ISSN 2541-223X
Volume 4 2022

POTENCY OF INDIGENOUS MICROBES OF PIYUNGAN LANDFILL YOGYAKARTA FOR MICROPLASTIC BIODEGRADATION

A S Putri[1], S Najah[1], A N Lathifah[1,3*], A A Asmara[1,2], D Wulandari[1,3]

[1] Environmental Engineering Department, Universitas Islam Indonesia, Yogyakarta 55584, Indonesia
[2] Center for Environmental Studies, Universitas Islam Indonesia, Yogyakarta 55584, Indonesia
[3] Environmental Biotechnology Laboratory, Universitas Islam Indonesia, Yogyakarta 55584, Indonesia

ABSTRACT
Research on the bacteria and fungi potentially degrading microplastic in Piyungan Landfill has not been done much. Meanwhile, plastic pollution is one of many environmental problems faced in the Special Region of Yogyakarta. The accumulation of plastic waste is getting more significant daily, and the capacity of landfills is decreasing. Bacterial and fungal testing was carried out by taking soil samples from landfills and isolating dilute Nutrient Broth, Malt Extract Agar, and Nutrient Agar media. Bacterial and fungal were identified by morphology colony and cell, then tested for plastic biodegradation by selected bacterial and fungal. The result of this research, there are 18 bacterial isolate and five isolate fungi, but for the degradation testing, chosen just three bacterial (Isolate A, F, J) degrade Polypropylene for ±30 days and three fungal (Isolate PA, PK, RJ) to degrade LDPE for ±14 days. The indicator for determining plastic degradation is clear-zone formation. The test result showed that isolate F, which had similarity with bacteria in the genus of *Micrococcus*, had a higher potential to degrade microplastic with clear-zone formation ±1.23 mm. Meanwhile, fungal isolate PK, similar to *Penicillium sp.*, had a higher potential to degrade microplastic, marked by the most significant clear-zone appearance, ±2.3 cm.

Keywords: Indigenous Microbes, Piyungan landfill, Microplastic biodegradation

1. INTRODUCTION
The global problem that is often recognized by the public is plastic pollution. In 2016, plastic production reached 322 million tons of plastic fibre manufacture [1]. The resistant nature of plastic makes it complex and lengthy for the degradation process by nature [2]. Plastic in micro-size has many types, shapes, and sizes, one of which is microplastic. Microplastic is a type of plastic waste that has a size smaller than 5 mm [3]. Microplastics have persistent properties and contain toxic or carcinogenic chemical elements. The compound can seep into the soil through leachate and enter the food chain, impacting human health and the environment [4]. A study by Utami and Liani [5] found that samples of ground-well water in the Piyungan landfill area contained microplastics. The highest microplastic was found in an area of 0 – 1 km in Lingkong Village at 146 ± 109 particles/L. These microplastics have Polystyrene (PS) and Polyvinyl Chloride (PVC) polymers. If the use of plastic continues but is not balanced with the calculation of the

[1*] Corresponding author's email: annisa.lathifah@uii.ac.id
DOI: https://doi.org/10.20885/icsbe.vol4.art44

length of the plastic decomposition process, the high level of plastic consumption, and the reduced availability of landfill land as a last resort in waste disposal, the problem of waste generation will not be avoided [2].

One solution to overcome microplastic pollution is the bioremediation method using microorganisms to degrade microplastics. Bioremediation is a way to clean up pollutants that is not difficult to do and has a low cost. Biodegradation by microorganisms can be obtained from an area polluted by plastic [6]. Previous research has shown that bacteria and fungi can be used as plastic biodegradation agents. It is known that the bacteria *Micrococcus rubidea, Pseudomonas putida, Micrococcus marcescens,* and *Vibrio fluvialis* have a plastic degradation ability of 94%-98%. On the other hand, the fungus that can degrade plastic is *Aspergillus terreus*, whose capability is 3.25% [7][8]. Research by Wardani [9] also proved that fungi in the genus *Aspergillus* could degrade LDPE by 5.7% in 30 days.

In 2018, the waste at the Piyungan landfill reached 500,000 tons or 549.74 tons/day. Plastic waste in the Piyungan landfill that has been piled up for a long time will break down into microplastics and contaminate water sources and the surrounding environment. In addition, the cattle grazing at the Piyungan landfill, the cows there often eat plastic, which is very dangerous for the health of animals and the food chain of organisms [10]. Based on the problems that occurred, this study was conducted to explore bacteria and fungi from the Piyungan landfill, which have the potential to degrade microplastics. The results of this study are expected to help provide information on the process of handling plastic waste by utilising microorganisms in the Piyungan landfill.

