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The effect of microplastic contaminated compost on the growth of rice seedlings

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ABSTRACT

Microplastics (MPs) are pollutants that have contaminated compost, but there are few studies on the interaction between compost contaminated with MPs (MPsC) and rice seedling performance. This study investigated the effect of MPsC on the planting activity in rice. In this Three different treatments were applied: treatment 1 (P1) as a control without the addition of MPs, Treatment 2 (P2) with the addition of a 1 % PET MPs concentration, and Treatment 3 (P3) with the addition of a 2 % PET MPs concentration. Rice seedlings were planted in the planting medium of each treatment and observed for 24 days after planting (dap). Treatment with the addition of PET MPs showed a significant reduction 38 %, 25 %, 25 % at root length, height, and fresh weight respectively. Additionally, in chlorophyll content there was a decrease of 42 %, 45 %, 55 % in Chl a, Chl b, and total Chl. This decrease may be caused by disturbances in nutrition and photosynthesis processes due to exposure to MPs. The addition of PET MPs to compost as a planting medium can inhibit the growth and health of rice seedlings. These findings underscore the critical need for effective management of plastic waste in agricultural compost to mitigate its adverse effects on plant growth and environmental sustainability. Proper disposal and treatment of plastic contaminants are essential to maintain the integrity of compost used in agriculture, thereby ensuring optimal plant health and ecological balance.

1. Introduction

The impact of microplastics (MPs) on plant seeds is highly relevant to the awareness of human welfare and environmental sustainability in today's society (Azeem et al., 2021). Plant seeds serve as the starting point in the plant's life cycle, and when contaminated by MPs (De Silva et al., 2022), the effects can disseminate throughout the growing plant and potentially be consumed by humans (Jin et al., 2021). This impact not only affects public health due to exposure to MPs in food, but also threatens adequate food availability because the quality of the harvest is compromised (Aydın et al., 2023). Furthermore, awareness of the importance of environmental preservation is burgeoning in society (Lamichhane et al., 2023; Osiako et al., 2022; Zhu Hang & Hadibarata, 2022). Additionally, the economic repercussions of MPs contamination on agricultural products could imperil farmers' income and the economic well-being of local communities (Sutanto et al., 2024). Therefore, the impact of MPs on plant seeds is not only pertinent in the context of public health and environmental sustainability but also represents a shared responsibility to ensure a better future for generations to come (Li et al., 2023b).

In addition, study on the impact of MPs on plant seeds has become a major focus of scientists in recent years. Several previous studies have investigated various aspects of this occurrence. For example, several studies have looked at the impact of exposure to MPs on the growth and development of plant seeds, including rice (MPs can be absorbed by roots thereby inhibiting root activity and photosynthesis) (Dong et al., 2020; Liu et al., 2022), vegetables (MPs can inhibit the internal activities of seed germination and seed root growth) (Bosker et al., 2019; De Silva

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et al., 2022) and other plants (MPs can cause disruption of photosynthesis and oxidative damage to seedlings, thereby inhibiting seed germination and seedling growth and reducing the shoot biomass ratio of seedlings) (Li et al., 2023a; Li et al., 2023c; Lian et al., 2020). The results of this study show that exposure to MPs can inhibit root growth, nutrient absorption and plant metabolism, as well as affect hormonal balance and plant stress responses (Jia et al., 2023). Other research has tried to understand how MPs carried by irrigation water or rain can affect plant seeds growing on agricultural land (Gan et al., 2023). MPs pose significant threats to plant ecosystems by affecting plant growth, nutrient uptake, and soil properties (Zhou et al., 2021c). MPs pose a significant threat to plant ecosystems by entering agroecosystems through various pathways (Dai et al., 2022). Studies indicate that MPs can hinder plant growth and nutrient absorption, leading to reduced crop yields (Jin et al., 2022). These particles can alter soil properties dan affecting biophysical characteristics (Yuan et al., 2023). Apart from that, there is also research exploring how MPs trapped in soil can affect the diversity of soil microorganisms and nutrient cycles in soil ecosystems (Chang et al., 2022). Furthermore, MPs in soil can change the composition of plant communities and impact plant performance (Khalid et al., 2020).

