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Synergistic integration of Kayu apu (*Pistia stratiotes*) and EM4 for sustainable landfill leachate treatment: A green approach to pollutant reduction

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ABSTRACT

This study evaluates the synergistic integration of phytoremediation using *Pistia stratiotes* (Kayu Apu) and microbial bioaugmentation with Effective Microorganisms 4 (EM4) for treating landfill leachate. A batch system was employed with EM4 concentrations of 0%, 10%, 20%, and 30% and residence times of 3, 6, and 9 days. The results indicate that the highest pollutant removal efficiency was achieved with 30% EM4 and a 9-day residence time, resulting in 41.5% BOD reduction, 37.3% COD reduction, and 38% TSS reduction. Additionally, DO levels increased significantly by 488.24%, indicating improved aerobic conditions, which are essential for microbial activity. These findings demonstrate the potential of integrating *Pistia stratiotes* and EM4 as a sustainable, eco-friendly approach to treating landfill leachate. Further research is recommended to optimize operational parameters, scale up the system, and ensure compliance with regulatory discharge standards.

1. Introduction

Landfill leachate is highly contaminated wastewater that poses significant environmental risks if not managed properly. It contains high concentrations of organic matter, inorganic pollutants, and suspended solids, contributing to water pollution when discharged untreated [1].

The primary indicators of leachate pollution are Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), and Total Suspended Solids (TSS), which deplete oxygen in water bodies, causing severe ecological disturbances [2,3].

In addition, wastewater that is discharged directly into the environment can increase river pollution [4]. Conventional treatment methods, such as chemical coagulation and advanced oxidation, are effective but often expensive and generate secondary waste [5].

Thus, there is growing interest in sustainable, cost-effective alternatives, such as phytoremediation, which utilizes plants to absorb and degrade contaminants [6]. This technology is popular due

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to its large remediation scope, ease of operation, and minimal environmental disruption [7,8].

Phytoremediation technology efficiently removes heavy metals from the soil, but it generates substantial biomass enriched with these harmful elements. Research shows that the leaching concentration of heavy metals in untreated phytoremediation residue can reach several hundred milligrams per liter (mg/L), surpassing hazardous waste limits in numerous countries and regions [9,10]. Managing the disposal of phytoremediation residue produced during remediation has emerged as a significant challenge [11,12]. Phytoremediation has gained attention as a green technology for wastewater treatment due to its effectiveness in reducing pollutants and improving water quality [13]. Aquatic macrophytes, such as *Pistia stratiotes* (Kayu Apu), enhance oxygenation, support microbial activity, and promote organic matter degradation [14]. However, plant-based remediation alone may not achieve regulatory standards for wastewater discharge. Adding activators, such as Effective Microorganisms 4 (EM4), can enhance degradation by introducing beneficial microorganisms that accelerate the breakdown of complex organic compounds [15].

Leachate treatment poses a significant challenge for landfill management, particularly in developing regions where waste segregation and advanced treatment technologies are limited. The Kaliabu Madiun Indonesia landfill receives unsorted waste from multiple districts, accumulating high-strength leachate with excessive BOD, COD, and TSS levels that exceed the standards. Traditional biological treatments, such as sedimentation tanks, fail to meet discharge standards set by the Ministry of Environment and Forestry Regulation No. P.59/Menlhk/Setjen/Kum.1/7/2016 [16]. This raises concerns about contamination of nearby water sources and underscores the need for more effective, sustainable treatment solutions. Phytoremediation, using *Pistia stratiotes*, has emerged as a viable strategy for wastewater treatment due to its ability to remove pollutants through absorption, accumulation, and rhizofiltration [17]. The effectiveness of plant-based systems can be enhanced through microbial augmentation. EM4 is a bioactivator containing lactic acid bacteria (*Lactobacillus sp*.), fermentation fungi (*Saccharomyces sp*.), and photosynthetic bacteria (*Rhodopseudomonas sp*.) that has the primary function to accelerate organic matter degradation in waste and wastewater treatment applications [15].

Studies have demonstrated the potential of phytoremediation to treat various wastewater types [7,18]. A study found that leachate treatment using a constructed wetland with *Hippochaetes lymenialis* reduced COD and BOD levels by 55% and 85%, respectively [19]. *Pistia stratiotes* treatment of domestic wastewater reduced BOD (82%), COD (81%), and TSS (82%) [20], suggesting its promise for phytoremediation. Microbial augmentation has also been explored as a means to enhance wastewater remediation. All compost samples formulated with 40% FB material and supplemented with both TAK and EM bio-activators exhibited lower final moisture content than the samples containing 20% FB material [15]. Echinodorus palaefolius–based constructed wetlands effectively reduced BOD and COD in wastewater, including tofu-industry effluent, through plant–microbe interactions and mechanisms such as phytoextraction, phytofiltration, and enhanced microbial biodegradation [21]. These studies highlight EM4's effectiveness in accelerating the microbial degradation of organic matter.

Despite growing research on phytoremediation and microbial augmentation, few studies have examined their combined use for treating landfill leachate. Given the high organic load and complex pollutants in landfill leachate, integrating *Pistia stratiotes* and EM4 may offer a more effective solution. This study builds on previous research by examining the optimal EM4 dosage and residence time to enhance phytoremediation efficiency in leachate treatment. Existing studies have separately examined phytoremediation and EM4-based treatments for various wastewater types. However, research on their combined application for landfill leachate is limited. For example, previous phytoremediation studies have primarily focused on CW and floating plant systems, often without microbial supplementation [19, 20]. While *Pistia stratiotes* has been tested in domestic and industrial wastewater treatment, its use for high-strength landfill leachate has not been extensively studied. Microbial bioaugmentation with EM4 has shown promising results in organic waste degradation, but has mainly been applied to domestic and industrial effluents rather than landfill leachate [15,22].

