit degree increase in temperature on C mineralization of isolated SOM fractions and whole soil. The analysis showed that one degree Celsius increase in temperature enhanced decomposition of isolated SOM fractions and whole soil to a greater extent at low temperatures and the effect diminished with increase in temperature. All the three models showed that the decomposition of cPOM fraction is likely to be influenced to the greatest extent (7–15 per cent increase) per °C rise in temperature between 10 and 15 °C followed by fPOM (6–12% increase). The MinOM is likely to be least affected with about 4–6 per cent increase in its decomposition per °C increase in temperature between 10 and 15 °C. Carbon mineralization of whole soil will increase by

**Table 2**

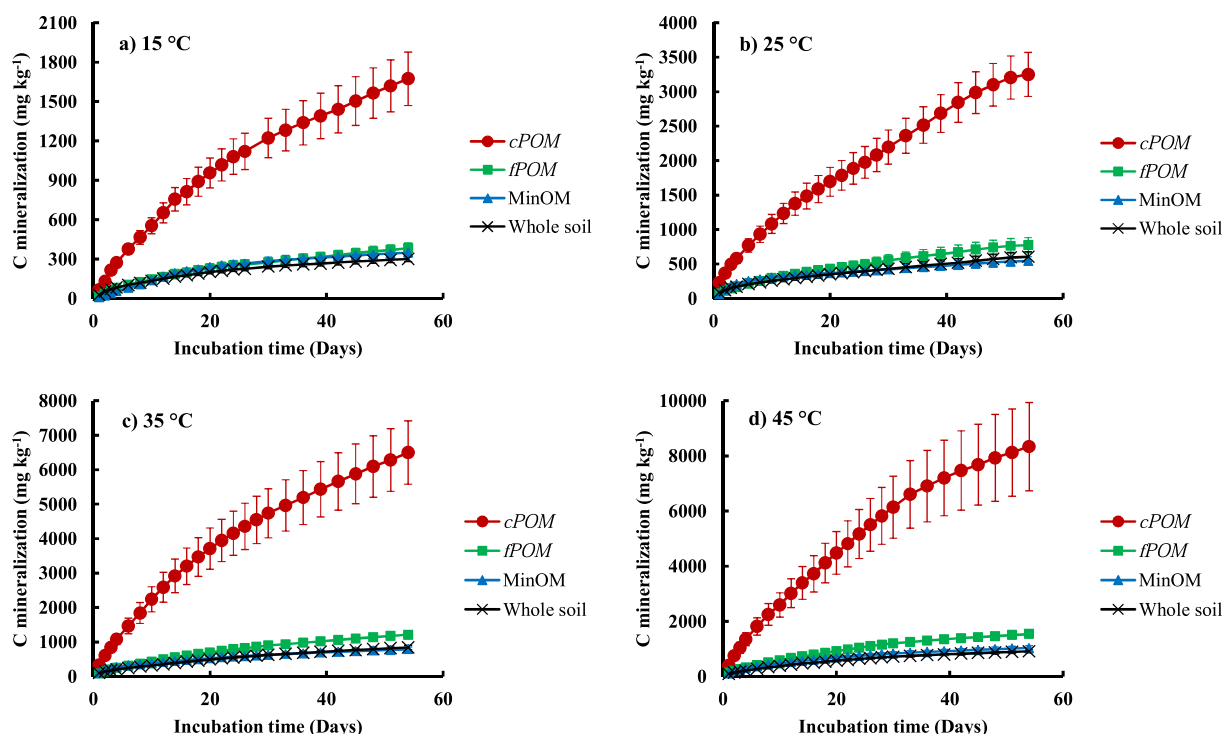
C:N ratios of soil organic matter fractions in differentially fertilized soils. Numbers in parenthesis indicate standard error.

Soil/Treatment <sup>a</sup>	cPOM	fPOM	MinOM	Whole soil
RS	15.5 (±0.4)	10.6 (±0.7)	6.6 (±0.2)	11.3 (±0.6)
IN	11.5 (±0.6)	7.5 (±0.1)	6.7 (±0.1)	9.5 (±0.4)
FYM	13.4 (±0.9)	9.3 (±0.5)	7.6 (±0.4)	10.9 (±0.3)
CONT	10.5 (±0.8)	7.2 (±0.4)	7.5 (±0.1)	11.3 (±0.5)
Mean	12.7 (±0.64)	8.7 (±0.47)	7.1 (±0.18)	10.8 (±0.30)
LSD	2.2	1.6	0.8	NS

NS: non-significant.