Although there has been a lot of study on the impact of MPs on plant seeds, there are still several problems and weaknesses that need to be overcome. For example, MPs mixed with distilled water and exposed to rice seeds can increase root length and significantly reduce the activity of antioxidant enzymes (Zhang et al., 2021). Exposure to polyethylene (PE) MPs in the rice seeding process using a hydroponic system can inhibit root growth (Tang et al., 2022; Zaidi et al., 2022). Tan et al. (2022) suggested that PE impact the growth, physiological, and biochemical attributes of rice seedlings. Rice seedlings exposed to polystyrene (PS) nano plastics (NPs) resulted in reduced root length and increased number of lateral roots (Zhou et al., 2021a). The impact of polystyrene (PS) and polytetrafluoroethylene (PTFE) MPs particles on hydroponic rice seedling can cause a decrease in root (leaf) mass and inhibit root activity (Dong et al., 2020). The presence of PS MPs impact on arsenic (As) uptake and accumulation in rice tissues (Mamathaxim et al., 2023). All these findings describe the impact of MPs on rice seedlings (*Oryza sativa L.*) carried out using hydroponic cultivation. However, the impact of using compost growing media contaminated with MPs on plant vegetative growth is still unknown. In addition, various kinds of compost products will have different types and sizes of MPs. More specifically, there is currently little information regarding the impact of the toxicity of microplastic-contaminated compost on plant growth.

Rice (*Oryza sativa L.*) is the main food crop for most of the world's population, especially in Asia (Muthayya et al., 2014). Rice plants have an important role in providing food for the community and in maintaining the balance of the agricultural ecosystem (Yuan et al., 2022). However, rice plants are also exposed to environmental contamination, including MPs (Wu et al., 2022). Compost is an organic material that is often used in agricultural practices to increase soil fertility and plant productivity (Ho et al., 2022). However, when the raw material for compost comes from plastic waste, especially single-use plastic, the possibility of contamination by MPs becomes a significant risk factor (Braun et al., 2021). Although there have been many studies investigating the impact of MPs on plants, study on the impact of compost contaminated with MPs on rice seedlings is still limited.

Therefore, this study aims to fill this knowledge gap by investigating the impact of MPs contaminated compost on the growth and development of rice seedlings. This study is expected to provide a deeper understanding of the interaction between contaminated MPs in compost and rice plants in the early stages of growth.

2. Materials and method

2.1. Materials

The cow dung was collected at a small-scale cattle farm in Pamekasan, East Java Province, Indonesia (7°07′ N, 113°26′ E). In the comprehensive composting process, cow urine was not included because the farm uses manure-urine separation technology. Commercial composting additives (organic fertilizer fermentation microbial agent) Songgolangit Persada Co., Jakarta, Indonesia. Chemicals methanol, acetone, hydrogen peroxide (H_2O_2) and ferrous sulfate liquid (Fe_2SO_4) were purchased from Kimia Farma company (Pamekasan, Indonesia). Plastics used in this study were purchased from plastic distributors Sinar Plastik (Pamekasan, Indonesia) and plastics brand Gading. The Rice seeds were (*Oryza sativa* L.) Bintara-08 variety.

2.2. Experimental setup and treatment

In this study, MPs particles of polyethylene (PET), with a diameter of 100–1000 μm and a purity of ≥97 % were obtained by crushing plastic materials with a blender (Iswahyudi et al., 2024). The MPs particles were subjected to a methanol wash to eliminate surface chemicals soluble in the solvent. Following the approach outlined by Bandow et al. (2017), the MPs underwent artificial aging at 80 °C for 360 h to simulate their outdoor degradation and were subsequently stored at 4 °C prior to utilization.

Composting was carried out on the cattle farm in December 2023. Approximately 500 kg of fresh cow dung was arranged in a cone-shaped pile with a triangular cross-sectional area. The initial moisture content of the raw cow manure was approximately 75 %, necessitating preliminary drying to reduce the moisture content to around 60 % to facilitate optimal composting conditions. Commercial composting additives were used to ensure successful composting, and to regulate compost fermentation, the method proposed by Li et al. (2022) was used as a basis. The characteristics of the compost result were pH 7.5, C-organic 10.30 %, Nitrogen (N) 0.90 %, Phosphorus (P) 0.40 %, and Potassium 0.60 %.

