



Review

Microbial Bioremediation of Microplastic Pollution for a Sustainable Ecosystem and Greener Future: A Review

Babita Thakur ^{1,†} , Sukhminderjit Kaur ^{2,*} , Manikant Tripathi ^{3,†} and Pankaj Singh ^{3,*} ¹ University Center for Research and Development, Chandigarh University, Gharuan, Mohali 140413, Punjab, India; babithakur18may@gmail.com² Department of Biotechnology, University Institute of Biotechnology, Chandigarh University, Mohali 140413, Punjab, India³ Biotechnology Program, Dr. Rammanohar Lohia Avadh University, Ayodhya 224001, Uttar Pradesh, India; manikant.microbio@gmail.com

* Correspondence: sukhminderjit.uibt@cumail.in (S.K.); singhpankaj0984@rediffmail.com (P.S.)

† These authors contributed equally to this work.

Abstract

Pollution by emerging contaminants like microplastic is one of the major environmental concerns. Microplastics have become ubiquitous anthropogenic pollutants of aquatic, terrestrial and atmospheric ecosystems, and can generate considerable ecological and health-related risks. Conventional remediation regimes are widely ineffective, due to the physicochemical recalcitrance of polymer matrices. Recent advances in microbial biotechnology have revealed several contrasting microbial taxa and enzyme systems, which can convert or mineralize synthetic polymers through a variety of pathways of complex biochemistry. This review summarizes the current understanding of microbial–polymer interactions, including surface colonization, biofilm-mediated depolymerization, and intracellular uptake of degradation intermediates. It also discusses recent developments in enzyme engineering, strain optimization employing the CRISPR method, and synthetic biology approaches improving catabolic efficiency. The advent of a variety of multi-omics technologies of metagenomics, transcriptomics, and metabolomics has enabled the characterization of novel hydrolases and oxidoreductases with a high potential catalytic efficiency. Advances in nanobiocatalysis, enzyme immobilization, and bioreactor technology improve the scale-up of these processes. Related molecular developments and environmental applications will promote the application of microbial biotechnology as a selective and sustainable tool for the mitigation of microplastic accumulation and the development of a circular bioeconomy that interacts positively with ecosystem resilience.

Keywords: microplastic bioremediation; microbial catabolism; plastic-degrading enzymes; CRISPR; omics technologies; enzyme engineering; nanobiocatalysis; circular bioeconomy



Academic Editor: Gabriella Caruso

Received: 24 April 2026

Revised: 17 May 2026

Accepted: 23 May 2026

Published: 17 July 2026

Copyright: © 2026 by the authors.

Licensee MDPI, Basel, Switzerland.

This article is an open access article distributed under the terms and conditions of the [Creative Commons Attribution \(CC BY\)](https://creativecommons.org/licenses/by/4.0/) license.

1. Introduction

In the 70 years that global plastic production has rapidly increased, microplastics (particles <5 mm) have been accumulating throughout the global aquatic, terrestrial and atmospheric environments, making them one of the most widely available and persistent anthropogenic contaminants of the Anthropocene Epoch. Microplastics are both intentionally manufactured as a primary particle and as a result of mechanical fracturing of larger plastic debris due to abiotic weathering (e.g., UV irradiation, mechanical abrasion, and thermo-oxidative degradation) [1]. Microplastics are able to persist for long periods of time

in the environment, have a relatively high surface-area-to-volume ratio and have physico-chemical heterogeneity. Therefore, microplastics have a long-term capacity to serve as a fine vector of transport and bioaccumulation of persistent organic pollutants, heavy metals, and infectious microorganisms across different trophic levels throughout the environment [2].

Due to their pervasive occurrence in both environmental media and socio-ecological systems, there is increasing recognition of microplastics as an emerging environmental pollutant that is likely to have long-lasting implications for both ecological and human health and well-being; however, current methodologies to address this issue remain inadequate [3]. Methods currently implemented for the remediation of microplastic pollution in the environment are primarily based on conventional physicochemical approaches, such as mechanical separation, filtration, sediment removal, and advanced oxidation processes; each of these suffers from low selectivity, relatively high operational cost, secondary waste generation, and/or limited efficacy against the nano- and sub-micrometer fractions of microplastics [4].

In addition, the inherent chemical recalcitrance of the synthetic polymer backbones (characterized by high molecular weight, hydrophobicity, and strong carbon-carbon or ester bonds) of most microplastics makes them resistant to complete mineralization by these existing remediation methods. Therefore, most of the currently used methodologies to remedy microplastic pollution in the environment simply redistribute these microplastics into the environment as opposed to achieving true degradation, which adds to the long-term ecological risk associated with the continued accumulation of plastic debris in the world's environment [5]. Microbial and biotechnological interventions may therefore represent an innovative alternative to conventional methods of remediating plastic pollution through sustainable means. Several diverse groups of microorganisms have the ability to colonize plastic surfaces, create structured biofilms, and initiate polymer depolymerization due to the secretion of specialized hydrolytic and oxidative enzymes that allow them to convert recalcitrant polymers into lower molecular weight intermediates that can be assimilated into their central metabolic pathways through this biological process [6]. The recent advances made in microbial biotechnology, particularly in the fields of enzyme engineering, CRISPR-based genome editing, and synthetic biology, have allowed for expanded opportunities to apply microbial bioremediation to plastic pollution through the rational optimization of catabolic pathways and the construction of tailored microbial systems with enhanced degradation efficiencies [7].

Furthermore, with the advent of multi-omics technologies, including metagenomics, transcriptomics, and metabolomics, researchers are now able to more effectively discover and functionally characterize novel plastic-degrading enzymes and microbial consortia from complex environmental matrices. Coupled with continued innovation in nanobio-catalysis, enzyme immobilization, and bioreactor engineering, these developments are progressively overcoming the logistical limitations associated with the reaction kinetics, process stability, and scalability of these bioremediation processes [8]. Together, microbial biotechnology is emerging as an ecologically sustainable and transformative strategy to mitigate microplastic pollution and to integrate the goals of plastic remediation with the principles of sustainability, circular bioeconomy, and ecosystem resilience [9]. The recent increase in studies regarding the biodegradation of microplastics is gaining traction among researchers; however, the majority of these studies cover the environmental aspects (e.g., where microplastics are found, what they do chemically, and how to analyze them) while not paying much attention to understanding the relationships between all the components involved in the biodegradation of microplastics through microbes [10]. In addition, previous studies fail to understand the links between emerging research areas such as synthetic biology, CRISPR/Cas genome engineering, and enzyme technologies

(e.g., multi-omics) and microbe-based biodegradation methods [11]. Hence, this review aims to provide a comprehensive and integrative account of the biodegradation of microplastics using microbial-based bioremediation systems, examines achievements along with some of the research that is currently in a bottleneck stage, and discusses the feasibility of transferring existing microbial-based microplastic bioremediation technologies to the field for inclusion in a sustainable and scalable bioremediation strategy, consistent with the principles of a circular bioeconomy.

2. Microplastics in the Environment

Microplastics are a chemically diverse and environmentally persistent class of particulate pollutants that differ in origin, polymer type, and physicochemical properties. Microplastic pollutants are released into the environment, where they will then undergo numerous transport, transformation, and aging processes that modulate their bioavailability, toxicity, and susceptibility to biodegradation by living organisms. Thus, understanding the source, environmental distribution, and intrinsic material properties of microplastic pollutants is essential for the rational design of effective remediation strategies using bioremediation [12].

2.1. Sources and Classification of Microplastics

Microplastics are generally divided into two types, primary and secondary, based on how they originated and how they have entered into the environment. The distinction between primary and secondary microplastics has important implications for the physicochemical properties, environmental fate or behavior, and biodegradation potential of microplastics [13]. Microplastics can be grouped into two categories: primary and secondary, as shown in Figure 1.

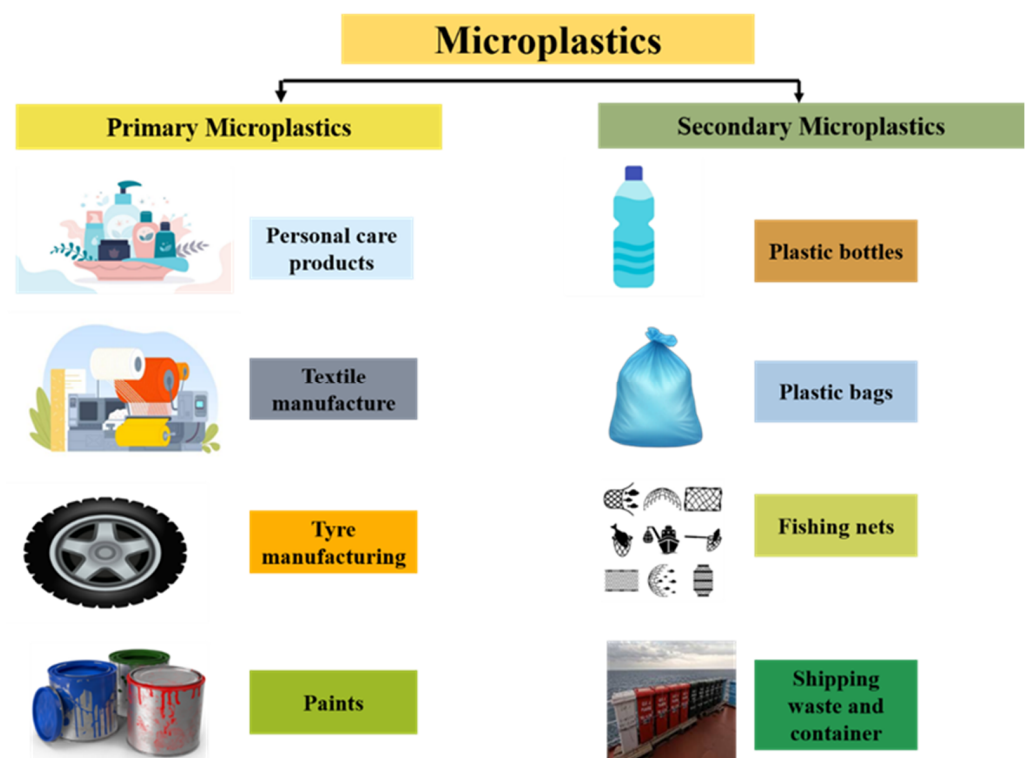


Figure 1. Microplastics classification and sources. (1) Primary microplastics are produced commercially; examples include personal care products, textile production, tire abrasion, and paint. (2) Secondary microplastics are the result of breaking up larger plastic items that deteriorate in our environment (examples include single-use plastic bottles, bags and fishing nets, shipping waste, etc.).

Primary microplastics are formed for use in a given industry or product as a result of human activity. Examples of primary microplastic products include exfoliating agents and polymer-base cleaners or scouring agents. The characteristics of primary microplastics are determined by their size, shape, and the chemical makeup of their surfaces when they are manufactured. Their uniform sizes allow manufacturers to produce them in a uniform shape and chemical composition for all types of industrial uses. Because of their ability to remain in the environment for long periods, primary microplastics often undergo predictable chemical changes when they are released into the environment [14].

Secondary microplastics are derived from macroplastics being broken down into smaller pieces through biological and chemical processes. They can be broken down into smaller pieces and degraded through photodegradation, photooxidation, and thermo-oxidation processes. Both the mechanical forces of the environment and biological processes also contribute to reducing the sizes and shapes of secondary microplastics. Biological processes also assist in the attachment of microorganisms to secondary microplastics, but the complete degradation of secondary microplastics by microorganisms is rare [15].

Secondary microplastics are mainly derived from single-use packaging, fibers of synthetic textiles, fishing nets, tire wear, agricultural mulch films, and greenhouse films. The vast majority of secondary microplastics are created from the continual breakdown of these sources of debris. Microplastics contain many different types of synthetic polymers. Polyethylene (PE) and polypropylene (PP) are highly resistant to chemical and biological degradation and can remain in the environment for long periods. The rigid structure of polystyrene (PS) tends to make it hydrophobic [16]. Other types of polymers, such as polyethylene terephthalate (PET) and polyurethane (PU), polycaprolactone (PCL), and polyamide polymers, have ester/urethane linkages and are therefore much more susceptible to hydrolytic and enzymatic cleavage than PE and PP. PVC is classified as a polymer that contains a relatively high quantity of chlorine-substituted carbons and is therefore also of concern for the potential release of toxic components into the environment [17].

Other classifications of microplastics take into account their size and shape; however, they can generally be divided into three categories: microplastics (or particles smaller than 5 mm), nanoplastics (particles smaller than 1 μm), and macroplastics. Because nanoplastics have smaller sizes and can be moved through the environment more easily than larger particles, they have increased potential to attach to other substances and be readily incorporated into living organisms. Thus, they also have greater potential to be absorbed into cells and transported into other parts of the body. Microplastics, regardless of their morphological characteristics (i.e., fibers, fragments, film, foam, and pellets), will also be affected by their age and by the environmental impact of other organisms attaching to them [18]. In addition, both the chemical composition of the microplastics and the environment in which they are located contribute to the formation of eco-coronas made up of organic matter, inorganic ions, and microbial polymers, which change the physical and chemical properties of microplastics. Therefore, it is necessary to develop a performance-based classification system for microplastics that takes into account all of the different characteristics of microplastics to effectively design bioremediation techniques [19].

2.2. Environmental Distribution of Microplastics

Microplastics occur in all ecosystems, including aquatic, terrestrial and atmospheric, which shows they have been produced in large quantities and can easily move throughout the environment. The aquatic environment is the most studied ecosystem when it comes to microplastics because they collect at the surface of the water, within the water column, in sediment, and in the organisms of both freshwater and marine ecosystems [20]. The forces that move microplastics vertically and horizontally are determined by hydrodynamic

forces, salinity gradients and biofouling, which allow microplastics to be transported over large geographic distances within ocean basins. In addition to aquatic ecosystems, the terrestrial companion of microplastic accumulation is rapidly gaining more recognition. Agricultural soils, landfills, and urban areas are being recognized as major sinks for microplastics. There are many ways to introduce microplastics to agricultural soil, including using sewage sludge as fertilizer, using plastic mulch in farming, irrigating with water that contains microplastics, and deposition of microplastics in the atmosphere [21]. Within the soil environment, there is potential for long-term accumulation of microplastics due to a lack of removal by physical means and limited degradation by ultraviolet (UV) light (photodegradation). The relationship between microplastics, mineral particles in the soil, organic matter and soil microorganisms alters the physical properties of soil, nutrient cycling, and the composition of soil microorganisms. Atmospheric microplastics are a newly identified and important pathway for global distribution of microplastics [22]. Fibrous and fragmented microplastics can become aerosolized through wind, urban activities or from abrasion of textiles, and can be transported through the atmosphere over significant distances. Through both dry and wet deposition, microplastics can be introduced into remote and pristine areas of the world such as in polar and mountainous ecosystems, demonstrating how microplastics are distributed over the earth [23].

2.3. Physicochemical Properties Influencing Biodegradability

Microplastics' capacity for biodegradation is directly correlated to the physicochemical characteristics that engage microorganisms in adhesion and enzymatic accessibility, as well as susceptibility to depolymerization. Physicochemical characteristics are dictated by the chemistry of the polymer chain backbone, the polymer's molecular weight, the polymer's degree of crystallinity or amorphosity, the degree of hydrophobicity of the polymer, the roughness of the polymer surface, and the presence of functional groups on the polymer chain that can be targeted by enzymes [24]. Polymers made up predominately of carbon-carbon backbones such as PE and PP are exceptionally resistant to biodegradation due to having no hydrolysable bonds and being highly stable structurally [25].

