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Identification and Screening of Enzymatic Activity of Degrading Fungi Microplastics in Three Final Processing Sites (TPA) in the Province Lampung

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Abstract

One of the main causes of problems in the Final Processing Site (TPA) of Lampung Province is the increasing pile of plastic waste produced by human activities. This can be a source of microplastic pollution and is dangerous if it settles in the body of organisms. Fungi can be an alternative to help the natural biodegradation process reduce microplastic pollution in the soil ecosystem. This study aims to identify and characterize fungal isolates that can degrade microplastics in the Final Processing Site (TPA) of Lampung Province. The sampling method was purposive sampling. The growing colonies were identified by observing the clear zone, then characterized morphologically. Data analysis using qualitative descriptive methods and measuring the clear zone index. This study successfully identified 13 fungal isolates that showed clear zone areas. Four isolates (Ba1.2, Ba2.5, Bu3.4, Ka2.3) could degrade three types of microplastics. The highest clear zone: Bu3.4 for PET (2,00 mm), Ka2.4 for PE (1,50 mm), and Ba2.3 and Ba2.5 for PP (1,25 mm). Identification and morphological characterization showed 8 isolates belonging to the genus *Aspergillus*, 3 isolates of *Penicillium*, 1 isolate of *Fusarium*, and 1 isolate of *Sclerotium*. Degradation occurs through hyphal adhesion and secretion of polymer-degrading enzymes.

Keywords: biodegradation; clear zone index; fungi; landfill; microplastics.

INTRODUCTION

Lampung Province faces significant problems in waste management in several landfills, such as Bakung Landfill (Bandar Lampung City), Bumiayu Landfill (Pringsewu Regency), and Karangrejo Landfill (Metro City) (Umayasari & Sandy, 2024). Operational facilities are old and unable to accommodate the increasing volume of waste (Supratikno et al., 2023). Although designed with a sanitary landfill system, the current implementation of the open dumping system hurts the environment and surrounding communities (Malihah et al., 2023).

Plastic is the main contributor to the problem in the Lampung Province TPA (Nisa et al., 2023). Plastic waste comes from various human activities, such as packaging, household appliances, and disposable products. Types of plastic such as polyethylene terephthalate, polyethylene, polypropylene, polyvinyl chloride, polystyrene, and nylon (Shadrina, 2024). This type of plastic is carcinogenic and contains hazardous chemicals, such as dibenzodioxins and Polychlorinated dibenzofurans which can cause cancer (Fathulloh et al., 2021). The accumulation of potential plastic that exceeds capacity not only hinders management efficiency, but also has the potential to create microplastics that endanger the ecosystem and the health of organisms (Shafira et al., 2022).

Microplastics are plastic fragments with a diameter of <5mm which are divided into two, namely primary microplastics and secondary microplastics (Jamika, 2023). Primary microplastics are plastic products intentionally made microscopic, such as microbeads found in skin care products (Trivantira, 2022). Secondary microplastics result from the fragmentation of larger plastics in the form of fibers, fragments, granules, and films (Syahadatina et al., 2024).

Febriyanti et al., (2024), stated that plastic contributes to microplastic pollution in various ecosystems, including soil and water. Plastic waste not only remains on the surface of the soil, but is also degraded into microplastics by wind and the activity of living organisms below the surface (Suharsono et al., 2021). Microplastics that accumulate in landfill soil can migrate through air, water, wind, dust, erosion, and surface water runoff creating a chain impact in the food chain (Rezaei et al., 2019). Microplastics take hundreds of years to decompose through various physical, chemical and biological processes.

Based on the results of the VOSviewer analysis, it is known that the problem of microplastics is still largely unresolved. One potential solution is to accelerate the degradation of microplastics through biological agents, such as microorganisms. Fungi are known to break down microplastics into simple compounds with the help of specific enzymes, thus offering a more sustainable approach to reducing the impact of microplastic pollution. The role of soil microorganisms, especially fungi as agents of microplastic biodegradation still requires further development (Arista, 2023). Previous studies have shown the potential of indigenous fungal isolates, such as *Aspergillus* sp., *Penicillium* sp., and *Fusarium* sp. in degrading microplastics (Karimah, 2023; Magnin et al., 2020; Najah, 2022; Rohmah et al., 2019). Therefore, this study aims to identify and characterize fungal isolates that can degrade microplastics at the Final Processing Site (TPA) of Lampung Province.