2. MATERIALS AND METHODS

2.1. Study sites and sampling locations

Soil samples were taken from the Piyungan landfill which is located at Ngablak, Sitimulyo Village, Piyungan District, Bantul Regency, Special Region of Yogyakarta Province. The sampling points were presented in Figure 1.

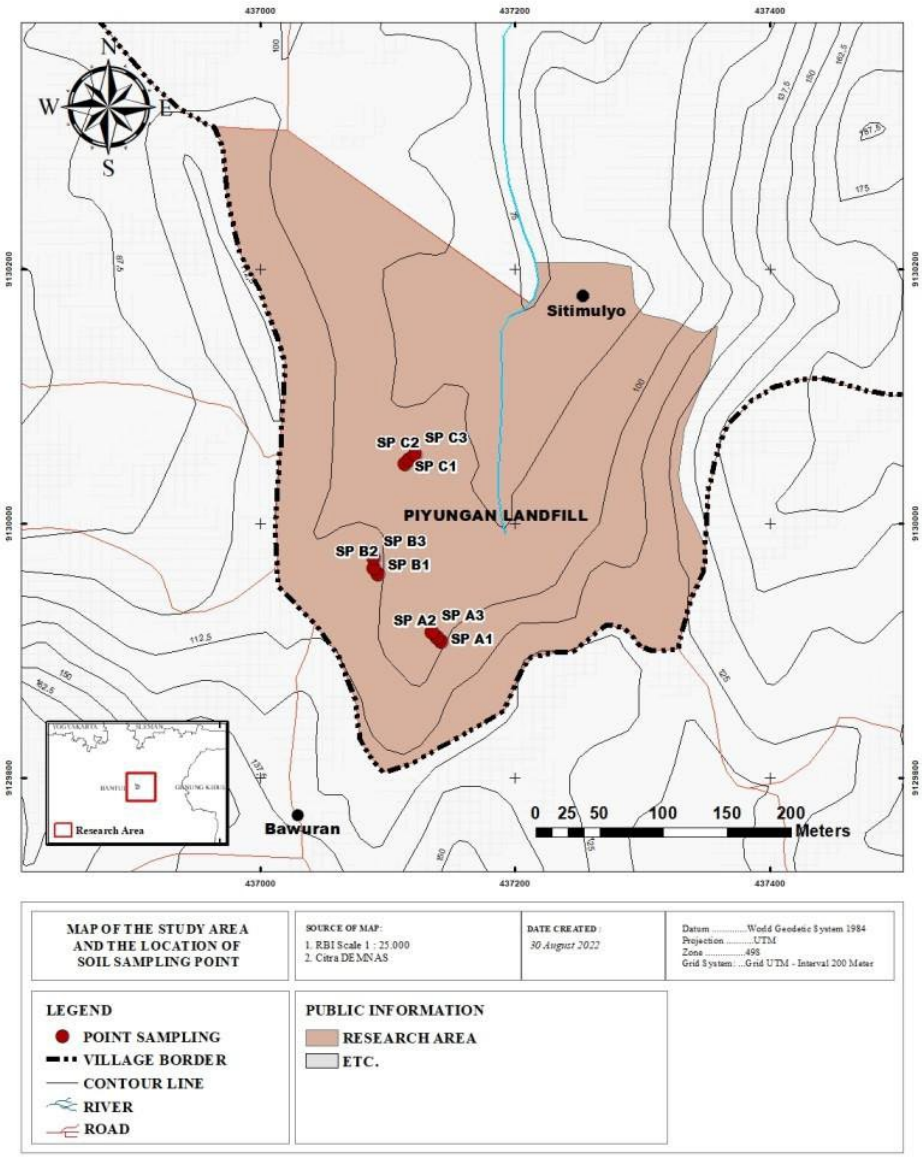


Figure 1 Sampling Points in the Piyungan Landfill

2.2. Sample preparation

Soil samples were taken using the grab sampling method at three sampling points (SP1, SP2, and SP3). Samples were collected on March, 8th 2022 from three points (15 cm in depth) with 1 m interval between them (Figure 2). The samples were kept in a cooler box containing an ice pack [11]. Finally the samples were stored in a refrigerator at 4°C for further analysis [12].

