BIODEGRADATION OF POLYETHYLENE TEREPHTHALATE (PET): A CRITICAL REVIEW

by

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Abstract

Polyethylene terephthalate (PET) is a highly persistent plastic pollutant that undergoes minimal natural degradation, prompting increasing interest in biological strategies as alternatives to conventional recycling methods. This critical review systematically analyzed 152 peer-reviewed studies, categorized into four methodological groups: enzyme-only, microbial-only, combined, and unconventional approaches to PET biodegradation. Studies were identified through structured searches and selected based on relevance, methodological quality, and availability of degradation data. Enzymatic systems - particularly engineered variants of PETase and cutinase - demonstrated high depolymerization efficiencies under optimized laboratory conditions. Microbial approaches effectively metabolized PET breakdown products across diverse environments but generally exhibited slower performance. Hybrid systems integrating enzymes, microorganisms, or pretreatments showed enhanced overall conversion, while unconventional strategies offered proof-of-concept insights with limited validation. Despite these advances, key challenges remain, including the lack of standardized degradation metrics, difficulties in scaling processes, and inconsistent substrate characterization. This review consolidates existing findings into systematic pathways, highlights recurring limitations, and outlines application contexts. The future of PET biodegradation will depend on continued innovations in enzyme engineering, microbial design, and integrated systems, with emphasis on improving efficiency and scalability to enable practical, real-world implementation within sustainable plastic waste management frameworks.

Keywords

Polyethylene terephthalate (PET), PET biodegradation, Enzymatic depolymerization, Microbial degradation, biological recycling

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List of Abbreviations

BbPETase	*Bacillus subtilis*-derived PETase
BHET	Bis(2-hydroxyethyl) terephthalate
BMHETase6M	Engineered MHETase variant with six mutations
Bs	*Bacillus subtilis*
BhrPETase	Bacterium HR29-derived PETase
cCALB	Chimeric Candida antarctica Lipase B
CBM	Carbohydrate-Binding Module
cPETase	Chimeric PETase
cScaffold (CBM3)	Cohesin-dockerin interaction scaffold protein
DAQI	Engineered LCC variant with DuraAQI mutations
EG	Ethylene Glycol
EstB	Esterase B from *Pseudomonas sp.*
FoCut	Computationally modeled Cutinase (*Fusarium oxysporum*)
GPI	Glycosylphosphatidylinositol Anchor
HDPE	High-Density Polyethylene
HFBI	Hydrophobin I from *Trichoderma reesei*
HGFI	Hydrophobin from Grifola frondosa
HiC	*Humicola insolens* Cutinase
ICCG	Industrially Composted Cutinase Variant (engineered LCC)
ICCM	Industrially Composted Cutinase Mutant
Is	*Ideonella sakaiensis*
KT2440	*Pseudomonas putida* KT2440 (model strain)
LCC	Leaf-Branch Compost Cutinase
LDPE	Low-Density Polyethylene
MD	Molecular Dynamics
MHET	Mono(2-hydroxyethyl) terephthalate
MHETase	Mono(2-hydroxyethyl) terephthalate Hydrolase
MtCut	Marine-derived Cutinase
NMR	Nuclear Magnetic Resonance
OaCut	Cutinase from *Oleispira* antarctica
PA	Polyamide (nylon)
PC enzyme complex	PETase + CALB + CBM3 scaffold enzyme assembly
PET	Polyethylene Terephthalate
PETase	Polyethylene Terephthalate Hydrolase
PHA	Polyhydroxyalkanoate
PHB	Polyhydroxybutyrate
PMID	PubMed Identifier
Pp	*Pichia pastoris*

PP	Polypropylene
PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-Analyses
PS	Polystyrene
PVC	Polyvinyl Chloride
QM/MM	Quantum Mechanics/Molecular Mechanics (hybrid computation)
R. *jostii*	*Rhodococcus jostii*
RolA	Hydrophobin from *Aspergillus oryzae*
SbPETase	Schlegelella brevitalea-derived PETase
ScPETase	PETase from Schlegelella brevitalea
SDS-PAGE	Sodium Dodecyl Sulfate–Polyacrylamide Gel Electrophoresis
SEM	Scanning Electron Microscopy
TfCut2	*Thermobifida fusca* Cutinase 2
TPA	Terephthalic Acid
WT	Wild Type

1. Introduction

1.1. Global plastic pollution and the growing burden of PET

1.1.1. Overview of major plastic types & plastic pollution

The problem of plastic pollution represents one of the most significant green issues in the 21st century. Globally, it is expected that 380 million tonnes of plastics are produced annually; only a small portion (9%) of the plastic is recycled very well, the rest end up in the landfills, natural habitat, and the marine environment (1). This mass immobilization has left ubiquitous plastic rubbish in all the ecological sections, including coastal beaches and deep-sea sediments, hence jeopardizing biodiversity, ecological stability, and human health. It is known that organisms from different trophic levels ingest macroplastics and microplastics, leading to various toxicological effects (1), (2).

The six major polymer types are polyethylene terephthalate (PET), high-density polyethylene (HDPE), polyvinyl chloride (PVC), low-density polyethylene (LDPE), polypropylene (PP), and polystyrene (PS), each designated by a specific resin identification code. These polymers have their difference in chemical structure, their use, and impact on the environment. PET and HDPE are primarily used in packaging and textiles because of their strength and ability to be resistant to moisture, while PVC is commonly used in pipes, construction materials, and cables. Plastic bags and films contain LDPE; PP is used in automotive and household products and PS can be found in the rigid application as well as expanded foam applications such as food containers and insulation. Other plastics outside of the six major groups include polycarbonate, acrylic, nylon, bioplastics, among others and should be referred to the other category (3). In simplicity, **Figure 1** illustrates

the various principal resin codes used in plastic categorization, with PET (Code 1) marked due to its widespread use in packages and applicability in this review.

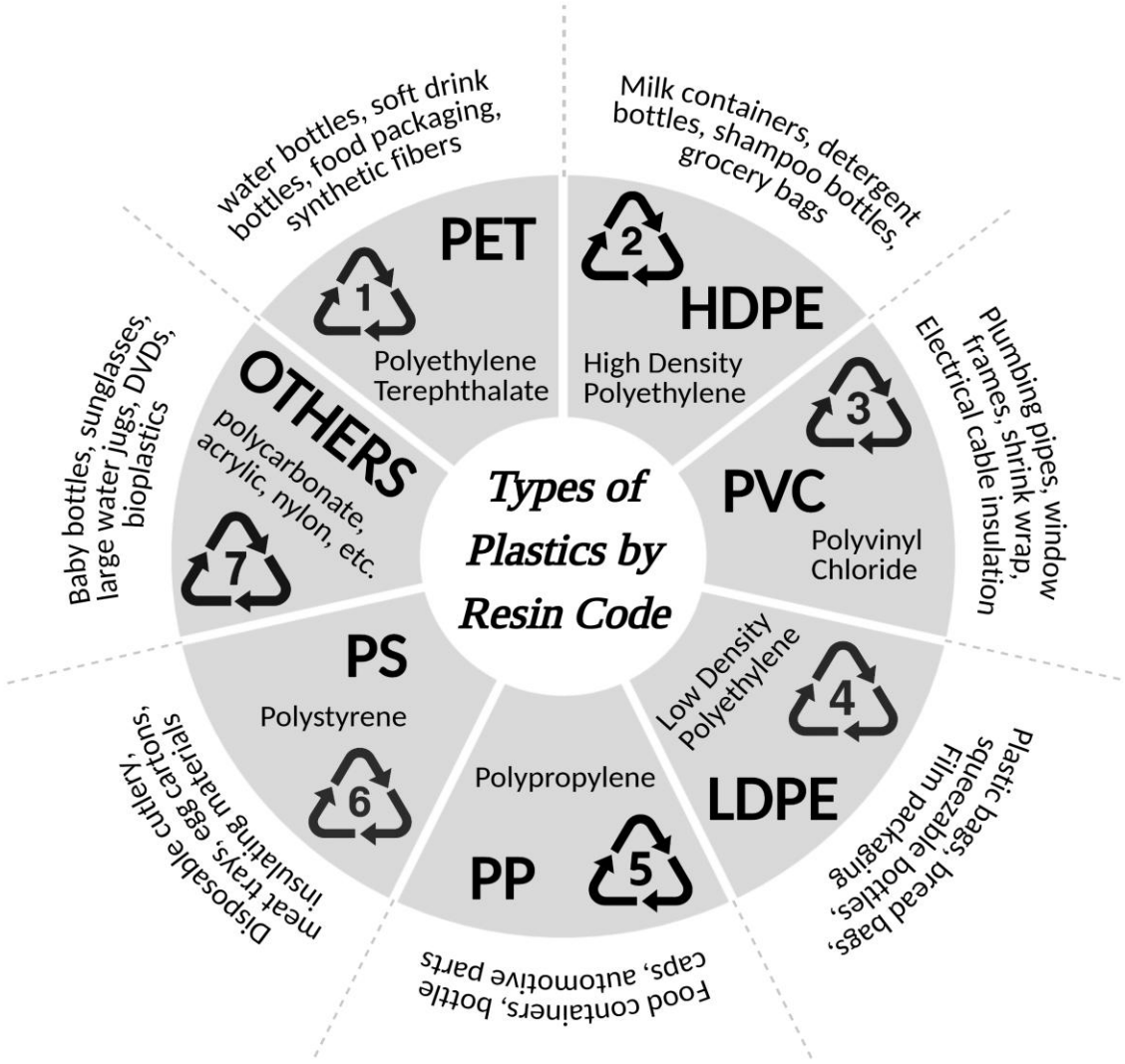


Figure 1. Classification of common plastic types by resin identification code. Among these, polyethylene terephthalate (PET, resin code 1) is widely used in bottles, packaging, and textiles. Created in BioRender.

Although PET is technologically recyclable, its recovery remains inefficient. Globally, the recycling rate of PET bottles is low due to contamination, limitations in sorting systems, and unfavorable economic factors (4). HDPE reclamation is a little more successful, whereas LDPE and PP are characterized by processing challenges, which limit reclamation. PS, especially in

expanded foam form, is virtually indestructible and degrades easily into little fragments, to become one of the biggest pollutants in the watery environment. The “other” category presents the greatest challenge for waste management, as it often includes complex or multi-layered materials that are incompatible with existing recycling systems (3). The very stubbornness, prevalence, and magnitude of plastic waste point to the relevance of positive changes in this area and effective solutions to eliminate the danger. Such interventions require a good comprehension of the chemical character and environmental fate of various forms of plastic, such as their degradation.

1.1.2. Rationale for focusing on PET

1.1.2.1. Production & application of PET

Among the multitude of synthetic polymers that have caused pollution in the world in terms of plastic, the polyethylene terephthalate (PET) has proved unique by its immense use rate in addition to its wide spectrum application. PET was originally used on synthetic fibers but through technological advancements and increasing commercialization it has become one of the most commercially important thermoplastics today and is used in packaging, textile and other industrial sectors. Its low weight, high tensile strength, resistance to chemical attacks, and clarity have predisposed its use in beverage bottles and food packages, and the robust nature of the material has further predisposed its domination with respect to polyester fibers in clothing and household material.

Human ubiquity to this material can be captured by productive reports that designate global PET production in excess of 82 million metric tons per year (5). Much of this amount is split into two main applications i.e. about 24 million metric tons of plastic is estimated to be used to create containers and single use beverage bottles, and about 44 million metric tons to create textiles and

synthetic fibers. PET use is extremely ubiquitous in most short-life and long-life consumer goods, hence there is a high possibility that plastic waste is produced at different stages of product life cycle. Specifically, PET textiles can shed durable microplastic during washing, whereas packaging materials can slip through the hands of recovery systems since they lack adequate sorting and recycling systems.

Besides increasing the environmental burden of PET, its scale and diversity in its use render it a vital center of interest in terms of intervention. Although this is a good thing as far as the product performance is concerned, it makes it extremely resistant to degradation in natural environments. Therefore, poorly handled PET waste ends up in the land and water systems, which leads to decades-long persistence and division of microplastics that are difficult to identify or loop back.

The volume, durability, and well-known chain of supply, as well as the uniformity in polymer properties, make it reasonable to pay attention to PET in biodegradation work. It is traceable and sortable using its resin identification code (#1) with logistical benefits to recycling, as well as scientific research. All these considerations of prevalence, persistence, and practicality make PET a top and unavoidable agenda in dealing with the overall plastic pollution problem.

1.1.2.2. Ubiquity of PET in pollution

PET is a highly stable plastic because it has a semi-crystalline association and its strong resistance towards weathering. This strength, although it makes it useful in its commercial use, implies that the PET waste is long-lasting in the land and marine environments. For example, studies indicate that much of the plastic entering the oceans originates from land-based activities, with an estimated 8 million tonnes discharged annually. A substantial proportion of this input consists of PET, which remains a dominant contributor to marine pollution. PET particles ultimately break down into

microplastics that have since been found in a variety of habitats in the oceans and shore sediments, freshwaters, and in the air itself.

These microparticle strands are a result of gradual decomposition of PET bottle, packaging and fibers, which is catalyzed by UV light, rubbing and thermal cycling. Gradually, small plastic elements are collected in layers of sediment and the water column, the level of which increased considerably because of the popular use of PET in disposable containers and textiles. The widespread nature of the global presence of PET contamination was revealed when microplastics were found in such ecologically sensitive territory as the Arctic, in addition to urban rivers. In addition to the high prevalence in the environment, PET-derived micro plastics increasingly endanger wildlife as well as humans. A large variety of organisms such as plankton and fish and seabirds consume them with food chain transfer potential. During microplastic surveys, PET is one of the most detected polymers. It validates PET's important role in health-related and ecological issues for plastic pollution (6).

The fact that PET is everywhere, and that it cannot be broken down naturally, highlights the importance of coming up with specific intervention techniques to remove and biodegrade it. Being one of the major components of microplastics in the environment, PET needs special attention to be considered: to reduce the already existing effects and to prevent its further accumulation.

1.2. Urgency of addressing PET waste

1.2.1. Environmental impact

PET (polyethylene terephthalate) is extremely persistent in natural environmental conditions due to the strong influence of the semi-crystalline structure and its insensitivity to biodegradation. PET waste that enters the water can remain in the aquatic ecosystems over the decades or even centuries,

during which it is slowly fragmenting into microplastics. Makeup macroplastics made of PET have now been found in a massive range of ecosystems, including coastal sediments and ocean gyres, lakes and rivers, and even glacier snow, indicating just how widespread and problematic the material is in the environment.

This kind of fragmentation worsens ecological hazards in several ways. First, the microplastic is easily consumed by the organisms of different trophic levels, such as zooplankton, fish, seabirds, and incurs physical threats and can add toxic additives and adsorbed pollutants to the organism (7). Second, it has been shown that the particles may accumulate inside organisms with the possibility of bioaccumulation, and trophic transfer, which further exposes the environment and increases the effect of the impacts through food webs (6). Consequently, PET pollution does not only present itself as apparent waste alone, but it is also a permanent reservoir of microplastic pollution that disrupts animal health, biodiversity, and ecosystem robustness. It is, therefore, crucial to realise this sort of pollution at an early stage in addition to recognizing that it is persistent and pervasive and hence proceed with the effort to degrade PET.

1.2.2. Health impact

The microplastics and chemical additives released by the polyethylene terephthalate (PET) product are known to leach in the soil, water bodies, and food chain that produces a possible form of entry into the human body through the consumption, inhalation, and direct contact of the skin. Although the daily exposure of environmental microplastics by diet is finally acknowledged, there is still very scarce knowledge regarding long-term consequences of chronic microplastic accumulation. Other reports have expressed concern that substances in PET like antimony, phthalates and

bisphenols can be set free due to ingestion of this material, which can possibly impact endocrine systems or subject cells to cellular stress (8).

Some of the recent studies regarding bioaccumulation of microplastics have revealed the presence of the particles in human tissues and body fluids, which sparked speculation regarding their long-term impact on body systems and metabolism. Even though the data remain sparse, researchers are increasingly realizing that continuous exposure to PET microplastic may contribute to the response of inflammation, microbiome levels, and even reproductive or developmental outcomes, particularly when an individual is exposed early in life or occupationally (9). Despite limited existing evidence, the emergence in the research interest points towards the potential health risks of prolonged exposure to PET microplastic and future research is needed about the long-term biological impacts of such exposure.

1.2.3. Economic impact

The economic consequences of PET waste go far beyond what one can see as pollution and introduce huge liabilities on the waste management systems and are resources that would have been efficiently used. The municipalities also have the burden on them in that they must spend so much on collecting, sorting and disposing PET and they must do the environmental remediation as well as many investments being made- this is because they must clean the silted-up waterways and clean up the affected habitats. Such costs take away precious financing of essential infrastructure and neighborhood amenities.

Meanwhile, low PET recycling rates compound the number of economic losses. Regardless of being the most common recycled plastic resin, in most areas, PET bottles are not collected to more than 30 percent of their level thus most of the substance cannot be reused as part of the economy

(10). Such inefficiency comes at the cost of both manufacturers and recycling industries, which lose the material value of this system, and at the cost of ever more virgin PET produced via petroleum feedstocks. Consequently, the world has recycling systems, which resemble an erratic supply chain, and a promise of a circular economy in PET is yet to be largely fulfilled. It is imperative to know these economic dynamics. Understanding these economic dynamics underscores the need for improved waste management infrastructure and innovative degradation technologies, both of which form central themes of this review on PET biodegradation.

1.3. Need for sustainable PET waste management

1.3.1. Limitations of conventional PET recycling methods

1.3.1.1. Landfilling drawbacks

Although landfilling is a common method, it is not a good long-term method of discarding PET waste. The semi-crystalline nature and hydrophobicity make PET resist the growth of microbes and environmental degradation; hence it can last several centuries in the landfills without considerable breakdown (3). It results in the long-term use of space in landfills and slow build-up of the stable plastic substance that cannot be considered part of natural cycles. With time, embedded PET can degrade due to physical and chemical stressors, cracking into microplastics that migrate into neighboring soils, spread into the groundwater. This kind of pollution is destructive to the environment and drinking water. Therefore, research is needed to identify sustainable and effective methods of degrading PET.

1.3.1.2. Incineration drawbacks

Incineration of PET waste has the potential of recovery of energy but comes at high environmental and social health costs. Care should be taken over the combustion process of toxic byproducts in

which dioxin, furan, particulate matter such as heavy metal, greenhouse gas and dioxin are the greatest risks despite having emitted controls in the combustion process (11). These are pollutants that may spread to long distances and cause buildup in ecosystems, leading to respiratory diseases and environmental pollution as well as climatic changes. Incinerators also require high capital investment and operational costs. Due to its hazardous emissions coupled with the cost of infrastructures, incineration of PET waste is not a sustainable, acceptable procedure of managing wastes. These are some of the intrinsic disadvantages which drive the necessity of more eco-friendly sustainable alternatives to PET degradation.