MATERIALS AND METHODS

Materials

The equipment used is spades, clip bags, *Lion Star* brand cooler boxes, *Hirayama* brand autoclaves, *Pyrex* brand petri dishes, *Pyrex* brand beakers, *Pyrex* brand measuring cups, *OneLab* brand Erlenmeyer flasks, *Pyrex* brand test tubes, test tube racks, ose needles, *OneLab* brand object glass, *OneLab* brand cover glass, *Biosan* brand hot plates, *Polygon* brand magnetic stirrer bars, tweezers, bunsen lamps, *Pyrex* brand glass droppers, *Eppendorf* brand micropipettes, *Eppendorf* brand blue tips, *Kern* brand analytical scales, *Thermo Scientific* brand vortex ovens, *Thermo Scientific* brand incubators, *ESCO* brand Laminar Air Flow and *Optika Italia* brand binocular microscopes. The materials used include soil samples from three different landfills, sterile distilled water, alcohol, *Paseo* brand tissue, *Joyko* brand label paper, wrapping paper, cotton pads, *Whatman* brand filter paper, aluminum foil, plastic wrap, gauze, 70% alcohol *OneMed* brand, *Himedia* brand instant Potato Dextrose Agar (PDA) media, Minimal Salt Medium (MSM) media, HUFA brand chloramphenicol, polyethylene terephthalate (PET) plastics, polyethylene (PE), polypropylene (PP) and Lactophenol Cotton Blue (LPCB).

Methods

Soil Sample Preparation

Sampling used the purposive sampling method (Roflin & Liberty, 2021). Soil samples were taken as much as 1 kg in an area surrounded by decomposed plastic waste with a depth of 11-15 cm (Karimah, 2023). Samples were stored in a cooler box using labeled plastic (Kahula et al., 2024).

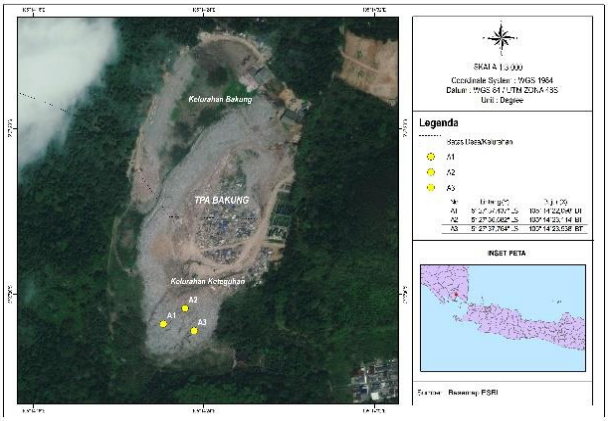


Figure 1 The sampling location is at Bakung Landfill, Bandar Lampung City.

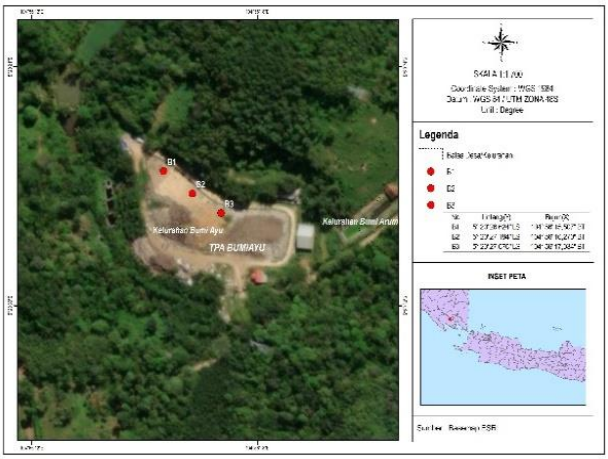


Figure 2. The sampling location is at Bumiayu TPA, Pringsewu Regency.

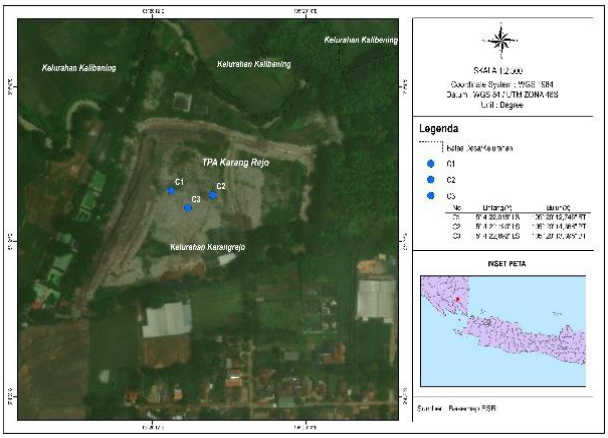


Figure 3. The sampling location is at Karangrejo TPA, Metro City.

