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Physical disturbance accelerates carbon loss through increasing labile carbon release

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Abstract: Labile carbon (C) is a major source of C loss because of its high vulnerability to environmental change. Yet its potential role in regulating soil organic carbon (SOC) dynamics remains unclear. In this study, we tested the effect of physical disturbance on SOC decomposition using soils from two abandoned farmlands free of management practice for more than 28 years. The soil respiration rate was measured in undisturbed and disturbed soil columns and was inversely modeled using the two-compartment model. We found that the C loss was 16.8~74.1% higher in disturbed than in undisturbed soil columns. Physical disturbance increased the total amount of labile C (C_1) loss by 136~241%, while had no effect on the kinetic decomposition rate constants of both labile (k_1) and stable (k_2) SOC decomposition. Physical disturbance fragmented the large macroaggregates into small macroaggregates, microaggregates, and free silt and clay-sized fractions. This indicates that C loss was derived from the initially protected labile C, and there was no change of SOC fraction being decomposed. Our results give insights into the understanding of the extent of labile C loss to physical disruption and demonstrate the potential effect of physical disturbance on SOC dynamics.

Keywords: carbon model; organic carbon decomposition; physical protection; soil incubation; soil organic matter

Labile soil organic carbon (SOC) accounts for less than 5% of the total SOC (Davidson and Janssens 2006, Zakharova et al. 2015, Godde et al. 2016), but is vulnerable to microbial degradation during physical disturbance due to loss of physical protection by soil aggregates or lack of adsorption by clay minerals (Krull et al. 2003). However, the issue of the physical disturbance on carbon (C) loss remains controversial (Zakharova et al. 2014, Tian et al. 2015).

In most studies, soil organic matter (SOM) decomposition was increased by physical disruption during the initial few weeks of incubation (Gregorich et al. 1989, Hassink 1992). However, the response of SOM to physical disruption is also much dependent on soil types (Hassink 1992, Drury et al. 2004, Tian et al. 2015). A possible cause is that SOM fractions associated with different sized particles (sand, silt, and clay) often have different structures and functions (Christensen 2001, Luo et al. 2017). The electrostatic adsorption of SOM by clay is regarded as the most efficient interaction in protecting SOM from being decomposed during aggregate disruption, which will not disappear after aggregate disruption (Sollins et al. 1996). In addition, the distribution of particle size is concerned with the formation of the organomineral complexes and the soil aggregation, and ultimately determines the stability of SOC (Sollins et al. 1996).

However, few studies have assessed the potential effect of physical disturbance on labile and stable C pools (Zakharova et al. 2014, 2015). In previous studies, the incubated soils were subjected to physical disturbance by crushing, sieving, and air-drying before incubation (Tian et al. 2015, Zakharova et al. 2015, Somasundaram et al. 2018), which may under-

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estimate the effect of physical disturbance on SOC dynamics due to the damage of initially protected labile C before incubation. A method with minimal disturbance to the original soils is necessary to accurately test the effect of physical disturbance on SOC dynamics. Therefore, we conducted an incubation experiment on undisturbed and disturbed soil columns and compared the soil respiration in these two soil treatments to investigate the potential effect of physical disturbance on soil C loss.

MATERIAL AND METHODS

Sampling sites. Soil samples were collected from two abandoned farmlands free of management practices for more than 28 years. One was taken from a meadow mixed with *Phragmites australis* (Cav.) Steud., at Shenyang Agricultural Experimental Station (41°32'N, 122°23'E), Liaoning province, Northeast China. The soil was sandy loam and classified as an Aquic Alfisol developed from silty sediments. The other soil was obtained from a mesic meadow dominated by graminoids at Hailun Agricultural Experimental Station (47°26'N, 126°38'E) located near Harbin, Heilongjiang province, Northeast China. The soil was clay loam and classified as an Aquic Mollisol with a deep A-horizon and predominantly Montmorillonite clay.

Soils from Shenyang station contained 1.66% C, 0.14% N, and a gravimetric C/N ratio of 12.08. Soils from Hailun station contained 3.56% C, 0.27% N, and a gravimetric C/N ratio of 13.45. The clay content in Alfisol and Mollisol soil was 12.61% and 35.19%, respectively.

