ELSEVIER

Scopus

doi

Journal of Vibration Engineering

ISSN:1004-4523

Registered

- SCOPUS
- GOOGLE SCHOLAR
- DIGITAL OBJECT IDENTIFIER (DOI)
- IMPACT FACTOR 6.1

Our Website
www.jove.science

Pretreatment-Enhanced Biodegradation of Polypropylene (PP) by *Bacillus cereus*

Sridevi Veluru[a*], Geethika Gudapati[b], Shaik Khasim Beebi[c], Nomi Yuktha Yedla[d], Tanmayi Bora[e], Tukaram Bai M[f] Praneetha Yelamanchili[g], Venkata rao Poiba[h]

[a*bdefg]Department of Chemical Engineering, AU College of Engineering (A), Andhra University, Visakhapatnam-530003, India.

[c] Department of Biotechnology, GITAM School of Core Engineering, Visakhapatnam

Abstract

Biodegradation was carried out in this work using *Bacillus cereus* (H23B00108), which was isolated from marine soil that was polluted with plastic and obtained from Tenneti Park in Visakhapatnam, India.To make commercial polypropylene (PP) beads more vulnerable to microbiological breakdown, they were pre-treated with chemicals and radiation.Weight loss and physical structural characterization using X-ray diffraction (XRD), scanning electron microscopy (SEM), and Fourier Transform Infrared spectroscopy (FTIR) were used to track degradation.Over the course of six weeks, the *Bacillus cereus* isolate (H23B00108) degraded untreated PP by 3%, chemically pretreated PP by 17%,combined UV and chemical pretreatment by 8.25%.Taken together, these results demonstrate that *Bacillus cereus* (H23B00108) has great promise as a PP degrader, lending credence to the idea that microbes can biodegrade plastic waste in a sustainable way, which is great news for PP waste management.

Keywords: Biodegradation; SEM; *Bacillus cereus ;*XRD; FTIR.

1.Introduction

The persistent nature of plastic waste has intensified its impact on environmental quality and aquatic ecological integrity. Production of plastic was estimated to reach 390.7 million metric tons in 2021 (1) and is projected to virtually triple, reaching around 1124 million metric tons by 2050.At least 8 million metric tons of plastics end up in the ocean every year; that's the same as emptying a garbage truck's load into the water every minute.This is projected to reach two per minute by 2030 and four per minute by 2050 (2) if no measures are implemented.The ubiquitous plastics and their variants are now an integral part of man-made demands across all industries because to their benign impact on humans (3).With its exceptional packaging properties, poly propylene is quickly becoming one of the seven most used forms of plastic.The worldwide plastic pollution situation has been exacerbated by the increasing use of single-use plastics, such as polypropylene.The non biodegradability of PP materials and the 6.3% annual growth in PP production sales since 2013 (4) raise serious environmental issues.

Particularly during the COVID-19 epidemic, these PP polymers were widely employed to make disposable things including packaging, rope, bottles, caps, fishing gear, carpet, strapping, straws for drinking, and personal hygiene items like masks, gloves, and hairnets.Regrettably, the presence of polypropylene microplastics (MPs), which are shards of plastic smaller than 5 mm in diameter, can be caused by the incorrect disposal and increased production of these PP plastics, which are worsened by natural processes. Microplastics can alter the distribution of energy and food, which in turn can reduce the generative productivity and physical fitness of marine organisms (Sussarellu et al., 2016), and the widespread use of plastics in everyday life releases microscopic particles into the environment, which pose a greater threat to human health than environmental contaminants owing to direct contact. Microplastics have been found in a wide variety of environmental matrixes, such as soils, freshwater bodies, deep sea sediments, soil, and the air we breathe.

New ways of dealing with plastic pollution are required because of its far-reaching effects.Not all solutions are without their flaws. Some plastics can't be recycled, burning plastics emits a cocktail of dangerous chemicals into the air, landfills take up valuable real estate that could be better used, and chemical treatments end up polluting our water supply (7).On the other hand, additional contamination issues may arise due to these technologies' inefficiency and limited treatment capacity.Because they are not biodegradable, PP materials in particular pose serious environmental risks.

Research into novel approaches to plastic waste management is, hence, essential.A sustainable, eco-friendly, and long-term solution that doesn't hurt the environment is being considered thanks to new research that suggests microbes or enzymes may be able to biodegrade plastics (8).Bacteria and fungus are just two examples of the microbes that engage in plastic biodegradation (9).

Developing effective and sustainable solutions for plastic waste management may depend on our ability to understand and use microbial pathways for PP decomposition.Colonization of the plastic surface by microbes initiates biodegradation of PP by secreting enzymes that hydrolyze the ester linkages in the polymer (10).The enzymatic mechanisms employed by these microbes are essential for overcoming the recalcitrant nature of polypropylene, characterized by its linear chain structure of propylene monomers that enhances its resistance to environmental degradation. There are several steps to the degradation process, including colonization by microbes, breakdown by enzymes, fragmentation of polymers, and mineralization (11).

This study set out to biodegrade PP beads that had been purchased from a commercial source by treating them with a new strain of *Bacillus cereus* (H23B00108), which had been found in Tenneti Park along the Visakhapatnam shoreline in India.Furthermore, the possibility of the strain forming a biofilm on a polypropylene surface was examined.Incorporating technical research like as FTIR, SEM, and XRD further confirmed that our indigenous strain could use PP as the only carbon

source.In addition, the use of pretreatment procedures such as UV irradiation, chemical oxidation, or a mix of the two to improve polypropylene degradation was also evaluated.

