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Effects of microplastics on sedimentary geochemical properties and microbial ecosystems combined with hydraulic disturbance

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HIGHLIGHTS

- Sediment physicochemical properties had different responses to MPs.
- First simulation of real water environment disturbance in MPs-exposed sediment.
- Disturbance shifted nitrogen cycling functional microbes in MPs-exposed sediment.
- Disturbance affected sediment bacterial communities in MPs-contaminated sediments.

GRAPHICAL ABSTRACT

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ABSTRACT

Microplastics (MPs) pollution is widely investigated owing to its potential threats to river ecosystems. However, it remains unclear whether hydraulic disturbance deepens or mitigates the effects of MPs-contaminated sediments on the river environment. Herein, we studied the impact of sediment aggregates, organic matter, and enzyme activity, with emphasis on microbial community structure and function in sediments exposed to MPs (1 %, 5 %, and 10 % *w*/w) in conjunction with hydraulic disturbance. The experimental results showed that the influence of MPs on the sediment under hydraulic disturbance is more significant than that of static culture, especially for various environmental factors (MWD, MBC, and sucrase activity etc.). The proportions of the >0.05 mm-fraction aggregates increased from 74–76 % to 82–88 % in the sediment throughout the entire disturbance process. It has been found that the disturbance generally promotes the interaction between MPs and sediments. FAPROTAX analysis demonstrated that the disturbance reduced the difference in effects on microbial functional genes between the control group and the MPs-added groups by up to 10 times, suggesting that the effects of disturbance on MPs-contaminated sediments are relatively complex. This work provides new insights

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into the effects of hydraulic disturbance on physicochemical properties and microbial communities of MPs-contaminated sediment.

1. Introduction

In recent decades, the relentless production and use of plastics have led to incessant microplastics (MPs) pollution in the environment (MacLeo et al., 2021). Large quantities of microplastic pollutants have been reported in oceans, rivers, lakes, and other aquatic ecosystems. Due to the high surface area, MPs can easily adsorb organic/heavy metal pollutants or directly enter the food chain, threatening ecosystem security (He et al., 2023; Huang et al., 2019b; Liu et al., 2022a; Zarfl and Matthies, 2010). A considerable number of studies have been conducted to investigate the risk of MPs to aquatic organisms (Avio et al., 2017; Liu et al., 2023; Liu et al., 2022b; Shi et al., 2019a). Meanwhile, river sediment is an important "sink" of MPs in the freshwater environment. MPs could enter the river via many pathways, including industrial discharge, surface runoff, and sewage treatment effluent, and eventually get deposited in sediments (Ding et al., 2022). Maheswaran et al. (2022) reported that the abundance of MPs extracted from the sediment of the Kavery River, South India, ranged from 187.00 ± 103.00 to 699 ± 66.00 items kg^{-1}. There has been an increasing number of investigations into the effects of MPs on river sediments over the past few years. However, knowledge on the influence of MPs under various environmental factors in river sediments is still limited.

Sediment dynamics play a critical ecological role in river systems by affecting all nutrient levels and geomorphic processes (Droppo et al., 2016; Grabowski et al., 2011; Huang et al., 2019c). Hydraulic disturbance, as an important factor interfering with sediment dynamics, can also lead to changes in the state of MPs in sediments. Hurley et al. (2018) found that 70 % of MPs pollution can be effectively eliminated in riverbeds by extreme flooding events. Besides, hydraulic disturbance may also change the effects of MPs on the physicochemical properties in sediment aggregates and microbial communities, which may be different from those in long-term stable river environments. For instance, the turbulent flow process can increase the content of dissolved oxygen in the water body, oxidizing iron and manganese ions in sediment particles. These cause an iron-manganese oxide layer to form on the suspended particles surface, which could absorb abundant heavy metals (Atkinson et al., 2007). Besides, Chen et al. (2023) confirmed hydraulic disturbance could affect the growth of biofilm on MPs. Higher-speed water flow also contributed to MPs fragmentation and release, then change the nutritional level of the environment (Zhang et al., 2022). Overall, hydraulic disturbance can disrupt the dynamic balance of nutrient release and precipitation between the "sediment-water", which cannot be ignored. In this regard, the impact of MPs on the environmental properties of sediments under hydraulic disturbance in actual freshwater environments has not been clearly defined. Therefore, the study of MPs on different physical and chemical properties of sediments in disturbed systems should be carefully considered. In this experiment, we focused on the impact of disturbance on various properties of MPs-contaminated sediments.

Polystyrene (PS) is one of most frequently detected polymer types in freshwater sediment environments due to its versatile properties and utility (Chen et al., 2021; Di and Wang, 2018). Given the environmental relevance, PS-MPs were selected for the experiment and PS among sediment incubated with 1 %, 5 %, and 10 % *w*/w for 80 days. Subsequently, a comprehensive turbulence device was used for intermittent disturbance, and the effects of different amounts of PS on sediment bacterial community structure, diversity by high-throughput sequencing technology, and physicochemical properties under hydraulic disturbance were investigated. The aim of this study was to explore the impacts of disturbance on various properties of MPs-contaminated sediments, in the following three steps: (1) analyzing the effects of different MPs concentrations on the physicochemical properties and enzyme activities in sediments; (2) exploring the impact of disturbance to geochemical properties in MPs-contaminated sediments; (3) investigating the changes of bacterial community structure in MPs-contaminated sediments involving disturbance. It is expected that the results will contribute to evaluating the ecotoxicological effects and associated risks in the river system.

2. Materials and methods

2.1. Sample preparation

Riverine sediment (0–5 cm) was collected using metal shovels and columnar samplers from the Changsha section of the Xiangjiang River (112°57′39.422″ E, 28°11′6.091″ N) in June 2022. According to Yin et al. (2022), the average abundance of MPs in Xiangjiang sediments was 288 ± 60 items/kg. The MPs concentration detected in the sediment used in the experiment was 184 items/kg, indicating that MPs pollution in this area is relatively low. Collected sediments were air-dried in a clean environment for two weeks before the experiment. The dried samples were sieved (<2 mm) to exclude any large debris. The physicochemical properties of sediment are shown in Table S1. The virgin PS MPs (density:1.04–1.06 g/cm^3, size: 89 μm) were purchased from Zhongcheng Plastic Co. Ltd., China. The surface morphology of MPs was characterized by a Scanning Electron Microscope (SEM, ZEISS Sigma 300, Germany).

2.2. Sediment incubation test

Beaker experiments were conducted for incubation in our study. According to investigation studies, doses of MPs were set as 1 %, 5 %, and 10 % (*w*/w) for lower, medium, and higher MPs-polluted sediments, respectively. For each group, a total of 400 g riverine sediment samples amended with MPs in duplicate were added to a 500 mL beaker as well as a control treatment group without MPs. All treatment groups are displayed in Table S2. The sediment was maintained at the submerged state of riverine sediments in shallow freshwaters, deionized water was added to obtain the water surface at around 2 mm above the sediment surface and stabilized in the dark for 80 days. After incubation, approximately 10 g of sediment (dry weight) was collected from each beaker at 5 random points and thoroughly mixed. After freeze-drying, the sample is stored at −20 °C for further testing. The reagents purchased and used meet the standards of analytical grade and above.