Soil sampling and incubation. In each of the two plots, we collected soils from five randomly chosen sites. Within each site, paired sub-samples were collected with minimal disturbance using a soil core sampler embedded with a lucite pipe (20 cm in length and 5.2 cm in diameter). For each pair of sub-samples, one sub-sample was subjected to the disturbance treatment, and the other one was served as the undisturbed control. In each undisturbed control soil core, plant litter and growing plants were carefully removed using long handle tweezers after sampling. After that, these undisturbed soil core was immediately placed in a polypropylene column and stored at 4 °C before incubation. For the disturbance treatment, plant litter and growing plants were also removed in the same way as in the undisturbed control treatment. After then, each soil core was broken up into small pieces manually and made them to pass through a 2-mm sieve, thoroughly homogenised and air-dried. Then each soil sample was placed into a polypropylene column and stored at 4 °C before incubation.

The moisture content in each column was maintained at 60% of the water holding capacity. Soil columns were incubated in processor-controlled incubators (Shellab LI20-2, Sheldon Manufacturing Inc., Cornelius, USA, with a temperature control accuracy and evenness of ± 0.02 °C) at 20 °C for 128 days. Anaerobic condition in each column was maintained *via* an automatic timer-controlled aeration system by providing fresh air for 1-h every 6-h during the entire incubation experiment.

Soil respiration. Respiration rate was measured after 3, 8, 14, 28, 45, 60, 75, 90, and 128 days of incubation using a CO_2 trapping method (Zhang et al. 2017) with the trapping efficiency of 99% (Lin et al. 2015). The CO_2 was trapped using 12 mL of 0.5 mol/L NaOH solution during an entire 24-h period. Empty columns were processed as blanks. The concentration of accumulated CO_2 was analysed using multi N/C® 2000 (Analytik Jena, Jena, Germany). The concentration of CO_2 was corrected for the initially present CO_2 in NaOH using the values from empty columns.

Inverse modeling of C pools using respiration data. The cumulative respired C of each soil sample was expressed as mg C/g initial C and was fitted individually to the following model:

$$C_{cum}(t) = C_1 \times (1 - e^{-k_1 \times t}) + C_2 \times (1 - e^{-k_2 \times t})$$

where: C_{cum} – cumulative C mineralised until time t (mg C/g initial C); C_1 – proportion of the most labile C pool (mg C/g initial C); C_2 – larger more stable C pool; k_1 and k_2 – first-order kinetic decomposition rate constants (per day) of the labile and stable SOC fractions, respectively; t – incubation time (day). In order to fit the model to the data, we made the assumption that the total initial C comprises C_1 and C_2, which was equal to 1 000 mg/g initial C (i.e., labile and stable C pools add up to the total amount of initial C in the soil) (Rey and Jarvis 2006).

Soil aggregate distribution. Soil aggregate size separation in disturbed and undisturbed soil treatments was performed before the incubation using the wet sieving method (An et al. 2013) with some modifications. Briefly, 100 g soil in each column was submerged in deionised water in a device assembled by three sieves differing in mesh size, i.e., a 2 mm, a 0.25-mm, and a 0.053-mm sieve in the order from top to bottom. After slaking for 5 min, the sieves were moved up and down for 50 times in 2 min before the wet soil samples in each sieve were collected in

