Research Paper

Effect of microplastics on organic matter decomposition in paddy soil amended with crop residues and labile C: A three-source-partitioning study

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ABSTRACT

Microplastics (MPs) are a widespread pollutant in terrestrial ecosystems. However, knowledge on how MPs impact soil organic matter (SOM) decomposition and the priming effect (PE) in rice paddy soil remains limited. By employing a three-source-partitioning approach, we investigated the interactive impact of MP dosage (none, low [0.01% w/w] or high [1% w/w]), labile C (^{14}C-labeled glucose), and ^{13}C-labeled rice straw addition on SOM decomposition and PE. Compared to soil without C addition (i.e., control), total SOM-derived CO_2 in low-MP soil declined by 13.2% and 7.1% after straw and glucose addition, respectively. Under combined glucose and rice straw addition, glucose-induced PE was up to 10 times stronger in the presence of low-MPs compared to that in high-MPs. However, glucose induced negative PE on rice straw decomposition in the presence of MPs. SOM decomposition was much higher under low MP dosage than under high MP dosage. However, MPs had a negligible effect on the mineralization of exogenous C substrate (glucose or straw). This study provides a novel and valuable insight on how MPs affect SOM turnover and C sequestration in paddy soil, highlighting the significance of interactions between environmental pollutants and biogeochemical processes that affect CO_2 fluxes.

1. Introduction

Plastic pollution, also called "white pollution", is a pervasive and growing problem worldwide (Liu et al., 2014). Even with immediate and concerted action, 710 million tons of plastic waste is predicted to enter the environment over the next 20 years (Lau et al., 2020). Gradually, plastic debris breaks down into smaller plastic fragments or particles <5 mm, typically referred to as microplastics (MPs), which are a widespread pollutant in terrestrial ecosystems (Qi et al., 2020; Rillig and Lehmann, 2020; Zhou et al., 2020). Yet, plastic film mulching, which primarily made from polyethylene, is being widely adopted in agricultural ecosystems, due to its benefits for crop production; consequently, it has become a major source of MPs and, therefore, soil pollutants (Huang et al., 2020; Kim et al., 2021).

Although plastic is resistant to degradation, it can fragment into smaller particles through tillage and exposure to UV radiation, which are then retained in the soil for a long period (Liu et al., 2014; Rachman,

2018). In addition to plastic film mulching, biosolids employed as fertilizers are another source of MP addition to agricultural soils. According to some sources, approximately 44,000–430,000 tons of MPs have been estimated to enter the agroecosystem annually through biosolids in North America and Europe (Ng et al., 2018). These MPs also contain large amounts of C within their molecular structure; however, this is unlikely to perform similar eco-functional services as soil organic matter (SOM) (Rillig, 2018). Currently, it remains unclear how the addition of MPs to the agricultural system impacts SOM decomposition, particularly when different organic substrates are added to soil during crop growth and cultivation (e.g., labile C through rhizodeposition, litter input from previous crops, or straw being returned to the soil).

Indeed, the mechanisms related to the impact of MP pollutants on microbial SOM decomposition processes are rarely studied. For example, MPs under straw residue addition have been shown to reduce soil dissolved organic C (DOC) content (Chen et al., 2020); however, it remains unclear whether MPs inhibit the release of DOC from SOM or

accelerate the decomposition of pre-existing DOC. A previous study reported that polyvinyl chloride MPs in soil promote the carbon use efficiency (CUE) of labile substrates (e.g., glucose and amino acids in DOC), indicating their relevance for C sequestration (Zang et al., 2020). MPs might also induce the formation of microbial biofilms, which represent a new microbial niche termed the "plastisphere", which impacts microbiota and microbial function (Miao et al., 2019). This microbial niche constitutes hotspots of microbial activity, and results in the heterogeneity of the biogeochemical cycling in soil (Awet et al., 2018; Hope et al., 2020). However, it is unclear whether, and to what extent, MPs induce changes to abiotic and biotic soil conditions that affect SOM decomposition.

Polyethylene is a high molecular organic polymer that contains almost 90% C, and is potentially a source of DOC in soil (Rillig, 2018; Zhu et al., 2020). Thus, polyethylene addition might affect the mineralization of native SOM through a phenomenon that is usually referred to as the "priming effect (PE)" (Kuzyakov, 2010). However, previous research revealed that the proportion of DOC released from polyethylene MPs is 0.11–0.48% of initial plastic C, of which only 22–46% is considered biolabile substrate (Zhu et al., 2020). Only small amounts of biolabile DOC are released from polyethylene MPs, which is unlikely to induce a real PE. However, MP addition might induce apparent PE that relates to the changes in microbial biomass turnover without impacting native SOM decomposition (Blagodatskaya et al., 2007; Blagodatskaya and Kuzyakov, 2008). MP addition alters various soil properties, such as improving the size of soil water stable aggregates, soil porosity (aeration), and water saturation capacity (de Souza Machado et al., 2019). Changes to these soil properties could alter microbial communities and their function, thus affecting SOM decomposition (Six et al., 2004; Gao et al., 2021; Han et al., 2021). However, paddy soils (flooded anaerobic) have different characteristics compared with upland soils, including soil structure, porosity, and water saturation. Thus, the effect of MPs on soil properties, microbial communities, and functions might be different (or less relevant) in paddy soils compared to upland soils (Li et al., 2021a, 2021b).

SOM decomposition is controlled by several factors, such as the type of organic substrate added and the interdependence of various SOM components (Shahbaz et al., 2018b, 2018a). Freshly added organic components, such as those from rhizodeposits (labile substrates) and crop residues, could modulate the decomposition of existing SOM, thus inducing PE on SOM decomposition (Zhu et al., 2018a; Zhu et al., 2018b). However, the mechanisms responsible for controlling the impact of MPs on PE in the presence of other substrates remain elusive. Furthermore, it is unclear how interactions between the decomposition processes of labile substrates and crop residues impact the direction and intensity of PE during SOM decomposition under MP pollution. In the present study, three doses of MPs (none, 0.01% w/w, and 1% w/w) were added to soil in combination with labile C (^{14}C-labeled glucose) and rice straw residues (^{13}C-labeled). The objectives were to: 1) examine how MPs affect the decomposition of SOM, added glucose (as labile substrate), and rice straw; and 2) investigate the combined impact of glucose and rice residues on SOM priming under MP addition. We hypothesized that if MPs negatively impacted microbial growth and activity, the presence of MPs in soil would reduce the decomposition of SOM, glucose, and rice residues.