Making PDA Media and Modifying MSM

The cultivation media used in the study consisted of Potato Dextrose Agar (PDA) and Mineral Salt Medium (MSM). PDA media was made with 39 grams of instant PDA in 1000 mL of aquades, added with chloramphenicol as an antibiotic (Adiningrum, 2024). MSM media is made with a composition of ingredients,

such as K_2HPO_4 (1 g), KH_2PO_4 (0.2 g), NaCl (1 g), $(NH_4)_2SO_4$ (1 g), $MgSO_4.7H_2O$ (0.5 g), $CaCl_2 \cdot 2H_2O$ (0.002 g), $FeSO_4.7H_2O$ (0.01 g), $MnSO_4.H_2O$ (0.001 g), $CuSO_4.5H_2O$ (0.001 g), $ZnSO_4.7H_2O$ (0.001 g), chloramphenicol, agar (15 g) in 1000 mL of distilled water (Wardani, 2021). The media is homogenized on a magnetic hot plate stirrer and autoclaved at 121°C and 1-2 atm for 20 minutes (Basarang et al., 2018). MSM media was modified by adding microplastic powder, namely polyethylene terephthalate (PET), polyethylene (PE), and polypropylene (PP) which had been sterilized by soaking in alcohol for 30 minutes, rinsing with distilled water for 20 minutes, and airing in laminar air flow for 15 minutes (Al Ikhsani, 2021).

Isolation of Microplastic Degrading Fungi

A soil sample weighing 1 gram was prepared in a test tube and diluted to 10 mL with sterile distilled water. The suspension was homogenized and serially diluted up to eight (10-8) for dry, non-agglomerated soil samples (Karimah, 2023). Samples from dilutions 10-5, 10-6, 10-7, and 10-8 were inoculated on PDA media with a volume of 1 mL using the pour plate method. Incubate for 3–7 days at room temperature (Abna et al., 2024). Selected fungal colonies were purified on new PDA media.

Screening of Enzymatic Activity of Microplastic Degrading Fungi

The degradation potential was tested using MSM agar media with a microplastic powder content of 1% (w/v). Fungal isolates were inoculated at 30-35°C for 8-10 days. The clear area around the colony is characterized by rapid growth and spread over the surface of the medium. This shows the potential of this fungus as a decomposer (Wardani, 2021).

Identification of Fungal Morphology

Fungal identification is carried out through macroscopic and microscopic observations. Macroscopic observations were carried out directly on fungal colonies grown for 7 days, including the characteristics of the shape, color and texture of the colony (Novitasari et al., 2021). The slide culture method with Lactophenol Cotton Blue (LPCB) staining was used for microscopic observations (Fitria et al., 2023). Pieces of media are placed on microscope slides. The fungal isolate was streaked around the corner of the media with a loop needle, and covered with a sterile cover glass. The slide is placed in a petri dish with filter paper and a slide holder. An incubation period of 3-5 days will see the growth of fungus around the media. The cover glass of the culture slide was stained with Lactophenol Cotton Blue (LPCB) dye and placed on an object glass with 100x magnification for genus identification (Fahmi et al., 2021). Characters that can be observed include the shape of the hyphae, branching patterns, spore structure, the presence of septa on the hyphae, exudates and water droplets.

Data analysis

Data analysis includes data obtained from field observations and laboratory results. The data was analyzed descriptively qualitatively, then presented as tables or pictures of the results of macroscopic and microscopic morphological characterization, as well as the formation of clear zones for each fungal isolate (Rohmah et al., 2019). Clear zone index measurements were carried out by measuring the enzymatic index value on the diameter of the clear zone formed around fungal colonies on MSM media containing microplastic powder (Najah, 2022).

$$\mathrm{EI} = \frac{\text{Diameter koloni} + \text{zona bening}}{\text{Diameter koloni}}$$

RESULTS AND DISCUSSION

Results

Isolation of Microplastic Degrading Fungi

Isolation of fungi from three landfills produced 15 isolates with different morphologies, of which 13 isolates showed the potential for microplastic degradation through the formation of clear zones, with the degradation process involving extracellular enzymes such as laccase, peroxidase, and lipase whose effectiveness is influenced by environmental factors and the physicochemical characteristics of microplastics.

