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The effects of diverse microplastics on adzuki bean (*Vigna angularis*) growth and physiologic properties

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Abstract Globally, microplastic pollution of soil ecosystems poses a major risk. The early studies found that the impact of microplastics on different plants could vary depending on the type of microplastic, the mass concentration or the plant species. This study investigated the effect of 3 mass concentrations (0.1%, 1%, and 2.5%) and 3 types of microplastics (PE MPs, PLA MPs, and PVC MPs) on adzuki bean biomass, root traits, Chlorophyll content and antioxidant enzymes. According to our findings, all

microplastics had an impact on biomass, but PLA MPs had the strongest inhibitory effect. The high mass concentration of microplastics had a significant influence on chlorophyll content. Adzuki beans exhibited varying degrees of damage upon exposure to microplastics, but they were able to withstand the oxidative stress brought on by PE MPs by increasing the activity of antioxidant enzymes (SOD and POD). Comparing the adverse effects of PE MPs on adzuki beans to those of PLA MPs and PVC MPs, principal component analysis and membership function value analysis revealed that the former had fewer impacts. Disparities in the observed effects may be attributed to variations in the properties of microplastics. Subsequent investigations into the mechanisms underlying microplastic toxicity need a more comprehensive exploration.

Supplementary Information The online version contains supplementary material available at <https://doi.org/10.1007/s10653-024-02157-2>.

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Introduction

Plastic items have been used extensively globally since their large-scale commercial production began in the middle of the twentieth century because of their low cost, ease of processing, and versatility. Approximately 90% of the demand for hundreds of plastic materials is met by polyethylene (PE), polypropylene (PP), polyvinyl chloride (PVC), polystyrene (PS), and

polyethylene glycol terephthalate (PET) (Andrady & Neal, 2009). It is estimated that the yearly output of disposable plastic items worldwide may exceed 12.01 billion tons, of which only 12% are recycled, 70% are burned, and over 9% are thrown into the soil, air, and oceans (An et al., 2023; Prata et al., 2019). By the middle of the century, landfills and natural ecosystems will have amassed around 12,000 metric tons of plastic debris due to the continuous large-scale manufacture of plastic waste and negligent disposal of it, which will cause serious ecological problems (Geyer et al., 2017). Larger plastic waste usually breaks down into microplastics (MPs, particles < 5 mm in size) due to their resistance to degradability (Thompson et al., 2004).

MPs are pervasive pollutants in various environmental compartments, including aquatic and terrestrial ecosystems, which raise significant concerns regarding their potential ecological and toxicological impacts (Sethia et al., 2024). The toxicity of MPs to aquatic organisms has been extensively explored (Reyes & Medina, 2024; Xue et al., 2024). Study found that the amount of MPs released on land each year is 4–23 times more than that in the ocean (Horton et al., 2017). Worldwide, the stock of MPs in agricultural soils could be 1.5–6.6 million tons (Kedzierski et al., 2023). Numerous studies have investigated their potential toxic effects in terrestrial ecosystems. Exposure of MPs to the environment can affect soil physicochemical properties (soil bulk weight, water-holding capacity, pH), plant growth and soil biodiversity (Machado et al., 2018; Urbina et al., 2020; Yang et al., 2021; Zhang et al., 2019). Plants, being an essential component of terrestrial ecosystems, are inevitably exposed to MPs that exist in the soil. Investigations have demonstrated that MPs are harmful to plants. For instance, plant roots have the capacity to absorb and transport sub-micron polystyrene (PS; size 1 μm) microplastics to above-ground components (Lian et al., 2020). It was found that PS and polytetrafluoroethylene (PTFE) microplastics cause mechanical damage to rice roots, resulting in the generation of reactive oxygen species (ROS) (Dong et al., 2020). The ROS accumulation in plant cell leads to impairments of plant growth, photosynthesis, and biochemical processes (Choudhury et al., 2013). The effects of MPs on plant growth have been studied in different species, mainly including lettuce (*Lactuca sativa*) (Zhang et al., 2024a, 2024b), wheat (*Triticum*

aestivum) (Han et al., 2024; Wang et al., 2024), and maize (*Zea mays*) (Yang et al., 2024), etc. The phytotoxicity of MPs is clearly plant species-dependent (Gong et al., 2021). Additionally, during the manufacturing process of MPs, different additives like plasticizers and antioxidants are added to enhance its performance. However, because of the weak link between the additives and the base polymer, these chemicals can be easily removed, which has hazardous effects on plants (Lee et al., 2022; Li et al., 2022). The alteration of soil environment by microplastics can indirectly impact plant (Shang, 2023; Wang et al., 2020b).