2. Materials and Methods

2.1 Collection of samples and Isolation of microorganisms:

The coastal soil at Tenneti Park along the Visakhapatnam shoreline was sampled at a depth of 15 cm, collected in glass vials, and kept in the laboratory for further examination. The coordinates of the park are 17.7486° N, 83.3424° E.

2.2 Characterisation of the isolated organism capable of degrading PP:

Serial dilution and plating on Luria-Bertani agar were subsequent steps in isolating the enhanced cultures.The plates were kept at 30°C for 24-48 hours, and then colonies with different morphologies were selected and isolated by streaking them many times.The morphology of the bacteria was identified as gram positive using a staining process and a medical binocular microscope.The genotypic features of the potential bacteria were then identified.The most similar isolate was located using the EzBioCloud 16S database, which compares partial 16S rRNA gene sequences to species that have been described before.

2.3 Pre-treatment of PP:

The plastics supplier supplied the commercial grade polypropylene (PP). Round beads with a particle size of 1 mm make up the PP.Prior to drying and weighing, the samples were disinfected with 70% ethanol.The PP, after being weighed, was immersed in a 69% nitric acid solution for a period of six days.To remove any possible contaminants, the beads were washed with distilled water before being immersed in a 99.9% ethanol solution. They were then oven-dried at 70 °C for 30 minutes.Shorter durations of 5, 15, and 30 minutes of exposure to 256 nm UV light were applied to a small number of PP samples that had been chemically pre-treated in a UV chamber.Degradation is enhanced over the 30 minutes because functional groups are formed to their fullest (12).

2.4 Biodegradation of PP by isolated bacterium :

Based on KS M3100-1:2002; (13), the biodegradability of PP was investigated.A rotary shaker incubator was used to cultivate pure bacterial strains in LB broth that contained tryptone, yeast extract, and sodium chloride at 30 °C for 24-48 hours until they reached the mid-log phase.The control group continued to use a flask that contained LB broth that had been inoculated with bacterial culture but did not contain PP beads.Afterwards, in conical flasks, 1% (v/v) of the culture was added to 500 mL of LB broth that already included 0.4 g of polypropylene (PP) beads. The beads were colored as follows: untreated, chemical pre-treated, and UV + chemical pre-treated. The incubation time was set at 30 minutes. A spectrometer (Genesys 50, Thermo Scientific, USA) was utilized to assess the development of bacterial strains using PP beads as a carbon source at an optical density of 540 nm (OD_{540}).The biodegradation experiments lasted for forty days at a temperature of 37 °C.

2.5 Weight loss determination:

After removing the residual PP microplastics from the medium using filtration, they were washed in a specific order with 70% ethanol to ensure that all cells and debris were removed. The samples were then oven-dried at 60 °C overnight, or until they were completely dry.Using an analytical balance with a readability of 0.0001 g (Sartorius ENTRIS 224-1S), the weight of the leftover polymer was calculated to quantify the level of degradation (14).Using Equation (1), we were able to calculate the percentage of weight loss for PP microplastics.

$$\% \text{ Weight loss} = \frac{W_o - W}{W_o} \quad (1)$$

where W_0 is the initial weight of the polymer (g) and W is the residual weight of the polymer (g).

2.6 Characterization studies of PP

2.6.1Spectroscopic evaluation of PP beads using FTIR:

Before and after a 40-day biodegradation period, FTIR spectra of PP films were collected (untreated, chemically pretreated, and UV+chemicallypretreated). The adherent bacterial biomass was then removed by washing the films sequentially three times with sterile distilled water and three times with 70% ethanol. The films were then oven dried at 60°C until they reached a constant weight according to the established procedure.The functional group analysis was carried out with the use of a Vertex 70v FT-IR spectrometer and a Hyperion 2000 microscope from Bruker in attenuated total reflectance (ATR) mode. The microscope had a field of view of 400 × 400 μm.FT-IR spectra were recorded by signal-averaging 32 scans across 4000–500 cm^{-1} at 4 cm^{-1} resolutionquantified carbonyl index: CI = A_{1710} / (A_{2918} + A_{2850}).

2.6.2 Scanning electron microscopy (SEM) of PP beads:

After the experimental time (40 days), the morphology of the degraded PP microplastics was examined using SEM.Their preparation for CO2 drying in critical-point ovens included soaking in 5% glutaraldehyde for an hour, washing in 100 mM phosphate buffered saline (pH 7.0), and dehydrating with 30-100% graded ethanol for 15 minutes.With the help of a sputter coater (MSP-1S, SHINKKU VD, Japan), a thin layer of gold was sputtered onto dehydrated PLA films.Following this, SEM EDS (SEM MODEL) pictures were produced (15).

2.6.3 X ray diffraction pattern of PP:

The PP crystallinity was examined using the PANalytical—X 'Pert' Pro X-ray diffraction technique.Cu Kα radiation (1.54060 A) was applied while the procedure was conducted at 45 kV and 40 mA with a θ/θ geometry.The size of thedivergence slit was set to 0.4785°. A scan rate of 10 m^{-1} and a step size of 0.0170 were used to capture the XRD patterns, which ranged from 10 to 60°, at 25 °C.By dividing the integrated area of the diffraction peaks (the crystalline component) by the integrated area of the full diffractogram, the total crystallinity was calculated (16).

