Microplastic pollution promotes soil respiration: a global-scale meta-analysis

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Abstract

Microplastic (MP) pollution likely affects global soil carbon (C) dynamics, yet it remains uncertain how and to what extent MP influences soil respiration. Here, we report on a global meta-analysis to determine the effects of MP pollution on the soil microbiome and CO_2 emission. We found that MP pollution significantly increased the contents of soil organic C (SOC) (18%) and dissolved organic C (DOC) (13%), the activity of fluorescein diacetate hydrolase (FDAse) (9%), and microbial biomass (17%), but led to a decrease in microbial diversity (3%). In particular, increases in soil C components and microbial biomass further promote CO_2 emission (+24%) from soil, but with a much higher effect of MPs on these emissions than on soil C components and microbial biomass. The effect could be attributed to the opposite effects of MPs on microbial biomass *vs*. diversity, as soil MP accumulation recruited some functionally important bacteria and provided additional C substrates for specific heterotrophic microorganisms, while inhibiting the growth of autotrophic taxa (e.g., *Chloroflexi*, *Cyanobacteria*). This study reveals that MP pollution can enhance soil CO_2 emission by causing shifts in the soil microbiome. These results underscore the potential importance of plastic pollution for terrestrial carbon fluxes, and thus climate feedbacks.

Key words: Microplastics; Carbon cycling; Microbial community, Soil CO_2 emission, Meta-analysis

1. Introduction

About 80% of plastic wastes have been emitted to terrestrial ecosystems due to improper waste management and insufficient recycling since Anthropocene, and soils are a major repository of microplastics (MPs, particle size < 5 mm) (Thompson et al., 2004; Nizzetto et al., 2016; Geyer et al., 2017). MPs are relatively persistent in soils because of their chemical inertness, and MPs in polluted soils could reach up to 6.7% of soil weight in extreme cases (Fuller and Gautam, 2016; Chamas et al., 2020). Because of the C-enriched and persistent properties of MPs in environment, the impacts of MPs pollution on soil carbon (C) emissions become important to study, and may contribute to global "missing sources" in the context of global C-climate feedbacks (Stubbins et al., 2021).

MPs enter terrestrial ecosystems as C sources independent of photosynthesis and primary productivity, thus emerging as an important factor or component of C cycling (Rillig, 2018; Rillig and Lehmann, 2020). Studies have shown that MPs could influence the stability and bioavailability of soil organic C (SOC), subsequently affecting the C utilization by microorganisms, and potentially contributing to global climate change (Li et al., 2021; Ng et al., 2021;). Specifically, MPs typically enhance on soil dissolved organic C (DOC), which could further accelerate CO_2 production by providing available C for microorganisms (Gao et al., 2021; Shi et al., 2023). Additionally, MPs particles can be colonized by microorganisms and form a plastisphere, leading to the changes in microbial community composition and function (Zettler et al., 2013; Amaral-Zettler et al., 2020; Rillig et al., 2024). For instance, the occurrence of MPs

43 may drive the enrichment of plastic-degrading bacteria (e.g., *Actinobacteria*), which
44 may promote CO_2 emission from soils (Eilers et al., 2010; Stubbins et al., 2021).
45 However, MPs can also release toxic additives that restrain microbial activity and
46 reduce community diversity, which may decrease soil CO_2 emission (Guo et al., 2020;
47 Yu et al., 2021; Li et al., 2023). In addition, MPs in soils can cause nitrogen
48 immobilization because most kinds of MPs have a high C/nitrogen (N), which
49 consequently reduce microbial activity (Rillig et al., 2021b). Despite these critical
50 uncertainties, a comprehensive assessment of the effects of MPs on soil C release is
51 currently lacking.

52 The effects of MPs on soil CO_2 emission will likely depend on various factors,
53 such as MPs type, size, and concentration, soil type, as well as experimental conditions
54 (e.g., duration, method). For example, biodegradable plastic is regarded as a green
55 replacement for conventional plastic to alleviate the pollution of soil MPs (Lambert and
56 Wagner, 2017). However, they are generally degraded into smaller MPs rather than
57 being completely decomposed into CO_2, meaning that potential effect on soil C
58 emission remain uncertain (Zhou et al., 2021; Nelson et al., 2022). In general,
59 noticeable effects on soil C emissions or shifts in the soil microbial community are
60 typically observed only when the concentration of MPs exceeds a certain threshold
61 (Lenz et al., 2016). High concentration of low-density polyethylene (LDPE, 1.00%)
62 stimulated while low concentrations (0.01% and 0.10%) did not affect CO_2 emission in
63 farmland soil (Zhang et al., 2022a). MP size and exposure duration could also be
64 important in affecting soil C cycling because they determine the adsorption surface area

and number of oxygen-containing functional groups of MPs (Ren et al., 2021). In addition, the interactions between MPs and microorganisms are sensitive to various factors (Rillig et al., 2021b). Therefore, it is a high priority to synthesize existing evidence to reveal how MP pollution mediates CO_2 emissions from soils.

Here, we conducted a global meta-analysis with 1027 observations to test the effects of MPs on soil C emission and the microbial community. Based on a proposed framework (Fig. 1), we hypothesized that i) MPs can promote soil CO_2 emission by stimulating microbial activity and increasing microbial biomass; ii) some functional microbial taxa may play a key role in soil CO_2 emission under MPs exposure; and iii) the effects of MPs on soil CO_2 emission could greatly depend on the type, size, concentration, and exposure duration of MPs, soil type, and experimental factor. This study provides insights into how MPs affect soil CO_2 emission by regulating the soil microbiome at a global scale.

2. Materials and methods

2.1 Data collection

We conducted a topic search to collect peer-reviewed studies published before June 2023 from the Web of Science (All databases, https://apps.webofknowledge.com/), Google Scholar (https://scholar.google.com), and the China Knowledge Infrastructure (http://www.cnki.net/). Specific search terms were combined with regard to "(microplastic* OR nanoplastic*) AND (carbon OR organic matter OR CO_2 emission OR microorganism OR microbial biomass OR microbial diversity OR enzyme activity) AND soil". The studies in the meta-analysis had to meet the following inclusion criteria:

(i) at least one of the response variables is reported, including SOC, DOC, CO_2 emission, microbial biomass C (MBC), microbial biomass N (MBN), fluorescein diacetate hydrolase (FDAse), Saccharase, Cellulase, microbial diversity indicators (Chao, Shannon, abundance-based coverage estimator (ACE), and Simpson indexes); (ii) the experiments present the concentration, type, and size of MPs; (iii) the paired control and MPs treatments are reported in the study; any other additional factors were excluded; and (iv) if there are different harvest time points or test soils from the same article that meets the above requirements, each study was considered independent. Chao and ACE indexes were used to assess microbial community richness, and Shannon and Simpson indexes were used to indicate the diversity of the microbial community (Kim et al., 2017).