<sup>a</sup> The treatment acronyms RS, IN, FYM and CONT respectively represent annual input of rice straw at 10 Mg ha<sup>-1</sup> along with fertilizer NPK (RS), application of fertilizer NPK only (IN), annual input of FYM at 40 Mg ha<sup>-1</sup> (FYM), and no input of organic and inorganic sources (IN).





**Fig. 1.** Temporal trends of C mineralization from coarse particulate organic matter (cPOM), fine particulate organic matter (fPOM), mineral associated organic matter (MinOM), and whole soil at 15, 25, 35 and 45 °C. Values averaged across field treatments for different fractions and whole soil. Line bars indicate standard error.

4–9 per cent at 10–15 °C temperature and the effect will diminish to about 2–3 percent at 35 °C.

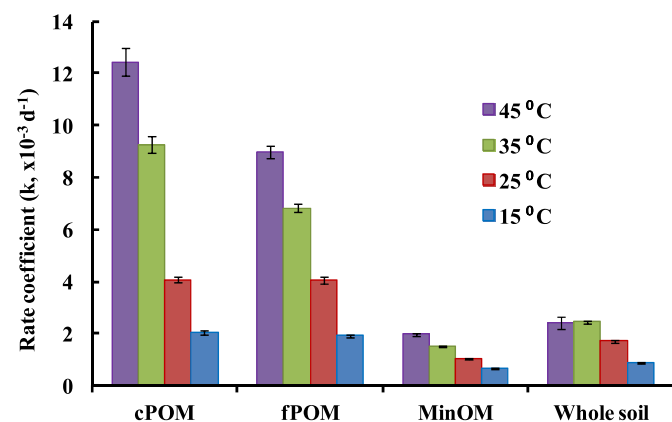
## 4. Discussion

### 4.1. Physical fractions and carbon mineralization

The results on C concentrations and C stocks showed that the soils included in the study represented a range of values comparable to those reported from the experimental area with similar agricultural management (Benbi et al., 2012). Addition of organic amendments generally resulted in higher C stocks in isolated SOM fractions and whole soil compared to unamended soils. Similarly, the differences in C/N ratio were more obvious in soils receiving

organic amendments in the field. The relatively wider C/N ratio of the cPOM than the fPOM and MinOM fractions suggests that the crop mediated and added organic C is first transferred to cPOM, which on decomposition, is progressively stabilized into fPOM and silt and clay- sized MinOM with narrowing down of C/N ratio. These results are similar to those reported by Bird et al. (2002) and Benbi et al. (2012). Using  $^{14}\text{C}$  isotope analysis, Bird et al. (2002) showed that  $^{14}\text{C}$  activities decrease with decreasing particle size, indicating increasing residence times of C in the finer fractions. Benbi et al. (2012) reported that the C/N ratio of SOM fractions decreased in the order cPOM > fPOM > MinOM suggesting that cPOM was more decomposable than the fPOM and MinOM.

The occurrence of MinOC as the largest pool (~53%) and cPOC as the smallest pool (~10%) of SOC showed that the majority of SOC was in stable form and less decomposable. Our estimates of MRT also confirmed that the cPOM was short-lived and was influenced by temperature to a greater extent as opposed to MinOM that was long-lived and was least affected by temperature. Earlier studies have also shown that turnover time of coarse fraction POM was lower than those for mineral associated organic matter (Garten and Wulfschleger, 2000). Using  $^{13}\text{C}$  natural abundance technique, Balesdent (1996) concluded that POM has a short mean residence



**Fig. 2.** Mineralization rate coefficients ( $\text{d}^{-1}$ ) for soil organic matter fractions and whole soil at different temperatures. Values averaged across field treatments for different fractions and whole soil. Line bars indicate standard error.

**Table 3**

Mean residence time (MRT) for soil organic matter fractions and whole soil, averaged across field treatments, at different temperatures.