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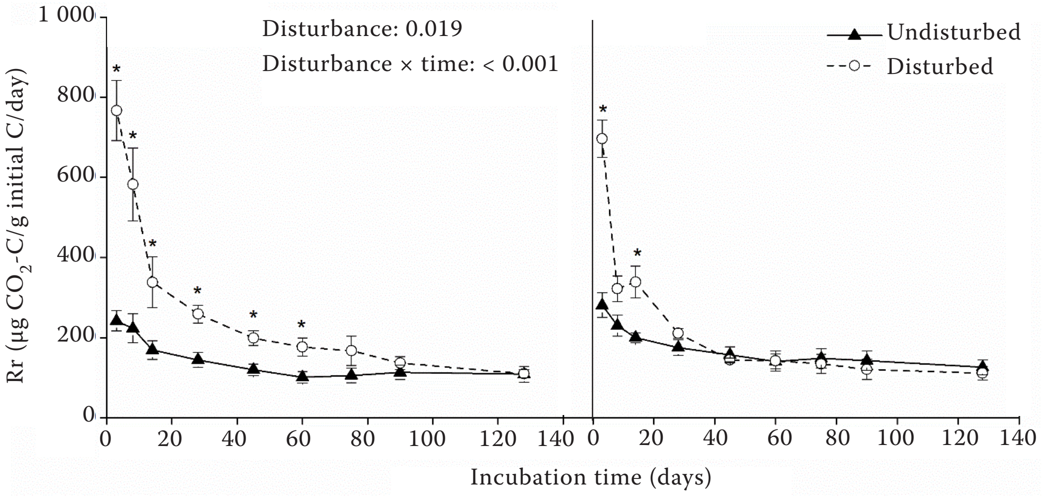

Figure 1. Dynamics of soil respiration (Rr) in undisturbed and disturbed soils from Shenyang (left) and Hailun experimental station (right). Error bars show ± standard error. *Significant at the 0.05 probability level

each respective pre-weighed salver (> 2 mm large macroaggregate, 0.25~2 mm small macroaggregate, 0.053~0.25 mm microaggregates, and < 0.053 mm free silt + clay-sized fraction). The soils in each salver were dried at 60 °C and weighted to determine the proportion in each size fraction.

Calculation and statistical analysis. We used repeated-measures ANOVA to test for the effect of physical disturbance on soil respiration. We used ANOVA to test for the effect of physical disturbance for each sampling time separately. One-way ANOVA was used to assess the effect of physical disturbance on soil aggregates distribution. The two-compartment model was used for the inverse modeling of CO_2 efflux from soil respiration. All curves of the dynamics of cumulative CO_2 efflux were fitted using nonlinear procedures using Origin 8.5 (Originlab Origin, Northampton, USA). We used a two-tailed *t*-test to test if the difference between observed and expected values of C_1, $k_{1,}$ and k_2 significantly deviated from zero. We used a two-tailed independent-samples *t*-test to determine the effect of disturbance on C_1, k_1, and k_2. Statistical analyses were performed using SPSS Statistics 20 (IBM SPSS Statistics, Armonk, USA). Differences were considered significant when $P < 0.05$. Origin 8.5 was used to create the figures.

RESULTS

Soil respiration. Soil respiration rate was significantly increased by physical disturbance ($P = 0.019$,