Based on these criteria, a total of 113 articles were selected, comprising 1027 paired experimental observations (Fig. S1; Table S1). We extracted the mean values, standard deviations (SDs), and sample sizes of each response variable in the control and treatment groups from the papers. Additionally, data from 55 articles that showed significant differences in soil microbial species at the phylum level between treatments were compiled, along with information on the dominant species in the MPs treatment. The data were extracted directly from tables and text or indirectly from figures and supplements using the WebPlotDigitizer (https://automeris.io/WebPlotDigitizer/). The sampling locations were extracted in each article or obtained through OvitalMap v9.3.4 (https://www.ovital.com/). Global distribution of sampling sites showed that most of the studies focused on agricultural soils and China was the research hotspots (Fig. 2A).

Moreover, the units of MPs concentration (e.g., g kg^{-1}, mg kg^{-1}) were converted into percentage to better compare the influence of MPs concentrations in different studies. Conventional polymers have a low degree of natural aging and are not easily decomposed in the environment compared to biodegradable polymers (Sun et al., 2022). As such, the type of MPs was divided into conventional MPs and biodegradable MPs (Table S2). The size of MPs included two groups: ≥ 100 and < 100 μm; and the exposure duration of MPs was divided into three groups of < 30, 30-60, and > 60 days. The soil types were classified as natural soils and agricultural soils based on the study area ecosystem types. Considering the differences in experimental conditions (without or with plants), we divided the experimental methods into incubation and pot experiment (Table S3). All the data collected in this work are from laboratory or greenhouse experiment. Due to the limited data, we did not consider the influence of climatic factors such as temperature and light conditions, as well as MP shape.

2.2 Data analyses

The effect size was measured by the natural log response ratio (RR), calculated as (Hedges et al. 1999):

$$RR = ln\left(\frac{X_t}{X_c}\right) = ln(X_t) - ln(X_c) \quad (1)$$

where X_t and X_c are the mean of treatment and control groups, respectively. Values of RR > 0 indicate positive effects, while those of RR < 0 indicate negative effects. The corresponding error variance (v) of the RR was calculated as follows:

$$v = \frac{{S_c}^2}{n_c {X_c}^2} + \frac{{S_t}^2}{n_t {X_t}^2} \quad (2)$$

where S_t and S_c denote the SDs of the treatment and control, respectively; n_t and n_c denote the repeated sample size of the treatment and control, respectively. When the standard error (SE) was presented instead of SD in the paper, we used the following equation to transform it:

$$SD = \sqrt{n} \times SE \tag{3}$$

The weighting factor (w_i) is the reciprocal of v_i. The weighted RR (RR_w), SE, and the 95% confidence interval (CI) were estimated by equations 4, 5, and 6 respectively.

$$RR_w = \frac{\sum_{i=1}^{k} w_i RR_i}{\sum_{i=1}^{k} w_i} \tag{4}$$

$$S(RR_w) = \sqrt{\frac{1}{\sum_{i=1}^{k} w_i}} \tag{5}$$

$$95\%CI = RR_w \pm 1.96 S(RR_w) \tag{6}$$

The RR of each variable was also converted into a percentage change caused by MPs, using the following equation:

$$(e^{RR} - 1) \times 100\% \tag{7}$$

The statistical analyses were conducted in Origin 2021 software and R (version 4. 3. 1) software using "metagear" (Lajeunesse, 2016) and "metafor" packages (Viechtbauer, 2010). The missing SDs were filled with the coefficient of variation from all studies with complete information (Bracken, 1992). The overall effects of MPs were estimated by the mixed-effects model using the *rma. mv* function ("study" as a random effect) and the restricted maximum likelihood (REML) method (Hedges et al. 1999). The between-group heterogeneity (Q_m) was used to test whether the effects of MPs differed significantly among the groups, wherein a significant Q_m value indicates

144 significant differences among groups. To test for publication bias, we used the funnel
145 plot method by plotting meta-analytic residuals against the inverse of their precision
146 (i.e., inverse of the sampling error) (Fig. S2). An asymmetric funnel plot indicates the
147 presence of publication bias. We used the 'randomForest' package to build the random
148 forest models (Breiman, 2001), and then we quantified the contributions of MPs
149 characters, experimental methods, and soil type to soil CO_2 emission, SOC, DOC, MBC
150 and MBN contents.

151 **3. Results**

152 *3.1 Overall effects of MPs exposure*

153 In the presence of MPs, the contents of SOC and DOC significantly increased by
154 17.9% (mean effect size = 0.17, 95% CI = 0.10–0.23) and 12.6% (0.12, 0.08–0.16),
155 respectively (Fig. 2B). Notably, MP exposure promoted soil CO_2 emission by 24.4%
156 (0.22, 0.12–0.32). The effects of MPs on soil enzyme activity depended on the enzyme
157 types. The activities of FDAse and cellulase increased by 9.4% (0.09, 0.02–0.16) and
158 27.0% (0.24, 0.13–0.35), respectively, while saccharase activity decreased by 8.6% (-
159 0.09, -0.18–0.00) (Fig. 2B). Moreover, MPs had significantly positive effects on soil
160 MBC (0.08, 0.00–0.16) and MBN (0.23, 0.10–0.36). By contrast, MP exposure had a
161 negative effect on bacterial diversity, as evidenced by a 1.1% decrease in the Shannon
162 index and a 3.3% decrease in the ACE index (Fig. 2B). However, there was no
163 significant effect of MPs on fungal diversity, including ShannonF, SimpsonF, and
164 ChaoF indexes (Fig. 2B). Additionally, *Proteobacteria* and *Actinobacteria* were the
165 dominant bacterial phyla, and *Ascomycota* was the dominant fungal phlyum in MP-
166 polluted soils (Fig. S3). Bacterial phyla such as *Proteobacteria* and *Actinobacteria*

increased frequently high (ranking in the top 25%), while *Acidobacteria* and *Chloroflexi* decreased frequently high.

3.2 Effects of MPs on the contents of SOC and DOC, and CO_2 emission

The type, size, exposure duration of MPs, experimental methods and soil type distinctly affected the contents of SOC and DOC, and CO_2 emission (Fig. 3, Table S4). Specifically, biodegradable MPs significantly increased DOC content (0.34, 0.25–0.43) and CO_2 emission (0.68, 0.46–0.90) compared to conventional MPs (Fig. 3A and C). Conversely, biodegradable MPs had a smaller positive effect on SOC relative to conventional MPs (Fig. 3B). Both MPs sizes had significant positive effects on SOC content, and bigger MP particles increased DOC content and promoted CO_2 emissions. The positive effects of MPs on DOC and CO_2 emission occurred during 30-60 days exposure, while the effect decreased with exposure duration (Fig. 3A and C). Soil CO_2 emission (0.25, 0.15–0.35) significantly increased in the incubation experiment (Fig. S4). The effect sizes of soil CO_2 emission exhibited a quadratic relationship with the increase in MPs concentration, reaching a peak at ~5%; and the effect sizes of SOC ($P > 0.05$) and DOC ($P < 0.05$) increased with MPs concentration, with thresholds of 10% (Fig. 4A–C). In addition, MPs increased SOC and CO_2 emission in agricultural soils, but not in natural soils. DOC content increased in both soils under MPs exposure, with a higher effect in the agricultural soils (Fig. S5).