The rice seeds used are the Bintara-08 variety. A total of 500 uniform rice seeds were sterilized with 3 % H_2O_2 for 15 min, rinsed with deionized water 2–4 times, and soaked in distilled water for 24 h at 26 °C in the dark. The soaked seeds were then placed into nine trays, then 50 seeds were placed in each tray, and compost, as well as three concentrations of 0 % w/w PET MPs (P1/control), 1 % w/w PET MPs (P2), and 2 % w/w PET MPs were added to the trays (3 treatment × 3 repetition) were conducted. The method proposed by Liu et al. (2021) was used as the basis. These trays were randomly arranged in the greenhouse, where the temperature was controlled at 21 °C ± 5 °C and the humidity was about 65 %. Watering was given as much as 60 % for 48 h. No additional fertilization was added.

The investigate in trays aims to evaluate the impact of MPs on rice seed growth. At days after planting (dap) 9, 12, 15, 18, 21, and 24, the number of germinations was counted and used to calculate the germination rate on each planting medium. The experiment was completed on day 24. Seedling parameters (including the germination percentage (GP), seedling height, number of leaf and fresh weight) were then determined. Briefly, three seedlings were randomly selected for each planting medium, and seedling height (distance from the surface of the planting medium to the top of the seedling), and root length were measured using a caliper.

2.3. Determination chlorophyll and identification MPs

Chlorophyll *a* (Chl *a*), chlorophyll *b* (Chl *b*), and total chlorophyll were measured after a 24 dap. In the analysis, 0.1 g of fresh leaves from each treatment was weighed. The leaf samples were crushed with a porcelain mortar until smooth and mixed with 10 ml of acetone solution.

The solution was then placed in the tube. Then, mixture was subsequently subjected to centrifugation to effectively separate the extract from the residual leaf material. A spectrophotometer was used to measure the absorbance after the extract was put into a cuvette (Shimadzu UV-1800 UV–VIS Spectrophotometer, Tokyo, Japan), at a wavelength of 662 and 644 nm. The ultraviolet–visible (UV–VIS) spectra of acidified supernatants were measured within the specified wavelength range. The measurements were taken immediately after the addition of the acidic solution and afterwards at two-minute intervals for a duration of 20 min. Additionally, a final scanning procedure was conducted 24 h later to assess potential temporal variations. Prior to the final analysis, the samples were stored in a refrigerated environment within sealed, opaque, dark-brown containers. The standard concentration UV–VIS for extracts in acetone solvents was Chl *a* max 86.3 g^{-1} cm^{-1} and Chl *b* max 20.49 g^{-1} cm^{-1} (Lichtenthaler & Buschmann, 2001). Calibrated a UV–VIS spectrophotometer for measuring chlorophyll concentrations typically involved preparing a set of standard solutions of known Chl concentrations and then using the spectrophotometer to measure the absorbance of these standards (Sobiechowska-Sasim et al., 2014). The detailed procedure for this analysis used a method proposed by Zhu et al. (2019).

2.4. Statistical analysis

The results of each triplicate sample are presented as the mean. Significant differences between the treatments and control were determined by one-way analysis of variance (ANOVA, $p < 0.05$), followed by an LSD test (least significant difference) using SPSS 20.0. Origin 9.5 was used to create and modify all the graphs.

3. Results and discussion

3.1. Effects of MPs on the germination percentage (GP)

Fig. 1 shows the germination percentage in various treatments, namely P1, P2, and P3. The germination percentage in treatment P1 was 17.5 %, in treatment P2 was 12.5 %, and in treatment P3 was 12.7 %. Treatment P1 had the highest percentage compared to other treatments, while treatments P2 and P3 had lower.

The observed variance in germination among treatments P1, P2, and P3 may be attributed to multiple factors associated with PET exposure. PET are capable of adsorbing and subsequently releasing toxic chemical compounds into the compost medium. Ge et al. (2021) States that PET can absorb and release harmful chemical compounds into the environment, which can damage plants. These chemical compounds can interfere with plant growth and development processes, inhibit the absorption of nutrients and water by plant roots, and disrupt overall plant metabolism (Martín et al., 2023). In addition, PET MPs may also disrupt the structure of growing media and reduce the availability of pore space, which can inhibit plant root growth and reduce the soil's ability to provide sufficient nutrients for plants (Han et al., 2022). This can result in a decrease in the growth percentage of plant seeds in treatments exposed to PET MPs. This difference in rice seeds germination percentage due to the impact of MPs was proven by Colzi et al. (2022) that reported effect of PET MPs on the growth percentage of plant seeds can vary depending on several factors, including plant type, MPs concentration, and environmental conditions. Strengthening this, several studies have found that exposure to PET MP can inhibit plant growth (Pignattelli et al., 2021a).