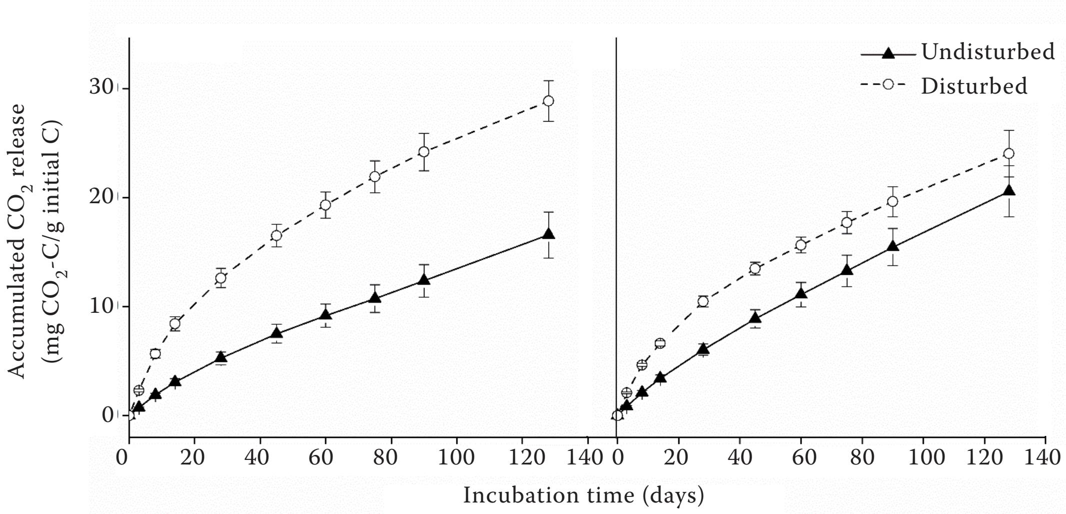

Figure 2. Cumulative CO_2 efflux from soil organic carbon decomposition in undisturbed and disturbed soils from Shenyang (left) and Hailun experimental station (right). Error bars show ± standard error

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Table 1. Values of the parameters obtained for the fitted carbon mineralisation model to cumulative carbon mineralisation data for the undisturbed (Un-Dis) and disturbed (Dis) treatment

Treatment	C_1				k_1				k_2 ($\times 10^{-5}$)			
	mean ± SE	*df*	*t*-value	*P*	mean ± SE	*df*	*t*-value	*P*	mean ± SE	*df*	*t*-value	*P*
Shenyang												
Un-Dis	3.342 ± 1.048	4	3.19	0.033	0.080 ± 0.021	4	3.82	0.019	10.408 ± 1.817	4	5.73	0.005
Dis	11.404 ± 1.114	4	10.24	0.001	0.060 ± 0.005	4	11.00	< 0.001	14.168 ± 1.066	4	13.30	< 0.001
Hailun												
Un-Dis	3.642 ± 0.845	4	4.31	0.013	0.054 ± 0.015	4	3.64	0.022	13.466 ± 2.034	4	6.62	0.003
Dis	8.607 ± 1.044	4	8.24	0.001	0.069 ± 0.008	4	8.81	0.001	12.342 ± 2.283	4	5.41	0.006

C_1 – proportion of the most labile C pool (mg C/g initial C); k_1 and k_2 – first-order kinetic decomposition rate constants (per day) of the labile and stable SOC fractions, respectively; SE – standard error

Figure 1). In disturbed soil treatment, the soil respiration declined sharply during the initial 28 days of incubation (Figure 1). During the earlier stage of incubation, the soil respiration rate was significantly higher in the disturbed than in the undisturbed soils. At the end of the incubation, the C loss in the undisturbed soils was 1.66% and 2.06% of the total SOC in Shenyang and Hailun, respectively (Figure 2). During the whole period, the C loss was increased by 74.1% and 16.83% in soils from Shenyang and Hailun, respectively (Figure 2).

Inverse modeling of soil respiration. The soil respiration rates fitted well with the two-pool C model (all $R^2 > 0.999$, $P < 0.001$). The proportion of labile C ranged from 0.33% to 1.14% in these soils (Table 1). Physical disturbance significantly increased the proportion of labile C in the total SOC in both two soil sites (Figure 3A). Compared to the undisturbed