1.3.1.3. Conventional mechanical recycling methods

Recycling of PET the most widespread mechanism includes such processes as collection, sorting, cleaning, shredding and remelting. Although it consumes less energy as compared to producing virgin PET, its application is somehow crippled by the degradation of the polymer, discoloration as well as contamination at every cycle. Exposure to heat and shear during processing causes chain scission, which reduces molecular weight and leads to embrittlement, discoloration, and loss of transparency in recycled PET. These quality losses limit recycling of PET to lower-value usages like fibers or liner materials of a package, instead of food grade bottles.

Due to the restrictions, mechanical recycling may be downcycling rather than recycling because the recycled material is less valuable and available to more uses. To make matters worse, practical recycling systems have not performed well in reducing the inefficiencies of sorting PET as well as contamination leading to recycling of a small portion of the PET that is recycled more than once with the other part discarded or diverted to landfilling and incineration (12). Such systemic wastefulness is indicative of the fact that mechanical recycling cannot be the sole method to deal

with PET waste consumption, and other methods must be utilized, like chemical recycling and biological degradation.

1.3.1.4. Conventional chemical recycling methods

One potential salvaging approach includes chemical depolymerization methods that theoretically should allow the re-refinement of PET into its original molecular components (terephthalic acid and ethylene glycol), including hydrolysis, glycolysis, and methanolysis methods. Such processes hold the prospect of completing the material loop, but reality must face vicissitudes plagued by their inexplicably involved nature. Key issue here is the multiple steps involved in purification of reaction products, which in many cases contain residual oligomers, solvents, catalysts and byproducts. Recycling of high-purity monomers requires series processes such as filtration, crystallization, distillation, and pH adjustment, which increase the cost of operation, energy consumption, and complexity of infrastructure.

Also, the approaches are still untapped on industrial scale, because of the intense capital and operating costs. In economic terms, pilot plants find it difficult to sustain operations due to the extreme temperatures usually needed in the depolymerization reactors, corrosive chemicals and extreme waste-stream management (12). Greater complexity of purification, usage of harmful reagents and energy-consuming conditions only serve to destroy the scalability of chemical recycling. Such barriers, in their turn, point to the importance of finding alternative solutions in the realm of PET management, particularly, biological or enzymatic processes that could possibly provide less complicated, less intensive, more environmentally friendly ways.

Table 1 outlines these conventional plastic waste treatment practices and respective shortcomings for each practice, highlighting the widespread technical, economic, and environmental disadvantages of each practice, including material degradation, leaching of microplastics, limited space, toxic emissions, and expensive operating costs. All together, these age-old concerns have created more worldwide motivation toward alternative practices, especially those biological in nature, like enzymatic and microbial PET biodegradation.

Conventional Method	Key Limitations
Landfilling	Space usage, microplastic leaching, groundwater risk, long-term burden
Incineration	Toxic emissions, high setup cost, air pollution, limited public acceptance
Mechanical Recycling	Quality loss, downcycling, thermal damage, limited reuse, inefficient sorting
Chemical Recycling	High cost, complex processing, intensive energy use, still emerging at scale

Table 1. Overview of conventional plastic disposal methods and their key limitations. Created in BioRender.

1.3.2. Biodegradation as a sustainable solution

1.3.2.1. Concepts

The biodegradation of PET represents a sustainable alternative to conventional plastic waste management methods. In this method, biological agents - mainly enzymes or microorganisms, are utilized which can catalyse the degradation of PET into its monomeric forms, namely terephthalic acid (TPA) and ethylene glycol (EG). Such monomers can then be either repolymerized in PET or fed to other plastic industrial applications and create a more circular pathway of plastic utilization (13).

The difference between biodegradation and the mechanical or chemical treatment method of recycling is that it operates under relatively mild and environmentally friendly conditions. The most common form of biological depolymerization is performed at ambient temperatures, neutral pH, and in water with little additional energy input required, and a minimised requirement of hazardous reagents. This is especially attractive both ecologically and technologically, especially in the situations when the traditional practices are not economically or logically viable.

It has also developed the concept to include simple microbial metabolism into advanced forms of innovation in enzyme engineering, the design of microbial consortia and bioreactor complexation. In the present day, biodegradation of PET is not only considered as an option in the waste management processes but a pillar in creating a sustainable material management. In this context, it underscores the broader interest in biological solutions for synthetic polymer degradation and serves as a key foundation for this review's exploration of next-generation PET degradation strategies.

1.3.2.2. Advantages over conventional methods

There are unique benefits of the biodegradation of PET compared to the more conventional mechanical or chemical recycling approaches. To begin with, it does not require use of fossil fuels, high temperatures, and toxic chemicals because enzymes and microorganisms work under mild, neutral, or watered conditions. This saves abundant energy and environmental risks during high temperatures or solvent treatments. Another major value is an upcycling potential. Due to providing a great degree of purity of the monomers, biological depolymerization may also enable the production of virgin-grade PET, or specialty chemicals. This is compared with mechanical recycling, in which downgraded materials are common, and compared to chemical recycling, in which the monomer yields may be impure because of side-reactions or contamination. There is

also the potential to process PET waste that is not good in mechanical recycling like colored, multilayer, highly contaminated streams via biodegradation. Today, these materials are often disregarded by existing systems and left to rot up in landfills, but with the use of enzymatic reactions, the impurities can be put abreast with, and the mixed waste can be broken down into valuable monomers (14).

Lastly, such benefits serve as direct contributions to the aims of a circular economy, providing profitable recovery of resources and, at the same time, limiting the number of greenhouse-gas emissions. Life-cycle assessments in a case of implementing higher levels of PET biodegradation and biological recycling, life-cycle assessments demonstrate that carbon footprints can be reduced by a substantial amount relative to mechanical methods without biological recycling and biodegradation (15). Biodegradation, therefore, comes out as not only a treatment alternative but also a core solution to the shift in the waste management system to sustainability and circularity.

1.4. Overview of PET biodegradation approaches

To appreciate the common objective of PET biodegradation methods, it is essential to first consider the underlying chemical pathway by which PET is broken down, whether through enzymatic, microbial, or hybrid processes. This review examines biological PET degradation strategies, most of which are grounded in the fundamental depolymerization pathway illustrated in **Figure 2**.

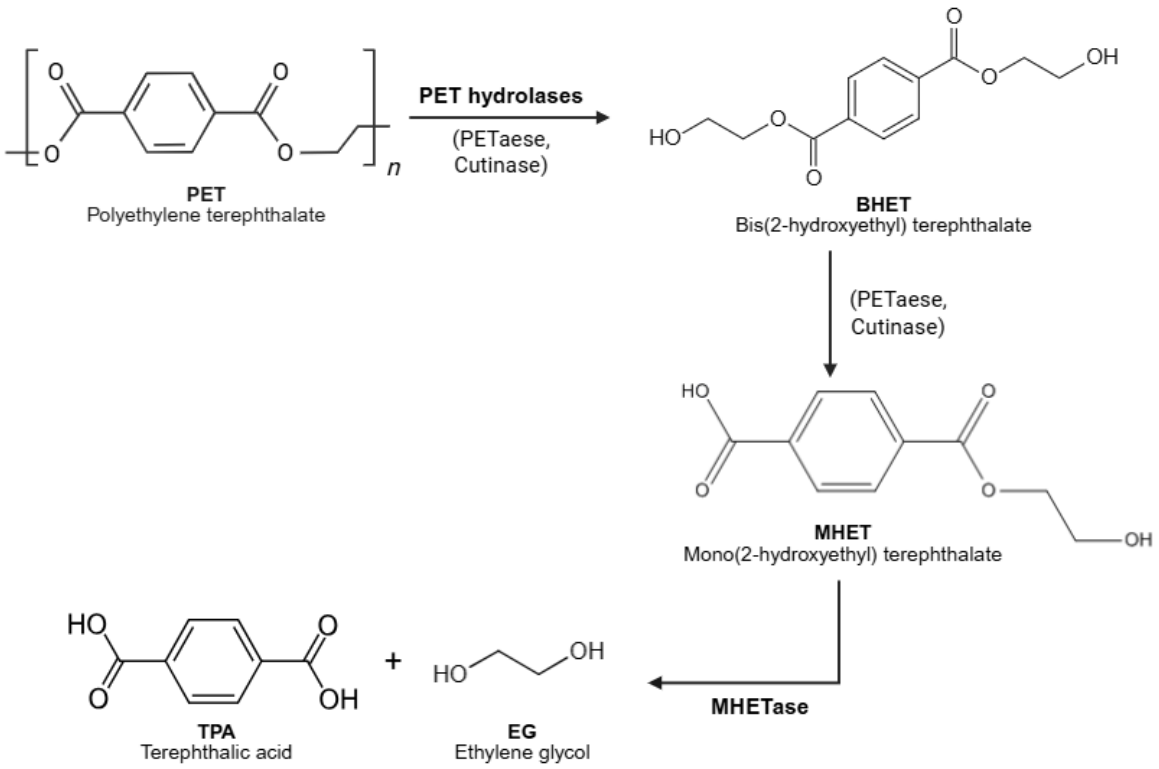


Figure 2. Hydrolysis pathway of PET into BHET, MHET, TPA, and EG; a meaningful chemical route in PET biodegradation. PET is broken down into both BHET intermediates and also MHET intermediates in enzymatic systems and in microbial degradation. These intermediates are further hydrolyzed into monomeric products: terephthalic acid (TPA) along with ethylene glycol (EG). Created in BioRender.

1.4.1. Enzymatic-only approaches

The enzymatic-only approaches reflect one of the main strategies investigated in PET biodegradation studies since it deals specifically with the application of purified enzymes to help depolymerize PET. These systems work on the enzyme such as PETase, MHETase, cutinases and leaf-branch compost cutinase (LCC), which can break the ester bond in PET and liberate its basic components TPA and EG. Reaction parameters, like temperature, pH, and concentration of the

substrate can be controlled very precisely using the isolated enzymes. This replication will allow reproducible experiments and allow the investigation of both the mechanism and molecular basis of enzymatic PET hydrolysis. This has made enzyme-based systems useful vehicles to explore structure-function relationships and to improve biocatalytic performance either by protein engineering or by immobilization schemes.

PET biodegradation is an enzymatic process under increasing research interest because it could assist in environmentally compatible recycling processes. With the growing amount of research in the enzyme-substrate interaction process, structural modifications, catalytic activities, etc., this has become a significant framework in the enhancement of the biological plastic disposal methods. Enzymatic techniques are also beneficial to the overarching study on how biological tools may be used in sustainable waste management systems through provision of a controlled and modular system. In this part, it is important to introduce enzymatic degradation of PET within the framework of this review as the one of the discrete lines of inquiry.

1.4.2. Microbial-only approaches

There are microbial-only methods, which utilize living microorganisms that depolymerize PET by their inherent metabolic pathways. The techniques depend on the innate capacity of a particular strain of bacteria or fungi to produce PET-dissolving enzymes or to break down the by-products of the degradation process. In comparison with enzymatic-only systems, microbial systems do not need to use externally purified enzymes but allows in situ degradation of PET in a microbial way. The studies conducted on the topic of PET degradation have mainly been stressing on the isolation of PET-degrading microbes in a few environments including landfills, composting facilities, the sea, and wastewater. Genera commonly studied include *Pseudomonas*, *Rhodococcus*,

Comamonas, *Bacillus* and *Ideonella*, among others, which exhibit signs of adaptive response towards synthetic polymers. At least some of these microorganisms can grow on PET or its monomers as sources of both carbon and energy, and they thus are attractive targets of waste treatment systems in an environmentally relevant setting.

The biodegradation of PET by microbial-only systems also forms an understanding about the ecology of plastic-polluted environments and it can be used to establish a bioaugmentation or bioremediation process. Such methods are commonly investigated to determine natural biodegradation routes, sources of genetic control of enzyme manufacture, and metabolic pyrones of degradation intermediates. On its own, microbial-only methods are one of four main pathways investigated through this review and form part of an evolving knowledge base based on biologically mediated PET hydrolysis.

1.4.3. Combined approaches

Integrated methods of PET biodegradation, the combination of two or more methods is often used where two strategies may be combined, such as enzymatic degradation/microbial assimilation, or chemical pretreatment/biological conversion. The advantage of these hybrid systems is that they manage to surpass the drawbacks of one-approach mechanisms because they allow more efficient fragmentation and exploitation of PET. These methods enhance an improved and balanced degradation process by combining enzymatic hydrolysis and microbial metabolism. Preliminary chemical treatments may in some cases be applied to increase accessibility of polymers of subsequent depolymerization or assimilation via biological means. With such integration, enhanced plastic waste flexibility and even scalability to deal with complex and/or contaminated plastic waste is possible. With the growing research, such multi-step systems are being explored

in a potential course of sustaining plastic waste management under a circular economy. Combinational approaches are one of the future prospective fields of PET biodegradation research and play a significant role in the general attempt to decrease the amount of plastic in the environment.

1.4.4. Unconventional methods

In addition to mainstream technologies of PET biodegradation, there are a number of unconventional or emerging technologies that have started to emerge. Such approaches tend to be beyond established enzymatic or microbial approaches and utilize experimental methods that cross several scientific fields. Although most of them are in a developmental stage (early stage or proof-of-concept), they witness the diversity and creativity enjoyed by PET biodegradation research. Possible examples are the use of artificial digestive conditions to emulate the in vivo conditions of degradation, the use of surface display technology to enhance the interaction of PET, and the modeling of the metabolic pathways in silico to engineer artificial microbial networks. Other initiatives look at marine microbial consortia, genetic circuit design, or even degradation environments mediated through biofilms. These methods tend to include synthetic biology, systems biology and environmental microbiology, a notion illustrating the interdisciplinary nature of this research area.

Though their application in practice is not still wide, such alternative approaches provide useful information and present new ways of understanding and thinking that could be applied in addition to or in strengthening common approaches. With the development of research, they can be scaled up to be solutions or be merged into hybrid PET biodegradation formats.

1.5. Objective & hypothesis of the essay

1.5.1. Objective

The primary objective of this thesis is to conduct a systematic literature review that critically evaluates and compares enzyme-only, microbial-only, combined, and other biological approaches to PET biodegradation in terms of degradation efficiency, biochemical mechanisms, and technological advancements. The review systematically analyzes experimental peer-reviewed studies to map key biochemical pathways, assess applied strategies, and identify performance trends. Findings are compiled to highlight innovations that enhance biodegradation efficiency, while also documenting current limitations. By consolidating this evidence, the study provides a comprehensive understanding of PET biodegradation, identifies knowledge gaps, and outlines directions for future research and industrial application.

1.5.2. Hypothesis

This thesis hypothesizes that biological methods can degrade PET more sustainably, efficiently, and at greater scale than conventional recycling approaches, as they operate under mild biochemical conditions. Through advanced biotechnological innovations and enzyme engineering, these approaches are expected to achieve higher depolymerization efficiency, enable high-purity monomer recovery, and address the shortcomings of mechanical and chemical recycling. Consequently, biologically driven PET degradation has the potential to deliver more sustainable waste management solutions by reducing environmental impact, lowering energy demands, and advancing a true circular economy for plastics.

2. Methodology

A comprehensive systematic literature search was conducted using the PubMed database to find peer-reviewed research studies on the biodegradation of PET using the PRISMA (Preferred Reporting Items for Systematic reviews and Meta-Analyses) guidelines.

2.1. Literature search

A structured search strategy was designed to focus specifically on PET biodegradation, incorporating a range of biological approaches. The following search query was applied in PubMed:

((Polyethylene Terephthalate[Title/Abstract]) OR (Terephthalate[Title/Abstract]) OR (TPA[Title/Abstract])) AND ((Bacteri*[Title/Abstract]) OR (Prokaryot*[Title/Abstract]) OR (Actinomyce*[Title/Abstract]) OR (*Pseudomonas*[Title/Abstract]) OR (*Rhodococcus*[Title/Abstract]) OR (*Bacillus*[Title/Abstract]) OR (*Comamonas*[Title/Abstract]) OR (*Ideonella*[Title/Abstract])) AND ((Biodeg*[Title/Abstract]) OR (Bioreme*[Title/Abstract]) OR (Recycl*[Title/Abstract])) NOT (Review[Publication Type])

To enhance coverage, a secondary manual screening of reference lists from relevant review papers was also conducted. The initial search retrieved 336 research articles.

2.2. Screening and study selection

A two-step screening procedure was used to identify all records: first, titles and abstracts were reviewed, and then the complete text was thoroughly examined to ascertain eligibility. Three records (two non-English publications (only in Chinese) and one study awaiting peer review) were excluded initially before the first screening started, leaving 333 records for additional evaluation.

181 studies were subsequently excluded based on the following reasons:

- Focusing exclusively on PBAT (n = 49)
- Addressing non-PET polymers (n = 71)
- Studies solely on non-biodegradation or chemical or physical degradation (n = 35)
- Computational or modeling-only studies (n = 13)
- Other studies misaligned with biodegradation criteria (n = 13)

Ultimately, 152 studies met all criteria and were included for comprehensive analysis, as summarized in the attached PRISMA flow diagram **(Figure 3)**.

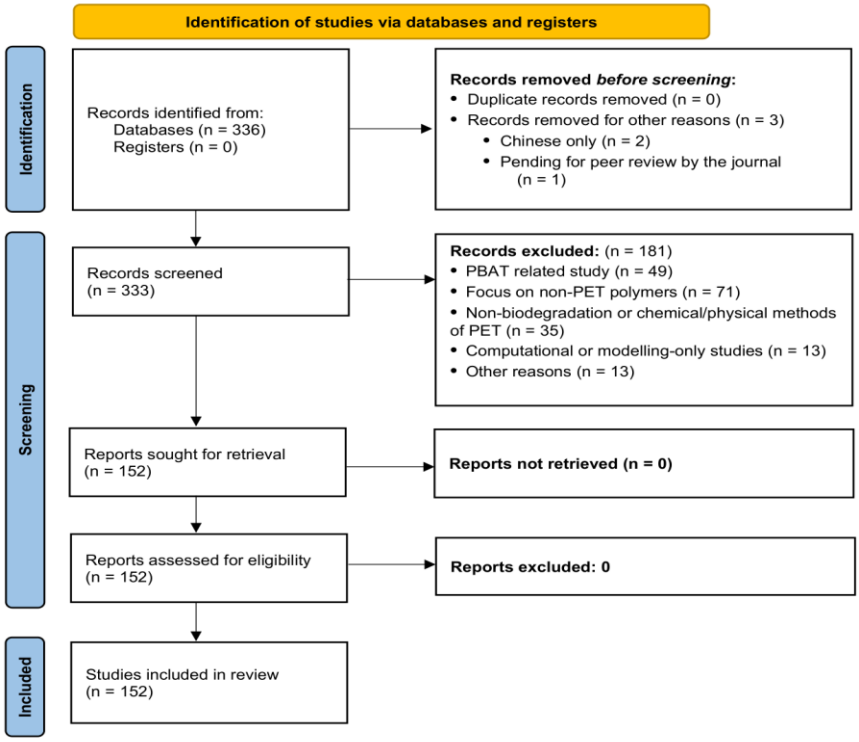


Figure 3. PRISMA diagram outlining the selection process of 152 PET biodegradation studies included in this study.