Fabaceae is one of plant species commonly recommended for toxicity tests (Agency, 2012). Adzuki bean (*Vigna angularis*), an important food legume, is grown in more than 30 countries (Wang et al., 2020a). To ensure food safety and protect human health, it is imperative to understand how microplastics affect adzuki bean growth. In the investigation, we used a variety of widely used microplastics, including polyethylene microplastics (PE MPs), polylactic acid microplastics (PLA MPs), and polyvinyl chloride microplastics (PVC MPs). The objectives of our study were: (i) to determine the impact of MPs on growth parameters such as biomass, root traits, and chlorophyll content in adzuki beans, (ii) to investigate the oxidative damage of different MPs on adzuki bean, and (iii) to evaluate the phytotoxicity of different types and mass concentrations of MPs by membership function value (MFV).

Materials and methods

Soils and microplastics preparation

Adzuki bean seeds for testing were provided by the Water Conservancy Institute of Hebei Agricultural University. The microplastics used were purchased from Dongguan Huachuang Plasticizing Material Co., Ltd., China. Soils were sampled from the farmland in Dagudian village, Mancheng district, Baoding, China (115°34'E, 38°87'N). The soil's characteristics were as follows: pH 7.89, organic matter content 19.70 g/kg, total nitrogen content 0.60 g/kg, available potassium content 40.33 mg/kg, and accessible phosphorus content 12.84 mg/kg. The soil was air-dried and sieved to a 5-mm size.

Experimental design

Three mass concentrations (0.1%, 1%, and 2.5%) and 3 types of MPs (PE MPs, PLA MPs, and PVC MPs) were included in the completely randomized block group design of the study. There were ten treatments in total, with 4 replicates for each treatment (Fig. 1). Soil without microplastic addition was denoted by CK, while soil containing 0.1% mass concentration of added PE MPs was indicated by 0.1% PE MPs; and so on. To formulate the MPs utilized in the test, 30 mesh and 100 mesh MPs were combined in a mass ratio of 6:4. Following a thorough mixing of the MPs with the soil in accordance with the intended dose, the soil was packed into pots weighing 0.5 kg each. In order to ensure sufficient nutrients, a nutrient solution containing the following was given to the soil once as a base fertilizer: $(\text{NH}_4)_2\text{SO}_4$; 944 mg/kg, KH_2PO_4 ; 220 mg/kg, K_2SO_4 ; 335 mg/kg, CaCl_2 ; 126 mg/kg, $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$; 43 mg/kg, $\text{MnSO}_4 \cdot 4\text{H}_2\text{O}$; 6.7 mg/kg, $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$; 10 mg/kg, and $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$; 2.0 mg/kg (Pu et al., 2024).

The pot experiment was conducted in semi-controlled climate conditions in a greenhouse of Hebei Agricultural University (115°45'E, 38°82'N). The light intensity was 9000 Lux throughout the reproductive period, with continuous lighting for 14 h per day, the temperature ranged from 25 °C to 27 °C, while the relative humidity of the air was maintained between 44 and 55%.

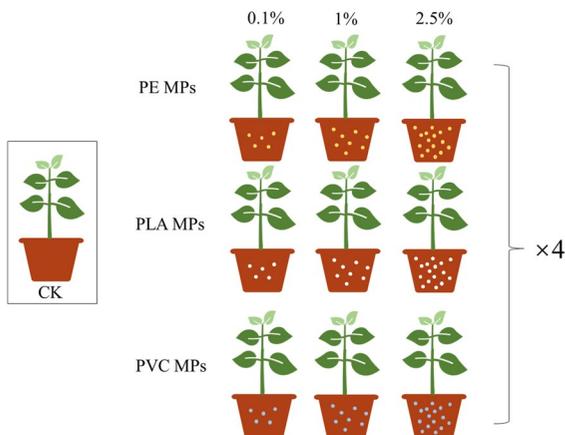


Fig. 1 Schematic diagram of microplastic-stressed *V. angularis* culture experiment

Following a 20-min soaking period in a 30% H_2O_2 solution and several rinses with distilled water to eliminate any residual liquid, the sterilized adzuki bean seeds were transferred to a culture dish and allowed to grow using distilled water. After that, the dish was kept in a dark area for one to 2 days to encourage germination. One plant per pot was left after 3 carefully chosen, germinated seeds were placed in the soil, nurtured for a week, and then interplanted. To maintain proper soil moisture levels, plants were frequently irrigated during growth. One month following seeding, shoots and roots were harvested for analysis (Fig. S1).

