Eco. Env. & Cons. 30 (February Suppl. Issue) : 2024; pp. (S369-S372)

ISSN 0971–765X

DOI No.: http://doi.org/10.53550/EEC.2024.v30i02s.073

Isolation of High Density Polyethelene Degradating Bacteria from Soil

Sonika Sandip Patil*[1], Monika Sandip Patil[1] and G.R. Pathade[2]

[1,2]Krishna Institute of Allied Sciences (KVVDU) Karad, M.S., India

(Received 24 July, 2023; Accepted 17 September, 2023)

ABSTRACT

Plastics are the most commonly used polymers for routine applications. These plastics are less biodegradable in the environment. So the plastics accumulated drastically in the environment and had bad effects on it. High-density polyethylene (HDPE) is a major cause of persistent and long-term environmental pollution. The main objective of the present study is the isolation and screening of bacterial species with the capability to degrade high-density polyethylene (HDPE). The two bacterial isolates were identified from soil collected from the dumping yard of village Atake (Tal-Karad, Dist-Satara). These two bacterial isolates were obtained on minimal agar medium incorporated with HDPE. These isolates are further tested for degradation of the plastic. The isolate 1 (*Bacillus cereus*) could degrade the plastic to the extent of 16% in terms of % weight loss, while the isolate 2 (*Pseudomonas halobacterium*) could degrade the plastic to the extent of 10% in terms of % weight loss, thus the isolate 1, which was identified as *Bacillus cereus* was found to be the best among the two isolates.

Key words *: Polyethylene, HDPE, Soil.*

Introduction

Plastics are organic polymers of high molecular mass. The name plastic is derived from its property of plasticity. Plasticity is the property of any material by which the material is able to irreversibly deform without breaking. The term plastic is derived from the Greek word „plastikos which means "capable of being shaped or molded (Divyalakshmi *et al.*, 2016). Plastic products have brought tremendous convenience to our lives. Polyethylene (PE) has been utilized in making grocery bags, food packaging films, and toys; polypropylene (PP) for the creation of straws, car seats and container caps; and lastly, polystyrene (PS) for making disposable food trays, and laboratory wares. The exceptional mechanical properties of PE and its relatively low cost led to the abundance and popularity of the mentioned polymer to the general public. It is also known that plastics, specifically PE, are inert materials (Bolo *et al.*, 2015)

The most common types of polyethylene are HDPE (High-density polyethylene), LLDPE (Linear low-density polyethylene) and LDPE (Low-density polyethylene). Polyethylene waste accumulation in the environment causes blockages in fishes, mammals, and birds, endangering a large number of species (Alamer *et al*., 2023). Polyethylene (PE) is manmade polymers used in many aspects of human life. It is widely used due to its light weight, inexpensive, strong and durable nature. The worldwide production rate of synthetic polymers is about 140 million

[1]Research Scholar, [2] Dean

tons and its utility is increasing at the rate of 12% per annum (Maroof *et al.*, 2021).

The by products of plastic degradation by physical and chemical methods are toxic irritants and harmful gaseous foams that not only damage the ozone layer in the upper atmosphere but also destroy the underground water resources, cause infertility of soil, prevent degradation of normal substances and are proved dangerous to human, animals and the ecosystem (Nadeem *et al*., 2021). Microbes are naturally performing degradation on complex organic matters, converting them into simpler organic substances and gaining nutrients during this process. Plastic waste has energy and carbon sources that help microbes in their survival. Plastics can degrade in aerobic or anaerobic conditions, and they can be used as a major natural clean up at locations where toxic waste is disposed. Additionally, biodegradation of polymers is defined as any change in polymer characteristics, such as molecular weight, mechanical strength, and surface features, or, in other words, the breakdown of material into pieces through microbial digestion into a simpler form via biochemical transformation (Alamer *et al.*, 2023).

Research on microbial degradation of plastic wastes, especially polyethylene which is highly resistant to biodegradation has been steadily growing. Some microorganisms have been shown to produce enzymes capable of polyethylene plastic degradation, including moulds such as *Aspergillus* and *Fusarium*, yeast such as *Cryptococcus* and bacteria in the genera *Pseudomonas and Bacillus* (Montazer *et al*. 2018).

Plastic wastes accumulating in the environment pose an ever-increasing ecological threat. Plastics that are biodegradable can be considered environmentally friendly. So the present project work was undertaken with the following principal objectives as Isolation of bacteria from plastics dumped in soiltheir plastic biodegradative ability.

Material and Methods

Collection of soil sample from dumping yard

The Soil samples were collected from the dumping yard of village Atake (Tal-karad, Dist-Satara) in a clean polythene bag and brought to the laboratory, where they were kept at room temperature for further processing.

Enrichment and isolation of plastic degrading bacteria

The plastic bottles were cut into pieces using scissors. The pieces and soil samples were added to minimal broth for enrichment at room temperature. After the 10 days of incubation, the 0.1 mL enrichment culture was spread and inoculated on the minimal agar plate and incubated at room temperature for 6-7 days for the isolation of plastic-degrading bacteria. Well-isolated representative colonies from minimal agar were picked up, studied for morphological and cultural characteristics, and aseptically transferred to sterile minimal agar slants.