Screening of Enzymatic Activity of Microplastic Degrading Fungi

The results of enzymatic activity screening of 13 fungal isolates from Bakung TPA (Ba), Bumiayu TPA (Bu), and Karangrejo TPA (Ka) showed a high degradation ability. Varies against polyethylene terephthalate (PET), polyethylene (PE), and polypropylene (PP) microplastics. This variation in ability is caused by differences in the enzymatic mechanisms possessed by each fungal isolate. The degradation ability is identified by forming a clear zone on MSM agar media, indicating the activity of extracellular hydrolytic enzymes (Istiqomah, 2020).

Table 1. Clear zone diameter by fungal isolates on MSM media.

Isolate Code	Diameter (mm) Clear Zone		
	PET	**PE**	**PP**
Ba1.2	1,33	1,05	1,19
Ba2.3	1,27	-	-
Ba2.4	1,03	-	-
Ba2.5	1,04	1,40	1,25
Ba3.6	-	1,25	1,17
Bu1.1	1,06	-	-
Bu2.2	1,21	-	1,17
Bu2.3	1,22	-	-
Bu3.4	2,00	1,21	1,00
Ka1.1	1,05	-	-
Ka2.3	1,25	1,05	1,08
Ka2,4	-	1,50	1,07
Ka3.5	1,10	-	-

Note**:** PET: Polyethylene terephthalate; (PE) Polyethylene; (PP) Polypropylene.

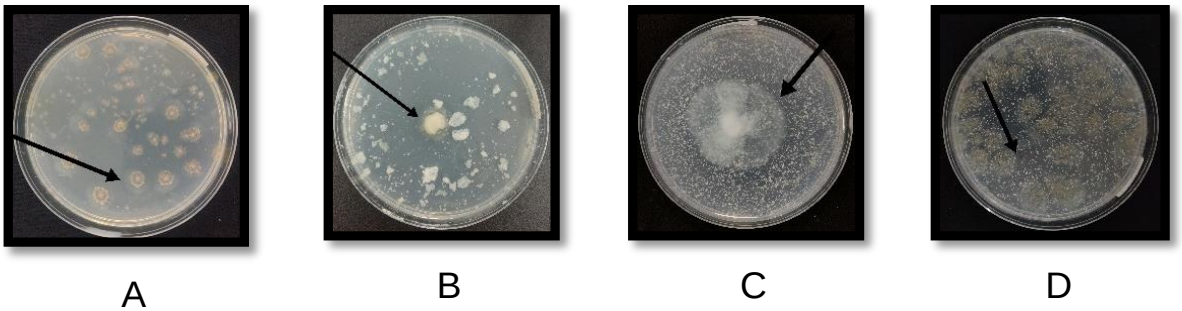


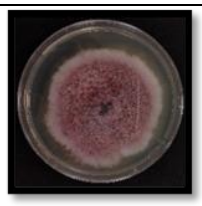
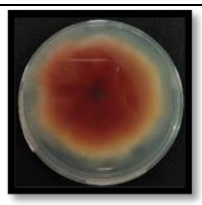
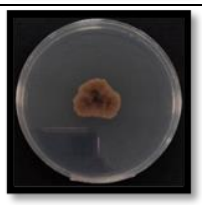
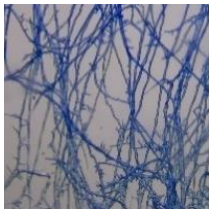
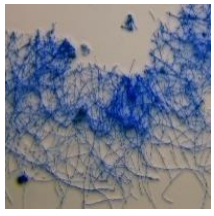
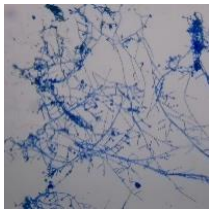
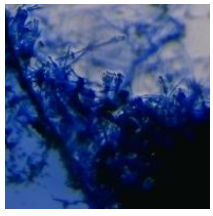
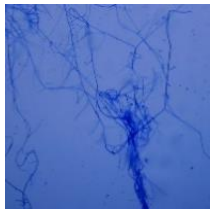
Figure 4. Fungal isolates showing the highest clear zone according to the content of microplastic types: (A) Isolate Bu3.4 the highest clear zone for PET reached 2,00 mm (B) Isolate Ka2.4 the highest clear zone for PE reached 1,50 mm (C) Isolate Ba2.3 the clear zone for PP reached 1,25 mm (D) Isolate Ba2.5 the clear zone for PP reached 1,25 mm.