2.3. Disturbance design

Disturbance studies were conducted using an approximately homogeneous turbulence simulation system (AHTS). The design of the AHTS system in this experiment was based on previous literature (Kang et al., 2019; Lucas et al., 2016). The detailed operation of the AHTS systems was described in text S2, turbulence is a purely diffuse, almost isotropic, and transversely uniform zero-mean flow in this device (Fig. S1) (Kang et al., 2019). The perforated plate was driven to vibrate up and down by a variable-speed motor, which simulated the resuspension of the sediment. Sediment contaminated with MPs is placed in a disturbance device. From 8:00 am to 11:00 am, the disturbance was continued for 3 h at 145 r/min, and about 25 g sediment (dry weight) was taken from five points in each device after standing for 24 h after the end of each disturbance, and thoroughly mixed. Samples were kept at −20 °C for following tests. After the disturbance experiment began, intermittent disturbance was performed on days 1, 3, 7, 15, 30, and 50. The detailed flow chart of the entire experiment is shown in Fig. S2.

2.4. Analytical methods

2.4.1. Measurement of sediment aggregate size and organic matter

The sediment samples were separated into four aggregate size fractions at 0 d, 30 d, and 50 d disturbance: including >1 mm, 1–0.25 mm, 0.25–0.053 mm, and <0.053 mm using the wet sieving method (Huang et al., 2019a). Soil organic matter (SOM) was determined by the external heating method of potassium dichromate and concentrated sulfuric acid. The mean weight diameter (MWD) of treated aggregate as an indicator of the sediment aggregate's structural stability, is calculated as follows (Zhu et al., 2018):

$$MWD = \sum_{i=1}^{n} x_i w_i \quad (1)$$

where w_i represents the weight percentage of aggregate particle size, i and x_i mean the average diameter of each aggregate particle size.

2.4.2. Measurement of microbial biomass and enzyme activity

Microbial biomass carbon (MBC) and Microbial biomass nitrogen (MBN) were measured by the fumigation-extraction method (Deng et al., 2016; Witt et al., 2000). The atmospheric pressure chloroform fumigation extraction method suitable for flooded sediment was used for determination. Specifically, about 5 g of fresh sediment was mixed with 300 alcohol-free chloroform in a 50 mL centrifuge tube to lyse microbial cells and release cellular content. The centrifuge tube was placed in a dark room for 24 h at 25 °C. After removing the chloroform, the sediment was transferred to a conical flask, and 50 mL of 0.5 M K_2SO_4 extractant was added. The mixture was shaken for 30 min before centrifugal treatment. Subsequently, the supernatant was filtered and the total organic carbon in the filtrate was determined by a total organic carbon analyzer. Another 12.5 g of sediment without fumigation was taken as the control group, following the same extraction procedure. Microbial nitrogen was measured by ultraviolet spectrophotometry with potassium persulfate digestion, and the difference between the extracted liquid and the blank group was calculated using fumigation-imitation steaming.

Soil MBC was calculated as:

$$MBC = \frac{EC - EC_0}{K_{EC}} \quad (2)$$

where EC means organic carbon extracted from fumigated sediment, EC_0 means organic carbon extracted from non-fumigated soils, K_{EC} is conversion coefficient, 0.45.

Soil MBN was calculated as:

$$MBN = \frac{EN - EN_0}{K_{EN}} \quad (3)$$

where EN means organic carbon extracted from fumigated sediment, EN_0 means organic carbon extracted from non-fumigated soils, K_{EN} is the conversion coefficient, 0.54.

Changes in two enzymes in sediments were quantified throughout the incubation to explore the related microbial activities among different treatment groups. Urease activities, as a common indicator evaluating MP effects on soil/sediment ecosystems, were determined by the sodium phenolate-sodium hypochlorite colorimetric method (Liu et al., 2023; Qin et al., 2022; Shi et al., 2019b). Sucrase activity was tested by 3,5-dinitrosalicylic acid colorimetric and expressed as glucose mg/g (Xue et al., 2022). More details of the sediment enzyme measurements can be described in Text S2.

2.4.3. DNA extraction and 16 S rRNA gene analysis

DNA of sediment using the FastDNA®Spin Kit was applied for extracting sediment DNA (MP Biomedicals, USA) in accordance with the manufacturer's instructions. The extracted genomic DNA was checked by 1 % agarose gel electrophoresis. Primers 341F and 806R were used to target V3–V4 regions of the 16S rRNA gene. High throughput sequencing was performed using the Illumina MiSeq platform (Majorbio Bio-pharm Technology Co., Ltd., Shanghai, China).

2.5. Statistical analysis

Statistical procedures were performed in IBM SPSS 27.0 and Origin 2021. One-way ANOVA was used to evaluate significant differences ($p < 0.05$) among different treatments. Sobs, Chao 1 richness, and Shannon diversity were used to assess the alpha diversity between groups. Gene function annotation was according to Functional Annotation of Prokaryotic Taxa (FAPROTAX). Redundancy Analysis (RDA) and Heatmaps were carried out to understand correlations between sediment properties and microbial communities.

3. Results and discussion

3.1. Changes in MPs properties before and after exposure

Based on SEM, the surfaces of the PS MPs after 80 d of incubation and 50 d of disturbance were compared with the pristine PS-MPs. As shown in Fig. 1(a), the surface morphology of the pristine PS-MPs was relatively smooth. In contrast, after 80 days of sediment culture and the subsequent 50 days of disturbance experiments, the surface morphology of the aged PS-MPs in Fig. 1(b–c) shows varying degrees of cracking traces, and cracks with layered structures, forming rough and irregular surfaces. This may be due to the adsorption of clay minerals on the surface of MPs during culture. In particular, the surface changes of MPs after disturbance are more prominent and rougher. It is speculated that the disturbance promotes the collision and aggregation between MPs and sediment, and more clay minerals can be adsorbed on the surface of MPs (Atkinson et al., 2007). The high proportion of Si, Al, Fe, and Mn attached to the surface of aged PS MPs was confirmed by Energy Dispersive Spectrometer (EDS) (Fig. S3). Moreover, the hydraulic disturbance can also increase the friction of the system on MPs, further deepening the wear and aging of MPs.

3.2. Effect of disturbance on sediment aggregate distribution with MPs

As the basic structure and functional unit of sediment, the formation of aggregates is the result of the interaction of microorganisms, crop roots, and other complex physicochemical processes. Studies have shown that about 70 % of the MPs entering the soil are closely linked to soil particles and participate in the formation of aggregates (Zhang and Liu, 2018). In our experiment, it was found that PS-MPs had little effect on the distribution of aggregate particle sizes in the 80 d culture compared with the control group. However, the proportion of aggregates with a particle size >0.053 mm generally showed an increasing trend, especially in sediment aggregates with added MPs during the disturbance test. This may be because disturbance increases the collision and aggregation between agglomerates. PS is a macromolecular organic substance with high hydrophobicity, which may cause more sediment minerals to bond with sediment particles through chemical bonds or van der Waals forces to form larger granular aggregates under disturbance, it was further confirmed in EDS analysis (Fig. S3). Therefore, microplastic-contaminated sediment aggregates tend to condense and cement under disturbance.