2. Materials and methods

2.1. Soil preparation

Plow layer soil (0–20 cm) was collected from a subtropical rice paddy (113°20′8″E, 28°33′26″N, 113 m above sea level) located in Hunan, China. The soil was classified as typical Stagnic Anthrosol. Soil was air-dried and sieved to < 2 mm. Air-dried soil had a pH of 6.4 (1:2.5, soil to water ratio), organic C (OC) content of 16.8 g C kg^{-1}, atom percentage of ^{13}C of 1.078%, total N (TN) content of 1.10 g N kg^{-1}, and total

phosphorus (TP) content of 0.50 g P kg^{-1}. It was composed of 12% silt, 82% clay, and 6% sand.

2.2. Rice straw

Rice plants were subjected to continuous ^{13}CO$_2$ (2.8 atom% of ^{13}C value) labeling to produce ^{13}C-labeled straw. Twenty-five day old rice seedlings were transplanted to pots in an air-tight control chamber. Rice plants were labeled for the entire vegetative stage, and then grown until maturity. At the ripening stage when plants were harvested, rice shoots were cut at the base of the plant and collected, and were subsequently dried at 60 °C for 48 h. For homogeneous mixing, shoot residues were chopped and sieved to < 2 mm. The C content in shoots was 48.6% (C/N: 25.6). The atom percentage of ^{13}C values for rice shoots was 2.268%.

2.3. Experimental layout and soil incubation

A dry equivalent of 60 g of 2 weeks of pre-incubated soil (water content, 34.3%) was thoroughly mixed with MPs and ^{13}C-straw, and then transferred to a 500 mL incubation bottle. The ratio of added deionized water to soil was 2:1 (w:v). A 2–3 cm water layer was maintained above the soil surface throughout incubation to keep the soil in a flooded condition. Three MP treatments (polyethylene; 40–48 μm; ^{13}C atom % = 1.073%; Sigma-Aldrich, St. Louis, USA; Fig. S1) were established; specifically, M0: no MP addition; M1: 100 mg [0.01% w/w] MPs kg^{-1} dry soil [low MPs]; and M2: 10,000 mg [1% w/w] MPs kg^{-1} dry soil [high MPs]. Each MP treatment had four sub-treatments (soil without substrate addition, soil + glucose, soil + straw, and soil + straw + glucose). Each treatment had three replicates, resulting in 36 experimental units. Rice straw was amended at a dose of 2.5 g kg^{-1} soil (1.215 mg C g^{-1} soil), corresponding to a rice straw returning rate ca. 6 t ha^{-1}. ^{14}C-labeled glucose solution (419 μg C g^{-1}, ^{13}C atom % = 1.074%, corresponding to 2.5% SOM in the control soil) was amended in aliquots every day during the first 30 days of incubation to simulate the continuous release of root exudates under field conditions. Soils without glucose addition were amended with an equal amount of distilled water as a reference to estimate the PE.

2.4. Sampling

After introducing distilled water or glucose to the soils, small vials containing 5 mL of 1 M NaOH were placed in the incubation bottles to trap CO$_2$ emissions (including three control bottles without soil). The bottles were immediately sealed so that they were air-tight and incubated at 25 °C for 100 days. Fresh NaOH vials were used to replace the previous vials at 1, 3, 5, 7, 10, 15, 20, 25, 30, 35, 42, 52, 65, 83, and 100 days of incubation. Samples of NaOH were used to analyze ^{14}C, ^{13}C, and the total amount of trapped CO$_2$. CO$_2$ that was trapped in the control bottles without soil was used to correct the total CO$_2$ flux. Other sets of replicates were prepared for soil sampling after 3, 15, 30, 65, and 100 days of incubation. Soil samples were carefully mixed to determine microbial biomass C (MBC).

2.5. Microbial biomass

Soil MBC was measured using the chloroform fumigation-K$_2$SO$_4$ extraction method (Wu et al., 1990). Extracts were analyzed using a total organic carbon/total inorganic carbon (TOC/TIC) analyzer (Multi N/C 2100, Analytik Jena, Germany) to determine C concentrations. Extracts were freeze-dried for ^{13}C analysis.

2.6. Isotopic and chemical analyses

To quantify CO$_2$ flux, 1 mL of CO$_2$-trapping NaOH solution was used, with 0.1 M HCl used to titrate off excess NaOH after precipitation with 1 M BaCl$_2$ solution. Phenolphthalein was used as an indicator (Zibilske,

1994).

For ^{13}C determination, 2 mL of CO_2-trapping NaOH solution was precipitated with 1 M $SrCl_2$ solution. The mixed solutions were shaken thoroughly, and were then centrifuged at 4000 rpm for 5 min. The precipitates were washed repeatedly with distilled water to remove excess NaOH, until the pH of the washing water reached 7. $SrCO_3$ precipitates were then dried at 60 °C and stored for ^{13}C analysis.

All samples were ground and passed through a 0.147 mm sieve before ^{13}C analyses. ^{13}C analysis of rice shoots that were prepared to add to the soil, freeze-dried extracts of MBC and $SrCO_3$ precipitates were analyzed using an isotope ratio mass spectrometer (IRMS) (MAT253, Thermo-Fisher Scientific, Waltham, MA, USA) coupled with an elemental analyzer (EA) (FLASH 2000, Thermo-Fisher Scientific, Waltham, MA, USA). The oxidation column of the EA system was filled with Cr_2O_3/Co_3O_4 as the oxidant. The temperature of the oven and combustion tube was 50 and 980 ℃, respectively. He was used as a carrier gas with a flow rate of 100 mL min^{-1}. L-glutamic acid (δ^{13}C VPDB = −26.39‰; USGS, Reston, VA, USA) was used as the isotopic reference material. The isotope ratio was expressed as parts per thousand (‰), relative to the international standard Vienna Peedee belemnite (VPDB) using delta notation (δ^{13}C).

^{14}C activity in CO_2 was analyzed immediately after collecting CO_2-trapping NaOH solution. A 1 mL aliquot of NaOH containing trapped $^{14}CO_2$ was analyzed by a Wallac 1404 liquid scintillation counter (Wallac EG&G, Milton Keynes, UK) after mixing it with 3 mL Scintisafe 3 scintillation cocktail (Fisher Scientific, Loughborough, UK). A 5 mL aliquot of K_2SO_4 extract (fumigated and non-fumigated) with 15 mL of scintillation cocktail was also measured through scintillation counting for ^{14}C activity, to calculate glucose-derived C in MBC.

