



Biodegradation Of Plastics By Microbes: Nature's Defence

Dhakshayani G^{#1}, Arun Kumar R^{#2}, Vasanth Kumar E^{#3} Sebastian J^{#4}

#1,2,3,4 Department of Pharmaceutical Technology,

Jaya Sakthi Engineering College, Chennai, 602024, Tamil Nadu, India.

Abstract

Plastics are widely used in consumer goods, building and construction, agriculture, and health. They are significant in every economic area worldwide. The biosphere and human existence on earth are seriously threatened by the excessive usage of plastics. The buildup of plastics on land and in the ocean has sparked interest in breaking down these polymers. To lessen the environmental load of plastics, sufficient biodegradable techniques must be used. Understanding the relationship between bacteria and polymers is crucial to resolving environmental issues related to plastics. Numerous biological things, primarily microbes, have developed survival and degradation techniques for plastics. The varieties of plastics based on their thermal and biodegradable properties, types of deterioration and biodegradation, types of degradable plastics, biodegradation characterisation, and variables influencing biodegradation are the main topics of this review. These microbes are ideal for green chemistry to remove toxic plastics from the environment because of their capacity for plastic degradation and bioremediation.

Introduction

The word plastic had originated from “pliable” that can be defined as “easily shaped”. Plastics can be conveniently modified from one shape to another based on their desired functionality. Plastics are also known as polymers or “long chains of monomers,” which are bonded to other identical subunits to form a polymer (Evode et al., 2021). Plastics are inexpensive, lightweight, strong, durable, corrosion-resistant materials, with high thermal and electrical insulation properties. The diversity of polymers and the versatility of their properties are used to make a vast array of products that bring medical and technological advances, energy savings and numerous other societal benefits (Andrady and Neal, 2009). Plastics consist of synthetic and semisynthetic polymers produced from coal, natural gas, crude oil, and organic products including cellulose, salt, and can also come from renewable components such as grain, corn, potato, palm, sugar beet and cane, starch, seaweed, and vegetable oil.

Plastic can be categorized as mega plastic (>1 m), macro plastic (<1 m), mesoplastic (<2.5 cm), and micro plastic (<5 mm) (defined size varies according to different literature records) according to size variations (Ivar do Sul *et al.*, 2013; Wang *et al.*, 2018). Another scientific literature categorizes plastic litter according to the different length ranges, as mega plastics (>100 mm), macro plastics (20–100 mm),

mesoplastics (5–20 mm), and micro plastics (<5 mm) (Barnes *et al.*, 2009). Meso plastic is an intermediate size range between visible macro plastic and minute microscopic plastics. Larger size plastics visualized by the naked eye are called macro plastics or mega plastics.

Plastics can be divided into three groups based on the physical properties, which includes thermoplastics, thermosets, and elastomers (Lusher *et al.*, 2015). Thermoplastics are plastics that can be re-melted while thermosets are plastics that cannot be remelted. Thermoplastics are including Polyethylene Terephthalate (PET), Polyethylene (PE), Low-Density Polyethylene (LDPE), High-Density Polyethylene (HDPE), Polystyrene (PS), Expanded Polyethylene (EPE), Expanded Polystyrene (EPS), Polyvinylchloride (PVC), Polycarbonate (PC), Polypropylene (PP), Polylactic acid (PLA) and Polyhydroxyalkanoates (PHA). Thermosets are including Polyurethane (PUR), Phenolic resins (PR), Epoxy resins (ER), Silicone, Vinyl esters, Acrylic resins (AR), Urea formaldehyde (UF) resins (Andrady and Neal, 2009). Meanwhile, elastomers are elastic polymers like rubber and neoprene (Lusher *et al.*, 2015).