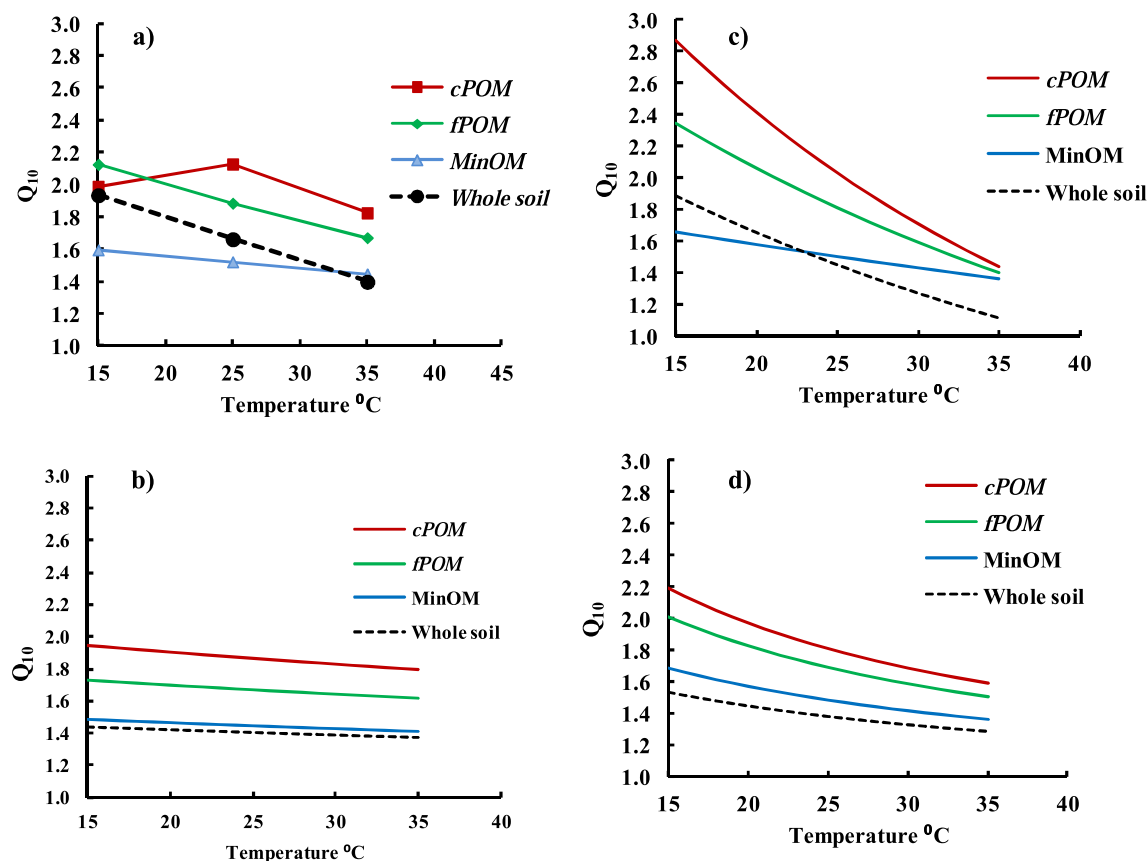
Organic matter fraction	MRT (Days)				
	15 °C	25 °C	35 °C	45 °C	LSD (0.05)
cPOM	490	246	108	81	182
fPOM	521	245	147	112	95
MinOM	1534	962	662	508	84
Whole soil	1131	585	410	415	155
LSD (0.05)	229	112	66	264	

**Table 4**  
Parameters estimates for temperature response models fitted to C mineralization rates of soil organic matter fractions and whole soil averaged across field treatments.  $R^2$  indicates goodness of fit between measured mineralization rates and those estimated by temperature response models.

Fraction	van't Hoff		Arrhenius		Llyod & Taylor			Gaussian			
	$b (\times 10^{-2})$	$R^2$	$E_a$	$R^2$	$A$	$E_0$	$R^2$	$a (\times 10^{-4})$	$b$	$c (\times 10^{-3})$	$R^2$
cPOM	6.24	0.935	47.7	0.947	0.628	340.6	0.975	1.88	0.175	−1.73	0.993
fPOM	5.13	0.947	39.3	0.958	0.362	303.5	0.984	3.63	0.137	−1.30	0.998
MinOM	3.69	0.984	28.2	0.990	0.026	227.6	0.999	2.50	0.0702	−0.49	0.999
Whole soil	3.36	0.763	26.0	0.786	0.023	186.1	0.856	2.16	0.116	−1.31	0.970