ISSN 2541-223X
Volume 4 2022

Table 1 Coordinates of Sampling Points

Sampling points	1	2	3
A	110 25'47.2"S 7 52'16"E	110 25'47.1"S 7 52'15.9"E	110 25'47"S 7 52'15.8"E
B	110 25'45.6"S 7 52'14.3"E	110 25'45.5"S 7 52'14.1"E	110 25'45.5"S 7 52'13.9"E
C	110 25'46.3"S 7 52'15.5"E	110 25'46.4"S 7 52'11.4"E	110 25'46.6"S 7 52'11.2"E

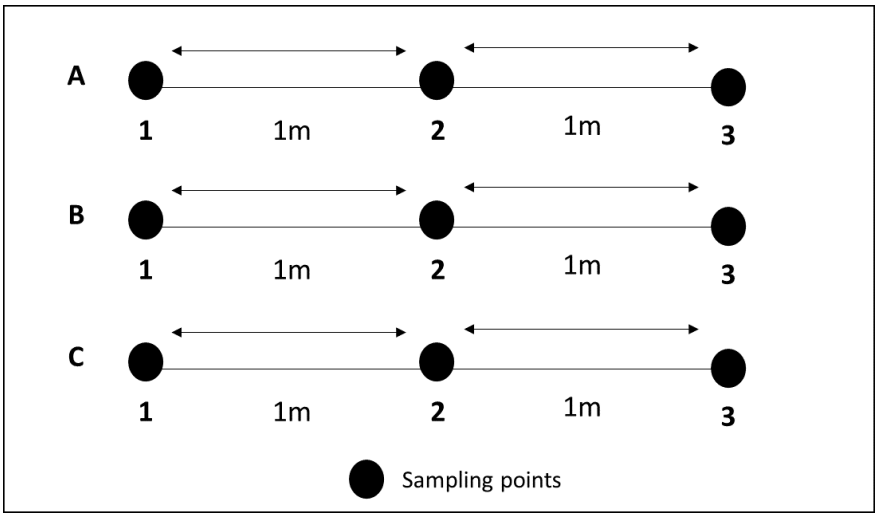


Figure 2 Interval Sampling Illustration

2.3. Media Preparation

In this study three kinds of media were used namely *Dilute Nutrient Broth* (DNB) agar, *Malt Extract Agar* (MEA) (MERCK, Germany), and *Nutrient agar* (NA) (MERCK, Germany). DNB agar and MEA were used for bacterial and fungal isolation, respectively. NA media were used for isolation cultivation and plastic biodegradation analysis. MEA and NA media were made according to the manufacturer's instructions. While DNB agar media were made by 100-fold diluted *Nutrient Broth* (NB) media (OXOID, UK) adding with 2% *Bacto agar* (OXOID, UK).

2.4. Isolation of Bacteria and Fungi

In this study, bacterial and fungal isolation was carried out using the *Direct Plating* method. In brief, the soil samples (5 g fresh weight) were suspended in 45 ml of *Buffered Peptone Water* (BPW) media (MERCK, Germany) and vigorously mixed for 6 minutes. The serial dilution of the samples was made until 10-7. Further, serial dilution from 10-4 to 10-7 was used for subsequent procedures [13]. Serially diluted samples (1 ml aliquots) were plated on DNB agar and MEA media using the pour plate method. The DNB media were used for bacterial isolation, while MEA media were used for fungal isolation. After 14 days of incubation, bacterial and fungal colonies were counted using a colony counter and recultured onto NA media to obtain pure cultures [14].

2.5. Bacterial and Fungal Identification

2.5.1 Bacterial morphology

In this study, bacterial and fungal identification was carried out based on morphological characterization. Observation of the morphology of the bacterial colonies was carried out using a colony counter to clarify the identification of the shape, colour, elevation, and

margins of the colonies, as well as to calculate the total bacterial colonies based on colony morphology. Furthermore, the identification of colony morphology was compared with the reference [15]. Further, the gram staining test was conducted to identify the bacterial cell by clarifying the cell shape and gram type.. The gram staining test was performed according to Singh *et al*. [16].

2.5.2 Fungal morphology

According to Barneth [17][18], fungal morphology was identified by macroscopic and microscopic observation. Fungal observations identified the fungal colonies' diameters, colony structure, and colour according to Hartanti [18]. In contrast, microscopic observations identified conidia, spores, hyphae, conidiophores, and sporangia, according to Iramayana [19]. Fungal microscopic identification was conducted using slide culture and *lactophenol cotton blue* (LPCB) staining. In brief, the sliced media was put on the observation glass, then a small number of fungi were placed in the media using a needle and scratched around. Further, the media were placed in a petri dish with cotton and distilled water. The media on Petri dishes were incubated for 3-5 days [20]. After the incubation, the fungal mycelium will appear at the edge of the agar. Further, the LPCB staining method was used to observe the fungal component, according to Hulse [21] [22].

2.6. Plastic Biodegradation Analysis

2.6.1 Plastic Biodegradation using Bacterial isolates

The ability of bacterial isolates to degrade plastic was observed by clear-zone/halo zone formation in the NA media containing plastic fragments. The type of plastic, namely Polypropylene (PP), was used. The degradation test begins with sterilised plastic with 70% alcohol (soaked for 1 hour), sprayed with distilled water, and then air-dried. Next, the plastic was inoculated on a petri dish with NA media which previously contained the selected bacterial isolates. Then it was incubated at 30°C for approximately 14 days. After that, the sample was dripped with Congo red reagent (pH adjusted to 6.7), then continued incubating for about 14 days to see the clear-zone formation around the plastic. Next, the clear zone was measured using a millimetre ruler [7].