Discussion

Isolation of Microplastic Degrading Fungi

The morphological characteristics observed in 13 isolates of microplastic-degrading fungi on PDA media showed colony diversity and could be grouped into four main suspected genera based on typical macroscopic and microscopic characteristics. Morphological identification is an important step in taxonomic characterization and understanding the biodegradation potential of fungal isolates (Haya, 2023). Based on morphological observations of microplastic degradation mushroom isolates, the result of characterization is obtained as follows:

Table 2. Characterization of Microplastic-Degrading Fungi Codes Ba1.2 – Ba3.6.

Character	**Isolate Code**				
Morphology	**Ba1.2**	**Ba2.3**	**Ba2.4**	**Ba2.5**	**Ba3.6**
Top view					
Bottom view					
Form	Circular	Circular	Circular	Circular	Circular
Margin	Entire	Entire	Entire	Undulate	Filaform
Texture	Cottony	Cottony	Velvety	Powdery	Cottony
Elevation	Raised	Raised	Raised	Umbonate	Flat
Exodus	Aerial	Aerial	Aerial	Aerial	Aerial
Character microscopic					
Suspected genus	*Fusarium*	*Aspergillus*	*Penicillium*	*Penicillium*	*Sclerotium*

The morphological characterization observations in Table 2 show that isolate Ba1.2 has a purplish pink color on the upper surface and reddish brown on the lower surface. Isolate Ba2.3 is white on the upper surface and yellowish white on the lower surface. Isolate Ba2.4 appears transparent with some structure in the center on the upper surface and has a brown color in the center on the lower surface. All three isolates have circular colony

shapes with entire (intact) edges. Isolates Ba1.2 and Ba2.3 have a cottony texture, while Ba2.4 has a velvet texture. All isolates show raised elevation with aerial hyphae growth. Microscopic observation identified Ba1.2 as a suspected genus *Fusarium* with visible septate hyphae and characteristic crescent-shaped (fusiform) macroconidia, Ba2.3 as suspected *Aspergillus* with round conidial heads, and Ba2.4 as suspected *Penicillium* with brush-like branched conidiophores. Isolate Ba2.5 has a green colony with yellow edges (top view) and reddish orange (bottom view), a circular shape with undulate (wavy) edges, powdery texture, and umbonate elevation. Microscopic observation identified it as the genus *Penicillium*. Isolate Ba3.6 has a whitish gray color, circular shape with filiform edges, cottony texture, and flat elevation, and is suspected as *Sclerotium* based on its characteristic sclerotia structure and the presence of mycelium with septate hyphae.

Table 3. Characterization of Microplastic-Degrading Fungi Codes Bu1.1 – Ka1.1 .

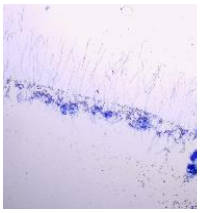
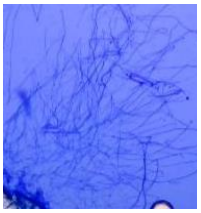
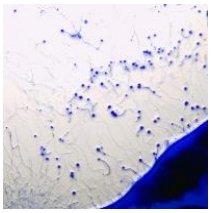
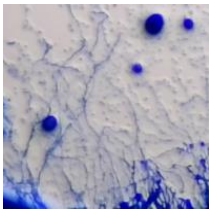
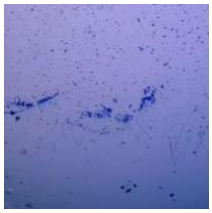
Character	**Isolate Code**				
Morphology	**Bu1.1**	**Bu2.2**	**Bu2.3**	**Bu3.4**	**Ka1.1**
Top view					
Bottom view					
Form	Filamentous	Circular	Circular	Circular	Circular
Margin	Filamentous	Entire	Entire	Entire	Filamentous
Texture	Velvety	Cottony	Powdery	Powdery	Powdery
Elevation	Raised	Raised	Convex	Convex	Raised
Exodus	Aerial	Submerged	Submerged	Submerged	Aerial
Character microscopic					
Suspected genus	*Aspergillus*	*Penicillium*	*Aspergillus*	*Aspergillus*	*Aspergillus*

Table 3 displays the morphological characterization observations of isolates Bu1.1, Bu2.2, Bu2.3, Bu3.4, and Ka1.1. Isolate Bu1.1 is blackish white with a spreading pattern, filamentous shape and edge, velvety texture, raised elevation, and suspected as *Aspergillus* with septate hyphae with upright conidiophores bearing conidial heads. Isolate Bu2.2 has a grayish white colony, circular shape with entire edge, cottony texture, raised elevation, and submerged hyphae growth. Microscopic observation identified it as *Penicillium*. Isolate Bu2.3 is reddish in the center and Bu3.4 shows a concentric circle pattern in orange. Both are circular with entire edges, powdery texture, convex elevation, submerged hyphae growth, and suspected as *Aspergillus*. Isolate Ka1.1 is black (top view) and dark gray with a circular pattern (bottom view), circular shape with filamentous edges, powdery texture, raised elevation, aerial hyphae growth, and suspected as *Aspergillus*.