On the other hand, polymers with functional groups such as esters, amides and/or urethanes (e.g., polyethylene terephthalate (PET), PUs, and polyamides) tend to have considerably greater enzymatic hydrolysis vulnerability than those that do not. The presence of oxygenated functional groups, increased roughness or decreased polymer molecular weight due to photooxidation by ultraviolet light, azoic agents and/or mechanical wear over time are potential augments/mediators for biodegradability [26]. Alternatively, the addition of agent types, such as plasticizers, stabilizers, flame retardants or fillers, may adversely affect or unpredictably alter pathways for microbial degradation. The assembly of eco-coronas composed of organic matter, metals, and microbial exopolymers on microplastics' surfaces can heavily impact surface chemistries, and subsequently the interactions and associations between microbial populations and microplastic substrates [27].

As a whole, the physicochemical characteristics of microplastics pose significant obstacles to effective biological degradation. Technological innovation using advanced biotechnology, including enzyme and strain development, as well as the creation of synthetic consortia, is necessary to overcome resistance to degradation from polymers. To facilitate rational applications of microbial and enzyme-based remediation strategies, scientists must possess the requisite knowledge regarding the characteristics of microplastic materials [28].

3. Microbial–Microplastic Interactions

The interaction of microorganisms with plastic is the primary mechanism of biological control over the fate of plastic in the environment and in engineered systems. These interactions are complex and depend on the chemical properties of plastics, the physical and chemical characteristics of their surroundings, and the metabolic capabilities of associated microbial communities. Microbial adhesion, biofilm production, extracellular depolymerization, and intracellular assimilation are sequentially interconnected; thus, the ability to biologically transform microplastics rather than simply colonizing them is determined by these processes combined [29].

3.1. Initial Microbial Adhesion and Surface Colonization

The process of organisms attaching themselves to microplastic surfaces is the first stage and very often the slowest step in terms of breaking down of microplastics by organisms. The adhesion of microorganisms to microplastics is a product of multiple types of interactions, including hydrophobic forces, electrostatic forces, van der Waals forces and hydrogen bonds, between the cells of the microorganisms and the polymer that make up the microplastics [30]. Hydrophobic polymers like polyethylene and PP naturally attract the hydrophobic cell envelopes of bacteria; thus, the bacteria can easily passively adsorb the polymers. When the microplastics have oxidized surfaces or aged in the environment, polar functional groups (e.g., -OH) are infused into their surfaces, which allows microorganisms to more easily identify and attach themselves to the microplastics [31].

The microplastic surface roughness, the size and shape of the microplastic particles, and the presence of organic material adsorbed to the particle surface all influence how easily a microorganism can adhere to a microplastic particle, since the surface roughness creates more opportunities for a microorganism to form a bond with the surface of the particle, as well as decrease the likelihood that the microorganism will be removed (i.e., detached) from the particle as a result of shear forces applied during fluid flow [32]. At the cellular level, adhesion of microorganisms to microplastic surfaces is mediated through specific surface structures, including pili, fimbriae, flagella, and outer membrane proteins. These structures provide a mechanism for microorganisms to form “irreversible” attachments and facilitate surface sensing. The development of a conditioning layer that consists of organic macromolecules and extracellular polymeric substances (EPSs) very rapidly alters the nature of the microplastic surface in terms of its physicochemical characteristics, which allow for colonization by a wide range of microorganisms. Once colonization occurs by these early colonizers, a distinct microbial community, or “plastisphere,” develops, which differs markedly from the microbial communities that are present in the surrounding water column (i.e., plankton) [33].

3.2. Biofilm Formation on Microplastic Surfaces

The accumulation of organisms and secreted EPS on surfaces of microplastic material after the adherence of single-celled organisms allows for the proliferation of the organism(s) and forms multiple structured layers of organismal material. The biofilm represents a three-dimensional structure that allows for a collection of microorganisms to remain metabolically active and resistant to extreme conditions such as decreasing nutrient supply or decreasing environmental moisture and resistance to oxygen exposure. Polysaccharide, protein, and lipid polysaccharide constituents primarily make up the biofilm matrix, enabling close proximity between microorganisms, thus permitting signaling and spatial organization of the collection of microorganisms via quorum-sensing mechanisms [34]. Microplastic-affiliated biofilms occur with pronounced spatial heterogeneity; gradients of oxygen, nutrients, and metabolic by-products have different stimulation effects on the performance and func-

tional specialization of different collections of microorganisms. Microplastic-associated biofilms act as biogeochemical hotspots by concentrating enzymes and metabolites at the polymer–microbe interface. Therefore, by providing a substrate for microorganisms, plastic-associated biofilms represent more than simply a “home” for the organisms; they also function in enzyme activity on the surface of the associated polymers [35].

3.3. Biofilm-Mediated Depolymerization Mechanisms

Microplastics are primarily depolymerized outside of the cell via enzymatic activity by bacteria contained within biofilms. The classes of enzymes that contribute to this process are hydrolases (esterases, lipases, cutinases, etc.) and oxidoreductases (laccases, peroxidases, monooxygenases, etc.), which catalytically cleave the polymer chain of microplastics through hydrolysis and oxidation. Under certain conditions over time due to abiotic factors that introduce new chemical functional groups and decrease the polymer crystallinity, the ability of these enzymes to degrade polymers is enhanced. Inside the biofilms, the proximity of the enzymes within the matrix of the biofilm creates a concentrated environment for the enzymes to work together, thereby allowing for increased catalytic activity and extended interaction between the substrate and enzyme [36]. Enzyme systems may work together in a sequential manner to convert larger microplastic polymers into smaller molecular components; however, the efficiency of polymer depolymerization is dependent on the arrangement of the polymer structures with carbon–carbon backbone polymers being more resistant to degradation when compared to a polymer that has a hydrolysable bond structure. Therefore, the microbial community structure found in biofilms, along with the redundancy of enzyme function among various bacterial species, may play a critical role in enabling microbes to successfully degrade certain polymer types that are more challenging to degrade than others [37].

3.4. Cellular Uptake and Metabolism of Degradation Intermediates

Low molecular weight intermediates of microplastics are formed via extracellular depolymerization and include oligomers, monomers and oxidized low molecular weight hydrocarbons. The intermediates formed from the extracellular depolymerization of microplastic polymers can be taken inside bacteria and subsequently metabolized. Microbes use either passive diffusion or active transport mechanisms to move the degradation products into the cells, based on the size and polarity of the compounds [38]. After entering the microbial cell, microplastic-derived materials can enter cell metabolic pathways via two mechanisms: they can be metabolized via β -oxidation, or a combination of glycolytic and TCA metabolic pathways, depending on the type of microplastic that formed the products. When the microplastic-derived compounds are metabolized, they are generally done so by a specific set of enzymes which are involved in the degradation of these compounds, e.g., dehydrogenases, hydratases and ring-cleaving enzymes [39]. Catabolic pathways for any of these groups of compounds allow the production of biomass, energy, or the complete mineralization of intermediates to carbon dioxide and water. However, many times during the catabolism of the microplastic-derived products, there are literally bottlenecks in the pathways (such as incomplete or toxic and less efficient pathways). These factors point towards further improving the efficiency of these pathways by either utilizing metabolic engineering or synthetic biology, or optimizing the pathways using techniques such as CRISPR or other biotechnological methods [40].

4. Microbial Diversity Involved in Microplastic Biodegradation

Microplastics degrade through microbial activity. These microbes are highly varied in phylogeny and function, and their different metabolic capabilities reflect extensive evolu-

tionary histories that have adapted the microbes to use very complex structures and highly resistant carbon sources as energy. Microbial diversity also determines the degradation efficiency, complete pathway breakdown, and environmental adaptability, as different taxa have unique enzymatic repertoires, ecological approaches, and mechanisms for coping with stress. Identifying the range of taxa and specialized functions of bacteria that can break down plastics will help develop methods for biologically based remediation [41].

4.1. Plastic-Degrading Bacteria

Microplastics biodegradation has been well established by microorganisms, specifically bacteria, which comprise the most metabolically adaptable and diverse group of species. Due to the speed with which they grow, their ability to adapt genetically and their versatile enzymatic capabilities, bacteria are suited for the degradation of plastics [26]. As such, plastic-associated microbial communities typically consist of members of the phyla *Proteobacteria*, *Firmicutes*, *Actinobacteria*, and *Bacteroidetes*, many of which are bacterial species that can form biofilms and attach to polymer substrates. Several Gram-negative bacteria have been reported to have the potential for degradation of plastic substrates. Bacteria from the genus *Pseudomonas putida*, *Pseudomonas aeruginosa* (for example, *P. putida*, *P. aeruginosa*) have been recognized for their ability to oxidatively modify polyethylene (PE) and PP and for their ability to metabolize hydrocarbons derived from plastics [42]. Bacteria from the genera *Rhodococcus ruber*, *R. opacus* possess highly hydrophobic cell wall components and demonstrate the ability to secrete extracellular oxidases that promote the erosion of the surface of polyethylene [43]. The complete degradation of PU and polyamides (PA) by the bacteria *Alcaligenes faecalis* and *Comamonas testosteroni* has been attributed to the activity of esterases and amidases. The landmark example of plastic biodegradation by bacteria is represented by *Ideonella sakaiensis*, which produces two enzymes known as PETase and MHETase, which can selectively depolymerize PET into monoethylene glycol (MEG) and terephthalate acid (TPA) [44]. Examples of other PET-degrading bacteria include *Thermobifida fusca*, *Clostridium botulinum*, and *Bacillus subtilis*. The cutinase-like enzymes produced by these bacteria do not possess the same level of thermal stability or catalytic efficiency. The genera *Bacillus*, *Paenibacillus*, and *Lysinibacillus* provide additional contributions through the production of extracellular enzymes and when it comes to environmental adaptability and resistance. While these advances have been made, most Gram-negative bacteria are not capable of degrading polyolefins environmentally; through the application of genetic approaches, including the use of CRISPR technology to develop enhanced pathways and the use of laboratory-based adaptation to improve on catabolic flows and enzyme secretion, improved plastic degradation can take place through the development of polyolefin-degrading bacteria [45,46].

4.2. Fungal and Actinomycetes Systems

Numerous climatic, ecological, and hydrological factors affect the rate at which fungi and actinomycetes contribute to the biodegradation of plastics by their interaction with the plastics during their initial stages of surface modification. Fungi, with their extensive networks of hyphae, have a larger surface area and stronger attachment to polymers than bacteria because they have the ability to physically penetrate into the polymers. In addition, fungi continuously secrete enzymes that help to degrade polymers, while bacteria often do not [47]. Many fungi have been reported to be able to colonize and degrade polyethylene, PS, PU, and polycaprolactone to varying degrees, including *Aspergillus* (such as *A. niger*, *A. flavus*), *Penicillium* (such as *P. simplicissimum*), *Fusarium*, *Cladosporium*, *Phanerochaete*, and *Trametes*, while white rot fungi, such as *Phanerochaete chrysosporium* and *Trametes versicolor*, are particularly noted for their ability to produce ligninolytic enzymes such as laccases, manganese peroxidases, and lignin

peroxidases, which allow them to introduce oxidative functional groups into polystyrene and other previously inert polymer backbones [48]. Additionally, actinomycetes, especially *Streptomyces*, *Nocardia*, *Amycolatopsis*, and *Micromonospora*, possess both filamentous and bacterial characteristics and produce a wide variety of hydrolytic, oxidative, and esterolytic enzymes that aid the depolymerization of PU, PT, and PA. The ecological resilience, secondary metabolite production, and ability to survive in nutrient-limited environments of actinomycetes make them attractive candidates for bioremediation efforts through the use of these organisms in soil and over the long term. However, both fungal and actinomycete-mediated bioplastics degradation processes take time and are often incomplete, thereby suggesting a need for the development of synergistic relationships between these two groups of organisms and the incorporation of sophisticated biotechnologies that enhance the efficiency and completeness of the biodegradation process [49].

Microplastics in nature and through human activity have been mostly biodegraded by complex groups of microorganisms working together (consortia) instead of specific microorganisms on their own. There are interaction pathways between different groups of microorganisms that work together to provide each other with parts necessary to complete degradation of microplastics. This allows many microorganisms to use different metabolic pathways, work together simultaneously and create a greater level of catabolism than can be accomplished by any one microorganism [50]. In a consortium, the first microbes breaking down the plastic will modify the surface of the polymer so that they can pull it apart (depolymerization) either through oxidation (adding oxygen) or hydrolysis (using water), to create intermediate products like oligomers and monomers. These are then taken up and metabolized by other microbes in the community (the secondary consumers/allied group) that use the degraded products as a carbon source, detoxify potential intermediates, and enhance the carbon flow between all members of the consortia via metabolic networks [51].

The functional performance of consortia is improved through cross-feeding (the sharing of metabolites between species), spatial compartmentalization (e.g., biofilm) and cooperation (quorum-sensing or other signaling mechanisms). Organisms in biofilms form their own microenvironments within the overall biofilm, which are characterized by chemical gradients and allow groups of organisms (the “degraders”) to continue oxidizing, hydrolyzing and assimilating their products while working together in close proximity. This spatially organized cooperative effort increases the efficiency of the enzymatic reactions that degrade the microplastic, establishes metabolic pathways that allow utilization of numerous sources of energy, and sustains the microbes’ viability under non-optimal conditions due to changing environmental conditions [52]. The advances in synthetic biology and systems biology will now allow researchers to rationally engineer synthetic microbial consortia for use in degrading microplastics, i.e., by combining organisms that have complementary enzymatic access and metabolic capabilities, and to create consortia that can degrade all forms of plastic and overcome the issues related to polymer recalcitrance. Omics-based technologies such as metagenomic sequencing and reconstruction of existing metabolic networks have provided unprecedented insights into the functional capacities, functional redundancy and major constraints in the degradative process of plastic microorganisms (microplastics). This has enabled researchers to optimize the composition and performance of engineered consortia, resulting in a robust, replicable and environmentally compatible approach to advancing the biodegradation of microplastics through the use of engineered consortia [40,53].

4.3. Engineered Microbial Consortia for Multi-Polymer Degradation

Often, single isolated microbial species lack the diversity of enzymes necessary for efficient degradation of mixed sources of microplastic pollution. With synthetic biology, synthetic microbial consortia can be designed, where multiple species will be used together

to increase their metabolic versatility through complementary metabolic pathways. For example, some of the organisms in a consortium might perform hydrolytic depolymerization (i.e., bacteria with PET-like activity), while some perform oxidative priming, (i.e., laccase), and others perform monomer creation, assimilation, and mineralization [54]. The way the consortia are spatially organized can also contribute to increasing the metabolic activities of each of the consortia organisms, when they communicate with each other through a form of signaling (quorum sensing) or by developing their own synthetic signaling molecules, substrate separation, and metabolic cooperation. This division of labor results in reduced metabolic stress for each of the individual microbes and decreased accumulation of intermediates, and allows simultaneous degradation of several different types of polymers, including PET, PE, PP, and PS [55]. The use of computational modeling and synthetic ecology as tools to optimize community composition, stability, and resilience offers a predictive model for creating multifunctional hybrid microbial consortia to use for bioremediation of large amounts of microplastic waste [56].

5. Enzymatic Mechanisms of Microplastic Degradation

Microbial biodegradation of plastics is dependent upon enzymatic depolymerization as the main biochemical process. Polymer crystallinity, hydrolysable bonds and substrate accessibility are all obstacles to effective enzymatic attack due to the recalcitrant nature of synthetic polymers. Plastic-degrading enzymes therefore have evolved specific catalytic characteristics that permit them to initiate bond cleavage on hydrophobic, high-molecular-weight substrates in the environments in which they occur. Plastic-degrading enzymes fall into two main categories: hydrolases and oxidoreductases. These enzymes typically act together to achieve full or partial polymeric degradation [57], as shown in Figure 2.