In addition to changing the particle size distribution of aggregates, Fig. 2d revealed MWD was generally improved after disturbance. Suggesting that the structural stability of MPs-sediment aggregate increased compared to static culture. Meanwhile, sediment aggregation is an important factor in soil organic carbon (SOC) stability (Zhao et al., 2018). Studies have shown that SOC (or SOM) is an important binder for formatting water-stable aggregates (Zhang et al., 2012). Our results also showed that the addition of MPs slightly enhanced the organic carbon concentration of aggregates involving disturbance (Fig. 3), the OM in

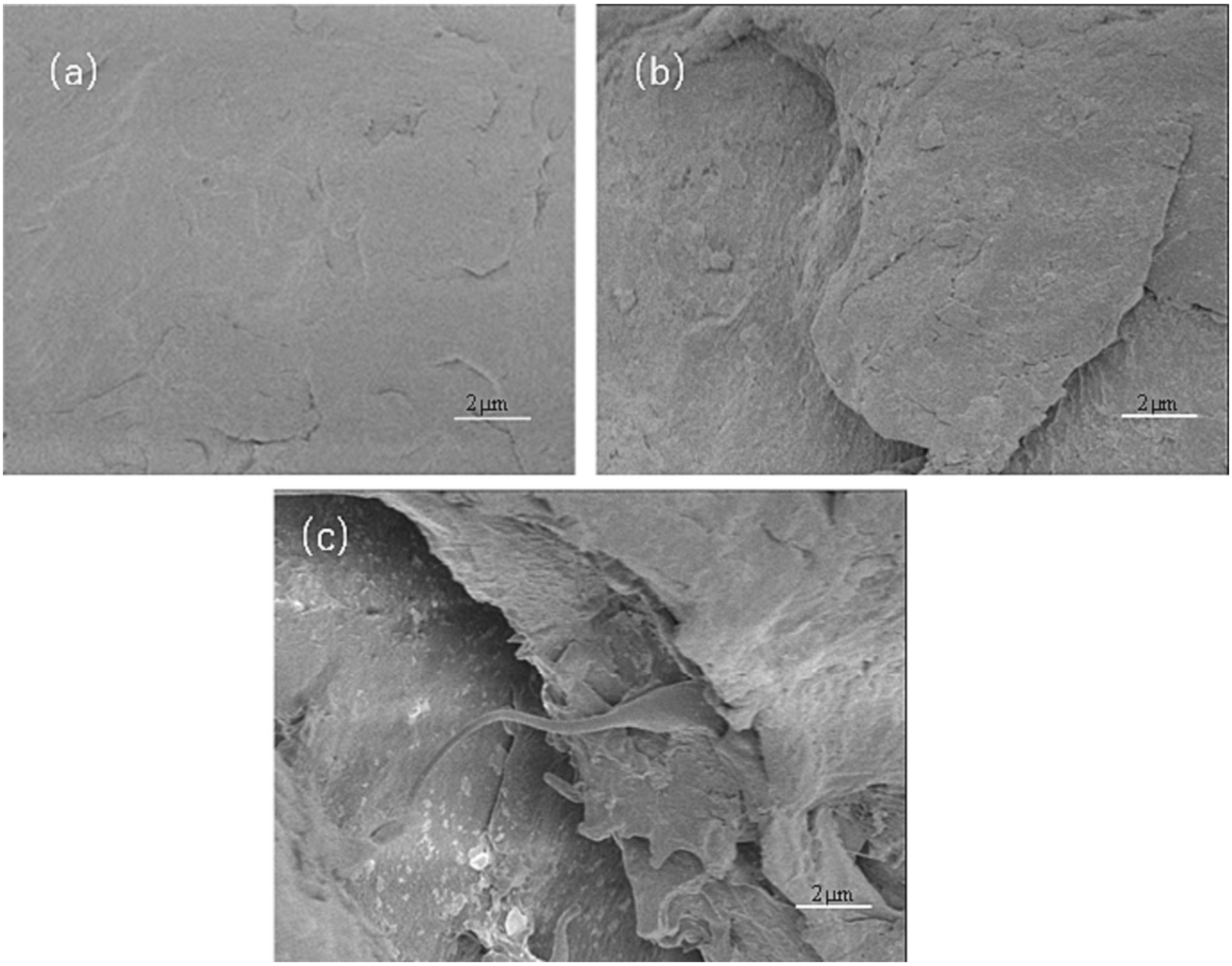

Fig. 1. SEM images of virgin PS MPs (a), aged PS MPs extracted from sediment (b) after 80 d incubation, and sediment (c) after 50 d disturbance.

the M10% group increased significantly at 50th d disturbance, while the macroaggregates (larger than 0.25 mm) and MWD decreased relative to the other groups. It is speculated that the effect of MPs on the stability of sediment aggregates weakens when collision and aggregation reach relative equilibrium in the late disturbance period. As mentioned above, the interaction between MPs and sediments is promoted under disturbance, and the OM and proportion of macroaggregate in sediments are changed, which could reduce the sediment bulk density.

3.3. *Effects of MPs and disturbance on microbial biomass and enzyme activity*

Soil microbial biomass (SMB), is used to describe the total mass of microorganisms present in soil. MBC is proposed as a sensitive indicator for measuring the adverse effects of contaminants on the soil microbial community (Broos et al., 2007). The addition of PS-MPs significantly affected MBC and MBN as compared to controls, where PS-MPs added reduced MBC and MBN in all treatments (Fig. 4(a–b)). Our findings agree with Khalid et al. (2023), who found that bioaccumulation of MPs can significantly decline microbial biomass carbon and nitrogen content. During the initial disturbance, the sediment state suddenly changes from static to dynamic, the oxygen content in the water body increases, then the microorganisms in the sediment are stimulated, resulting in enhanced activity and a corresponding increase in MBC and MBN, which ranged from 34 ± 15.5 to 95 ± 19.6 and 5.9 ± 1.75 to 14.17 ± 1.21, respectively. As the disturbance continues, the interaction between MPs and sediments is promoted, the inhibitory effect of MPs on sediment increases, MBC and MBN decrease. The variation of MBC and MBN is not completely consistent, the peak value of MBC in groups supplemented with MPs is one cycle compared with MBN involving disturbance. This is speculated the inhibition of PS on microorganisms involved in the nitrogen cycle is greater than that involved in the microbial carbon cycle. In addition, our study also found that the order of MBC content among the components was the same sequence before and after the entire disturbance cycle: M1% > M10% > C > M5%, and there was little difference among the components. This phenomenon may be because disturbance can change the chemical properties in the sediment, such as dissolved oxygen (DO), pH, and affect microbial communities (Eggleton and Thomas, 2004, Peng et al., 2021, Yi et al., 2023), leading to gradually narrowing the difference in the impact of MPs on the sediment environment between groups.