3 Results and discussion:

3.1 Molecular identification and phylogenetic characterization

By utilizing UV-Visible spectroscopy at 540 nm, the bacterial growth curve was quantified.When exposed to PP carbon, the strain grew swiftly, as seen by the growth curve. Noticeable progress was made in just two days.After reaching its peak within five days, the bacterial population began a slow but steady drop that persisted for the following week.The growth of bacterial degrading strain with respective time. It was determined that this particular strain belonged to the genus *Bacillus cereus* (H23B00108) based on its morphological traits. Biotechnology relies heavily on *Bacillus cereus* (H23B00108), a rod-shaped, aerobic, gram-positive bacteria with a unipolar flagellum.*Bacillus cereus* (H23B00108) was shown to have the highest potential to biodegrade PP films, according to this investigation.Primers BF27 (AGA GTT TGA TCC TGG CTC AG) and B765R (CTG TTT GCT CCC CAC GCT TTC) were used to amplify and sequence the 16S rRNA genes. By comparing the incomplete 16S rRNA gene sequences to the EzBioCloud 16S database, which includes all validly described species, the most similar isolate could be found.Through the use of the neighbor-joining method, the evolutionary history was inferred (17).The phylogenetic tree is displayed in Figure 1.The evolutionary distances (18) were calculated using the greatest composite likelihood technique, with each site representing the number of base substitutions. This analysisinvolved 11 nucleotide sequences. First, second, third, and noncoding codon locations were all covered.The partial deletion option was considered to be present in any region where the site coverage was less than 95%.Thus, no location could contain missing data, unresolved bases, or alignment gaps smaller than 5%.In the end, 711 locations made it into the dataset.The evolutionary analysis was performed using the MEGA11 software (19).

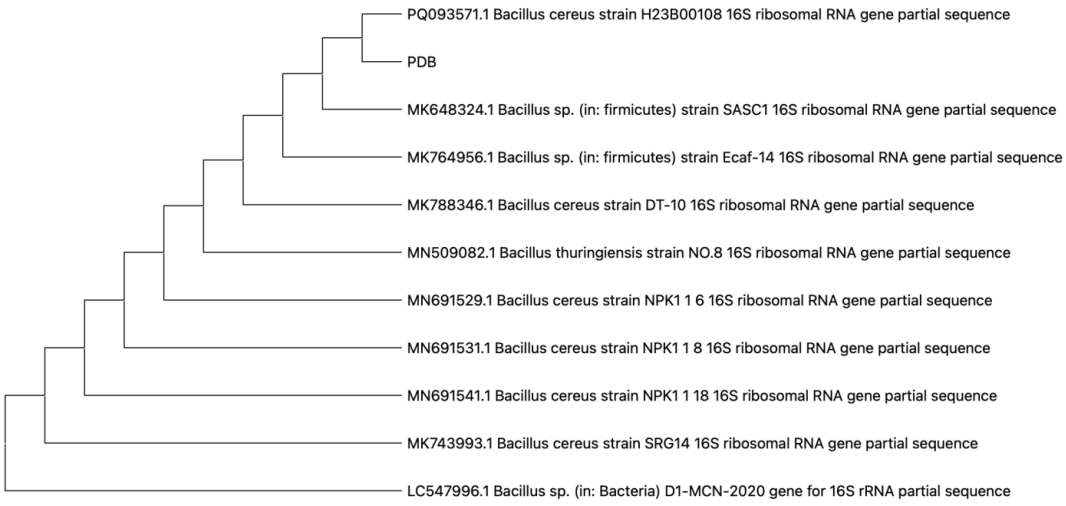


Fig 1 Identification of PP degrading bacteria through Phylogenetic tree

3.2 Weight loss determination:

Bacillus cereus (H23B00108) was likely involved in the biodegradation process that sped up the breakdown of PP.Throughout the 10-to 40-day duration, the PP film's weight dropped dramatically.One possible way to determine the biodegradation of PP is to measure their weight loss.*Bacillus cereus* (H23B00108) inoculation resulted in weight loss after 32 days (Figure 2).Consumption of PP reduces carbon content, which leads to weight loss in pre-treated PP infected with *Bacillus cereus* (H23B00108).After 30 days of incubation, untreated polypropylene (PP) lost about 3% of its weight, demonstrating relatively little degradation.However, after being chemically pretreated, the breakdown of PP increased dramatically, reaching around 17% at 30-32 days. This suggests that the degradation susceptibility of PP was greatly boosted by the pretreatment. A maximum weight loss of around 8.25% was noted around 30 days of incubation after combining chemical pretreatment with combined UV (30 min) exposure, which demonstrated significant deterioration.Particularly in the samples that had been chemically treated, the *Bacillus cereus* (H23B00108) was able to persist on the surfaces of the PP beads and aid in their breakdown.

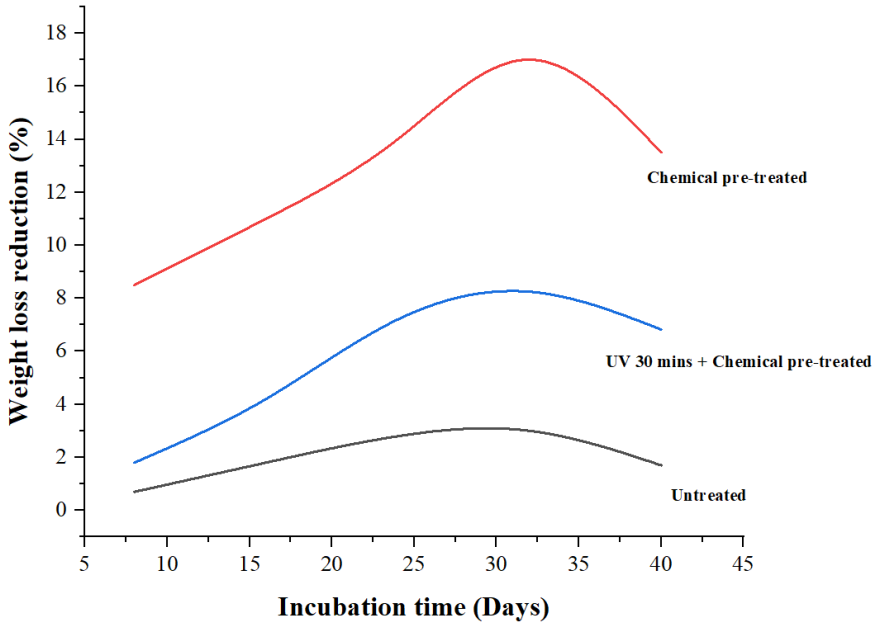


Figure 2.Weight loss reduction % of PP that has been incubated with *Bacillus cereus* (H23B00108)