Against this background, a clear research gap remains regarding the combined application of phytoremediation with *Pistia stratiotes* and microbial bioaugmentation with EM4 for the treatment of high-strength landfill leachate. While previous studies have primarily examined plant-based systems or EM4-based treatments independently, their synergistic integration and optimization for landfill leachate have not been sufficiently explored. Therefore, the primary objective of this study is to evaluate the effectiveness of combining *Pistia stratiotes* and EM4 in reducing BOD, COD, and TSS while increasing dissolved oxygen (DO) levels in landfill leachate. This study further investigates the influence of varying EM4 dosages and residence times on treatment performance to identify optimal operating conditions. The findings are expected to contribute to the development of sustainable, cost-effective, and environmentally friendly leachate management strategies, particularly for landfill sites with limited access to advanced treatment technologies.

2. Materials and methods

2.1. Experiment set-up

This study was conducted using a batch system with EM4 levels (0%, 10%, 20%, and 30%) and residence times (0, 3, 6, and 9 days) to evaluate the effectiveness of phytoremediation in reducing these pollutant parameters. The research was conducted in several main stages: plant acclimatization; initial toxicity testing using the Range-Finding Test (RFT); leachate treatment via phytoremediation; and analysis of water quality parameters before and after treatment. This research was conducted at two central locations: the Kaliabu Madiun Indonesia Landfill in Madiun Regency for leachate sampling, and the Laboratory of the Madiun Regency Environment Agency, where water quality was tested before and after treatment.

Pistia stratiotes is a free-floating aquatic macrophyte belonging to the family Araceae, characterized by rosette-shaped leaves and a dense fibrous root system that provides a large surface area for pollutant adsorption and microbial attachment, making it suitable for phytoremediation applications. Each treatment reactor consisted of a plastic tank with a practical working volume of 24 L and was operated under batch conditions without mechanical aeration. The experiment was conducted under ambient laboratory conditions, with wastewater maintained at 32-35 °C and natural daylight. Each reactor contained 30 uniformly sized Pistia stratiotes plants, and EM4 was applied as a volumetric percentage of the commercial EM4 stock solution, as specified in the experimental design.

2.2. Materials and tools

The primary materials used in this study were leachate from the Kaliabu Madiun landfill, which served as the research object, and *Pistia stratiotes*, used as a phytoremediation agent. *The leachate originated from mixed municipal solid waste and was characterized by high organic content and elevated suspended solids concentrations*. Additionally, EM4 was utilized as a bioactivator to accelerate the degradation of organic compounds in leachate. The tools used in this study include an acclimatization reactor and a treatment reactor, which consisted of five plastic tanks measuring 60 cm × 20 cm × 20 cm. A pH meter was used to measure the acidity level of the leachate before and after treatment. In this study, the average pH of leachate waste ranged from 6.4 to 7.3. A thermometer was used to monitor temperature during the treatment process. During the study, the wastewater temperature was observed at 32-35℃. A 500 mL sample bottle was used to store the leachate and test it in the laboratory.

2.3. Acclimatization and Range Finding Test (RFT)

Before being used in a leachate treatment reactor, *Pistia stratiotes* must undergo acclimatization to adapt to the leachate environment and reduce stress from environmental changes.

The acclimatization process was carried out in the following steps: (1) Choosing healthy plants of a uniform size; (2) Cleaning the plants of soil and dirt attached to the roots; and (3) Soaking the plants in clean water for seven days to ensure the plants are in optimal condition before use in research. All *Pistia stratiotes* plants used in this study were uniform in size, physiological age, and health before acclimatization.

Following plant acclimatization, an RFT was conducted to determine the tolerance of *Pistia stratiotes* to landfill leachate. Plants were exposed to leachate concentrations of 0%, 20%, 40%, 60%, and 80% and monitored for seven days to assess survival. The results indicated that *Pistia stratiotes* remained viable up to a leachate concentration of 40%, whereas higher concentrations caused significant plant mortality. Based on these findings, a leachate concentration of 40% was selected for subsequent phytoremediation experiments. The RTF process is shown in Figure 1.

2.4. Leached treatment by phytoremediation

The primary focus of this study is the treatment of leachate using phytoremediation. The research was conducted by preparing four reactor units, each filled with 18 L of diluted leachate based on the RFT test results (leached concentration: 40%). The leachate concentration selected was the higher COD value (635 mg/L), based on initial observations at the landfill, and was diluted with deionized (DI) water to 254 mg/L.

Each reactor was filled with 30 stems of *Pistia stratiotes* and subjected to EM4 levels of 0%, 10%, 20%, and 30%. The treatment was conducted over 9 days, with sampling on days 0, 3, 6, and 9 to analyze COD, BOD, TSS, and DO using grab sampling. Table 1 presents the study's treatment design.