Characterization and identification of plastic degrading bacteria

Bacteria were chosen for characterization and identification by microscopic and morphological observation and biochemical tests with respect to Bargey's Manual of determination bacteriology.

Studies on plastic degradation capacity of bacterial isolates

The pre-weighted high density polythene strips of 1.00 g were aseptically transferred to the conical flask containing 100 ml of minimal broth medium and separately inoculated with the selected bacterial strains. Control was maintained with high density polythene strips in the microbe free medium. Four different flasks were maintained for each treatment.

Dry weight determination of plastic (Nadeem *et al.*, 2021)

After 3 weeks of incubation, the polythene strips were collected, washed thoroughly using distilled water, and shade dried till a constant weight was obtained. From the collected data, the weight loss of the polythene strip was calculated as follows:

$$\frac{\text{Initial weight} - \text{Final weight}}{\text{Intial weight}} \times 100$$

Determination of percentage weight of degraded (HDPE)

From the calculated value of polyethylene degradation the percentage of degradation was determined.

Results and Discussion

As per the cultural, morphological, and biochemical characteristics, (Table 1) the bacterial isolates were

designated as isolate 1, isolate 2, and tentatively identified as shown in Table 1. The selected isolate was characterised according to Bergey's Manual of Systematic Bacteriology, Volume II, Section 12.

Bacterial isolate 1 was found to be *Bacillus cereus*.

Bacterial isolate 2 was found to be *Pseudomonas halobacterium*.

It can be seen from Table 1 that all two bacterial isolates associated with plastic that was dumped in the dumping yard were isolated and were designated as Isolate 1 and Isolate 2. It is further seen from Table 1 that the isolates were identified as *Bacillus cercus* (isolate 1) and *Pseudomonas halobacterium* (isolate 2). It becomes clear from Figures 1 and 2 that

Table 1. Identification of isolates

Sr.No	Isolate	Tentatively identification
1	Isolate 1	Bacillus cereus
2	Isolate 2	Pseudomonas halobacterium

all two isolates were capable of degrading the plastic (HDPE). It can be also observed from Fig. 1 and 2 that the isolate 1 (*Bacillus cereus*) could degrade the plastic to the extent of 16% in terms of % weight loss, while the isolate 2 (*Pseudomonas halobacterium*) could degrade the plastic to the extent of 10% in terms of % weight loss, thus the isolate 1, which was identified as *Bacillus cereus*. *Bacillus cereus* was found to be the best among the two isolates.

Conclusion

The present study deals with the isolation, identification, and degradative ability of plastic-degrading bacteria from plastic dumped in soil. The organisms identified were further inoculated into different culture media, and their biodegradative ability was determined by weight loss after a period of 21 days. The plastic strip was degraded by using isolate 1 (*Bacillus cereus*) at approximately 16% andisolate 2

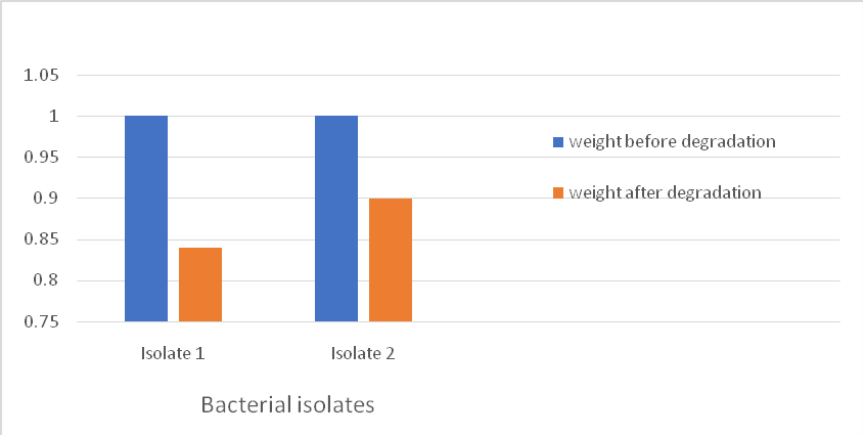

Fig. 1. Weight of HDPE before and after degradation

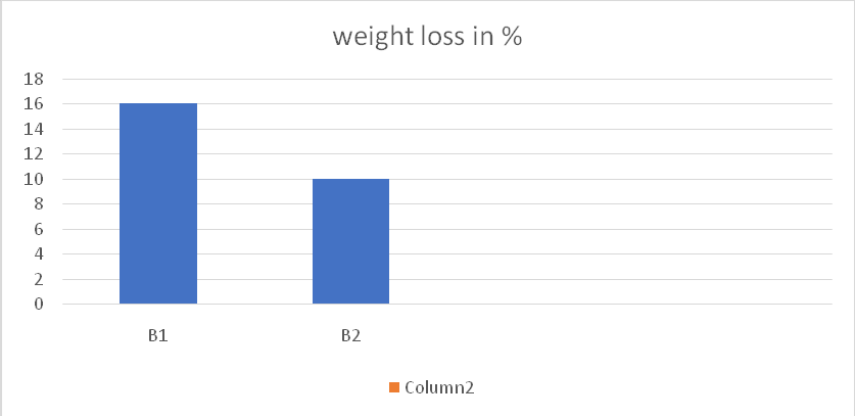

Fig. 2. Percentage of weight loss HDPE strips after treatment

(*Pseudomonas halobacterium*) at 10%. From this project, we conclude that the bacterial strains isolated from different soil samples can have the ability to degrade the plastic.