Sample analysis

Biomass measurements

The plant's aboveground portion was cut with scissors to remove it from the root, and then it was cleaned, dried, and baked at 70 °C to a consistent weight. Similar procedure was carried out on the root following the completion of the morphological examination.

Root morphology measurements

In a steel mesh frame, root samples were cleaned with distilled water after being rinsed with tap water to remove any adhered soil. Following washing, the adzuki bean's root nodules were counted. Following the uniform distribution of the roots in the transparent tray filled with distilled water, the tray was scanned using an Epson Perfection V600 Photo scanner. With the use of WinRHIZO, the scanned pictures were examined, and measurements were made of the root morphology, including root length and diameter (mm).

Chlorophyll content measurements

Weighed 0.1 g of fresh leaves crushed with liquid nitrogen in a centrifuge tube, added 5 mL of 80% acetone to completely submerge the leaves, placed in a dark environment for 24 h. When the leaves in the tube completely faded, the supernatant was taken and the absorbance values at wavelengths of 663 nm (chlorophyll *a*), 645 nm (chlorophyll *b*), and 470 nm (carotenoids) were determined using an enzyme-linked immunosorbent assay (ELISA). The

calculation formula are as follows (Lichtenthaler & Wellburn, 1983):

$$\text{Chlorophyll } a : \text{Chla} = 12.21 \times A_{663} - 2.81 \times A_{645} \quad (1)$$

$$\text{Chlorophyll } b : \text{Chlb} = 20.13 \times A_{645} - 5.03 \times A_{663} \quad (2)$$

$$\text{Carotenoids: Car} = (1000 \times A_{470} - 3.27 \times \text{Chla} - 104 \times \text{Chlb}) / 229 \quad (3)$$

$$E_x = \sum_{y=1}^n (L_{xy} \times C_{xy}) \quad (4)$$

$$W_x = E_x / \sum E_x \quad (5)$$

In the formula, L_{xy} is the loading of the index on the y -th principal component, C_{xy} is the contribu-

Antioxidant enzymes measurements

Malondialdehyde (MDA) content was measured using the thiobarbituric acid colorimetric method. Superoxide dismutase (SOD) activity was measured with the nitrogen blue tetrazolium method. Catalase (CAT) activity was measured via the ultraviolet absorption method, and Peroxidase (POD) activity was measured with the guaiacol method. Ascorbate peroxidase (APX) activity was measured by spectrophotometric assay. Soluble protein (SP) content was determined by a Coomassie Brilliant Blue G-250 reaction using the enzyme solution (Bradford, 1976).

Statistical analysis

IBM SPSS Statistics 26 and Excel 2019 were used to process and perform statistical analyses on all the data. The significant differences between the control and treatment groups (LSD) were found using a one-way ANOVA. The primary and interaction effects of microplastic type and mass concentration on adzuki bean growth were examined using 2-way ANOVA. The Origin 2023b was used to draw the graphs. To evaluate the effects of MPs on *V. angularis*, the membership function values (MFV) was calculated using principal component weighted analysis as calculated in previous studies (Li et al., 2023; Zhang et al., 2022a). Weight (W_i) is the relative importance of an indicator in the overall evaluation. On the basis of principal component analysis, the weights were determined by calculating the effect of MPs addition on plant's each indicator according to the loading (L) and contribution rate (C) of each indicator:

tion rate of the y -th principal component, n is the number of extracted principal components, and W_x is each weight of the indicator. MFV calculation in fuzzy mathematics is as follows:

$$M_y = (X_y - X_{\min}) / (X_{\max} - X_{\min}) \quad (6)$$

where M_y is the membership function value of the index y , X_y is the measured value of the index y , X_{\min} and X_{\max} is the minimum and maximum values of the y -index measurements. The final MFV of comprehensive evaluation:

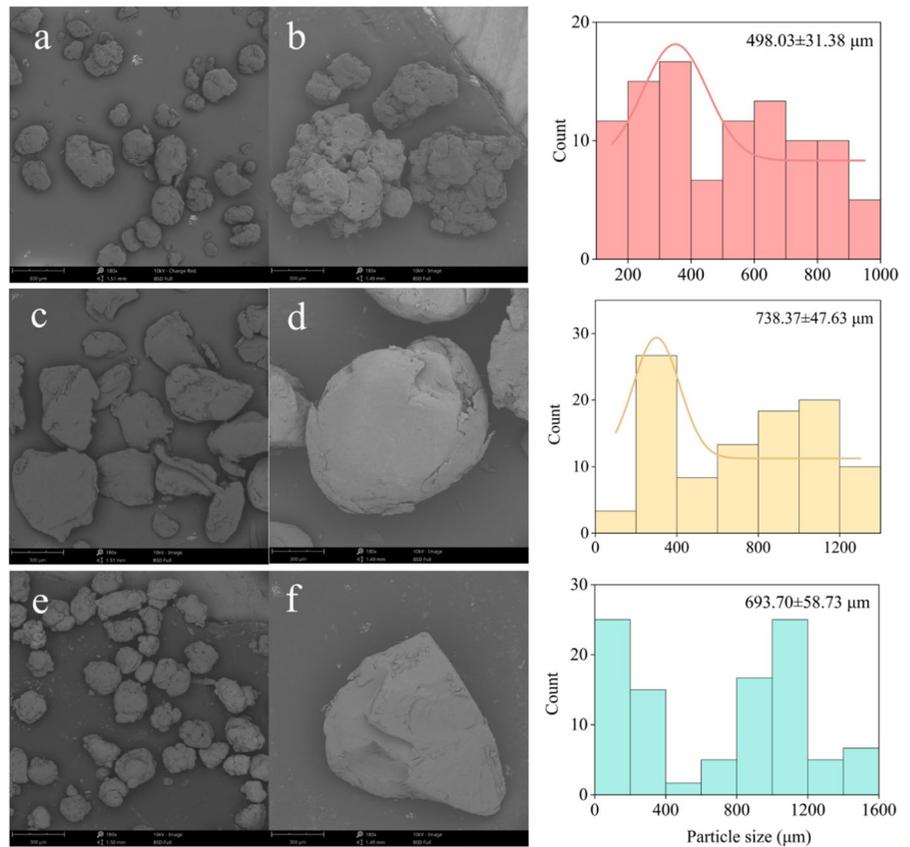
$$MFV = W_x \times M_x \quad (7)$$

Results

Morphology characterization and particle size of MPs

Scanning electron microscopy (SEM) was used to determine the size and shape. The average particle size of PE MPs, PLA MPs, and PVC MPs was $498.03 \pm 31.38 \mu\text{m}$, $738.37 \pm 47.63 \mu\text{m}$, and $693.70 \pm 58.73 \mu\text{m}$, respectively (Fig. 2). Every plastic particle had an asymmetrical shape. Whereas PE MPs and PLA MPs were comparatively smooth (Fig. 2a, c), the surfaces of PVC MPs with a particle size of 100 mesh had more pores and cracks (Fig. 2e). PVC MPs had sharp edges for large size (30 mesh) microplastic pellets, implying that they had a greater effect than the other 2 types (Fig. 2b, d, and f).

Fig. 2 Microplastic characterization. Scanning electron microscope (SEM) images of 100 mesh **a, c, e** and 30 mesh **(b, d, f)** PE MPs, PLA MPs and PVC MPs and size distribution after proportional mixing



Biomass of *V. angularis*

According to the type or mass concentration of MPs, we discovered in our research that the biomass of *V. angularis* varied (Fig. 3). The shoot biomass of *V. angularis* was considerably reduced by 0.1% PLA MPs, 1% PLA MPs, 0.1% PVC MPs, and 2.5% PVC MPs when compared to CK; however, PE MPs had no noticeable impact on shoot biomass. PLA MPs produced the least amount of biomass in the shoots among the treatments at 0.1% and 1% mass concentration of the 3 microplastics. PLA MPs on root biomass remained the microplastic that had the biggest reduction (Fig. 3).

Root traits of *V. angularis*

The findings of 2-way ANOVA indicated that root length and root diameter had significant relationships with mass concentration, type, and their interaction ($P < 0.05$) (Table 1). Compared to CK, root

diameter considerably increased at 1% PE MPs, whereas root length was dramatically reduced at 0.1% PLA MPs, 0.1% PVC MPs, 1% PLA MPs, and 1% PVC MPs by 35.88%, 37%, 37.69%, and 46.30%, respectively. Taking into account the variations in the 3 types of microplastics, the frequency of nodules was decreased by 2.5% PVC MPs; at 0.1% and 1% mass concentration, PE MPs considerably enhanced root length (Table 1).

Chlorophyll content of *V. angularis*

The contents of Chl_a and Car showed a decreasing tendency with increasing mass concentration from 0.1% to 2.5% when compared to CK. 2.5% PVC MPs were found to have the lowest Car content, while 2.5% PLA MPs had the lowest level of Chl_a content (Fig. 4). Following treatment with 1% PVC MPs and 2.5% PE MPs, there was a significant increase in Chl_b relative to CK (Fig. 4).