3.3. FTIR :

The FTIR study presented strong molecular evidence that *Bacillus cereus* (H23B00108) microbes are causing chemical changes on polypropylene (PP) films.In order to analyze the chemical and structural changes in polypropylene (PP) films subjected to various treatments—including untreated, chemically pretreated, and chemically pretreated combined with UV irradiation—a series of important spectral shifts and intensity changes were observed in the films before and after biodegradation. These changes reflect the underlying biochemical alterations that occur during

degradation.Several changes in peak intensity and band locations were noticed in the spectra, which were in the range of 4000-500 cm^{-1} (Figure 3).The presence of prominent peaks in the 2950-2850 cm^{-1} region of the FTIR spectrum of untreated polypropylene prior to biodegradation was ascribed to the symmetric and asymmetric C-H stretching vibrations of the methyl ($-CH_3$) and methylene ($-CH_2-$) groups, respectively.Polypropylene is characterized by CH_3 bending and deformation vibrations, which were detected at peaks at 1450-1375 cm^{-1}. These vibrations show that a stable hydrocarbon backbone is present (20).

Observations of changes in the fingerprint region (1500-500 cm^{-1}) following biodegradation suggest structural modifications and the potential formation of oxygen-containing functional groups, like carbonyl and hydroxyl groups, due to oxidative degradation processes (21).When comparing spectral changes in untreated PP to chemically pretreatment samples prior to biodegradation, very small alterations were noted Nevertheless, upon biodegradation, chemically prepared PP samples exhibit additional spectrum alterations, such as a notable diminution in the strength of the C-H stretching bands (2950-2850 cm^{-1}) and methyl deformation bands (1450-1375 cm^{-1}).In addition, after biodegradation, the most noticeable changes in spectrum were observed in polypropylene samples that had been pretreated with both ultraviolet light and chemicals.The presence of additional bands (about 1700 cm^{-1} and 3200-3500 cm^{-1}) implies the establishment of carbonyl and hydroxyl groups.Accelerated oxidation and significant breakdown of long polymer chains are indicated by the FTIR spectra, which display substantial distortion and reduction in peak intensities (22,23).There were minimal spectral changes in untreated PP, moderate structural modification in chemically pretreated PP, and the highest degree of polymer degradation and alteration in characteristic absorption bands in UV combined with chemical pretreatment. Microplastics made of polypropylene can be more easily broken down by microbes when subjected to certain pretreatment techniques, such as a mix of chemical treatment and ultraviolet light (24).Russell et al. (2011) found similar findings, such as the absence or loss of peaks linked to ester bonds in polymer degradation (25).

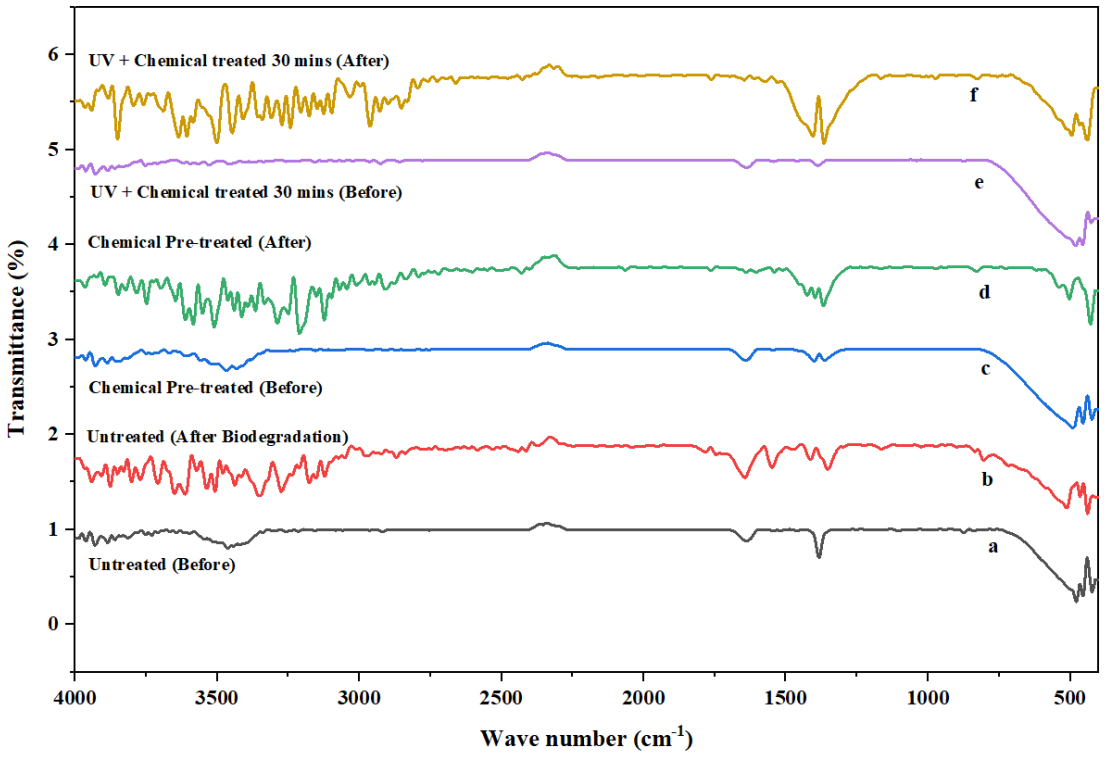
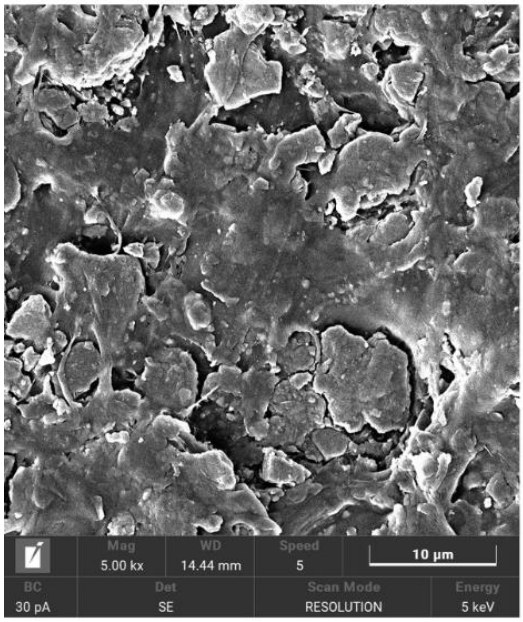