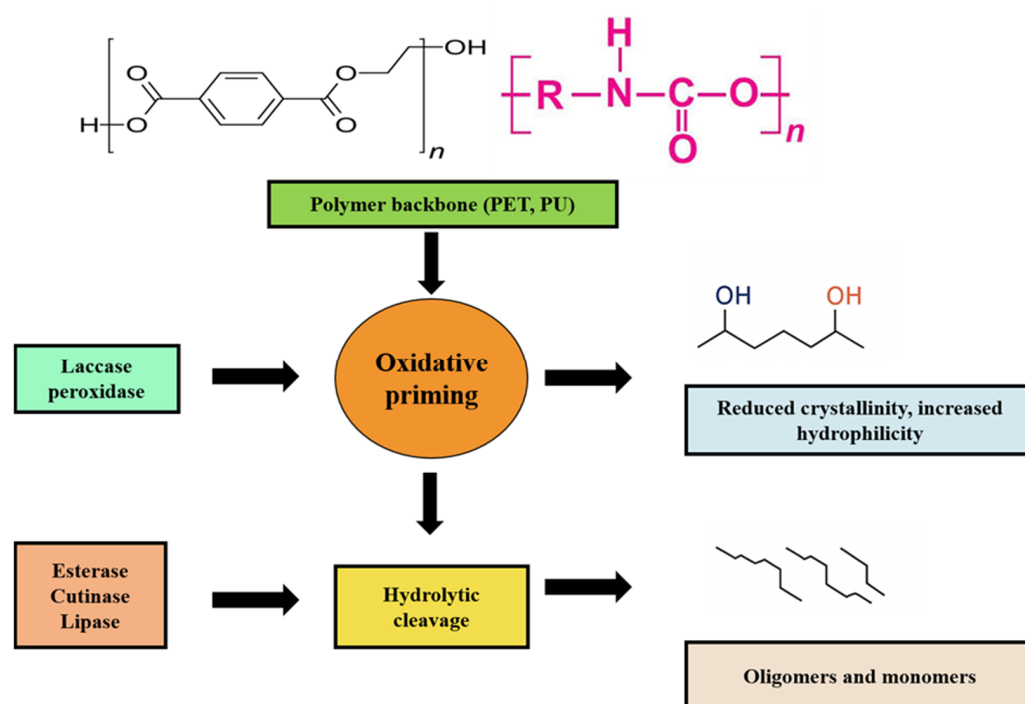


Figure 2. A synergistic enzymatic system with oxidative and hydrolytic activity is responsible for degrading microplastics. The oxidative enzymes, laccase and peroxidase, activate polymer backbones through oxidation (oxidative priming), including polyethylene terephthalate (PET) and polyurethane (PU), by reducing their crystallinity and increasing their hydrophilicity. Subsequently, the hydrolytic enzymes, esterase, cutinase, and lipase, cleave the polymer chains to generate small oligomers and monomers for further assimilation by microbes.

5.1. Hydrolases Involved in Polymer Depolymerization

Hydrolases are the major class of enzymes that catalyze the breakdown of synthetic polymers containing hydrated functional groups through depolymerization. They do this by hydrolyzing ester, amide, and urethane bonds via nucleophilic attack, which is typically facilitated by three amino acids (serine, histidine, aspartate) at the binding site of the enzyme, which creates oligomeric and monomeric intermediates that may enter into microbial metabolism [58]. The major categories of hydrolases studied to date regarding degradation of plastics and microplastics include esterases, lipases, cutinases, carboxylesterases, and proteases. Each group has varying degrees of specificity against different polymers, i.e., PET, PU, PCL, and polyamide polymers. Hydrolytic enzymes that are effective for degrading PET (PETase, mono-(2-hydroxyethyl) terephthalate hydrolases (MHETase)) exhibit a distinct enlarged substrate-binding site, allowing them to accept large, rigid aromatic polymer structures into the active site to enable access to the ester bonds located within shut-in crystalline areas [59].

Hydrolytic enzymes of other types are less specific for polymers than esterase-hydrolytic enzymes. Cutinase-like enzymes have been derived from both bacteria and fungi and have broader substrate promiscuity, suggesting they can hydrolyze aliphatic-aromatic polyesters and biodegradable plastics through surface erosion mechanisms. Their more open active-site architecture and interfacial-activated nature provide them with the ability to interact with insoluble polymer substrates [60]. Urethanase and amidase enzymes are involved in degrading polyurethane and polyamide materials through carbohydrate and amide-bond recognition, respectively. In many cases, these enzymes work in concert with other hydrolases and oxidoreductases to facilitate sequential depolymerization, particularly with respect to complex polymer blends containing many different types of polymers. Although hydrolases have this depolymerizing ability, their ability to do so is still strongly limited by the polymer crystallinity, molecular weight, hydrophobic surface properties, and limitations to enzyme accessibility [61]. Environmental factors, such as temperature and pH, and substrate availability, all greatly influence the activity of hydrolases and frequently result in the slow and incomplete depolymerization under ambient conditions. Therefore, these limitations indicate the need for enzyme engineering, surfactant modification, and improvement of the hydrolytic efficiencies of these enzymes to make them suitable for the bioremediation of microplastics [62].

5.2. Oxidoreductases and Auxiliary Enzymes

Hydrolases are directly responsible for the hydrolysis of polymeric bonds. Oxidoreductases and their auxiliary enzymes initiate a number of oxidative forms of modification to the polymer, thereby allowing them to become susceptible to the hydrolytic effects of hydrolases. Oxidoreductases and their auxiliary enzymes achieve these forms of modification by catalyzing redox reactions that produce reactive oxygen intermediates (ROS) as byproducts of the reaction. Furthermore, the production of polar functional groups on the polymer backbone provides the opportunity for additional hydrolytic modifications via subsequent oxidation [63]. Hydroxylation, carbonylation, and epoxidation are examples of oxidative on polymer backbones; when polymers undergo oxidation to introduce a hydroxyl or carbonyl group, the polymer chains will become less crystalline and more hydrophilic, and therefore create a more open and flexible polymer chain. The addition of functional groups on the polymer surface provides greater mobility, thus allowing for greater permeability of the polymer. In turn, the increased flexibility and the increase in the polymer surface area provide a much larger opportunity for the hydrolytic effect of hydrolases on the polymer. The incorporation of laccases onto polyethylene and PP has been observed to increase the surface area of the substrates due to the presence of hydroxyl

and carbonyl groups along the polymer backbone [64]. Auxiliary enzymes (glycosyl hydrolases and cellulases) and small-molecule electron mediators (redox-active phenolics) frequently act together with oxidoreductases, destabilizing polymer crystalline domains and shuttling electrons between the polymer–enzyme interface, enhancing the rate of the radical-mediated depolymerization pathway. The use of electron mediators (e.g., ABTS or HBT) to enhance the turnover of laccases and broaden the range of polymers that can be attacked illustrates the cooperative or synergistic nature of oxidoreductases and auxiliary enzymes in an engineered system. The combined action of oxidoreductases and auxiliary enzymes constitutes a coordinated, “priming” mechanism for hydrolytic cleavage due to the lowered activation energy requirement for hydrolytic cleavage, which subsequently facilitates the degradation of high molecular weight microplastics in environmentally relevant conditions [65].

5.3. Structure Function Relationships of Plastic-Degrading Enzymes

The efficient catalytic activity of the enzymes that can break down plastics can be explained by their three-dimensional geometric configuration at the molecular level as well as how they interact with the surface of polymers, which includes the surface areas of the polymers that bind to the enzyme’s surface. Studies of the structure of the enzyme using X-ray crystallography, cryo-EM, and nuclear magnetic resonance spectroscopy (NMR) have shown that the configuration of the enzyme’s active site, the size of the tunnel the substrate passes through, and the hydrophobicity or hydrophilicity of the surface of the enzyme have a direct effect on how well it will recognize and attach to the polymer [66]. An example of an enzyme that can break down plastics is *Ideonella sakaiensis* PETase, which has a surface configuration that allows for a shallow, solvent-accessible active site, allowing for the binding of PET and a nucleophilic attack on the ester bond, while lipases are more rigid enzymes that typically metabolize smaller aliphatic esters [67].

The studies of molecular dynamics and computer modeling of molecular docking have demonstrated the ability of certain enzymes to adapt their flexibility to the complex structures of polymer chains, enabling the polymer chain to be held in the correct orientation, bind to the enzyme, and create transition states through the organic components, such as the interfacial loop residues and surface residues around the catalytic triad [68]. The use of structural information generated by X-ray crystallography, cryo-EM, NMR, and other studies has been used to further develop the thermostability, substrate specificity, and solvent tolerance of the enzymes through site-directed mutagenesis, enabling the enzymes to retain their catalytic abilities in various extreme situations. The structure can also be affected by the addition of covalent modifications that occur after translation, such as glycosylation and the formation of disulfide bridges, which stabilize the enzyme and help prevent them from being degraded by proteases [69]. Many enzymes combine their catalytic activities through synergistic relationships to increase catalytic efficiency; for example, peroxidases work synergistically with laccases, while lipases and oxidoreductases create a sequential oxidative approach, which increases their respective substrate affinity and catalytic efficiency. The basic aspects of these enzymatic mechanisms will be used to devise and engineer enzymes that will be effective for producing stable biocatalysts that can degrade a variety of recalcitrant microplastics, such as PET with high crystallinity, polyethylene, PP, and PE [70].

The build-up of microplastics across the globe within land-based and aquatic environments poses serious risks to the environment as well as human health due to their resistance to chemical breakdown, long-term presence in the environment, and potential for bio-concentration in living organisms. Many traditional methods for the removal of microplastics which typically employ physical or chemical means fail to adequately address

this challenge through the promotion of alternative methods of pollution control such as environmentally compliant disruption of plant materials; as a result, researchers are looking to employ biotechnological approaches to remove polymers [71]. In order for microbes to degrade microplastics they must work together to activate both hydrolytic and oxidative enzymes; however, much of the catabolism of many naturally occurring microbial types would not be adequate to biodegrade high-crystallinity polymers or highly engineered synthetic blends. As a result, there have been several exciting new possibilities afforded by advances in CRISPR/Cas technology due to its ability to allow for targeted and effective design of microbial strains with enhanced plastic-degrading capabilities [72]. Through its capacity to create precise adjustments to metabolic pathways, regulate metabolic pathways, and regulate the synthesis of the enzymes within microbe chassis, the CRISPR-based approach results in the development of microbial chassis with the ability to remove microplastics from the environment via efficient, effective, and safe bioremediation processes. The following section will outline the available CRISPR/Cas toolset, as well as describe CRISPR/Cas toolset approaches for the creation of next generation plastic-degrading microbes and the advantages to utilizing biosafety criteria when creating microbial chassis for sustainable microplastic bioremediation [73]. While much progress has been made in our understanding of the microbial enzymatic breakdown processes of microplastics, naturally producing microbial enzymes have low catalytic efficiencies and limited substrate specificity and environmental stabilities (i.e., function outside of the laboratory), and presently face challenges in degrading highly crystalline and chemically resistant polymers, such as PE and PET [74]. Consequently, researchers are utilizing advanced genetic engineering techniques (i.e., targeted modification of metabolic pathways, optimization of enzyme expression, improvement of substrate utilization efficiency, etc.) to enhance microbial degradation capabilities. Furthermore, CRISPR/Cas-based genome editing is being utilized as an innovative method to create advanced future generation microbial systems with enhanced plastic biodegradation capabilities [75].

6. CRISPR/Cas Systems in Microbial Genome Editing

Microbial synthetic biology has been greatly impacted by the advancement of CRISPR/Cas technology, specifically through the use of Class II Effectors (Cas9 and Cas12a) to provide highly specific programmable genome-editing capabilities for microbes [76]. Whereas other enzymes mediated by these systems will create double-stranded breaks (DSBs) in the target DNA in order to facilitate recognition of Protospacer-Adjacent Motifs (PAMs), the engagement of the Effectors Cas9 and Cas12a represents the initiation of a sequence specific targeting mechanism within the microbe's genome. For example, when used within the context of microplastic bioremediation, CRISPR/Cas can allow for the incorporation of natural or synthetic plastic-degrading genes, the disruption of biochemical pathways that inhibit the breakdown of plastics, or the improvement/modification of a microbe's native metabolic pathways involved in anaerobic degradation of plastics [77], as shown in Figure 3. CRISPR/Cas also provides methods that allow for the nearly precise incorporation (i.e., insertion) of synthetic operons that encode PETase, MHETase, cutinases, and laccases through homology-dependent repair (HDR) pathways as well as for the random deletion of competing metabolic pathways through non-homologous end joining (NHEJ).

Most recently, CRISPR/Cas technology has evolved to base editing and prime editing; these newer approaches allow for the targeted substitution of single nucleotides, without the associated generation of double-stranded breaks, which would otherwise create unwanted off-target effects, thereby potentially increasing the stability of the resulting genome. Additionally, CRISPR provides the capability of multiplex genome editing (i.e., simultaneous engineering of multiple genes, operons, and regulatory elements) to produce organisms

with the potential for producing synergistic enzyme systems for the better degradation of microplastics [78,79].

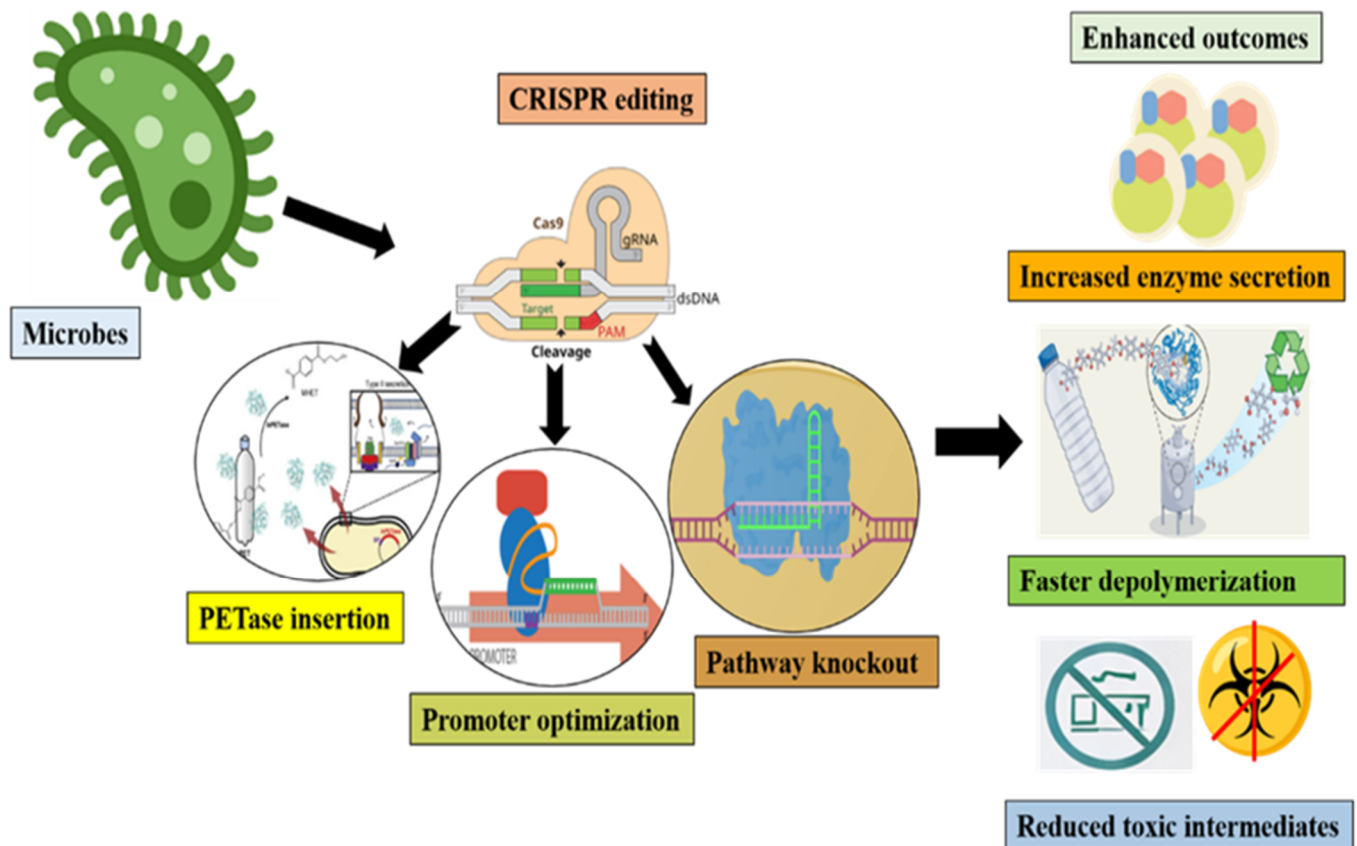


Figure 3. CRISPR/Cas-directed improvement of microbial systems that can enhance microplastic biodegradation. Methods for genome editing using CRISPR, such as adding PETase, optimizing promoters, or knocking out metabolic pathways, could lead to improved degradation of microplastics by increasing secretions of enzymes improving depolymerization rates and reducing accumulation of toxic intermediates during biodegradation of microplastics. PETase: polyethylene terephthalate hydrolase; CRISPR: clustered regularly interspaced short palindromic repeats; Cas: CRISPR-associated protein.