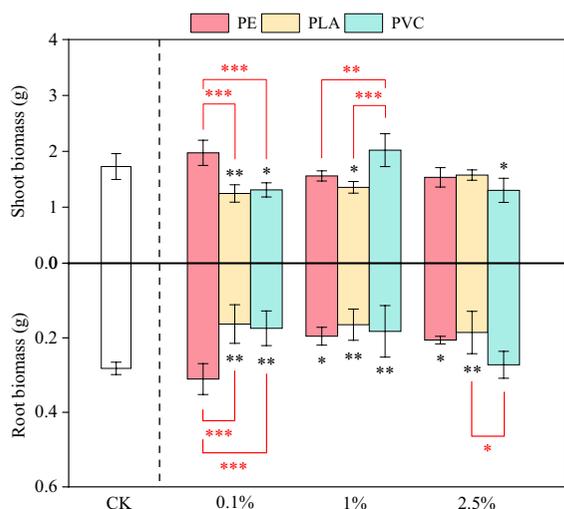


Fig. 3 Effects of microplastics on shoot and root biomass of *V. angularis*. CK represents control plants without any treatment; Data are the means of 4 replicates (mean \pm SD). Significant differences between control and MPs treatments are marked with * ($p < 0.05$), ** ($p < 0.01$) and *** ($p < 0.001$). For a given concentration mass, a significant difference among different types of microplastics is marked with red * ($p < 0.05$), ** ($p < 0.01$) and *** ($p < 0.001$) above the connection line

Antioxidant enzymes of *V. angularis*

In *V. angularis* leaves, a gradual increase in MDA content was noted within the mass concentration range of this study. The MDA content reached its maximum level when 2.5% PVC MPs were applied, and it was 1.07 times greater than CK. When 3 microplastics were compared, it was discovered that, at the same mass concentrations (1% and 2.5%), PVC MPs treatments were considerably higher than the other 2 MPs treatments (Fig. 5). With increasing mass concentration, the SOD and POD activities at the treatment of PE MPs increased. At 2.5% compared to CK, the increases in SOD and POD activities were 25.69% and 49.48%, respectively (Fig. 5). Two-way ANOVA findings indicated that, with the exception of APX, mass concentration was significant for all antioxidant enzyme activities ($p < 0.05$) (Table S1). In comparison to CK, the increases in CAT activity with 1% PLA MPs and 2.5% PVC MPs treatments were 31.6% and 45.2%, respectively. CAT activity was substantially higher at PVC MPs treatment than at PLA MPs and PE MPs by 33.51% and 1.2 times, respectively, when the mass concentration was the same (2.5%) (Fig. 5). On the other hand, SP content treated with various kinds of microplastics showed the opposite variations. PE MPs treatment resulted in a much

Table 1 Effects of microplastics on root traits of *V. angularis*

Mass concentration (%)	Microplastic type	Number of nodules per plant	Root length (mm)	Root diameter (mm)
CK	–	9.00 \pm 1.00	2391.97 \pm 203.84	0.46 \pm 0.16a
0.1%	PE	1.33 \pm 1.03	3006.24 \pm 757.44a	0.44 \pm 0.07
0.1%	PLA	2.00 \pm 1.15	1533.69 \pm 297.90b*	0.40 \pm 0.05
0.1%	PVC	2.33 \pm 0.88	1507.01 \pm 170.44b*	0.42 \pm 0.02
1%	PE	2.33 \pm 1.86	2184.39 \pm 458.31a	0.64 \pm 0.13a*
1%	PLA	8.67 \pm 3.53	1284.37 \pm 64.37b**	0.38 \pm 0.02b
1%	PVC	2.00 \pm 0.58	1490.36 \pm 616.88ab*	0.42 \pm 0.02b
2.5%	PE	14.00 \pm 7.09a	1831.04 \pm 346.99b	0.37 \pm 0.08
2.5%	PLA	11.33 \pm 2.60a	3198.23 \pm 463.13a*	0.41 \pm 0.05
2.5%	PVC	3.00 \pm 1.53b	1788.21 \pm 707.48b	0.40 \pm 0.02
Mass concentration (DF=2)		0.015	0.045	0.034
Microplastic type (DF=2)		0.136	0.015	0.019
Mass concentration*Microplastic type (DF=5)		0.252	0.001	0.005

