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The Effectiveness of Plastic Degrading Bacteria in Different Types of Soils in Indonesia for Different Types of Plastic Waste.

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Introduction

Plastic waste is one of the biggest components in trash of around 10% in the world [1]. Among the many, plastic waste consists mostly of polyethylene, polyvinyl, polypropylene, and polystyrene. These materials can be harmful towards the environment when mismanaged including respiratory issues like asthma, endocrine disruption, and damage to internal organs such as the liver and kidneys [2].

Indonesia is the second largest plastic polluter next to China [16]. In 2024, Indonesia has a population of 283.5 million people, who produce between 6.8 and 7.8 million metric tons of plastic waste annually [1], [4]. In response, the Indonesian government strives to reduce plastic waste by 70% through efforts by offering tote bags or paper utensils as replacements [3]. However, plastic trash from years ago still persists across the country, making up 19.4% to 19.8% of waste in Indonesia by 2024 [5].

Due to their chemical structures, consisting of long-chain polymers of carbon-bound bonds, it's a lot more difficult for the environment to degrade them. Plastics are also mostly hydrophobic, causing enzymes and microbes to have a difficult time trying to attach to them to degrade the material [3], [7]. Furthermore, plastic degradation often leaves traces of microplastics in the environment. and living organisms [7]. When ingested, microplastics can cause gut blockage, reduce feeding, cause weight loss, and reduce fitness. It also inhibits growth on primary producers by reducing chlorophyll content, and damage photosynthetic efficiency in algae, reducing primary productivity [4], [6].

Fortunately, we have microbes that can degrade plastic waste. Through the formation of biofilms on plastics, bacteria can attach to the material and disrupt its chemical bonds. This is first done by producing oxidative enzymes (such as oxygenases, peroxidases, and laccases) that insert oxygen into polymer chains, which creates carbonyl (-C=O) or hydroxyl (-OH) groups [7] that act as structural weak points in plastics. Then, the bacteria produce enzymes to break down plastic. One example of such bacteria, *Bacillus spp.*, breaks down plastic by producing lipases, proteases, cutinases, and esterases. Meanwhile, *Pseudomonas spp.* breaks down plastic with hydrolases and oxygenase or PETase-like enzymes. The enzymes then break them down to smaller chains before being digested by the bacteria [8], [9].

Table 1. Microbial Enzymes in Plastic Degradation

Microbe	**Key Enzymes Produced**	**Plastic Types Affected**	**Mode of Action**	**References**
Bacillus spp.	Lipases, cutinases, estrases	PE, PP, PS, PVC	Oxidation, hydrolysis, biofilm attack	[5], [6], [10]
Pseudomonas spp.	PETase-like enzymes, oxygenases, hydrolases		Oxidative cleavage, surface pitting	[7], [15], [17]

Indonesia is a diverse country especially in terms of ecosystems such as paddy fields, mangroves, wetlands, tropical soils, and coastlines, hosting unique microbial communities [10], [11]. In particular, microbes *Bacillus spp.,* and *Pseudomonas spp.,* which can be easily found in Indonesian soils and landfills [7], [6]. The abundance of these microbes, in addition to previous evidence suggesting their efficacy in promoting plastic degradation [6], [7], [9], presents an opportunity to address Indonesia's plastic waste issue.

Therefore, in this research, we aim to reduce the amount of plastic pollution by testing the effectiveness of *Bacillus spp.* and *Pseudomonas spp.* in different soils with different plastics. We use these two for they are not only well studied, but quite common in not only

local soil but throughout the world. With this knowledge, we may be able to learn how to increase effectiveness of the bacteria.

Literature Review

Among diverse types of plastic, polyethylene, polyvinyl, polypropylene, and polystyrene are commonly used in the production of plastic-based items (source). These four have similar structures, with polyethylene having the simplest one, being more widely used, consisting of mainly long chains of –CH_2– units. Polyvinyl, polypropylene, and polystyrene have an addition of a chlorine atom, methyl group, or a phenyl ring respectively [12], [13]. For this reason, polyethylene is the easiest to degrade whereas polyvinyl is one of the hardest to degrade with an addition of a chlorine atom, making it a lot more stable than the other types of plastic. With an addition of side-groups, enzymes have a difficult time trying to reach the backbone of the molecules [14], [15].