3.3 Effects of MPs on microbial communities and enzyme activity

The type, size, exposure duration of MPs, experimental methods, and soil type influenced the effects of MPs exposure on soil microbial community and enzyme

189 activity (Fig. 5, Table S4). Biodegradable MPs significantly increased MBC (0.43,
190 0.26-0.60) and MBN (1.28, 0.95-1.61) compared to conventional MPs; larger MP size
191 significantly increased soil MBC (0.12, 0.02-0.21) and MBN (0.51, 0.34-0.68) in
192 contrast to smaller MPs. Notably, the positive responses of soil MBC and MBN were
193 observed only within the exposure time of 30-60 days. Below 10% MP concentration,
194 the effect sizes of MBC and MBN exhibited a quadratic relationship with effects
195 peaking at a MP concentration of ~5% (Fig. 4D and E). Moreover, MPs increased MBC
196 and MBN in the incubation experiments and in agricultural soils (Fig. S4 and S5).
197 Conventional MPs and larger MPs significantly decreased the Shannon index of
198 bacteria, but did not affect that of fungi (Figs. 5C and S5A). When MP exposure
199 duration was less than 30 days or exceeded 60 days, the bacterial Shannon index
200 exhibited a significant decrease (Fig. 5C), while the fungal Shannon index decreased
201 with the exposure time of MPs (Fig. S6A). Smaller MPs had a significantly positive
202 effect on the Simpson indices of bacteria and fungi (Figs. 5D and S5B). However, the
203 Chao indexes of bacteria and fungi were not sensitive to MPs exposure (Figs. 5E and
204 S5C). Moreover, the negative effect of MPs on the bacterial ACE index increased with
205 exposure duration, with a significant decrease (5.6%) when duration was > 60 days.
206 The RRs of Shannon indices of bacteria and fungi decreased with an increase in MPs
207 concentration (Fig. 4F and G). Bacterial Shannon and ACE indexes exhibited a decline
208 in incubation experiments, while Shannon and Chao showed increases in pot
209 experiments (Fig. S4). In addition, MPs increased the Simpson index of bacteria but
210 decreased the ACE index in the agricultural soils, and decreased Shannon index in the

natural soils (Fig. S5).

Conventional MPs had more effects on soil enzyme activity compared with biodegradable MPs (Fig. S6D-F, Table S4). Specially, conventional MPs exposure increased the activities of FDAse and cellulase significantly by 10.1% (0.10, 0.02–0.18) and 30.5% (0.27, 0.15–0.38), respectively, but decreased the activity of saccharase by 16.7% (-0.18, -0.24–-0.12). Larger MP size resulted in an increase in the activities of FDAse and cellulase, but a decrease in saccharase activity (Fig. S6 D-F). The activities of FDAse and cellulase significantly increased during 30-60 days, but were not affected when incubations were longer than 60 days (Fig. S6 D-F). Moreover, cellulase activity significantly increased in the incubation experiment, but the saccharase activity displayed the opposite pattern (Fig. S4). With MPs, the activities of FDAse and cellulase increased in agricultural soils, while saccharase activity decreased in natural soils (Fig. S5). The activities of FDAse, Saccharase and cellulase did not show significant effect with MP concentration (Figs. 4 and S7).

3.4 Linkages among soil C, microbial community, and CO_2 under MP exposure

The ACE index of bacteria was negatively related to SOC ($p < 0.01$), and the Shannon ($p < 0.05$), chao ($p < 0.01$), and ACE ($p < 0.05$) indices were negatively related to soil DOC (Fig. 6A). The Shannon ($p < 0.001$), Simpson ($p < 0.01$), and Shao ($p < 0.05$) indices of fungi were positively related to SOC. SOC, DOC, MBC, and MBN displayed a significant positive correlation ($p < 0.001$) with soil CO_2 emission (Fig. 6B–E). There was a positive relationship ($p < 0.001$) between DOC and SOC, and MBC, MBN and FDAse were positively related ($p < 0.001$) to DOC (Fig. S8). In addition,

233 random forest analysis demonstrated that MPs concentration had the highest
234 contribution to soil CO_2 emission, SOC, DOC, MBC, and MBN (Fig. S9).

235 **4. Discussion**

236 *4.1 MP pollution promotes CO_2 emission from soil*

237 MPs significantly promoted soil CO_2 emission, and this is mainly due to the
238 increase in soil C and microbial biomass (Fig. 7). The evidence comes from the positive
239 correlation between soil CO_2 emission and SOC/DOC/MBC/MBN (Fig. 6B-E). Indeed,
240 MPs are polymers with approximately 80% C content, which can be used as potential
241 C sources for microorganisms (Rillig, 2018). We found that DOC significantly
242 increased and SOC is significantly and positively associated with DOC under MP
243 exposure (Figs. 2 and S7A), suggesting that MPs may facilitate the mineralization of
244 organic matter, thereby enhancing the bioavailability of soil C. Furthermore, it has been
245 shown that the carbon-based substrates released by abiotic and biotic degradation of
246 plastics are available for heterotrophic growth (Romera-Castillo et al., 2018; Sheridan
247 et al., 2022; Qiu et al., 2024). Therefore, an abundant C source is assimilated by
248 microorganisms into their biomass and produces CO_2 through metabolic processes.
249 This was supported by the positive correlation between soil DOC and
250 MBC/MBN/FDAse (Fig. S8), implying that the surge in DOC levels induced by MPs
251 fosters the proliferation of microorganisms, thereby promoting soil CO_2 emission.
252 Moreover, the presence of MPs has been shown to improve soil structure, leading shifts
253 in the stability of soil aggregates (de Souza Machado et al., 2019; Wang et al., 2022).
254 Therefore, this alteration in soil structure provides microbial communities with greater

amounts of pore and better aeration potentially contributing to the observed increase in soil microbial biomass and CO_2 emission (Rillig et al., 2024).

The unique MP habitat, the plastisphere, selectively enriches certain microorganisms with effects that likely radiate into the soil. We observed that the Shannon and ACE indexes of soil bacteria significantly decreased, indicating that MP reduced bacterial richness and diversity (Fig. 2). The potential reason is that MPs or the additives they release (e.g., bisphenol A (BPA), bis (2-ethylhexyl) phthalate)) are toxic to soil microorganisms, making it difficult for some microorganisms to struggle to survive, resulting in a "neighbor avoidance effect" (Wang et al., 2021; Yu et al., 2021). The photosynthetic bacteria *Chloroflexi* and *Cyanobacteria* were decreased at a higher frequency, while heterotrophic bacteria with strong adaptability, such as *Proteobacteria* and *Actinobacteria*, are the dominant bacterial phlya in MP-polluted soil (Fig. S3A, C). MPs often lower soil bulk density, especially fibers, thus improving soil porosity and oxygen supply (Rillig et al., 2021a; Shi et al., 2023). These results indicated that MPs could promote soil CO_2 emission through reducing CO_2 utilization by photosynthetic microorganisms and increasing heterotrophic microbial respiration.