Data are the means of 4 replicates (mean \pm SD). Significant differences between control and MPs treatments are marked with * ($p < 0.05$), ** ($p < 0.01$) and *** ($p < 0.001$). The lowercase letters indicate significant differences ($p < 0.05$) between different types of microplastics at the same mass concentration

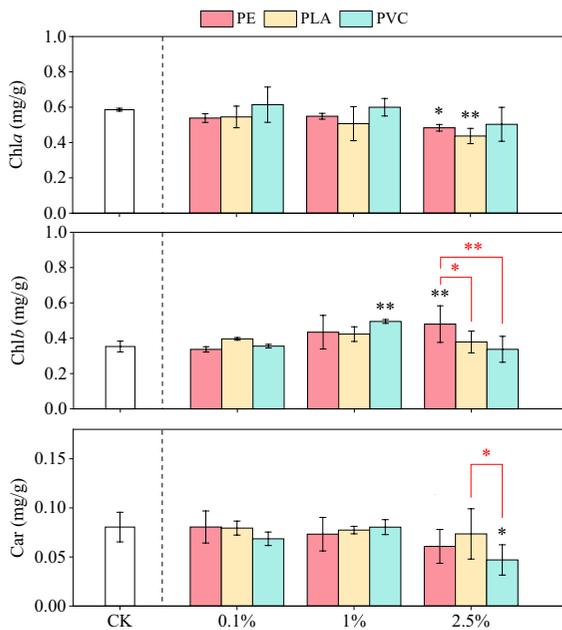


Fig. 4 Effects of microplastics on photosynthetic parameters of *V. angularis*. CK represents control plants without any treatment; Data are the means of 4 replicates (mean ± SD). Significant differences between control and MPs treatments are marked with * ($p < 0.05$), ** ($p < 0.01$) and *** ($p < 0.001$). For a given concentration mass, a significant difference among different types of microplastics is marked with red * ($p < 0.05$), ** ($p < 0.01$) and *** ($p < 0.001$) above the connection line

higher SP content than CK, whereas PLA MPs and PVC MPs treatment resulted in a significantly lower SP content (Fig. 5).

Evaluation of integrated effect of MPs in *V. angularis*

In this study, we divided the indicators into 2 major categories, growth and physiology, and calculated the MFV separately. Taking the growth indicators as an example, the data were distinguished into 4 principal components (PC), PC1, PC2, PC3 and PC4, with eigenvalues greater than 1 selected. The cumulative contribution rate of the first 4 PCs was 72.16% (Table S2). Results of MFV and PCA were used to evaluate the response of indicators to microplastics and distinguished differences in the response of adzuki bean to different microplastics. And we found that 1% PE MPs had the highest MFV followed by 0.1% PE MPs and 1% PVC MPs, whereas the MFV of 2.5% PVC MPs was lower (Table 2). A higher MFV indicated a lower toxicity of MPs on plant growth.

For physiological indicators, the maximum membership value was treated with PE MPs (Table 2), indicating that PE MPs increased antioxidant enzyme activities in response to oxidative stress. In conclusion, the toxic effects of PE MPs on adzuki bean were less than those of PLA MPs and PVC MPs.

Discussion

Influences of MPs on plant growth

The impact of the surrounding environment on plant growth and development can be observed through the change of biomass. All microplastics, with the exception of PE MPs, were observed to diminish shoot biomass (Fig. 3). In line with the results of 98.6 and 26.5 μm PE MPs that produced minimal effects on plant biomass (Meng et al., 2021; Yu et al., 2022). On the other hand, it was clear that microplastics inhibited root biomass (Fig. 3). According to Zang et al (2020), the amount of MPs introduced to the soil significantly influenced the amount of response that 2 popular types of MPs (PVC and PE, 1 μm) had on wheat shoot and root biomass. PLA MPs with different mass concentration (1%, 5%, and 10%, 74 μm) resulted significant reductions in alfalfa root biomass by 25.6%, 33.7%, and 59.3%, respectively (Wang, 2022). A decrease in biomass accumulation during the plant’s growth stages can be attributed to the impact of microplastics on soil properties altering the soil conditions, which in turn affects plant uptake of water and nutrients (Koskei et al., 2021). However, further investigation is required to determine the precise impact mechanisms.