Table 2. Common Plastics and Their Relative Degradability

Plastic Type	**Abbreviation**	**Structure Highlight**	**Relative Degradability**	**Reference**
Polyethylene	PE	-CH_2- backbone	Easiest to degrade	[11], [12]
Polypropylene	PP	-CH_2- backbone + CH_3	Moderate	[13]
Polystyrene	PS	-CH_2- backbone + phenyl ring	Moderate-hard	[17]
Polyvinyl chloride	PVC	CH_2 - backbone + Cl	Hardest to degrade	[14], [16]

Studies show that *Bacillus spp.* can degrade multiple types of plastic. Against PE (polythelene), strains like *B. subtilis, B. licheniformis, and B. cereus* cause surface oxidation, pitting, reduced crystallinity, and modest mass loss, especially after pretreatment or in consortia [6], [7]. Against PVC, *Bacillus spp.* tend to cause surface oxidation affecting additives [15]. With polypropylene, specific strains such as *B. cereus* colonizes PP films, chemically altering it which produces measurable yet limited changes unless under pretreated conditions or long incubations [7]. In polystyrene, *Bacillus spp.* degrades it through biofilm creation and enzymatic attacks, with SEM-visible erosion, FTIR chemical changes, and moderate weight loss, particularly in mixed cultures [11].

Pseudomonas spp. shows varying results and effectiveness, showing partial breakdown instead of a complete mineralization. Studies with PE shows that *Pseudomonas spp.* report modest weight loss, surface pitting, and oxidation detectable by FTIR, especially after UV/thermal pretreatment or when strains are enriched from polluted soils or insect guts [14], [16]. Results in PVC's degradation, due to it's strong C-Cl bonds, tend to reflect additive removal and surface oxidation rather than polymer backbone cleavage, with up to ~19% mass loss reported [15], [17]. *Pseudomonas spp.,* also colonizes PP films, oxidizing it, and under pretreatment or specialized enrichment conditions, laboratory isolates produce SEM-visible damage and FTIR shifts [8], [16]. Polystyrene shows somewhat strong evidence of an oxidative attack as *Pseudomonas* strains alter surfaces, producing fragments and demonstrating measurable chemical changes [18].

Previous Indonesian studies on plastic degradation tend to use only one or two type of plastics, often being LDPE and HDPE [14], [16]. To address this gap in research, the current research aims to investigate other types of plastic such as PVC, PS, PP. These materials are

often used in Indonesia for medical packaging, construction, food, automative parts, and etc. This is what we can figure out in this specific research as it not only uses the common types of plastic [7], [13]. Other than that, we'll also combine methods such as gravimetric loss, FTIR, SEM, ad CO_2 assays to ensure reliability since most research tend to rely on weight loss only due to the creation of biofilm, capturing a different aspect of plastic biodegradation. CO_2 assays seem to be one of the most reliable ones as a demonstration to mineralization, the ultimate goal of biodegradation. It also validates microbial activities and provides quantitative data overtime, allowing comparisons between isolates or conditions [9].

Research Question

Is there a significant difference in the plastic-degrading ability of bacterial isolates from different types of plastic and soil environments?

Proposed Methodology

In this study, we'll be using a quantitative method by purposive sampling of soil from distinct ecosystems in Indonesia. Each soil sample will be incubated along with the bacteria *Pseudomonas spp.* and *Bacillus spp.*, which are reported to be effective plastic degraders. This approach will target isolated bacteria, allowing systematic collection of bacteria most likely to degrade plastic. The assays, (gravimetric weight loss, FTIR, SEM, and CO_2 evolution assays) will provide measurable evidence of degradation efficiency across different soil environments and plastic types.

Project Practicalities: For this project to pursue, we need a microbiology lab with access to incubators, analytical balances, drying ovens, and basic microbiological tools. Consumables will include sterile plastics, media, plates, reagents for enzyme assays, and sequencing services. Proper biosafety approval, PPE, and waste disposal procedures must be in place. Since the experiments will be generating hundreds of samples, careful scheduling, labeling, and data management are critical. Consumables should be budgeted, sequencing costs and possible access fees for advanced instruments.

Soil sampling: Sample soil from landfills, roadside, riverbank. Pick atleast one site, like a landfill, that is known to have plastic contamination. At each site, collect around 50g of topsoil (0-5 cm) into sterile bags. Keep cool at 4-10 degrees Celsius and process within 48 hours. For longer storage, store below 4 degrees Celsius for less than a week.

Enrichment culture to select plastic-utilizing microbes: Cut polyethylene, PVC, polypropylene, polystyrene plastics into uniform strips (ex: 1x2 cm). Wash with detergent before rinsing it with 70% ethanol. Let it air dry. Sterilize with UV on both sides for 15-30 minutes or rinse with 70% ethanol. In a 250 mL flask, add 50 mL MSM (Methylsulfonylmethane) + 1–2 sterile plastic strips + 1 g soil. Incubate at 30°C, 150 rpm, for 7–14 days. After 7–14 days, transfer 5 mL culture into fresh MSM + fresh sterile plastic strips (1:10 dilution). Repeat 2–3 transfers (each 7–14 d). This enriches for microbes that can persist when plastic is the main carbon source. Record turbidity (OD600) and note visible biofilm on plastic.