3. Results

3.1. Systematic categorization and pathway selection

The collected dataset of 152 PET biodegradation studies was systematically categorized to facilitate detailed and structured analysis aligned with the objectives of this review. The studies were divided into four primary groups: degradation method, degradation efficiency, type of plastic, and source of plastic (**Figure 4**).

The classification by type of plastic revealed that most studies (n = 125) concentrated on PET, which comes in a variety of forms, including amorphous, film, crystalline, bio-based, and textile fibers. While some research (n = 17) concentrated on PET degradation products, such as mix monomers (3), oligomers (1), TPA (3), BHET (2), MHET (2), and terephthalate (6). A smaller number of studies included mixed plastics alongside PET (n = 9). Only one study involved other plastics entirely (n = 1).

In the grouping by degradation method, studies were classified as enzymatic-only approaches (n = 66), microbial-only approaches (n = 45), combined approaches (n = 29), and other or unconventional methods (n = 12). PETase variants (37), MHETase variants (2), dual-enzyme systems (3), Cutinase/LCC variants (9), mixed enzyme approaches (10), and other types (5) were among the enzymatic-only studies. Microbial-only studies included work on *Ideonella sakaiensis* (4), single-species bacteria (20), defined co-cultures and mixed isolates (12), environmental microbial aspects (8), and other microbial approaches (1). Combined approaches integrated enzyme-microorganism systems (23), chemical-biological systems (4), and other combinations (2). The unconventional methods group encompassed innovative or non-traditional approaches (12).

For degradation efficiency, the studies were grouped into high efficiency (≥80-90% conversion or complete degradation, n = 16), moderate efficiency (50-80%, n = 7), low efficiency (20-50%, n = 9), and very low efficiency (<20%, n = 23). Among high-efficiency studies, degradation time ranged from less than a day (4) to 1-7 days (5), over a week (4), and more than a month (3). Additionally, several studies focused on degradation by-products and intermediates (n = 20), degradation of such intermediates (n = 17), and changes in polymer characteristics (n = 17). Some studies employed comparative methods (n = 27), while others did not report efficiency or key degradation parameters clearly (n = 16).

In terms of the source of plastic used in research, 78 studies used virgin PET, 28 used recycled PET, and 18 utilized environmental samples. The environmental samples were also geographically categorized as Asia (10), Africa (1), North America (1), South America (3), Europe (1), Pacific Ocean (1), and global (1). Six studies investigated two or more plastic source types, while 22 studies fell into other categories such as computational studies, non-PET-related research, or studies that did not specify the plastic source.

A broader and clearer understanding of the prevalent approaches, effectiveness, and kinds of plastic materials examined was made possible by this thorough classification, which offered a solid basis for the methodical comparison and interpretation of PET biodegradation research. As the 'grouping by degradation method' provides an ideal framework for comparing enzymatic, microbiological, mixed, and unconventional strategies applied to PET biodegradation, I have chosen to use it to structure and analyze the collected papers for the purposes of this review.

To ensure systematic extraction and organization of findings across all studies, structured Tables 2–22 (Appendix) were created using uniform columns because they capture each study's PMID,

key findings, type and source of plastic, study location, degradation method, used enzyme or microorganism, degradation efficiency, time required, environmental impact, and reported limitations. This structure ensures a clear as well as comparable presentation. Data for PET biodegradation is presented consistently in all ways.

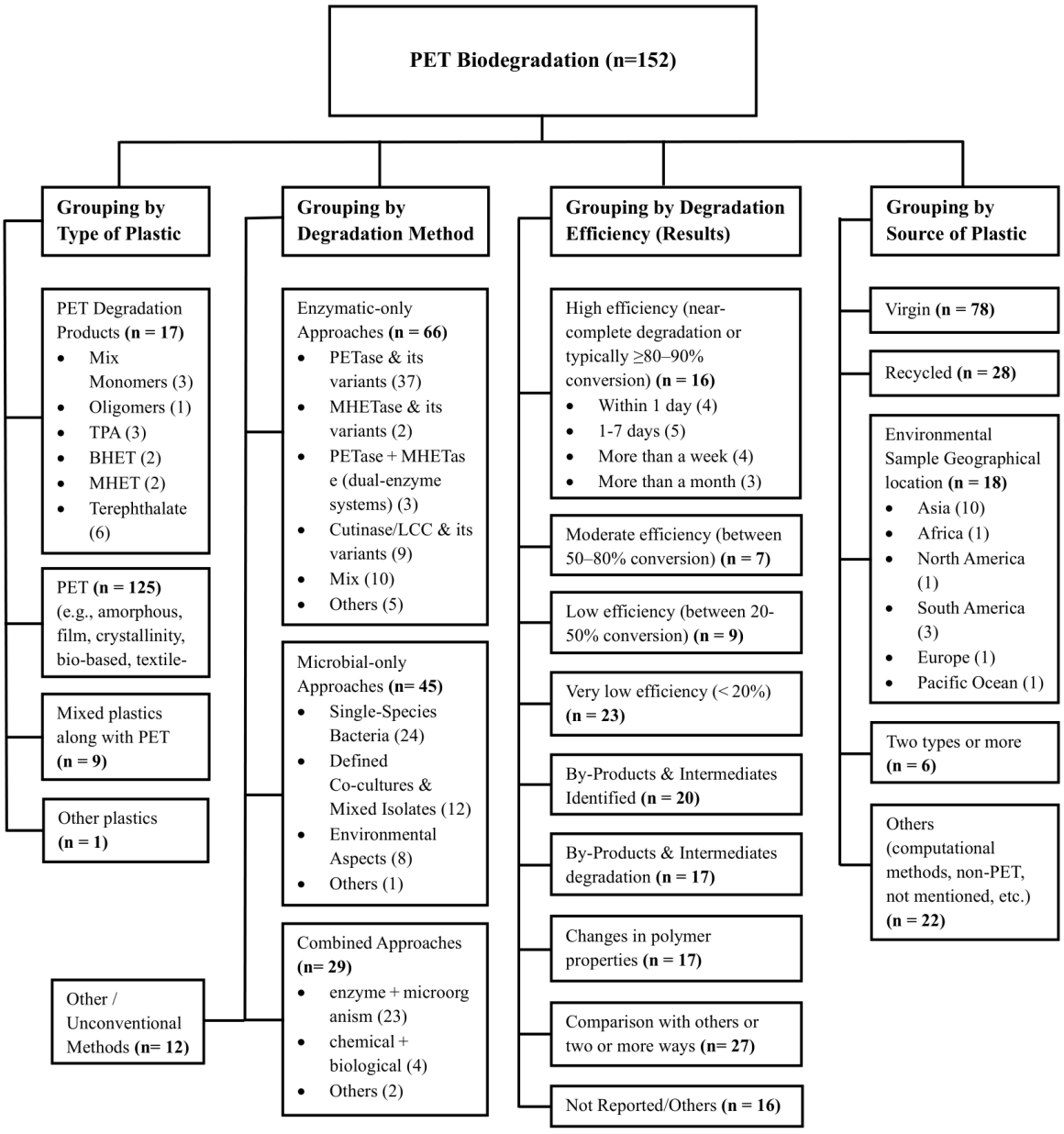


Figure 4. Categorization of 152 PET biodegradation studies by type of plastic, degradation method, degradation efficiency, and source of plastic.

3.2. Enzymatic-only approaches findings

3.2.1. PETase & its variants

3.2.1.1. Common findings

Across the 37 studies on PETase from *Ideonella sakaiensis* and its engineered variants, a consistent pattern was the higher degradability of amorphous or low-crystallinity PET under mild reaction conditions compared to high-crystallinity forms. For example, IsPETaseMut acting on recycled PET bottles (~50 mg, low crystallinity) achieved ~19.6 mg weight loss (~39%) over 2 weeks (16). TurboPETase also acted effectively on recycled PET flakes (17). Virgin PET powder was degraded over a 4-week period, with TPA release reflecting more accessible low-crystallinity regions (18).

Mutations to PETase (engineered variants) consistently enhanced PET hydrolysis. For instance, wild-type PETase gets "overloaded" and slows down at high amounts under certain pH (pH 9), but HotPETase keeps working steadily (19). TurboPETase, an engineered redesign, efficiently degrading for 98.20% recycled PET flakes in 8 h (17). PETaseR280A expressed in a marine diatom demonstrated greater depolymerization rates (~80 fold higher) than the unmodified enzyme on PET substrates (20). At 30 °C, engineered TSP-PETase exhibited a 30% increase in PET degradation relative to the wild-type (21). The W159H/S238F double mutant showed on amorphous PET film, achieved ~0.5-1.5%/h product release, with up to ~5× higher conversion on crystalline powder than amorphous PET (22). Thermostability engineering further boosted performance: thermostable PETase variants such as the triple mutant (TM), TM1 (IsPETaseTMK95N/F201I), TM3 sustained rapid hydrolysis (e.g., nanoparticles in ~24 h) (23);

HLCB (HP-BaCBM fusion) improved PET hydrolysis by ~25.5% over HotPETase after 50 h (24). Structural analysis showed PETase degrades PET via nick generation and terminal digestion, with the R280A variant increasing degradation efficiency by 22.4% in 18 h and 32.4% in 36 h (25). Improvements reported for additional engineered constructs further confirmed the trend of enhanced performance via active-site and stability mutations (24) (26).

Surface display and immobilization improve PETase stability, reuse, and activity retention. PETase co-displayed with hydrophobin (from *Trichoderma reesei*) degraded low-crystallinity PET within 18 h at 30 °C and high crystallinity PET within 10 days (27) and released TPA and MHET. Yeast (*Pichia pastoris*) surface-displayed PETase with glycosylphosphatidylinositol (GPI) anchors achieved ~36× higher hcPET degradation than purified PETase, retaining 85% activity after three and 50% after seven cycles (28). Another *P. pastoris* surface-displayed PETase degraded 55% in 18 h at 30 °C (29). In another study, hydrophobin (RolA and HGFI) pretreatment enhanced binding, enabling ~34% weight loss in 5 days under mild conditions (30). Functionally expressed in the green alga *Chlamydomonas reinhardtii* (using strain CC-124) PETase acted on virgin PET powder and film for 4 weeks (18).

Across studies, PETase-mediated hydrolysis predominantly released monomeric products in some cases. Hydrophobin pretreatment increased MHET and but very low in TPA yields under mild conditions (30). Expressed-PETase hydrolyzed virgin PET powder to release 9.12 mg of TPA from 30 mg of PET (conversion rate of 35.17%) (18). At 55 °C, engineered TSP-PETase produced 2.4 mM combined TPA and MHET in 105 min at 28.8 g/L PET (21). BhrPETase from bacterium HR29, produced 0.17 mM BHET, 3.66 mM MHET, and 2.47 mM TPA within 20 h (31). PETase treatment yielded 2.3 mM MHET and 1.3 mM TPA after 96 h (32). Fusion of a cellulose-binding domain to IsPETaseEHA increased total product formation from 17.9 to 174.1 μM (71.5%

increase) and from 30.7 to 251.5 μM at 40 °C (44.5% increase) with CBM fusion (26). Also in one study, D-HLCB (surface-displayed form) reached 1.03 mM monomeric products versus ~271.5 μM for HP alone in 71 h (24).

3.2.1.2. Differences

Different PET crystallinity, physical forms, and sources strongly influenced degradation efficiency. In a co-display system, PETase hydrolyzed 55% lcPET, whereas just 10.9% of hcPET under the same conditions (27). The TS-ΔIsPET variant degraded PET microparticles (~30 μm) with 26% yield, but 80% for nanoparticles (~80 nm) where reaction conditions varied with particle morphology (33). Wild-type PETase degraded ~15% crystallinity film less efficiently than engineered variants (34).

Thermal stability varied among PETase studies. Many enzymes operated optimally at ~30 °C, but several retained activities at higher temperatures. For instance, engineered versions performed well at 40 °C (22), 45 °C (33) and 50 °C (35). Interestingly, a few were stable at 55 °C (21) and even 60 °C (23). Improved stability beyond mesophilic ranges was also noted with the report of elevated-temperature stability (50 °C) (36). Reaction times for PETase and its variants varied widely, though many studies reported optimal degradation within 3-5 days or shorter. Longer durations were observed for certain systems, extending to 2 weeks (16) or several weeks (20) (37). Multiple studies reported dual timing linked to substrate type or crystallinity -examples include 18 h for lcPET vs. 10 days for high-crystallinity (27), 1.5 h for nanoparticles vs. 2 days for microparticles (33), 1 day vs. several days depending on setup (38) and 59.2% higher product yield in 18 h vs. ~27.7% higher after 5 days with PcAA14A compared to IsPETaseEHA alone (39). High-performance enzymes also achieved rapid hydrolysis in hours (21) (23) (31).

Engineered and newly discovered PET hydrolases generally outperformed wild-type PETase under comparable conditions. Classic benchmarks show modest wild-type activity improved by point mutations: on ~15%-crystallinity PET film, wild type gave ~10% crystallinity loss in 96 h at 30 °C, while S238F/W159H reached ~14% (34); IsPETaseMut fusion with NusA protein achieved ~1.4× wild type (16). Also, IsPETase I139R degraded ~3.6× more PET than wild type in 72 h at 40-50 °C (35), W159H/F229Y increased activity by nearly 40-fold (38), and rational designs showed 3.3-fold higher degradation (40) (41). While cyclized cPETase achieved ~10-fold rate increases with enhanced stability (42). Novel enzymes broadened the landscape: BbPETase was active within 1-3 days and shows 3.5 higher fold (43), SbPETase from *Schlegelella brevitalea* showed ~8% higher PET film degradation than wild-type IsPETase (44), and additional hydrolases plus engineered variants showed measurable conversion (3.1-fold faster) even over longer incubations of 30 days (45). In addition, a novel PETase, EstB from *Pseudomonas sp.* (37), demonstrated PET degradation over a 24-48 day incubation.

Several studies reported atypical product distributions and partial hydrolysis, alongside computation-only analyses. Experimental papers showed shifts in product balance or incomplete conversion under specific setups: rationally engineered variant S136EPETase produces MHET which is 5× more than TPA (40), ancestral sequence reconstruction variants yielded less TPA at 30 °C but over 2× higher turnover at 60 °C (46), and active-site flexibility from a novel disulfide bond enabled PETase to achieve 5-120× higher PET film depolymerization at 30 °C than thermophilic hydrolases (47). By contrast, under higher-temperature operation, TPA became the dominant product after 2-3 days, indicating more complete depolymerization (35) (36). Molecular-dynamics binding and selectivity analysis (48) (49), PETase conformational mapping and mutations (50) (51), and in-silico mutational hotspot detection (52) constitute a separate body of

work that is purely computational. These experiments support but do not complement the lab data by giving mechanistic understanding and product-forming predisposition prediction but cannot give experimental product ratios or yields. **Table 2** presents detailed information on the referenced articles.

3.2.2. MHETase & its variants

3.2.2.1. Common findings

Both studies reported heterologous expression of *Ideonella sakaiensis*'s MHETase. One investigation (53) cloned the MHETase gene into an expression vector, transformed *E. coli* BL21 (DE3), and confirmed protein expression by SDS-PAGE and activity assay, using recycled PET and virgin PET films as substrates. The other study (54) used a yeast whole-cell biocatalyst (surface display) with MHET derived from virgin PET and achieved nearly 100% conversion of MHET to TPA in exactly 24 hours.

3.2.2.2. Differences

The host systems, substrates, and quantification methods varied. In the *E. coli* study (53), MHETase was expressed in a bacterial system using recycled PET and virgin PET films, but no time-course data or degradation efficiency percentage was reported. In contrast, the yeast display study (54) used MHET from virgin PET and measured ~100% conversion in 24 hours. **Table 3** has more detailed information about these articles.

3.2.3. PETase + MHETase (dual-enzyme systems)

3.2.3.1. Common findings

Three studies investigated PET depolymerization using a combination of PETase and MHETase. In the first study (55), PET biodegradation was achieved through a chimeric PETase-MHETase with a 12-residue linker, producing intermediates over 96 hours, with efficiency dependent on PET crystallinity and substrate accessibility. The second study (56) presented a dual-method analytical strategy using PETase and MHETase to quantify hydrolysis products, reporting ~30 μM MHET, 10 μM TPA and 2 μM BHET after 4 hours. The third study (57) used a thermostable PETase triple mutant (S121E/D186H/R280A) in combination with IsMHETaseSM (W397A) to depolymerize PET, achieving a maximum of 70% depolymerization in 1 hour at 50 °C.

3.2.3.2. Differences

The first study (55) optimized a chimeric PETase-MHETase construct and reported results after 96 hours, while the second (56) focused on analytical quantification using NMR. The third (57) employed a thermostable PETase triple mutant with MHETase variant to achieve rapid depolymerization of virgin PET. **Table 4** provides detailed information about these three article.

3.2.4. Cutinase/LCC & its variants

3.2.4.1. Common findings

Across the nine studies, cutinase enzymes - particularly thermostable and engineered variants such as LCC-ICCG, ICCG DAQI, ICCM, MtCut, and TfCut2 - consistently demonstrated high PET depolymerization under elevated temperatures. In one case, LCC-ICCG when fused with PelB secretion signal, achieved ~100% degradation of amorphous PET and extruded PET in 3 days at 65 °C, with ~80% degradation for recycled PET (58) and in another study, achieved 100% depolymerization of 20 g post-consumer PET flakes within 10 hours at 72 °C (59). Another engineered variant, ICCG DAQI, released 97.8% TPA and 98.6% EG from PET textile fibers in

18-20 hours (60). ICCM cutinase expressed in *E. coli* degraded PET films by ~100% within 4 days at 60 °C, pH 10 (61), while MtCut - a marine metagenome-derived cutinase - reached nearly complete conversion in 60-72 hours (62). Tfuc2-LCI, a thermostable variant of *Thermobifida fusca* cutinase fused with an anchor peptide, degraded 66.7% of PET substrate, compared to ~37% by Tfuc2 alone for 4 days at 65°C (63), while TfCut2 double mutant G62A/F209A, combined with a cationic surfactant (30 ppm), degraded complete lcPET in 30 hours at 40-70 °C, pH 7-9 (64). Temperature effects were evident; for example, ICCG produced 6.99% weight loss at 70 °C compared to 3.65% at 37 °C over 8 days (65). Several studies used recycled PET or virgin PET, and pretreatment methods such as amorphization were noted to improve enzymatic accessibility.

3.2.4.2. Differences

Enzyme type, substrate form, and performance varied across the studies. TfCut2 required a cationic surfactant to achieve high degradation levels (64), whereas LCC-ICCG and its variants (including ICCG DAQI) consistently reached near complete depolymerization in ≤4 days (58) (59) (60) (61). Reaction times ranged from 18-20 hours (ICCG DAQI) to 8 days (ICCG), with optimal degradation often occurring between 60-72 °C. Some studies reported degradation as percentage PET conversion (58) (64) (65), while others quantified monomers release (60) (61) (62). One study (66) diverged from experimental methods by using only computational modeling (docking and molecular dynamics) to predict FoCut cutinase-PET binding. More detailed information of these articles are provided in **Table 5**.