Table 4. Characterization of Microplastic-Degrading Fungi Codes Ka2.3 – Ka3.5.

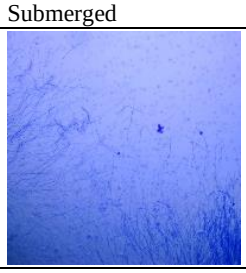
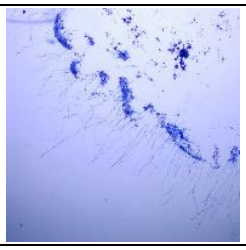
Character Morphology	Isolate Code		
	Ka2.3	**Ka2.4**	**Ka3.5**
Top view			
Bottom view			
Form	Filamentous	Circular	Filamentous
Margin	Filamenous	Entire	Filamentous
Texture	Powdery	Cottony	Powdery
Elevation	Flat	Raised	Flat
Exodus	Aerial	Submerged	Aerial
Character microscopic			
Suspected genus	*Aspergillus*	*Aspergillus*	*Aspergillus*

Table 4 shows the morphological characterization observations of isolates Ka2.3, Ka2.4, and Ka3.5. Isolate Ka1.1 is black (top view) and dark gray with a circular pattern (bottom view), circular shape with filamentous edges, powdery texture, raised elevation, and aerial hyphae growth. Isolate Ka2.3 is whitish brown to whitish gray, filamentous in shape and edge, powdery texture, flat elevation, and aerial hyphae growth. Isolate Ka2.4 is brownish green-gray with a dark center and cream brown on the bottom, circular shape with entire edge, cottony texture, raised elevation, and submerged hyphae growth. Isolate Ka3.5 has a brownish green colony with irregular pattern (top view) and whitish to grayish (bottom view). The colony shape and edge are filamentous, powdery texture, flat elevation, and aerial hyphae growth. This isolate is identified as genus *Aspergillus* on microscopic observation showing unbranched upright conidiophores with round conidial heads. All four isolates are suspected to be genus *Aspergillus*.

Discussion

Isolation of Microplastic Degrading Fungi

Isolation of fungi from soil samples of Bakung Landfill, Bumiayu Landfill, and Karangrejo Landfill was conducted at depths of 11-15 cm, using serial dilution methods (10^{-5} to 10^{-8}) and pour plate technique. The selection of 11-15 cm depth in soil microbiology research is based on previous studies showing that the composition and function of microbial communities are significantly influenced by soil depth, with significant changes in community structure and microbial activity as depth increases (Naylor et al., 2022).

The isolation process yielded 15 fungal isolates with different morphologies, with 13 isolate codes showing microplastic degradation potential by forming clear zones. Indigenous fungi from landfills have resistance to extreme conditions such as temperature fluctuations, high humidity, and the presence of diverse waste. Fungal growth is influenced by organic matter content, temperature, humidity, and soil pH (Nafiah, 2024).

The mechanism of microplastic degradation by fungi involves a gradual process that begins with colonization of the polymer surface, followed by secretion of extracellular enzymes that break down complex chemical bonds. The main enzymes involved in the degradation process include laccase, manganese peroxidase, alkane hydroxylase, lignin peroxidase, versatile peroxidase, and lipase which can break the chemical bonds of plastic

polymers (Azzahra, 2024). According to Rohmah et al., (2019) the isolate *Aspergillus terreus* (LM 1021) showed optimal degradation activity at pH 5 and a temperature of 25°C, capable of degrading PHB (44.96%), plastic bags (4.28%), and LDPE (4.9%). Another study stated that *Aspergillus niger* can degrade polyethylene up to 16% in 30 days (Safdar et al., 2024). Studies show that *Fusarium solani* can degrade PET by producing hydrolase enzymes (Erlambang et al., 2019).

The efficiency of microplastic biodegradation is influenced by the physicochemical characteristics of the polymer, including crystallinity, molecular weight, and functional groups. Polymers with high crystalline structures such as PET and HDPE tend to be more resistant to biological degradation compared to amorphous polymers such as PHB and PCL. Modification of microplastic surfaces through physical or chemical pretreatment can increase the rate of degradation by creating nucleation points for enzyme attachment. Optimal conditions for fungal growth are achieved on Potato Dextrose Agar (PDA) media with pH 4.5-5.6, temperature 24-29°C, and relative humidity 90-100% (Basarang et al., 2018). These parameters are important to optimize the cultivation of microplastic-degrading fungi in environmental biotechnology applications.