6.1. Engineering Plastic-Degrading Metabolic Pathways

Microplastics (i.e., plastics that are less than 5 mm) are notoriously difficult to biochemically degrade due to their polymeric structure. Because of this difficulty, the successful biodegradation of microplastics requires the co-ordination of various enzyme-catalyzed metabolic reactions to overcome this polymeric structure. Genomic editing technology (i.e., CRISPR) can aid in the design and optimization of the many enzymatic pathways involved in biodegradation of microplastics [80]. With the help of an array of hydrolytic (e.g., PETase, cutinase, lipase), and oxidizing enzymes (e.g., laccases, peroxidase, monooxygenase), the structure of the microplastics can be altered and destabilized through the introduction of polar functional groups (i.e., hydroxyls, themoacids, etc.), which increases the solubility of the microplastics, and permits their uptake and catabolism by microorganisms through the use of auxiliary enzymes (i.e., transporters, regulatory elements, etc.), as well as synthesizing energy and intermediates necessary for the microorganism's metabolism [81]. Using CRISPR technology to both identify the various metabolic bottlenecks and genetically manipulate the various genes involved in those various metabolic pathways (e.g., over-expressing genes encoding for rate-limiting enzymes, deleting genes encoding for competing pathways), and systematically optimizing metabolic pathways, is

expected to result in increased efficiencies for converting polymers into energy, reduced accumulation of intermediates, and improved microbial fitness under biologically relevant environmental conditions [82].

6.2. Enhancement of Enzyme Expression and Secretion

The biodegradation rate of plastics is primarily determined by the quantity of plastic-degrading enzymes found in the environment. By using CRISPR-based techniques, researchers have been able to manipulate the transcriptional and translational machinery of a given organism to allow it to produce and secrete plastic-degrading enzymes in much greater quantities than would occur naturally. The controlled overexpression of plastic-degrading enzymes (e.g., PETase, MHETase, cutinases, laccases) can be achieved without introducing an excessive metabolic burden through the use of appropriate promoter design, gene dosage variation, or transcription factor modulation [7]. Engineering the pathways that enable the secretion of proteins outside the cell (e.g., Sec, Tat) as well as co-expressing chaperones with the plastic-degrading enzymes, can help ensure that the enzymes properly fold and are exported efficiently to the environment, thereby maximizing the exposure of the insoluble polymer surface to the enzyme [83]. Enhancing the overall stability of the enzymes through post-translational modification (e.g., glycosylation) and/or optimizing the signal peptide sequence for heterologous expression can increase enzyme thermostability, resistance to solvents, and resistance to proteolytic degradation. Finally, the incorporation of dynamic expression systems that enable enzyme production based on the polymer presence and/or environmental factors can optimize enzyme production and resource utilization through the use of CRISPR-based techniques [84].

6.3. Biosafety and Ethical Considerations of CRISPR-Engineered Strains

Microbial engineering by CRISPR provides opportunities to transform how we remove microplastics from the environment; however, there are numerous biosafety and ethical issues associated with the engineering of microbes. First, before being released into the environment, these engineered microorganisms need to be evaluated for their ability to persist in the environment, transfer their engineered genes to other organisms and cause unintentional ecological problems. To minimize any potential impact on the environment, there are several containment strategies that can be employed, including using inducible kill switches and creating auxotrophs that can only grow if provided with certain essential nutrients [85]. There are many synthetic toxin–antitoxin kill switch systems that have been developed as a means to control the survival of engineered microorganisms outside of a controlled environment by inducing programmed cell death if specific environmental conditions are met. The use of self-targeting CRISPR circuits and auxotrophic dependence as biocontainment strategies have also been proposed to prevent the uncontrolled spilling into the environment of genetically modified microbial strains and to prevent horizontal gene transfer of those strains [86]. These biosafety methods can enhance the ecological safety and regulatory acceptance of genetically engineered microorganisms used in bioremediation on a large scale in the environment. Additionally, through the use of bioreactors, microbes can be deployed in bioremediation without direct contact with the environment [87]. Regulatory frameworks, such as the Cartagena Protocol on Biosafety, national regulations for genetically modified organisms (GMOs) and Environmental Risk Assessment Guidelines (ERAs), exist to regulate the release of engineered microbes in a responsible manner. Ethical considerations also include issues of transparency surrounding genetic engineering, equitable access to bioremediation technologies, and public perception of the use of genetically modified organisms in environmental applications [88]. Therefore, the integration of CRISPR design along with a thorough biosafety and ecological risk

assessment process, as well as ethical governance, will result in the creation of sustainable, safe and socially acceptable microbial platforms for the large-scale bioremediation of microplastics [89].

7. Synthetic Biology Approaches in Microplastic Bioremediation

Microplastics present a major dilemma for existing bioremediation methods because of their long-lasting effects on the environment and their chemical composition being made up of many different polymers. Synthetic biology can be utilized as a means of developing microorganisms to have improved abilities to degrade plastics. Through synthetic biology, microorganisms can be designed and constructed in an intentional way (i.e., through modularizing gene constructs), and then assembled into systems to degrade different types of microplastics through different types of environmental conditions (e.g., aerobically) [90]. By combining all of these different approaches, synthetic biology will create specific strains or consortia types that will allow microorganisms to have coordinated hydrolytic, aerobic, and transport functions, thereby increasing the efficacy, stability, and adaptability of bioremediation processes for microplastics [91].

7.1. Construction of Synthetic Degradation Pathways

The field of synthetic biology enables the design of new polymer degradation pathways and the reconstruction of these pathways within microbial hosts. Synthetic operons allow for the use of hydrolytic enzymes such as polyethylene terephthalate hydrolases (PETases), laccases, and peroxidases, along with oxidative enzymes such as monooxygenases, cutinase, and lipases, to create a series of sequential oxidation and hydrolysis reactions, which enhance the rate of degradation of plastic. Pathway modularity facilitates the optimization of enzyme ratios, the direction of substrate to enzyme, and the regeneration of co-factors to improve the kinetics of polymer degradation [92]. For example, engineered pathways for the degradation of PET use extracellular PETases coupled with intracellular MHETases (multi-hydroxyethyl terephthalate hydrolase) and lower-level catabolic enzymes to convert mono-converted products, such as TPA and ethylene glycol (EG), to central metabolites [93]. Genome-scale metabolic modeling and flux balance analyses provide the means to identify rate-limiting steps in pathways, which then guide the rational insertion, deletion or overexpression of genes to maximize carbon flux toward either microbial growth (and ultimately biofuel production) or bioenergy. Additionally, engineered pathways can be optimized further using inducible promoters or environmentally responsive switches for the context-dependent activation of these pathways and for the reduction in metabolic burden [94].

7.2. Modular Genetic Circuits for Polymer Catabolism

The modular makeup of genetic circuits creates a potentially adaptable system through which various aspects of enzyme expression, substrate sensing, and thus metabolic control over these enzymes can be coordinated based upon the introduction of a microplastic input. The synthetic regulatory element of the microbial chassis has enabled the delivery of several synthetic regulatory elements to control degradation pathways temporally and spatially through a number of new approaches [95]. Examples include using polymer-specific promoters to regulate the expression of the PETase and laccase only in the presence of the substrate(s) being broken down, allowing microbes to conserve cellular resources and improve the effectiveness of their enzymatic degradation of plastics. Using logic-based genetic circuits allows for efficient decision-making by microbes, enabling them to utilize various polymer degradation pathways dependent upon the polymer(s) being broken down [96]. Logic-based circuits will also allow for a proper balance between catabolic

activity and cellular homeostasis. The use of feedback loops and signal amplification methods will further enhance the reliability of these circuits, making them capable of functioning under variable environmental conditions. The modular nature of these circuits permits rapid adaptation and customization to allow for multiple polymer degradation and the utilization of microbial systems in environmental variations [97].

8. Omics-Driven Insights into Microplastic Biodegradation

Microplastics have a broad range of properties that makes them very difficult to break down (heterogeneous, resistant), and therefore having a complete understanding of the microbial population associated with these materials will help scientists develop the necessary strategies to effectively degrade them [98]. Omics technologies (metagenomics, transcriptomics, proteomics and metabolomics) provide researchers unprecedented abilities to understand the genetic, regulatory and metabolic basis behind the breakdown of these synthetic polymers, as well as allowing for identification of novel enzymes involved in plastic degradation, as shown in Figure 4. This information allows strain engineering and the development of new synthetic biology applications that can be used to assist long-term recovery of microplastics from the plant ecosystem [99].

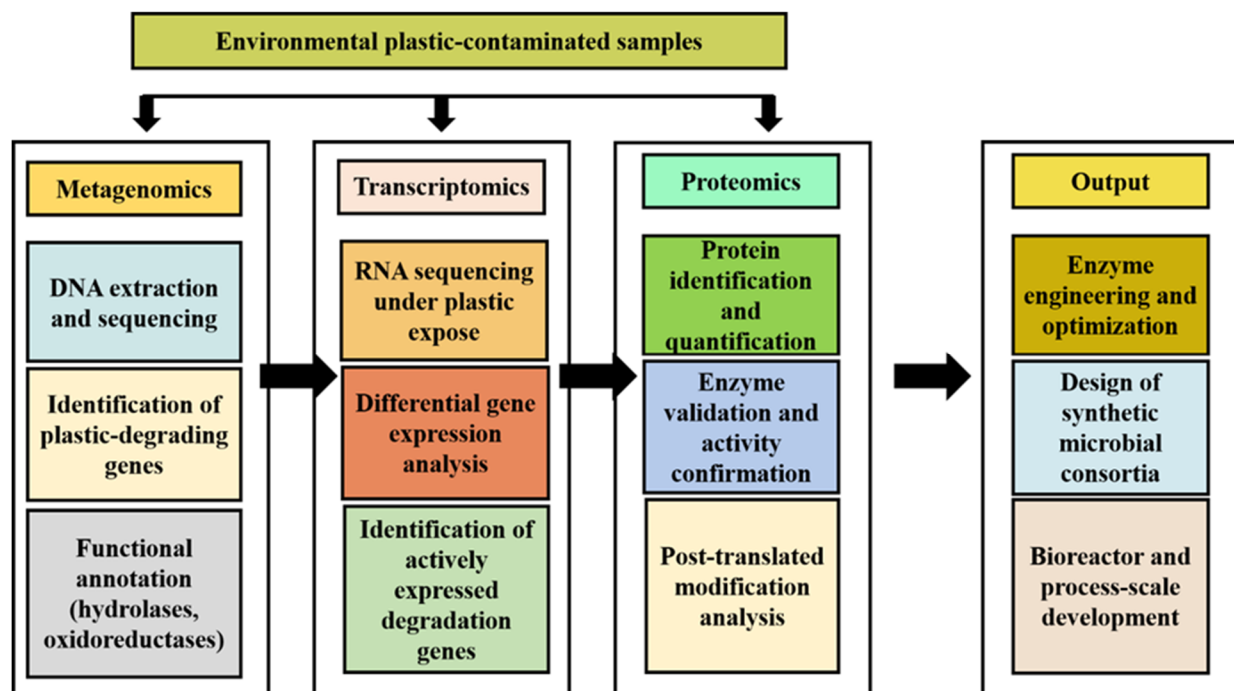


Figure 4. Integrated multi-omics pipeline for discovery and optimization of plastic-degrading systems.

8.1. Metagenomics for Discovery of Novel Plastic-Degrading Genes

Metagenomic analysis of the microbial community structure of microplastics-contaminated environments does not require the cultivation of the individual organismal members of the community and enables the identification and characterization of both commonly identified microorganisms and previously unobserved rarities that have adapted their metabolic capabilities specifically for utilizing this synthetic polymer, as well as developing biofilms that produce localized concentrates of enzymatic activity on the surfaces of microplastics [100]. Environmental DNA from the associated environments is sequenced using high-throughput shotgun sequencing methods to identify a combination of both previously identified hydrolytic enzymes as well as new, unidentified oxidoreductases such as laccases, peroxidases, and flavin-dependent monooxygenases, and to correlate specific DNA sequences of the identified/conceivably identified hydrolase enzymes to

their ability to degrade certain specific polycondensate polymers or oligomeric precursors through the application of functional metagenomics via the cloning and expression of environmental DNA fragments in heterologous microbe systems [101]. Functional metagenomics has facilitated not only the identification and characterization of previously known anaerobic enzymes, but also the identification and characterization of previously unidentified anaerobic enzymes. In addition, during comparative metagenomic studies, additional informative details will be gleaned regarding the gene clusters responsible for encoding catabolic enzymes that are organized in clusters according to their region and species, and the co-regulation of the catabolic gene cluster's uptake and assimilation of oligomers is demonstrated [102]. Horizontal genetic transfer events between microbial species provide evidence of the response of an evolving microbial community to anthropogenic microplastic pollution. The results from the metagenomic studies described will expand the potential enzymatic capabilities used for the bioremediation of microplastics, and additional product variations of the genes discovered via the metagenomic studies will create genetic "modules" to be utilized in developing engineered strains or consortia of microbes with enhanced enzymatic hydrolytic activities, greater substrate versatility, and superior capabilities under environmental stress conditions [103].