3.2. Effects of MPs on the root length

Fig. 2 illustrated impact MPs on the root length of plant seedlings. P1 exhibiting the greatest root length at 6.5 cm, followed by P2 at 5.7 cm, and P3 at 5.0 cm in 9 daps. By 12 daps, all treatments demonstrated a marked increase in root length. P1 displayed the most pronounced growth, reaching 8.9 cm, while P2 and P3 measured 7.7 cm and 6.6 cm, respectively. The variation in root length persisted at 18 and 21 daps, with P1 exhibiting significant fluctuations, whereas P2 and P3 maintained a more stable growth pattern. Meanwhile an increase in root length was noted in P1 and P2, contrasting with a decrease in P3. The impact of PET on roots was significant at 21 daps meanwhile study by Kang et al. (2023) demonstrated that 14-day photoaged PET hindered seed germination in *Pisum sativum L.* and root elongation in blackgram (*Vigna mungo L.*) and tomato (*Solanum lycopersicum L.*) seedlings (Sahasa et al., 2023). Furthermore, P1 had the most beneficial impact on root growth, with P2 showing intermediate results, and P3 displaying the lowest growth due to MP exposure.

These findings indicate PET MPs can also reduce the availability of nutrients to plants. MPs particles can absorb nutrient compounds from compost, thereby reducing their availability to plants (Zhang et al., 2022) cause plant roots having difficulty getting enough nutrients and affects their root growth. Additionally MPs causing blockage of plant roots. Yu et al. (2023) stated that exposure to MPs may trigger excessive production of reactive oxygen in plants, which can damage plant cells and inhibit root growth. PET MPs significantly impeded root growth in

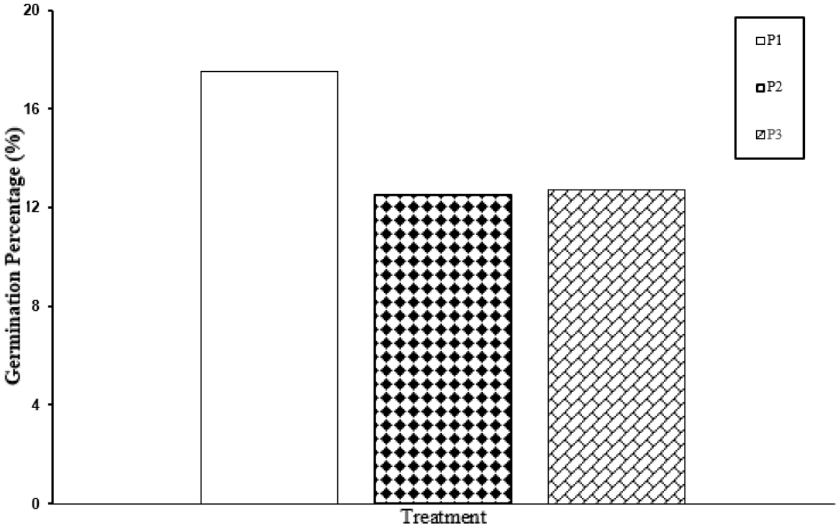

Fig. 1. Effects of different PET concentrations and compost on rice seed germination. Note: The germination rate of the rice seeds in each treatment observed on the ninth day. Germination percentage = (number of seeds germinated on the day/number of test seeds) × 100 %.