As a consequence of their chemical nature, MPs domesticate or recruit microorganisms with functions associated with the decomposition or degradation of organic compounds (Fig. S3; Li et al., 2024). The occurrence frequency of *Proteobacteria* and *Actinobacteria* increased under MP exposure (Fig. S3B), which might be because certain species within *Proteobacteria* and *Actinobacteria* possess the capability to degrade plastics (Eilers et al., 2010; Wang et al., 2023). Furthermore, the

activities of cellulase increased and the saccharase decreased (Fig. 2), indicating that microbial decomposition of plant residues and cellulose substances increased and that of sucrose substances decreased (Gunina and Kuzyakov, 2015). In general, microorganisms would preferentially decompose and utilize sucrose compared to cellulose, but the exposure of MPs allowed microbial community to make more use of more persistent substances. This is supported by the frequent occurrence of *Gemmatimonadetes* (Fig. S3B), as some members of the phylum participate in the decomposition of complex organic compounds (Hou et al., 2021).

Moreover, the decomposition of microbial necromass C may contribute significantly to soil CO_2 emissions. Although MP pollution caused a decline in certain microorganisms, the presence of MPs can decrease the content of microbial necromass (Chen et al., 2023). This is probably because microbial necromass is rich in N, and MPs entering the soil can result in a high C/N, which accelerates the decomposition of microbial necromass to obtain N (Kögel-Knabner, 2002; Liang and Balser, 2012). Consequently, the higher degradation and decomposition potential in MP-polluted soils also contributed to soil CO_2 emissions.

4.2 Factors influencing soil CO_2 emission under MPs exposure

The effect of MPs on soil CO_2 emission is variable, and this variability is largely caused by the different properties of MPs and the complexity of environmental conditions (Stubbins et al., 2021; Rillig et al., 2024). A random forest model showed that the concentration of MPs has the greatest effect on soil CO_2 emission, followed by the MPs type, experimental methods, soil type, exposure duration, and MPs size (Fig.

S9). The concentration of MPs broadly determines the input of soil plastic C, which is related to whether MPs stimulate or inhibit microbial activity. Our results showed that the effects of MPs on soil CO_2 emission first increased and subsequently decreased with MP concentration, with the largest effect at 5%, a trend consistent with the change in soil microbial biomass (Fig. 4). Although soil DOC was significantly positively correlated with MP concentration, microbial diversity exhibited a significant negative correlation (Figs. 4, 6, and S6). These results indicated that resilient microorganisms rapidly proliferate under abundant nutrient supply when the concentration of MPs is below 5%, and when it is above 10%, the majority of microorganisms may not survive normally due to the increased environmental pressure.

In terms of MP type, biodegradable MPs have a stronger effect on promoting soil CO_2 emission than conventional MPs (Fig. 3A), which can be attributed to their lower chemical stability. This allowed microorganisms to mineralize and degrade biodegradable MPs to available C, incorporating it into their own biomass, as suggested by the increase in soil DOC and microbial biomass (Figs. 3A and 5A, B). A recent study reported that PLA (Polylactic acid, biodegradable MPs) increased the number of unstable DOM molecules (e.g., carbohydrate-like compounds), while PE (Polyethylene, conventional MPs) increased condensed aromatic-like compounds, suggesting that biodegradable MPs provide more available C sources for microbial metabolism (Shi et al., 2023). Moreover, the higher electron transfer capability of DOM in biodegradable MPs soil may also stimulate its mineralization (Tan et al., 2017; Shi et al., 2023). Actually, the effect on soil CO_2 emission of MPs was time-dependent (Yu et al., 2022;

Shi et al., 2023). We found that the positive effects of MPs on CO_2 emission became more pronounced as time elapsed, with the largest effect observed between 30 and 60 days and the effect was not significant after 60 days (Fig. 3). This trend is similar to the content of soil DOC and the activity of FDAse. FDAse activity reflects total microbial metabolic activity, which is closely related to soil CO_2 emission (Green et al., 2006). Therefore, the reduced availability of nutrients and the release of more toxic extracts from MPs raise the pressure on microorganisms, resulting in lower CO_2 emissions after 60 days (Liu et al., 2023a). Given the limited data, we recommend long-term continuous observations better to understand the relationship between MPs and soil CO_2 emission.

In addition, soil CO_2 emission increased in agricultural soil but exhibited minimal variation in natural soils (Fig. S5), which may be related to the background value of soil MPs, fertilization history, and soil C storage (Li et al., 2021; Hu et al., 2023). Soils with high initial SOC content emitted less CO_2 under MPs exposure (Fig. S10). This may be explained by the priming effect (Bastida et al., 2019); the soils with more C have high microbial activity, and the addition of MPs does not strongly stimulate microbial decomposition. Therefore, soil conditions should be fully considered when studying effects of MP pollution.

4.3 Limitations

Although our meta-analysis elucidated the mechanisms of soil CO_2 emission mediated by MPs from the perspective of the soil microbiome, results still have several uncertainties, and there remain important knowledge gaps. Firstly, the concentrations of MPs added to the soil in laboratory experiments exceeded those typically found in

the natural environment, which might bias our ability to understand and predict the effects of actual MP pollution on soil C emissions and the microbial community (Phuong et al., 2016; Zhang et al., 2022b; Liu et al., 2023b). As such, the effects of MPs on soil C emission at environmentally relevant concentrations are worthy of further study; however, as with other global change factors, the interest is also in exploring effects of potential future levels of pollution. Secondly, our work primarily focused on exploring the impact of individual factors, without considering potential interactive effects. It is crucial to recognize that MPs pollution is increasingly acknowledged as a global change factor (Rillig and Lehmann, 2020) and should therefore be studied in the context of other global change factors rather than in isolation. Therefore, uncertainties exist when extrapolating the results of this work to the natural environment that is affected by multiple factors (Rillig et al., 2023).

5. Conclusions

We provide evidence that MPs increased the soil C pool (in part very likely because plastic-carbon was captured as soil C) and microbial biomass thereby promoting soil CO_2 emission. In general, bacteria exhibited a higher sensitivity to MPs than fungi, and as the MP concentration increased, the diversity of both showed a decline. Although MPs reduced soil microbial community diversity, the simultaneous increase in microbial biomass, along with the positive correlation between DOC and MBC/MBN/FDAse suggested that MPs could recruit plastic-degrading microorganisms or supply usable C sources for soil microorganisms, particularly heterotrophic microorganisms (e.g., *Proteobacteria*, *Actinobacteria*, and

365 *Gemmatimonadetes*). Consequently, the flourishing of these dominant populations
366 greatly enhanced the emission of soil CO_2. Specially, biodegradable MPs have a more
367 pronounced positive effect on soil CO_2 emission. The greatest effect of MPs on soil
368 CO_2 emission is observed at a MPs concentration of 5% or the exposure duration of 30-
369 60 days. Our meta-analysis not only advances our understanding of MP effects on soil
370 ecosystems, but given the global extent of this effect also highlights a potential feedback
371 mechanism with relevance to Earth system-level effects on climate.