Isolation of pure cultures: Serially dilute enriched cultures in sterile saline (10^-1 to 10^-6). Plate 100 µL aliquots on; MSM agar (no other carbon) with finely ground plastic (emulsified) or with plastic powder overlaid. LB agar for general heterotroph isolation (to catalog non-selective isolates). 25–30°C for 48–96 h. Look for colonies forming halos or colonies

attached to plastic particles. Pick distinct colonies, restreak 2–3 times on MSM agar to ensure purity. Assign isolate IDs (e.g., S1-1, S1-2 for site 1 isolates). Make glycerol stocks (15–20% glycerol) and store at −80°C.

Assays: Promising isolates then will be screened using clear-zone (halo) formation on MSM–plastic agar, esterase/lipase activity on tributyrin or Rhodamine B agar, and biofilm formation on plastic strips. The one to undergo quantitative assays are a subset of strong candidates where standardized plastic strips will be incubated with the bacterial cultures. Mass loss of plastics will be recorded after 0, 14, 28, and 42 days. The results will be calculated after drying and weighing strips, including abiotic and media controls to ensure reliability.

Complementary analyses: To provide deeper insight into degradation mechanisms, we can conduct extracellular enzyme assays (pNPA hydrolysis), biofilm quantification by crystal violet staining, and surface chemistry changes by ATR-FTIR or SEM. We can conduct molecular identification of efficient isolates by performing PCR amplification and sequencing of 16S rRNA genes before following with a BLAST analysis for taxonomic assignment. The data that will be systematically recorded will include isolate ID, plastic type, degradation percentage, enzyme activity, and sequencing results.

Statistical analyses: Using Two-way ANOVA, we can compare our isolates (isolate, plastic type). We can also use linear mixed-effects models to compare between the isolates, plastics, and timepoints with post-hoc tests. With this we can test correlations between enzyme activity, biofilm formation, and degradation efficiency of the microbes. With controls, replication, and a consistent sample, we can ensure accuracy. The expected outputs will include a list of candidate degraders, degradation kinetics, enzyme-degradation correlations, and visual/structural evidence of plastic breakdown.

Roadblocks and Potential Limitations: Plastics degrade very slowly. Therefore, measurable weight loss may be small and hard to detect in a limited time. There's also chances for contamination by non-target microbes, producing false positives since the methods might favor fast-growing bacteria rather than heavy degraders. Last but not least, the methodology is resource-heavy, requiring a lot of space and time. Without advanced tools, confirming true plastic bond breakdown may be difficult.

Post Program Plan: After the experiment, promising isolates will be preserved in glycerol stocks for future research and results will be written up in a final report and presentation, potentially submitted for publication. We can share findings with local schools, NGOs, or environmental agencies to raise awareness of plastic pollution. In the long term, the work can be extended by testing bacterial consortia, scaling up to soil or bioreactor trials, or performing genetic analysis to identify plastic-degrading genes, ensuring both scientific and community impact.

Conclusion: This research will highlight the potential of soil bacteria, especially *Bacillus* and *Pseudomonas,* in degrading common plastics such as polyethylene, polypropylene, polystyrene, and PVC under controlled conditions. Plastic's chemical stability may make it difficult for our environment to breakdown naturally, our methodology offers a structured approach to isolate, screen, and quantify microbes with promising degradation abilities. To understand their role in mitigating plastic pollution and providing possibilities for eco-friendly waste management strategies, we need to identify and characterize these bacteria. Although there are challenges present in scaling lab results to real-world conditions, this

research provides an important step towards developing sustainable microbial solutions to a big pressing environmental problem.

References

[1] World Bank, *Plastic Waste Discharges from Rivers and Coastlines in Indonesia*, 2021. [Online]. Available: https://www.worldbank.org/en/country/indonesia/publication/plastic-waste-discharges-from-rivers-and-coastlines-in-indonesia

[2] UNEP, *Single-use Plastics: A Roadmap for Sustainability*, United Nations Environment Programme, 2018. [Online]. Available: https://www.unep.org/resources/report/single-use-plastics-roadmap-sustainability

[3] UNEP and IGES, *National Plastic Waste Reduction Strategic Actions for Indonesia*, 2020. [Online]. Available: https://www.unep.org/ietc/resources/policy-and-strategy/national-plastic-waste-reduction-strategic-actions-indonesia