time relative to C associated with clay- and silt- sized organo-mineral complexes, indicating the relatively high lability of POM. Commensurate with the lability of SOM fractions, the greatest quantity of C was mineralized from cPOM followed by fPOM and the least from MinOM, which increased with rise in temperature; the increase being greater between 10 and 35 °C compared with 35–45 °C. The growing inequality in mineralization rates between labile and stable pools with increase in temperature suggest labile pools of SOM had greater temperature sensitivity than the stable pool and the whole soil. The difference in mineralization rates between cPOM and MinOM more than doubled as the temperature increased from 15° to 45 °C. Our results on increased disparity in mineralization rates and MRTs between cPOC, fPOC and MinOC with increase in incubation temperature validate our hypothesis that labile pools of SOC were more sensitive to temperature than the recalcitrant pool of C. The greater decomposition temperature sensitivity of POM, which represents uncomplexed and unprotected pool of SOM consisting of plant material in various stages of decomposition along with microbial biomass and microbial debris

may be attributed to its easy availability to microorganisms and rapid decomposability. As the dead plant material resides for longer time in the soil system, it experiences greater physico-chemical transformation and is stabilized in aggregates and bonded onto the surface of minerals such as clay and silt (Six et al., 2002) and may even become biochemically recalcitrant with low bioavailability and long turnover times (Krull and Skjemstad, 2003). This conceptual framework of SOM stabilization supports our hypothesis that labile fractions with a shorter turnover time will respond more rapidly to temperature than stabilized recalcitrant fractions. However, characterization of recalcitrant C is still debatable. Kleber et al. (2011) showed that C age is not necessarily related to molecular structure or thermodynamic stability. They (Kleber et al., 2011) showed that passive pools of relatively old C had larger proportion of easily metabolizable organic molecules with low thermodynamic stability than the young C that had higher proportion of stable organic structures. It was suggested that SOM turnover rate may be viewed as codetermined by the interaction between substrates, microorganisms, and abiotic variables. Conant



**Fig. 3.** Decomposition temperature response ( $Q_{10}$ ) of different SOM fractions and whole soil estimated from a)  $Q_{10}$  model, b) Arrhenius model, c) Gaussian model, and d) Llyod and Taylor model.

et al. (2011) defined resistance of SOM to decomposition as being due either to its chemical conformation or its physico-chemical protection, both of which determine the temperature sensitivity of decomposition. This supports our contention that physical fractionation of SOM reflects both physico-chemical protection and chemical conformation and thus could provide a better insight into the decomposition temperature sensitivity of SOM. In addition to SOM characteristics, enzyme kinetics, and potential for microbial adaptation are critical for predicting temperature sensitivity of organic matter decomposition (Billings and Ballantyne, 2013).

#### 4.2. Predicting temperature dependence of C mineralization

We used four commonly used temperature response models to upscale our results and to enumerate the impact of warming on SOM decomposition (see Appendix A). While all the four models provided good fit to the C mineralization data, the shape of the temperature response curve for different models varied considerably. van't Hoff's model has the limitation that it assumes constant temperature sensitivity over a temperature range. The use of the other three models showed that the temperature sensitivity of C mineralization declined with increase in temperature, though the estimates of  $Q_{10}$  and magnitude of decline (in  $Q_{10}$  values) differed with the choice of the model. While there was a small linear decrease with Arrhenius model, the Lloyd and Taylor and the Gaussian models showed relatively large curvilinear decline. For the whole soil, the Arrhenius model yielded almost constant decomposition  $Q_{10}$  (1.44–1.38) between 15 and 35 °C whereas the  $Q_{10}$  estimated by the Lloyd and Taylor and the Gaussian models declined from 1.54 to 1.29 and 1.89 to 1.12, respectively at 15 °C and 35 °C. In the literature, a considerable range of decomposition  $Q_{10}$  values (2–4.5 between 5 and 35 °C), employing different models have been reported, with the greatest relative sensitivity having been observed at low temperatures (Kirschbaum, 1995; Kätterer et al., 1998). However, the results from different studies that employed different models may not be directly comparable as the estimates of decomposition  $Q_{10}$  vary with the choice of the model.