8.2. Transcriptomics and Proteomics for Pathway Elucidation

Although metagenomic data provide insight into the genetic potential of microbial communities exposed to microplastics, transcriptomic and proteomic data provide concrete information about gene expression patterns and protein production during degradation processes. Transcriptomic studies, which utilize Next Generation Sequencing (NGS) platforms such as RNA-seq (RNA sequencing), have provided evidence that upregulation of specific genes involved in the degradation of plastics coincides with increased expression of genes that code for extracellular depolymerizing enzymes, transport mechanisms for oligomeric and monomeric substrates, and central metabolic processes/energy pathways [104]. These investigations have also revealed several mechanisms whereby microorganisms counteract the stress associated with colonizing recalcitrant polymeric material, including oxidative stress, membrane remodeling, and biofilm production, thereby demonstrating that adaptation is essential for microbial survival on these substrates. Proteomic investigations through both shotgun and secretome methods corroborate transcriptomic findings by confirming production and post-transcriptional regulation of the same enzymes. Profiling studies have indicated a disconnect between transcriptionally induced enzymatic activity and secretion, emphasizing that control of stability and turnover rates at the protein level is crucial for substrate degradation [105]. Furthermore, detecting multi-enzyme complexes and accessory factors, such as binding domains and redox partners, has assisted in further refining existing models of polymer degradation and assimilation. The combination of transcriptomic and proteomic methodologies allows for the construction of a complete degradation pathway of the polymers and reconciles gene presence with the functional expression of the genes of interest [106].

8.3. Metabolomics for Tracking Degradation Intermediates

Metabolomics serves to validate the functional processes that occur during the bioremediation of microplastics by allowing the use of mass spectrometry and other metabolomic technologies, both targeted and untargeted, to identify and quantify the intermediate products produced through the degradation process, as well as the end products of the biodegradation process. Metabolomics currently provides access to an extensive array of chemical structures produced during the degradation of various types of polymers through means of polymer cleavage (e.g., oligomers, monomers, and subsequent metabolites),

including but not limited to terephthalic acid, ethylene glycol, and adipic acid and their derivative aliphatics, as well as fatty acid derivatives [107]. Temporal metabolomic profiling has been utilized to define the kinetics of degradation using temporal metabolomic profiles to define and understand the degree of degradation of each type of polymer and aid in understanding where bottlenecks occur during degradation. While confirming the breakdown of plastics into their respective building blocks, metabolomics has also provided insight into the degree to which the assimilated plastic-derived carbon contributes to the growth of microbes and to the production of microbial metabolites, such as fatty acid derivatives [108]. Currently, the extent of carbon assimilation from plastic has been evaluated using stable isotope-labeling techniques to establish a clear causal link between the carbon released during the depolymerization of plastics and the metabolism by microbes of the assimilated plastic-derived carbon via both central and peripheral pathways. When the metabolomic data are integrated with genomic, transcriptomic, and proteomic data, it provides the complete framework to allow for the development of predictive models and rationally designed microbial production platforms designed to efficiently facilitate the bioremediation of microplastics [109].

9. Enzyme Engineering and Nanobiocatalysis

While enzymes that degrade plastics occur naturally, the capacity of these enzymes to catalyze the degradation of different types of plastics (i.e., demonstrate proof-of-concept activities) is limited when applying these enzymes under conditions experienced in the environment and industry. The usefulness of these enzymes for the remediation of microplastics is hindered due to their low turnover rates, narrow substrate specificity, limited thermal and pH stability, and susceptibility to denaturation [110]. Therefore, the development of enzyme engineering and nanobiocatalysis technologies has enabled significant advancement towards eliminating these barriers to their effective use by improving catalytic efficiencies, stabilizing the enzymes, and increasing their reusability. By combining methods of protein engineering with the principles of nanoscale material science, the next generation of biocatalytic systems will be designed that will allow for the translation of biocatalytic processes from the laboratory to actual commercial processing environments [111].

9.1. Protein Engineering for Improved Catalytic Efficiency

The advancement of the catalytic performance of plastic-degrading enzymes is due to the application of protein engineering, which is the modification of protein structure and function. The use of rational design strategies that utilize high-resolution structural data and molecular dynamics simulations has facilitated the different modifications to active site residues, substrate binding sites, and access tunnels in order to enhance enzyme and polymer interaction. The modifications made using rational design techniques have resulted in an improved affinity of the enzyme for the substrate, a better fit of the polymer chain within the enzyme, and a faster rate of catalytic turnover, particularly with regard to high-crystallinity and hydrophobic plastics [112]. At the same time, directed evolution strategies have been successful in producing variant enzymes that have greater activity towards plastics under harsh environmental conditions. The use of multiple cycles of random mutagenesis and high-throughput screening produces variants that are more thermally stable, tolerant to a wider range of pH, and have greater resistance to many of the inhibitors found in contaminated environments [113]. It is critical to note that protein engineering shows that the increase in catalytic efficiency is due to the cumulative effects of distal mutations on protein flexibility and dynamic conformational states rather than simply altering the active site (catalytic center). Thus, it is necessary to consider the dynamics of enzymes and long-distance interactions when rationally designing new plastic-degrading

bio-catalysts. The amount and strength of this identification of catalytic efficiency may vary from study to study depending on the studies' approaches and methodologies used [114].

9.2. Nanobiocatalysts for Enhanced Enzyme Stability

Nanobiocatalysis is a new approach to improving the performance and stability of enzymes by taking advantage of the specific physicochemical characteristics of various nanomaterials. There are several types of enzyme/nanomaterial conjugates, which include metal nanoparticles, metal oxide nanoparticles, carbon nanostructures, and biopolymeric nanocarriers. These have resulted in the enhancement of enzyme activity retention, enzyme stability and resistance to environmental stressors. High surface-to-volume ratios at the nanometer scale allow for improved enzyme loading without compromising enzyme accessibility, thus increasing effective local enzyme concentration [115]. Additionally, it is thought that nanomaterials exhibit the capacity to modulate enzyme behavior through good nano-bio interactions, thereby providing active stabilization of the selected enzyme. Surface functionalization of the nanoparticles permits the controlled orientation of the enzyme on the nanoparticle surface and reduces the conformational entropy loss, as well as minimizing the aggregation that would otherwise lead to the loss of enzyme activity [116]. In addition, the use of nanomaterials in certain instances has resulted in increased rates of electron transfer, localized enhancement of substrate concentrations, and the establishment of microenvironments for catalysis. The utilization of nanobiocatalysis would be beneficial for accelerating the degradation of microplastics due to increased accessibility of enzymes to insoluble and heterogeneous polymer surfaces. Therefore, nanobiocatalysis serves as a stable platform for long-term biocatalytic activity in environmentally dynamic and contaminated conditions [37].

9.3. Enzyme Immobilization Strategies for Reusability

The strategy of enzyme immobilization has become increasingly important for enhancing the operational stability, re-usability and economic viability of enzyme systems used to degrade microplastics. By adopting various enzyme immobilization techniques (i.e., through adsorption, covalent bonding, entrapment and cross-linking), enzymes can be retained in a well-defined matrix, while still retaining catalytic function. Enzymes that have been immobilized are generally more resistant to thermal denaturation, proteolytic degradation and shear stress, and therefore are better suited to longer-term and continuous flow remediation operations [117]. The development of new immobilization materials has increased the potential for utilizing these techniques. Biopolymer-based support materials, hybrid organic-inorganic matrices, and nanostructured scaffolds possess tunable physicochemical properties, making it possible to optimize the performance of the enzyme by maximizing the enzyme load, mass transfer and substrate diffusion. In addition, enzyme immobilization has been shown to facilitate the recovery and re-use of enzymes after numerous catalytic cycles, with very little loss in enzyme activity, thus lowering the operational cost dramatically. When combined with protein engineering and nanobiocatalyst design, the immobilization strategy has laid the foundation for building integrated, scalable enzyme systems, which can achieve sustained, efficient biodegradation of microplastics [75].

10. Bioreactor Technologies and Process Scale-Up

To implement the success of biotechnology developed at a laboratory scale to fully commercial microplastic biodegradation operations, a reliable bioreactor system must be developed that allows for the sustainable growth of microbes or enzymes at a controlled and scalable level. The use of microplastic subunits creates a distinct range of biochemical

engineering challenges as these materials do not behave as soluble substrates, resulting in the need to use multiple approaches in the bioreactor design process [118]. Thus, it would be necessary to integrate biochemical engineering principles, materials science technologies, as well as microbial ecologies to create a bioreactor system designed for optimal substrate biocatalyst interaction, stability, and reliability in terms of degradation performance. Recent advances in bioreactor design, bioprocessing optimization technologies, and scale-up modalities have recently begun to move experimental microplastic biodegradation technologies into the commercial marketplace [119].

10.1. Bioreactor Designs for Microplastic Biodegradation

Microplastic biodegradation efficiency and consistency are primarily determined by the design of the bioreactor. Conventional stirred-tank bioreactors are commonly used in laboratory settings because they provide easy operation and accurate control of key physicochemical parameters (temperature/pH/dissolved oxygen). The poor wettability and buoyancy of microplastics prevent adequate mixing and microbial and substrate interaction in conventional stirred-tank bioreactor systems, leading to poor performance. Therefore, to mitigate these issues, alternative reactor designs, such as airlift reactors, packed-bed reactors, and fluidized-bed bioreactors, have been developed to improve solid and liquid contact and increase the biofilm-mediated biodegradation of microplastics [120]. Biofilm reactors are among the most promising alternatives, as surface-attached microbial communities have superior resilience to environmental variations and increased access to the surface of polymer material. Rotating biological contactors and membrane-supported biofilm reactors allow for continuous contact between microplastics and living enzymes while minimizing biomass washout. In addition, in enzymatic systems, reactors utilizing immobilized enzymes or nanobiocatalysis afford precise control of the time a microplastic substrate remains in the reactor and the product formed per enzyme. In summary, the described reactor types illustrate the significance of tailoring hydrodynamic conditions and reactor configurations to the physicochemical properties of microplastics and their associated biological systems [121].

10.2. Process Optimization and Operational Challenges

To achieve the highest degradation efficacy while achieving the greatest operational stability when using a reactor over time, the optimization of processing of microplastics to be degraded is critical. The physical characteristics of the microplastic, chemical properties, and biological activity present significant variability that will influence the degradation rates of microplastics. Microplastic degradation will be influenced by a number of parameters such as particle size, polymer crystallinity, microbial and/or enzymatic loading, availability of nutrients, the rate of O₂ transfer and hydraulic retention time [122]. When optimizing processing, it is important to balance the conditions required to promote optimal microbial growth and the kinetics of degradation as excessive biomass build-up in the reactor can result in an adverse effect on the mass transfer rate and overall reactor performance. There remain many operational issues that adversely affect the large-scale application of the biological degradation of microplastics to recycled plastics. The variability of composition in the feedstock of microplastics affects the degradation rates and makes it challenging to maintain stable processing conditions [123]. Other factors include biofouling, enzyme inactivation or competing products (inhibitors) that can build up and affect the long-term viability of reactor operation. Automation through the use of advanced monitoring and control systems (online sensors, automatic feedback controls and data-driven models of the degradation process) are becoming more widely used to resolve these issues. Innovative

use of omics will complement monitoring and control systems to allow for the dynamic adjustment of processing conditions to enhance the efficiency of biocatalytic actions [124].

10.3. Industrial Feasibility and Scalability

The industrial practicality of using microorganism-related methods to bioremediate pollution caused by microparticles can also involve looking at how are they financially achievable and whether they can be built at scale or not, but most importantly if there are ways in which current waste/landfill facilities can be used. Enzyme engineering innovation has greatly increased the capacity for microplastics to be biodegraded. The leaf-branch compost cutinase (LCC) enzyme variant developed was able to achieve about a 90% PET depolymerization in 10 h at 72 °C under the best available conditions [125]. The engineered PETase variants that have been produced from *Ideonella sakaiensis* have been found to exhibit much higher rates of PET degradation than the wild-type PETase produced by the same bacterium [67]. The results of these studies provide support for the continued advancement of engineered microbes and enzymes as a cost-effective and viable method for sustainably and systematically remediating microplastics on a large scale. In addition to laboratory-scale studies, microbial and enzyme-mediated bioremediation systems are being developed for numerous practical uses, such as in municipal waste water treatment facilities (e.g., sewage treatment plants), marine remediation, and soil with plastic contamination. Microbial biofilm-based consortia and bioreactor-assisted degradation (biodegradation) systems can facilitate continuous microplastic removal under controlled environmental conditions. However, problems such as heterogeneous waste types, volatility of environmental conditions, and scalability of operations hinder broad scale adoption across industries [126]. When research moves beyond the lab scale to pilot or industrial scales, this generates new challenges regarding mixing/alloying of ingredients, obtaining oxygen(s) in the material, and being able to keep the biological catalyst active. There have been multiple types of engineering that have been suggested to help mitigate the risks associated with scaling up and providing additional operational flexibility with systems that could have potentially been lost at the pilot levels [127]. The studies compare how much it costs to produce the biocatalyst and how much it costs to operate the reactor, and finally evaluate what the operational cost might be for the downstream processes relative to the environmental benefits that are achieved through bioremediation of microparticles. The coupling of biodegradation technologies with liking (bio-contained) waste valorization techniques, such as the recovery of biopolymer or monomer or some other energy-rich metabolite, can positively impact the economy of the process and increase the likelihood of sustainable practices being put in place [128]. This can assist with identifying suitable areas for investment and determining how to implement these methods into regulations within the industry, carrying out life cycle assessments and conducting cost-benefit analysis. Ultimately, as bioreactor engineering progresses, so too will be the ability to transfer biodegradation technology from experimental to commercial scales, and researchers will be able to determine what aspects, if any, from the regulatory and economic sides need to be addressed before implementing microplastics mitigation strategies [28].

11. Current Challenges in Microplastics Biodegradation

Many technological and scientific obstacles still prevent the large-scale biodegradation of microplastics through enzymes and microbes, even though considerable advances have been made in this area. One of the key impediments to large-scale biodegradation of microplastics is the difficulty associated with the biodegradation of PE, PP and PS, which contain very stable carbon-carbon (C-C) backbones that resist enzymatic attack or

degradation [129]. Other properties of PE, PP and PS, such as a high degree of crystallinity, demonstrated hydrophobicity, and limited surface access to microbes, have an additional inhibitory effect on microbial colonization and on microbial and enzymatic depolymerization efficiency in the natural environment. Environmental parameters such as temperature, salinity, pH, and the availability of oxygen and the presence of other contaminating materials have significant effects on microbial activity and degradation of plastics in natural environments [62]. A second set of important obstacles to large-scale microbial biodegradation of plastics includes the low catalytic activity, specificity and thermal stability of most naturally occurring plastic-degrading enzymes. While some engineered forms of naturally occurring enzymes such as PETase have been developed, and exhibit a very high degree of biodegradation of plastics under laboratory conditions, long-term retention of enzyme activity and operational stability during industrial-scale applications are difficult to maintain. Additional factors that complicate large-scale biodegradation of plastic waste include the fact that the waste stream is heterogeneous; in addition to the presence of the various plastics in the waste stream, there are also many additives including dyes and plasticizers, and different types of polymeric materials, which create difficulties in biodegradation of plastics and generate toxic intermediate products [125]. The large-scale use of engineered microbial systems for biodegradation of plastic waste also raises biosafety and regulatory concerns related to potential horizontal gene transfer, ecological imbalance, and unintentional persistence of genetically modified microorganisms in the environment. There are significant obstacles to maintaining stable microbial communities in dynamic or changing environments because of inter-species competition for nutrients, nutrient constraints and other factors that may stress the microbes. Thus, further advancement in the fields of synthetic biology, enzyme engineering, bioreactor design and optimization, and integrated pretreatment of waste will be needed to make microbial bioremediation of microplastics an efficient, safe and commercially viable system for waste treatment [130].