Acknowledgements

I take this opportunity to express my profound gratitude and indebtedness to Dr.S. C. kale, Prof., Head and Dean, Krishna institute of Allied sciences, (KVVDU) Karad for availing the laboratory facilities in the institute to carry out the experimental work and his guidance. I wish to express my sincere thanks to teacher in charge of my project work Mrs. Jayashree P. Nanaware for the help and valuable suggestion in preparation of this project.

I wish to express my thanks to Dr. G. R. Pathade who moderated this paper and, in that line, improved the manuscript significantly. We are also grateful to teaching, non-teaching staff of the institute for their co-operation during this course of my work.

Conflicts of interest

Authors hereby declare that there is no any conflict of interest among authors.

References

Alamer, N. J., Aldayel, M. F. and Khalifa, A. 2023. Isolation and Characterization of Brucella spp., Low-Density Polyethylene (LDPE) Plastic Degrading Bacteria in Al-Ahsa Region, Saudi Arabia. *Applied Sciences*. 13(7): 4629.

Arias-Villamizar, C. A. and Vázquez-Morillas, A. 2018. Degradation of conventional and oxodegradable high density polyethylene in tropical aqueous and outdoor environments. *Revistainternacional de contaminaciónambiental*. 34(1): 137-147.

Balasubramanian, V., Natarajan, K., Rajeshkannan, V. and Perumal, P. 2014. Enhancement of in vitro high-density polyethylene (HDPE) degradation by physical, chemical, and biological treatments. *Environmental Science and Pollution Research*. 21: 12549-12562.

Balasubramanian, V., Natarajan, K., Rajeshkannan, V. and Perumal, P. 2014. Enhancement of in vitro high-density polyethylene (HDPE) degradation by physical, chemical, and biological treatments. *Environmental Science and Pollution Research*. 21: 12549-12562.

Bolo, N. R., Diamos, M. A. J. C., SIA, S., Ocampo, M. A. B., and Suyom, L.M. 2015. Isolation, identification, and evaluation of polyethylene glycol and low density polyethylene-degrading bacteria from Payatas Dumpsite, Quezon City, Philippines. *Phil J Health Res Dev*. 19(1): 50-59.

Divyalakshmi, S. and Subhashini, A. 2016. Screening and isolation of polyethylene degrading bacteria from various soil environments. *IOSR J Environ Sci Toxicol Food Technol*. 10(12): 01-7.

Gajendiran, A., Krishnamoorthy, S. and Abraham, J. 2016. Microbial degradation of low-density polyethylene (LDPE) by Aspergillus clavatus strain JASK1 isolated from landfill soil. *3 Biotech. 6*: 1-6.

Gupta, K. K., Devi, D. and Rana, D. 2016. Isolation and screening of low density polyethylene (Ldpe) degrading bacterial strains from waste disposal sites. *World Journal of Pharmaceutical Research*. 5(11): 1633-1643.

Hussein, A. A., Al-Mayaly, I. K. and Khudeir, S. H. 2015. Isolation, Screening and Identification of Low Density Polyethylene (LDPE) degrading bacteria from contaminated soil with plastic wastes. *Mesopotamia Environmental Journal*. 1(4): 1-14.

Maroof, L., Khan, I., Yoo, H. S., Kim, S., Park, H. T., Ahmad, B. and Azam, S. 2021. Identification and characterization of low density polyethylene-degrading bacteria isolated from soils of waste disposal sites. *Environmental Engineering Research*. 26(3).

Montazer, Z., Habibi-Najafi, M. B., Mohebbi, M. and Oromiehei, A. 2018. Microbial degradation of UV-pretreated low-density polyethylene films by novel polyethylene-degrading bacteria isolated from plastic-dump soil. *Journal of Polymers and the Environment*. 26: 3613-3625.

Nadeem, H., Alia, K. B., Muneer, F., Rasul, I., Siddique, M. H., Azeem, F. and Zubair, M. 2021. Isolation and identification of low-density polyethylene degrading novel bacterial strains. *Archives of Microbiology*. 203(9): 5417-5423.

Shimao, M. 2001. Biodegradation of plastic. *Curr. Opin. Biotechnol*. 12: 242-277.

Skariyachan, S., Manjunatha, V., Sultana, S., Jois, C., Bai, V. and Vasist, K. S. 2016. Novel bacterial consortia isolated from plastic garbage processing areas demonstrated enhanced degradation for low density polyethylene. *Environmental Science and Pollution Research*. 23: 18307-18319.