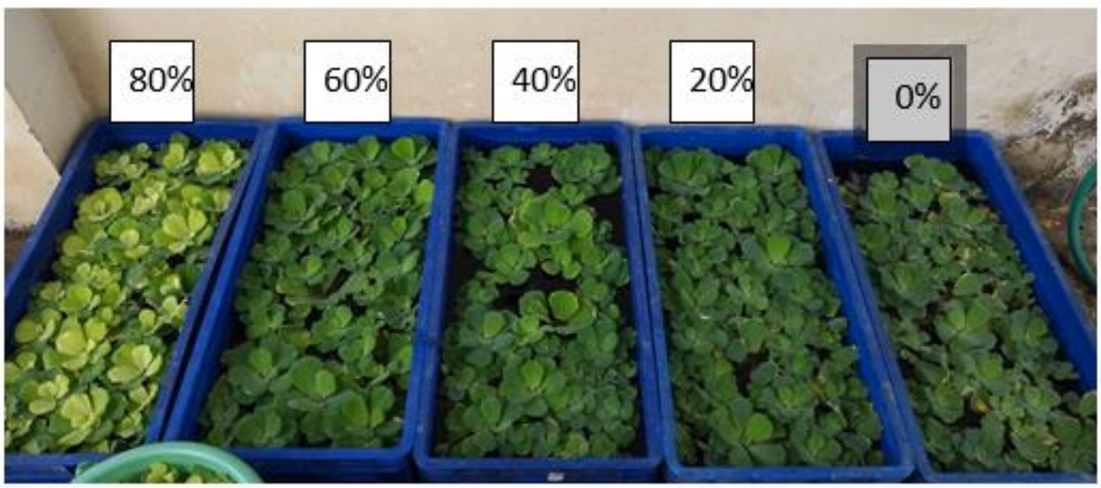

Fig. 1. RFT process on the *Pistia stratiotes*.

Table 1. Design of the study for leached treatment via phytoremediation.

Reactor (R)	Number of plants	Leachate concentration (mg/L)	EM4 concentration (% vol.)
R1	30	254	0
R2	30	254	10
R3	30	254	20
R4	30	254	30

2.5. Data analysis method

After treatment, the leachate samples were analyzed in the laboratory for changes in BOD, COD, TSS, and DO. Initial concentrations of BOD, COD, TSS, and DO were measured before treatment to establish baseline conditions, and these values are presented in Table 2. Phytoremediation effectiveness is calculated using equations [22,23]. Based on the pollutant reduction efficiency formula. The analysis of BOD and COD pollutants followed the guidelines set by Standar Nasional Indonesia, SNI 9689.72:2009 [22]. TSS measurement was based on SNI 6989.3:2019 [23], and DO analysis adhered to SNI 06-6989.14-2004 [24]. The obtained data were processed using

OriginPro 2025 software (OriginLab Corporation, Northampton, MA, USA).

$$\text{Removal (mg/L)} = co - c \quad (1)$$

$$\text{Removal ratio (\%)} \varepsilon = \frac{Co - C}{Co} \quad (2)$$

where ε is the efficiency, C_0 is the initial concentration, and C is the final concentration.

3. Results and discussion

3.1. Characteristics of landfill leachate

The leachate from the Kaliabu landfill in Madiun, East Java, Indonesia, exhibited pollutant concentrations that exceeded environmental quality standards, as shown in Table 2.

Table 2. The initial characteristics of leachate in this study.

Parameter	Quality standards [16]	Value
BOD (mg/L)	150	275
COD (mg/L)	300	635
TSS (mg/L)	100	158
DO (mg/L)	-	1.05
pH	6 - 9	7.2

The BOD level was recorded at 275 mg/L, exceeding the standard limit of 150 mg/L. High BOD reflects elevated organic matter levels in water, which stimulate aerobic microbial activity and consequently increase dissolved oxygen consumption [25]. The COD level of 635 mg/L was more than double the permissible limit of 300 mg/L, reflecting the presence of both biodegradable and non-biodegradable organic pollutants [26]. The TSS concentration reached 158 mg/L, exceeding the maximum allowable standard of 100 mg/L. Critically low DO levels (e.g., 1.05 mg/L) reflect severe oxygen depletion driven by microbial degradation of labile dissolved organic carbon, which can induce hypoxic or anoxic conditions and associated impacts, such as water acidification and elevated carbon dioxide (CO_2) emissions [3,27]. However, the leachate's pH meets quality standards. These findings underscore the necessity for effective treatment strategies to reduce pollutant levels and mitigate environmental risks associated with leachate discharge. These findings emphasize the need for integrated treatment approaches that simultaneously address organic, inorganic, and suspended pollutant loads. The findings highlight the critical environmental challenges of untreated landfill leachate, particularly its potential to contaminate nearby water sources and disrupt aquatic ecosystems. The elevated BOD and COD levels indicate a substantial organic pollutant load that can deplete oxygen in receiving water bodies, threatening aquatic life and biodiversity [3]. Similarly, the high TSS concentration can lead to turbidity and hinder photosynthesis in aquatic systems, further compromising ecological balance [17]. The low DO level observed in the leachate exacerbates these risks by limiting oxygen availability for microbial degradation and aquatic organisms.

The elevated BOD and COD concentrations observed in the leachate are primarily attributed to the decomposition of organic matter from mixed municipal solid waste, including food residues, paper, and biodegradable household materials. During anaerobic degradation within landfill cells, complex organic compounds are converted into soluble organic substances, increasing oxygen demand in the leachate. High TSS levels result from suspended particulates, fine soil fractions, and decomposed waste materials that are mobilized by percolating rainwater. Meanwhile, the low DO concentration reflects intense microbial oxygen consumption associated with the high organic load, which limits oxygen availability and creates predominantly anaerobic conditions within the leachate.

3.2. The RFT analysis

The RFT was conducted on the *Pistia stratiotes* plant for seven days as a phytoremediation medium for leachate wastewater absorption. The RFT test was conducted to assess the tolerance of the *Pistia stratiotes* plant to various concentrations of leachate. The concentrations of leachate wastewater used in the test were 0%, 20%, 40%, 60%, and 80%. The test results shown in Figure 2 indicate that at concentrations of 0%, 20%, and 40%, all plants remain alive, with no mortality (% death = 0.0%).