Extracellular enzymatic activities are vital indicators of sediment functioning, including nutrient cycling, metabolic functioning, and other microbial activities (Cao et al., 2020; Pang et al., 2023; Qin et al., 2022). MPs effects on soil or sediment urease activity show more complex patterns, depending on the properties of MPs and environmental conditions. In general, enzyme activity is reduced in soils that have been chronically contaminated with plastic debris (Liu et al., 2023; Qian et al., 2018). The sediment urease activity in different treatments is illustrated in Fig. 4c. In comparison, the 80-day incubation test showed that PS-MPs significantly inhibited sediment urease activity and sucrase activity at 1 % *w*/w, and slightly stimulated sediment urease activity at 5 % and 10 % w/w, which may be attributed to inverse dose-dependent effects. Enzyme activities related to the nitrogen cycle generally

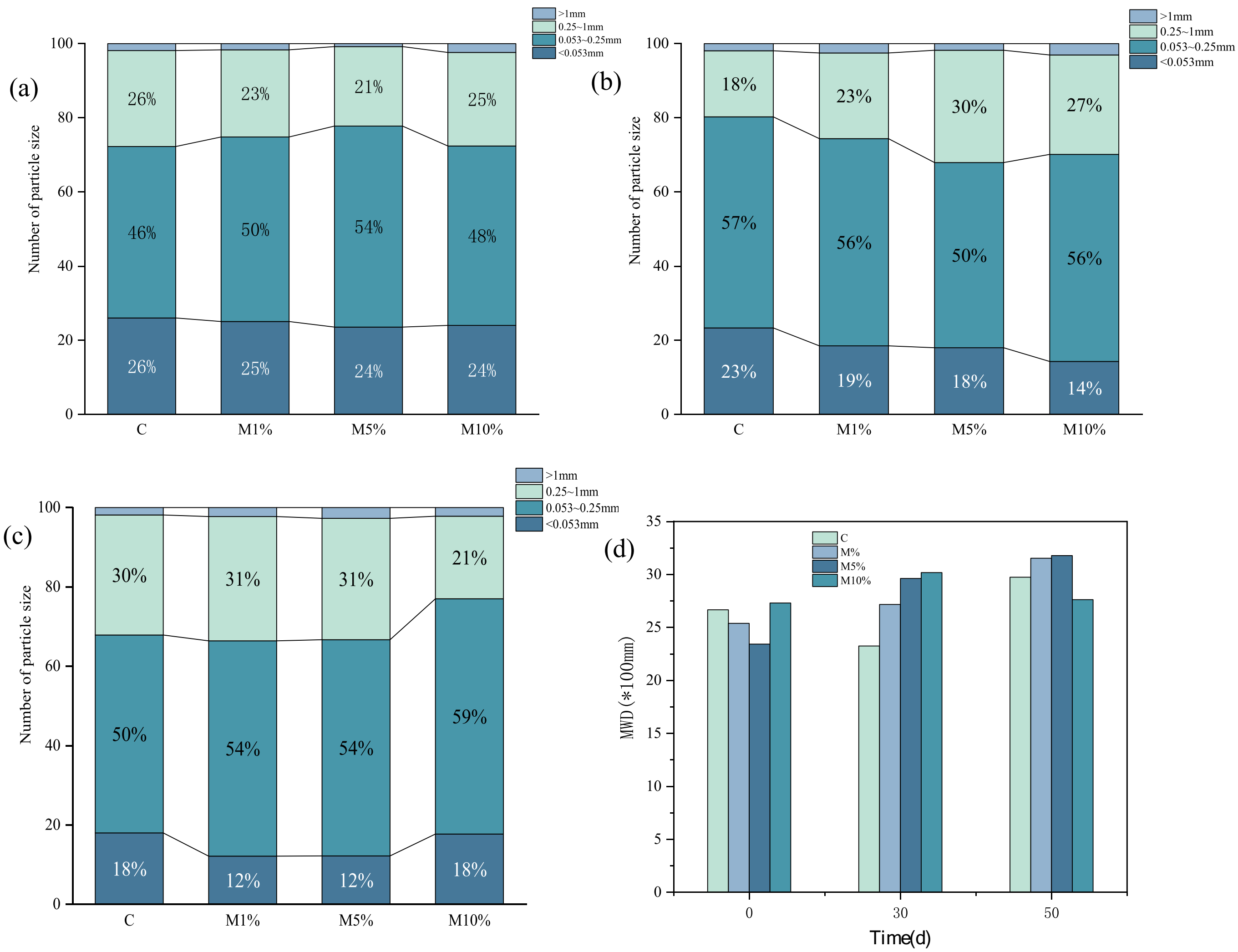

Fig. 2. Disturbances of aggregates in control and PS-MPs treated sediment, (a): 0 d; (b):30 d; (c):50 d, and the mean mass diameter of sediments under different PS-MPs pollution at 0 d, 30 d, 50 d disturbance (d).

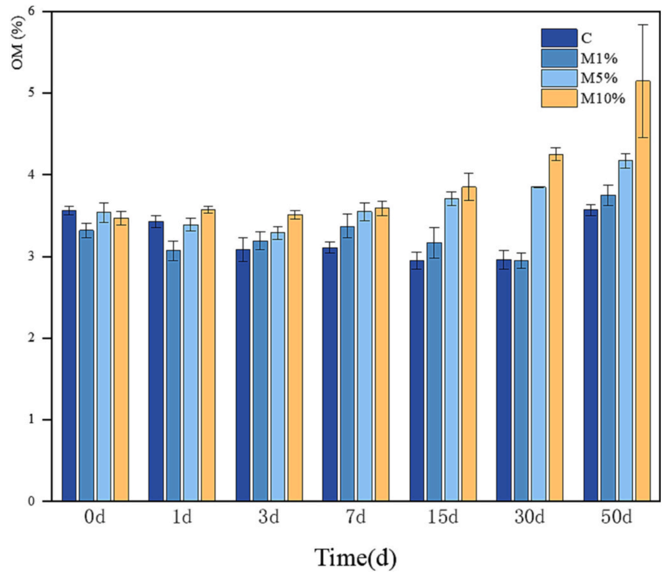

Fig. 3. Organic matter content in different disturbance processes.