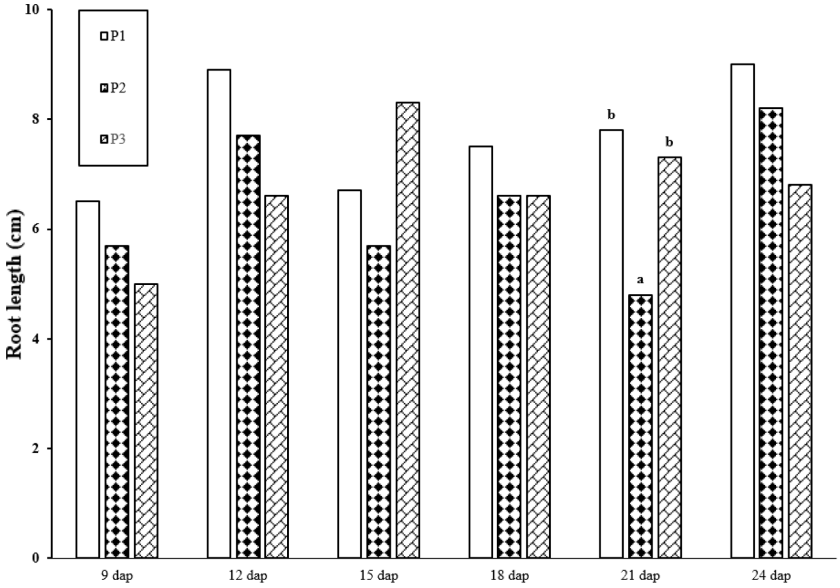

Fig. 2. Effects of different PET concentrations and compost on root length. Different letters for each parameter indicate significant difference at $P < 0.05$.

hyperaccumulators, *Solanum photeinocarpum* and *Lantana camara*, when added to heavy metal-contaminated soil.

3.3. *Effects of MPs on the growth of rice height*

Effect of MPs on rice height in this study are shown in Fig. 3. On 9 dap, plant height in all treatments was still relatively low but had begun to show signs of growth. Then, at 12 daps, there was a significant increase in plant height in all treatments, indicating faster growth. Deviations in plant height between treatments began to appear in this period, where treatment P1 had the highest plant height, followed by P3, and the lowest was P2.

A similar pattern of increased growth continued to occur at 15, 18, 21, and 24 daps. However, the differences between plant heights in each treatment became clearer as time went by. Treatment P1 continues to show the highest plant height, followed by P3 and P2. Thus, the data shows that treatment P1 tends to have the most positive impact on plant growth than MPs contaminated compos. The buildup of MPs in compost can inhibit plant root penetration and gas exchange in the soil, which can affect plant growth and development. This can be reflected in the data pattern with treatments P2 and P3 showing slower growth compared to P1. Furthermore, The decrease in root length in treatments P2 and P3 was also caused by a reduction in microbes due to the presence of MPs. MPs can alter the soil microbial community, which is essential for nutrient cycling and root health. Disruptions in microbial activity can negatively affect root growth and plant resilience. Research indicates that the presence of MPs in soil significantly reduces bacterial populations and enzyme activity related to soil health (Zhou et al., 2021b). Additionally, studies investigating the effects of MPs at various concentration levels (1.5 %, 7.5 %, 15 %) on soil properties and Vicia faba plants have shown a decrease in root length and plant biomass (Elbasiouny et al., 2023). Meanwhile, Mps can alter soil structure by affecting soil pores (Yang et al., 2023), resulting in a decrease in soil density and changes in water and air storage. These changes induce stress in plant roots as their ability to absorb water and nutrients is compromised. Consequently, plant root growth is disrupted because soil microbes play a crucial role in supporting plant nutrition.

The PET MPs have varying impacts on plant height depending on the plant species and concentration of MPs. Study indicates that PET MPs did not significantly affect the plant height of *Solanum photeinocarpum* and *Lantana camara* (Yu et al., 2023). In contrast, a study on blackgram (*Vigna mungo L.*) and tomato (*Solanum lycopersicum L.*) showed that PET