2.6.2 Plastic Biodegradation using Fungal isolates

The ability of fungal isolates to degrade plastic was carried out using low-density polyethylene (LDPE) powder in minimal nutrient media divided into 1% LDPE and 5% LDPE. The LDPE powder and media were autoclaved at 121˚C for 15 minutes and then placed on a petri dish. A small number of fungal isolates were inoculated at the centre of the media, then incubated at 28˚C for seven days. After that, the clear-zone formation around the colonies was observed. The isolates with maximum clear-zone diameter were targeted as microplastic degraders [23].

3. RESULT AND DISCUSSIONS

3.1. Characteristics of Bacterial and Fungal Isolates

Based on the isolation of bacteria and fungi taken from soil samples from the Piyungan landfill, Bantul, Special Region of Yogyakarta, based on sampling points SP1, SP2, and SP3, 18 bacterial isolates and five fungal isolates were obtained. Isolation was carried out with an incubation time of ±14 days with a temperature of 30^{o} C for bacteria and incubation time of ±2 days with a temperature of fungal isolates that had different variations depending on the colony type and cell morphology. Microbial growth in the soil can be influenced by temperature, humidity, pH, and O_2 requirements [24]. In addition, soil texture can affect the effectiveness of bacterial and fungal growth. This study selected three bacterial isolates and three fungal isolates for plastic biodegradation

ISSN 2541-223X
Volume 4 2022

analysis. The morphology characteristics of cell bacteria are presented in **Table 2**. Furthermore, the characteristics of the cell morphology are shown in Table 3. Both bacteria and fungi colonies are shown in Figure 2.

Table 2 The characteristics of selected bacterial isolates from Piyungan Landfill

Sampling point	Isolate code	Colony morphology				Cell morphology	
		Shape	Elevation	Margin	Colour	Cell shape	Gram type
SP1	A	Circular	Flat	Entire	White	Cocci	+
SP2	F	Irregular	Convex	Undulate	White	Cocci	+
SP3	J	Circular	Flat	Entire	Yellow	Cocci	-

Table 3 The characteristics of selected fungal isolates from Piyungan Landfill

Sampling point	Isolate code	Cell morphology						
		Texture	Observe	Reverse	Exudate drops	Form	Hyphal	Conidia
SP1	PA	Velvetly	Greyish blue with white color on the edges	Yellow	Yes	Rugose	Insulated, short, branched	Rounded conidia, single phialides and grow in clusters on the metule
SP2	PK	Velvetly	Gray with white edges then turns yellow	Yellow	Yes	Rugose	Insulated, short, branched	Rounded conidia, single phialides and grow in clusters on the metule
SP3	RJ	Cottony	Slow white myceliu m growth and does not spread over the dish	Yellowish white	Yes	Verrucose	Not insulated, branched, very long stolon hyphae	Rounded sporangium at the tip of the sporangiophore

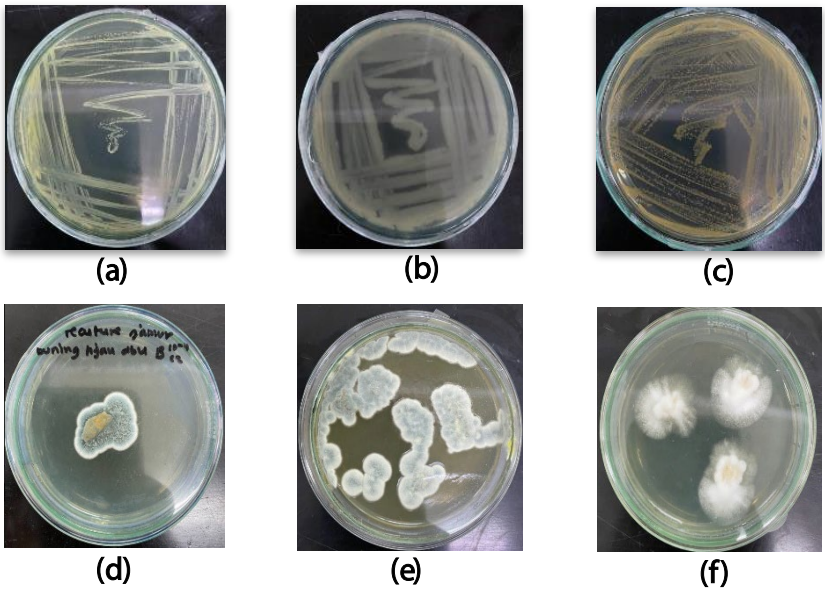
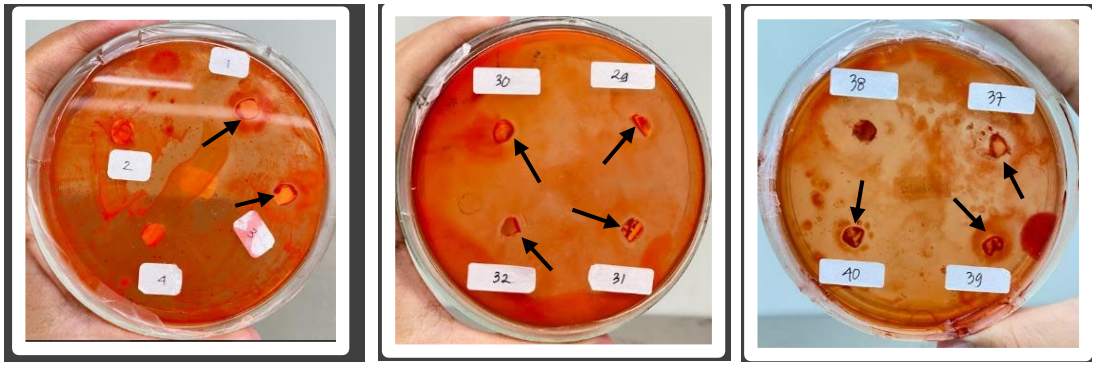