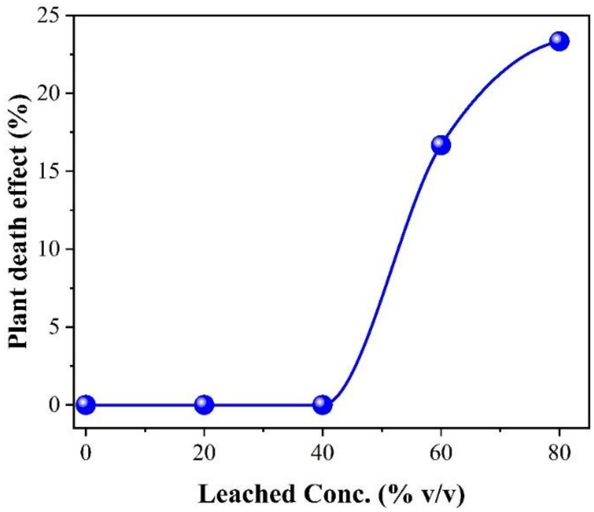

Fig. 2. Range Finding Test on the *Pistia stratiotes*.

However, at a concentration of 60%, 5 of 30 plants died, yielding a mortality rate of 16.7%. At a concentration of 80%, the number of dead plants increased to 7, resulting in a mortality rate of 23.3%. Based on these results, *Pistia stratiotes* plants can survive up to 40% concentration but experience a significant increase in mortality at concentrations of 60% and above. This indicates that *Pistia stratiotes* begin to experience toxic effects from leachate wastewater at concentrations of ≥60%, suggesting that the optimal concentration for phytoremediation should be below this value. Based on these results, a leachate concentration of 40% was determined as the threshold at which *Pistia stratiotes* could thrive without significant mortality. This concentration was, therefore, selected for subsequent phytoremediation experiments.

The results of the RFT align with previous studies, which demonstrate the tolerance limits of aquatic plants to wastewater pollutants. *Pistia stratiotes* is highly adaptable to various polluted environments, but experiences stress at elevated pollutant concentrations [14]. Aquatic plants show optimal growth and contaminant uptake at moderate concentrations but experience high mortality at elevated levels; for instance, *Eichhornia crassipes* can remove up to 84.38% of ciprofloxacin via root uptake without short-term growth inhibition [28]. Similarly, leachate concentrations above 50% often cause plant stress due to high pollutant loads, such as heavy metals and organic matter, which interfere with metabolic and photosynthetic processes [29]. Compared to these studies, the tolerance limit observed in the current research (40% leachate concentration) reinforces the suitability of *Pistia stratiotes* as a phytoremediation agent for moderately polluted wastewater. However, it also highlights the necessity of determining pollutant thresholds to optimize plant survival and treatment efficiency in landfill leachate applications.

The RFT results are essential for defining operational parameters in landfill leachate phytoremediation using *Pistia stratiotes*. A leachate concentration of 40% was identified as the maximum tolerable level that allows plant survival and effective pollutant removal, as higher concentrations may compromise plant viability and treatment efficiency. Moreover, the findings emphasize the importance of pre-treatment or dilution strategies to reduce leachate concentrations before initiating phytoremediation. The system can achieve sustainable and effective remediation by maintaining pollutant levels within the plant's tolerance range. This study also underscores *Pistia stratiotes'* adaptability to moderately polluted environments, making it a promising candidate for large-scale leachate treatment projects, particularly in regions lacking advanced wastewater treatment facilities [29].

3.3. The effect of adding EM4 on the BOD alleviation

The study of the effects of EM4 concentrations (0%, 10%, 20%, and 30%) on BOD_5 reduction in leachate wastewater is presented in Figure 3. The experiment used four reactors, R1 (without EM4), R2 (10% EM4), R3 (20% EM4), and R4 (30% EM4), which were observed on days 3, 6, and 9, respectively. The initial BOD_5 concentration was 275 mg/L, and measurements were taken to assess the effectiveness of each treatment in reducing the organic pollutant load. The results (Figure 3a) showed that the reactor without EM4 (R1) had the lowest remaining BOD_5 among the reactors. On day 3, the BOD_5 in R1 was 32 mg/L, leaving 234 mg/L of the initial concentration. In EM4 reactors, the BOD_5 values set aside were slightly higher: 77 mg/L (R2), 93 mg/L (R3), and 84 mg/L (R4). On day 6, the BOD_5 reduced in R1 increased to 35 mg/L, while R2 showed 92 mg/L, and R3 and R4 each 102 mg/L. On day 9, the BOD_5 reduced in R1 decreased to 19 mg/L, while in R2, R3, and R4, the reduced BOD_5 values were 88, 103, and 114 mg/L, respectively.

In terms of removal efficiency (Figure 3b), R1 had the lowest efficiency: 11.64% on the 3rd day, 12.73% on the 6th day, and decreased to 6.91% on the 9th day. This indicates that the BOD_5 removal process in R1 was less effective, as the remaining BOD_5 value was still close to the initial value. In contrast, reactors using EM4 had slightly higher initial removal efficiencies than R1, with a larger increase over time. On the 3rd day, the removal efficiencies of R2, R3, and R4 were 28%, 33.82%, and 30.55%, respectively. On the 6th day, the efficiency of R2 was 33.45%, whereas R3 and R4 were 37.09%. After 9 days, R2 decreased slightly to 32%, then increased to 37.45% and 41.45% in R3 and R4, respectively.

This demonstrates that reactors with EM4 are more effective at treating BOD_5 in leachate wastewater over the long term.

The lower remaining BOD values observed in R1 (without EM4) at early observation times may be attributed to initial plant uptake, adsorption, and rhizofiltration processes that occur immediately after reactor startup. In contrast, reactors supplemented with EM4 likely experienced a short acclimatization or lag phase, during which introduced microorganisms adapted to the leachate environment and competed with indigenous microbial populations. Consequently, the enhanced degradation effect of EM4 became more pronounced at longer residence times rather than during the initial treatment period.