3.2.5. Mix

3.2.5.1. Common findings

Ten research studies used mixed enzyme combinations to approach PET biodegradation; PETase was frequently paired with complementing enzymes such MHETase, LCC, or new hydrolases. These combinations outperformed single-enzyme setups in a number of instances in terms of depolymerization efficiency. Examples include dual enzyme system with LCC + immobilized TfCa, which improved PET degradation (2.4× higher) over TfCut2 alone within 24 hours (67), and BMHETase6M, an engineered variant with 8-fold higher activity for MHET hydrolysis and 2-fold increase for BHET release compared to wild-type BMHETase when incubated for 24 hours at 55 °C and pH 8.0 (68). Mors1 degraded amorphous PET films to 2.5% in 10 days at 25 °C (69), while MoPE hydrolyzed semi-crystalline PET powder to ~1.3 μM TPA in ~3 days (70). Engineered bacterial systems included *Pseudomonas putida* KT2440, which showed ~1.5% weight loss in 6 days on virgin commercial PET film (71), and a bacterial extracellular recombinant system (BECRS) co-expressing FAST-PETase and MHETase that released up 14.6 mM product release from amorphous PET film in 7 days (72). Additionally, FAST-PETase + MHETase achieved 71.3% weight loss of 200-mesh PET powder in 24 hours and up to 84.6% when co-displaying FAST-PETase and MHETase (73). PC enzyme complex (cPETase-chimeric PETase from *Ideonella sakaiensis* + cCALB-chimeric *Candida antarctica* lipase B + CBM3-scaffold assembled via cohesin-dockerin interactions) partially degraded crystalline PET, waste PET and BHET at 30 °C over 4 days (74), while disulfide bridge engineering in BbPETaseCD improved activity by ~11-fold over 14 days at 40 °C (75). Computational work included QM/MM modeling of LCC-ICCG and PETase oligomer hydrolysis (76) to probe catalytic pathways.

3.2.5.2. Differences

Although all studies aimed to improve PET degradation, their strategies varied. Dual-enzyme synergy was evident in systems like dual enzyme system + immobilized TfCa (67) and

BMHETase6M (68). Reaction conditions varied widely, from 24-hour incubations (67) (68) (73) to 14 days (75). Quantification methods included weight loss (71) (73) and monomers release (70) (72). Substrate sources ranged from virgin PET (69) (70) (72) and recycled PET (68) to environmental PET waste (74). Computational-only studies (76) provided mechanistic insight but no direct degradation data. **Table 6** provides more details pertaining to the articles discussed in this section.

3.2.6. Others

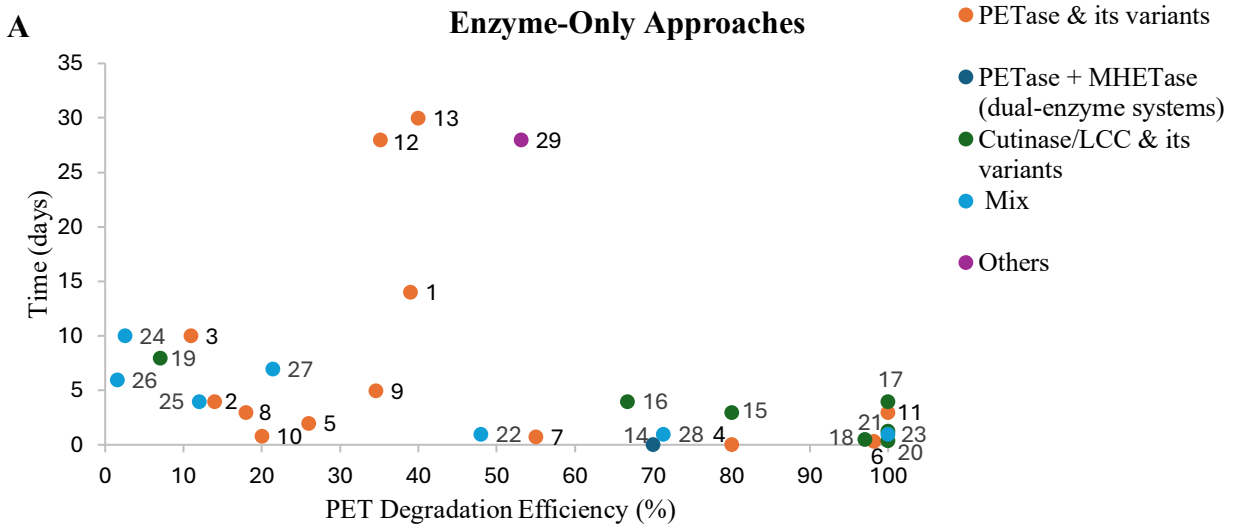
3.2.6.1. Common findings

The five studies in this category explored PET degradation through less conventional enzymes, hosts, or approaches compared to the main categories. A putative PET hydrolase from *Staphylococcus epidermidis* released MHET and TPA from BHET but did not directly degrade PET polymer, showing activity after 24 hours at pH 8 and 37 °C (77). Screening work on family IV polyesterases identified GEN0105 as a notable variant, with activity detectable in 12-hour reactions on virgin PET using HPLC analysis (78). A cold-active esterase, PET30 from *Kaistella jeonii*, demonstrated potential for PET bioremediation, although catalytic efficiency was lower than that of *Ideonella sakaiensis* PETase (79). An engineered *Yarrowia lipolytica* strain expressing PETase degraded virgin PET (44.7%) over 7-10 days, with increased activity (53.1%) when supplemented with 1% olive oil (80). Structural and mutational analysis of Ple629 led to a D226A/S279A double mutant with a 5.5-fold improvement in PET hydrolysis over 24 hours (81).

3.2.6.2. Differences

The studies varied in enzyme source, PET form, and experimental aims. The *Staphylococcus epidermidis* hydrolase (77) targeted BHET rather than intact PET, whereas other tested activity directly on PET. Only the engineered *Y. lipolytica* PETase strain (80) used a whole-cell yeast system for PET powder and flake degradation, while Ple629 engineering (81) focused on structure-guided mutagenesis to enhance activity. Reaction times ranged from short incubations of 12-24 hours (78) (81) to multi-day processes (80). Reported limitations included lack of direct PET polymer testing (77), weak-to-moderate activity in many family IV enzymes (78), low catalytic efficiency compared to benchmark PETase (79), dependence on additives for optimal yeast activity (80). More information of articles mentioned in this section is provided in **Table 7**.

Using the collected data, a consolidated scatter plot and table **(Figure 5)** were made to show the differences in PET breakdown efficiency between enzyme-based methods. Degradation efficiency (%) is plotted against reaction time (days) for PETase variants, dual-enzyme formulations, cutinases, mix variants, and other enzymes. A unique experimental investigation using a particular enzyme or combination of enzymes is represented by each data point. Importantly, in order to guarantee data integrity and comparability, the figure only included research that specifically reported or permitted the calculation of PET degradation efficiency (%).



B

No.	Approach	Used Main (best-performing) Variant	PMID	Degradation Efficiency (%)	Time (days)
1	PETase & its variants	NusA-IsPETaseMut	35577005	39	14
2		Double mutant PETase S238F/W159H	29666242	14	4
3		Codisplayed PETase (+ HFBI hydrophobin)	36414665	10.9	10
4		TS-ΔIsPET (W159H/F238A/ S121E/D186H thermostable variant)	35008691	80(~80 nm)	0.0625
5				26(~300 μm)	2
6		TurboPETase	38360963	98.2	0.33
7		Codisplay (IsPETase + HFBI)	33579416	55	0.75
8		W159H/F229Y (double mutant)	33753197	18	3
9		IsPETaseWT	33561457	34.6	5
10		BhrPETase	33375975	20	0.83
11		Partial-deglyco Fast-PETase-Pp	36639437	100	3
12		Wild-type PETase	32345276	35.17	28
13		Wild-type ScPETase	38749251	40	30
14	PETase+MHETase (dual-enzyme systems)	IsPETaseTM (triple mutant S121E/D186H/R280A), IsMHETaseSM (single mutant W397A)	37350932	70	0.042
15	Cutinase/LCC & its variants	PelB-LCC	37594123	80	3
16		Tfuc2-LCI (engineered fusion of Tfuc2)	39653629	66.7	4
17		ICCM cutinase	38944965	100	4
18		ICCG DAQI	39390488	97	0.5
19		ICCG (engineered LCC variant)	35039943	7	8
20		LCC-ICCG	32269349	100	0.42
21		TfCut2 G62A/F209A double mutant	31690819	100	1.25
22	Mix	Polyester hydrolase (TfCut2/LCC) + immobilized TfCa	27214855	48	1
23		LCC-ICCG + BMHETase6M	39357610	100	1
24		Mors1 (from Moraxella sp.) and OaCut (from Oleispira antarctica)	34705547	2.5	10
25		PC enzyme complex = cPETase + cCALB + CBM3-scaffold	35753492	12	4
26		LCC, HiC (Humicola insolens cutinase)	35717313	1.5	6
27		BECRS = FAST-PETase + CBM3 surface display + intracellular MHETase	38703683	21.4	7
28		FAST-PETase + MHETase	37804764	71.3	1
29	Others	IsPETase (wild type) expressed in Y. lipolytica AJD2 strain	35850328	53.1	28

Figure 5. PET biodegradation efficiency and enzymatic variants in enzyme-only degradation systems. **(A)** A scatter plot showing the relationship between PET biodegradation efficiency (%) and time (days) for enzyme-only PET degradation studies. Each dot represents a distinct study using specific enzymes or combinations, with color-coded categories representing PETase and its variants, dual-enzyme systems (PETase+MHETase), cutinases and its variants, mix and other engineered enzymes. **(B)** Tabulated summary of the enzyme variants corresponding to each data point in **Figure 5(A)**, listing the exact variant used, PubMed ID (PMID), degradation efficiency (%), and reaction time in days.

3.3. Microbial-only approaches findings

3.3.1. Single-species bacteria

3.3.1.1. *Ideonella sakaiensis*

3.3.1.1.1. Common findings

Four studies investigated *Ideonella sakaiensis* for PET degradation using microorganism-based systems rather than purified enzymes. The wild-type strain 201-F6 was tested under both aerobic and anaerobic conditions, with PET powder, PET film, virgin PET granules, or environmental PET as the sole carbon sources. The role of PETase in facilitating polymer breakdown was evident in one study (82), where the wild-type strain degraded roughly 8% of PET film in 10 days, whereas the ΔPETase mutant displayed 20% of the degradation capacity of the wild-type strain. Another study (83) showed that *I. sakaiensis* degraded 75% of PET film in 30 days under anaerobic fermentation conditions. Regarding bioconversion, one study team degraded 7.9 g/L (~79%) of virgin PET granules and also converted 9% of PET-derived carbon into polyhydroxyalkanoate (PHA) over the course of a 6-day fermentation (84). Furthermore, transcript-level analyses (85) demonstrated that the presence of PET increased expression of genes involved in the transport and degradation of MHET and TPA, demonstrating microbial digestion of PET monomers even in the absence of measured mass loss data.

3.3.1.1.2. Differences

These four studies differed greatly in terms of substrate type, culture conditions, degradation endpoints, and timeframes. For instance, PET film in a controlled lab environment demonstrated low degradation efficiency (8% in 10 days) (82). In contrast, PET film used in anaerobic conditions resulted in higher degradation (75% in 30 days) (83). Virgin PET granules were employed in the

biopolymer manufacturing study (84), which focused on product conversion. One study (85), on the other hand, shifted from quantifying physical degradation to gene expression profiling, validating the expression of TPA transporter and MHETase but not finding real substrate breakdown. The timing also varied significantly. Degradation was recorded by some systems in as short as 6 days and by others in as long as 30. Furthermore, the genetic background utilized was directly related to degradation performance: PETase-deficient knockout mutant was unable to degrade PET at all (82), demonstrating the importance of this enzyme even in microbial-only systems. Additional specific details on theses articles are included in **Table 8**.

3.3.1.2. *Bacillus strains*

3.3.1.2.1. Common findings

Five studies in this category investigated PET degradation by *Bacillus strains*, either isolated from environmental sources or tested in laboratory settings, and in many of these applications, detectable PET depolymerization or structural change was noted. PET was used in forms such as films and nanoparticles. In one study (86), an engineered *Bacillus subtilis* strain within an artificial consortium (Bs_PETase + Bs_MHETase + R. *jostii*) degraded 31.2% of PET film in 7 days. In another (87), co-inoculation of *Bacillus cereus* and *Bacillus subtilis* in soil led to a 14% reduction in PET's carbonyl index over 6 months. PET nanoparticle degradation by *Bacillus safensis* YX8 was observed by monomers release such as TPA (4. μM), MHET (3.3 μM), BHET (0.4 μM) over 15 days at 45 °C (88). Another study (89) identified *Bacillus muralis* from plastic-contaminated soil and confirmed its growth on PET-containing media, though no quantitative degradation data were reported.

3.3.1.2.2. Differences

The five investigations varied greatly in terms of strain selection, experimental methodology, PET form and degradation assessment, even though they were all from the same genus. The fastest degradation (31.2% PET film loss in 7 days) occurred with the engineered B. *subtilis* strain in a consortium (86). Also, pretreatment enhanced degradation. In one study, natural strains such as *Bacillus safensis P9* degraded PET film slowly (1.1%) without pretreatment, which increased to 1.9% after thermal (80 °C, 10 days) or sulfuric acid pretreatment (~1.7× improvement) for 28 days (90). PET nanoparticle breakdown was reported for *B. safensis YX8* at 45 °C in 15 days whereas PET film for 2 months with very low efficiency (88). Some studies (88) (89) presented only qualitative or indirect PET degradation evidence without exact weight-loss measurements. The choice of PET type (film versus nanoparticles) also influenced the degradation rates reported. **Table 9** has more detailed information regarding the articles discussed in this section.

3.3.1.3. *Comamonas testosteroni*

3.3.1.3.1. Common findings

Four of the five studies in this category tested PET degradation using whole-cell *Comamonas testosteroni* systems frequently in conjunction with additives or surface modification techniques, while one study focused solely on TPA mineralization without PET polymer. This bacterium was tested on PET in microplastic, microparticle, or film form, and in one instance, it degraded the monomer TPA completely. For example, when Tween-20 was used as a surfactant, *C. testosteroni* F6 degraded PET microplastics by 11.04% in 5 days; without Tween, degradation was 6.32% (91). In another study, PET microparticles (~7.3 μm) were reduced to ~1.58 μm after 48 h exposure to *C. testosteroni* F4, indicating significant particle size reduction without mass loss (92).

C. testosteroni strain 3a2 mineralized 107 ± 3 mg/L of TPA completely in 12 h, but no PET polymer was used (93). The engineered strain RW31 achieved 37.1% PET weight loss in 7 days in vitro, and 0.83% in vivo in 30 days (94). Additionally, PET weight loss of 5.67% in 28 days was recorded when *C. testosteroni* F4 was with renewal of bacterial cultures on Span-80 coated PET surfaces (95).

3.3.1.3.2. Differences

The studies varied substantially in degradation substrates, enhancement strategies, and evaluation metrics. While most studies used PET microplastics or films, one focused solely on TPA degradation which part of para-toluic acid (pTol) without PET polymer involvement (93). Degradation timelines ranged from 12 h (TPA mineralization) to 28 days (PET film degradation). Additives such as Tween-20 or Span-80 were employed to improve microbial adherence and substrate accessibility.

The engineered strain RW31 achieved the highest PET film degradation at 37.1% in 7 days (94), whereas native strains such as F6 and F4 showed lower efficiencies - 11.04% in 5 days and 5.67% in 28 days, respectively (91) (95). In one case (92), degradation was inferred from particle size reduction rather than direct mass loss. Studies performing both in vitro and in vivo tests (94) reported significantly lower efficiencies in vivo under real-world conditions. **Table 10** provides detailed information each article presented in this section.

3.3.1.4. *Rhodococcus species*

3.3.1.4.1. Common findings

Three studies used *Rhodococcus species* capable of breaking down PET polymer or its breakdown product in a lab setting. According to two investigations, *Rhodococcus erythropolis* MTCC 3951

entirely degraded 5 mM TPA in 14 hours (96) (97), however higher doses, such 120 mM, took 84 hours to degrade nearly completely (97). In one investigation, *Rhodococcus* IITR131 was found to degrade PET polymers, causing a 19.7% weight loss in PET films after 60 days at 30 °C (98).

3.3.1.4.2. Differences

The three studies differed mainly in their substrates, degradation timeframes, and microbial targets. Two studies targeted soluble TPA (5-120 mM), while one used intact PET films. Degradation of 5 mM TPA was consistently achieved within 10 h, whereas PET film degradation required 60 days to reach 19.7% weight loss. The PET polymer work used *Rhodococcus* IITR131, while TPA degradation was performed by R. *erythropolis* MTCC 3951. Only one study (96) applied an immobilization method (freeze-thaw treatment) to potentially improve reusability (~92% removal in cycle 10). More details regarding these articles are available in **Table 11**.

3.3.1.5. *Pseudomonas species*

Across the two studies in this category, *Pseudomonas species* were examined for their ability to degrade PET-related monomers, particularly TPA and its esters, rather than intact PET polymer. In the first study (99), *Pseudomonas putida* KT2440 underwent adaptive laboratory evolution combined with genetic modification, resulting in a 4.1-fold increase in growth rate on TPA, although PET film or microplastics were not tested and EG utilization remained significantly lower. In the second study (100), a mixed culture of *Pseudomonas fluorescens*, *Pseudomonas testosteroni*, and *Pseudomonas acidovorans* achieved complete degradation of synthetic terephthalate within 7 days including timing of growing on terephthalate (1-2 days). While both studies demonstrated strong TPA-associated degradation capabilities, neither provided data on the breakdown of intact PET polymer. **Table 12** contains further specific information on these articles.

3.3.1.6. Other single species

3.3.1.6.1. Common findings

The five studies categorized as “Other Single-Species” investigated various bacterial genera, such as *Brucella*, *Serratia*, *Gordonia*, *Paracoccus*, and a mixed anaerobic culture, assessing their ability to break down PET or PET-derived intermediates in laboratory settings. *Gordonia sp.* CN2K demonstrated the greatest efficiency in PET degradation, reaching a 40.43% reduction in PET weight after 45 days in mineral salt media with shaking incubation (101). Likewise, *Brucella* intermedia IITR130 resulted in a 26.06% decrease in PET sheet mass over 60 days (102), while *Serratia plymuthica* IV-11-34 reached ~7% biodegradation in 40 days, assessed through CO_2 evolution as a measure of PET degradation (103). A study focused on *Paracoccus pantotrophus* DSM 2944 and evaluated monomer utilization, noting full consumption of EG in 30 hours and approximately 20% conversion of TPA under optimal conditions (104), although the PET polymer was not evaluated. In a further study (105), the incorporation of PET into a mixed microbial community from seed sludge where *Thermotogae* (42.9%) & *Firmicutes* (49.6%) were dominant phyla and *Tepidanaerobacter syntrophicus* and *Clostridium_sensu_stricto_7* were potential degrader led to approximately 4.1% weight reduction of PET over 25 days.