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Figure captions:

Fig.1 Hypothesized processes by which MPs can affect soil carbon emissions.

Fig. 2 The distribution of sampling locations (A) and the effects of MPs on soil CO_2 emission, the contents of SOC, DOC, MBC, and MBN, enzyme activity, and microbial diversity (B).

Fig. 3 Effects of MPs on soil CO_2 emission, SOC, and DOC content based on MPs type, size, and exposure duration (days).

Fig. 4 Effects of MP concentration (%) on the effect size (RR) of soil CO_2 emission, SOC, DOC, MBC, MBN, Shannon indices of bacteria and fungi, and FDAse.

Fig. 5 Effects of MPs on soil microbial biomass and bacterial diversity based on MP type, size, and exposure duration (days).

Fig. 6 Relationships between the effect size (RR) of soil CO_2 emission and SOC (A), DOC (B), MBC (C), MBN (D); relationships between the effect size (RR) of soil C and microbial diversity (E).

Fig. 7 Mechanisms by which the soil carbon pool and microorganisms drive CO_2 emissions under the influence of microplastics.

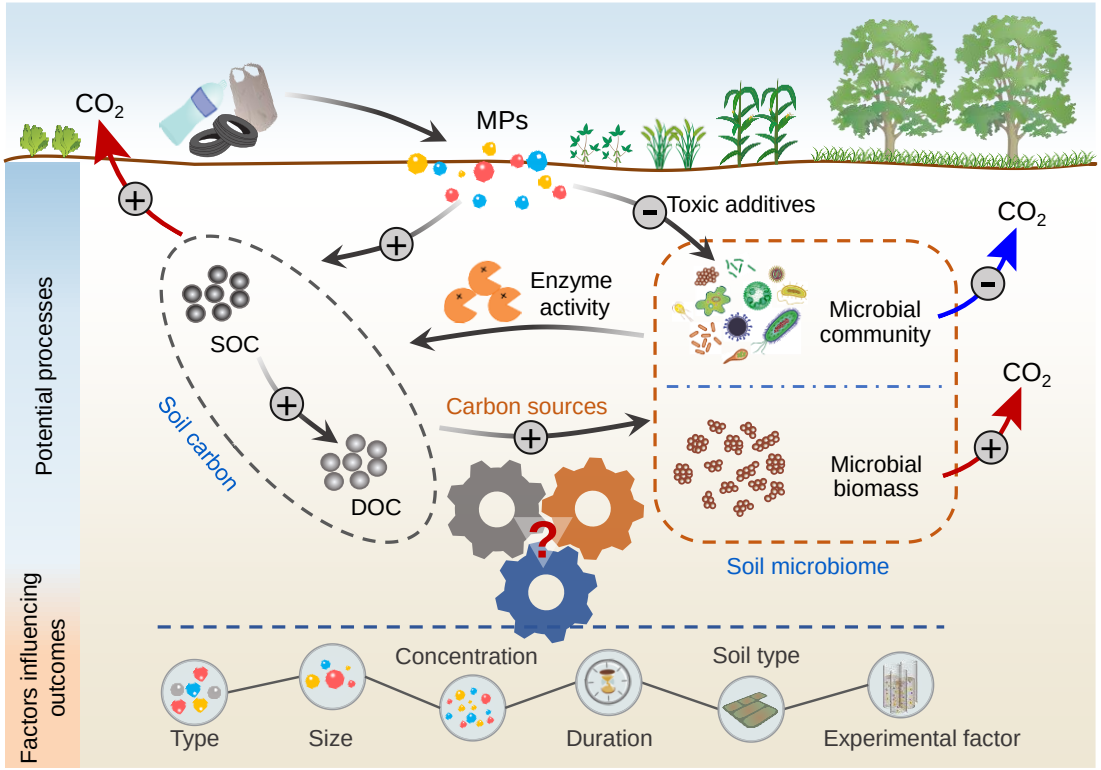


Fig.1 Hypothesized processes by which MPs can affect soil carbon emissions. MPs promote the transformation of soil organic carbon (SOC) to dissolved organic carbon (DOC), thereby providing more readily available carbon sources for microorganisms. Nutrients in sufficient supply increase the soil microbial biomass and promote soil CO_2 emission. However, toxic additives released from MPs could decrease microbial community diversity by inhibiting specific microbial taxa, thus decreasing soil CO_2 emission. Moreover, the various factors (e.g., MP properties, soil types, and experimental conditions) could change the effects of MPs on soil CO_2 emissions.