Figure 3.FTIR spectrum of PP beads a) Untreated PP beads before biodegradation b) Untreated PP after biodegradation c) Chemical pre-treated PP before biodegradation d) Chemical pre-treated PP after biodegradation e) UV 30 mins and chemical pre-treated PP before biodegradation f) UV 30 mins and chemical pre-treated PP before biodegradation

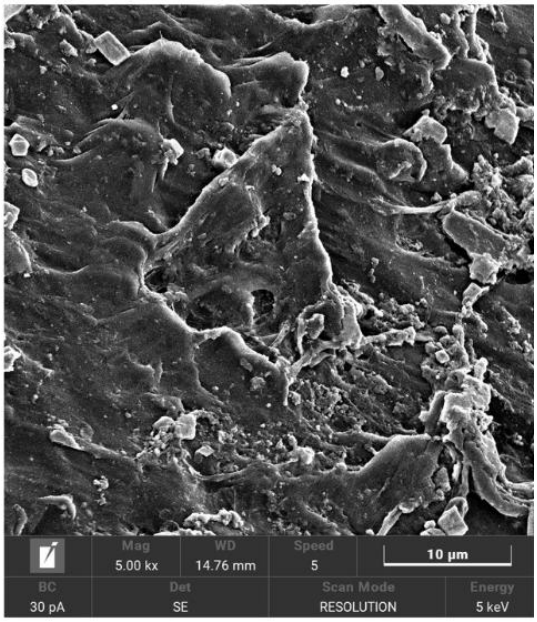
3.4 SEM:

By employing scanning electron microscopy, we were able to see the bacterial colonization and the subsequent changes in the surface micromorphology of PP films.A ZEISS scanning electron microscope was used to analyze all of the materials.Polypropylene (PP) surface morphological changes were examined using Scanning Electron Microscopy (SEM) before and after deterioration in untreated, chemically pretreated (HNO_3), and UV-chemically treated samples.Prior to degradation, the SEM micrograph of untreated polypropylene (Figure 4a) showed a consistent, smooth, and compact surface morphology with few surface flaws.The structural deformations that occur during degradation are usually caused by microorganisms that inhabit the surface of the polymer. These deformations include surface modifications such small fractures and shallow pits (Figure 4b).Figure 4c shows that chemically prepared polypropylene had microcracks and increased surface roughness after being treated with HNO_3 before deterioration.More morphological abnormalities, including deep fissures, holes, pores, and broken areas, were seen in the chemically treated PP during degradation (Figure 4d).The combination of ultraviolet light and chemical pretreatment of PP samples resulted in more noticeable morphological alterations (Figure 4e).Surface imperfections and grooves were clearly visible in the scanning electron micrograph of the pre-degradation sample. Figure 4f shows that after biodegradation, the surface micrographs revealed significant erosion, big holes, deep fissures, and

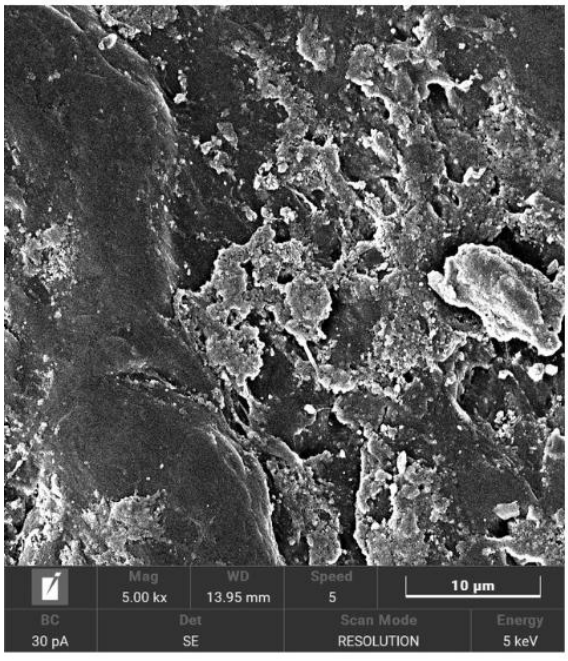
cavities, all of which pointed to advanced polymer degradation and strong microbial activity.Previous reports have shown that surface erosion, oxidation, and molecular fragmentation of polymers occur during microbial colonization, which is consistent with the modifications that have been observed (26,27,28).