3.3.1.6.2. Differences

The five studies differed greatly in bacterial types, degradation materials, and incubation periods. Materials included PET films, sheets, or monomers (TA and EG), with degradation evaluated via mass loss (101) (102), CO_2 emission (103), or monomer reduction (104). The degradation time varied from 30 hours for EG metabolism to 60 days for notable PET film degradation. Significantly, the highest polymer degradation efficiency (40.43%) was achieved with *Gordonia*

sp. CN2K, whereas other species like *Serratia* and *Tepidanaerobacter* reached lower efficiencies ranging from approximately 4-7%. Experimental configurations varied: *Paracoccus* was examined for monomer degradation, whereas *Brucella* and *Serratia* were studied with PET films in aerobic static or stirred cultures. **Table 13** offers more detailed information for all articles from this section.

3.3.2. Defined co-cultures & mixed isolates

3.3.2.1. Common findings

The studies in this category demonstrated that defined co-cultures or mixed microbial isolates can enhance PET degradation efficiency compared to single strains, sometimes incorporating fungal partners or sequential chemo-biological methods. Several co-cultures degraded PET intermediates such as BHET or TPA, while others depolymerized PET films or powders. For example, *Enterobacter sp.* HY1 with EstB esterase gene degraded 1000 mg/L BHET by 80.8% in 5 days (106), and *Pigmentiphaga kullae* CHJ604 using two dioxygenase enzymes: TphA1A2A3 (terephthalate 1,2-dioxygenase) and OphA1A2 (phthalate 4,5-dioxygenase) degraded 332.26 mg/L TPA in 5 days (107). An engineered co-culture of *Fusarium solani*, *P. aeruginosa* RBM21 and *Bacillus subtilis* grew for 14 days on PET as the sole carbon source, though without quantified PET mass loss (108). A hydrostatic pressure system with *Flavobacterium*, *Rhodococcus* and tphA3 reduced PET particle size by 366.62 μm (13.66%) in 30 days (109). Five *Comamonas testosteroni* strains degraded 100 mg/L TPA completely within 6-8 h, with the C8 strain having lowest time (6 hours) (110).

Environmental consortia also showed activity - Apatlaco river microbial communities (111), bacterial strains isolated from insect guts (112) metabolized PET and a consortium containing *Bacillus thuringiensis* C15, *Bacillus albus* PFYN01, *Pseudomonas sp.* B10, and two strains of

Pseudomonas sp. SWI36 (113) achieved ~3% PET weight loss after 8 weeks at 30 °C and completely converted BHET to TPA and EG, and *Bacillus sp.* HY-75, *Xanthomonas sp.* HY-74 which derived from insect guts degraded PET films by 1.44% and 1.57% in 6 weeks, respectively (112). A two-step process combining chemical glycolysis and microbial degradation by *Pseudomonas* (3 unidentified species) and *Bacillus* (2 unidentified species) achieved complete BHET breakdown in 1.5 h and continued PET depolymerization over 10 weeks with having maximum ~56.5% degradation including hydrolysis (2 weeks) (114). In vivo, a consortium containing *Sarcina aurantiaca*, *Bacillus subtilis*, *Aspergillus flavus* and *Aspergillus niger* degraded more PET with 28.78% efficiency in compared to single species in 60 days (115). *Rhizospheric* co-isolates *Priestia aryabhattai*, *Bacillus pseudomycoides* and *Bacillus pumilus* degraded PET sheets by 40%, 36%, 32% in 28 days and powder by 69%, 66%, 64% in 18 days respectively (116).

3.3.2.2. Differences

The studies varied in microbial combinations, degradation targets, and system setups. Some co-cultures targeted PET polymer degradation by weight loss, size reduction, or end-product detection, while others focused solely on monomers such as BHET or TPA. Depending on the system, degradation efficiencies varied from nearly total monomer breakdown to PET polymer weight loss, which was typically between less than 1% and 40%. Consortia of fungi and bacteria, such as *Fusarium solani* and *Bacillus subtilis*, demonstrated growth on PET without causing detectable PET loss (108). Laccase and esterase production occurred by degradation of PET by *Dermacoccus* and *Bacillus sp.* BS5 and caused 4.44% PET weight loss in 4 weeks (117). **Table 14** presents additional details on each article presented in this section.

3.3.3. Environmental aspects

3.3.3.1. Common findings

Strains isolated from environmentally significant ecosystems, including mangrove sediments, marine caves, deep-sea sediments, hot springs, and chemically worn environments, were used in all eight of the studies in this group to assess the microbial PET breakdown capacity. Measurable physical or chemical changes, such as surface erosion, mass loss, the release of monomers like MHET or TPA, and microbial growth under PET-dependent conditions, were used in many investigations to establish PET breakdown.

Bacillus cereus and *Bacillus gottheilii* isolated from mangrove sand demonstrated 6.6% and 3.0% PET weight loss, respectively, after a 40-day incubation period in one study (118). In the western Pacific Ocean, PET films were broken down by multiple marine bacteria with having 1.2-1.8% degradation rates (1.8% by *Nocardioides marinus*) in 30 days (119). This resulted in the production of degradation intermediates such as MHET and TPA as well as visible surface erosion. Similarly, after three weeks, *Pseudomonas chengduensis* BC1815, which was isolated from the Pacific Ocean, showed PET degradation and lost about 1.43% of its weight (120).

Using a qualitative agar-based approach, hot spring isolates from Peru (121) showed PET hydrolysis at 55-60 °C, as evidenced by the creation of a clear zone in under two days. Similarly, weight loss increased over the course of 90 days due to PET film degradation by marine *Bacillus* species AIIW2, rising from 0.42% after 15 days to 1.93% after 90 days (122). In another examination, PET weight loss ranged from 1.8% to 16.2% over more than 60 days by deep-sea sediment samples from Pacific Ocean where BC14 (dominated by *Alcanivorax*, *Pseudomonas*, *Nitratireductor*, *Thalassospira*) showed highest degradation (123). Although no precise

quantitative data was provided, microbial strain I18 from Portuguese marine cave samples (124) showed PET film deterioration during a 45-day period. After 136 hours, *Rhodococcus* spp.-dominated microbial communities in one study that used chemically deconstructed PET (125) demonstrated enhanced growth which subtly supported the utilization of monomers from PET-derived products.

3.3.3.2. Differences

The source ecosystems, bacterial strains, substrate types, degradation circumstances, and quantification techniques used in the investigations differed greatly. PET films, virgin sheets, PET that had been chemically broken down, and mineral salt medium formulations were among the substrates. While some investigations tracked by visible clear-zone creation (121), others utilized gravimetric weight loss (e.g., up to 16.2%) (123). Depending on the microbiological species and environmental adaptation, degradation times ranged from 2 days to 90 days (gradual PET weight loss).

While some isolates came from more moderate biological niches like mangroves or marine caves, others came from more extreme settings (such as thermophiles from hot springs and deep-sea bacteria). Three studies reported surface erosion or microbial growth without directly quantifying PET loss, hence not all of them gave degradation efficiency in exact percentage figures. Furthermore, in at least two cases (123) (125), microbial consortia or communities were evaluated; in the remaining cases, specified single-species isolates were used. Further information on the articles can be found in **Table 15**.

3.3.4. Other

One study in this category (126) employed metagenomic investigation of the gut microbiota of pigs, identifying the existence of microbial taxa that break down PET, such as Lacto*Bacillus*, *Bacteroides*, and *Clostridium*. Genes encoding putative PET hydrolases were found, suggesting the possibility of enzymatic hydrolysis of PET despite the lack of direct PET polymer breakdown tests. The study raises the possibility that pigs and other comparable gut microbiota could be new sources of PET-degrading microbes, with potential environmental implications for PET biodegradation techniques. However, the verification of actual deterioration performance is limited due to the lack of experimental PET weight loss data. **Table 16** gives additional article-related details.

With the compiled data, the combined scatter plot and supporting table (**Figure 6)** were made to demonstrate the comparison of PET degradation performance between diverse microbial degradation systems. The scatter plot **(Fig. 6A)** includes PET degradation efficiency (%) against reaction time (days), categorizing microbial strains into groups such as *Ideonella sakaiensis*, *Bacillus* species, *Comamonas testosteroni*, *Rhodococcus* strains, established co-cultures, environmental consortia and other microbial isolates. Each location on the graph corresponds to a single study employing a single microbial species or population under predetermined conditions. For consistency and comparison purposes, only those studies which reported PET degradation efficiency as measurable percentages were included in the graphical plot.

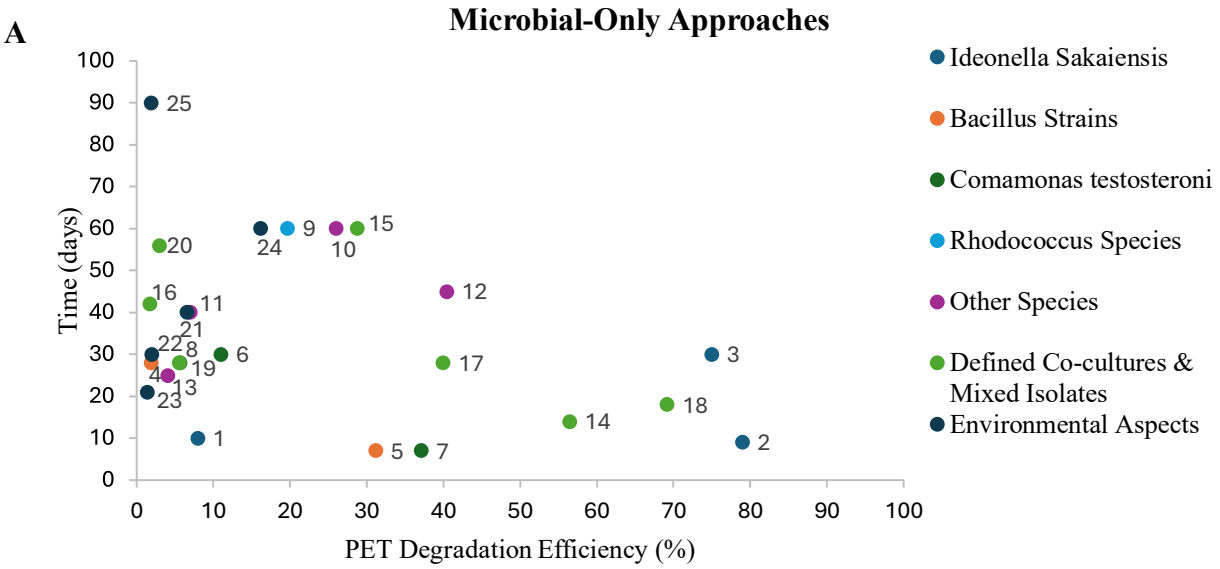


Figure 6. PET biodegradation efficiency and microbial strains in microbial-only degradation systems. **(A)** A scatter plot showing the relationship between PET biodegradation efficiency (%) and time (days) for microbial-only PET degradation studies. Each dot represents a specific study using specific microbial strains or communities, with color-coded categories representing *Ideonella sakaiensis*, *Bacillus strains*, *Comamonas testosteroni*, *Rhodococcus species*, other individual species, defined co-cultures and mixed isolates, and environmental consortia. **(B)** Tabulated overview of microbial strains and communities corresponding to **Fig. 6(A)**, listing the exact microbial variant used, PubMed ID (PMID), degradation efficiency (%), and experimental time (days).

B

No.	Approach	Used Main (best-performing) Variant	PMID	Degradation Efficiency (%)	Time (days)
1	*Ideonella sakaiensis*	*Ideonella sakaiensis* 201-F6 (parent strain DpyrF)	34260304	8	10
2		*Ideonella sakaiensis* 201-F6 (wild-type strain)	34620982	79	9
3		*Ideonella sakaiensis* 201-F6 (wild-type strain)	36103351	75	30
4	*Bacillus* Strains	*Bacillus* safensis strain P9	39379655	1.9	28
5		Engineered *Bacillus subtilis* Bs_PETase, engineered *Bacillus subtilis* Bs_MHETase (secretes MHETase)	35003008	31.2	7
6	*Comamonas testosteroni*	*Comamonas* testosterone strain F6	35293270	11.04	30
7		Engineered *Comamonas testosteroni* MO-19 MP expressing fusion enzyme FAST-PETase + IsMHETase (MPase)	39708916	37.1	7
8		*Comamonas testosteroni* F4	39708756	5.67	28
9	*Rhodococcus Species*	*Rhodococcus* rhodochrous IITR131 (wild-type strain)	39706601	19.7	60
10	Other species	Brucella intermedia IITR130	38459363	26.06	60
11		Serratia plymuthica IV-11-34	34678385	7	40
12		Gordonia sp. CN2K	38944007	40.43	45
13		Mixed thermophilic microbial community (Thermotogae, Firmicutes, etc.)	34320761	4.1	25
14	Defined Co-cultures & mixed isolates	3 Pseudomonas spp., 2 *Bacillus* spp.	38002377	56.5	14
15		Sarcina aurantiaca (TB3) + *Bacillus subtilis* (TB8) + Aspergillus flavus (STF1) + Aspergillus niger (STF2)	36228720	28.78	60
16		Xanthomonas sp. HY-74 (from Xylocopa appendiculata) *Bacillus* sp. HY-75 (from Eumenes decoratus)	37688172	1.78	42
17		Priestia aryabhattai VT3.12 *Bacillus* pseudomycoides VT3.15 *Bacillus* pumilus VT3.16	35460002	40(sheet)	28
18			35460002	69.2(powder)	18
19		*Bacillus* BS5	38519709	5.56	28
20		*Bacillus* thuringiensis strain C15 + *Bacillus* albus strain PFYN01 + Pseudomonas sp. B10 + Pseudomonas sp. SWI36 + Pseudomonas sp. SWI36	33361127	3	56
21	Environmental aspects	*Bacillus cereus* (mangrove sediment, Malaysia)	28964604	6.6	40
22		Nocardioides marinus BC14_2_R3 (Pacific Sea ~5000m)	37421171	2	30
23		Pseudomonas chengduensis BC1815 (deep-sea bacterium)	40252025	1.43	21
24		BC14 (Alcanivorax sp. A02-7, Pseudomonas sp. A09-2, Thalassospira marina, Nitratireductor sp.) (deep-sea)	38734049	16.2	60
25		Marine *Bacillus* sp. AIIW2 (isolated from plastic waste)	34646260	1.93	90

3.4. Combined approaches findings

3.4.1. Enzyme + microorganism

3.4.1.1. Common findings

Across 23 studies, combined enzymatic-microbial strategies consistently depolymerized PET films, powders, oligomers, or monomers under varied conditions. *Ideonella sakaiensis* 201-F6 featured prominently, achieving near-complete PET hydrolysis in 6 weeks at 30 °C via PETase and MHETase, with comparable performance in engineered consortia expressing both enzymes (127) (128).

Material properties strongly influenced outcomes. Using *I. sakaiensis*, transparent PET from food containers reached ~96% conversion in ~4 weeks, whereas colored PET required ~7 weeks for 63.9% degradation (129). Enhancing enzyme presentation also helped: hydrophobin-fused PETase delivered ~52% more TPA in 7 days than the non-displayed enzyme (130). Additionally, sequential microbial-enzyme workflows delivered high conversions. *Pseudomonas* pretreatment for 7 days followed by engineered cutinase ICCG gave 94.5% PET depolymerization in 6 days (131). Thermophilic whole-cell systems based on engineered *Clostridium thermocellum* reached ~62% PET-film loss over 14 days (132) and up to ~96.7% conversion in 10 days (133) with LCC enzyme. In compost-style settings, PET microplastics were reduced by 35% in 50 days with WCCG (26% without WCCG) (134). For PET oligomers, a co-culture produced PHA at 0.11 g per g oligomer in 72 h (135).

Multiple orthogonal readouts confirmed PET breakdown, including TPA/MHET/EG release, gravimetry, carbonyl-index changes, SEM, and clear zones. A two-step route using PES-H1 hydrolysis followed by *Pseudomonas putida* KT2440 conversion operated within 72 h with having

no quantitative data (136). A DuraPETase-Est_B system yielded 38.04 µM total products from PET microplastics over 30 days, and Est_B alone achieved complete BHET hydrolysis (0.23 mg/mL) in 4 h at 30 °C (137). Also, bio-upcycling appeared repeatedly. One study reported ~47% PET depolymerization within 10-24 h with downstream PHA/HAA bioplastics production (138). In another study, engineered *Pseudomonas sp.* JY-Q with FASTPETase-MHETase chimerase, BHET was fully hydrolyzed within 72 h, and PET films showed visible erosion after 28 days (139). A *Yarrowia lipolytica-Pseudomonas* co-culture released 0.31 g/L TPA from 10 g/L PET (3%) over ~9.5 days (140). Under prolonged incubation (90 days), PET oxidation was evident from a carbonyl-index rise to 5.35 (141).

Additional whole-cell routes complemented these findings. *Comamonas testosteroni* KF-1 induced biofragmentation of PET pellets/films with ~56% BHET release over up to 42 days (142). Pretreatment with *Stenotrophomonas pavanii* JWG-G1 drove ~91.4% PET-film weight loss in 14 days (2.73-fold higher than non-pretreated films) (143). Metagenomic/enzyme-annotation work identified relevant PET-active lineages and enzymes supporting these systems without any quantitative data (144) (145) (146). While not targeting PET polymer, *Pseudomonas aeruginosa* PP4/PPD rapidly degraded terephthalate/isophthalate (20-24 h) (147), and *Pseudomonas umsongensis* GO16 utilized TA and EG - useful for downstream assimilation after enzymatic depolymerization (148).

3.4.1.2. Differences

Performance varied widely across systems, substrates, and readouts. Reported durations ranged from 4 h for complete BHET hydrolysis with Est_B (137) to 90 days for PET-film aging with a carbonyl-index increase to 5.35 (141). *Piscinibacter sakaiensis* reduced PET weight by around 40-

50% in 5 weeks under optimal conditions (149). The structure of the substrate was important: transparent PET performed better than colored PET in similar circumstances, and amorphous or oligomeric PET generally breakdown more quickly than crystalline or bottle-grade materials (129). Process design also shaped outcomes. Pretreatment or sequential coupling improved overall conversion-e.g., *Pseudomonas nitroreducens* S8 + *P. monteilii* S17 pretreatment followed by ICCG hydrolysis (131) and *S. pavanii* JWG-G1 pretreatment giving a 2.73× increase (143).