OC and TN of the soil used in the present study were determined by dry combustion with an elemental analyzer (Vario EL III, Elementar, Germany). Soil TP was determined based on NaOH fusion, analyzed at 880 nm by molybdate colorimetry on a spectrophotometer (UV2300II, Techcomp Ltd, China). Soil texture was assessed by a laser diffraction particle size analyzer (Mastersizer 2000, Malvern Instruments Ltd., Malvern, UK).

2.7. Calculations and statistics

Partitioning of the three C sources (straw (^{13}C), glucose (^{14}C), and SOM (^{12}C)) in the total CO_2 efflux and MBC were performed step-by-step, following the procedure of Shahbaz et al. (2018b). The amount of glucose-derived C (C_{G-d}, $\mu g\ g^{-1}$) in the pools (CO_2 and MBC) was calculated as:

$$C_{G-d} = C_G \times {}^{14}C_{curr}/{}^{14}C_G \qquad (1)$$

where $^{14}C_{curr}$ is current ^{14}C radioactivity (in DPM), $^{14}C_G$ is the total initial radioactivity of added glucose (in DPM), and C_G ($\mu g\ g^{-1}$) is the amount of the total applied glucose.

The amount of SOM-derived C (C_{SOM-d}) was then calculated as:

Soil without straw: $C_{SOM-d} = C_{total} - C_{G-d}$ (2)

Soil with straw: $C_{SOM-d} = C_{total} - C_{G-d} - C_{str-d}$ (3)

where C_{total} is the total amount of C in glucose-amended soils with or without straw addition in the corresponding pool, and C_{str-d} is straw-derived C, which was calculated from the atomic percentage values of ^{13}C (Shahbaz et al., 2018b) determined by IRMS (see Eq. (5)).

The fraction of C (f C) from straw in CO_2 and MBC was calculated as:

$$f\ C = [(At_{total} - At_{con}) / (At_{str} - At_{con})] \qquad (4)$$

where At_{total} is the atomic percentage value of ^{13}C (K_2SO_4 extracts and CO_2 trapped in NaOH) in straw-amended soil; At_{str} is the specific atomic percentage value of ^{13}C of straw used in the present study; and At_{con} is the atomic percentage value of ^{13}C in the control soil without straw

addition. The amount of straw-derived C (C_{str-d}) was calculated as:

$$C_{str-d} = f\ C \times [TC] \qquad (5)$$

where [TC] represents total C ($\mu g\ g^{-1}$) in the CO_2 and MBC pools in straw-amended treatments. Similarly, the amount of SOM-d C (C_{SOM-d}) was calculated by subtracting C_{str-d} and C_{G-d} (with straw and/or glucose amendment) from total C.

PE ($\mu g\ C\ g^{-1}$) was calculated according to Shahbaz et al. (2018b). The PE of the separate addition of glucose and straw on SOM decomposition was calculated as:

$$PE = C_{SOM-d,\ amended} - C_{SOM-d,\ con} \qquad (6)$$

where $C_{SOM-d,\ amended}$ is the amount of SOM-derived CO_2 ($\mu g\ C\ g^{-1}$ soil) from soil with exogenous substrate and $C_{SOM-d,\ con}$ is the amount of CO_2 from the control soil without exogenous substrate. PE in soil containing straw + glucose was calculated to examine the simultaneous effects of glucose on SOM and straw decomposition:

$$PE\ on\ straw = C_{str-d,\ str+G} - C_{str-d,\ str} \qquad (7)$$

$$PE\ on\ SOM = C_{SOM-d,\ str+G} - C_{SOM-d,\ str} \qquad (8)$$

where $C_{str-d,\ str+G}$ and $C_{str-d,\ str}$ represent the amount of straw-derived CO_2 ($\mu g\ C\ g^{-1}$) in the soil + straw + glucose and soil + straw treatments, respectively; and $C_{SOM-d,\ str+G}$ and $C_{SOM-d,\ str}$ represent the amount of SOM-derived C in the soil + straw + glucose and soil + straw treatments, respectively. The cumulative amount of PE and C efflux (derived from straw, SOM, or glucose) was calculated by summing the calculated values of all sampling points during incubation.

Data (PE and C fluxes) were analyzed by one-way ANOVA with Fisher's least significant difference (LSD) test to compare the means of variables between MP treatments, after checking for normality with a Shapiro–Wilk test and homogeneity of variance with the Levene's test. Data were log_{10} transformed to reduce heteroscedasticity, when necessary. The standard error of the PE and C flux values was calculated to estimate the precision of mean values. Statistical analyses were performed using SPSS 22 (IBM, Armonk, NY, USA).

3. Results and discussion

3.1. Effect of MPs on glucose-induced priming of SOM decomposition

Total soil CO_2 efflux ranged from $989 \pm 14\ \mu g\ C\ g^{-1}$ to $1663 \pm 5\ \mu g\ C\ g^{-1}$ (Fig. S2). To partition the contribution of different sources in total CO_2 efflux, glucose- and rice straw-derived CO_2 were calculated based on ^{14}C activity and ^{13}C values, respectively (Shahbaz et al., 2018a, 2018b). The cumulative glucose- and rice straw-derived CO_2 efflux was approximately 60–67% that of amended glucose C and approximately 33–35% that of amended straw C at the end of incubation (Fig. 1). SOM-derived CO_2 efflux ranged from $929 \pm 11\ \mu g\ C\ g^{-1}$ to $1019 \pm 11\ \mu g\ C\ g^{-1}$, which was calculated by subtracting glucose- or/and rice straw-derived CO_2 from the total CO_2 efflux (Fig. 2).