PE has high molecular weight polymers, complex three-dimensional structures, and hydrophobic components (Aamer *et al.*, 2009). PET is a synthetic plastic that was first developed in 1941 which is commonly used as food containers, bottles, and others. HDPE is a synthetic plastic that has been produced since 1939. LDPE which has been developed and produced since 1939 is commonly used as drink containers. PS has been produced since 1930, widely used as disposable cups and wrapping tools. PP is used as bottle caps, medicine bottles, car benches, batteries, disposable syringes, and carpets. Plastics contain many additives used to reinforce those polymers (Lusher *et al.*, 2017). Those components used to increase the resistance of plastics toward biotic and abiotic factors (Campanale *et al.*, 2020). Additives that commonly used in plastic production are phthalates, bisphenol A (BPA), flame retardants (FRs), polybrominated diphenyl ethers (PBDEs), and nonylphenols (Lusher *et al.*, 2017). Commonly used Phthalates are Di-2- (ethylexyl) phthalate (DEHP), Dibutyl phthalate (DBP), and Diethyl phthalate (DEP) (Lusher *et al.*, 2017). Flame Retardants (FRs) are additional components used to reduce the flammability of plastic polymers. The widely used FRs include polybrominated diphenyl ethers (PBDEs) and hexabromocyclododecane (HBCD). Nonylphenol ethoxylate (NPE) and Nonylphenols (NP) are used as stabilizers in food packaging and as antioxidants in several polymers including rubber, vinyl, polyolefins, polystyrenes, and PV (Lusher *et al.*, 2017).

Besides synthetic plastic, there are also bioplastics. Bioplastics are divided into biodegradable plastics, oxo-biodegradable, and bio-based plastics based on biodegradability. Biodegradable plastics are plastics that can be degraded by microorganisms into water and carbon dioxide or methane, such as poly (butylene succinate-co-butylene adipate) with various degree of crystallinity (Baidurah *et al.*, 2012) and poly (3-hydroxybutyrate-co-3-hydroxyhexanoate) (Baidurah *et al.*, 2019). Oxo-biodegradable plastics are plastics that belong to the polyolefin group which contain a metal salt and will be fragmented into smaller fragments. Also, bio-based plastics are plastics that are easily degraded by microorganisms because they are made of easily degraded materials (Lusher *et al.*, 2017).

Biodegradation of plastics

Biodegradation is a new strategy for plastic waste remediation that has been increasingly discussed as an eco-friendly technology. Microbial potential (mainly bacteria and fungi) for plastic degradation through enzymatic hydrolysis has been intensively studied in recent years (Buchholz *et al.*, 2022; K. Zhu *et al.*, 2020; L. Zhu *et al.*, 2020). The exploration of new microbial enzymes and further mechanistic elucidations are crucial for enhanced MNP remediation by biodegradation (Buchholz *et al.*, 2022). Thus, identifying plastic-active enzymes for further application in biotechnological processes and elucidating their actual action in nature is an emerging research field, which is still in its infancy (Chow *et al.*, 2023). The process of MNP biodegradation is divided into four main steps (Rai *et al.*, 2021). Biofilm development

on plastispheres that decreases the hydrophobicity and buoyancy of plastic particles, followed by biodeterioration through exopolysaccharides and enzymatic action (endo-/exoenzymes). The third step is the destabilization of the carbon skeleton in MNP particles through enzymatic depolymerization, using oxidases, amidases, peroxidases, and laccases, then assimilation of monomers by microbial biomass (Gómez-Méndez et al., 2018). Hydrolases were reported to play a vital role in plastic polymer hydrolysis and therefore in determining MNP biodegradation rates (Ho et al., 2018). Certain microbes, such as the alkane-degrading marine bacterium *Alcanivorax borkumensis*, were reported to have a key role in LDPE degradation (Delacuvellerie et al., 2019), resulting in significant physicochemical alterations. In addition, other bacterial strains, such as *Bacillus gottheilii* and *Bacillus cereus*, showed high potential to effectively remove a wide range of MNPs (PP, PET, PS, and PE) from mangrove sediments (Auta et al., 2017). Fungi, such as *Zalerion maritimum*, also showed high efficiency in plastic degradation (Paço et al., 2017) using different mechanisms, through the release of sticky natural biosurfactants, such as hydrophobins (Sánchez, 2020). In some cases, pretreatment is required to enhance the degradation process. In this context, PP biodegradation by two different fungal strains (*Phanerochaete chrysosporium* and *Engyodontium album*) was enhanced after starch/pro-oxidant pretreatment (Jeyakumar et al., 2013). Despite their promising roles, the enzymes available at present act mainly on high-molecular-weight polymers of PET and ester-based PUR, with moderate turnover rates, and no enzymes acting on other high-molecular-weight polymers, such as PS, PP, ether-based PUR, and polyethylene are known (Danso et al., 2019). Bioengineering of bacterial strains for enhanced protein production could further enhance MNP biodegradation by increasing enzyme activities (Wei et al., 2016). In addition, the application of targeted microbial strain engineering can accelerate cellular enzymatic activities towards enhanced plastic degradation. In this respect, an engineered strain of *Bacillus subtilis* showed enhanced PETase activity (ca. four-fold) by inactivating the twin arginine translocation complexes, which further enhanced MNP degradation (Huang et al., 2018). An integrated microalgae–bacteria system also showed potential to enhance the degradation of MNPs through enzymatic action. In this context, the photosynthetic diatom *Phaeodactylum tricornutum* was used as a cell factory for engineered PETase isolated from *Ideonella sakaiensis*, a known bacterium with a high capability for plastic degradation and for consuming it as both a carbon and energy source (Moog et al., 2019). Although microbial biodegradation of MNPs is a promising approach, few studies have been conducted on microalgae, which require further investigations.