Evidence types spanned monomer/intermediate release (TPA/MHET/EG), direct weight-loss measurements, and materials metrics such as the carbonyl index, capturing both chemical depolymerization and physical transformation of the PET matrix (141) (149). Host implementations were diverse - hydrophobin-based surface display in *E. coli* (130); thermophilic whole-cell catalysts (*C. thermocellum*) (132) (133); engineered *Pseudomonas sp.* JY-Q (139); and *Y. lipolytica* co-cultures (140). Studies covered films, pellets, and microplastics, with objectives ranging from fast monomer recovery to bio-upcycling (PHA/HAA) and compost-compatible processing, as reported across the dataset. **Table 17** includes more comprehensive information on the articles presented here.

3.4.2. Chemical + biological

3.4.2.1. Common findings

Across all four studies in this group, chemical pretreatment of PET - whether through ammonium hydroxide hydrolysis or deep eutectic solvent (DES) treatment - either produced soluble aromatic monomers such as TPA, which microbes could metabolize, or enhanced composting weight loss under DES conditions. The ammonium hydroxide procedure was used to convert PET into a "deconstructed PET" (DCPET) combination of TPA and monoamide. Within seven days,

microbial consortia (LS1_Calumet and LS2_Calumet), with chemical pretreatment of ammonium hydroxide (NH_4OH), broke down ~99.8% of monomers (150). In a similar vein, TPA uptake of ~1-2 g/L produced by chemically deconstructed PET (CDPET) from PET cups over a 48-hour period (151). When choline chloride: glycerol DES pretreatment was followed by bioaugmentation in composting systems, PET weight decreased by 22-23% in 42 days at the lab scale and by up to 25% in 65 days at the pilot scale (152). Although one article also emphasizes hybrid chemical-dual enzyme system pathways - which combine chemical pretreatment PETase and MHETase - for complete BHET conversion to TPA and EG (153).

3.4.2.2. Differences

The two-step chemical-and-biological process is the same for all experiments, but the pretreatment chemistry, substrate type, incubation conditions, and degradation metrics vary. The ammonium hydroxide pretreatment method achieved nearly total monomer release (150) by microbial consortia over 7 days. Unlike film mass-loss targets in compost, monomer consumption experiments on CDPET (151) measured ~1-2 g/L TPA uptake in 48 hours. The composting investigation highlighted the cumulative effect of chemical pretreatment by separating three bioremediation situations over the same time period: with DES + bioaugmentation (22-23% loss), with bioaugmentation alone (23% loss), and with natural attenuation (10% loss) (152). The review (153) notes near-complete BHET conversion but lacks standardized cross-study performance metrics. More specific information of the articles is summarized in **Table 18**.

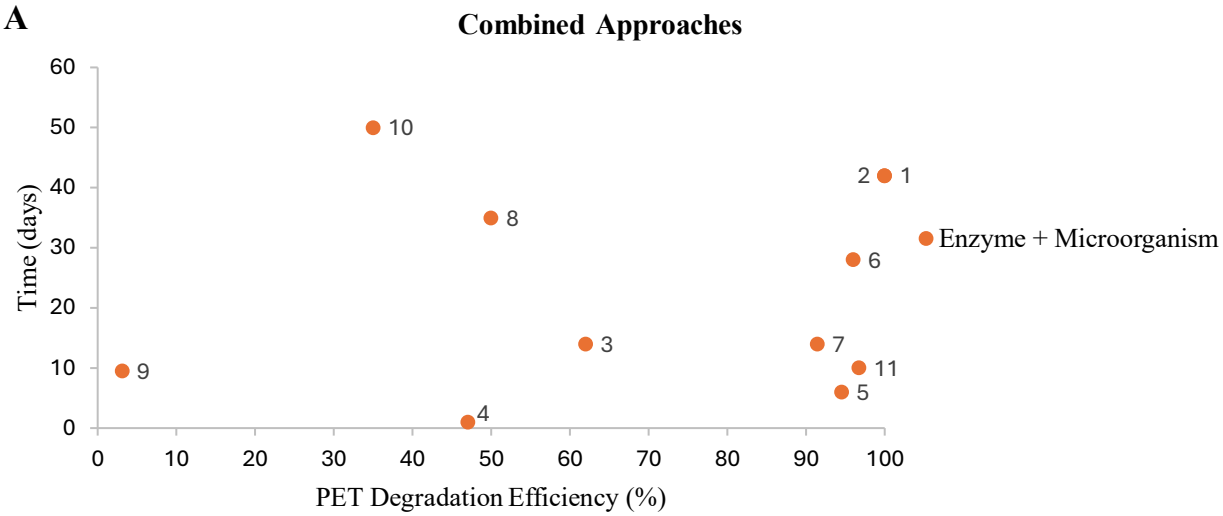
3.4.3. Others

Two studies in this category used hybrid strategies outside standard enzyme-microbe or chemical-bio workflows. One coupled biofilm modulation with enzymatic PET degradation using Dh3

(*Alkalilimnicola ehrlichii*), Dh5 (*Aquabacterium sp.*), and Thc_Cut2, yielding TPA and MHET detected at 230-260 nm after 5 days at 37 °C (154). The other showed *Rhodococcus sp.* RHA1 completely metabolizes terephthalate as the sole carbon source under aerobic conditions, with transcriptomics indicating a bifurcated TPA-catabolic pathway; PET polymer degradation was not directly tested, and time was not reported (155). **Table 19** provides further article information in detail.

To visualize the performance of combined PET degradation approaches, **Figure 7** provides an overview of combined systems. The scatter plot **(7A)** shows degradation efficiency (%) plotted against reaction time (days) for each combined study, where a PET-degrading enzyme was combined with a suitable microbial strain. Each point corresponds to a single study aimed at testing the synergy between enzymatic catalysis and microbial assistance, be it through co-incubation, sequential degradation, or engineered expression systems. Such hybrid systems are increasingly considered for their potential in alleviating limitations of single-component systems. Notably, only studies that reported or enabled calculation of PET degradation efficiency (%) were included, to ensure consistency and comparability of the dataset.

A



B

No.	Approach	Used Main (best-performing) Variant	PMID	Degradation Efficiency (%)	Time (days)
1	Enzyme + Microorganism	PETase, MHETase *Ideonella sakaiensis* 201-F6	26965627	100	42
2		PETase, MHETase *Ideonella sakaiensis* 201-F6	33579403	100	42
3		LCC (Leaf-branch compost cutinase) Clostridium thermocellum DSM1313 (genetically engineered)	32343496	62	14
4		LCC Pseudomonas umsongensis GO16	33865980	47	1
5		ICCG Pseudomonas nitroreducens S8, Pseudomonas monteilii S17	36474116	94.5	6
6		PETase, MHETase *Ideonella sakaiensis*	36385587	96	28
7		*Thermobifida fusca* cutinase (TfC) Stenotrophomonas pavanii JWG-G1	34864034	91.4	14
8		IsPETase, IsMHETase Piscinibacter *sakaiensis* 201-F6	39992123	50	35
9		IsPETase Yarrowia lipolytica Po1f, Pseudomonas stutzeri TPA3P	39629164	3.1	9.5
10		WCCG (quadruple mutant of LCC) Natural compost microbial consortium enriched with *Bacillus*, Acinetobacter, Cladosporium, Aspergillus, Penicillium	38040305	35	50
11		LCC F243I Clostridium thermocellum DSM1313	39904161	96.7	10

Figure 7. PET biodegradation efficiency and hybrid systems in combined enzyme-microorganism degradation approaches. **(A)** A scatter plot showing the relationship between PET biodegradation efficiency (%) and time (days) for combined approaches utilizing both enzymes and

microorganisms. Each dot represents a distinct study using a specific pairing of microbial strains with PET-degrading enzymes. **(B)** Tabulated summary of the combined systems corresponding to each data point in **Fig. 7(A)**, listing the exact microbial strain(s), enzyme(s), PubMed ID (PMID), degradation efficiency (%), and reaction time in days.

3.5. Unconventional methods

3.5.1. Marine origin

3.5.1.1. Common findings across studies

Under various experimental settings, all five research that used marine-derived bacteria or enzymes to carry out PET biodegradation techniques showed quantifiable PET depolymerization or monomeric conversion. In one study, when *Thioclava sp.* BHET1 and *Bacillus sp.* BHET2 were isolated from marine sediments and co-cultured for 6 weeks, they showed 22-fold and 2.6-fold increase in BHET conversion respectively (156). Engineered PETase and MHETase-expressing *Vibrio natriegens* Vmax strains showed activity on PET microplastic particles; after 8 days, the whole-cell system yielded ~10-11 mg/L BHET/MHET, while crude enzyme systems released up to 127 mg/L BHET and 14 mg/L MHET (MPP construct, 24-60 h) and 128 mg/L BHET and 76.5 mg/L MHET (TPL construct, 24 h) (157). The effective conversion of MHET was confirmed by the marine MHETase-like enzyme Mle046, which degraded 92% MHET in around 21 minutes at 30°C (158).

Under conditions like those of saltwater, PET surface colonization and possible breakdown were likewise fostered by biofilm-based marine systems. Within 45 days, PET films treated to consortia of *Alcanivorax*, *Cycloclasticus*, and *Thalassospira* developed biofilms, and observed morphological damages under ambient sea conditions (159). The ability of the *Pseudomonas*

aestusnigri marine enzyme PE-H to degrade PETase was shown in a different investigation. The wild-type PE-H released approximately 4.2 mg/L of MHET from PETase in 48 hours, but the intended Y250S mutant yielded 5.4 mg/L MHET from PETb; WT had no activity in the same time frame and activity on PETb was ~45× lower than on PETa (160).

3.5.1.2. Differences

The five studies differed greatly in terms of timelines, substrate types, microbial systems, and degradation goals. The enzymatic assay employing Mle046 achieved near-complete MHET breakdown in about 21 minutes (158), while co-culture systems (such as *Thioclava-Bacillus*) took 6 weeks to obtain increased BHET conversion (156). While marine consortia formed biofilms over a period of 45 days without measurable PET weight loss (159), PETase/MHETase-expressing *Vibrio natriegens* strains targeted PET microplastic particles over an 8-day period where crude enzyme system took upto 5 days; whole-cell system needed 8 days (157).

Additionally, the substrates varied: some research only employed model intermediates like MHET or BHET, while others included PET microplastics or PETa film. Applications included isolated enzyme reactions, microbial whole-cell transformations, and multispecies marine biofilms. Enzyme types included naturally occurring hydrolases (like PE-H) and synthetic variations (like Y250S). Also, many studies employed various detection and validation methods, including HPLC quantification of released chemicals, morphological observation of PET film degradation, and biofilm formation evaluations. Additional article-specific details are available in **Table 20**.

3.5.2. Larvae

Both investigations that used Tenebrio molitor larvae to study insect-mediated PET biodegradation found quantifiable PET degradation, which was confirmed by mass decrease and microbial

participation. PET was significantly reduced in weight when consumed by *T. molitor* in the first investigation, and the material passed through the gut almost entirely in 12 to 15 hours and an average mass reduction of 71.03% (PET-1) and 73.28% (PET-2) after passing through the digestive tract over a 36-day feeding trial, with a PET half-life of 3.36 h for PET-1 and 4.32 h for PET-2 (161). Under regulated feeding settings, PET1100 demonstrated 73.3% mass reduction in 13 days, PET27100 in 15 days, and PET34100 in 16 days, confirming PET degradation over longer time periods (162). In both situations, *T. molitor* played a consistent role, showing promise in plastic consumption with noticeable substrate loss. Furthermore, only one study (161) profiled microbial diversity, identifying *Spiroplasma*, *Dysgonomonas*, and *Hafnia-Obesumbacterium* as the predominant degraders; the other study mainly reported degradation efficiency without mentioning the specific microbial agents. **Table 21** highlights more detailed descriptions of the articles presented in this section.

3.5.3. Others

3.5.3.1. Common findings across studies

Each of the five investigations investigated indirect or non-traditional methods of PET degradation in either natural or laboratory settings. Using native *Bacillus gottheilii*, one study examined PET breakdown in mangrove soil conditions and found that, over the course of 90 days, there was a significant 18% weight loss of PET microplastics in treated mangrove soil, compared to a 16.4% loss in the control soil (163). In freshwater (Lake Maggiore), PETase and MHETase activity was detected alongside microbial communities such as *Pseudomonas* and *Bacillus*, and DSC/TGA confirmed chemical and structural changes in PET microplastics over 43 days (164). Lens-less digital holographic microscopy tracked PETG film exposed to enzymatic degradation, showing

observable surface alterations mainly within the first 11 days of a 43-day monitoring period (165). In a human gastrointestinal simulation (INFOGEST model) particularly due to interactions with colonic microbiota, PET particles exhibited structural fragmentation and amorphous region degradation within 72 h, but no full mineralization occurred (166). In a unique method, PET waste was electrospun into air filtration membranes, resulting in 100% material reuse (167). With deterioration assessed by metrics like weight loss, thermal behavior, or microscopy, these investigations validate detectable alterations or degradation processes in PET or PETG under simulated or non-traditional settings.

3.5.3.2. Differences

The experimental setups, methods of degradation, and efficiency differed substantially. PET microplastics lost 15% of their mass during 90 days in mangrove soil due to biodegradation by native microorganisms (163). In contrast, lake habitats did not exhibit any discernible biodegradation, although molecular evidence of microbial activity was discovered (164). The PETG enzymatic monitoring experiment documented progressive changes over 43 days but did not quantify degradation percentages (165). Within 72 hours, PET structural degradation was shown in the human gastrointestinal system simulation; however, complete breakdown was not attained, and only amorphous regions were impacted (166). But by focusing just on physical repurposing and ignoring PET breakdown, the electrospinning study (167) discovered a distinct kind of PET transformation. **Table 22** gives more in-depth information about these articles.

4. Discussion

4.1. Summary of key findings

This review analyzes a systematically selected set of 152 PET biodegradation studies, categorized as enzyme-only (n = 66), microbial-only (n = 45), combined enzyme–microorganism systems (n = 29), and unconventional methods (n = 12). Collectively, these studies encompass diverse degradation strategies, PET substrate types, and performance outcomes, providing a comprehensive overview of current biological approaches to PET degradation (**Figure 4**).

In every category, amorphous PET was consistently degraded better than crystalline, textile, or composite. Most of the studies used virgin PET (78), some used recycled (28) and environmental samples (18). Among the 63 studies which reported or allowed calculation of the degradation efficiency, 15 of them showed large efficiency (higher) (≥80%) whereas 23 were below 20% based on the type of enzymes/microbes, form of PET and temperature. The PET degradation was proven by the releasing of monomers TPA, MHET, and EG.

The PET depolymerization was the most effective and quickest in enzymatic-only systems. Several designed variants, such as TurboPETase (17), FAST-PETase (36), and thermostable PETase derivatives (e.g., S121E / D186H / R280A) (57), repeatedly showed robust degradation in 1-3 days. Cutinase variants such as LCC-ICCG and ICCG DAQI achieved near-complete depolymerization in 10-20 hours, while TfCut2 mutants required slightly longer times under aided conditions. The performance of degradation was also increased by the multi-enzyme system, particularly the one involving the favorable combination of PETases and MHETases. The systems dominated by microbes were more variable. Whereas the *Ideonella sakaiensis* achieved the level of 79% breakdown in 6 days (84). *Comamonas testosteroni* modified variant - RW31 strains

attained 37.1% in vitro (94). Several studies concentrated on the monomers release (fully or partially) instead of complete polymer degradation.

Synergistic results were achieved with combined enzyme-microbe systems. FAST-PETase and MHETase expressing engineered *E. coli* were able to degrade 71.3% within 24 hours (73), and surface displays and co-cultures ports also displayed immensely positive results (68) (72). The monomers of PET were occasionally transformed to bio-based value-added products like PHB (84) or engineered hosts such as *Yarrowia lipolytica* and *Pseudomonas putida* (80) (99). Unconventional approaches such as those involving gastrointestinal fluid emulations (166) and natural microbe consortia (163) (164) had less substantial degradation though showed applicability in real-life or passive applications.

In general, the presented results indicate the obvious correlation between efficiency of degradation and such factors as design of the biocatalyst, the form of PET, and the conditions of the reaction. Hybrid enzymes-microbe systems and engineered enzymes are currently the most successful with environmental systems and microbial methods also promising to be useful in practice. This synthesis serves as the basis of the critical assessments which are introduced in the following paragraphs where strengths, limitations, and comparative analysis of each approach to biodegradation are discussed.

4.2. Efficiency, engineering, and limitations of enzymatic-only approaches

Of the total 66 enzyme-only PET biodegradation studies evaluated, many indicated a high level of degradation and resulted in a rapid reaction time, especially under n optimized conditions. Engineered PETase and thermostable cutinase generally showed better rate of degradation and

time than the wild-type enzymes. This method takes advantage of rational enzyme engineering and thus it is possible to enhance catalytic activity, capacity to bind PET, and structural durability.

Many enzyme systems, which performed well, were greater than 70% degraded in 2 to 5 days and occupied the lower right quarter of the graph (visualized as **Figure 5A** and tabulated as **5B**). Among them, present the best results with TurboPETase (98.2% in 0.33 days) (17), the PETase triple mutant coupled with MHETase (70.0% in 0.042 days) (57) and ICCG cutinase (100.0% in 0.4 days) (59). Such a quick results were obtained with amorphous or cryo-ground PET at high temperature leading to the validity of the enzymatic strategy to be representative in controlled laboratory environments.

The mechanism behind the improvement of performance in engineered PETases is associated with the specific mutations, which increase thermostability and substrate binding. To illustrate, R280A has increased the entrance of its active site by reducing loop interference; and S121E and D186H enhance local hydrogen bonding and rigidity of the enzyme. Such mutations increase the plastic-binding and catalytic efficiency, and it is also valid in the TM variant (57). On the same note, the thermostable variants of LCC use disulfide bridge formation and loop stability to preserve intactness when temperatures reach up to 72 °C (59). Synergetic conversion of PET into TPA and EG without intermediates such as MHET accumulating was mitigated with PETase + MHETase systems (73). Other thermostable cutinases including LCC-ICCG and ICCG DAQI achieved more than 90% degradation at 65-72 °C (60) (61), which exploited the thermal transition of PET to degrade it successfully. Majority of the responses were in a pH range of 7.5-9.0 under tightly buffered environments.

Along with such successes, there are some limiting factors. Although there have been rational advances made, there are still difficulties in translating this success to real world PET types. Process of depolymerization was increased via the use of enzyme combinations. Most of these enzymes fared badly on crystalline, or textile PET material or multilayer PET that was not pretreated. Also, the work itself was measured mostly to short response times and the reuse of enzymes in a series of subsequent reactions was not often analyzed. However, it still has some obstacles concerning widespread deployment and robustness in the face of changing environmental conditions. The performance of wild-type enzymes was usually low, including ~10% on semi-crystalline PET (34), highlighting the importance of sophisticated engineering towards application success.

A small area of future innovation is enzyme engineering to make it more useful to crystalline or composite forms of PET or relating enzymatic PET depolymerization to downstream microbial upcycling systems. Such measures can be of assistance in closing existing performance gaps between laboratory potential and actual practice.

Summing up, the enzymatic-only methods are reviewed as the best due to the highest and the most complete PET degradation in the most favorable conditions. As therapeutic development of mutation design improves, as well as condition tolerance and substrate range, they possess an excellent promise to be applicable to plastic waste valorization at scale.