Negative PE during the early incubation period was observed in soil amended with glucose, i.e., before day 10, day 7, and day 15 for soils with no MPs, low MPs, and high MPs, respectively (Fig. 3a). The amount of total glucose used in this study (419 $\mu g\ C\ g^{-1}$ soil) corresponded to 164% that of original soil MBC (i.e., at the beginning of incubation). However, because we added an aliquot for 30 days, the daily added amount of glucose (i.e., 14 $\mu g\ C\ g^{-1}$ soil per day) was equivalent to 5.5% original soil MBC. This small amount of easily available substrate triggered microbial growth, and resulted in negative apparent PE (Blagodatskaya and Kuzyakov, 2008). With the continual addition of glucose, microorganisms might have had sufficient energy, but limited nutrients (e.g., N) (Wei et al., 2020). SOM becomes the main source of N acquisition for microbes in moderately labile C soil, with extracellular enzymes being released to decompose SOM to acquire necessary nutrients,
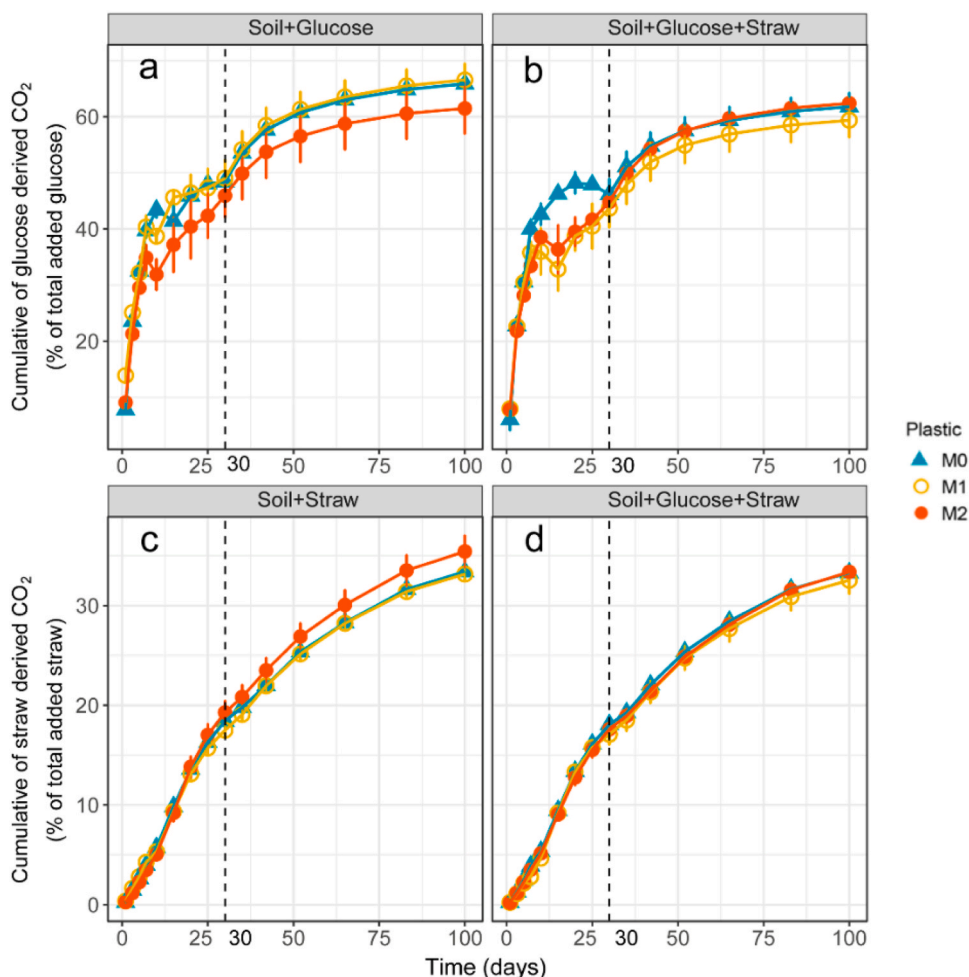
Fig. 1. Cumulative CO2 release from amended C sources in soils with and without microplastics. M0 = no microplastics; M1 = low (0.01%) microplastics; M2 = high (1%) microplastics. Error bars represent the standard error of the mean (n = 3).

inducing a positive PE after negative PE (Fontaine et al., 2003; Wei et al., 2020). The large amount of labile C input can cause the proliferation of fast-growing r-strategists (Blagodatskaya et al., 2007). However, a substantial number of r-strategist microorganisms died due to substrate exhaustion or severe N deficiency; thus, N-rich necromass might have become available to slowly-growing K-strategists (Ziegler et al., 2005; Cui et al., 2020). The decline in MBC after glucose addition eventually stopped, indicating the re-utilization of microbial necromass (from lysed microbial cell), may have partly released the requirement of labile C and nutrients from SOM (Shahbaz et al., 2017; Cui et al., 2020). This consequently reduced SOM decomposition and led to an intensive negative PE after day 42 of incubation in soil with low MPs (Fig. 3a). MBC increased over the first 30 days and subsequently sharply declined, indicating the microbial succession of mechanisms governing PE (Fig. 4b). However, SOM-derived MBC increased slightly after day 65, whereas glucose-derived MBC decreased (Fig. 4b), indicating that the decomposition of SOM facilitated microbial growth. Slowly-growing microorganisms (i.e., K-strategists) are more consistent in degrading complex SOM. After labile resources are exhausted, microbial succession from fast-growing to slow-growing microorganisms produced extracellular enzymes to decompose recalcitrant native SOM (Wei et al., 2020). This process likely explains the persistent and slow-growing MBC during the late stage of incubation in the present study (Fig. 4a–d).

The addition of MPs resulted in a cumulative negative PE of 76.5 ± 11.4 µg C g^{-1} induced by glucose at the end of incubation, which accounted for 6% of total CO$_2$ efflux; however, higher dose of MPs had little effect on PE after day 42 of incubation (Figs. 3a and 5). The surface

of MPs has different charges that attract and adsorb oppositely charged hydrophobic organic compounds and chemicals (Mato et al., 2001; Sun et al., 2020; Zhang et al., 2020). Glucose is a hydrophilic substance and carries no charge; therefore, it is unlikely to interact strongly with MP surfaces, limiting the impact of MPs on glucose decomposition. This supported the finding that similar amounts of glucose-derived CO$_2$ and MBC were observed in MP amended soils (p > 0.05; Figs. 1a and 4 e). However, organic and inorganic chemicals in soil can be accumulated by MPs, which then attracting microorganisms that form a favorable niche for C and the cycling of other elements, this niche being referred to as the "plastisphere" (Amaral-Zettler et al., 2020). It is plausible that the plastisphere creates a microbial activity hotspot that accelerates the decomposition of SOM. However, SOM-derived CO$_2$ was not affected by high MP contents (Fig. 2), indicating that high MPs had a negligible effect on SOM decomposition. The significant decline in MBC on days 30 and 65 showed that high MPs inhibited microbial growth in glucose-amended soil (Fig. 4b). Slower microbial growth indicates less C and nutrient demand from the soil, and therefore, less SOM decomposition and negligible PE in soil amended with glucose (Figs. 2b and 3 a).