decrease with decreasing carbon availability (Liu et al., 2023; Yi et al., 2021). As an important extracellular nitrogen mineralize, urease is widely used in nutrient cycling and microbial nutrient requirement assessment (Song et al., 2020). Low-dose MPs affected sediment carbon consumption, leading to a decrease in urease activity. However, disturbance can promote hydraulic circulation and increase the water content between aggregates. On one hand, urease is related to sediment moisture. Studies have shown that for every 21 % reduction in sediment moisture, urease decreases by 10–67 % (Sardans and Peñuelas, 2005). On the other hand, oxygen content is also key to nitrogen conversion and nitrogen cycling processes (Shen et al., 2022), the disturbance causes a sudden increase in oxygen content, which may inhibit the circulation of the anaerobic part of the nitrogen cycle, thereby affecting urease activity. Under these two actions, the influence of MPs on the urease activity of sediments gradually weakened. Similar result analysis to urease, the response sensitivity of sediment sucrase activity in different components to PS-MPs was dependent on the addition dose, among which the sucrase activity increased most significantly in M10% compared to control groups, with the difference ranging from 1.15 ± 0.14 to 4.45 ± 0.36 mg/d, as shown in Fig. 4d. A high dose of MPs may promote the hydrolysis rate of polysaccharides to monosaccharides in sediments and increase invertase activity (Chen et al., 2022; Zhou et al., 2021). Chen et al. (2022) also demonstrated that invertase increased by the supplemented dose of MPs (10%MPs > 5%MPs > CK). Meanwhile, M1%

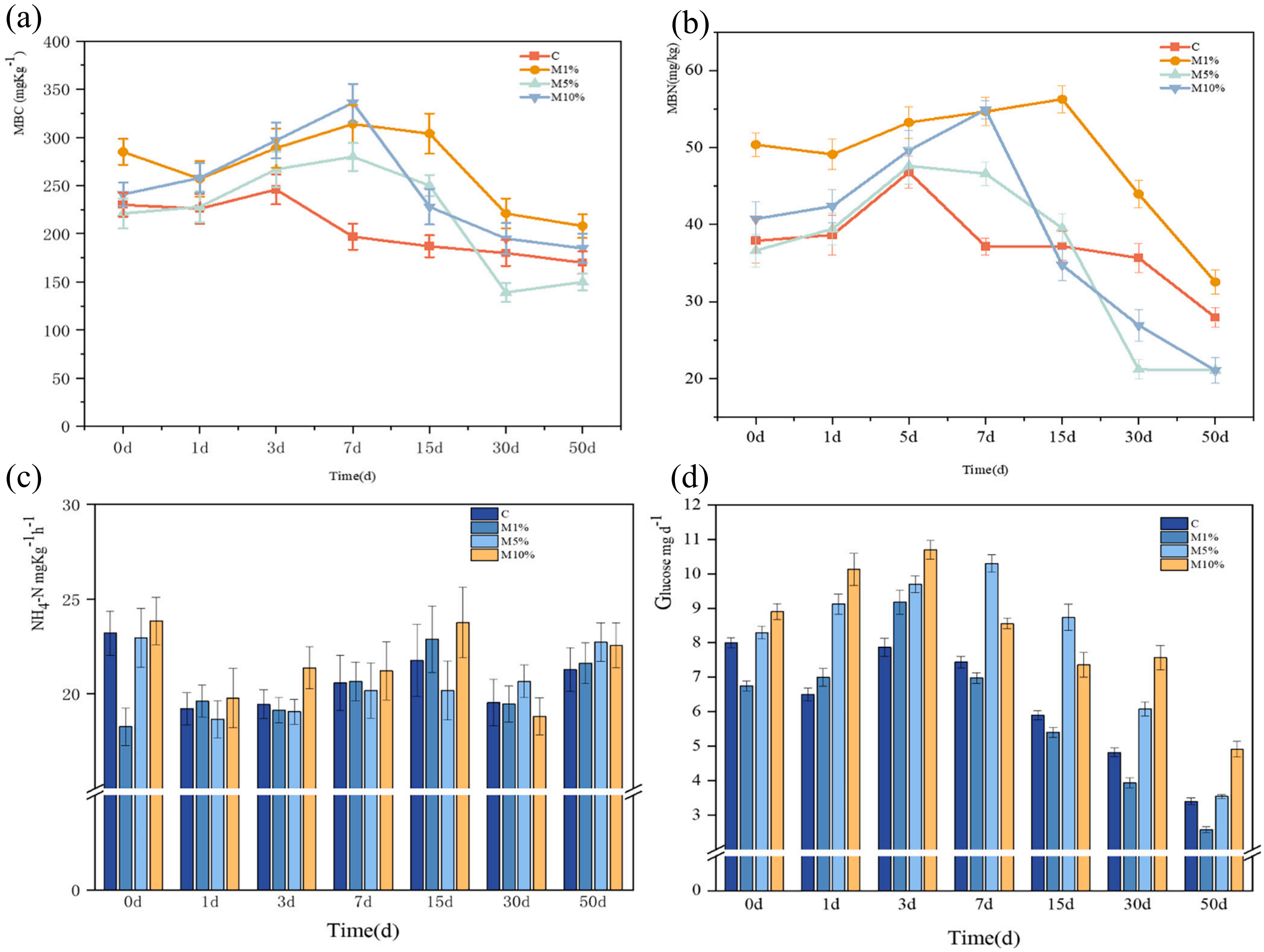

Fig. 4. Sediment MBC (a), MBN (b), urease (c), and sucrase (d) activities in different disturbance periods.

reduced the sucrase activity relative to the control group. Some research has also found that 1 % PS MPs significantly decreased soil sucrase activities (Yu et al., 2021). The effect of MPs on enzyme activity depends not only on MPs dose but also on the sediment texture. Disturbance increased the collision and binding frequency between various aggregates and MPs, indirectly promoting the interaction between MPs and sediment microorganisms, the sediment sucrase activity increased in the high-dose MPs group during the first seven days of disturbance. By changing the physical and chemical properties of the sediment environment, the disturbance destroyed the original microbial balance and inhibited microbial enzyme activity (Eggleton and Thomas, 2004). When the influence of MPs on sediments tends to be balanced, the inhibition effect of disturbance continues to increase, resulting in a decrease in the sucrase activity in overall sediment.

3.4. Effects of MPs and disturbance on sediment bacterial community

3.4.1. Diversity analysis of sediment bacterial community

Soil microbial community can indicate the change in soil quality and the stability of aggregates partly depends on the diversity and composition of soil microbiota (Feng et al., 2022). Thus, exploration about the overall response of the bacterial community and in treated sediment to MPs and hydraulic disturbance was necessary. Rarefaction curves, shown in Fig. S4, became relatively stable as the number of sequencing increases, indicating that the sequencing in our study was sufficient to reflect the bacterial community in the sediments. The alpha diversity indices can reflect the richness and diversity of sediment bacterial communities under different treatments. Differences in bacterial community richness and diversity between control and MPs amended sediment were found. Ace and Chao 1 (Table S3) indices added with MPs were lower than in the control group at the early disturbance stage, especially with 10 % PS-MPs added, suggesting an inhibiting effect on the richness of microbe under PS-MPs treatment. Nevertheless, Ace and Chao 1 indices in sediments with MPs were less reduced compared to the control group and even increased in the group with 10 % MPs at 50 d disturbance. The result may be that the MP treatment group has higher stability by hydraulic disturbance, that is essentially consistent with the results of MWD. Shannon index and Simpson index reflect the diversity of the community (Gong et al., 2019; Li et al., 2022), and there is a slight difference in biodiversity between the components.