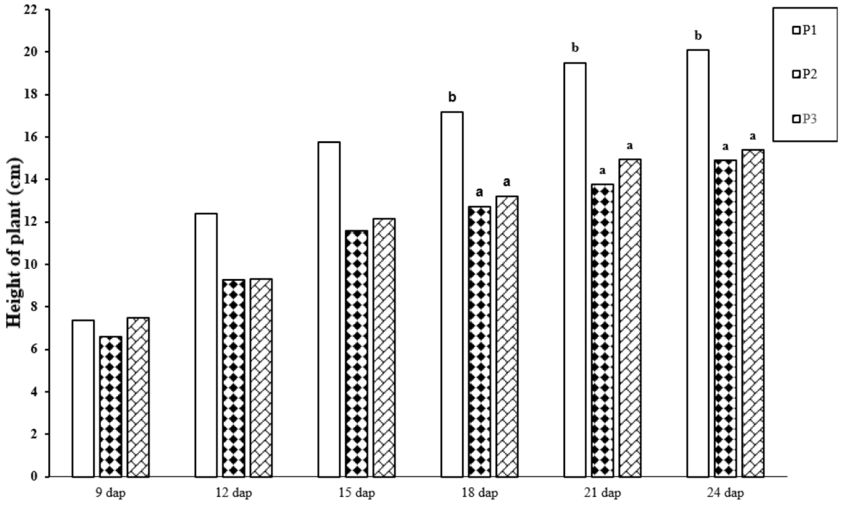

Fig. 3. Effects of different PET concentrations and compost on rice height. Different letters for each parameter indicate significant difference at $P < 0.05$.

MPs had minimal influence on the shoot growth of tomato plants (Sahasa et al., 2023). Additionally, the presence of PET MPs did not effectively reduce shoot height in *Carex stenophylla Wahlenb* (Gharahi & Ahmadmahmoodi, 2022). However, the study on blackgram (*Vigna mungo L.*) revealed a distinct reduction in root and shoot characteristics with increasing concentrations of PE-MPs in different growth media types (Sahasa et al., 2022).

3.4. Effects of MPs on the number of leaf

The data presented in Fig. 4 illustrate the number of leaves on rice seedlings. No significant difference in number of leaves was observed across all treatments. However, Treatments P1 and P2 exhibited an increase to 3 leaves per seedling, whereas P3 remained at 2.5 leaves per seedling. This suggests variation in leaf growth patterns among the treatments, with P1 and P2 demonstrating superior growth compared to P3. These findings illustrate the growth pattern of leaf number in rice seedlings over the observation period, showing a gradual increase from 9 daps to 24 daps, with notable differences in growth between treatments P1, P2, and P3 becoming evident after 18 daps.

The impact of microplastics on leaf number in plants varies depending on the particular plant species and the size of the microplastic particles. Research has shown that microplastics, especially nanoplastics (NPx), can have varying effects on different plant parameters (Sahasa et al., 2022). The MPs in this study were still in micro form with a size of 100 – 1000 μm, which indicates that this size was not able to inhibit the formation of new leaves. While some plants may not show significant changes in leaf number when exposed to microplastics, other growth parameters such as root morphology and photosynthetic pigments may be negatively affected (De Silva et al., 2021). MPs can cause stress in plants, but the number of leaves remained relatively stable in the various MPs doses tested (Mikail, 2023). Additionally, (Hadiuzzaman et al., 2022) support this finding, stating that varying the dose of microplastics did not have a significant effect on the number of leaves in several plant species tested.

3.5. Effects of MPs on the fresh weight

The fresh weight of rice seedlings during the dap observation period in the three treatments (P1, P2, and P3) (Fig. 5) shows consistent variations in the weight of rice seedlings. On 9 dap and 12 dap, all treatments had a relatively similar fresh weight of rice seedlings, with a value of around 0.4 to 0.5 g. This shows that at the beginning of growth, the seedlings in all treatments had quite similar weights. At this stage, the negative impact of MPs has not yet affected rice plant biomass. In contrast, there was a slight variation at 15 daps, where the P3 treatment showed an increase in the fresh weight of rice seedlings to 0.5 g, while the P2 treatment showed a decrease to 0.3 g. On 18 dap and 21 dap, the fresh weight of rice seedlings was again stable in all treatments, with values almost the same as on 15 daps. PET MPs in compost may interfere with the activity of microorganisms that are important for the decomposition of organic matter and plant nutrient cycles. High concentrations of MPs in soil environment can reduce plant biomass by affecting soil microbial activity, which is crucial for organic matter decomposition and nutrient provision to plants. The presence of MPs in soil can also disrupt soil structure and inhibit microbial growth, thereby diminishing the efficiency of organic matter decomposition and reducing the availability of essential nutrients for plants (Zhou et al., 2021c). Additionally, Study shows that PET particles can absorb and transport pollutants such as naphthalene and phenanthrene, thereby affecting the growth of plants such as blackgram (Sahasa et al., 2022). In addition, PET MPs that are resistant to environmental degradation can enter the food chain and potentially harm human health (Abbasi et al., 2021). Pignattelli et al. (2021b) stated that PET MPs can affect the physiology of *lepidium sativum* plants and the fresh weight of shoots. Study has simulated the passage of PET MPs through roots, stems and fruit, showing changes in the fresh weight of the fruit and if consumed by humans would show health effects on digestion (Dainelli et al., 2023). The presence of PET MPs in the environment poses a risk to terrestrial ecosystems, affecting the health of soil organisms and disrupting normal biological processes (Dissanayake et al., 2022).