3.2. Effect of MPs on rice straw decomposition and straw-induced PE

Straw-derived CO$_2$ remained unaffected by the addition of MPs (p > 0.05; Fig. 1c); however, straw-derived MBC decreased under MP addition at the end of incubation (Fig. 4g). Previous research revealed that MPs inhibit the activity of enzymes related to soil C and N decomposition, reducing rice straw decomposition in paddy soil (Chen
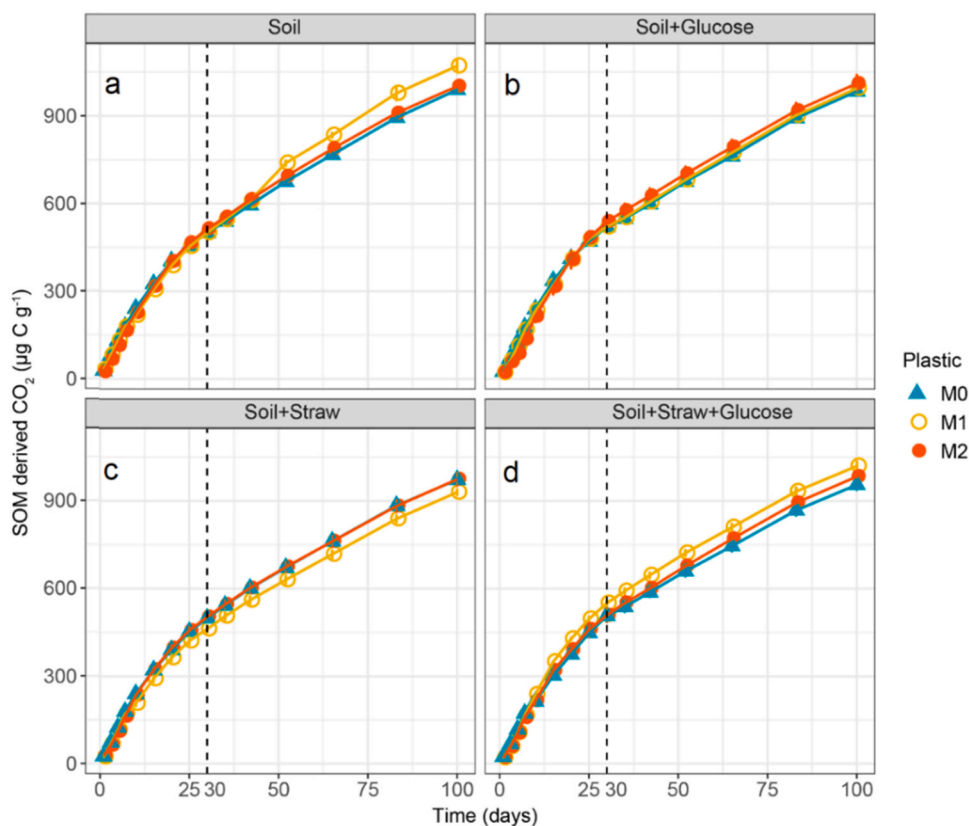
Fig. 2. Cumulative SOM-derived CO_2 in soils with and without microplastics. M0 = no microplastics; M1 = low (0.01%) microplastics; M2 = high (1%) microplastics. Error bars represent the standard error of the mean (n = 3).
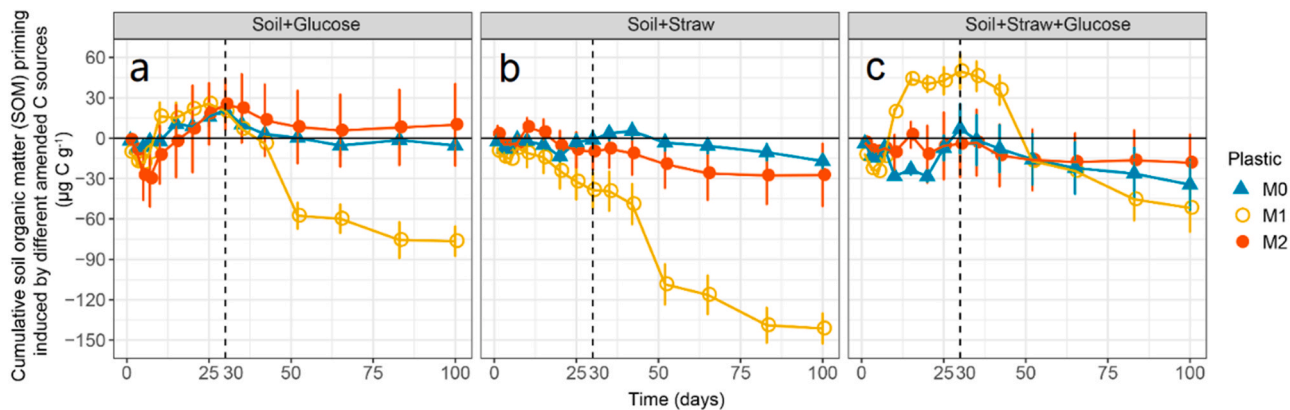

Fig. 3. Cumulative soil organic matter (SOM) priming induced by single straw or glucose addition or the combination of glucose + straw addition over 100 days. SOM priming is the difference in SOM decomposition between soils amended by straw, glucose, or glucose + straw with the no-addition control. M0 = no microplastics; M1 = low (0.01%) microplastics; M2 = high (1%) microplastics. Error bars represent the standard error of the mean (n = 3).

et al., 2020). The presence of MPs might alter the composition of microbial communities and their functions in paddy soil. For instance, MPs might cause a shift in the status of anabolic (i.e., microbial biomass growth or extracellular enzyme production) and catabolic (i.e., energy yield processes associated with CO_2) pathways of microbes (Chen et al., 2020; Guo et al., 2020; Zang et al., 2020). Rice straw-amended soil with low MPs contained more MBC than soil without MPs during the first 30 days of incubation; however, CO_2 release was similar (Figs. 1, 2 and 4). This indicates that low MPs induced C substrate for microbial biomass growth because r-strategist microorganisms proliferated rapidly and outcompeted the K-strategists in using the labile part of rice straw (Fontaine et al., 2003). Thus, MBC increased in soil with low MP

addition during the first 30 days of incubation. However, higher doses of MPs did not enhance the assimilation of C by microorganisms in rice straw-amended soil (Fig. 4c, g). Zang et al. (2020) reported that increasing the amount of polyethylene MPs in soil reduced the CUE of labile C. This supports the finding that MBC (straw derived) in high-MP soil was lower than that in low-MP soil on day 30 of incubation (Fig. 4c). However, the labile part of straw C can be exhausted over multiple weeks of incubation (Shahbaz et al., 2018a), potentially driving the strong decline in MBC after 30 days (Fig. 4c).