4.3. Critical evaluation of microbial-only approaches

In 45 microbial-only PET biodegradation studies reviewed, a great range of degradation patterns was identified with regards to microbial strain, PET form, incubation duration, and environmental conditions. Microbial systems that have been described took longer periods of time-mostly

between 7 and 30 days, and several concentrated on downstream metabolism of intermediates derived by the depolymerization of PET or partial depolymerization. Both native plastic-degrading strains and engineered strains with PETase and MHETase homologs were included in the species. There were very few microbial systems that could achieve PET degradation efficiencies of greater than 50% in a very short period (**Figure 6A** and **6B**). Many of strains needed amorphous or cryo-ground PET to enable significant levels of depolymerization and minimal salt or M9 media was often used. These trends verify the microbial strategy of being dependent on the geometry of bioavailable substrates and induction of PET-degrading enzymes which depends on the nutrient profile.

In contrast to enzymatic only systems which were designed to be fast, microbial investigations tended to focus on combining the depolymerization with biological conversion of the monomers of PET. As an example, in *I. sakaiensis*, the broken-down PET molecules have been used to create PHB in the intracellular environment in the presence of nutrient limitation (84). This indicates the possibility of the circular metabolic cycling, in which the PET-derived carbon undergoes the upcycling to the value-added polymers. This pathway was confirmed by downstream metabolite tracking in multiple experiments through intermediate synthesis and polymer buildup.

Microbial attachment and PET type usually affected interpretations on the outcomes of degradation. Microbial colonization and the efficiency of enzymatic hydrolysis were directly affected by PET form differences in the ease of reaching the surface due to differences in shape, powder, granule, or film. Other studies emphasized that it was not always easy to quantify the level of PET degradation even in cases where the latter was clearly visible, because of the absence of common metrics, or the inability to use mass balance methods (88) (89). These inconsistencies are

reasons why standardization of the degradation procedures should be adopted in microbial studies in PET.

Comparative biodegradation tests proved that engineered *Pseudomonas* described as a strong monomer utilizer whereas *Ideonella sakaiensis* strains occurred to be the best ones concerning the application of heterologous PET-degrading enzymes. The combination of these findings supports the argument that strain specific expression of enzymes, time of incubation and the structure of the PET substrate are key factors in determining successful activity of microorganisms in the PET biodegradation process. Also, most high-efficiency strains were most effective in laboratory conditions, and little information was available as to how the strains would work in variable natural or industrial conditions.

Finally, microbial-only PET degradation techniques provide the benefit of combining intracellular absorption with polymer breakdown, opening a novel avenue for biologically closed-loop recycling. Yet, lower rates of degradation, substrate form dependency and limited standardization are the major concerns. These limitations can possibly be addressed through co-culture systems and the use of hybrid enzyme-microbe approaches as discussed further in the yet to be mentioned sections.

4.4. Analysis of combined approaches

The combination of enzymatic hydrolysis with microbial metabolism is a rationally designed advance in PET biodegradation, the two-pronged benefit of which is the rapid depolymerization of the polymer and the subsequent assimilation of the monomers. Of the 29 articles covered under this category, those that demonstrated effective integration of enzyme activity with microbial

uptake mechanisms generally performed better than compared to those in which either the enzyme or the microbe acted as a limiting factor. Some of the top-performing configurations coupled highly active PET hydrolases with metabolically significant microbes to reach near-total degradation in 6 to 14 days. For instance, a thermostable cutinase of LCC with improved enzymatic properties was identified in LCC F243I, *Clostridium thermocellum* DSM1313 (thermophilic bacterium), and 96.7% of PET degradation by this organism after 10 days (133) was observed when compared to *Pseudomonas nitroreducens* S8 and *Pseudomonas monteilii* engineered with ICCG LCC that decomposed 94.5% PET in 6 days (131). These results indicate that the thermophilic microorganisms and engineered consortia can be more efficient and fast in breaking down as their natural abilities are combined optimally.

Although a helpful scatter plot and table were produced (**Figure 7**), the few points of data restricted what could be seen to general trends. Most systems were in the moderate-to-high degradation (40-95%) and reaction timeline usually broad. Chart-based interpretation cannot, however, capture the strategic subtlety of enzyme-microbe pairing. Detailed results indicated that some systems fully depolymerized PET (e.g., *Ideonella sakaiensis* with native PETase-MHETase in over 42 days) (127) (128), while others facilitated surface hydrolysis with carbon assimilation, effectively closing the loop from polymer back to cell uptake.

However, not all the combinations were equal in performance. An engineering that co-cultivated *Yarrowia lipolytica* AJD2 with *Pseudomonas stutzeri* and IsPETase only managed 3.1% degradation in 9.5 days (140) due to expression inefficiencies or metabolic mismatches. Compost-based consortia with LCC variants operated non-uniformly and only managed 35% degradation within 50 days (134), demonstrating slower dynamics in non-engineered microbial communities.

These illustrations show that mere co-application of microbes and enzymes is insufficient; biological compatibility and simultaneous kinetics are necessary.

Successful combined systems used a two-step model: enzymatic depolymerization of PET and microbial metabolism and utilization of PET intermediates. This modularity (enzyme makes PET monomers; the microbe immediately consumes them) can limit intermediate accumulation and support higher end-conversion. In general hybrid enzyme-microorganism strategies have great potential, but how they will be effectively used is tied to matching systems which can integrate enzyme functionality, microbial metabolic requirements and the process environment.

4.5. Experimental innovations and early insights in unconventional approaches

A large percentage of the published studies of PET biodegradation is dominated by enzyme, microbial, or combined categories. In a minority of these studies, sometimes, more unconventional methodologies are used, such as using novel bioengineering technologies, or atypical environments in which to attempt to extend the scope of PET degradation. Such methods are usually more focused on innovation than instant efficiency, as a means of overcoming long-term weaknesses of contact, conversion, or connectivity to the system. To give an example, enzymes have been taken nearer to their plastic substrates with the help of surface display technologies. These strategies reflect a shift towards whole-cell catalyst systems that eliminate purification processes and enable direct substrate interaction. Another trend in this research is attempting to simulate biologically relevant environments, such systems do allow for exploration of PET degradation in host organisms or natural gut-like systems. Computational models are also gaining popularity as inexpensive screening systems. However, the majority of such unconventional methods are at the initial stage and typically rely on simplified PET substrates, ideal lab conditions,

or theoretical assumptions. Despite that, their conceptual influence is significant; they extend the biodegradation horizon beyond efficiency metrics, embracing new interdisciplinary perspectives and framing PET degradation as a programmable, modular, and systems-level biological challenge.

4.6. Comparative insights across all approaches

Across the full spectrum of 152 studies examined in this review, the enzymatic-only, microbial-only, combined, and unconventional approaches each present distinct performance profiles, technical advantages, and implementation challenges. One of the prevailing trends that can be derived from the scatter plots and tables is that enzymatic-only systems dominate the fast PET degradation group. These results were primarily obtained on low- to mid-crystallinity substrates under buffered laboratory conditions, generally with mutants of PETase, dual-enzyme systems like PETase + MHETase, or thermostable cutinases like ICCG and LCC-ICCG. In contrast, microbial-alone systems utilized longer incubation times (7-30 days), comparatively lower degradation efficiency, and broader ecological applications like downstream assimilation or production of biopolymers. In general, microbial systems emphasize assimilation/upcycling potential and operation in more complex matrices, albeit with stronger dependence on PET form/crystallinity.

Combined enzyme-microbe approaches represent a convergence of speed and metabolic versatility. Although fewer in number (n=29), many reports in this category aim to pair rapid depolymerization with biological sinks for intermediates to mitigate product inhibition. These two-stage systems - comprising enzymatic depolymerization followed by microbial uptake - conceptually reduced product inhibition and allowed metabolic assimilation. However, not all combinations were successful; low-performance cases (e.g., 3.1% degradation in 9.5 days for a

Yarrowia-Pseudomonas-IsPETase system) (140) underscore the importance of biological compatibility and synchronized kinetics.

Unconventional approaches, though fewer in number, introduced conceptual innovations that broadened the PET biodegradation landscape. These early-stage strategies optimized system integration, programmability, and spatial control at the expense of short-term efficiency. Representative themes include host-centered catalysis and ecologically inspired reactors (e.g., gut- or lake-like settings), along with monitoring innovations that resolve early-stage surface and chemical changes. While not primarily designed to maximize short-term conversion, these studies act as feeders for contact engineering, compatibility, and observability that can inform mainstream workflows. Although not scalable yet, these strategies are indicators of the evolution to modularized, unorthodox platforms processes of PET.

Synthesizing findings across all four categories, it becomes evident that rather than a single dominant strategy, each PET biodegradation approach demonstrates distinct strengths under specific conditions. Enzymatic-only methods dominate in terms of degradation speed and yield, particularly under controlled laboratory conditions, but often require low-crystallinity substrates and stable environments. Microbial-alone systems, as slower and PET-form-dependent, permit complete mineralization and present promise in natural or semi-controlled settings based on environmental adaptation capability. Enzyme-microbe mixtures achieve a functional equilibrium, achieving high degradation efficiency within days through the balance of fast depolymerization and biological assimilation. Although frequently being proof-of-concept in nature, nonconventional strategies present out-of-the-box innovations: the solvent pretreatment, bio-upcycling, or synthetic gene circuits, in addition to others, that can change how bioprocessing is

done in the next generation. Specifically, the scope of each form of study was different: enzyme-based and mixed systems aimed at identifying a way to rapid depolymerization, and microbial and nonconventional ones explored circular metabolism, or true resilience like composting. These variances in goals of experiments (bio-upcycling, environmental integration, or yield maximization) underscore that performance is not merely driven by biological tools but also by design priorities and operating conditions. Finally, methodological inconsistencies within each category (like variability in PET type, pH reporting, and mass balance computation) are hindrances to direct comparisons and again justify the need for standard degradation protocols. In the future, the most effective PET biodegradation strategies will be ones that are both enzymatically specific and microbially robust, with a toolbox mentality that leverages the complementary strengths of each strategy to offer scalable, long-lasting solutions for reducing PET waste in numerous environmental and industrial applications.

4.7. Limitations of reviewed studies

While the systematic review aimed for comprehensive inclusion, several recurring limitations across the 152 studies undermined data comparability, reproducibility, and overall clarity. A primary concern lies in the inconsistent and sometimes incomplete characterization of PET substrates. While most studies reported the PET's physical form - such as amorphous, crystalline, powder, or film-based - a number of studies (around 11%) still failed to specify the source, such as whether the PET was virgin, post-consumer, or environmentally collected. This omission limits accurate comparison, as polymer properties can vary significantly depending on origin. These inconsistencies were observed across all approach types, including enzyme-only systems such as PETase studies, demonstrating that this issue is not confined to microbial or unconventional strategies.

A second key limitation involved PET degradation reporting metrics. Several studies reported only relative improvements in enzyme activity or product release (e.g., mg/L of TPA or MHET), without providing the actual initial PET weight or reaction surface area. As a result, it was not possible to calculate absolute degradation percentages in many cases. Even when degradation percentages were reported, supporting information such as PET mass used, PET concentration in solution, or recovery yields of degradation products was frequently lacking. Moreover, mass balance irregularities were noted, particularly in microbial and co-culture systems, where downstream metabolite assimilation or unaccounted loss processes were not accounted for. Such variations prevented useful quantification and interpretation of PET depolymerization outcomes.

Data representation was another critical bottleneck. While some studies reported precise PET weight loss curves, others relied solely on proxy measurements such as TPA or BHET release, optical density changes, or CO_2 evolution, with no cross-verification to PET loss. Units varied widely (mg, %, mM, or absorbance values) often with no clear reference to initial substrate quantity, resulting in fragmented datasets that were difficult to graph or compare quantitatively. Notably, many studies had to be excluded from chart-based analysis simply because they lacked complete or standardized degradation data. In addition, methodological inconsistencies further complicated comparative evaluation. Reaction conditions such as pH and buffer concentration were not always specified or vaguely described (e.g., "pH maintained neutral"), and incubation temperatures were listed at times without qualification of whether they were optimized or standard. In certain microbial and enzyme-alone research, degradation assays were conducted without stating whether PET was the sole carbon source or co-substrates had been added. Besides that, certain research merged engineered systems with wild-type controls without adequately

distinguishing the experimental setup, and therefore it was not possible to assess the distinctive contribution of strains, enzyme variants, or consortia.

Lastly, most of the studies were not ecological or industrially relevant considering that oversimplified experimental designs were relied upon. The studies of enzymes alone often favored idealized laboratory conditions-with purified proteins, homogeneous PET films, systems that are highly buffered, and sometimes high temperatures-that are far away to practical implementation conditions. On the other hand, microbial research examined either in composited or environmental specimens frequently displayed inadequate monitoring, biological replication, or even control groups, thus challenging the statement of degradation consistency and reproducibility. Some of the less conventional work, especially on gene circuits or synthetic biofilms, provided conceptually promising proof-of-concept innovations, but were distinctly premature with no applications in the field, no durability testing, and no lifecycle analysis. A larger restriction in the totality of methodologies was the lack of standardized procedures, which produced a lot of variances in measures of PET (ranging between less than 1 mg and over 50 mg), unreliable timings, and mismatched units. In addition, few long-term degradation studies (longer than two weeks) were present in the literature although they are critical in the evaluation of scalability and environmental relevance of a study. This disadvantage identifies the fact that harmonization of reporting standards is urgently needed, and an improved explanation of methodology needs to be achieved in future studies of PET biodegradation.

These limitations not only hinder the comparability and replicability of current findings but also mirror basic structural flaws in the area. These deficits need to be overcome in order to develop more effective, scale-up, and environmentally responsive PET biodegradation systems. The

following section outlines key areas for future research and outstanding research questions that have emerged from this review.

4.8. Future directions and research gaps

PET is one of the most durable synthetic polymers and conventional recycling processes continue to present extreme difficulty in the form of contamination, power use, and operating costs. An alternative, lower-energy and greener path, consisting of biological degradation through enzymes or microbes is an option that also complies with the global trends towards less plastic pollution. This review systematically analyzed 152 peer-reviewed studies, grouped by degradation approach to facilitate structured comparison. The direction in these fields is the creation of engineered PETase variants, multi-species bacterial consortia, and two-stage enzyme-microbe technologies, and they all lead to increasingly sustainable and efficient plastic management.

In order to inform further developments, the domain needs to resolve a number of research gaps. One of the most immediate needs is the standardization of evaluation metrics and methodological transparency. Across the reviewed studies, degradation performance was reported using inconsistent units (mg, %, mM, absorbance) often without reference to the initial PET quantity. This heterogeneity hindered comparative analysis and led to the exclusion of otherwise promising studies from graphical evaluations. Future studies should adopt a unified framework for reporting PET form, source, crystallinity, and reaction setup, along with absolute degradation percentages and product yields. Clear and reproducible reporting will enhance comparability across enzyme-based, microbial, and hybrid systems.

Another high-priority direction is the optimization of enzyme performance for industrial relevance. PETase, MHETase, and LCC variants have been deeply researched, yet reproducible and high-

yield degradation against broad ranges of PET substrates (especially high-crystallinity and textile-grade varieties) is limited. Future projects should target next-generation protein engineering, for example, thermostability regulation, improved substrate affinity, and improved catalytic turnover by directed evolution or computer-aided modeling. Enzyme immobilization and co-factor engineering are also deserving of consideration for improving operation stability, enabling continuous biocatalytic processes. In microbial systems, the focus should move towards developing advanced microbial consortia capable of fully mineralizing PET and its intermediates under different environmental conditions. While partial degradation by single strains was demonstrated in certain studies, complete breakdown typically requires synergistic consortia with directed metabolic cooperation. Synthetic biology tools like modular gene circuits and horizontal pathway transfers are to be explored in future work to build long-lasting and engineerable PET-degrading microbial communities.

Finally, but not least, the increasing enzymatic and microbial toolkit remains a long-term aspiration. Much of the work used well-characterized enzymes or isolates, and vast metabolic pathway and environmental source diversity have remained unexplored. Enzyme mining and metagenomic screening enzymes of extreme or polluted environments may prove to lead to new enzymes with improved properties that are PET-active. In a similar manner, poorly comprehended microorganisms can expose complete delivery systems, especially by high pH, heat, or saline environments.

One of the key bottlenecks of the field is the lack of scale-up of laboratory success to industrial. Most reviewed studies operate under idealized lab conditions (purified proteins, buffered media, short timeframes, high temperature) which do not reflect the complexities of large-scale waste

processing. There is a clear need for pilot-scale validation, including the design of microbial bioreactors, immobilized enzyme columns, and solid-state setups compatible with mixed or contaminated plastic waste. Addressing substrate diversity - particularly textile PET and layered packaging - will also require integration of pretreatment steps or tailored dual-stage degradation workflows. Another critical area is system-level integration with circular economy frameworks. The distinct advantage that biological degradation of PET holds is that the degradation process is capable of efficiently recovering monomers that are of high purity and can be re-used either in reused or recycled into new products. Few studies have however performed lifecycle or techno-economic analyses, nor do we know very much about their scalability, cost competitiveness and how they would interface with the current infrastructure. Further or follow up research including LCA, risk assessment, and output testing relevant to industry, needs to be carried out to promote the involvement of environmental scientists, engineers, and policymakers to facilitate its large-scale deployment.

Biodegradation presents high-purity recycling avenues for PET and extended use in a sustainable manner. Future activities should continue to build upon the nascent platforms - engineered enzymes, microbial consortia, and integrated systems - yet direct primary focus on real-world deployment and global circular economy goals.

4.9. Final critical summary

This review systematically reviewed 152 PET biodegradation studies across enzyme-only, microbial-only, combined, and unconventional approaches. Within each category, there were unique strengths and compromises. Enzyme processes achieved rapid depolymerization under ideal conditions but were limited by long-term functionality and substrate versatility. Microbial

systems showed improved metabolic versatility but were limited by low rates of degradation and environmental sensitivity. Hybrid strategies exhibited synergistic advantages but were nevertheless limited by heterogeneous procedures and integration issues. Unconventional techniques introduced innovation but were not necessarily scalable or proven.

Common roadblocks to all the strategies were uneven degradation practices, convergent reporting methods, short datasets, and limited scalability. However, the sector is advancing from engineered enzymes, synthetic communities, and modular breakdown platforms. The integration offers a structured foundation to address longstanding problems and guide the development of scalable, real-world PET degradation solutions.

5. Conclusion

The global issue of plastic pollution, in particular PET, has instigated an expanding area of research within biologically mediated degradation approaches. This review systematically examined 152 peer-reviewed studies on enzymatic, microbial, combined, and unconventional PET biodegradation methods. All the categories showed different advantages and gaps depending upon the reaction conditions, the nature of substrates and the type of mechanism. The rapid depolymerization was detected in enzyme systems though those based on engineered variants of PETase/cutinase were more effective, and the best laboratory conditions were used so that the catalytic activity was maximized. Microbial processes could act on a variety of PET breakdown products and function under variable environmental conditions but were generally slower in terms of degradation rate, which was frequently substrate crystallinity- and limited PET accessibility-dependent. Hybrid approaches took advantage of synergistic advantages of enzymatic hydrolysis and microbial consumption or pretreatment, yet issues of reproducibility and scale-up remain. Novel approaches introduced new concepts but were not sufficiently validated or reproducible.