Enzymatic Activity Screening of Microplastic Degrading Fungi

Table 1 shows that the potential for degradation of polyethylene terephthalate (PET) is the type of microplastic that is most degraded with 11 isolates, followed by polypropylene (PP) with 8 isolates, and polyethylene (PE) with 6 isolates. This degradation pattern is consistent with the molecular characteristics of each related polymer. Polyethylene terephthalate (PET) has an ester bond that is relatively easier to hydrolyze by cutinase and enzymes generally produced by soil fungi. Polyethylene (PE) and polypropylene (PP) have a more stable and hydrophobic hydrocarbon structure, making them more difficult to degrade (Ningrum et al., 2023). Isolate Bu3.4 was able to degrade polyethylene terephthalate (PET) with the highest clear zone reaching 2,00 mm after 10 days of incubation. This ability can be attributed to the secretion of specific hydrolytic enzymes such as PETase and MHETase work synergistically to break the ester bond in the PET structure. The PETase enzyme catalyzes the hydrolysis of PET into MHET (mono-2-hydroxyethyl terephthalate), which is then converted into terephthalic acid and ethylene glycol by MHETase. The high degradation ability of the Bu3.4 isolate is in line with the research of Danso et al. (2019), which reported that several species of fungi from the genus *Fusarium* and *Humicola* can produce the enzyme A potent PETase. Isolate Ka2.4 showed superiority in degrading polyethylene (PE) with a clear zone of 1,50 mm. Polyethylene (PE) is known as a polymer that is very resistant to biological degradation because its saturated hydrocarbon chain structure does not have functional groups that are easily attacked by enzymes. The ability of Ka2.4 isolates to indicate the production of oxidative enzymes such as laccase or peroxidase that can oxidize the PE surface through the formation of carbonyl groups (Asmi, 2020). *Lysinibacillus xylanilyticus* and *Aspergillus niger* can degrade polyethylene plastic (Arista, 2023). Isolates Ba2.3 and Ba2.5 showed the ability to degrade polypropylene with a clear zone reaching a size of 1,25 mm. The mechanism of PP degradation by fungi begins with oxidoreductase enzymes such as laccase and peroxidase catalyzing the oxidation of the polymer surface, producing carbonyl and carboxyl groups on the PP chain. Monooxygenase and alkane hydroxylase enzymes insert oxygen atoms into the CH bond, forming primary alcohols which are then oxidized to aldehydes by alcohol dehydrogenase. The aldehyde dehydrogenase enzyme converts aldehydes to carboxylic acids, while esterases and lipases facilitate the cleavage of the polymer chain at the ester group formed (Nurmalasari & Kebumian, 2018). Figure 4 shows that there are four isolate codes (Ba2.3, Ba2.5, Bu3.4, and Ka2.4) that can potentially degrade the three types of samples tested. This multi-degradation ability is very valuable in biodegradation applications in overcoming microplastic pollution in the environment (Fachrul et al., 2021). The fungal isolates that have the highest clear zone according to the content of microplastic types are as follows: isolate code Bu3.4 with the highest clear zone for polyethylene terephthalate (PET) reaching 2,00 m, isolate code Ka2.4 with the highest clear zone for polypropylene (PE) reaching 1,50 mm and isolate codes Ba2.3 and Ba2.5 with the highest clear zone for polypropylene (PP) reaching 1,25 mm.

MSM agar media with 1% microplastic powder content with essential inorganic components such as mineral salts K_2HPO_4, KH_2PO_4, $(NH_4)_2SO_4$, $CaCl_2$), dan trace elements ($FeSO_4$, $ZnSO_4$, $MnSO_4$), but deliberately made deficient in organic carbon sources. The composition of this media forces fungi to develop an enzyme system that can degrade microplastics into monomer units that can be used as a source of carbon and energy (Wei & Zimmermann, 2017). The ability of microplastic biodegradation by fungi is identified by forming a clear zone around the colony on MSM media. This clear zone is seen as a transparent area that contrasts with the opaque area around it. MSM media adjusted at a pH range of 6.5-7.0 supports optimal activity of degradative enzymes such as laccase, peroxidase, and hydrolase produced by microplastic degrading fungi.