The observed findings align with previous studies highlighting the effectiveness of combined phytoremediation and microbial augmentation in wastewater treatment. This symbiosis is more effective if the number of microorganisms increases and their environmental adaptation improves [17].

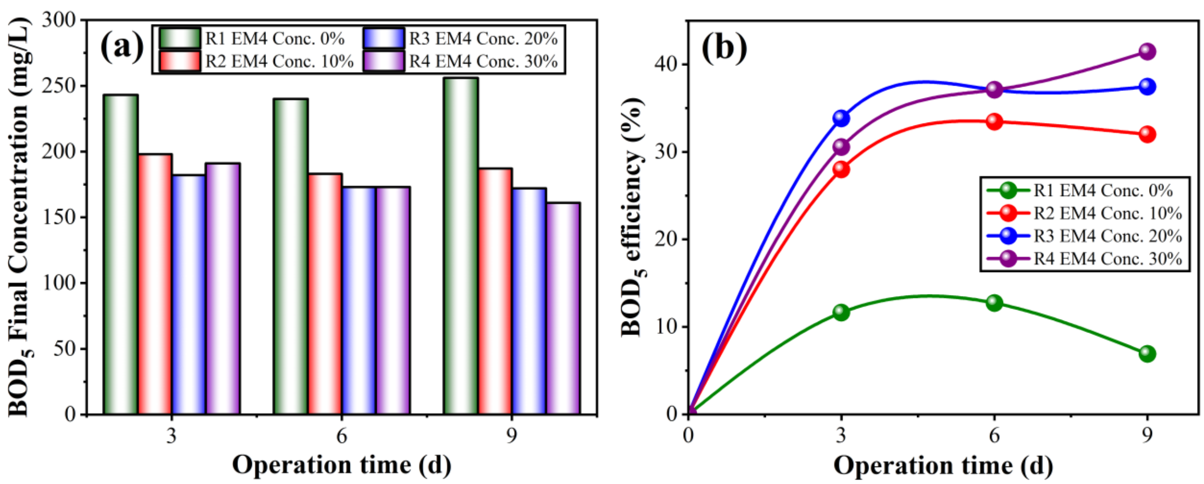

Fig. 3. Alleviation of BOD_5 from leached wastewater: (a) BOD_5 Final Concentration (mg/L) and (b) BOD_5 Efficiency (%).

The roots of pumice wood plants provide habitat for microorganisms that degrade organic matter in the leachate. Pumice wood plant roots supply significant organic carbon to soil via rhizodeposition, with exudates and root turnover contributing an estimated 10–100 mg C g^{-1} root material [29,30].

CO_2 and water (H_2O) are used by the apu wood plant in photosynthesis, producing oxygen (O_2). This oxygen is used by microorganisms for respiration, thereby optimizing bacterial growth and promoting the degradation of organic matter in leachate [26].

The *Pistia stratiotes* plant has long, fine roots, providing an adequate habitat for microorganisms to grow [14]. The decrease in BOD is also caused by rhizofiltration, namely, the absorption of organic matter by plant roots, which is then distributed to the stem, spreads throughout the plant, and undergoes biological processes until it accumulates in the plant stem [31]. EM4 accelerates the degradation of organic matter due to its high abundance of beneficial microorganisms, such as

Lactobacillus sp. and *Saccharomyces sp.*, which enhance phytoremediation.

However, compared to earlier studies, the BOD removal efficiency in this research is higher. For example, it was reported that leachate BOD decreased by 24.11% after using *Pistia stratiotes* for three days [32]. In contrast, this study achieved a 41.45% reduction in R4 with the combined application of EM4 and phytoremediation over nine days. This comparison highlights the advantages of integrating microbial bioactivators, such as EM4, into plant-based treatment systems to enhance leachate remediation.

The findings demonstrate the potential of combining *Pistia stratiotes* with EM4 to reduce BOD levels in landfill leachate. The highest BOD reduction achieved in R4 (41.45%) indicates that adding EM4 enhances the degradation of organic pollutants, which is critical for improving leachate quality and minimizing its environmental impact. This system's ability to lower BOD concentrations underscores its applicability in wastewater treatment, particularly in resource-constrained settings where advanced treatment technologies may not be feasible. These results also highlight the importance of optimizing EM4 dosage and residence time to achieve maximum treatment efficiency. The study provides evidence that a 30% EM4 addition and a nine-day residence time are optimal conditions for BOD reduction in leachate treatment. This integrated approach offers a sustainable, cost-effective, and environmentally friendly solution to mitigate the ecological risks posed by untreated landfill leachate.

3.4. The effect of adding EM4 on the COD reduction

A similar early-stage behavior observed in COD removal can be explained by the initial plant-driven removal mechanisms and the acclimatization phase of EM4-associated microorganisms, as discussed in the BOD section. COD was observed using the same method as for BOD removal. The initial COD concentration in wastewater was 635 mg/L, and the study results were analyzed to determine the relationship between the amount of COD removed (mg/L) and its removal efficiency (%), as shown in Figure 4. The results in Figure 4a indicate that reactor R1 (without EM4) achieved lower COD removal than the other reactors. On the 3rd day, the amount of COD successfully removed in R1 was 15 mg/L, whereas in the reactors with EM4 addition, the COD values removed were 35 mg/L in R2, 70 mg/L in R3, and 74 mg/L in R4. On the 6th day, the amount of COD removed in R1 increased slightly to 20 mg/L, whereas in R2, R3, and R4, it was 34 mg/L, 115 mg/L, and 124 mg/L, respectively. Meanwhile, on the 9th day, the COD removed in R1 increased again to 39 mg/L, whereas in the reactor with EM4, the amounts removed were 63 mg/L in R2, 133 mg/L in R3, and 235 mg/L in R4. When evaluated by COD removal efficiency (Figure 4b), R1 exhibited the lowest efficiency: 2.36% on the 3rd day, 3.15% on the 6th day, and 6.14% on the 9th day. The COD remaining in this reactor was still relatively high compared with the other reactors, indicating that COD removal in R1 was not yet fully optimized.