Mechanism of biodegradation

The adherence of the microorganism to the surface of the polymer, preceded by the colonization of the external surface, growth of the microbial, use of the polymer as a source of carbon and energy, and final degradation of the polymer is the primary mechanism for microbial degradation (Oliveira et al., 2020; Restrepo-Flórez et al., 2014). Microorganisms can stick to the surface of a polymer if it is hydrophilic. Once anchored to the surface, the organism can grow by utilizing the polymer as a source of carbon and energy. Polymer biodegradation happens by hydrolysis after colonization; first, the enzyme catalyzes the substrate material and then facilitates the hydrolysis reaction. Polymers degrade into small molecular weight oligomers, dimers, and monomers before finally mineralization to CO₂ and H₂O (Tokiwa et al., 2009). The surface composition can quantify the scope of colonization on the polymer, as hydrophilic areas are much more conveniently colonized by microbes. This is a restriction since the polymer's water-repellent surface contradicts the porous structure of the microorganisms (Fig. 1) (Restrepo-Flórez et al., 2014).

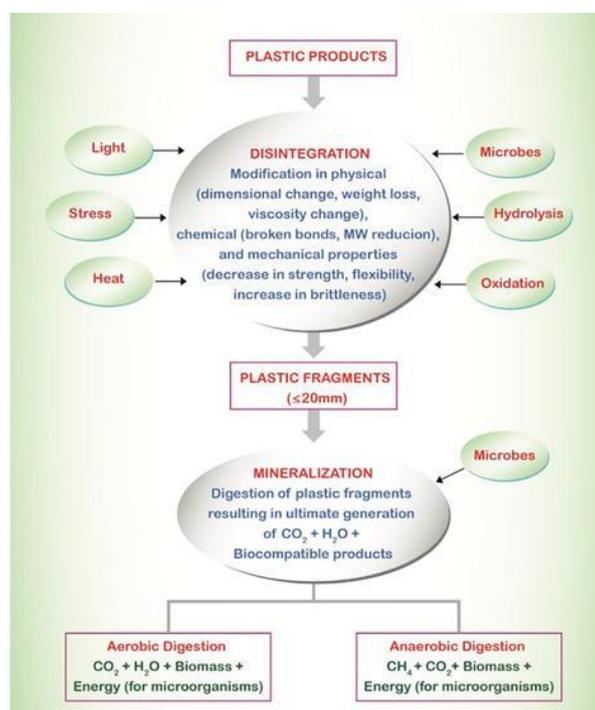


Fig. 1. Mechanism of plastic biodegradation

Biotic degradation

Microplastic biodegrades as a consequence of degradation by microbes in the marine environment. However, because of their size, macroplastics (larger plastic debris) do not make the optimum source of nutrients for biotic degrading agents; either the enzymes secreted by the microbes are insufficient to denature the macroplastics, or they contain not easily and quickly biodegradable for biological cell uptake (Haque and Fan, 2023). Synthetic polymer plastics must first be changed into carbon molecules prior to being mineralized by microbial pathogens during the degradation reaction. Plastics' (polymers') organic molecules' size is bigger than the particle sizes of a microorganism's cellular membrane. As a result, they must be metabolized into tiny pieces before being assimilated and biodegraded within microbial cells. As a result, finer particles of plastic created as the result of environmental factors degradation are of sufficient size to be broken down even more by microbial cells (Zhang et al., 2021).