Cumulative negative PE accounted for 11% of total CO_2 efflux in soil with low MPs at the end of incubation. In contrast, cumulative negative PE in soil with high MPs was 2% of the total CO_2 efflux (Fig. 5). Previous
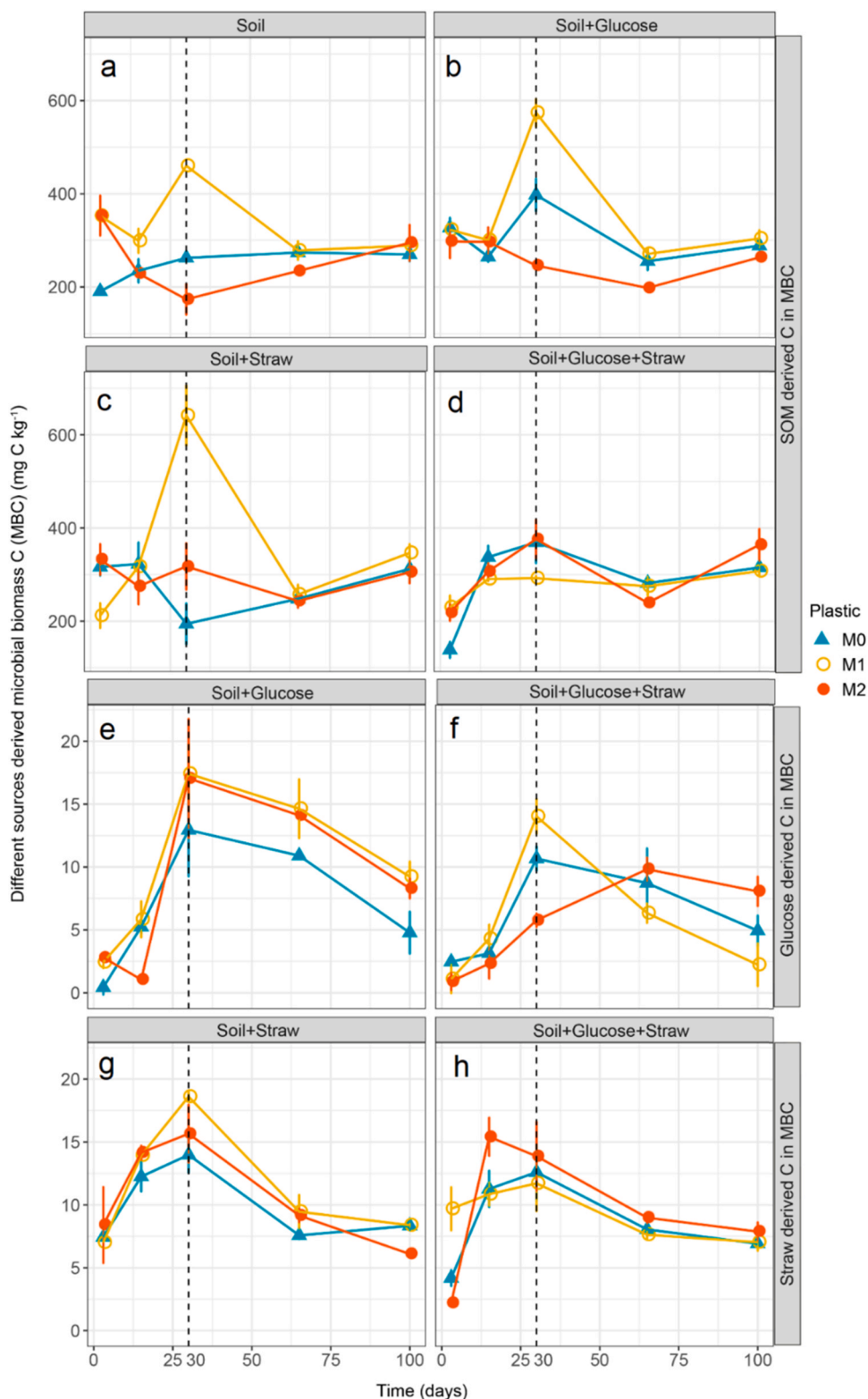
Fig. 4. Contribution of soil organic matter (SOM)-, straw-, and glucose-derived carbon (C) to the total microbial biomass C (MBC). M0 = no microplastics; M1 = low (0.01%) microplastics; M2 = high (1%) microplastics. Error bars represent the standard error of the mean (n = 3).

research revealed that rice straw induces both positive and negative PEs (Ye et al., 2015; Zhu et al., 2016, 2018b). Although the mechanisms of PE are complex and remain elusive, the direction and intensity of PE are mainly related to the quality and quantity of the substrate (e.g., rice straw in the present study) (Blagodatskaya and Kuzyakov, 2008; Shahbaz et al., 2017).