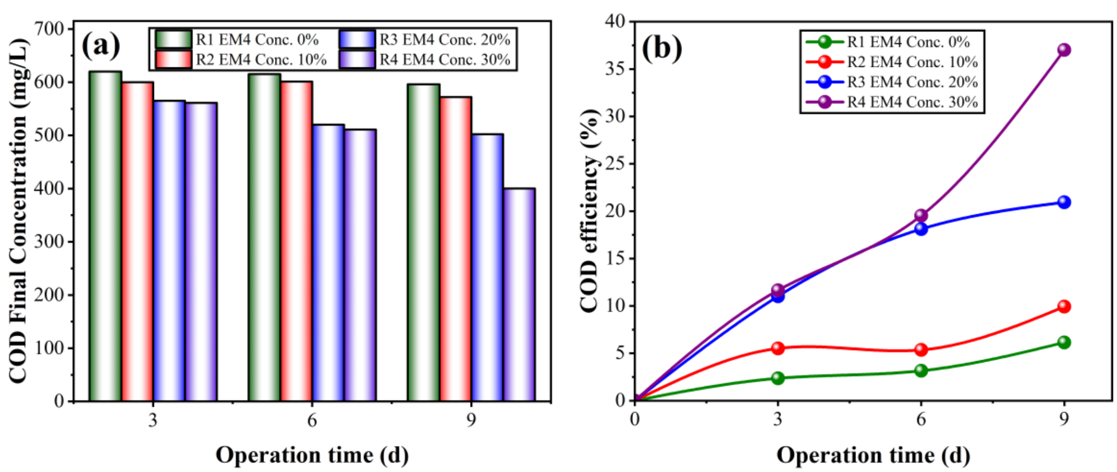

Fig. 4. Alleviation of COD from leached wastewater: (a) COD Final Concentration (mg/L) and (b) COD Efficiency (%).

In contrast, the reactor's COD removal efficiency increased over time with the addition of EM4. On the 3rd day, the COD removal efficiencies in R2 were 5.51%, in R3, 11.02%, and in R4, 11.65%, respectively. This value continued to increase until the 9th day, when the COD removal efficiencies at R2, R3, and R4 reached 9.92%, 20.94%, and 37.01%, respectively. This indicates that the COD removal efficiency was higher than that in R1. In the long term, EM4 enhances the effectiveness of organic matter degradation in wastewater. The results are consistent with previous studies that emphasize the effectiveness of combining phytoremediation with microbial augmentation in wastewater treatment. *Pistia stratiotes* enhances COD reduction by fostering symbiotic interactions between plant roots and microorganisms [14]. The additional introduction of EM4, which contains beneficial microbes such as *Lactobacillus sp.* and *Saccharomyces sp.*, further accelerates the degradation of organic pollutants. Compared to earlier research, the COD removal efficiency in this study (37.30% in R4) aligns with findings from [15], who reported an 83.0% COD reduction in laundry wastewater using microalgae with EM4. While the efficiency in this study is lower, this can be attributed to the more complex composition and higher initial COD levels of landfill leachate compared to those of domestic wastewater. This highlights the challenges of treating landfill leachate and the importance of optimized treatment systems. The findings highlight the potential of integrating *Pistia stratiotes* and EM4 for the effective treatment of landfill leachate. The highest COD reduction observed in R4 demonstrates that increasing the EM4 dosage enhances microbial activity, thereby accelerating the degradation of both biodegradable and non-biodegradable organic matter. This is particularly significant for landfill leachate, which often contains complex pollutants. Moreover, the results highlight the importance of optimizing operational parameters, such as EM4 dosage and residence time, to achieve higher treatment efficiency. The findings support the adoption of this combined treatment approach as a sustainable and cost-effective solution for leachate management in resource-constrained regions. Future studies could focus on scaling up the system and evaluating its performance in real-world settings.

3.5. The effect of adding EM4 on the TSS decline

The high concentration of suspended particles in wastewater indicates elevated TSS levels, which can degrade water quality and pollute aquatic environments. The initial TSS concentration in the leachate wastewater was 158 mg/L. Figure 5 shows the TSS removal efficiency. Based on Figure 5a, the remaining TSS in each reactor was calculated by subtracting the TSS removed from the initial concentration.