Bacteria, fungi, and algae are the most common microorganisms found in marine ecosystems. Microbial enzymes are responsible for biotic degradation. Chemical compounds are converted into simplified chemical compounds, metabolized, and deposited in primary-level cycles, such as carbon, nitrogen, and sulfur through microbial degradation. Carbon dioxide, methane, and microbial extracellular matrix components are among the by-products of this system (Chandra, 1998; Banker et al., 2016). Microbial character traits, such as microbe form, propagation, developmental stage (temperature, pH, availability of oxygen, essential minerals, etc.), and enzymatic categories (intracellular and/or extracellular enzymes contributing to exo or endo polymer cleaving). Surface conditions (size, water-soluble, and hydrophilicity properties), first-order frameworks (chemical composition, molecular mass, and molecular dissemination), and relatively high structures (thermodynamic stability, melting temperature, fracture toughness, crystalline structure, and degree of crystallinity) are among the chemical and physical properties of polymers (Tokiwa et al., 2009).

Bacterial biodegradation of plastics

Many bacteria genera that are commonly found in the marine environment like *Bacillus* species e.g., *Bacillus subtilis* and *Bacillus cereus*, and *Bacillus megaterium*, *Brevibacillus*, *Streptomyces*, *Amycolatopsis*, *Clostridium*, *Methanosarcina barkei*, *Schlegelella*, *Pseudomonas aeruginosa*, *Azotobacter* spp., *Alcanivorax*, *Hyphomonas*, and *Cycloclasticus* species, *Rhodococcus ruber*, *Serratia marcescens*, *Staphylococcus aureus*, and *Streptococcus pyogenes*, and other bacterial strains also lead to the microbial

degradation of plastics (Arefian et al., 2023; Haque and Fan, 2023; Nakamiya et al., 1997). The *Bacillus* species were discovered to secrete extracellular hydrolytic enzymes, such as lipase, xylanase, keratinase, chitinase, and protease, which resulted in the biodegradation of microplastics (Chandra et al., 2020). *Methanosarcina barkei* bacteria strain can degrade the most commonly used plastic polymer, PVC. They can stick to the surface of PVC surfaces and discharge exopolymeric compounds to produce a biofilm, preceded by the discharge of enzymes to breakdown the plastic through enzymatic hydrolysis of the synthetic polymer bonds which resulted in the biodegradation of PVC (Nguyen et al., 2016, Reisser et al. 2014). Likewise, *Rhodococcus ruber* will also degrade PE by producing an enzyme laccase, which ultimately resulted in PE degradation (Santo et al., 2013). *Azotobacter* spp., which releases hydroquinone peroxidase, could also degrade PS. PET can also be degraded by *Alcanivorax*, *Hyphomonas*, and *Cycloclasticus* species, which could also alter the physiochemical properties through the use of ester bond hydrolysis (Denaro et al., 2020).

Biodegradation by fungi

Many fungal genera, such as *Acrimonious*, *Zalerion maritimum*, *Curvularia sp.*, *Cladosporium*, *Debaryomyces*, *Emericellopsis*, *Eupenicillium*, *Fusarium*, *Mucor*, *Paecilomyces*, *Pullularia*, *Rhodosporidium*, *Verticillium*, *Aspergillus sp.*, *Aureobasidium*, *Chaetomium*, *Cryptococcus*, *Fusarium*, *Rhizopus arrhizus*, *Trichoderma*, *Penicillium sp.*, *Thermoascus*, *Tritirachium album*, *Humicola insolens*, *Rhodotorula aurantiaca*, and *Kluyveromyces sp.* (Alariqi et al., 2006; Motta et al., 2009; Ojha et al., 2017; Tokiwa et al., 2009; Webb et al., 2000) also contribute to the microbial degradation of plastics. It has been demonstrated that *Aspergillus clavatus* can biodegrade LDPE (Mor and Sivan, 2008). *Zalerion maritimum*, the ocean's dominant fungal species, could also degrade PE (Zeghal et al., 2021). The main mechanism of plastic degradation by fungi, such as bacteria, involves fungi adhering to the polymer surface, in which they grow to create a biofilm and produce enzymes that degrade the carbon-carbon bonds occurring in the plastic. The above enzymes have the potential to accelerate the oxidation process as well as degrade plastic into tiny pieces (e.g., oligomers, dimers, and monomers). For example, fungi found in marine habitats, such as *Penicillium citrinum* and *Fusarium oxysporum*, breakdown PET, and *Trichoderma harzianum* release manganese peroxidase, lignin peroxidase, and laccase that breakdown PE and PU (Zeghal et al., 2021).