The use of Arrhenius, the Lloyd and Taylor and the Gaussian models for upscaling the results in context to global warming showed that one degree Celsius increase in temperature will enhance decomposition of isolated SOM fractions and whole soil to a greater extent at low temperatures and the effect will diminish with increase in temperature. The Gaussian model showed that the decomposition temperature response of mineralization of isolated SOM fractions mainly differed at temperatures below 25 °C beyond which  $Q_{10}$  tended to converge to a value of about 1.4 at 35 °C for isolated SOM fractions. This suggests that the impact of temperature on SOC decomposition will be primarily limited to temperatures below 25 °C beyond which the effect will be small. Therefore, warming will enhance decomposition of isolated SOM fractions and whole soil to a greater extent at low temperatures and the effect will diminish with increase in temperature. The decomposition of labile pools will be influenced to the greatest extent as opposed to stable pool that will be least affected due to warming. Similar to estimates of  $Q_{10}$ , the models differed considerably in predicting relative increase in decomposition. While Arrhenius model predicted the smallest relative increase in the decomposition of isolated SOM fractions (e.g. 7.4–7.1% increase for cPOC between 10 and 15 °C) as a result of warming, the Gaussian model showed the largest increase (14.8–12.9% for cPOC between 10 and 15 °C). The modeling analysis showed that C mineralization of whole soil will increase by 4–9 per cent at 10–15 °C temperature and the effect will diminish to about 2–3 percent at 35 °C. Kirschbaum (1995) using Gaussian model suggested that a 1 °C increase in temperature could lead to a loss of over 10 per cent of SOC in regions of the

world with an annual mean temperature of 5 °C whereas the same temperature increase would lead to a loss of only 3 per cent of SOC for a soil at 30 °C. Similar to our results, Kirschbaum (1995) reported that loss of SOC per degree warming will decrease with increasing temperature. However, our results with the use of different models show that the choice of a model impacts the predicted response of SOM decomposition to warming. In the literature, one of these four models has, generally, been advocated to describe temperature dependence of organic matter decomposition. However, the evaluation of models has generally been limited to goodness of fit to the experimental results and robustness of statistical estimates (Lloyd and Taylor, 1994; Leirós et al., 1999; Fang and Moncrieff, 2001; Tuomi et al., 2008) without emphasis on the differences in the estimated temperature response. It is important to investigate into the reasons for differential behavior of the models and identify conditions under which a given model may be applied for upscaling experimental results.

The results of our study have shown that the labile pools of SOC viz. cPOC and fPOC were more sensitive to temperature than the stable MinOC, especially at temperatures below 25 °C. The mineralization of cPOC exhibited the highest temperature sensitivity. This is a convenient finding as the cPOC comprises a very small fraction of total SOC and is short-lived. In the present study, cPOC accounted for only 10 percent of SOC with MRT ranging between 81 and 490 days at different temperatures. Davidson and Janssens (2006) contended that if temperature sensitivity of a soil is attributable mainly to the labile SOM pool, then the impact on CO<sub>2</sub> evolution will be relatively small and short-lived. Conversely, if temperature sensitivity is attributable mainly to the stable SOM pool then the response in CO<sub>2</sub> evolution will be large and long-lived. Our results are in contrast to those reported by Knorr et al. (2005) and Conant et al. (2008a), who reported that the decomposition of stable or slow pools of C are more sensitive to temperature than the labile pools causing a potentially larger positive feedback in response to global warming. However, Reichstein et al. (2005) and Fang et al. (2005) argued that it is premature to conclude that stable soil carbon is more sensitive to temperature than labile carbon and the decomposition of both the pools may show similar response. The conflicting results from different studies may partially be attributed to the differences in methodology employed to estimate decomposition temperature sensitivity, and the inability to precisely define and quantify labile and stable SOM (Kirschbaum, 2006; von Lützow and Kögel-Knabner, 2009). Conant et al. (2008b) distinguished the response of labile and recalcitrant soil C to increased temperature by incubating soils for varying durations to deplete the samples of labile soil C. The assumption was that the changed lability is the dominant factor driving changes in the respiration rate under constant temperature and moisture conditions. Plante et al. (2010) studied temperature sensitivity of SOC decomposability by assessing responses of soil fractions of different lability isolated by acid hydrolysis and concluded that POM had lower  $Q_{10}$  than the whole soil. But these studies did not characterize well-defined fractions of SOM rather relied on ease of decomposition (either through incubation or acid hydrolysis) to distinguish labile and recalcitrant pools. During incubation of soils to deplete labile soil C, the evolved CO<sub>2</sub> could arise from various pools of SOM rather than a specified pool leading to confounding of results. Recently, it has been shown that, in short-term incubations, it is the integrated effect of different SOM pools rather than any individual fraction that contributes towards C mineralization (Benbi et al., 2012). Acid hydrolysis, which indirectly relies on molecular structure, may not specifically distinguish SOM fractions of different lability as the C turnover in soils is only weakly controlled by the molecular structure (Marschner et al., 2008). For example, turnover of lignin or certain biochar, which were supposed to be stable, was similar to