The decomposition of the easily degradable fraction (e.g., water soluble substances) and less-degradable or resistant fraction (e.g., cellulose and lignin) of rice straw are controlled by different microbial successional groups (Aoyama, 1996; Noll et al., 2005). Negative PE was observed during this period (i.e., 1–30 days in soils without MPs and 3–7 days in 1% MPs amended soil; Fig. 3b), indicating a pool substitution
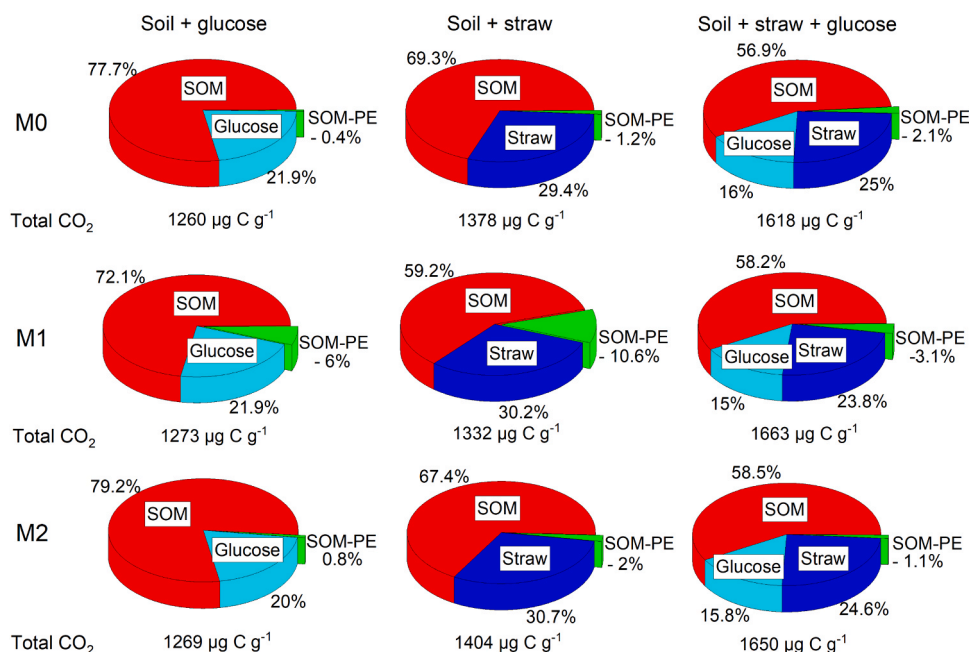
Fig. 5. Contribution (%) of three C sources (glucose-, straw-, and soil-derived C (SOM)) to cumulative CO2 efflux. The priming effect (PE) on SOM induced by straw or the combination of straw + glucose is also shown. SOM-derived CO_2 includes the proportion of SOM-PE CO_2. M0 = no microplastics; M1 = low (0.01%) microplastics; M2 = high (1%) microplastics.

phenomenon, as microorganisms benefit from labile substrate derived from residues (Blagodatskaya and Kuzyakov, 2008; Shahbaz et al., 2017). Soluble substances released from straw might activate SOM feeding microorganisms, because these compounds are also found in SOM; thus, enzymes produced to decompose straw might also degrade SOM (Fontaine et al., 2003). This supported the positive PE on SOM decomposition that induced by rice straw in the present study at 35–42 days in non-MP amended soil and 10–15 days in 1% MP amended soil (Fig. 3b). However, rice straw induced negative PE on SOM decomposition for most of the incubation duration, especially in low MP amended soils (Fig. 3b). The decomposition of SOM was also correlated to differences in C:N stoichiometry between SOM and exogenous substrates (Zhu et al., 2018b). Returning rice straw with a high C:N ratio may inhibit the microbial mineralization of native SOM (negative PE) because N and P are often insufficient in paddy soil to meet the demands of microbes (Li et al., 2012; Wei et al., 2020). Therefore, rice straw might have stimulated SOM-decomposing microorganisms; however, this effect would have been gradually overtaken by microorganisms that utilize nutrients from SOM for anabolism to support accelerated microbial growth. In addition, SOM-derived MBC in soil with straw was 2.0 and 3.3 times higher in low MP soil compared to high and no MP soil, respectively (Fig. 4c). Higher polyethylene MPs in soil might have reduced the CUE of labile C by inhibiting the growth of r-strategists, which mainly feed on labile C (Fontaine et al., 2003; Zang et al., 2020). In contrast, large amounts of necromass from accelerated r-strategists could be re-utilized by K-strategists during microbial succession in low MP amended soil, efficiently releasing energy and nutrients from SOM decomposition (Cui et al., 2020). The release of straw-derived CO_2 accounted for 33–35% of added straw C over 100 days incubation (Fig. 1c). This result indicates that negative PE and high proportions of incomplete decomposition of exogenous rice straw residue favor the sequestration of SOM in paddy soil (Wei et al., 2021).