3.4.2. Changes in bacterial community structure

To examine the effects of MPs on bacterial community composition in amended sediment, the response ratios of bacteria were analyzed successively for each phylum and class level based on their relative abundance. As shown in Fig. 5(a). The top ten dominant bacterial phyla, identified from the bacterial communities of 6 sediment samples, with the highest relative abundance were Proteobacteria, Chloroflexi, Acidobacteria, Actinobacteria, Bacteroidetes, Firmicutes, Myxococcota, Patescibacteria, Gemmatimonadetes, unclassified_k_norank_d_Bacteria. Among the different samples, Proteobacteria (18.68–27.52 %), Chloroflexi (16.79–23.2 %), and Acidobacteriota (16.77–20.76 %) were

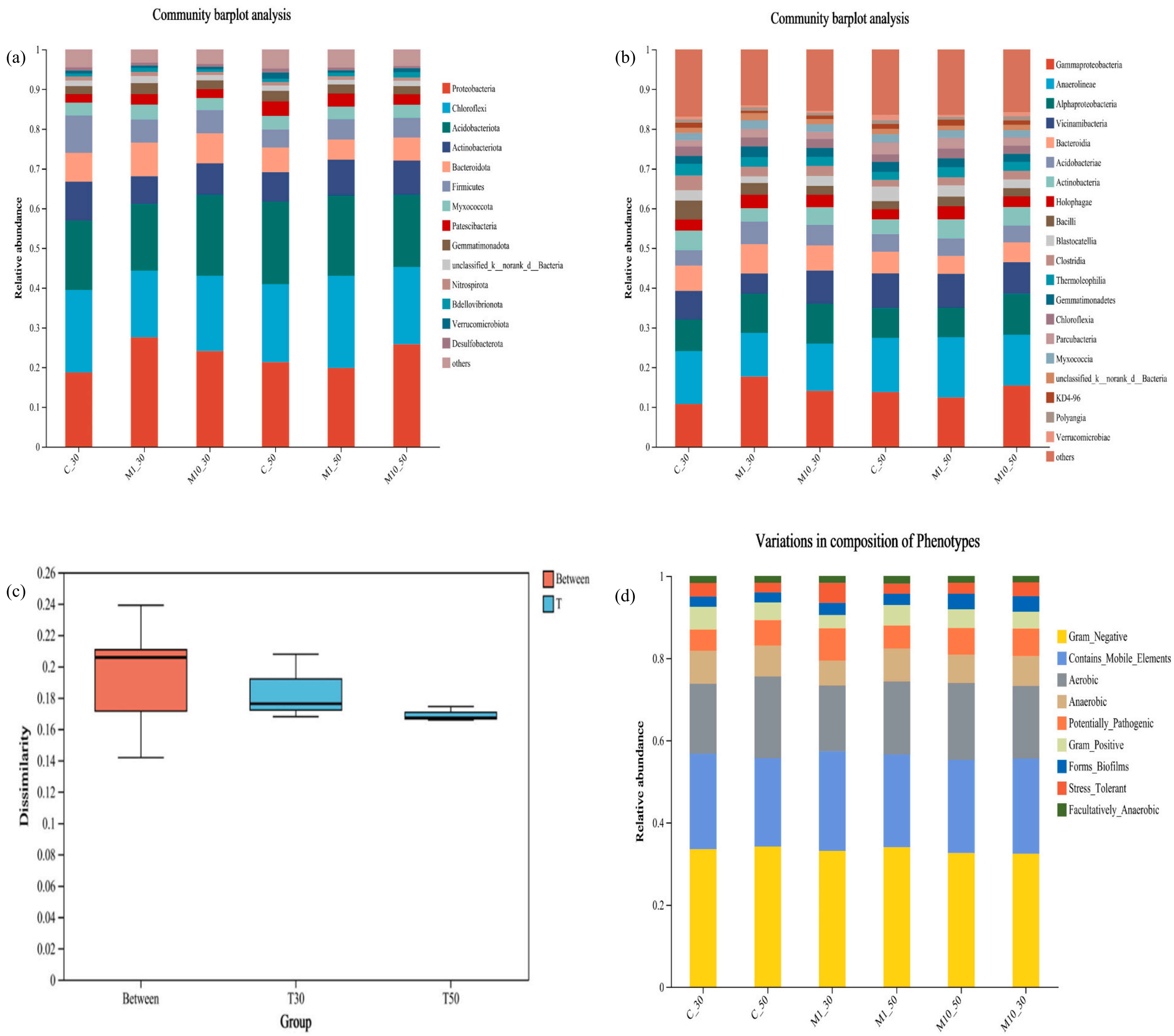

Fig. 5. The relative abundances of bacterial community at phylum level (a) and class level (b) on different treatment sediment under disturbance; Box plot (c) of microbial diversity on 30 d and 50 d disturbance; Bacterial community phenotypes in (d) different treatment sediment under disturbance (C_30, C_50: control samples under 30 and 50 d of disturbance; M1_30, M1_50: added 1%PS-MPs samples under 30 and 50 d of disturbance; M10_30, M10_50: added 1%PS-MPs samples under 30 and 50 d of disturbance).

the three dominant bacteria, accounting for >56 % of the total bacterial relative abundance in each sample. Many studies reported that Proteobacteria tend to dominate soil communities (Qin et al., 2022), Wang et al. (2023) found that the majority of sequences in the soil samples, affected by LDPE, belonged to the phyla Proteobacteria and Actinobacteriota. As can be seen from the box plot (Fig. 5(c)), there was a slight difference in microbial diversity between sediments with different PS-MP content under disturbance, especially at the end of the disturbance. PS-MPs with low biodegradability are unlikely to affect sediment microbial community structure directly in a short period, but they can drive the selection of special microbial classes with resistance and potential degradation (Wang et al., 2022; Zhang et al., 2019). Our results were similar to those of other studies, in which the relative abundances of *Proteobacteria* showed general increases with the elevated concentrations of MPs under disturbance. This finding may be attributed to the fact that *Proteobacteria* prefers to colonize in nutrient-rich and low-bulk-density soils (Eilers et al., 2010). The anaerobic phylum, Chloroflexi, is competitive in low-carbon soil conditions and related to carbon sequestration (Lv et al., 2014), relative abundances of Chloroflexi in the MPs added group were reduced due to the increase of OM and oxygen content.