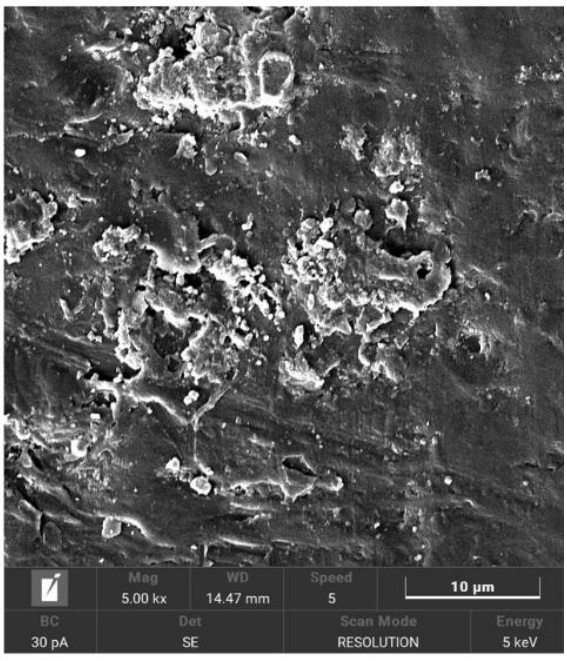
a



b



c



d

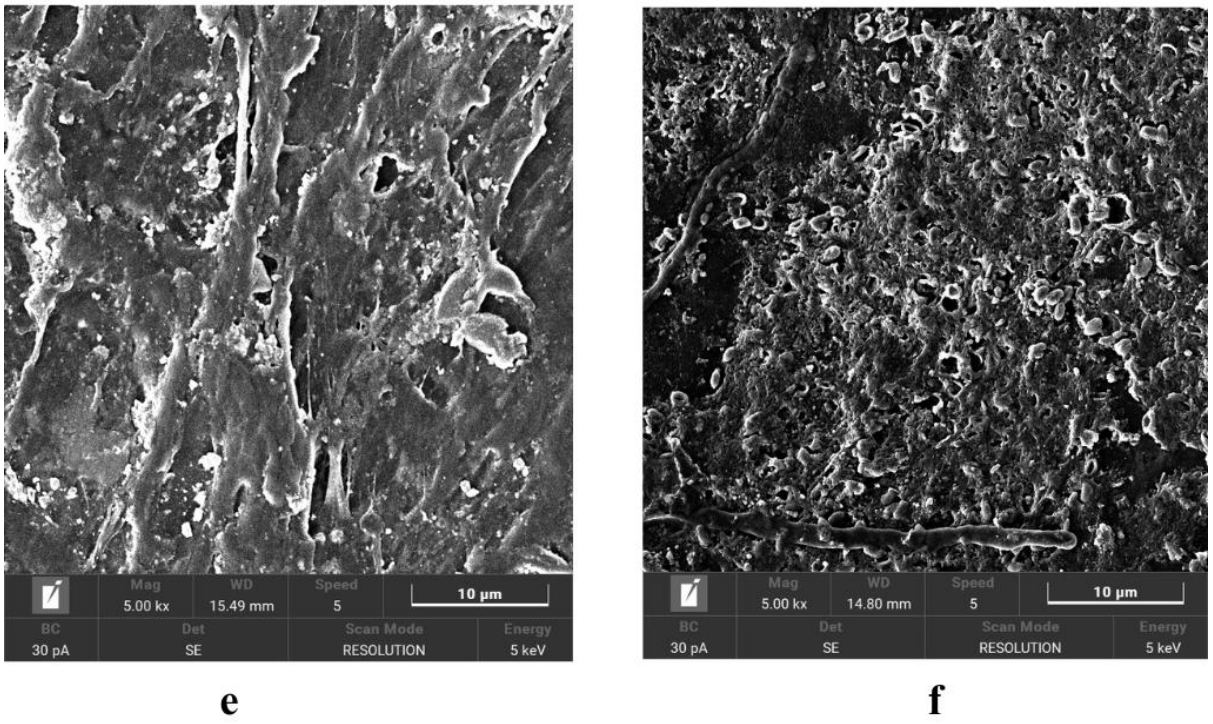


Figure 4: SEM images depicting the colonization, biofilm formation, and biodegradation of polypropylene by bacterial strain *Bacillus cereus* (H23B00108) a-b) Untreated PP before degradation and after 40 days of degradation c-d) chemical pre-treated PP before degradation and after ofdegradation e-f) UV 30 mins and chemical pre-treated PP before degradation and after 40 days of degradation