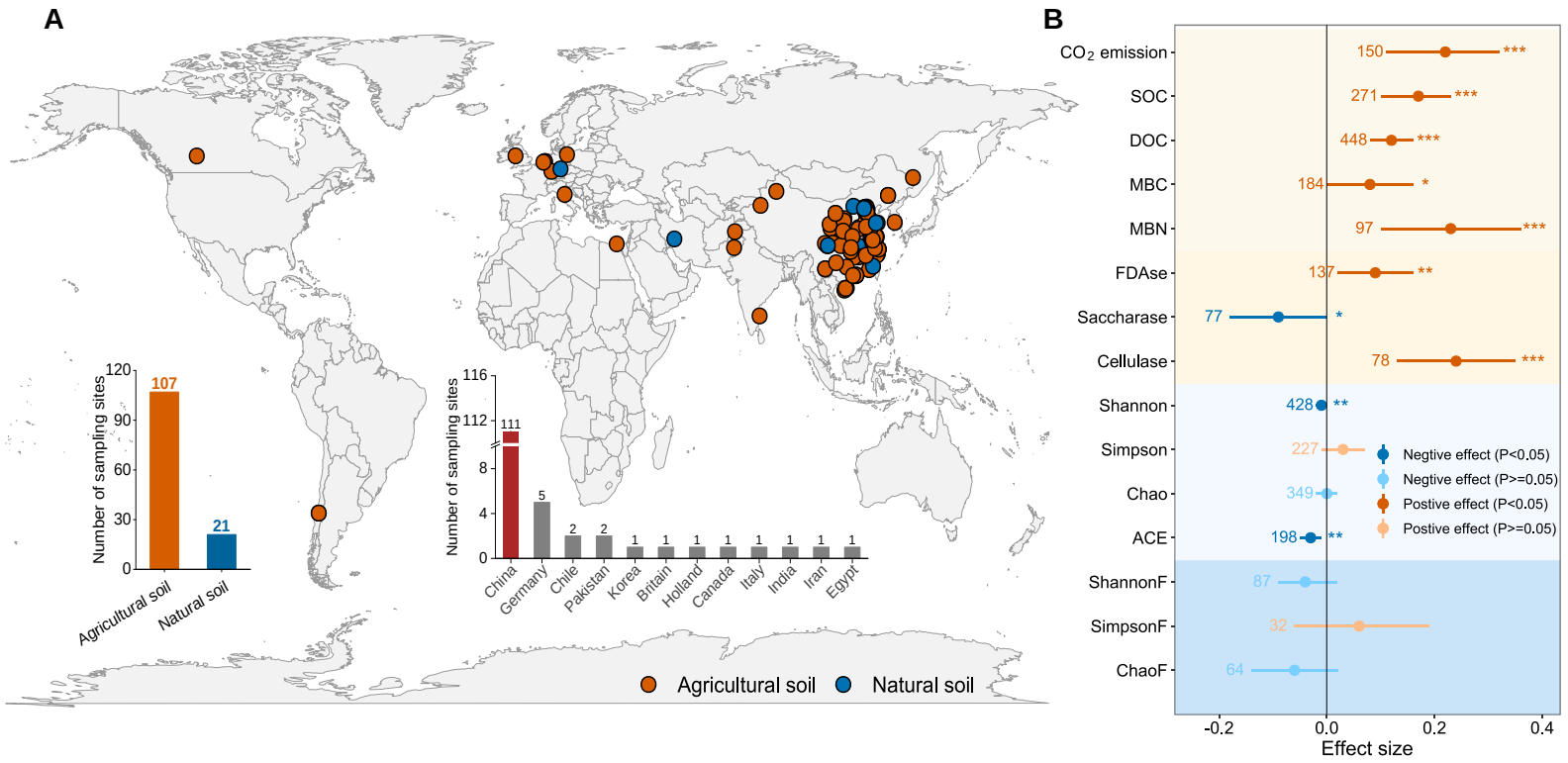


Fig. 2 The distribution of sampling locations (A) and the effects of MPs on soil CO_2 emission, the contents of SOC, DOC, MBC, and MBN, enzyme activity, and microbial diversity (B). SOC, soil organic carbon; DOC, dissolved organic carbon; MBC, soil microbial biomass carbon; MBN, soil microbial biomass nitrogen; FDAse, fluorescein diacetate hydrolase; Bacterial diversity includes Shannon, Simpson, Chao, and ACE indexes; Fungal diversity includes ShannonF, SimpsonF, and ChaoF indexes.

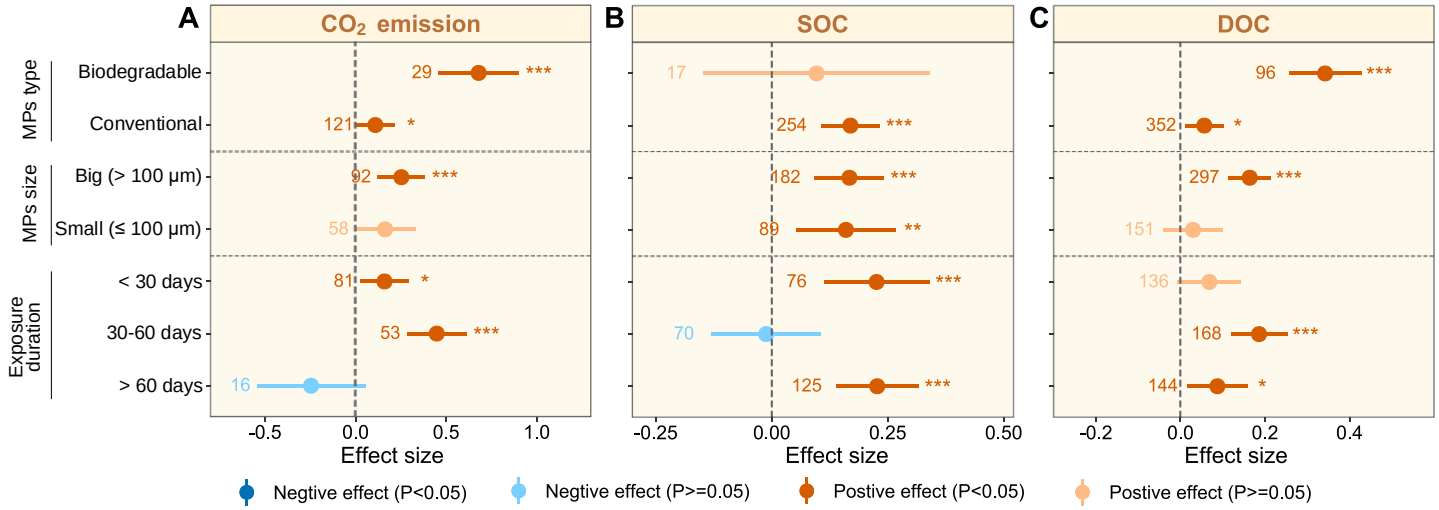


Fig. 3 Effects of MPs on soil CO_2 emission, SOC, and DOC content based on MPs type, size, and exposure duration (days). SOC, soil organic carbon; DOC, dissolved organic carbon.