other organic compounds (Gleixner et al., 1999; Heim and Schmidt, 2007). Conversely, physical fractionation according to size yields well-defined pools that reflect both physico-chemical protection and chemical conformation. The incubation of physically separated SOM pools in isolation can provide better insight into the decomposition temperature sensitivity and mechanism of C turnover in soils. It may be argued that the incubation of isolated fractions is not representative because the soil matrix environment has been destroyed, but the approach provides estimates closer to intrinsic temperature sensitivity of different fractions rather than apparent temperature sensitivity (Davidson and Janssens, 2006). However, intrinsic temperature sensitivity of decomposition can be fully explored only through understanding the phenomena like SOM protection, varying substrate availability and diffusion constraints (Billings and Ballantyne, 2013). Further, the protection of organic matter from decomposition depends on the physical nature of the soil mineral fraction, especially the presence of surfaces capable of adsorbing organic materials and the arrangement of pores and soil particles (Baldock and Skjemstad, 2000). Skene et al. (1996) showed that over a period of 170 days, a decrease in the decomposition of straw occurred with the inclusion of a mineral matrix such as sand, kaolinite or a loamy sand. However, the mineral matrix will probably have less effect on decomposition of POM as by definition it represents uncomplexed and unprotected pool of SOM. Physical fractionation coupled with direct assessment of molecular structure has been proposed as an effective approach for interpreting C quality–temperature relationship for short-term decomposition (Wagai et al., 2013).

Similar to several published studies our results also confirm that the temperature sensitivity of C mineralization was greater at low temperatures than the high temperature. However, most of the studies available in the literature on temperature dependence of SOM are limited to maximum 30–35 °C temperature. In semi-arid tropical/sub-tropical regions where temperature in summer often exceeds 40 °C it is imperative to study the temperature dependence of organic matter at temperature greater than 40 °C. One of the limitations of our study could be that it has been conducted over short-time. However, incubations over extended periods may lead to exhaustion of more readily decomposable material, which would be most pronounced under the most favorable conditions. This could reduce decomposition rates and also lead to underestimation of its temperature dependence (Kirschbaum, 2006). In our study the cumulative amount of C mineralized had attained a plateau by the end of incubation and the per day rate of C mineralization for a given SOM fraction was almost constant at a specified temperature. In longer-term incubations there is also the confounding factor of substrate supply (Gu et al., 2004). When conditions are favorable for organic matter decomposition, the faster decomposition rate also depletes the pool of labile substrate, thus reducing the subsequent rate of organic matter decomposition.

It may be mentioned that under our short-term laboratory incubation microbial thermal acclimation and community compositional shift probably did not occur, which may occur under long-term warming (Bradford et al., 2008). Our incubation experiments at constant moisture without simultaneous changes in temperature and moisture did not mimic the field conditions. In the field soil moisture and temperature generally change simultaneously. Studies in the past have documented an interaction between soil moisture and temperature in influencing organic matter decomposition (Curtin et al., 2012). Therefore, further studies are needed to characterize the interactive effect of temperature and moisture on decomposition of SOM fractions. The initially high rates of decomposition observed in our study may be due to drying and rewetting of the SOM fractions and whole soil. The SOM fractions after isolation were dried and rewetted prior to initiation of