[4] World Economic Forum, "Indonesia unveils action plan to prevent 16 million tonnes of plastic from entering the ocean," *WEF*, Apr. 2020. [Online]. Available: https://www.weforum.org/press/2020/04/indonesia-unveils-action-plan-to-prevent-16-million-tonnes-of-plastic-from-entering-the-ocean

[5] Antara, "Indonesia races to end plastic waste by 2029 amid rising crisis," *Antara News*, Sep. 12, 2025. [Online]. Available: https://en.antaranews.com/news/358689/indonesia-races-to-end-plastic-waste-by-2029-amid-rising-crisis

[6] A. A. Shovitri, A. Yuliani, S. N. Fajri, A. R. Kusumawardhani, and T. W. Agustini, "Plastic-degrading bacteria isolated from contaminated mangrove sediment in Wonorejo, Surabaya," *AIP Conf. Proc.*, vol. 2199, no. 1, p. 030006, 2019, doi: 10.1063/1.5141290.

[7] F. Hening, R. Kamelia, and R. Kesuma, "Identification and characterization of microplastic degrading bacteria in three landfills of Lampung Province," *J. Biol. Biotechnol. Indones.*, vol. 10, no. 1, pp. 79–90, 2024. [Online]. Available: https://ejournal.brin.go.id/JBBI/article/view/11318

[8] T. Mulyaningsih, A. R. Rahardjo, and D. Ismayati, "Pemanfaatan Pseudomonas putida untuk mendegradasi plastik," *Jurnal Teknologi Kesehatan*, vol. 20, no. 1, pp. 12–19, 2024. [Online]. Available: https://e-journal.poltekkesjogja.ac.id/index.php/JTK/article/view/986

[9] P. Danso, J. Chow, and W. Streit, "Plastics: Environmental and biotechnological perspectives on microbial degradation," *Appl. Environ. Microbiol.*, vol. 85, no. 19, pp. e01095-19, Oct. 2019, doi: 10.1128/AEM.01095-19.

[10] A. Budiani, A. S. Shovitri, E. Nursanty, and A. R. Kusumawardhani, "Analysis of soil bacterial diversity from tropical rainforest and oil palm plantation in Jambi, Indonesia by 16S rRNA-DGGE profiles," *IOP Conf. Ser. Earth Environ. Sci.*, vol. 948, no. 1, p. 012099, Aug. 2021, doi: 10.1088/1755-1315/948/1/012099.

[11] H. Logan and P. Vos, "Bacillus," in *Bergey's Manual of Systematics of Archaea and Bacteria*. Hoboken, NJ: Wiley, 2015, pp. 1–163.

[12] J. R. Jambeck et al., "Plastic waste inputs from land into the ocean," *Science*, vol. 347, no. 6223, pp. 768–771, Feb. 2015, doi: 10.1126/science.1260352.

[13] M. Auta, C. Emenike, and S. Fauziah, "Distribution and importance of microplastics in the marine environment: A review of the sources, fate, effects, and potential solutions," *Environ. Int.*, vol. 102, pp. 165–176, Apr. 2017, doi: 10.1016/j.envint.2017.02.013.

[14] B. M. Kyaw, T. Champakalakshmi, J. S. Sakharkar, A. P. Lim, and K. R. Sakharkar, "Biodegradation of low density polythene (LDPE) by *Pseudomonas* species," *Indian J. Microbiol.*, vol. 52, no. 3, pp. 411–419, Sep. 2012, doi: 10.1007/s12088-012-0250-6.

[15] L. Giacomucci, S. Raddadi, F. Soccio, F. Lotti, M. Sannino, and F. Fava, "Polyvinyl chloride biodegradation by *Pseudomonas citronellolis* and *Bacillus flexus*," *New Biotechnol.*, vol. 52, pp. 35–41, Jan. 2019, doi: 10.1016/j.nbt.2019.04.005.

[16] H. M. Lee et al., "Evaluation of the biodegradation efficiency of four various types of plastics by *Pseudomonas aeruginosa* isolated from the gut extract of superworms," *Microorganisms*, vol. 8, no. 9, p. 1341, Sep. 2020, doi: 10.3390/microorganisms8091341.

[17] Y. Zhang et al., "Multi-omic analyses provide insights into microbial PVC biodegradation," *mSystems*, vol. 7, no. 4, pp. e00491-22, Jul. 2022, doi: 10.1128/msystems.00491-22.

[18] H. R. Kim, J. M. Yu, H. C. Lee, S. Jeon, and D. H. Kim, "Biodegradation of polystyrene by *Pseudomonas* sp. DSM 50071: Surface modification and molecular changes," *Environ. Sci. Technol.*, vol. 54, no. 20, pp. 12662–12673, Oct. 2020, doi: 10.1021/acs.est.0c03397.

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