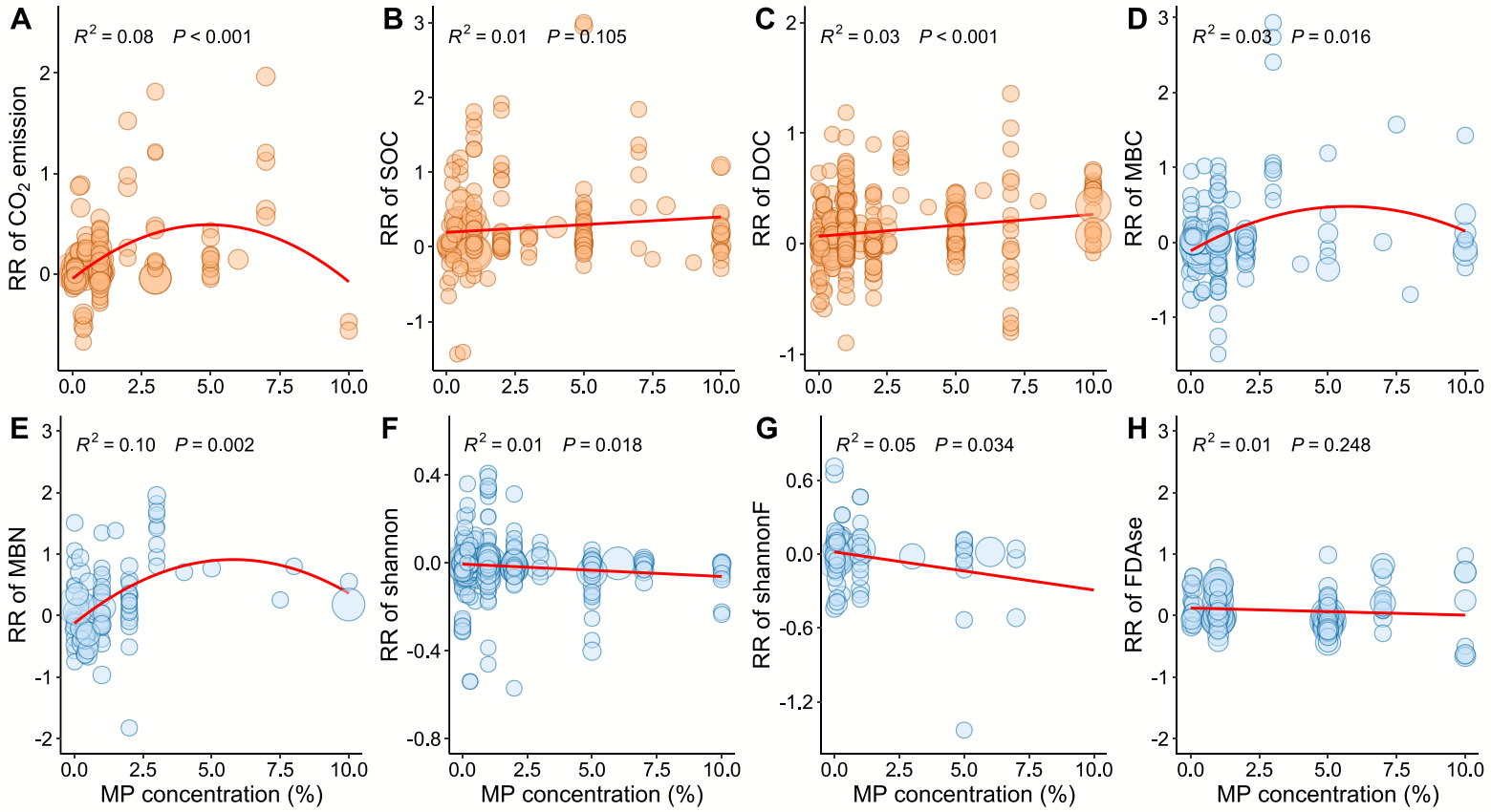
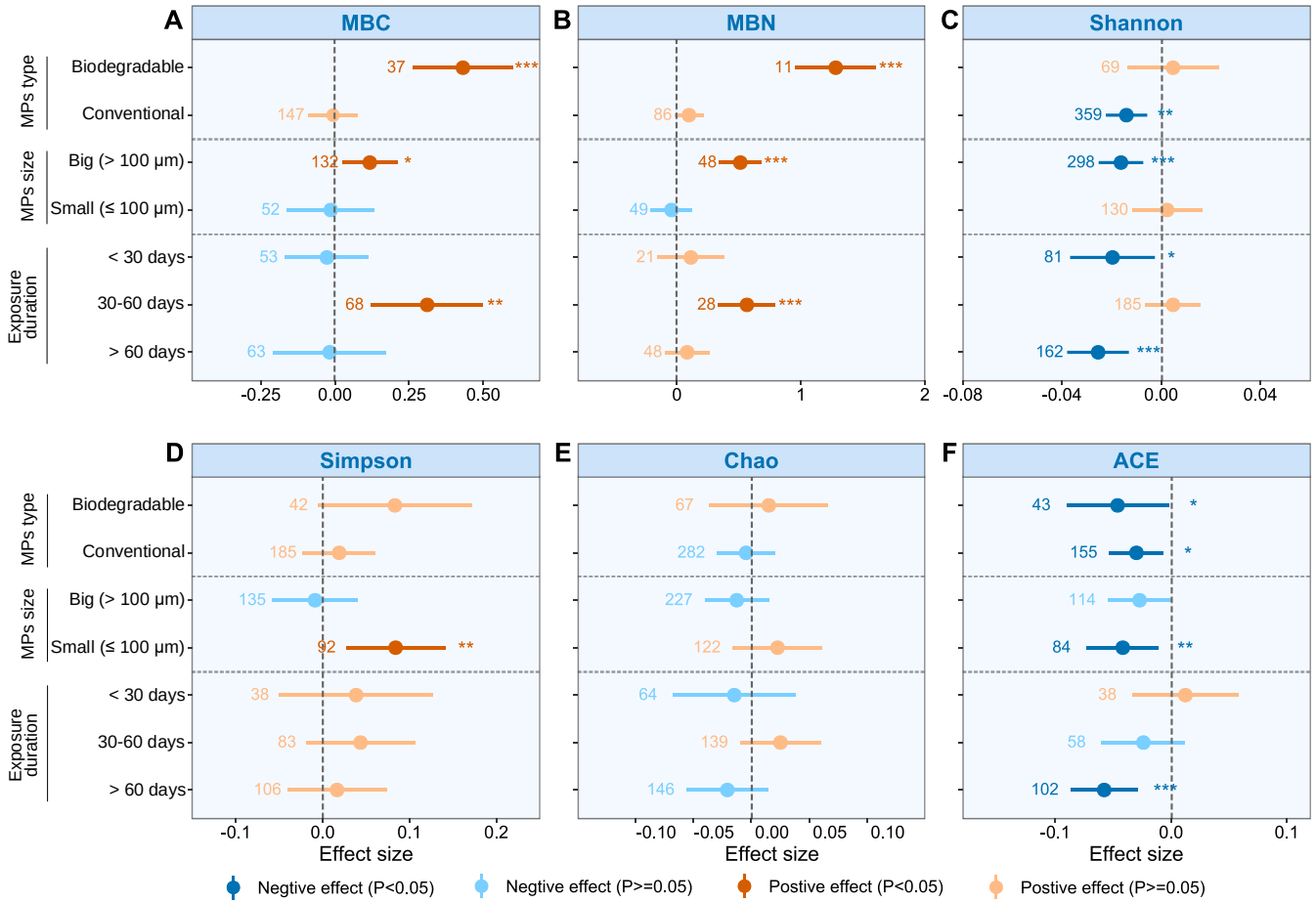


Fig. 4 Effects of MP concentration (%) on the effect size (RR) of soil CO_2 emission, SOC, DOC, MBC, MBN, Shannon indices of bacteria and fungi, and FDAse. SOC, soil organic carbon; DOC, dissolved organic carbon; MBC, soil microbial biomass carbon; MBN, soil microbial biomass nitrogen; FDAse, fluorescein diacetate hydrolase.