Enzymatic mechanisms involved in plastic biodegradation by fungi

The mechanism of biodegradation involves the action of microbial enzymes on the surface of the plastics. The microbes such as bacteria and fungi attach to the plastic film and inert the enzymes and grow on it by utilizing it as substrate and source of nutrition. Therefore, the polymers slowly get depolymerized and degradation will be compiled by mineralization process, where H₂O (water), CO₂ (carbon dioxide), CH₄ (methane) are end products (Frazer, 1994; Montazer et al., 2019). The ability of fungi was they can invade substrates using enzymes that can detoxify pollutants. Fungi can also produce some surface-active proteins, i.e., hydrophobins to coat hyphae to hydrophobic substrates. The growth of many fungi can also cause small-scale swelling and bursting, as the fungi penetrate the polymer solids (Griffin, 1980). The degradation of plastics by some fungi occurs through the intracellular and extracellular enzymatic systems. the intracellular enzymatic system acts as an internal mechanism for detoxification and plays a major role in fungal adaption (Jeon et al., 2016; Olicón-Hernández et al., 2017; Schwartz et al., 2018; Shin et al., 2018). The extracellular enzymatic system consists of a hydrolytic system that produces hydrolases that are involved in polysaccharide degradation and the unspecific oxidative system involved in breaking down complex structures, such as lignin degradation (Sánchez, 2009). The enzymatic complex is produced mainly by wood degrading fungi, such as *Basidiomycetes* (Sánchez, 2009). the action of fungi on the surface of plastics can be affected by environmental factors such as moisture, pH, temperature, etc. sufficient moisture is required for activation of fungi, appropriate pH environment is required for the action of enzymes on plastic polymer and equally, temperature plays a vital role in this biodegradation process, polymers of high melting point take more time to degrade than polymers of low melting point.

Biodegradation by algae

Algae are frequently used throughout tested microorganisms for investigating the harmful effects of microplastics. However, various algae, both photorespiration and heterotrophic, have been extensively researched for their key responsibilities in the microbial degradation of microplastics (Miloloža et al. 2020; Amobonye et al. 2021). They are capable of removing both inorganic and organic contaminants from a diverse range of environments by soaking up, removing impurities, or metabolizing them into healthy and safe levels (Hoffmann et al., 2020; Hwang et al., 2020).

Future prospects and Conclusion

Investigations into plastic trash of all sizes are necessary to better understand the effects on the environment and human health. According to a Nova Institute study, the world's capacity to produce bioplastics was estimated at 2.11 million tonnes in 2019 and may rise to approximately 2.43 million tonnes in 2024 (Chia et al., 2020). According to Beckstrom et al.'s techno-economic and life cycle effect estimate (Beckstrom et al., 2020), bioplastics might be marketed for as little as \$970 per tonne-1 while also lowering greenhouse gas emissions by 67–116%. It is anticipated that the market for bioplastics would grow and eventually supplant traditional plastics. The sustainable bioplastic materials are currently under development and it is essential to explore innovative materials or polymers from renewable sources for the sustainable manufacturing of bio-based and biodegradable plastics. This is because although some types of bioplastics are bio-based, they are not fully biodegradable and may bring a serious negative environmental impact. The materials used for manufacturing bioplastics must be biodegradable and preferably made from natural renewable resources like biomass, plants, waste resources, microalgae and bacteria (Sidek et al., 2019). These renewable materials must not compete with traditional sources used as foods while being able to reduce the use of non-renewable resources in the long term. The sustainable bioplastic materials are currently under development and it is essential to explore inno On top of that, bioplastics produced must be environmentally and consumer-friendly, such as being free of odor throughout its life-time. These types of bioplastics will have excellent thermal properties that are suitable as packaging material in food and beverage industry.

Abbreviation:

PP- Polypropylene

PET-Polyethylene terephthalate

PS-Polystyrene

PE-Polyethylene

PUR- Polyurethane

HDPE- High density polyethylene

LDPE- Low density polyethylene

PVC- Polyvinylchloride

TPA- Terephthalic acid

PBAT- Polybutylene adipate terephthalate

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