3.3. Combined effect of glucose and straw on SOM decomposition with the addition of MPs

The combination of rice straw and glucose intensified the positive PE on the decomposition of SOM in low-MP soil at days 10–42 of incubation. However, negative PE was recorded during the later stages of incubation (Fig. 3c). Soil microorganisms are predominantly C-limited; therefore, the addition of glucose accelerates microbial growth. This in turn leads to microbial nutrient limitation, because microbial growth requires a suitable amount of nutrients (particularly N) to attain stoichiometric ratios (Zhu et al., 2018b; Wei et al., 2020). Rice straw could also serve as a primer; thus, the release of the labile part of rice straw coupled with the presence of glucose stimulated fast-growing microorganisms, which mainly invested energy in reproduction (r-strategists) (Tian et al., 1992; Ye et al., 2015; Shahbaz et al., 2018b). Total cumulative positive PE on SOM decomposition induced by glucose in soil with low MPs was 9.7 times more intensive than that in soil with high MPs (Fig. 6a). However, MPs resulted in a negative PE on rice straw decomposition induced by glucose, especially in soil with high MPs (Fig. 6b). The variable PE of SOM and rice straw induced by glucose highlights the difference in lability and metabolic status of these two substrates for microorganisms. Increasing the dose of MP addition can reduce bulk soil density, promote the saturated hydraulic conductivity (de Souza Machado et al., 2018, 2019). This probably promoted the diffusion of glucose and dissolved labile substrates in rice straw. Higher saturated hydraulic conductivity also favors the movement of microorganisms to hunt for nutrients, which facilitating fast-growing microorganisms (Wang and Or, 2013). Slower-growing microorganisms are the main group that degrade SOM; however, they grow too slowly compared to fast-growing microorganisms when competing for labile substrates (Fontaine et al., 2003). When sufficient substrates from glucose and the dissolved labile part of rice straw are present, the growth of fast-growing microorganisms is facilitated; however, the utilization of SOM is lower by slower-growing microorganisms because of their slow growth rate. As the glucose and labile substrates in straw were exhausted after day 30, and the recalcitrant part became dominant, the co-metabolism of straw and glucose occurred. This phenomenon was supported by the sharp decline in straw-derived MBC after a peak on day 15, whereas glucose-derived MBC continued to increase until it peaked on day 65 in high MP soil (Fig. 4f, h). This highlighted that straw served as a C source for microbial growth; however, the mineralization of C from straw declined in high MP soil. (Fig. 7).
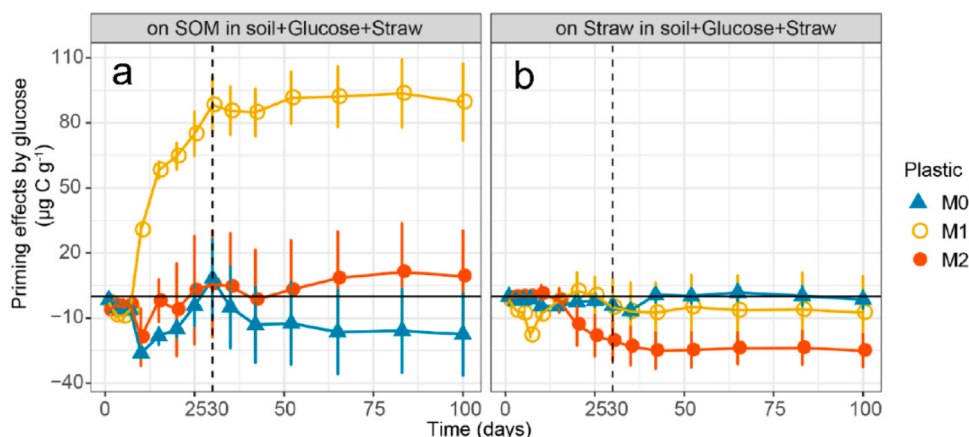
Fig. 6. Priming effects of glucose addition on the decomposition of straw and soil organic matter (SOM). Priming effects are the differences in SOM or straw decomposition between soil + straw + glucose and soil + straw amendments. M0 = no microplastics; M1 = low (0.01%) microplastics; M2 = high (1%) microplastics. Error bars represent the standard error of the mean (n = 3).
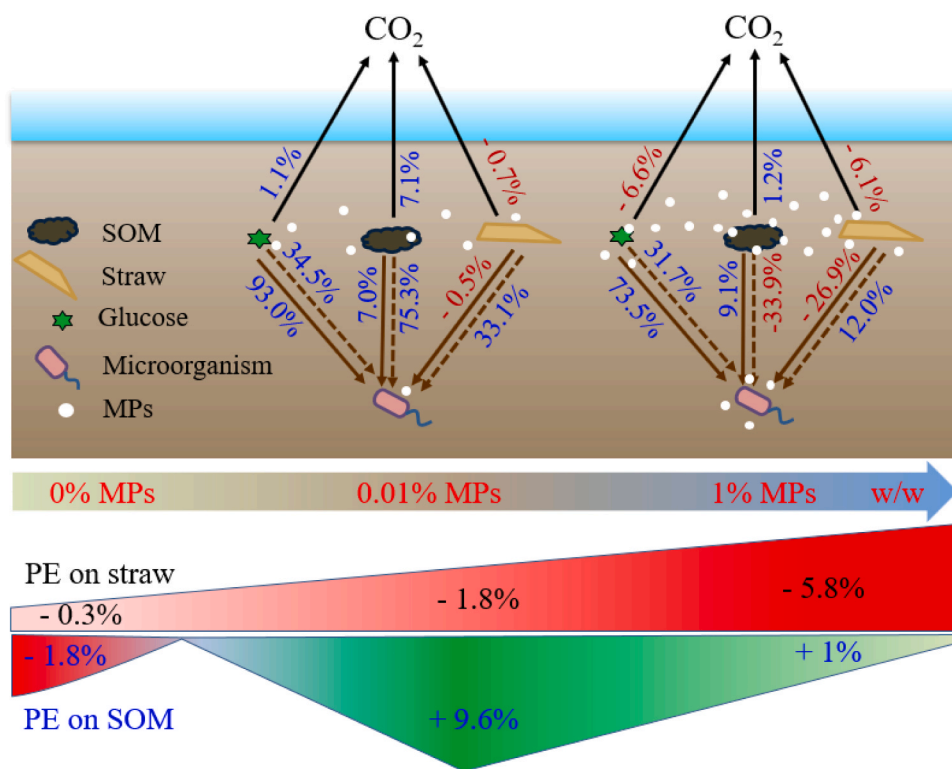
Fig. 7. Schematic presenting an overview of the effect of microplastics (MPs) on the priming effect (PE) of soil organic matter (SOM) and rice straw decomposition induced by glucose. Values in the upper part of the figure show the effect of MPs on glucose-, rice straw-, and SOM-derived C allocated to the microbial biomass C and CO_2 pool, in soil with or without a sole exogenous substrate (glucose or rice straw). That is, MP effects are indicated by differences between the substrate-derived C in MPs amended soil and soil without MPs. Dotted and solid arrows represent the effects on day 30 and 100, respectively. Values in the lower part of the figure indicate the effect of MPs on the PE induced by glucose in soil amended with both glucose and rice straw.

4. Conclusion

Our results clearly show that low doses of MP pollutants (e.g., polyethylene) strongly promote SOM decomposition in paddy soil. However, MP addition did not affect the mineralization of glucose and rice straw; rather, it drove the intensity of negative PE on native SOM decomposition, which is favorable for C sequestration in paddy soil. However, this was not the case under higher MP addition, which led to lower microbial activity and C utilization. Our results provide new insights on how MP pollutants impact SOM decomposition and PE by exogenous C (i.e., glucose and rice straw) in paddy soil. Future research should focus on clarifying the complex interactions among microbes, enzyme activity, physicochemical properties, and MP niche effects, as well as assessing the contribution of these processes to long-term soil C sequestration.

CRediT authorship contribution statement

Mouliang Xiao: Methodology, Investigation, Data curation, Writing - original draft, Writing - review & editing. **Muhammad Shahbaz**: Formal analysis, Writing - review & editing. **Yun Liang**: Formal analysis, Visualization, Writing - review & editing. **Jian Yang**: Writing - review & editing, Funding acquisition. **Shuang Wang**: Writing - review & editing. **David R. Chadwicka**: Conceptualization, Writing - review & editing. **Davey Jones**: Conceptualization, Supervision, Resources, Funding acquisition, Writing - review & editing. **Jianping Chen**: Project administration, Resources, Funding acquisition, Writing - review & editing. **Tida Ge**: Conceptualization, Supervision, Project administration, Writing - review & editing.

Declaration of Competing Interest

Acknowledgements

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.jhazmat.2021.126221.

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