Figure 2 Morphology colony of bacterial and fungal
(a) bacterial isolate A, (b) bacterial isolate F, (c) bacterial isolate J, (d) fungal isolate PA, (e) fungal isolate PK, and (f) fungal isolate RJ

3.2. Plastic Biodegradation Ability of the Bacterial and Fungal Isolates

The potency of selected bacterial and fungal isolates in plastic degradation were observed in the mediaThe clear zone or halo zone formation around the bacterial and fungal culture indicates plastic fragments or debris. For the bacterial isolates, the analysis was carried out using Polypropylene (PP) plastic from soil samples taken from the Piyungan landfill, Bantul, Special Region of Yogyakarta. In Indonesia, Polypropylene (PP) plastic is the third biggest type of plastic produced as packaging for food and beverages [22]. Thus, we used the Polypropylene plastic-type in our study. The results showed that after 14 days of incubation, the clear-zone formation could be observed in all of the tested media containing the selected isolates (Figure 3). The clear zone is formed due to the activity of using plastic substances as a source of energy and carbon for the metabolism of the bacteria [23]. Isolate F has the greatest potential for plastic degradation because it has an average clear zone distance formed in the plastic area, which is 1.23 mm. While the bacteria with the smallest potential in degrading plastic is isolate A with an average value of the clear zone distance formed in the plastic area, which is 0.35 mm (Table 4). Based on previous research, the research was conducted using the purification method and degradation test to obtained the type of microplastic-degrading bacteria, *Micrococcus rubidaea* which had a degradation ability of 98% and *Micrococcus marcescens* had a degradation ability of 96% [7].



(a) (b) (c)

Figure 3 Clear zone formation of selected bacterial isolates around the PP plastic after 30 days incubation in NA media: clear zone of bacterial isolate A (a), clear zone of bacterial isolate F (b), and clear zone of bacterial isolate J (c).

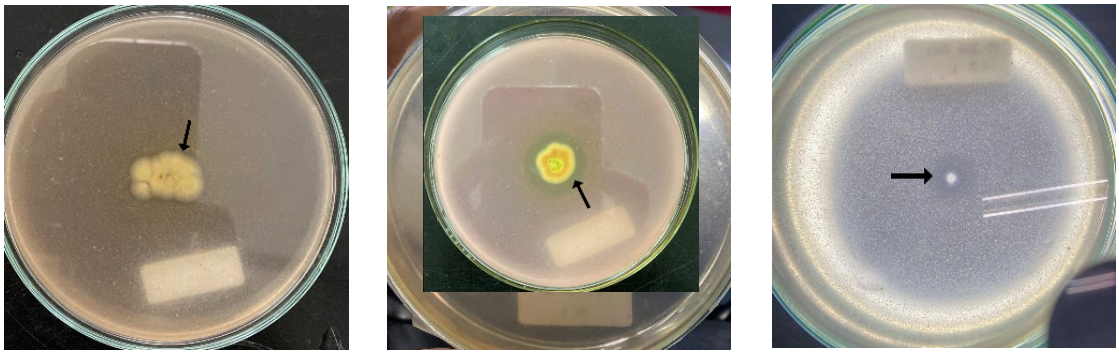
Table 4 The clear zone size of the selected bacteria isolates