incubation. Drying of soils causes changes in soluble organic matter due to killing of microbial biomass, which on rewetting is rapidly mineralized leading to initially high rates of C mineralization (Benbi and Richter, 2002). We separated organic matter fractions by dispersing with sodium hexametaphosphate, which may influence the composition of isolated SOM fraction and being an antimicrobial agent could damage the cell membranes of bacteria (Fukao et al., 2000). Though it is a commonly used chemical dispersant (Gregorich and Beare, 2007), studies enumerating its effect on SOM composition and soil solution chemistry are lacking. Agents other than sodium hexametaphosphate such as sonication and shaking with glass beads have been used to disperse the soil. The potential problem associated with the use of ultrasonic dispersion is the redistribution of organic matter among size/density fractions; and it is reported to seriously affect the amount and properties of POM fractions (Elliott and Cambardella, 1991; Oorts et al., 2005). Further, the sonication energy has to be standardized for each soil type to minimize redistribution among organic matter fractions (Cerli et al., 2012). Shaking without any dispersing agent will probably cause fewer artefacts. However, shaking speed will have to be optimized to achieve realistic separation of SOM fractions without breaking the soil structure.

## 5. Conclusions

The cPOC, representing labile pool of C comprised the smallest pool of SOC and the MinOC representing stable pool accounted for more than half of the bulk SOC. The cPOM was most decomposable and the MinOM was least decomposable. The labile pools of SOM (cPOM and fPOM) were more sensitive to temperature than the stable pool (MinOM). The temperature response of mineralization of isolated SOM fractions mainly differed at temperatures below 25 °C beyond which the response tended to converge suggesting thereby that the differential response of labile and stable pools to temperature will be foremost at temperatures below 25 °C beyond which the effect will be small and similar for SOM pools of different lability. Warming will influence decomposition of cPOM to the greatest extent and the MinOM the least. Because cPOC comprises a small fraction of bulk SOC and is short-lived its high temperature sensitivity will have little impact on CO<sub>2</sub> evolution. Choice of a temperature response model could influence upscaling of experimental results and predicted response to warming emphasizing the need to employ appropriate model for simulating C fluxes.

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## Appendix A

Description of temperature response models fitted to experimental results

### i) The $Q_{10}$ function

The temperature dependence of soil respiration, commonly referred to as the  $Q_{10}$  value, has been the focus of many studies. The  $Q_{10}$  for a reaction rate is defined as the factor by which the rate increases with a 10 °C rise in temperature. For the first-order exponential equation, which assumes  $Q_{10}$  is constant over the temperature, the  $Q_{10}$  value can be calculated as:



$$Q_{10} = (k_2/k_1)^{10/(T_2-T_1)}$$

Where  $k_2$  and  $k_1$  are respiration rates observed at temperatures  $T_2$  and  $T_1$ .

ii) van't Hoff's equation: assumes an exponential relationship between the rate of the process and the temperature

$$k = ae^{bT}$$

$$Q_{10} = e^{10b}$$

Where,  $k$  is the  $\text{CO}_2$  evolution rate ( $\text{d}^{-1}$ );  $a$  and  $b$  are fitted parameters.  $Q_{10}$  is the temperature coefficient that represents the relative growth of respiration as the temperature increases by  $10^\circ\text{C}$ .

iii) The Arrhenius equation: assumes the activation energy for the process to be constant

$$k = Ae^{-E_a/RT}$$

where,  $k$  is the reaction rate constant,  $A$  is a frequency or pre-exponential factor that represents theoretical reaction rate in the absence of activation energy,  $E_a$  is interpreted as the required activation energy,  $R$  is gas constant ( $8.314 \text{ J K}^{-1} \text{ mol}^{-1}$ ) and  $T$  is temperature in Kelvin.

iv) The Lloyd and Taylor model: assumes the energy of activation to vary as the reciprocal of temperature

$$k = ae^{-E_0/(T-T_0)}$$

where,  $a$  is an overall rate term;  $E_0$  here does not denote the activation energy as in Arrhenius Equation;  $T$  is temperature in Kelvin;  $T_0$  is some temperature that is being used as a reference temperature and it must be less than any measurement temperature. Lloyd and Taylor (1994) used this model with  $E_0 = 308.6$  and  $T_0 = 227.1 \text{ K}$

v) The Gaussian model: O'Connel (1990) presented an equation to describe the temperature dependence of biological processes like soil respiration or mineralization rates.

$$k = ae^{bT+cT_2}$$

where,  $a$  represents absolute rate of the process and  $b$  and  $c$  represent its temperature dependence. All three are fitted parameters.

## Appendix B. Supplementary data

Supplementary data related to this article can be found at <http://dx.doi.org/10.1016/j.soilbio.2013.12.032>.

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