12. Future Prospects

Future research on the bioremediation of microplastics will focus on creating effective microbial and enzyme-based solutions to reduce plastic waste through natural degradation and recovering valuable resources from these degraded microplastics. The use of advancements in integrating multi-omics technology, engineered CRISPR-based systems, manufacturer-produced microbial groups, and engineered enzymes should lead to improvements in plastic-degrading microbe/enzyme catalytic efficiency, stability and specificity. Harnessing advances in nanobiotechnology, artificial intelligence and smart bioprocessing systems will also increase large-scale biodegradation and optimization of biodegradation processes. More and more attention and focus are being directed towards using the waste from plastic materials for valorization, namely converting degraded microplastics into alternative useable products such as biomass, bioenergy and biochemicals for industrial use. As seen in Figure 5, microbes and enzymes can be used to degrade or decompose plastics into smaller molecular sizes, from which point they can be processed into alternative useable products according to the principles and concepts of the circular bioeconomy.

Future applications will require improved bioreactor technologies, biosafety testing and standards, effective regulatory frameworks, and collaborative interdisciplinary projects to successfully establish economically and environmentally sustainable remediation processes.

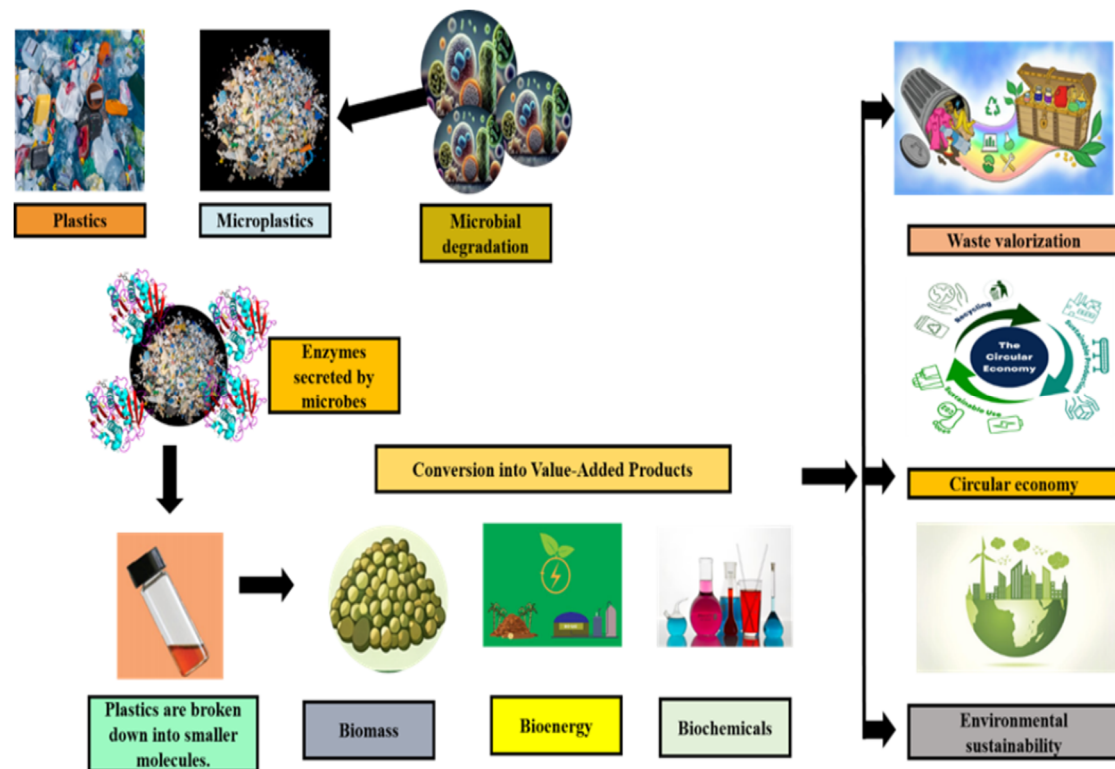


Figure 5. An integrated approach to bioremediation of microplastic waste through microbial degradation and the conversion of degraded microplastics into value-added products. Extracellular enzymes secreted by microbial communities during biodegradation of plastic waste breaks down microplastics into smaller molecules. These smaller molecules can then be converted to value-added products including biomass, bioenergy and /or biochemicals. The integrated approach to bioremediation of microplastic waste also supports waste valorization, circular bioeconomy strategies and long-term sustainability for the environment.

13. Conclusions

Microplastics now present a serious threat to the environment due to their ability to persist in the environment for long periods of time, to be widely distributed over large areas (i.e., have a global scale of impact), and to resist degradation by natural biological processes because of their synthetic polymer nature. This study reviews the use of microplastic degradation processes employing bacterial and enzymatic biodegradation pathways as viable, environment-friendly alternatives to traditional methods of bioremediating microplastics in the environment. Advances within scientific and technical fields have resulted in increased awareness regarding microbial degradation of plastic materials. Microbial ecology, new enzyme discovery, and use of genome-editing tools (e.g., CRISPR) are the areas that have contributed to the advancements in biodegradation processes. Additionally, the emergence of new omics technologies, nanobiocatalysis, and bioreactor design has assisted researchers in making useful discoveries related to microbial degradation of plastics.

Recent advances in bioprocess optimization and scaling bioreactor technologies indicate that biological treatment systems are now developing through laboratory trials to actual use in the environment. Despite these advances, a number of scientific and technical challenges remain, such as polymer recalcitrance and environmental variability, incomplete degradation of microplastics, system instability, and biosafety concerns with engineered microbial systems, that still prevent the large-scale implementation of bioremediation strategies. In addition, there remains a need for continued interdisciplinary research among microbiology, synthetic biology, materials science and process engineering to support the development of safe and commercially viable biodegradable materials.

Overall, microbial-based biotechnology is among the most promising platforms available for achieving sustainable management of microplastics and for providing long-term environmental protection.

Author Contributions: B.T. (writing—original draft, and revising—revised draft and prepared figures); S.K. (writing—original draft, revising—revised draft, supervision and validation); M.T. (Project administration, revising—revised draft, supervision, validation); P.S. (Project administration, revising—revised draft, supervision, validation). All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: No new data were created or analyzed in this study. Data sharing is not applicable to this article.

Conflicts of Interest: The authors declare no conflicts of interest.

References

1. Ashrafy, A.; Liza, A.A.; Islam, M.N.; Billah, M.M.; Arafat, S.T.; Rahman, M.M.; Rahman, S.M. Microplastics pollution: A brief review of its source and abundance in different aquatic ecosystems. *J. Hazard. Mater. Adv.* **2023**, *9*, 100215. [\[CrossRef\]](#)
2. Elawady, A.; Mohammed, J.A.; Abid, M.B. Advancements in Microplastics Detection Techniques and Their Multidimensional Impacts on Aquatic Ecosystems and Human Health. *J. Hazard. Mater. Plast.* **2025**, *2*, 100025.
3. Lebepe, J.; Buthelezi, N.M.D.; Manganyi, M.C. Occurrence and Control of Microplastics and Emerging Technological Solutions for Their Removal in Freshwaters: A Comprehensive Review. *Microplastics* **2025**, *4*, 70. [\[CrossRef\]](#)
4. Kumar, M.; Chaudhary, V.; Kumar, R.; Srivastav, A.L. Microplastics, their effects on ecosystems, and general strategies for mitigation of microplastics: A review of recent developments, challenges, and future prospects. *Environ. Pollut. Manag.* **2025**, *2*, 87–105. [\[CrossRef\]](#)
5. Jiao, H.; Xiong, M.; Al-Tohamy, R.; Schagerl, M.; Kornaros, M.; Ali, S.S. Rethinking plastics through microbial biodegradation and circular economy innovation—A review. *Environ. Chem. Ecotoxicol.* **2026**, *8*, 195–224. [\[CrossRef\]](#)
6. Lokesh, P.; Shobika, R.; Omer, S.; Reddy, M.; Saravanan, P.; Rajeshkannan, R.; Saravanan, V.; Venkatkumar, S. Bioremediation of plastics by the help of microbial tool: A way for control of plastic pollution. *Sustain. Chem. Environ.* **2023**, *3*, 100027. [\[CrossRef\]](#)
7. Palit, P.; Minkara, M.; Abida, M.; Marwa, S.; Sen, C.; Roy, A.; Pasha, M.R.; Mosae, P.S.; Saha, A.; Ferdoush, J. PlastiCRISPR: Genome Editing-Based Plastic Waste Management with Implications in Polyethylene Terephthalate (PET) Degradation. *Biomolecules* **2025**, *15*, 684. [\[CrossRef\]](#) [\[PubMed\]](#)
8. Viljakainen, V.R.; Hug, L.A. New approaches for the characterization of plastic-associated microbial communities and the discovery of plastic-degrading microorganisms and enzymes. *Comput. Struct. Biotechnol. J.* **2021**, *19*, 6191–6200. [\[CrossRef\]](#) [\[PubMed\]](#)
9. Narancic, T.; O'Connor, K.E. Microbial biotechnology addressing the plastic waste disaster. *Microb. Biotechnol.* **2017**, *10*, 1232. [\[CrossRef\]](#) [\[PubMed\]](#)
10. Kamalasekaran, K.; Sundramoorthy, A.K. A review on microplastics: Sources, environmental fate, degradation pathways, and analytical identification methods. *RSC Adv.* **2026**, *16*, 13851–13874. [\[CrossRef\]](#) [\[PubMed\]](#)
11. Cai, Z.; Li, M.; Zhu, Z.; Wang, X.; Huang, Y.; Li, T.; Gong, H.; Yan, M. Biological degradation of plastics and microplastics: A recent perspective on associated mechanisms and influencing factors. *Microorganisms* **2023**, *11*, 1661. [\[CrossRef\]](#) [\[PubMed\]](#)
12. Lalrinfela, P.; Vanlalsangi, R.; Lalrinzuali, K.; Babu, P.J. Microplastics: Their effects on the environment, human health, and plant ecosystems. *Environ. Pollut. Manag.* **2024**, *1*, 248–259. [\[CrossRef\]](#)
13. Muñiz, R.; Rahman, M.S. Microplastics in coastal and marine environments: A critical issue of plastic pollution on marine organisms, seafood contaminations, and human health implications. *J. Hazard. Mater. Adv.* **2025**, *18*, 100663. [\[CrossRef\]](#)
14. Ziani, K.; Ioniță-Mîndrican, C.B.; Mititelu, M.; Neacșu, S.M.; Negrei, C.; Moroșan, E.; Drăgănescu, D.; Preda, O.T. Microplastics: A real global threat for environment and food safety: A state of the art review. *Nutrients* **2023**, *15*, 617. [\[CrossRef\]](#) [\[PubMed\]](#)
15. Sutkar, P.R.; Gadewar, R.D.; Dhulap, V.P. Recent trends in degradation of microplastics in the environment: A state-of-the-art review. *J. Hazard. Mater. Adv.* **2023**, *11*, 100343. [\[CrossRef\]](#)
16. Hossain, M.; Engelhardt, I. Global plastic footprint: Unveiling property trends, environmental fate, and emerging threats of microplastic and nanoplastics pollution across ecosystems. *Energy Ecol. Environ.* **2025**, *10*, 637–674. [\[CrossRef\]](#)

17. Ali, S.S.; Alsharbaty, M.H.M.; Al-Tohamy, R.; Schagerl, M.; Al-Zahrani, M.; Kornaros, M.; Sun, J. Microplastics as persistent and vectors of other threats in the marine environment: Toxicological impacts, management and strategical roadmap to end plastic pollution. *Environ. Chem. Ecotoxicol.* **2025**, *7*, 229–251. [[CrossRef](#)]
18. Turkey, A.; Upadhyay, L.S.B. Microplastics: An overview on separation, identification and characterization of micro-plastics. *Mar. Pollut. Bull.* **2021**, *170*, 112604. [[CrossRef](#)] [[PubMed](#)]
19. Yang, H.; Chen, Z.; Kong, L.; Xing, H.; Yang, Q.; Wu, J. A Review of Eco-Corona Formation on Micro/Nanoplastics and Its Effects on Stability, Bioavailability, and Toxicity. *Water* **2025**, *17*, 1124. [[CrossRef](#)]
20. Rossatto, A.; Arlindo, M.Z.F.; de Moraes, M.S.; de Souza, T.D.; Ogradowski, C.S. Microplastics in aquatic systems: A review of occurrence, monitoring and potential environmental risks. *Environ. Adv.* **2023**, *13*, 100396. [[CrossRef](#)]
21. Kye, H.; Kim, J.; Ju, S.; Lee, J.; Lim, C.; Yoon, Y. Microplastics in water systems: A review of their impacts on the environment and their potential hazards. *Heliyon* **2023**, *9*, e14359. [[CrossRef](#)] [[PubMed](#)]
22. Zhang, Y.; Kang, S.; Allen, S.; Allen, D.; Gao, T.; Sillanpää, M. Atmospheric microplastics: A review on current status and perspectives. *Earth-Sci. Rev.* **2020**, *203*, 103118. [[CrossRef](#)]
23. Zhu, J.; Dong, G.; Feng, F.; Ye, J.; Liao, C.H.; Wu, C.H.; Chen, S.C. Microplastics in the soil environment: Focusing on the sources, its transformation and change in morphology. *Sci. Total Environ.* **2023**, *896*, 165291. [[CrossRef](#)] [[PubMed](#)]
24. Gupta, G.K.; Dixit, M.; Chot, E.; Shukla, P. Insights into Microbial Enzymatic Biodegradation of Plastics and Microplastics: Technological Updates. *ACS Environ. Au* **2025**, *5*, 520–542. [[CrossRef](#)] [[PubMed](#)]
25. Gewert, B.; Plassmann, M.M.; MacLeod, M. Pathways for degradation of plastic polymers floating in the marine environment. *Environ. Sci. Process. Impacts* **2015**, *17*, 1513–1521. [[CrossRef](#)] [[PubMed](#)]
26. Tokiwa, Y.; Calabia, B.P. Biodegradability and biodegradation of polyesters. *J. Polym. Environ.* **2007**, *15*, 259–267. [[CrossRef](#)]
27. Lumio, R.T.; Tan, M.A.; Magpantay, H.D. Biotechnology-based microbial degradation of plastic additives. *3 Biotech.* **2021**, *11*, 350. [[CrossRef](#)] [[PubMed](#)]
28. Yadav, P.; Kumar, A.; Ram, K.; Kumar, A.; Gupta, R.K.; Dufossé, L. Microbial degradation of microplastics: Effective-ness, challenges, and sustainable solutions. *Curr. Res. Microb. Sci.* **2025**, *9*, 100495. [[CrossRef](#)] [[PubMed](#)]
29. Parsaeimehr, A.; Miller, C.M.; Ozbay, G. Microplastics and their interactions with microbiota. *Heliyon* **2023**, *9*, e15104. [[CrossRef](#)] [[PubMed](#)]
30. Yang, W.; Li, Y.; Boraschi, D. Association between microorganisms and microplastics: How does it change the host—Pathogen interaction and subsequent immune response? *Int. J. Mol. Sci.* **2023**, *24*, 4065. [[CrossRef](#)] [[PubMed](#)]
31. Li, M.; Ma, W.; Fang, J.K.H.; Mo, J.; Li, L.; Pan, M.; Li, R.; Zeng, X.; Lai, K.P. A review on the combined toxicological effects of microplastics and their attached pollutants. *Emerg. Contam.* **2025**, *11*, 100486. [[CrossRef](#)]
32. Liu, J.; Zheng, L. Microplastic migration and transformation pathways and exposure health risks. *Environ. Pollut.* **2025**, *368*, 125700. [[CrossRef](#)] [[PubMed](#)]
33. Perchikov, R.; Cheliukanov, M.; Plekhanova, Y.; Tarasov, S.; Kharkova, A.; Butusov, D.; Arlyapov, V.; Nakamura, H.; Reshetilov, A. Microbial biofilms: Features of formation and potential for use in bioelectrochemical devices. *Biosensors* **2024**, *14*, 302. [[CrossRef](#)] [[PubMed](#)]
34. Rummel, C.D.; Jahnke, A.; Gorokhova, E.; Kühnel, D.; Schmitt-Jansen, M. Impacts of biofilm formation on the fate and potential effects of microplastic in the aquatic environment. *Environ. Sci. Technol. Lett.* **2017**, *4*, 258–267. [[CrossRef](#)]
35. Fortin, S.G.; Uhlig, K.; Hale, R.C.; Song, B. Microplastic biofilms as potential hotspots for plastic biodegradation and nitrogen cycling: A metagenomic perspective. *FEMS Microbiol. Ecol.* **2025**, *101*, fiae035. [[CrossRef](#)] [[PubMed](#)]
36. Sahith, V.N.; Kumar, A.; Sruthi, V.S.; Sathish, S.; Venkatesan, D.; Prabu, D.; Samrot, A. Microbial and enzymatic biodegradation of microplastics and nanoplastics: Advances, challenges, and sustainable solutions for Environmental Remediation. *Desalin. Water Treat.* **2025**, *324*, 101450. [[CrossRef](#)]
37. Tang, K.H.D.; Lock, S.S.M.; Yap, P.-S.; Cheah, K.W.; Chan, Y.H.; Yiin, C.L.; Ku, A.Z.E.; Loy, A.C.M.; Chin, B.L.F.; Chai, Y.H. Immobilized enzyme/microorganism complexes for degradation of microplastics: A review of recent advances, feasibility and future prospects. *Sci. Total Environ.* **2022**, *832*, 154868. [[CrossRef](#)] [[PubMed](#)]
38. Sridhar, S.; Murugesan, N.; Gopalakrishnan, M.; Janjoren, D.; Ganesan, S. Removal of microplastic for a sustainable strategy by microbial biodegradation. *Sustain. Chem. Environ.* **2024**, *6*, 100088. [[CrossRef](#)]
39. Zhou, L.; Ran, L.; He, Y.; Huang, Y. Mechanisms of microplastics on gastrointestinal injury and liver metabolism dis-order. *Mol. Med. Rep.* **2025**, *31*, 98. [[CrossRef](#)] [[PubMed](#)]
40. Rezaei, Z.; Dinani, A.S.; Moghimi, H. Cutting-edge developments in plastic biodegradation and upcycling via engineering approaches. *Metab. Eng. Commun.* **2024**, *19*, e00256. [[CrossRef](#)] [[PubMed](#)]
41. Wang, J.; Zhang, T.; Li, Y.; Ye, Y.; Tan, X.; Liu, X.; Hu, L.; Du, Z.; Ye, M. Microplastics and microbial interactions in marine environments: A critical review on biogeochemical cycling and ecological impacts. *Mar. Pollut. Bull.* **2026**, *222*, 118891. [[CrossRef](#)] [[PubMed](#)]
42. Zhai, X.; Zhang, X.-H.; Yu, M. Microbial colonization and degradation of marine microplastics in the plastisphere: A review. *Front. Microbiol.* **2023**, *14*, 1127308. [[CrossRef](#)] [[PubMed](#)]