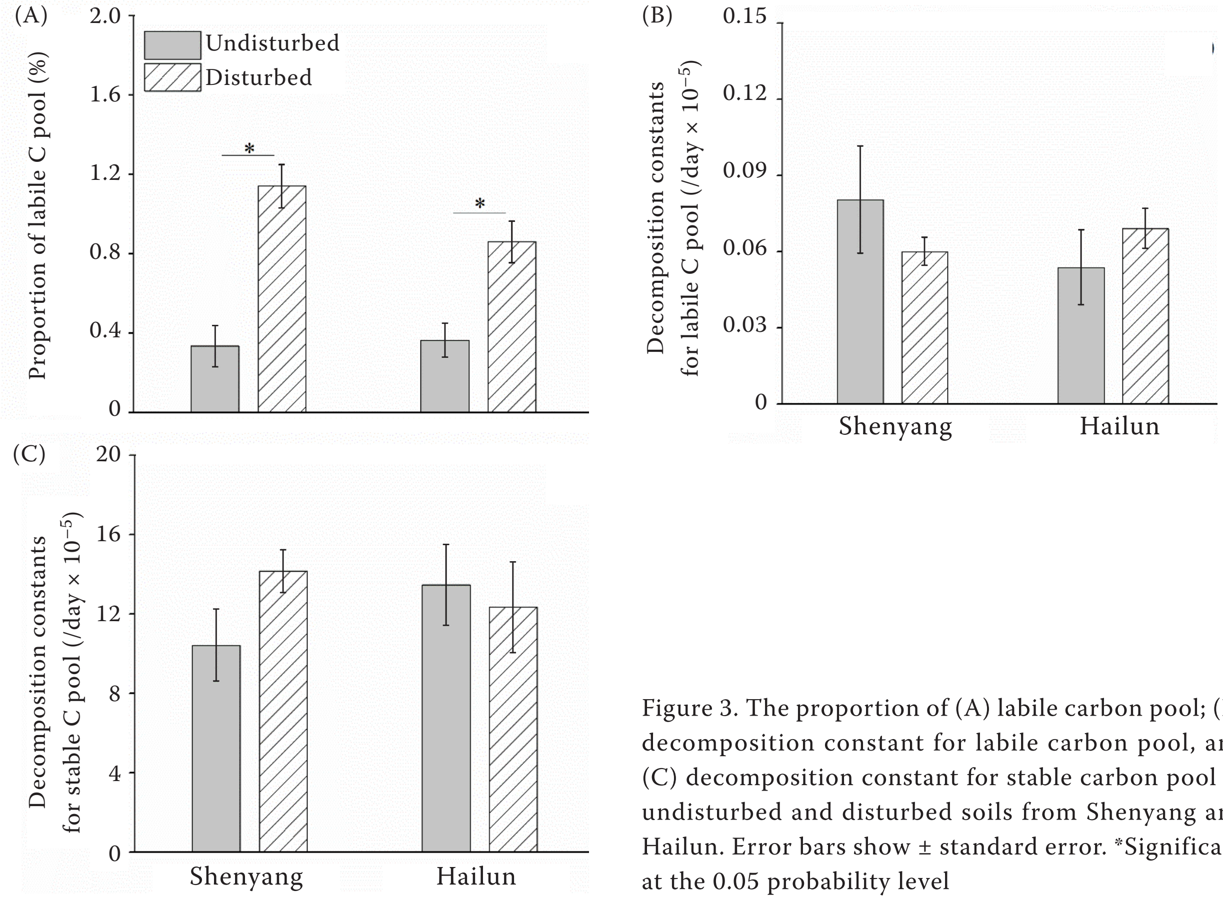

Figure 3. The proportion of (A) labile carbon pool; (B) decomposition constant for labile carbon pool, and (C) decomposition constant for stable carbon pool in undisturbed and disturbed soils from Shenyang and Hailun. Error bars show ± standard error. *Significant at the 0.05 probability level

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soils, the proportion of labile C in the disturbed soils was increased by 241.2% and 136.4% in soils from Shenyang and Hailun, respectively (Figure 3A). However, the turnover rate of labile SOC and stabilised SOC was not effected by physical disturbance (Figure 3B, C).

Changes of soil aggregates after disturbance. The distribution of soil aggregates was greatly changed by physical disturbance (Figure 4). At the end of incubation, the proportion of the large macroaggregate in the disturbed soils was 71–92% lower than that in the undisturbed soils. Conversely, the proportions of the small macroaggregate, microaggregate, and free silt and clay-sized fraction were higher in the disturbed soils than in the undisturbed soils. The changes in aggregate size distribution due to disturbance were similar in soils from Shenyang and Hailun.

DISCUSSION

The acceleration of SOC decomposition during the initial days of incubation as a result of physical disturbance was consistent with previous studies (Hassink 1992, Zakharova et al. 2015). And the difference of C loss in soils from Shenyang and Hailun may be related to the soil properties and cover plants. The results of the two-pool C model showed that disturbance significantly increased the proportion of labile C in total SOC (Figure 3A), which indicates the vulnerability of labile C to physical disturbance (Davidson and Janssens 2006). This is because, in well-structured soil, SOM is stabilised within aggregates and intimate association with soil minerals (Six and Paustian 2014, Lehmann and Kleber 2015). Physical disruption broke the large macroaggregates into smaller aggregates (Figure 4), and some of the initially protected soil C becomes available to soil organisms and thus degraded rapidly after a significant disruption of soil aggregates.