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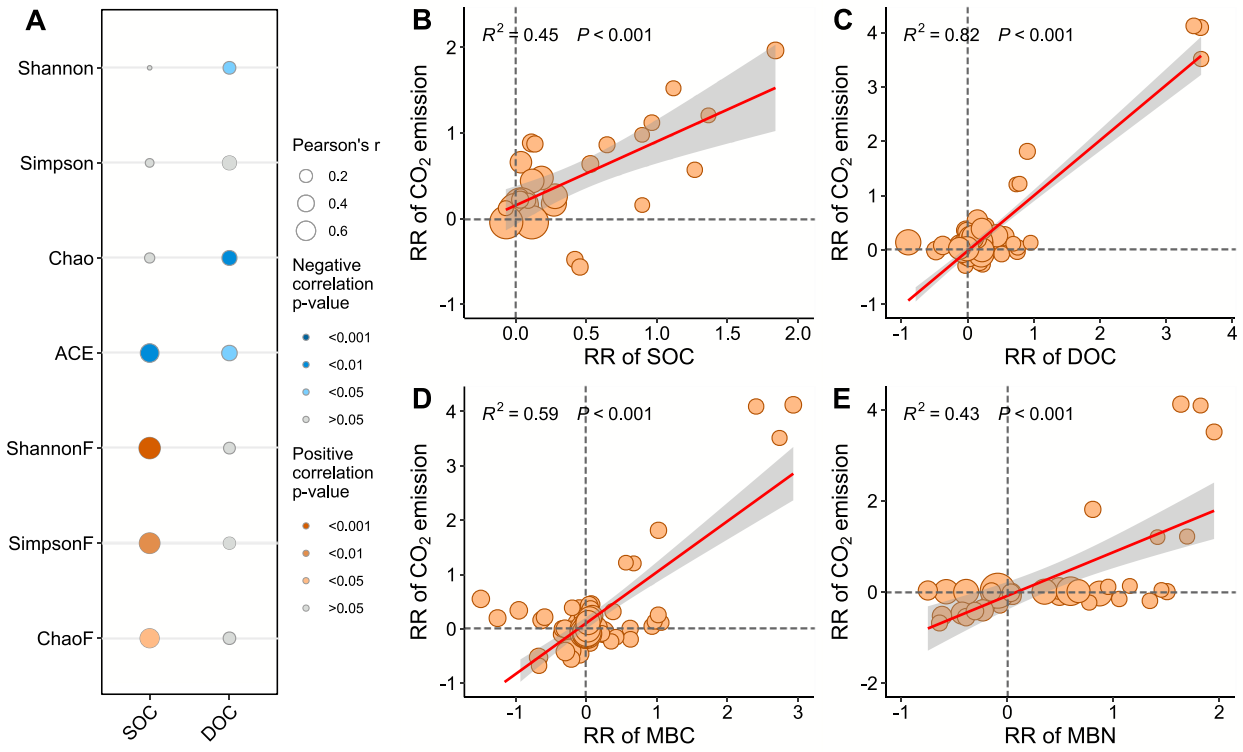

Fig. 6 Relationships between the effect size (RR) of soil CO_2 emission and SOC (A), DOC (B), MBC (C), MBN (D); relationships between the effect size (RR) of soil C and microbial diversity (E). SOC, soil organic carbon; DOC, dissolved organic carbon; MBC, soil microbial biomass carbon; MBN, soil microbial biomass nitrogen; Bacterial diversity includes Shannon, Simpson, Chao, and ACE indexes; Fungal diversity includes ShannonF, SimpsonF, and ChaoF indexes.

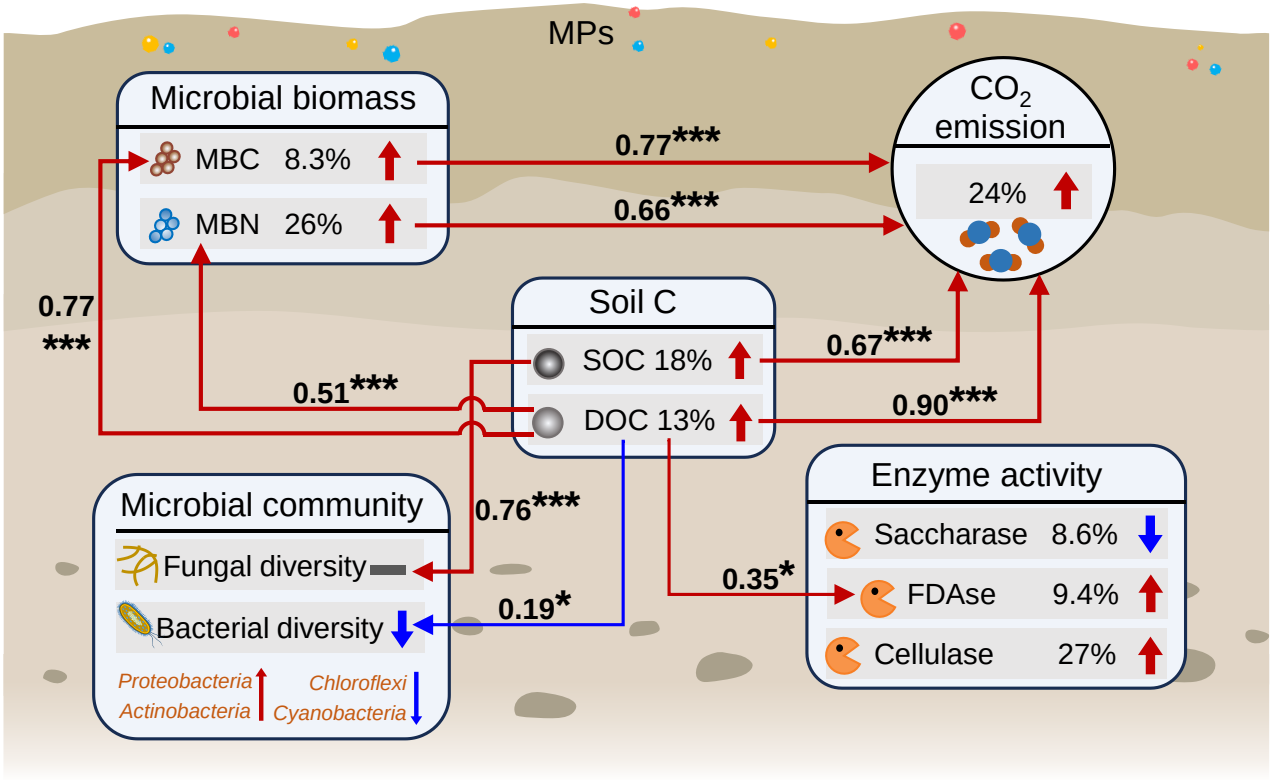

Fig. 7 Mechanisms by which the soil carbon pool and microorganisms drive CO_2 emissions under the influence of microplastics. Red and blue lines indicate positive and negative flows of causality, respectively. Figures on the line represent the correlation coefficient, and Shannon indexes of bacteria and fungi were used to calculate the correlation with soil SOC and DOC. ***, $P < 0.001$; **, $P < 0.01$; *, $P < 0.05$. SOC, soil organic carbon; DOC, dissolved organic carbon; MBC, soil microbial biomass carbon; MBN, soil microbial biomass nitrogen; FDAse, fluorescein diacetate hydrolase.