43. Zampolli, J.; Mangiagalli, M.; Vezzini, D.; Lasagni, M.; Ami, D.; Natalello, A.; Arrigoni, F.; Bertini, L.; Lotti, M.; Di Gennaro, P. Oxidative degradation of polyethylene by two novel laccase-like multicopper oxidases from *Rhodococcus opacus* R7. *Environ. Technol. Innov.* **2023**, *32*, 103273. [[CrossRef](#)]
44. Schmidt, J.; Wei, R.; Oeser, T.; Dedavid e Silva, L.A.; Breite, D.; Schulze, A.; Zimmermann, W. Degradation of polyester polyurethane by bacterial polyester hydrolases. *Polymers* **2017**, *9*, 65. [[CrossRef](#)] [[PubMed](#)]
45. Kotova, I.B.; Taktarova, Y.V.; Tsavkelova, E.A.; Egorova, M.A.; Bubnov, I.A.; Malakhova, D.V.; Shirinkina, L.I.; Sokolova, T.G.; Bonch-Osmolovskaya, E.A. Microbial degradation of plastics and approaches to make it more efficient. *Microbiology* **2021**, *90*, 671–701. [[CrossRef](#)]
46. Heris, Y.S. Different aspects of bacterial polyethylene terephthalate biodegradation. *Bull. Natl. Res. Cent.* **2025**, *49*, 28. [[CrossRef](#)]
47. Hari, S. Review on effect of fungi on plastic degradation. *JRAR* **2019**, *6*, 261–265. [[CrossRef](#)]
48. Philippe, A.; Salaun, M.; Quemener, M.; Noël, C.; Tallec, K.; Lacroix, C.; Coton, E.; Burgaud, G. Colonization and biodegradation potential of fungal communities on immersed polystyrene vs. bio-degradable plastics: A time series study in a marina environment. *J. Fungi* **2024**, *10*, 428. [[CrossRef](#)]
49. Decorosi, F.; Exana, M.L.; Pini, F.; Adessi, A.; Messini, A.; Giovannetti, L.; Viti, C. The degradative capabilities of new *Amycolatopsis* isolates on polylactic acid. *Microorganisms* **2019**, *7*, 590. [[CrossRef](#)] [[PubMed](#)]
50. Uthra, K.T.; Chitra, V.; Damodharan, N.; Devadoss, A.; Kuehnelt, M.; Exposito, A.J.; Nagarajan, S.; Pitchaimuthu, S.; Pazhani, G.P. Microplastic emerging pollutants—impact on microbiological diversity, diarrhea, antibiotic resistance, and bioremediation. *Environ. Sci. Adv.* **2023**, *2*, 1469–1487. [[CrossRef](#)]
51. López, M.J.; Jurado, M.M.; López-González, J.A.; Estrella-González, M.J.; Martínez-Gallardo, M.R.; Toribio, A.; Suárez-Estrella, F. Development of plastic-degrading microbial consortia by induced selection in microcosms. *Front. Microbiol.* **2023**, *14*, 1143769. [[CrossRef](#)] [[PubMed](#)]
52. Luo, A.; Wang, F.; Sun, D.; Liu, X.; Xin, B. Formation, development, and cross-species interactions in biofilms. *Front. Microbiol.* **2022**, *12*, 757327. [[CrossRef](#)] [[PubMed](#)]
53. Shokunbi, A.O.; Ezech, O.M.; Fadipe, D.P.; Olanrewaju, A.S.; Badru, O.O.; Eze, C.N.; Iwuala, L.C. Microbial Consortia: Synergistic Effects on Plastic Degradation and Enzyme Production. *J. Med. Sci. Biol. Chem.* **2025**, *2*, 149–156. [[CrossRef](#)]
54. Getino, L.; Revilla-Gómez, J.A.; Ariza-Carmona, L.M.; Thijs, S.; Didierjean, C.; Chamizo-Ampudia, A. Biodegradation of petrochemical plastics by microorganisms: Toward sustainable solutions for plastic pollution. *Bacteria* **2025**, *4*, 44. [[CrossRef](#)]
55. Armes, A.C.; Schaefer, A.L.; Hochanadel, L.H.; Klingeman, D.M.; Carper, D.L.; Abraham, P.E.; York, L.M.; Carrell, A.A.; Doktycz, M.J.; Pelletier, D.A. Quorum sensing modulates microbial community structure through regulation of secondary metabolites. *mSphere* **2025**, *10*, e0105024. [[CrossRef](#)] [[PubMed](#)]
56. Deter, H.S.; Lu, T. Engineering microbial consortia with rationally designed cellular interactions. *Curr. Opin. Biotechnol.* **2022**, *76*, 102730. [[CrossRef](#)] [[PubMed](#)]
57. Mohanan, N.; Montazer, Z.; Sharma, P.K.; Levin, D.B. Microbial and enzymatic degradation of synthetic plastics. *Front. Microbiol.* **2020**, *11*, 580709. [[CrossRef](#)] [[PubMed](#)]
58. Ramli, A.N.M.; Hong, P.K.; Manas, N.H.A.; Azelee, N.I.W. An overview of enzyme technology used in food industry. In *Value-Addition in Food Products and Processing Through Enzyme Technology*; Elsevier: Amsterdam, The Netherlands, 2022; pp. 333–345.
59. Mandal, M.; Roy, A.; Popek, R.; Sarkar, A. Micro-and nano-plastic degradation by bacterial enzymes: A solution to ‘white pollution’. *Microbe* **2024**, *3*, 100072. [[CrossRef](#)]
60. Verschoor, J.-A.; Arentshorst, M.; Regensburg-Tuink, A.J.G.; Seekles, S.J.; Hondel, C.v.D.; de Winde, J.H.; Ram, A.F.J. Heterologous expression and characterization of synthetic polyester-degrading cutinases from *Fusarium* spp. in *Aspergillus niger*. *Microbiol. Spectr.* **2025**, *13*, e0217725. [[CrossRef](#)] [[PubMed](#)]
61. Raczyńska, A.; Góra, A.; André, I. An overview on polyurethane-degrading enzymes. *Biotechnol. Adv.* **2024**, *77*, 108439. [[CrossRef](#)] [[PubMed](#)]
62. Carr, C.M.; Clarke, D.J.; Dobson, A.D.W. Microbial polyethylene terephthalate hydrolases: Current and future perspectives. *Front. Microbiol.* **2020**, *11*, 571265. [[CrossRef](#)] [[PubMed](#)]
63. Rovalletti, A.; De Gioia, L.; Fantucci, P.; Greco, C.; Vertemara, J.; Zampella, G.; Arrigoni, F.; Bertini, L. Recent theoretical insights into the oxidative degradation of biopolymers and plastics by metalloenzymes. *Int. J. Mol. Sci.* **2023**, *24*, 6368. [[CrossRef](#)] [[PubMed](#)]
64. Schwab, S.T.; Baur, M.; Nelson, T.F.; Mecking, S. Synthesis and deconstruction of polyethylene-type materials. *Chem. Rev.* **2024**, *124*, 2327–2351. [[CrossRef](#)] [[PubMed](#)]
65. Porter, S.S.; Bantay, R.; Friel, C.A.; Garoutte, A.; Gdanetz, K.; Ibarreta, K.; Moore, B.M.; Shetty, P.; Siler, E.; Friesen, M.L. Beneficial microbes ameliorate abiotic and biotic sources of stress on plants. *Funct. Ecol.* **2020**, *34*, 2075–2086. [[CrossRef](#)]
66. VL, D.S.; Rajmohan, G.; Nagasubramanian, K.; Venkatachalam, P. Improving the binding affinity of plastic degrading cutinase with polyethylene terephthalate (PET) and polyurethane (PU); an in-silico study. *Heliyon* **2025**, *11*, e41640. [[CrossRef](#)]
67. Sevilla, M.E.; Garcia, M.D.; Perez-Castillo, Y.; Armijos-Jaramillo, V.; Casado, S.; Vizuete, K.; Debut, A.; Cerda-Mejía, L. Degradation of PET bottles by an engineered *Ideonella sakaiensis* PETase. *Polymers* **2023**, *15*, 1779. [[CrossRef](#)] [[PubMed](#)]

68. Garduño-Juárez, R.; Tovar-Anaya, D.O.; Perez-Aguilar, J.M.; Lozano-Aguirre Beltran, L.F.; Zubillaga, R.A.; Alvarez-Perez, M.A.; Villarreal-Ramirez, E. Molecular dynamic simulations for biopolymers with biomedical applications. *Polymers* **2024**, *16*, 1864. [\[CrossRef\]](#) [\[PubMed\]](#)
69. Peccati, F.; Alunno-Rufini, S.; Jiménez-Osés, G. Accurate prediction of enzyme thermostabilization with Rosetta using AlphaFold ensembles. *J. Chem. Inf. Model.* **2023**, *63*, 898–909. [\[CrossRef\]](#) [\[PubMed\]](#)
70. Kim, S.; Ga, S.; Bae, H.; Sluyter, R.; Konstantinov, K.; Shrestha, L.K.; Kim, Y.H.; Kim, J.H.; Ariga, K. Multidisciplinary approaches for enzyme biocatalysis in pharmaceuticals: Protein engineering, computational biology, and nanoarchitectonics. *EES Catal.* **2024**, *2*, 14–48. [\[CrossRef\]](#)
71. Witczak, A.; Przedpeńska, L.; Pokorska-Niewiada, K.; Cybulski, J. Microplastics as a threat to aquatic ecosystems and human health. *Toxics* **2024**, *12*, 571. [\[CrossRef\]](#) [\[PubMed\]](#)
72. Choi, J.; Kim, H.; Ahn, Y.R.; Kim, M.; Yu, S.; Kim, N.; Lim, S.Y.; Park, J.A.; Ha, S.J.; Lim, K.S.; et al. Recent advances in microbial and enzymatic engineering for the biodegradation of micro-and nanoplastics. *RSC Adv.* **2024**, *14*, 9943–9966. [\[CrossRef\]](#) [\[PubMed\]](#)
73. Minhas, W.R.; Saleemi, H.; Raheem, M.F.; Khalid, S.S.; Aslam, H.; Ahmad, W. Crispr-Engineered Microbes for Enhanced Biodegradation of Recalcitrant Pollutants A Genomic Approach To Environmental Remediation. *Front. Health Inform.* **2024**, *13*, 8281–8304. [\[CrossRef\]](#)
74. Rafeeq, H.; Khokhar, A.M.; Afsheen, N.; Rameez, M.; Zia, M.A.; Mehmood, Y.; Ali, M.; Jamil, F.; Riaz, F.; ur Rehman, K.; et al. Microbial enzymes and nanotechnology: A synergistic platform for the detection, degradation, and detoxification of Micro (nano) plastics. *J. Hazard. Mater. Plast.* **2026**, *3*, 100047. [\[CrossRef\]](#)
75. Simran; Amin, G.; Kabir, M.G. Microbial engineering for sustainable microplastic biodegradation: From enzyme redesign to synthetic consortia. *Int. Microbiol.* **2026**, *29*, 23–38. [\[PubMed\]](#)
76. Thakur, S.; Mathur, S.; Patel, S.; Paital, B. Microplastic accumulation and degradation in environment via biotechnological approaches. *Water* **2022**, *14*, 4053. [\[CrossRef\]](#)
77. Arab, B.; Chen, J.; Khusnutdinova, A.N.; Chou, C.P.; Liu, Y. Advancing Bio-Recycling of Nylon Monomers Through CRISPR-Assisted Engineering. *Environ. Technol. Innov.* **2025**, *39*, 104267. [\[CrossRef\]](#)
78. Li, W.; Huang, C.; Chen, J. The application of CRISPR/Cas mediated gene editing in synthetic biology: Challenges and optimizations. *Front. Bioeng. Biotechnol.* **2022**, *10*, 890155. [\[CrossRef\]](#) [\[PubMed\]](#)
79. Zhang, S.; Guo, F.; Yan, W.; Dai, Z.; Dong, W.; Zhou, J.; Zhang, W.; Xin, F.; Jiang, M. Recent advances of CRISPR/Cas9-based genetic engineering and transcriptional regulation in industrial biology. *Front. Bioeng. Biotechnol.* **2020**, *7*, 459. [\[CrossRef\]](#) [\[PubMed\]](#)
80. Dayal, L.; Yadav, K.; Dey, U.; Das, K.; Kumari, P.; Raj, D.; Mandal, R.R. Recent advancement in microplastic removal process from wastewater-a critical review. *J. Hazard. Mater. Adv.* **2024**, *16*, 100460. [\[CrossRef\]](#)
81. Khairul Anuar, N.F.S.; Huyop, F.; Ur-Rehman, G.; Abdullah, F.; Normi, Y.M.; Sabullah, M.K.; Abdul Wahab, R. An overview into polyethylene terephthalate (PET) hydrolases and efforts in tailoring enzymes for improved plastic degradation. *Int. J. Mol. Sci.* **2022**, *23*, 12644. [\[CrossRef\]](#) [\[PubMed\]](#)
82. Cho, S.; Shin, J.; Cho, B.-K. Applications of CRISPR/Cas system to bacterial metabolic engineering. *Int. J. Mol. Sci.* **2018**, *19*, 1089. [\[CrossRef\]](#) [\[PubMed\]](#)
83. Zhu, B.; Wang, D.; Wei, N. Enzyme discovery and engineering for sustainable plastic recycling. *Trends Biotechnol.* **2022**, *40*, 22–37. [\[CrossRef\]](#) [\[PubMed\]](#)
84. Joho, Y.; Vongsouthi, V.; Gomez, C.; Larsen, J.S.; Ardevol, A.; Jackson, C.J. Improving plastic degrading enzymes via directed evolution. *Protein Eng. Des. Sel.* **2024**, *37*, gzae009. [\[CrossRef\]](#) [\[PubMed\]](#)
85. Sahoo, S.; Routray, S.P.; Lenka, S.; Bhuyan, R.; Mohanty, J.N. CRISPR/Cas-mediated functional gene editing for improvement in bioremediation: An emerging strategy. In *Omics Insights in Environmental Bioremediation*; Springer: Berlin/Heidelberg, Germany, 2022; pp. 635–664.
86. Chan, C.T.; Lee, J.W.; Cameron, D.E.; Bashor, C.J.; Collins, J.J. ‘Deadman’ and ‘Passcode’ microbial kill switches for bacterial containment. *Nat. Chem. Biol.* **2016**, *12*, 82–86. [\[CrossRef\]](#) [\[PubMed\]](#)
87. Nkhabindze, B.Z.; Magagula, C.N.; Earnshaw, D.; Mhlanga, C.F.; Matsebula, S.N.; Dladla, I.G. Regulatory framework for genetically modified organisms in the Kingdom of Eswatini. *GM Crop. Food* **2024**, *15*, 212–221. [\[CrossRef\]](#)
88. Brewer, A.M.; George, D.R.; Frow, E.K. Emerging governance considerations for the deployment of genetically engineered microbes. *Curr. Opin. Microbiol.* **2026**, *89*, 102685. [\[CrossRef\]](#) [\[PubMed\]](#)
89. Sajjad, M.; Huang, Q.; Khan, S.; Khan, M.A.; Liu, Y.; Wang, J.; Lian, F.; Wang, Q.; Guo, G. Microplastics in the soil environment: A critical review. *Environ. Technol. Innov.* **2022**, *27*, 102408. [\[CrossRef\]](#)
90. McCarty, N.S.; Ledesma-Amaro, R. Synthetic biology tools to engineer microbial communities for biotechnology. *Trends Biotechnol.* **2019**, *37*, 181–197. [\[CrossRef\]](#) [\[PubMed\]](#)
91. Liu, X.H.; Jin, J.L.; Sun, H.T.; Li, S.; Zhang, F.F.; Yu, X.H.; Cao, Q.Z.; Song, Y.X.; Li, N.; Lu, Z.H.; et al. Perspectives on the microorganisms with the potentials of PET-degradation. *Front. Microbiol.* **2025**, *16*, 1541913. [\[CrossRef\]](#) [\[PubMed\]](#)