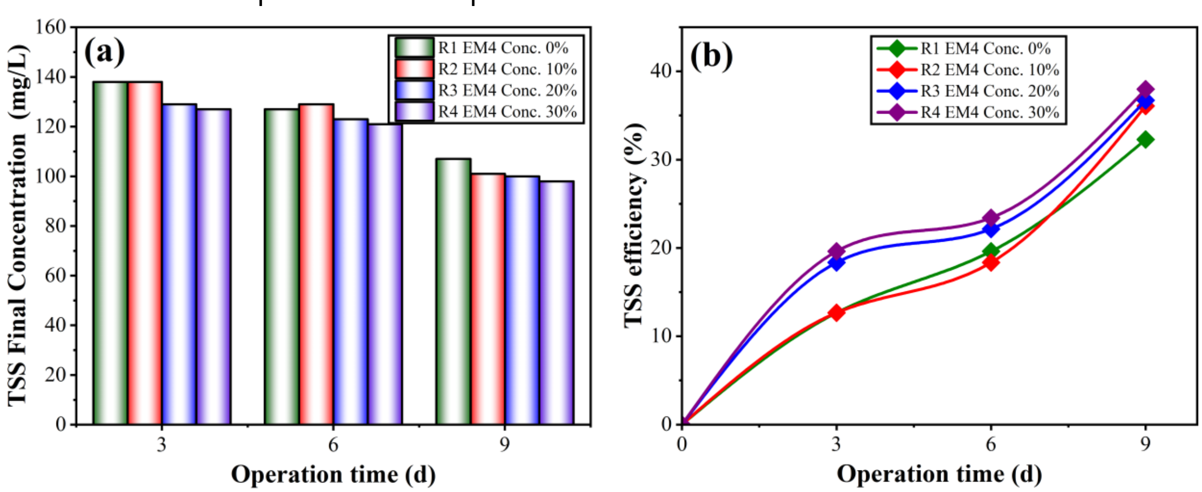

Fig. 5. Reduction of TSS from leached wastewater: (a) TSS Final Concentration (mg/L) and (b) TSS Efficiency (%).

On day three, the TSS removal in R1 was 20 mg/L, whereas the reactors with EM4 showed slightly higher reductions: 20 mg/L in R2, 29 mg/L in R3, and 31 mg/L in R4. On day six, the TSS reduction increased slightly across all reactors: R1 reduced TSS by 31 mg/L, whereas R2, R3, and R4 reduced TSS by 29 mg/L, 35 mg/L, and 37 mg/L, respectively. On day nine, the TSS reduction increased significantly, with R1 at 51 mg/L, R2 at 57 mg/L, R3 at 58 mg/L, and R4 at 60 mg/L, respectively. These data show that although all reactors experienced a decrease in TSS, reactors with EM4 had a greater reduction in TSS than R1. Figure 5b shows that R1 had the lowest TSS removal efficiency compared to the reactors using EM4. On day three, R1's efficiency was 12.66%, whereas that of reactors with EM4 was slightly higher, with R3 and R4 reaching 18.35% and 19.62%, respectively. By day six, the efficiency increased for all reactors: R1, 19.62%; R2, 18.35%; R3, 22.15%; and R4, 23.42%. On day nine, the efficiency increased: R1 reached 32.28%, while R2, R3, and R4 reached 36.08%, 36.71%, and 37.97%, respectively. These data indicate that the reactor without EM4 (R1) was suboptimal for TSS removal, whereas reactors with EM4 demonstrated more stable and effective removal rates.

The findings are consistent with previous studies that emphasize the roles of phytoremediation and microbial augmentation in reducing TSS. It has been reported that *Pistia stratiotes* effectively filters suspended solids through its dense root system, which retains particles and promotes sedimentation [33]. Additionally, it highlights the role of plant roots as a medium for microbial growth, which facilitates the breakdown of suspended particles [17]. The enhanced TSS reduction observed with the addition of EM4 aligns with previous findings that microbial augmentation accelerates coagulation and sedimentation in wastewater. The addition of EM4 accelerates coagulation and sedimentation in wastewater treatment by promoting microbial flocculation, thereby enhancing floc stability and improving TSS removal [34]. However, compared with previous studies, the 38% TSS reduction achieved in RF4 indicates higher efficiency, likely due to the synergistic effects of *Pistia stratiotes* and the microbial bioactivators in EM4. This underscores the advantage of integrating plant-based and microbial treatment systems for effective wastewater remediation.

Reducing TSS in landfill leachate is crucial for enhancing water quality and mitigating the risk of environmental pollution. The study demonstrates that integrating *Pistia stratiotes* and EM4 achieves significant reductions in TSS, thereby ensuring compliance with regulatory standards for reactors R3 and R4. This highlights the effectiveness of the combined treatment approach in addressing a key pollutant in landfill leachate. These findings have practical implications for the design of sustainable wastewater treatment systems. By optimizing EM4 dosage and residence time, this approach offers a cost-effective and environmentally friendly solution for TSS removal. Moreover, the ability to achieve regulatory compliance with relatively simple treatment methods makes this approach suitable for deployment in resource-constrained regions where advanced treatment technologies are unavailable. Future research could investigate the system's scalability and evaluate its performance across various leachate compositions.

3.6. *The effect of adding EM4 on the increase in DO level*

The amount of DO available to microorganisms during the biodegradation process of organic matter is crucial in wastewater treatment. Higher DO levels reflect better aerobic conditions, which are important for biological wastewater treatment. The initial DO concentration in the wastewater was 1.02 mg/L. The results showed that the DO level in each reactor increased with operating time, as shown in Figure 6. As shown in Figure 6a, on the 3rd day, the increase in DO levels in R1 (without EM4) reached 3.08 mg/L, whereas the reactors with EM4 showed higher increases: 3.2 mg/L in R2, 3.7 mg/L in R3, and 3.9 mg/L in R4. On the 6th day, DO levels continued to increase: 4.02 mg/L in R1, 4.15 mg/L in R2, 4.2 mg/L in R3, and 5.1 mg/L in R4. On the 9th day, DO levels reached their highest point: R1 at 4.1 mg/L, R2 at 4.7 mg/L, R3 at 4.9 mg/L, and R4 at 6.0 mg/L. These results indicate that higher EM4 concentrations result in greater increases in DO levels in wastewater. This may indicate that the addition of EM4 encourages aerobic microbial activity, thereby increasing DO as part of the biological metabolic process. Figure 6b shows the percentage increase in DO levels compared to the initial value (1.02 mg/L). Reactors with higher EM4 concentrations exhibit faster and greater DO increases. On the 3rd day, the DO increase in R1 reached 201.96%, while reactors with EM4 showed higher increases: 213.73% in R2, 262.75% in R3, and 282.35% in R4. By the 6th day, DO increases were greater: R1 by 294.12%, R2 by 306.86%, R3 by 311.76%, and R4 by 400%. On the 9th day, DO levels peaked with R1 at 331.37%, R2 at 360.78%, R3 at 380.39%, and R4 at 488.24%. These results demonstrate that adding EM4 significantly accelerates the increase in DO levels in wastewater, thereby improving aerobic conditions in wastewater treatment systems.