A well-developed root system is essential for plant growth and development, and usually plants adjust their root structure in response to soil environmental stresses. High mass concentration (2.5%) PE MPs and PLA MPs treatments in this study significantly increased the number of nodules in adzuki bean (Table 1). The study by Meng et al (2021) showed that LDPE MPs (1%, 1.5% and 2.5%) in soil stimulates root nodules formation, and this is further supported by a recent studies which reported that the MPs enhances the activity of rhizosphere bacteria which directly improves the soil nitrate content and nitrogen uptake in plants (Kim et al., 2023). In our results, root length was significantly reduced in PLA MPs (0.1%

Fig. 5 Effects of microplastics on antioxidant system of *V. angularis*. CK represents control plants without any treatment; Data are the means of 4 replicates (mean \pm SD). Significant differences between control and MPs treatments are marked with * ($p < 0.05$), ** ($p < 0.01$) and *** ($p < 0.001$). For a given concentration mass, a significant difference among different types of microplastics is marked with red * ($p < 0.05$), ** ($p < 0.01$) and *** ($p < 0.001$) above the connection line

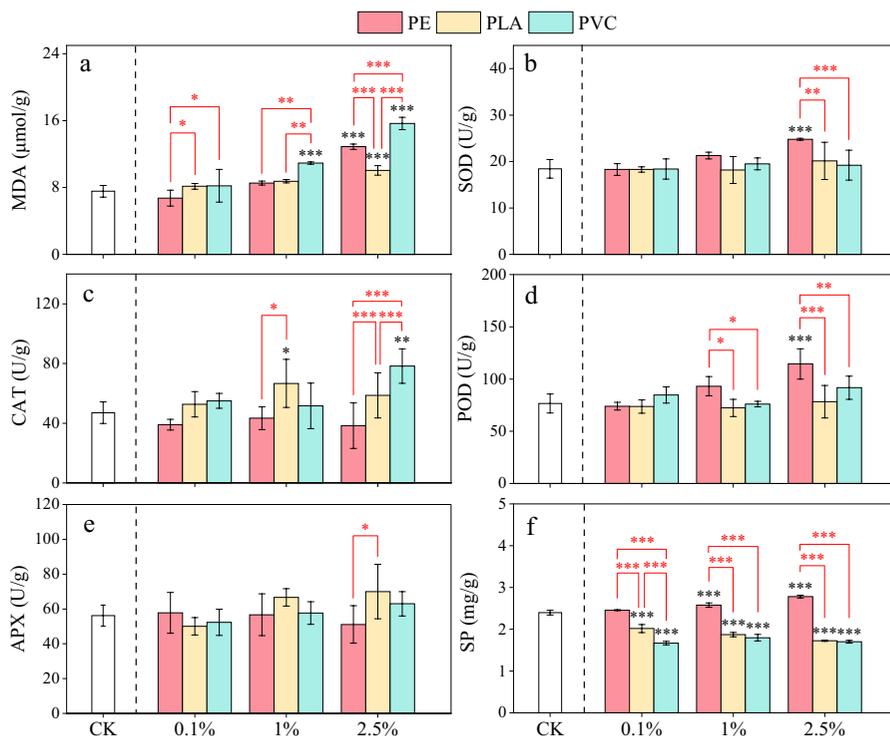


Table 2 The effect of MPs by membership function value (MFV)

Treatment	Index		
	Total	Growth	Physiology
0.1% PE	0.44	0.61	0.46
1% PE	0.58	0.62	0.63
2.5% PE	0.60	0.46	0.96
0.1% PLA	0.34	0.46	0.39
1% PLA	0.38	0.44	0.25
2.5% PLA	0.35	0.46	0.27
0.1% PVC	0.36	0.45	0.37
1% PVC	0.33	0.51	0.40
2.5% PVC	0.35	0.35	0.33

and 1%) and PVC MPs (0.1% and 1%) treated plants with comparison to CK, whereas the effect on root diameter was not significant (Table 1). A study about the effect of microplastics on alfalfa root morphology showed that PLA MPs (5% and 10%, mass concentration) reduced plant root length, highlighting the toxic effect of MPs on plant root system (Wang, 2022). Zhang et al (2023) demonstrated for the first time the enrichment effect of micron-sized PE ($> 10 \mu\text{m}$) in

crops by using the self-developed quantitative characterization method of Eu-MPs and in situ zymography. Although large microplastics (just like the MPs in this study) cannot be absorbed by plant roots, they may be enriched in plant roots (Chen et al., 2023), through contact with the root system. Hence, it is possible to cause mechanical damage to the plant roots, thereby inhibiting root activity and impeding root growth (Li et al., 2022; Rozman et al., 2021).