At the class level, the dominant species were also similar across each sediment (Fig. 5(b)). Both control and treatment samples were mainly comprised of Gammaproteobacteria, Anaerolineae, and Alphaproteobacteria except for M1_50. The proportion of Vicinamibacteria was slightly higher in M1_50 (8.52 %) than Alphaproteobacteria (7.43 %). Alphaproteobacteria and Gammaproteobacteria belong to the phylum Proteobacteria, but Alphaproteobacteria has great variability and few common points, Alphaproteobacteria have been reported to be the dominant bacteria in coastal waters (Du et al., 2022; Gómez-Pereira et al., 2012). Gammaproteobacteria can biodegrade plastics by synthesizing hydrolases or participate in the decomposition of complex organic matter (Hou et al., 2021). The experiment found that Gammaproteobacteria were enhanced in the PS-MPs, except for the M1_50 treatment. It has been reported that the addition of aromatic carboxylic acids can promote the growth of Proteobacteria (Ma et al., 2020), so the increase in Proteobacteria may be attributed to benzene in polystyrene in PS-treated, which is consistent with previous studies (Zhu et al., 2022). Among the treatments in each group, the relative abundance of Gammaproteobacteria in M1_30 was higher, while the relative abundance of Gammaproteobacteria in M1_50 was lower than that of the control group. The reason why the relative abundance of these gates differs in the PS group may be that the impact of MPs on sediments may not only be related to the nature of MPs but also depend on MPs dose, as well as other environmental factors. In addition, Anaerolineae belong to the phylum Chloroflexi and tend to survive in nutrient-rich and anoxic environments. In the early stages of disturbance, relative abundances of Anaerolineae in the control group were higher than that in the MPs group. As the disturbance frequency decreases, the anaerobic bacteria content on day 50 is higher than that in the control group with the increase in organic matter content.

The phenotypic shift of the sediment bacterial community was analyzed by using BugBase. Consistent with many previous results, MPs exposure increased the relative content of biofilm-forming bacteria in sediments, as shown in Fig. 5(d). Besides, PS decreased the relative abundance of aerobic bacteria and increased the abundance of anaerobic bacteria. These results agree with those reported by Zhu et al. (2022) that both PVC and PS decreased the aerobic bacterial community relative abundance. It is worth noting that exposure to PS-MPs significantly increased the relative abundance of Potentially_Pathogenic within 30 d. Potentially_Pathogenic enrichment on MPs may pose ecological risks. Nevertheless, the relative abundance of Potentially_Pathogenic in all components decreased on day 50, suggesting that the disturbance may be not conducive to the growth of relative microorganisms.

Additionally, some studies have shown that MPs may affect ecological cycles through the colonization of specific microorganisms on their surfaces (Huang et al., 2021). Based on high-throughput sequencing results, we used the Functional Annotation of Prokaryotic Taxa (FAPROTAX) to predict functions related to biochemical cycle processes for analyzing whether MPs will affect the functional diversity of ecosystems and the role of disturbance in this (Fig. 6). Comparing the abundances of the predicted functions, found that the addition of PS-MPs in sediment showed significant changes in genes related to the nitrogen cycle, including nitrogen_respiration, nitrate_respiration, and nitrate_reduction. Nitration is a series of processes that oxidize N from NH_3 or NH_4^+ to NO_3^- under aerobic conditions. The remarkable presence of nitrifying genes in PS-MP sediments implies that PS-MPs may affect the nitrification process in sediments, which is consistent with the previous analysis on urease activity. The gene abundances of methylotrophy and methanol oxidation in sediment samples were lower than those in the MPs group at 30th d. Nothing, the gene abundances of aromatic_compound_degradation in M1_30 was lower than those in M10_30 and control groups, it is speculated that compared with adding 10%*w*/w PS-MPs, adding 1 % w/w PS-MPs will have a greater impact on the sediment environment, which is not conducive to microorganisms with aromatic_compound_degradation growth. However, these gaps narrowed significantly after the 50 d disturbance, demonstrating that

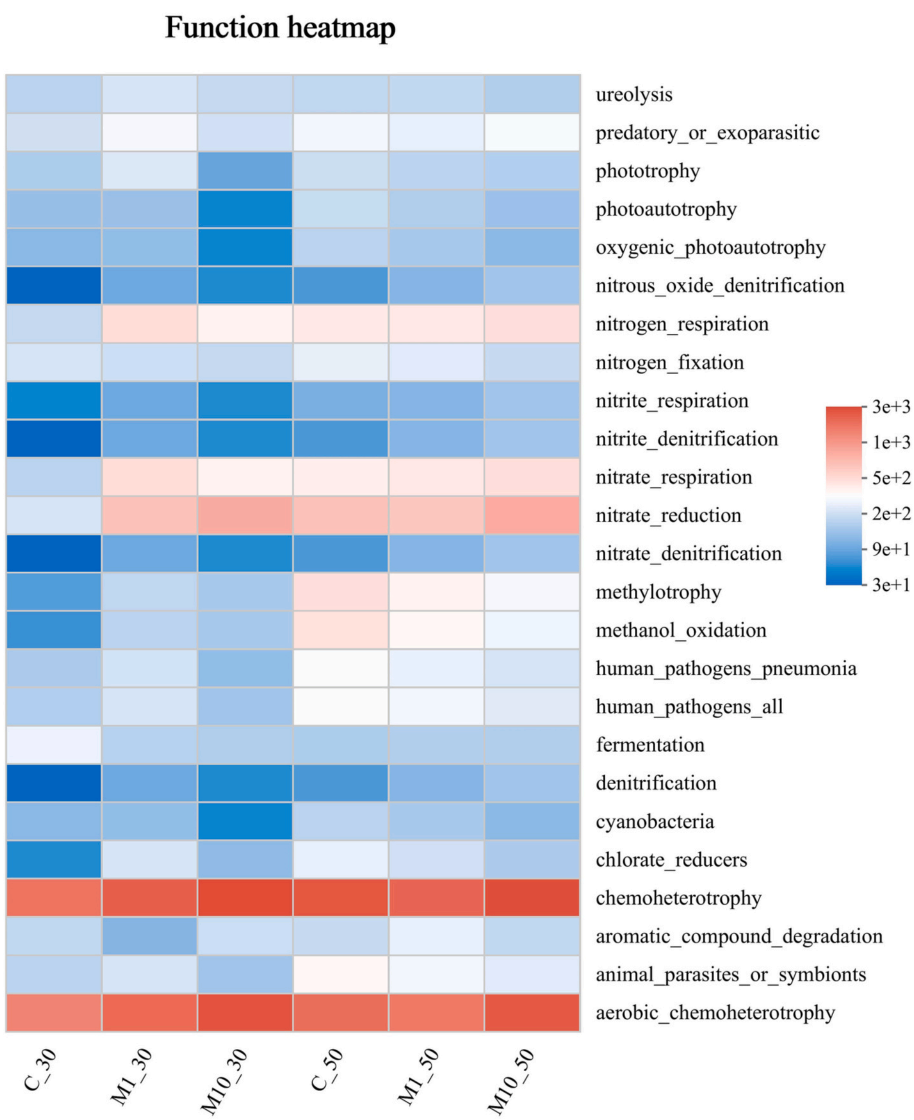

Fig. 6. FAPROTAX clustering heatmap among different treatment groups.

hydraulic disturbance can affect the microbial communities of the sediments containing MPs contaminants by enhancing dissolved oxygen and regulating the interaction between sediment and MPs.