Physical disturbance increased the proportion of labile C by 136% to 241% (Table 1), while the C decomposition rate (k_1) in the disturbed treatment remained almost unchanged, indicating no change of SOM fraction being decomposed in consideration of the varying inherent decomposition rate of different components of SOM. Moreover, despite the differences of soil structure between soils from Hailun (classified as clay loam soil) and Shenyang (classified as sandy loam soil) (Figure 4), the change of labile C loss by physical disturbance was similar in these two soils. This is because although the disturbance processes, including soil core breaking, sieving, and air-drying, disrupted the large macroaggregates fragmented into smaller aggregates, most of the soil remained structured (Figure 4). The C mineralised following disturbance was mostly derived from sand-size associated C fractions occluded within the large macroaggregates (Gregorich et al. 1989). While the interactions between SOM and clay minerals will not disappear after aggregate disruption (Christensen 2001, Kleber et al. 2015).

Moreover, the large macroaggregate is the most unstable structure compared with other smaller aggregates (Six and Paustian 2014). In our study, the change of the large macro-aggregate was similar in sandy loam and clay soil, which indicated the generality of the effect of physical disturbance on labile C loss. In most cultivated and continuous cropping soils, aggregate disruption often has no evident effect on extra CO_2 production. This may be ascribed

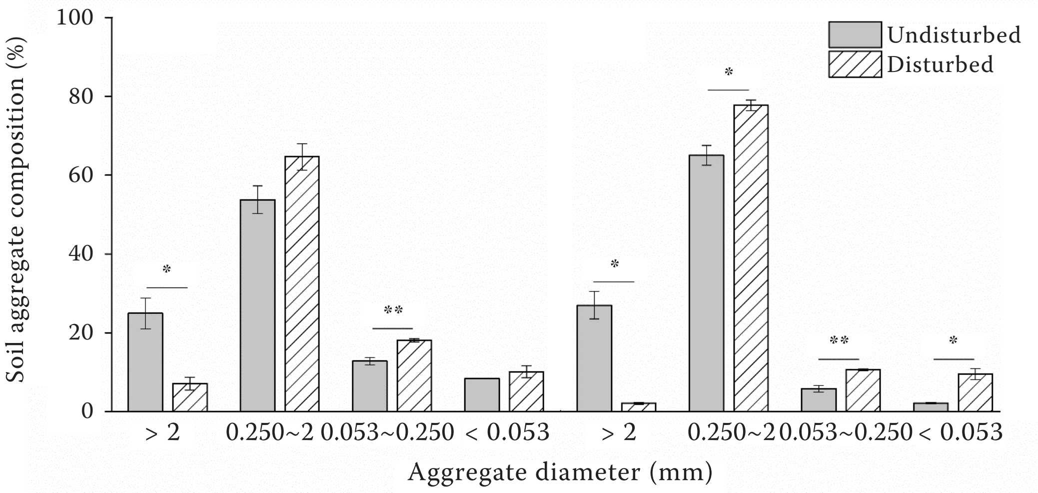

Figure 4. Soil aggregate composition in undisturbed and disturbed soils from Shenyang (left) and Hailun (right). Error bars show ± standard error

to the exhaustion of the easily decomposable SOC due to the constant disruption of the most unstable soil structure before incubation (Drury et al. 2004, Tian et al. 2015). In our study, we used soils from two abandoned farmlands free of management practices for more than 28 years and also used undisturbed soil cores as the control, which helps to eliminate the confounding effect of different management practices and incubation methods on labile C release.

In conclusion, physical disturbance increased labile C by 1.36- to 2.41-fold compared to the undisturbed soils, indicating a large potential effect on SOC dynamics. Furthermore, the C model showed the change of C pool contributions to total SOC and the decomposition rate related to each C pool, which provides a new approach to better predict the C loss following environmental change or management practice.

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