Isolate code	Average size of clear zone formation (mm)
A	0.35
F	1.23
J	0.94

In this study, isolates A, F, and J, which formed a clear zone in the plastic area, had similar characteristics to the bacteria in the genus of *Staphylococcus, Micrococcus,* and *Enterococcus,* respectively [24][25][26]. The observation of clear-zone formation on plastic biodegradation is the initial step to seeing the presence of bacterial activity in the plastic area. The biodegradation ability of the bacteria as plastic degrading agents can be further observed by looking at the formation of biofilms and the percentage of plastic weight reduction. Biofilms are formed from bacterial colonies attached to plastics to break down complex polymer bonds into simpler bonds such as monomers, dimers, and oligomers using the help of extracellular enzymes. Polymers can be degraded due to enzymes attached to the substrate. Moreover, the hydrolytic cleavage occurs and undergoes mineralization to turn into CO_2 and water. Plastic can be degraded over a long period because when the nutrients or carbon contained in the composition of the NA media run out, bacteria will use plastic as a carbon source to carry out metabolic processes [27].

The fungal degradation was examined on the 10-fold diluted MEA media added with LDPE powder [29]. This kind of plastic is widely used around the world for packaging [28]. Isolates were inoculated at the centre of the media and then cultivated in an incubator for seven days at 28˚C. The results showed that the fungal isolates positively impacted both 1% and 5% LDPE concentrations based on their clear-zone formation (Figure 4). The appearance of a clear zone indicates that isolates could use LDPE powder, thereby degrading polyethylene as a carbon source [30]. A clear area around the colony might show enzyme activity [31]. Isolate RJ had the maximum clear zone in 1% LDPE, while

isolate PK obtained the ultimate clear zone in 5% LDPE (Table 5). The higher concentration, the larger the clear zone received [32].



(a) (b) (c)

Figure 4 Clear zone formation of selected fungi around LDPE plastic after ±14 days incubation in minimal MEA media: clear zone of isolate PA (a), clear zone of isolate PK (b), and clear zone of isolate RJ (c).

Table 5 The clear zone size of the selected fungi isolates

Isolate code	Clear zone in 1% LDPE (cm)	Clear zone in 5% LDPE (cm)
PA	0.1-0.3	0.1-1.55
PK	0.05-0.35	0.55-2.3
RJ	0.2-1.1	0.1-0.6

All the isolates (PA, PK, and RJ) showed positive results based on their clear-zone formation on the minimal MEA media with LDPE powder. Isolates PA and PK had similar characteristics to fungi in the genus of *Penicillium* [37]. Isolate RJ had the closest similar feature to fungi in the genus of *Rhizopus* [38]. Fungi have apical hyphae that can expand their mycelium through various materials [34]. The penetration ability of hyphae is related to the secretion of exoenzymes and their hydrophobins. Those compounds and abilities cause fungal adhesion to the plastic's hydrophobic substrate [33]. Fungal strains with the maximum size of a clear zone had a better degradation ability [35]. Thus, based on this study, isolated PK had the best ability to degrade LDPE, followed by isolated PA. At the same time, isolated RJ had the minimum capability to degrade LDPE. The fungal examination as plastic biodegradation agents can be further observed by scanning electron microscopy (SEM), pH changes, weight reduction, FTIR, and soil microbial biomass carbon test [36].

4. CONCLUSIONS

To summarise, the investigation of this study to explore the potential of microorganisms from the Piyungan landfill to degrade plastic resulted in a culture collection of three selected bacterial isolates (isolate A, F, and J) and three selected fungal isolates (isolate PA, PK, and RJ). The bacterial isolates were able to degrade polypropylene (PP), while

the fungal isolates were able to degrade low-density polyethylene (LDPE). The highest ability of a microorganism to degrade plastic is indicated by the maximum size of the clear zone. The bacterial and fungal isolates with the largest clear zone diameter were isolated in F and J, thus promising for plastic biodegradation.

5. ACKNOWLEDGMENTS

The Environmental Quality Laboratory Universitas Islam Indonesia (UII) crews are thanked for help during analysis, especially Ms Rina Isnikarita, S.Si. We also sent our honour to the Directorate of Research and Community Service (DPPM) UII, which has funded this project by a research grant for beginners numbering 001/Dir/DPPM/70/Pen.Pemula/II/2020.