3.5. Effects of environmental factors on sediment microbial communities with MPs

The MPs properties and environmental conditions could both affect bacterial community composition and structure in sediment samples. In our study, redundancy analysis (RDA) was performed to explore the correlation between the top 5 phyla disturbances with different MP treatments and environmental factors. As shown in Fig. 7(a), all sediment samples were not clustered at the phylum level that occurred in each treatment. According to the disturbance time, the 50 d treatment was slightly more clustered than the 30 d treatment, which may be due to the disturbance further regulating the physicochemical properties of the environment in the treatment groups, such as oxygen content, and the differences caused by MPs between the treatments. Niu et al. (2021) found that plastic-degrading microbial communities on microplastic surfaces in river sediments were significantly correlated with TOC and dissolved oxygen (DO). Based on the dosage of PS-MPs added, the control groups and the low-MPs concentration groups dispersed relatively widely, while the higher initial PS-MPs concentration generally clustered together at different disturbance stages. The effects of changes in bacterial community composition and structure in sediment samples with different PS amounts on sediment environmental factors are not the same and also depend on the nature of MPs and other environmental factors which was accordant with the previous studies. Besides, the results showed that the two axes explained 89.35 % of the classification information, suggesting the obvious correlations between bacterial taxonomic composition at the phylum level and environmental factors. Among the various factors, MWD was the most significant factor affected by the bacterial community of the PS sample, especially for samples disturbed at 50 d, followed by urease, OM, MBC, and sucrase. This might be because hydraulic disturbance promotes aggregation between PS and sediment, changing the particle size of the aggregates. Hou et al. (2021) confirmed size of aggregates was a dominant factor influencing the relationships between sediment aggregate properties and bacterial community. This further implies that hydraulic disturbance can alter the physicochemical properties of sediments contaminated by MPs.

MPs can also affect microbial communities indirectly via changing sediment properties. In our present study, correlation analysis shows close correlations between sediment parameters and the relative abundance of specific microbial members. The correlation heatmap visually shows the relationship between the relative abundance of different bacteria in the sediment and the physical-chemical properties of the sediment treated in different PS amounts, through the Spearman rank correlation coefficient (Fig. 7(b)). Using this, the correlation between microorganisms and environmental factors is evaluated. It was observed that the abundance of dominant Nitrospirota was negatively correlated with sediment OM ($0.01 < p \leq 0.05$). The abundance of dominant Bacteroidota was negatively correlated with urease ($0.01 < p \leq 0.05$). Consistent with previous studies, the increase of total organic carbon (TOC) can indirectly improve the abundance of Bacteroidota and decrease urease activity (Fang et al., 2023), which results in a negative correlation between OM and Nitrospirota by altering urease activity. Besides, the abundance of dominant unclassified_k__norank_d__Bacteria was also negatively correlated with MWD ($0.01 < p \leq 0.05$). Also, the abundance of dominant Bacteroidota was a significant positive correlation with MBC ($0.001 < P \leq 0.01$), and the abundance of dominant Alphaproteobacteria was a positive correlation with sucrase at the class level as illustrated in Fig. S5. The above results showed that there were positive and negative correlations between environmental factors and the abundance of different flora. The decrease of OM, urease, MWD, and other environmental factors could increase the abundance of resistant bacteria positively correlated with them or decrease the abundance of sensitive bacteria negatively correlated with them. These further indicate that MPs can have positive or negative effects on certain species by changing environmental factors, thereby affecting the entire sediment bacterial community structure (Li et al., 2022; Song et al., 2018).

4. Conclusions

Based on high-throughput sequencing and comparative analysis of sediment's physicochemical properties, the effects of hydraulic disturbance on sediment environment with different dosages of PS-MPs were studied. Compared with relatively low PS content (1w/w%), high PS content (10w/w%) reduces community richness and diversity. Although the importance of microplastic was not as high as expected within the concentration range of this study, MPs have a greater influence on the sediment environment involving disturbance in contrast to static culture, and the variation range of various environmental factors is larger. These variations are the driving forces that promote the differentiation and succession of bacterial communities. Also, this research confirmed that the disturbance narrowed the difference in microbial functional genes between the control group and the MPs-added groups by up to 10

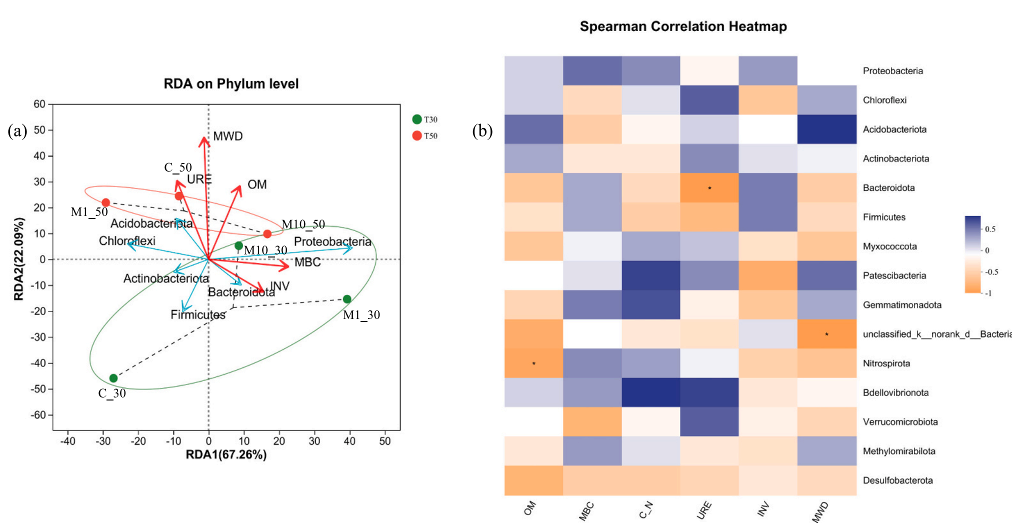

Fig. 7. (a) Redundancy analysis of the relationship between bacterial communities and environmental factors; (b) Heatmap of correlation analysis between microbial function and physicochemical index (*statistically difference at $P < 0.05$, **significant statistical difference at $P < 0.01$, ***a very significant statistical difference at $P < 0.001$).

times. In this experiment, an intermittent disturbance environment was set up for the first time to simulate the actual river sediment environment, providing new insights into the changes in the ecological environment metabolism of MPs-polluted sediments, and deepening the understanding of the role of MPs in sediments in the natural environment. Future studies should pay more attention to the combination with the actual sediment environment and diversified environmental factors.

CRediT authorship contribution statement

Wenjuan He: Writing – original draft, Validation, Methodology, Investigation, Formal analysis, Data curation. **Jinhui Huang:** Writing – review & editing, Supervision, Methodology, Funding acquisition, Conceptualization. **Si Liu:** Writing – review & editing, Validation, Formal analysis, Conceptualization. **Hanbo Yu:** Validation, Formal analysis, Conceptualization. **Enjie Li:** Methodology, Investigation. **Wei Zhang:** Validation, Formal analysis. **Kaixin Yi:** Visualization, Investigation. **Chenyu Zhang:** Writing – review & editing, Software. **Haoliang Pang:** Methodology, Formal analysis. **Xiaofei Tan:** Writing – review & editing, Formal analysis.

Declaration of competing interest

The authors declare that they have no potential competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The authors do not have permission to share data.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2024.171350.

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