92. Gercke, D.; Furtmann, C.; Tozakidis, I.E.; Jose, J. Highly crystalline post-consumer PET waste hydrolysis by surface displayed PETase using a bacterial whole-cell biocatalyst. *ChemCatChem* **2021**, *13*, 3479–3489. [\[CrossRef\]](#)
93. Liu, F.; Wang, T.; Liu, X.; Xu, N.; Pan, X. Efficient biodegradation and upcycling of polyethylene terephthalate mediated by cell-factories. *Front. Microbiol.* **2025**, *16*, 1599470. [\[CrossRef\]](#) [\[PubMed\]](#)
94. Cai, Y.; Wang, Y.; Hu, S. Synthetic Gene Circuits Enable Sensing in Engineered Living Materials. *Biosensors* **2025**, *15*, 556. [\[CrossRef\]](#) [\[PubMed\]](#)
95. Akram, M.A.; Savitha, R.; Kinsella, G.K.; Nolan, K.; Ryan, B.J.; Henehan, G.T. Microbial and enzymatic biodegradation of plastic waste for a circular economy. *Appl. Sci.* **2024**, *14*, 11942. [\[CrossRef\]](#)
96. Chulkov, V.; Gasanov, M.; Isakov, V.; Denisenko, A.; Nwosu, C.; Rodkin, S. Molecular and cellular effects of microplastics and nanoplastics in the pathogenesis of cardiovascular, nervous, urinary, digestive, and reproductive system diseases: A global systematic review. *Int. J. Mol. Sci.* **2025**, *26*, 11194. [\[CrossRef\]](#) [\[PubMed\]](#)
97. Ibrahim, N.; Rahman, A.M.N.A.A.; Shafiq, M.D.; Lockman, Z.; Jaafar, M.; Kameda, Y. Microplastic Pollution: Sources, Degradation Mechanisms, Analytical Advances, and Mitigation Strategies for Environmental Sustainability. *Rev. Environ. Contam. Toxicol.* **2025**, *263*, 27. [\[CrossRef\]](#)
98. Wu, X.; He, F.; Xu, X.; Wu, L.; Rong, J.; Lin, S. Environmental Health and Safety Implications of the Interplay Between Microplastics and the Residing Biofilm. *Environ. Health* **2024**, *3*, 118–132. [\[CrossRef\]](#)
99. Purohit, J.; Chattopadhyay, A.; Teli, B. Metagenomic exploration of plastic degrading microbes for biotechnological application. *Curr. Genom.* **2020**, *21*, 253–270. [\[CrossRef\]](#)
100. Eisen, J.A. Environmental shotgun sequencing: Its potential and challenges for studying the hidden world of microbes. *PLoS Biol.* **2007**, *5*, e82. [\[CrossRef\]](#) [\[PubMed\]](#)
101. Handelsman, J. Metagenomics: Application of genomics to uncultured microorganisms. *Microbiol. Mol. Biol. Rev.* **2004**, *68*, 669–685. [\[CrossRef\]](#) [\[PubMed\]](#)
102. Henawy, A.R.; Ismail, S.M.; Gharib, S.; Elarabi, N.I.; Abdelhadi, A.A.; Halema, A.A. Harnessing bacterial power and omics technologies for sustainable plastic waste biodegradation. *Biodegradation* **2026**, *37*, 46. [\[CrossRef\]](#) [\[PubMed\]](#)
103. Shukla, S.; Khan, R.; Chrzanowski, L.; Vagliasindi, F.G.A.; Roccaro, P. Advancing sustainable agriculture through multi-omics profiling of biosolids for safe application: A review. *J. Environ. Manag.* **2025**, *375*, 124292. [\[CrossRef\]](#)
104. Samrot, A.V.; Abubakar Mohamed, A.; Faradjeva, E.; Si Jie, L.; Hooi Sze, C.; Arif, A.; Chuan Sean, T.; Norbert Michael, E.; Yeok Mun, C.; Xiao Qi, N.; et al. Mechanisms and impact of biofilms and targeting of biofilms using bioactive compounds—A review. *Medicina* **2021**, *57*, 839. [\[CrossRef\]](#) [\[PubMed\]](#)
105. Wang, X.; Jiang, Y.; Liu, H.; Yuan, H.; Huang, D.; Wang, T. Research progress of multi-enzyme complexes based on the design of scaffold protein. *Bioresour. Bioprocess.* **2023**, *10*, 72. [\[CrossRef\]](#) [\[PubMed\]](#)
106. Hajnajafi, K.; Iqbal, M.A. Mass-spectrometry based metabolomics: An overview of workflows, strategies, data analysis and applications. *Proteome Sci.* **2025**, *23*, 5. [\[CrossRef\]](#) [\[PubMed\]](#)
107. Yang, J.; He, J.; Jia, L.; Gu, H. Integrating metagenomics and metabolomics to study the response of microbiota in black soil degradation. *Sci. Total Environ.* **2023**, *899*, 165486. [\[CrossRef\]](#) [\[PubMed\]](#)
108. Gu, J.-D.; Wu, E.K.W. Biodegradability of synthetic plastics and polymeric materials: An illusion or reality in waste managements? *Appl. Environ. Biotechnol.* **2021**, *5*, 9–27. [\[CrossRef\]](#)
109. Ruginescu, R.; Purcarea, C. Plastic-degrading enzymes from marine microorganisms and their potential value in recycling technologies. *Mar. Drugs* **2024**, *22*, 441. [\[CrossRef\]](#) [\[PubMed\]](#)
110. Jiménez, D.J.; Öztürk, B.; Wei, R.; Bugg, T.D.; Amaya Gomez, C.V.; Salcedo Galan, F.; Castro-Mayorga, J.L.; Saldarriaga, J.F.; Tarazona, N.A. Merging plastics, microbes, and enzymes: Highlights from an international workshop. *Appl. Environ. Microbiol.* **2022**, *88*, e0072122. [\[CrossRef\]](#) [\[PubMed\]](#)
111. Grigorakis, K.; Ferousi, C.; Topakas, E. Protein engineering for industrial biocatalysis: Principles, approaches, and lessons from engineered PETases. *Catalysts* **2025**, *15*, 147. [\[CrossRef\]](#)
112. Xiao, H.; Bao, Z.; Zhao, H. High throughput screening and selection methods for directed enzyme evolution. *Ind. Eng. Chem. Res.* **2015**, *54*, 4011–4020. [\[PubMed\]](#)
113. Casilli, F.; Canyelles-Niño, M.; Roelfes, G.; Alonso-Cotchico, L. Computation-guided engineering of distal mutations in an artificial enzyme. *Faraday Discuss.* **2024**, *252*, 262–278. [\[CrossRef\]](#) [\[PubMed\]](#)
114. Kim, J.; Grate, J.W.; Wang, P. Nanobiocatalysis and its potential applications. *Trends Biotechnol.* **2008**, *26*, 639–646. [\[CrossRef\]](#) [\[PubMed\]](#)
115. Sharifi, M.; Sohrabi, M.J.; Hosseinali, S.H.; Hasan, A.; Kani, P.H.; Talaie, A.J.; Karim, A.Y.; Nanakali, N.M.Q.; Salihi, A.; Aziz, F.M.; et al. Enzyme immobilization onto the nanomaterials: Application in enzyme stability and prodrug-activated cancer therapy. *Int. J. Biol. Macromol.* **2020**, *143*, 665–676. [\[CrossRef\]](#) [\[PubMed\]](#)
116. Abdelhamid, M.A.A.; Khalifa, H.O.; Yoon, H.J.; Ki, M.-R.; Pack, S.P. Microbial immobilized enzyme biocatalysts for multipollutant mitigation: Harnessing nature's toolkit for environmental sustainability. *Int. J. Mol. Sci.* **2024**, *25*, 8616. [\[CrossRef\]](#) [\[PubMed\]](#)

117. Hao, X.; Liu, P.; Chu, X. Recent advances in the strategies of simultaneous enzyme immobilization accompanied by nanocarrier synthesis. *Appl. Sci.* **2024**, *14*, 3702. [[CrossRef](#)]
118. Anand, U.; Dey, S.; Bontempi, E.; Ducoli, S.; Vethaak, A.D.; Dey, A.; Federici, S. Biotechnological methods to remove microplastics: A review. *Environ. Chem. Lett.* **2023**, *21*, 1787–1810. [[CrossRef](#)] [[PubMed](#)]
119. Parashar, N.; Hait, S. Recent advances on microplastics pollution and removal from wastewater systems: A critical re-view. *J. Environ. Manag.* **2023**, *340*, 118014. [[CrossRef](#)]
120. Yan, W.; Pengfei, H.U. Harnessing Biofilm-Mediated Bioremediation for Sustainable Wastewater Reuse: From Mechanisms to Policy Integration. *Water Res. X* **2025**, *29*, 100456. [[CrossRef](#)]
121. Wu, Y.; Yi, R.; Wang, Y.; Zhang, C.; Zheng, J.; Ning, P.; Shan, D.; Wang, B. Light-driven degradation of microplastics: Mechanisms, technologies, and future directions. *J. Hazard. Mater. Adv.* **2025**, *17*, 100628. [[CrossRef](#)]
122. Zhang, X.; Li, X.; Zhao, X.; Li, Y. Factors affecting the efficiency of a bioelectrochemical system: A review. *RSC Adv.* **2019**, *9*, 19748–19761. [[CrossRef](#)] [[PubMed](#)]
123. Bai, X.; Li, C.; He, S.; Zhou, J.; Hu, J. Combining advanced oxidation processes with biological processes in organic wastewater treatment: Recent developments, trends, and advances. *Desalin. Water Treat.* **2025**, *323*, 101263. [[CrossRef](#)]
124. Tournier, V.; Topham, C.M.; Gilles, A.; David, B.; Folgoas, C.; Moya-Leclair, E.; Kamionka, E.; Desrousseaux, M.-L.; Texier, H.; Gavalda, S.; et al. An engineered PET depolymerase to break down and recycle plastic bottles. *Nature* **2020**, *580*, 216–219. [[CrossRef](#)] [[PubMed](#)]
125. Ellis, L.D.; Rorrer, N.A.; Sullivan, K.P.; Otto, M.; McGeehan, J.E.; Román-Leshkov, Y.; Wierckx, N.; Beckham, G.T. Chemical and biological catalysis for plastics recycling and upcycling. *Nat. Catal.* **2021**, *4*, 539–556. [[CrossRef](#)]
126. Puteri, M.N.; Gew, L.T.; Ong, H.C.; Ming, L.C. Technologies to eliminate microplastic from water: Current approaches and future prospects. *Environ. Int.* **2025**, *199*, 109397. [[CrossRef](#)] [[PubMed](#)]
127. Dey, S.; Veerendra, G.T.N.; Babu, P.S.S.A.; Manoj, A.V.P.; Nagarjuna, K. Degradation of plastics waste and its effects on biological ecosystems: A scientific analysis and comprehensive review. *Biomed. Mater. Devices* **2024**, *2*, 70–112.
128. Bhattacharjee, U.; Baruah, K.N.; Shah, M.P. Exploring sustainable strategies for mitigating microplastic contamination in soil, water, and the food chain: A comprehensive analysis. *Environ. Chem. Ecotoxicol.* **2025**, *7*, 453–461. [[CrossRef](#)]
129. Jiao, H.; Al-Tohamy, R.; Xiong, M.; Schagerl, M.; Reinthaler, T.; Al-Zahrani, M.; Sun, J.; Ali, S.S. Microplastic biodegradation and environmental safety: From microbial mechanisms to engineered systems and circular bio-based implementation. *Ecotoxicol. Environ. Saf.* **2026**, *313*, 120016. [[CrossRef](#)] [[PubMed](#)]
130. Lerner, A.; Lieber, A.D.; Nelson-Dooley, C.; Leu, A.; Perro, M.; Koch, G.; Benzvi, C.; Smith, J. Genetically Modified Microorganisms: Risks and Regulatory Considerations for Human and Environmental Health. *Microorganisms* **2026**, *14*, 467. [[CrossRef](#)] [[PubMed](#)]

Disclaimer/Publisher’s Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.