6. REFERENCES

[1] Pico Y, Alfarhan A, and Barcelo D 2019 Nano- and Microplastic analysis: Focus on their Occurrence in Freshwater Ecosystems and Remediation Technologies. TrAC - Trends in Analytical Chemistry. 113: 409-425

[2] Shen M, Song B, Zeng G, Zhang Y, Huang W, Wen X, and Tang W 2020 Are Biodegradable Plastics a Promising Solution to Solve the Global Plastic Pollution. Environmental Pollution. 263: 114469

[3] Azizah P, Ridlo A, Suryono, C 2020 Mikroplastik pada Sedimen di Pantai Kartini Kabupaten Jepara, Jawa Tengah. Journal of Marine Research. 9: 326–332

[4] Sari R N 2017 Karakteristik Air Lindi (Leachate) di Tempat Pembuangan Akhir Sampah Air Dingin Kota Padang. Laboratorium Fisika Bumi. 6: 93–99

[5] Utami I and Liani M 2021 Identifikasi Mikroplastik pada Air Sumur Gali di Sekitar TPA Piyungan Yogyakarta. Jurnal Riset Daerah Badan Perencanaan Pembangunan Daerah Kabupaten Bantul, Provinsi Special Region Of Yogyakarta. 21: 4003-4014

[6] Megga, R P Festy, A R Arina F S, and Puji A N 2020 Isolasi Mikroorganisme Pendegradasi Mikroplastik dari Tempat Pembuangan Akhir (TPA) Sampah. In Angewandte Chemie International Edition. 6: 951–952

[7] Vianti, R O Melki, Rozirwan, and Purwiyanto A I S 2020 Purifikasi Dan Uji Degradasi Bakteri Mikroplastik Dari Perairan Muara Sungai Musi, Sumatera Selatan. Maspari Journal. 12: 29–36

[8] Rohmah U M, Shovitri M, and Kuswytasari K 2019 Degradasi Plastik Oleh Jamur Aspergillus Terreus (Lm 1021) Pada Ph 5 Dan Ph 6; Serta Suhu 25 Dan 35 Celcius. Jurnal Sains dan Seni ITS. 7: 5–10

[9] Wardani, D P A 2021 Isolasi dan Identifikasi Jamur Pendegradasi Polietilena Berdensitas Rendah (LDPE) dari Tempat Pemrosesan Akhir Supit Urang, Malang. Doctoral dissertation, Universitas Islam Negeri Maulana Malik Ibrahim

[10] Pramesti N K L, Berata I K, and Kendran A A S 2020 Profil Hematologi, Kadar Timbal dan Kadmium dalam Darah Sapi Bali yang Rumennya Mengandung Sampah Plastik. Indonesia Medicus Veterinus. 9: 526

[11] Lestari W, Ariyanti N, Pandu J, Saifuddin F, Utama W, Bahri A S, and Wijaya I P K 2018 Studi Kelayakan Perangkap CO2 Berdasarkan Analisa Fisik Sedimen (Studi Kasus: Formasi Kabuh, Cekungan Jawa Timur Utara . Iptek Journal Of Proceedings Series. 227-231

[12] Manasa K, Reddy R S, Triveni S, and Manasa C K 2017 Isolation and Characterisation of Pseudomonas Fluorescens Isolates from Different Rhizosphere