3.5 XRD:

Figure 5 shows the X-ray diffraction (XRD) patterns of three different types of polypropylene (PP): untreated, chemically pre-treated, and UV + chemical pre-treated PP, following incubation with the *Bacillus cereus* (H23B00108) bacterial strain.Figure 3a shows that the untreated polypropylene had diffraction peaks at 2θ = 9.58°, 14.23°, 17.09°, 18.77°, 21.79°, 25.60°, and 29.70°, which are indicative of the polypropylene's semi crystalline structure. The diffraction peaks of the untreated sample are at 9.51°, 11.80°, 14.21°, 17.05°, 18.65°, 21.79°, 24.09°, 25.55°, and 29.63°, indicating that the crystalline areas have been somewhat disrupted by the chemical treatment of the polymer surface.The PP that was pre-treated with chemicals and exposed to UV light for 30 minutes also showed peaks at 9.60°, 14.18°, 17.04°, 18.66°, 21.71°, 28.75°, and 29.64°.When contrasted with the control sample, the treated one showed a small change in the location of the diffraction peaks and a weakening of their strength.This decrease in crystallinity demonstrated that the polymer chains' orderly crystalline arrangement was disturbed by the combined actions of chemical preparation, UV irradiation, and bacterial activity.The space between the reflection sites (d) decreases as the sinθ value

increases, according to Bragg's law (29,30).Additionally, there was a little change in the interplanar spacing (d-spacing) determined from the XRD peaks following treatment.While the d-spacing values of the untreated PP ranged from 9.22 Å to 1.60 Å, the samples that were chemically treated and those that were UV + chemically treated revealed values ranging from 9.29-1.66 Å and 9.20-2.08 Å, respectively.Disruption of the crystalline lattice occurs during biodegradation, as indicated by the modest increase in d-spacing.

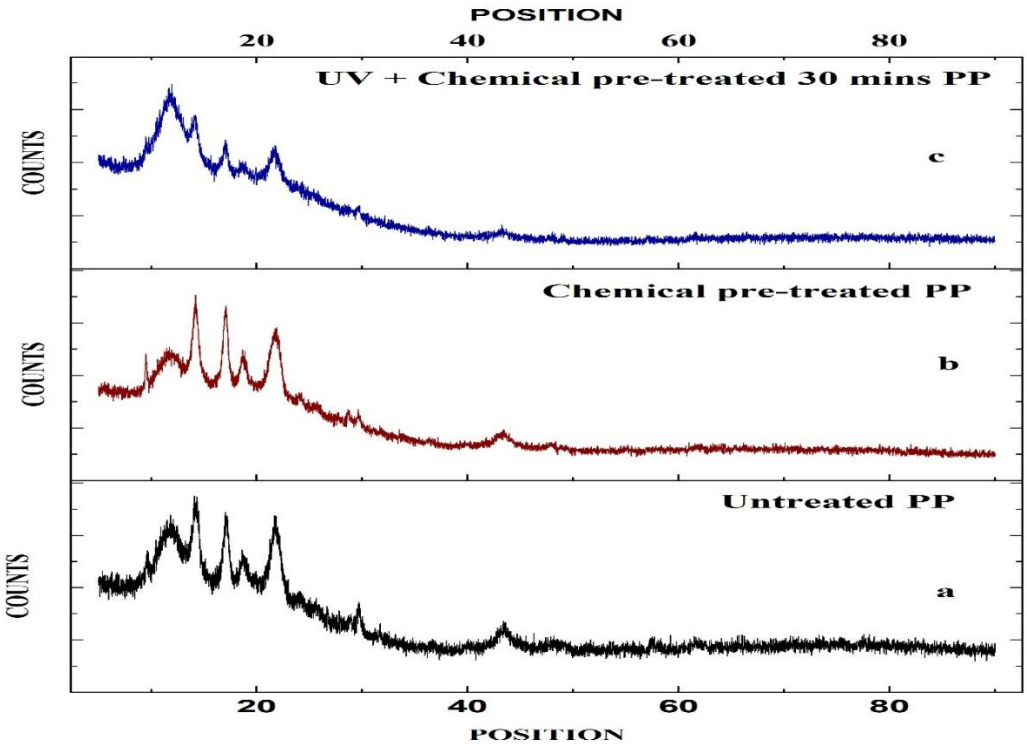


Figure 5 XRD patterns of a) Untreated PP b) Chemical Pre-treated PP
c) UV 30 mins + Chemical pre-treated PP (after biodegradation)

Conclusion:

Bacillus cereus (H23B00108) proved to be highly effective in breaking down chemically processed polypropylene (PP) after 40 days of incubation under ideal growth circumstances.Weight loss of 17% was found to be the most efficient degradation rate. Subsequently, an 8.25% reduction in weight was achieved by a mix of ultraviolet light and chemical treatments. The untreated PP, on the other hand, exhibited minimal deterioration, losing a mere 3% of its weight.Pure *Bacillus cereus* (H23B00108) used PP beads as a carbon source, according to characterization tests using FT-IR, SEM, and XRD. Chemical and photo

oxidative pretreatments enhance microbial colonization and enzymatic destruction of the polymer structure, according to these investigations' findings.When taken as a whole, this research shows that increasing the biodegradability of polymers like polypropylene (PP) by a combination of pretreatment methods and microbial breakdown is a significant step toward more sustainable plastic waste management.

Acknowledgments

The authors sincerely acknowledge the Department of Chemical Engineering, College of Engineering, Andhra University, Visakhapatnam, India, for providing the basic infrastructure and lab facilities for carrying out this research study.