3.6. Effects of MPs on photosynthesis

Chl content due treatment shown in Fig. 6. The data shown that the control treatment (P1) tends to have higher chlorophyll concentrations (both Chl *a*, Chl *b*, and total chlorophyll) compared to treatments P2 and P3. This indicates that increasing concentrations of PET MPs in compost can influence the absorption or production of chlorophyll in rice seedlings, with a decrease observed in treatments P2 and P3 compared to the control (P1). MPs may caused contamination into compost can disrupt the availability of nutrients for plants. MPs can bind nutrients in the compost, thereby reducing the availability of nutrients that can be absorbed by plants. As a result, the concentrations of Chl *a*, Chl *b*, and total chlorophyll in rice seedlings in treatments with the addition of MPs (P2 and P3) may decrease due to a lack of nutrients needed for

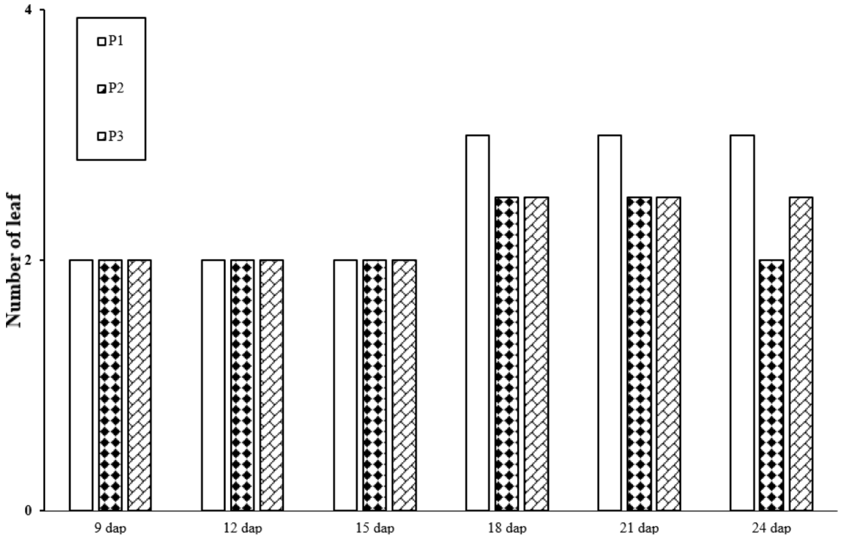

Fig. 4. Effects of different PET concentrations and compost on number of leaf seedling.

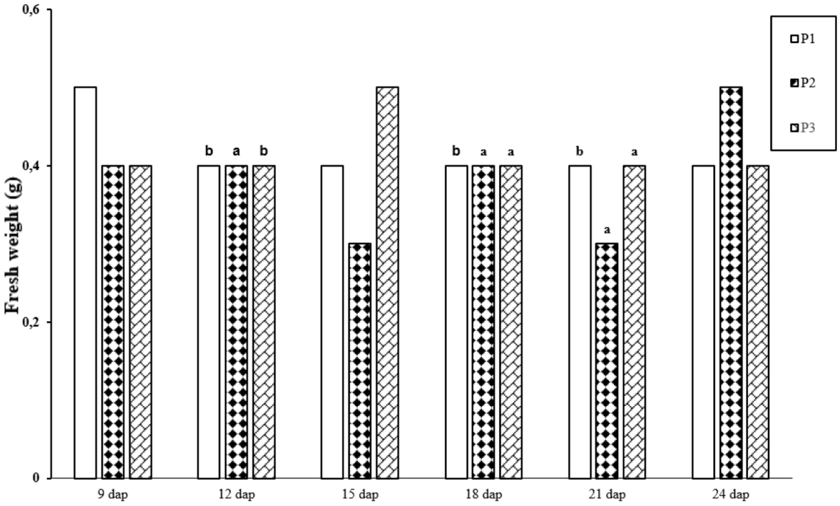

Fig. 5. Effects of different PET concentrations and compost on number of fresh weight seedling. Different letters for each parameter indicate significant difference at P < 0.05.