The relatively small size of the clear zone (1,00 mm to 2,00 mm) reflects the slow degradation process of microplastics in accordance with the recalcitrant nature of plastic polymers (Asmi et al., 2022). The rate of degradation is influenced by various factors including polymer crystallinity, molecular weight, particle size distribution, and the presence of additives such as plasticizers and UV stabilizers that can change the

accessibility of the substrate to enzymes. The time and size of the clear zone varied depending on the type of fungal isolate tested, with some isolates starting to form a clear zone from day 6, and the maximum size was reached after 14-21 days of incubation at 30°C (Wardani, 2021).

The formation of clear zones on nutrient-poor MSM agar media indicates that indigenous fungi from the landfill have adapted to use microplastics as an alternative carbon source. This adaptation involves developing an enzyme system that can degrade synthetic polymers into monomer units that can be utilized for mushroom growth. This adaptive ability makes indigenous fungi from landfills in Lampung Province potentially effective and sustainable biodegradation agents in overcoming microplastic pollution.

Morphological Identification of Microplastic Degrading Fungi

This research identified four suspected genera, namely *Aspergillus*, *Penicillium*, *Fusarium*, and *Sclerotium*. The *Aspergillus* genus dominates (isolates Ba2.3, Bu2.3, Bu3.4, Ka1.1, Ka2.3, Ka2.4, and Ka3.5) with the advantages of fast mycelial growth and production of antifungal compounds (Magnin et al. 2020; Wardani, 2021). *Aspergillus* produces laccase, esterase, peroxidase, lipase and urease enzymes which play a role in the degradation of complex polymers (Oliveira et al. 2020). *Penicillium* (isolates Ba2.4, Ba2.5, and Bu2.2) is the second most abundant genus, known to have significant synthetic polymer degradation capabilities through the production of enzymes such as cutinase, esterase, and lipase (Suresh et al., 2025). The typical morphological structure of *Penicillium* in the form of brush-like branched conidiophores makes it easier to identify microscopically.

Fusarium (isolate Ba1.2) with characteristic crescent-shaped (fusiform) macroconidia can degrade various polymers by producing laccase, peroxidase and lipase enzymes. *Sclerotium* (isolate Ba3.6) has the potential to degrade complex organic polymers via ligninolytic enzymes. According to Sakti et al. (2024), *Sclerotium rolfsii* has the potential to degrade complex organic polymers through the production of ligninolytic enzymes. The distinctive sclerotia structure is the main marker in identifying this genus. Although research on *Sclerotium*'s ability to degrade microplastics is still limited, the potential of its ligninolytic enzymes opens up new opportunities in the development of plastic biodegradation technology.

The dominant powdery texture in many isolates (Ba2.5, Bu2.3, Bu3.4, Ka1.1, Ka2.3, Ka3.5) is related to the production of large amounts of conidia which supports the spread and adaptation of the fungus to various environmental conditions. Aerial hyphal growth allows optimal oxygen access for oxidative reactions in polymer degradation, while submerged hyphal growth allows direct contact with the substrate for enzymatic degradation processes.

The morphological diversity of microplastic-degrading fungi found in this study provides insight into the different physiological adaptations to microplastic substrates. These isolates have the potential to have different degradation mechanisms and efficiencies. These four indigenous fungal genera have high adaptability and complex enzymatic systems, so they have the potential to be effective microplastic biodegradation agents for applications to restore plastic-polluted environments.

CONCLUSIONS

This study successfully isolated 15 fungal isolates with diverse morphologies from three landfills in Lampung Province, with 13 isolates showing microplastic degradation activity as indicated by the clear zone index value. PET type microplastics were degraded the most (11 isolates), followed by PP (8 isolates) and PE (6 isolates). Four isolates (Ba2.3, Ba2.5, Bu3.4, and Ka2.4) had the potential to degrade the three types of samples tested. The isolates with the highest clear zones were Bu3.4 for PET (2,00 mm), Ka2.4 for PE (1,50 mm), and Ba2.3 and Ba2.5 for PP (1,25 mm). Morphological characterization showed 8 isolates suspected of the *Aspergillus* genus, 3 isolates of *Penicillium*, 1 isolate of *Fusarium*, and 1 isolate of *Sclerotium*. Microplastic degradation involves hyphal adhesion to the plastic surface and the secretion of extracellular enzymes that break down the chemical bonds of the plastic polymer. Indigenous fungi from landfills in Lampung Province have the potential to be effective and sustainable biodegradation agents in overcoming microplastic pollution in the environment.

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