References:

1. Statista Research Department. (2023). *Plastic production worldwide 2021*. Retrieved from https://www.statista.com/statistics/282732/global-production-of-plastics-since-1950/
2. Ellen MacArthur Foundation, & World Economic Forum. (2016). *The new plastics economy: Rethinking the future of plastics* (pp. 1–120). Ellen MacArthur Foundation.
3. Horton, A. A. (2022). Plastic pollution: When do we know enough? *Journal of Hazardous Materials, 422*, 126885.
4. Sutkar, P. R., &Dhulap, V. P. (2025). Advancements in polypropylene biodegradation: A comprehensive microbial and analytical review. *Sustainable Chemistry for the Environment*, 100213.
5. Jeyavani, J., Al-Ghanim, K. A., Govindarajan, M., Nicoletti, M., Malafaia, G., & Vaseeharan, B. (2024). Bacterial screening for efficient polypropylene microplastics biodegradation. *Science of the Total Environment, 918*, 170499.
6. Sussarellu, R., et al. (2016). Environmental impact and sustainable development of polypropylene-based materials. *Resources, Conservation and Recycling*. https://doi.org/10.1016/j.resconrec.2025.108142
7. Ru, J., Huo, Y., & Yang, Y. (2020). Microbial degradation and valorization of plastic wastes. *Frontiers in Microbiology, 11*, 442. https://doi.org/10.3389/fmicb.2020.00442
8. Miloloža, M., Cvetnić, M., KučićGrgić, D., OcelićBulatović, V., Ukić, S., Rogošić, M., &Bolanča, T. (2022). Biotreatment strategies for removal of microplastics. *Environmental Chemistry Letters, 20*, 1377–1402. https://doi.org/10.1007/s10311-021-01370-0

9. Mohanan, N., Montazer, Z., Sharma, P. K., & Levin, D. B. (2020). Microbial and enzymatic degradation of synthetic plastics. *Frontiers in Microbiology, 11*. https://doi.org/10.3389/fmicb.2020.580709
10. Umamaheswari, M. M. (2014). Growth of biofilms on abiotically pretreated polypropylene surfaces. *European Journal of Zoological Research*.
11. Sutkar, P. R., &Dhulap, V. P. (2025). Stages of plastic degradation: Colonization, fragmentation, and mineralization. *Sustainable Chemistry for the Environment*.
12. Kong, L., & Wang, Y. (2015). Enhancing hydrophilicity of polypropylene membranes by nitric acid activation. *Journal of Membrane Science, 487*. https://doi.org/10.1016/j.memsci.2015.03.044
13. Jeon, H. J., & Kim, M. N. (2016). Isolation of mesophilic bacterium for biodegradation of polypropylene. *International Biodeterioration & Biodegradation, 115*, 244–249.
14. Mohan, A. J., Sekhar, V. C., Bhaskar, T., &Nampoothiri, K. M. (2016). Microbial assisted degradation of plastics. *Bioresource Technology, 213*, 204–207.
15. Sekhar, V. C., Nampoothiri, K. M., Mohan, A. J., Nair, N. R., Bhaskar, T., & Pandey, A. (2016). Microbial degradation of high-impact polystyrene. *Journal of Hazardous Materials, 318*, 347–354.
16. Sudhakar, M., Doble, M., Murthy, P. S., & Venkatesan, R. (2008). Marine microbial degradation of polyethylene. *International Biodeterioration & Biodegradation, 61*, 203–213.
17. Saito, N. (1987). The neighbor-joining method: A new method for reconstructing phylogenetic trees.
18. Tamura, K., Nei, M., & Kumar, S. (2004). Prospects for inferring large phylogenies. *Proceedings of the National Academy of Sciences, 101*(30), 11030–11035. https://doi.org/10.1073/pnas.0404206101
19. Tamura, K., Stecher, G., & Kumar, S. (2021). MEGA11: Molecular evolutionary genetics analysis version 11. *Molecular Biology and Evolution, 38*, 3022–3027.
20. Abbas-Abadi, M. S., Haghighi, M. N., & Yeganeh, H. (2014). Structural characterization of polypropylene using FTIR spectroscopy. *Polymer Testing, 34*, 1–7. https://doi.org/10.1016/j.polymertesting.2014.01.003
21. Sivan, A. (2011). New perspectives in plastic biodegradation. *Current Opinion in Biotechnology, 22*(3), 422–426.
22. Andrady, A. L. (2017). The plastic in microplastics: A review. *Marine Pollution Bulletin, 119*(1), 12–22.

23. Gewert, B., Plassmann, M., & MacLeod, M. (2015). Pathways for plastic degradation in marine systems. *Environmental Science: Processes & Impacts, 17*, 1513–1521.
24. Shah, A. A., Hasan, F., Hameed, A., & Ahmed, S. (2008). Biological degradation of plastics: A comprehensive review. *Biotechnology Advances, 26*(3), 246–265.
25. Russell, J. R., et al. (2011). Biodegradation of polyurethane by fungi. *Applied and Environmental Microbiology, 77*(17), 6076–6084.
26. Wróbel, M., Szymańska, S., Kowalkowski, T., &Hrynkiewicz, K. (2023). Microorganisms for polyethylene and polypropylene degradation. *Microbiological Research, 267*, 127251.
27. Jeon, J. M., Park, S. J., Choi, T. R., Park, J. H., Yang, Y. H., & Yoon, J. J. (2021). Biodegradation of polyethylene and polypropylene. *Polymer Degradation and Stability, 191*, 109662.
28. Basak, N., Meena, S. S., & Shilpa. (2024). Biodegradation of LDPE by *Pseudomonas aeruginosa*. *Biodegradation, 35*(5), 641–655.
29. Esmaeili, A., Pourbabaee, A. A., Alikhani, H. A., Shabani, F., & Esmaeili, E. (2013). Biodegradation of LDPE by mixed cultures. *PLoS ONE*. https://doi.org/10.1371/journal.pone.0071720
30. Musuc, A. M., Badea-Doni, M., Jecu, L., Rusu, A., & Popa, V. T. (2013). FTIR, XRD, and DSC analysis of polyethylene biodegradability. *Journal of Thermal Analysis and Calorimetry, 114*, 169–177.