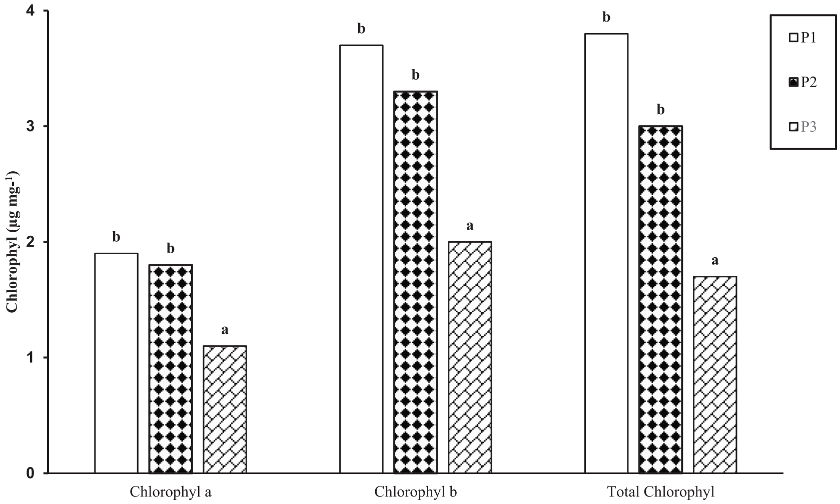

Fig. 6. Effects of different PET concentrations and compost on photosynthesis. Different letters for each parameter indicate significant difference at P < 0.05.

chlorophyll formation. MPs have been shown to release harmful additives that plants can absorb, disrupting metabolic processes and Chl synthesis. Yang and Ma (2023) found that MPs can reduce soil microbial activity, essential for organic matter decomposition and nutrient cycling. Additionally, MPs increase oxidative stress in plants, potentially damaging chloroplasts and thereby reducing Chl content (Sahasa et al., 2022). Furthermore, MPs can alter endogenous fluorescence derived from chlorophylls, impacting the sensitivity of chlorophyll fluorescence (De Silva et al., 2021).

4. Conclusion

The aim of this research was to evaluate the impact of PET MPs contamination in compost on the growth of rice seedlings. Based on the results of this study, treatment with the addition of PET MPs showed a significant reduction 38 %, 25 %, 25 % at root length, height, and fresh weight respectively. Additionally, in chlorophyll content there was a decrease of 42 %, 45 %, 55 % in Chl a, Chl b, and total Chl. This decrease may be caused by disturbances in nutrition and photosynthesis processes due to exposure to MPs. The addition of PET MPs to compost as a planting medium can inhibit the growth and health of rice seedlings. These findings underscore the critical need for effective management of plastic waste in agricultural compost to mitigate its adverse effects on plant growth and environmental sustainability. These findings highlight the importance of managing plastic waste and the need to find environmentally friendly alternatives in making growing media. For future research, investigating more deeply the mechanisms behind the influence of other types of MPs and their concentrations on plant health has recommended.

CRediT authorship contribution statement

Iswahyudi Iswahyudi: Writing – original draft, Conceptualization. **Adi Sutanto:** Writing – review & editing, Conceptualization. **Wahyu Widodo:** Writing – review & editing, Supervision, Methodology. **Warkoyo Warkoyo:** Writing – review & editing, Data curation. **Marchel Putra Garfansa:** Writing – review & editing, Visualization. **Syamsul Arifin:** Writing – original draft, Data curation. **Siti Holifah:** Writing – review & editing, Data curation. **Sugiono Sugiono:** Supervision, Methodology. **Mohammad Shoimus Sholeh:** Data curation. **Shefa Dwijayanti Ramadani:** Formal analysis.

Declaration of competing interest

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Ethical statement

This article does not contain any studies with human or animal subjects performed by any of the authors.

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