Soils of Telangana (Journal of Pharmacognosy and Phytochemistry JPP) 63: 224–229
[13] Friska, A 2019 Isolasi Bakteri Endofit dari Bruguira sp. Sebagai Sumber Senyawa Antibakteri (Fakultas Matematika dan Ilmu Pengetahuan Alam)
[14] Zegeye E K, Brislawn C J, Farris Y, Fansler S J, Hofmockel K S, Jansson J K, Wright A T, Graham E B, Naylor D, McClure RS and Bernstein H C 2019 Selection, Succession, and Stabilization of Soil Microbial Consortia. Msystems 4: 19
[15] Amelia N and Sulistiyaning T H 2021 Kajian Pengaruh Penggunaan Biosurfaktan Rhamnolipida dan Surfaktin pada Proses Bioremediasi Tanah Tercemar Crude Oil. Jurnal Teknik ITS. 10: 2
[16] Singh S, Dutta U, Gupta S, Jamwal S, Bhat A K, and Gupta V 2017 Morpho-cultural and Biochemical Identification of Pseudomonas sp. Isolated from the Rhizosphere of Different Vegetable Crops and Study its Efficacy on Solanum melongena (Brinjal). Journal of Pharmacognosy and Phytochemistry. 6: 22–28
[17] Barnett H L 1960 Illustrated genera of imperfect fungi. Illustrated genera of Imperfect Fungi.
[18] Hartanti A T, Susanti F, Prasasty V D and Radiastuti N 2021 Culturable Endophytic Fungal Diversity in Cassava Tubers of Indonesia. Biodiversitas Journal of Biological Diversity, 22: 3
[19] Iramayana T I and Arif A 2019 Keragaman jamur pada log dan kayu gergajian nyatoh (Palaquium sp.). Jurnal Perennial. 15: 8-15
[20] Valencia P E and Meitiniarti V I 2017 Isolasi dan Karakterisasi Jamur Ligninolitik serta Perbandingan Kemampuannya dalam Biodelignifikasi. Scripta Biologica. 4
[21] Hulse J D 2018 Review of Comprehensive Staining Techniques used to Differentiate Arbuscular Mycorrhizal Fungi from Plant Root Tissues. Acta Scientific Agriculture (ISSN: 2581-365X) 2: 7
[22] Wilkes T I, Warner D J, Edmonds-Brown V, Davies K G, and Denholm I 2020 A Comparison of Methodologies for the Staining and Quantification of Intracellular Components of Arbuscular Mycorrhizal Fungi in the Root Cortex of Two Varieties of Winter Wheat. Access Microbiology. 2: 2
[23] Alshehrei F 2017 Biodegradation of Low Density Polyethylene by Fungi Isolated from Red Sea Water. International Journal of Current Microbiology and Applied Sciences. 6(8): 1703-9.
[24] Mardyansah D and Trimulyono G 2021 Isolasi, Karakterisasi, dan Uji Potensi Bakteri Pelarut Fosfat dari Rhizosfer Tanaman Jati dan Sengon di Pegunungan Kapur, Daerah Selatan Kabupaten Tulungagung (Jurnal Lentera Bio) 10: 188–198.
[25] Reformanda S, Sipriyadi, Darwis W, Wibowo R H, Supriati R and Siboro R 2021 Isolation and Identification of Cellulase- Producing Encophytic Bacteria From Yellow Root Plants (Arvangelisia flava (L.) Merr) From Enggano Island (Proceedings of the 3rd KOBI Congress, International and National Conferences (KOBICINC 2020) 14: 523–528.
[26] Pinheiro E T, Candeiro G T, Teixeira S R, Shin R C, Prado L C, Gavini G and Mayer M P, 2015 RNA-based Assay Demonstrated Enterococcus faecalis Metabolic Activity after Chemomechanical Procedures (Journal of endodontics) 41:1441-1444.
[27] Barnett, H. L. (1960). Illustrated genera of imperfect fungi. Illustrated genera of Imperfect Fungi.

[28] Fibriarti B L, Feliatra F, Amin B, and Darwis D 2021 Biodegradation of LDPE Plastic by Local Strain of Bacillus sp. Isolated from Dump Soil of Pekanbaru, Indonesia. Biodiversitas Journal of Biological Diversity. 22: 12
[30] Verma N, and Gupta S 2019 Assessment of LDPE Degrading Potential Aspergillus Species Isolated from Municipal Landfill Sites of Agra. Applied Sciences. 1: 1-10.
[31] Rahim I, and Nasruddin A 2019 The Ability of Rot Fungi from Cocoa Plant in Producing Lignocellulosic Enzymes. IOP Conference Series: Earth and Environmental Science. 270(1): 012037
[32] Dhanam I D A G M, Fatmawati N N D, and Budayanti N N S 2021 Efek Aktivitas Antibakteri Ekstrak Etanol Daun Pepaya (Carica Papaya L.) Terhadap Pertumbuhan Bakteri Klebsiella pneumoniae ATCC 13883 dan Staphylococcus aureus ATCC 25923. E-Jurnal Medika Udayana. 10(2): 97-105
[33] Temporiti M E E, Nicola L, Nielsen E and Tosi S 2022 Fungal Enzymes Involved in Plastics Biodegradation. Microorganisms, 10: 1180
[34] Daccò C, Girometta C, Asemoloye M D, Carpani G, Picco A M, and Tosi S 2020 Key Fungal Degradation Patterns, Enzymes and Their Applications for the Removal of Aliphatic Hydrocarbons in Polluted Soils: A Review. International Biodeterioration and Biodegradation. 147: 104866
[35] Zhang J, Chen J, Jia R, Dun Z, Wang B, Hu X, and Wang Y 2018 Selection and evaluation of microorganisms for biodegradation of agricultural plastic film. 3 Biotech, 8: 1-8
[36] Ghosh D, Dey S, Chakraborty H, Mukherjee S, Halder A, Sarkar A and Sarkar J 2022 Mucormycosis: A new threat to Coronavirus disease 2019 with special emphasis on India. Clinical Epidemiology and Global Health
[37] Ristiari N P N, Julyasih, K S M and Suryanti I A P 2019 Isolasi dan Identifikasi Jamur Mikroskopis pada Rizosfer Tanaman Jeruk Siam (Citrus nobilis Lour.) di Kecamatan Kintamani, Bali. Jurnal Pendidikan Biologi Undiksha. 6(1): 10-19
[38] Yastanto AJ 2020 Karakteristik Pertumbuhan Jamur pada Media PDA dengan Metode Pour Plate. Indonesian Journal of Laboratory. 2(1): 33-39