The results align with previous studies demonstrating that the introduction of microbial bioactivators enhances wastewater oxygenation. It has been reported that the addition of EM4 increases microbial respiration and decomposition of organic matter, leading to higher DO levels [14]. Furthermore, it highlights that *Pistia stratiotes* contributes to oxygen production through photosynthesis, creating a favorable environment for aerobic microbial activity [31].

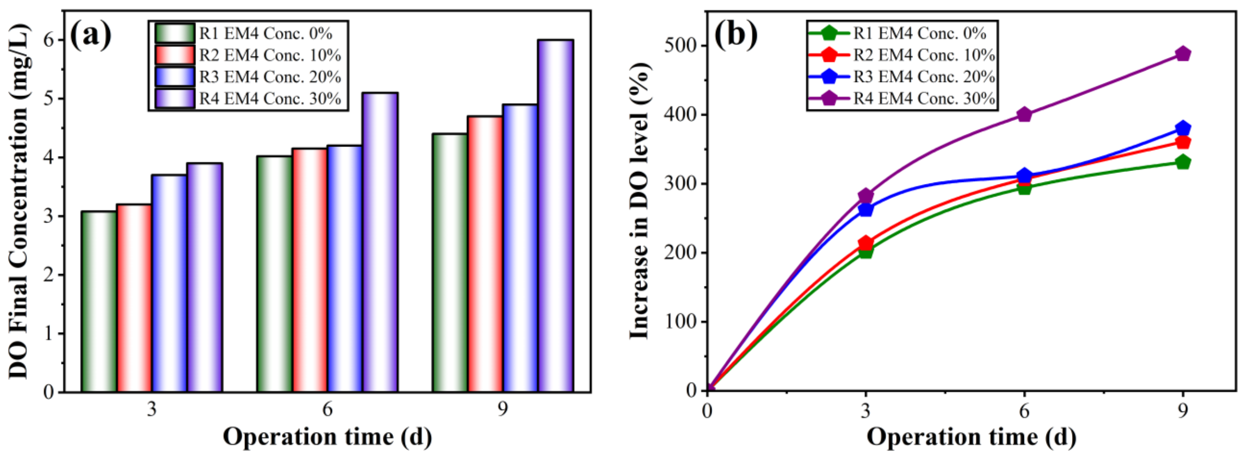

Fig. 6. The increase in the DO level: (a) DO Final Concentration (mg/L) and (b) Increase in DO level (%).

C Compared to earlier findings; the DO level of 6 mg/L achieved in R4 exceeds the typical range reported for phytoremediation systems without microbial augmentation. For example, liquid waste with *Pistia stratiotes* plants increases DO levels to between 6.8 and 7.2 mg/L [35]. This indicates that integrating EM4 and *Pistia stratiotes* provides a synergistic effect, enhancing oxygenation and more effectively supporting microbial metabolism. The significant increase in DO levels observed in all reactors underscores the effectiveness of combining *Pistia stratiotes* and EM4 in improving leachate quality. Enhanced oxygenation supports microbial degradation of organic matter and reduces ecological risks associated with the discharge of oxygen-depleted leachate into water bodies.

The findings emphasize the importance of optimizing EM4 dosage and residence time to maximize DO levels, which are critical for sustaining microbial activity and achieving efficient pollutant removal. The ability to achieve a DO level of 6 mg/L, as observed in R4, suggests that this integrated approach is a viable and sustainable solution for leachate treatment, particularly in regions with limited access to advanced treatment technologies. Future research could explore scaling this method for larger applications and assessing its long-term operational stability.

Although the combined *Pistia stratiotes* EM4 system demonstrated improved pollutant removal compared to plant-only treatments, several limitations should be acknowledged. This study did not evaluate heavy metal removal, which is an essential concern in landfill leachate management. Additionally, the experimental duration was limited to a 9-day batch system, and the performance under long-term or continuous-flow conditions remains unknown. Scaling this approach to field applications may also present challenges related to land availability, biomass management, and operational control.

4. Conclusion

This study demonstrates the synergistic effectiveness of integrating phytoremediation using *Pistia stratiotes* and microbial bioactivation with EM4 for landfill leachate treatment. The combined system effectively reduced BOD, COD, and TSS while significantly increasing DO levels, with optimal performance achieved at a 30% EM4 dosage and a nine-day residence time. These results confirm the potential of this approach as a cost-effective and environmentally sustainable alternative for improving leachate quality and mitigating environmental risks. From a practical perspective, this combined phytoremediation, EM4 system may be applied as a low-cost pretreatment or polishing step in landfill leachate management, particularly in regions with limited access to advanced wastewater treatment technologies. However, considerations regarding land availability, biomass handling, and operational control should be addressed. Despite its promising results, further studies are recommended to scale this method for large-scale applications, optimize operational parameters, and ensure compliance with regulatory discharge standards. Future research should also examine long-term system stability, plant-microbe interactions, heavy metal removal, and the performance of continuous-flow configurations.

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Author's contribution

Indah Nurhayati: Conceptualization, Methodology, Writing – Original Draft. **Evi Afifah:** Formal Analysis, Data Curation. **Muhammad Al Kholif:** Conceptualization, Methodology, Validation, Data Curation, Writing – Original Draft

Conflict of interest

No potential conflict of interest was reported by the authors.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

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