Influences of MPs on chlorophyll contents

As a crucial photosynthetic pigment, chlorophyll contents changes have a direct effect on photosynthesis. Generally, abiotic stresses could damage plant chlorophyll synthesis (Dalal & Tripathy, 2012). Exposure to the highest concentration of $150 \mu\text{m}$ PE MPs (1500 mg/kg) significantly reduced chlorophyll *a*, and chlorophyll *b* of *Viola philippica* leaves by 27.9% and 23.3% 26.7%, respectively (Niu et al., 2022). Our results are quite parallel to these studies, we found that the chlorophyll content of plants was susceptibly affected by high mass concentration MPs treatments, and different types of MPs have different impacts (Fig. 4). Pignattelli et al (2020) found

significant differences in photosynthetic pigments content of *Lepidium sativum* leaves by exposure to different types of microplastics (PE MPs, PP MPs and PVC MPs, 0.02%, 0.125 mm). Even microplastics with small particle sizes and high mass concentrations in soils, had no significant impact on the photosynthetic pigment content of plant leaves (Yu et al., 2022). There is no uniform conclusion on the effect of microplastics on the photosynthetic properties of plants at present.

Influences of MPs on oxidative damage

External environmental stress causes imbalance in plant ROS levels, once the level of ROS exceeds the antioxidant capacity of plants, oxidative stress occurs, resulting in lipid peroxidation of cell membranes and excessive production of MDA, that cause disruption of the cell membrane system (Zhang et al., 2022b). Thus, the level of MDA content helps to measure the plant oxidative damage induced by the ROS. The addition of microplastics (2.5%) significantly increased the MDA content depending on the types of MPs in Fig. 5, suggesting the accumulation of certain amount of ROS caused the membrane damage. A number of studies have reported similar to our findings (Niu et al., 2022; Shi et al., 2022, 2023).

SOD, CAT and POD are important enzymes in plants for removing oxygen free radicals in cells, and typically plants regulate their production to maintain normal physiological functions (Valavanidis et al., 2006). The promoting effect of microplastics exposure on plant antioxidant enzymes (SOD and POD) activities is noticeable (Li et al., 2020b; Liao et al., 2019; Xu et al., 2021). In this study, SOD and POD activities were significantly higher than that of CK after exposure to 2.5% PE MPs (Fig. 5). This indicated that adzuki bean can resist the oxidative stress caused by PE MPs through improving the activities of antioxidant enzymes (Table 2). In PLA MPs (1%) and PVC MPs (2.5%) treatment groups, CAT activities were higher as compared to CK (Fig. 5), indicating that CAT production under MPs stress may resist oxidative stress. As the main osmotic regulatory substance in plant, variation of SP content might be indicate the impact of external stress on biotic growth and development (Sairam et al., 2008). The results of this study revealed that PE MPs treatments significantly

increased SP content in adzuki bean leaves (Fig. 5), which may be the result of the organism's defense against oxidative stress. Whereas the decrease in SP content under the treatments of PLA MPs and PVC MPs may be due to their possible toxic effects on the plants.

In summary, we hypothesized that PE MPs has a low toxicity compared with the other 2 microplastics. This result may be attributed to the polymerization of PE, which has only carbon and hydrogen atoms (Li et al., 2023). The research used *Cucurbita pepo* L. as model plant to test the toxic effects of microplastics (PP, PE, PVC and PET) found PE as the less toxic material, PVC decreased the indicators to a higher extent compared to the other treatments and was considered as the most toxic material (Colzi et al., 2022). In addition to the structural properties inherent to MPs, it is imperative to consider their environmental implications. Research showed that PE caused minor effect on soil structure (Machado et al., 2019). Further research is needed to investigate the underlying mechanisms, given the variation in experimental conditions.

Conclusion

The results showed that PE MPs, PLA MPs, and PVC MPs affect adzuki bean growth in different ways. Microplasticity effects on plant biomass, root growth, and chlorophyll content are related to microplastic type and mass concentration. High mass concentrations of all MPs in this study caused oxidative damage to *V. angularis*. PE MPs showed the capability of increasing the most of significant variations in enzymes activities, thus considered the less toxic MP. Our study confirmed the potential negative effects of MPs on plant, but given the diversity of MPs and the wide range of soil–plant systems, more research is needed to clarify the mechanisms of MP toxicity and potential threats to human health via global food chain.

Author contributions Author contributions RL, FHS, and XXW designed and performed the experiments. RL analyzed the data and wrote original draft. XMY, HQL, and XXW polished the language of manuscript and revised the manuscript. HQL and XXW provided financial support.

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Data availability Data is provided within the manuscript or supplementary information files.

Declarations

Conflict of interest The authors declare no competing interests.

Ethical approval Not applicable.

Consent to participate The authors declare that they consent to participate in this study.

Consent for publication The authors declare that they consent to the publication of this study.

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