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Biodegradation of Polyethylene using *Bacillus subtilis*

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Abstract

Polyethylene is among the major plastics being dumped in the environment. The study explores methods to enhance the rate of biodegradation of polyethylene using physical and biological means. Bacterial species –*Bacillus subtilis* – was tested for its potential in utilizing polyethylene as their sole carbon source. The microbial species produced surface active compounds (Biosurfactants) that enhance the degradation process. Pretreatment of polymer films with Ultraviolet radiation aids its accessibility as food for the microorganisms thus enabling a much faster rate of biodegradation. Inoculation of pretreated polyethylene films of thickness 18μ with *Bacillus subtilis* with the addition of its biosurfactant (surfactin) proved to be most efficient with a weight loss percentage of 9.26% in 30 days

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Keywords: Polyethylene;Biodegradation;Biosurfactants

1. Introduction

Biosurfactants are surface-active compounds synthesized by a wide variety of microorganisms. They are molecules that have both hydrophobic and hydrophilic domains, comprising an acid, peptide cations, or anions, mono-, di- or polysaccharides and a hydrophobic moiety of unsaturated or saturated hydrocarbon chains or fatty acids Due to their amphiphilic structure, biosurfactants increase the surface area of hydrophobic water-insoluble substances, increase the water bioavailability of such substances and change the properties of the bacterial cell

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surface. Because of their potential advantages, biosurfactants are widely used in many industries such as agriculture, food production, chemistry, cosmetics and pharmaceutics.

There have been focuses on the recent hypotheses and experimental findings regarding the biodegradation of polyethylene. Ambika *et al*. (2009), made a review on different approaches to enhance the biodegradation of polyolefins. It discusses various physical, chemical and biochemical approaches that can be adopted to enhance their biodegradation. From this review it was inferred that biosurfactants can be used as an enhancing agent of biodegradation process [1]. Pretreatment of the polymer using physical means prior to biodegradation have been found to enhance the process considerably. UV radiation was used as a pretreatment by Mahalakshmi *et al.* (2012) and Sowmya *et al.* (2014) [2,3].

Research works on biosurfactants, its production, analysis and applications were also measured. There is ample research literature in the fields of polymer degradation and on the various aspects of biosurfactants such as its production, extraction from different microbes, its application in heavy metal removal and biodegradation of hydrocarbons. However, the use of biosurfactants in polymer degradation is an inadequate area of study.

The explicit objectives of this study are: To determine and compare the rate of biodegradation of Polyethylene (PE) films of two thicknesses

- Using mono culture of *Bacillus subtilis*
- With and without pretreatment of UV-rays on Polyethylene
- With and without addition of Biosurfactant

2. Materials and Methods

2.1. Collection of Polymer Sample

Polyethylene (PE) films of two thicknesses – 18μ LDPE (Low density Polyethylene) and 41μ HDPE (High density Polyethylene)-were purchased. PE films were cut in required size of approx. 2 cm x 2 cm and they were subjected to UV treatment for 72 hours.

2.2. Microorganisms

Bacterial species *Bacillus subtilis* was selected for the study based on their ability to produce biosurfactants. Agar slants of *Bacillus subtilis* MCC No. 2183 was obtained from Microbial Culture Collection, Pune. *Bacillus subtilis* produces biosurfactant known as surfactin. Bacterial species was cultured in nutrient broth (Nutrient Medium: Beef extract (10g), Peptone (10g), NaCl (5g), Distilled water 1L) and incubated for 24 hours at 32^{o}C [4, 5, 6].

2.3. Production of Biosurfactants

For the production of biosurfactants from each of the B.subtilis, freshly prepared nutrient medium was inoculated with cultural broth and was incubated at 32^{o}C for 24 hrs. On reaching the endogenous phase of bacterial growth, olive oil was added (30 ml/L). Conical flasks were kept in a shaking incubator for 3 days and 7 days at 32^{o}C, 180rpm [7].

2.4. Estimation of Biosurfactants

Screening test: oil spreading technique: Oil spreading assay, 10 μL of crude oil was added to the surface of 40 mL of distilled water in a petri dish to form a thin oil layer. Then, 10 μL of culture or culture supernatant were gently placed on the centre of the oil layer. The presence of biosurfactant would displace the oil and a clear zone would form [8].