Collectively, this review illustrates growing momentum into the area of PET biodegradation. Improvements have been realized in the field of enzyme, microbial engineering, and systems combination. However, practical development is still restricted by the absence of standard reporting, the limited number of metrics, and scalability problems. By integration of existing knowledge into methodological frameworks, this review shows where advances have been made and where issues persist.

In anticipation of implementation in the future, synchronizing laboratory performance with daily implementation will be critical. The overlap of synthetic biology, systems engineering, and

environmental biotechnology opens new avenues to overcome current constraints and achieve scalability, scale-up PET degradation. Increased innovation, supported by standardization and long-term validation, will be critical in enabling experimental success to be translated into applicable plastic waste utilization. This review provides the systematic foundation for that effort and underscores the need for coordinated biologically driven approaches to accelerate PET biodegradation into the sphere of practical application.

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Appendix

Table 2. Detailed information on articles related to PETase & its variants.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/ Challenges (If any)
35577005 (16)	1. Investigated overexpression strategies for *Ideonella sakaiensis* PETase (IsPETase) in *E. coli* to improve PET biodegradation efficiency. 2. Optimized expression of IsPETaseMut, a more active PETase mutant, using chaperone co-expression and fusion expression techniques.	Polyethylene terephthalate (PET)	Recycled PET (from plastic bottles) (~50 mg)	China	Enzyme-based depolymerization	IsPETaseMut (Mutant PETase from *Ideonella sakaiensis*) NusA-IsPETaseMut (fusion with NusA protein for improved solubility & altered catalytic properties)	~19.6 mg weight loss (~39%) 1.4 higher than unmodified PETase	2 weeks	Promotes efficient PET biodegradation	1. Slow degradation rate. 2. Lower efficiency on highly crystalline PET compared to amorphous PET.
29666242 (34)	1. Engineered PETase to improve PET degradation efficiency by modifying its active-site cleft. 2. The modified PETase double mutant (S238F/W159H) exhibited improved PET degradation.	Polyethylene terephthalate (PET)	Recycled PET film (~15% crystallinity)	University of Portsmouth, UK; National Renewable Energy Laboratory, USA	Enzyme-based depolymerization	PETase (from *Ideonella sakaiensis* 201-F6), Double mutant PETase S238F/W159H, Engineered PETase mutants for improved PET degradation	PET crystallinity loss: Wild type: ~10% Double mutant: ~14%	96 hours (4 days) at 30°C, pH 7.2	PETase specificity for aromatic polyesters could aid in selective plastic degradation.	PETase activity limited to specific temperature and pH conditions.
36174890 (35)	1. Engineered variants of *Ideonella sakaiensis* PETase using an electrostatic interaction-based strategy to improve thermostability and efficiency.	Polyethylene terephthalate (PET)	Recycled PET from postconsumer PET bottles	Tianjin University, China	enzyme-based degradation	Enzyme: IsPETaseI139R (a variant of PETase)	1. IsPETase I139R degraded 3.6 times more PET than wild-type PETase. 2. IsPETase S92K/D157E/R251A fully depolymerized PET into TPA.	72 hours at 40–50°C.	Enhances PET biodegradation efficiency	Higher temperatures (40–50°C) are required for optimal performance

36811288 (19)	1. Investigated concentration-dependent inhibition of PETases from *Ideonella sakaiensis* (IsPETase) and its homologues. 2. Inhibition is affected by incubation time, PET surface area, and solution conditions. 3. Adding DMSO or increasing PET surface area via cryomilling partially alleviates enzyme inhibition.	Polyethylene terephthalate (PET)	Virgin (Amorphous PET film, Cryomilled PET powder)	University of Portsmouth, UK & National Renewable Energy Laboratory (NREL), USA	enzyme-based depolymerization	HotPETase (engineered, thermostable)	Amorphous PET films incubated with IsPETaseWT at 30 °C for 96 h showed typical hydrolysis but suffered up to 93% inhibition at high enzyme concentration (500 nM) at pH 9.0. HotPETase completely eliminated concentration-dependent inhibition, maintaining consistent hydrolysis even at high enzyme concentrations.	96 hours at 30°C	Enzyme engineering can enhance PET hydrolysis efficiency, making biocatalysis more industrially viable.	1. Natural mesophilic PETases exhibit concentration-dependent inhibition, limiting their industrial application. 2. Inhibition mechanisms are not fully understood but relate to enzyme-enzyme interactions on PET surfaces.
36414665 (27)	1. The system co-displays PETase and hydrophobin (HFBI) on yeast cell surfaces, improving enzyme adhesion to PET. 2. Enhanced adsorption and enzymatic degradation were confirmed via molecular dynamics simulations.	Polyethylene terephthalate (PET) (both high-crystallinity and low-crystallinity)	Typically, virgin (PET bottles)	Tianjin University, China	Enzymatic degradation using a genetically engineered yeast system. Special Bacteria/Bacterium Used: *Pichia pastoris* yeast engineered to display PETase and hydrophobin (HFBI) on the cell surface.	PETase (from *Ideonella sakaiensis*) Hydrophobin (HFBI from *Trichoderma reesei*) – improves enzyme binding to PET.	High-crystallinity PET (hcPET, 45% crystallinity): 10.9% conversion Low-crystallinity PET (lcPET, 6.3% crystallinity): 55% conversion	hcPET: 10 days at 30°C. lcPET: 18 hours at 30°C.	1. Uses a yeast-based whole-cell system, reducing costs and energy consumption compared to purified enzymes. 2. Hydrophobin increases enzyme adhesion, minimizing enzyme loss during the reaction.	1. The enzyme is heat-labile, requiring mild reaction conditions (30°C–40°C). 2. PETase activity decreases in the presence of degradation byproducts.

35008691 (33)	1. The study highlights a significant improvement in the efficiency and thermal stability of PET hydrolases, leading to a more effective enzymatic PET degradation process.	Polyethylene terephthalate (PET)	Virgin	University of Insubria and Politecnico di Milano, Italy	Enzymatic degradation using IsPETase variants from *Ideonella sakaiensis*	TS-ΔIsPET variant TS-ΔIsPET (W159H/F238A/S121E/D186H thermostable variant) (engineered from *Ideonella sakaiensis* PETase)	26% depolymerization yield (PET microparticles (~30 µm) using 0.1 mg/mL enzyme. >80% PET degradation nanoparticles (~80 nm) using 20 µg/mL of TS-ΔIsPET	80% degradation in 1.5 hours at 45°C. 26% degradation in 2 days at lower concentrations	Potential for circular recycling: The process enables PET to be broken down into its monomers (terephthalic acid (TPA) and ethylene glycol (EG)), which can be reused to create new PET, reducing fossil fuel consumption and plastic pollution	1. Enzyme inactivation: TS-ΔIsPET showed a tendency to adsorb onto PET microparticles, leading to enzyme deactivation over time. 2. Temperature sensitivity: The enzyme lost activity after 24 hours at 45°C,
31887523 (28)	1. Developed a whole-cell biocatalyst by displaying bacterial PETase on the yeast (*Pichia pastoris*) surface. 2. Enhanced PETase's enzymatic activity, pH, and thermal stability compared to native PETase.	Polyethylene terephthalate (PET)	Recycled	Tianjin University, China	Enzymatic degradation	Bacterial PETase (from *Ideonella sakaiensis* 201-F6). PETase engineered for surface display on yeast cell walls using glycosylphosphatidylinositol (GPI) anchors.	Surface-displayed PETase on yeast cells → 36× higher hcPET degradation turnover than purified PETase. Maintained 85% activity after three recycling cycles, and 50% after seven cycles.	18 hours at 30°C (Consistent degradation efficiency observed across multiple reaction cycles)	1.Operates efficiently at moderate temperatures (~30°C), reducing energy consumption compared to thermal/chemical recycling methods. 2. Provides an eco-friendly PET degradation strategy using yeast surface display.	1. Whole-cell biocatalyst reuse efficiency decreased after seven cycles, indicating eventual loss of enzyme activity. 2. Surface display system may have steric hindrance issues, affecting full enzyme functionality.

37401198 (48)	1. The study uses molecular dynamics simulations to analyze how IsPETase binds to PET substrates of different polymerization degrees. Binding energy increases with substrate chain length up to a certain threshold, after which the increase slows down. 2. These findings explain why wild-type PETase has low hydrolysis efficiency and provide insights for designing more efficient PET-degrading enzymes.	Polyethylene terephthalate (PET)	Virgin	China	Enzymatic degradation	Enzyme: IsPETase (PETase from *Ideonella sakaiensis* 201-F6)	NA	NA	NA	The exact substrate binding mode of PET to PETase remains unclear. It does not experimentally measure PET degradation efficiency, conversion rates, or real-world degradation timelines.
38360963 (17)	1. A newly designed PET hydrolase, TurboPETase, was developed using computational redesign and deep learning models. The enzyme was engineered to have a more flexible PET-binding groove, allowing it to hydrolyze PET more efficiently.	Polyethylene terephthalate (PET)	recycled PET flakes	China	Enzymatic degradation	TurboPETase (engineered variant of PET hydrolase derived from bacterium HR29)	98.20%	8 hours	1. Biodegradation of PET at industrially relevant conditions provides an eco-friendly recycling alternative. 2. Avoids microplastic formation, as PET is completely depolymerized into monomers.	1. Requires high enzyme stability at industrial conditions (temperature, pH, reaction time). 2. Incomplete degradation of PET with very high crystallinity (>30%) remains a challenge.
33579416 (29)	1. The study demonstrates the use of yeast cell surface display to enhance PET degradation. 2. *Pichia pastoris* GS115/PETase-GCW51 was used to express PETase, which was tested for thermostability and solvent stability.	Polyethylene terephthalate (PET)	Commercial PET bottles (Virgin PET)	China, Germany	Enzymatic degradation	Enzyme: PETase Host Organism: *Pichia pastoris* GS115/PETase-GCW51 (yeast cell surface display system)	55%	after 18 hours at 30°C	NA	Stability of PETase under different chemical/solvent conditions was tested, implying potential challenges in industrial applications.

39628843 (44)	1. The study characterized a novel PET hydrolase (named Sb PETase) isolated from Schlegelella brevitalea *sp.* nov. and compared its activity with the well-known Is PETase from *Ideonella sakaiensis*.	Polyethylene terephthalate (PET)	Virgin Commercial PET Film	Qingdao, China	Enzymatic degradation	Enzyme: Sb PETase from Schlegelella brevitalea *sp.* nov. IsPETase from *Ideonella sakaiensis*	Not specific but 8% higher than the wild type	BHET: 18 hours incubation at 30°C PET Film: 24 hours incubation at 30°C	NA	The catalytic activity of Sb PETase is lower than that of Is PETase.
35951410 (46)	1. The study used ancestral sequence reconstruction (ASR) and a design method (PROSS) to trace the evolution of the PET-degrading enzyme IsPETase from ancestral cutinases. 2. It was found that low-level "promiscuous" PETase activity existed in ancestral enzymes well before PET was introduced into the environment, and later mutations (especially around the active site) enhanced PET-degrading activity.	Polyethylene terephthalate (PET)	Recycled	NA	Enzymatic depolymerization	IsPETase Enzyme	1. wild-type IsPETase produced approximately 119 µM terephthalic acid (TPA). 2. Although some ancestral variants (e.g., GrAnc8) showed lower TPA production at 30 °C, GrAnc8 demonstrated a more than 2-fold higher turnover rate when assayed at an elevated optimal temperature (60 °C).	over a 6-hour incubation period at 30 °C	1. it discusses that enzymatic depolymerization of PET is a low-energy, sustainable alternative to traditional methods (such as incineration or mechanical recycling) and holds promise for reducing plastic waste accumulation.	1. There is an inherent trade-off between enzyme thermostability and catalytic activity. While some engineered variants gained significant stability.
37050393 (41)	1. Rational design based on the 3D structure of wild-type IsPETase led to the introduction of single-point mutations (notably S238Y, I208V, and N212A). 2. DSC analyses demonstrated that the S238Y and N212A mutants induced a greater reduction in PET crystallinity (with S238Y showing a 3.3-fold improvement) compared to the wild type.	Polyethylene terephthalate (PET)	Virgin (Commercial PET Bottles)	Ecuador	Enzyme-based degradation	Engineered *Ideonella sakaiensis* PETase (specifically the S238Y mutant)	The S238Y mutant exhibits a 3.3-fold increase in PET-degrading activity compared to the wild-type enzyme.	3 days	The study emphasizes that enzymatic degradation offers a sustainable alternative to thermal recycling processes, which generate hazardous emissions. The approach is positioned as a promising strategy to manage PET waste in an environmentally friendly manner.	1. High crystallinity of commercial PET (around 31%) still poses a barrier by limiting enzyme accessibility. 2. The enzyme performs better when the PET surface is pre-treated.

33753197 (38)	1. The study addresses the limited stability and activity of wild-type IsPETase in PET degradation. 2. A novel mutation design tool, Premuse, was developed to mine evolutionary information from 1486 homologous sequences. Ten single point mutations were initially selected; among these, two mutations—W159H and F229Y—were found to enhance enzyme stability.	Polyethylene terephthalate (PET)	typically, virgin (PET bottles)	Chinese Academy of Agricultural Sciences, Beijing, China	Enzyme-based degradation	Engineered IsPETase (double mutant W159H/F229Y)	The W159H/F229Y variant exhibits a nearly 40-fold increase in degradation activity for amorphous PET relative to the wild type. PET bottle preform-23.4 mg PET/h/mg enzyme.	For amorphous PET- 24 hours at 40 °C For PET bottle-24 to 72 hours	By significantly enhancing the biodegradation of PET under mild conditions, the engineered enzyme offers a promising eco-friendly alternative to traditional thermal or chemical recycling methods, potentially reducing hazardous emissions and energy consumption.	Although the double mutant W159H/F229Y shows dramatically improved performance on amorphous PET and bottle preform, challenges remain: The enzyme's performance on highly crystalline PET may still be limited.
33561457 (30)	1. PETase from *Ideonella sakaiensis* 201-F6 was expressed and purified from *E. coli* to hydrolyze waste PET substrates for monomer recycling. 2. The intermediate BHET is efficiently converted to MHET, while further hydrolysis of MHET to TPA is very slow.	Polyethylene terephthalate (PET)	Environmental waste	Department of Chemical Engineering, National Taiwan University of Science and Technology, Taipei, Taiwan	Enzymatic hydrolysis	IsPETase (from *Ideonella sakaiensis* 201-F6) hydrophobins (RolA and HGFI) are used as pretreatment agents	Without hydrophobin pretreatment: ~18% weight loss of PET With hydrophobin pretreatment: ~34% weight loss	over 5 days	Enzymatic recycling of PET under mild conditions (pH 8, 30 °C) (using PETase) is a green and energy-efficient alternative to conventional chemical recycling methods.	1. PETase converts BHET to MHET very efficiently, but further hydrolysis of MHET to TPA is very slow, leading to accumulation of MHET. 2. Effective pretreatment with hydrophobins is required to enhance PETase activity, adding an extra step to the process.

33375975 (31)	1. BhrPETase, a PET hydrolase from bacterium HR29 (a close homologue of LCC with 94% sequence identity), was successfully secreted by *Bacillus subtilis*. Engineering the host strain via hrcA deletion boosted enzyme production approximately 20-fold, achieving titers of 0.66 g/L.	Polyethylene terephthalate (PET)	Virgin	Shanghai, China	Enzymatic hydrolysis	BhrPETase (The PET hydrolase characterized from bacterium HR29.)	BhrPETase generated a total of 6.3 mM degradation products (0.17 mM BHET, 3.66 mM MHET, 2.47 mM TPA)	20 hours	The high activity and exceptional thermostability of BhrPETase make it a promising candidate for the biological recycling of PET waste.	1. Although BhrPETase is highly thermostable, its activity decreases when the reaction temperature is raised above 70 °C. 2. The enzyme was tested on amorphous PET; efficiency on semi-crystalline or highly crystalline PET may differ.
34833184 (32)	1. The study demonstrates that enzymatic treatment of recycled bottle-grade PET with PETase leads to significant changes in the material's properties. Enzymatic action causes the formation of colloidal structures—identified as BHET dimer oligomers—on the PET surface. These changes are correlated with increased surface roughness, a decrease in crystallinity, and reduced mechanical integrity.	Polyethylene terephthalate (PET)	Recycled	University of Bayreuth, Germany	Enzymatic degradation	PETase from *Ideonella sakaiensis*	NA (MHET degraded 2.3 mM and TPA 1.3 mM after 96 hours)	96 hours	PET-degrading enzymes (like PETase) might contribute to the breakdown of macroplastic into microplastics.	1. The enzyme shows a lower efficiency in degrading smaller PET oligomers (specifically, the BHET dimer tends to accumulate), indicating substrate binding limitations. 2. The direct measurement of degradation remains challenging.

36639437 (36)	1. The predominant hydrolysis product is terephthalic acid (TPA) with almost complete conversion (up to ~94.7% TPA by product composition).	Polyethylene terephthalate (PET)	Environmental	Hubei University, Wuhan, China	Enzymatic degradation	IsPETase (derived from *Ideonella sakaiensis*) Partial-deglyco Fast-PETase-Pp	100% depolymerization, The hydrolytic reaction predominantly produces TPA with a conversion rate where approximately 94.7% of the product is TPA.	2–3 days at 50 °C	eco-friendly and energy-efficient	2. The enzyme efficiently degrades PET flakes and trays but shows little to no degradation activity against more crystalline PET products.
37555384 (42)	1. The study explores two strategies to enhance the thermostability of the native IsPETase enzyme from *Ideonella sakaiensis*.	Polyethylene terephthalate (PET)	Virgin	Singapore	Enzymatic degradation	IsPETase vGFP–FAST (scaffolded FAST PETase) the engineered FAST PETase	SC-ST Cyclized Variant (cPETase): 10-fold reduction in PET hydrolysis compared to native PETase as measured by degradation on low-crystallinity PET powder.	3 days	developing thermostable PET hydrolase enzymes offer potential for green recycling of single-use PET	The SC-ST cyclization approach, while enhancing thermostability, compromises PET hydrolysis efficiency due to enzyme dynamics.
31601227 (20)	1. The marine diatom Phaeodactylum tricornutum was successfully genetically engineered to produce and secrete an engineered PETase enzyme, which degraded PET-based substrates in a saltwater environment.	Polyethylene terephthalate (PET)	Recycled	Germany	Enzymatic	PETaseR280A (an engineered version of PETase from *Ideonella sakaiensis*)	The degradation rate of PETG was approximately 80-fold higher than that of conventional bottle PET under the tested conditions.	2 to 6 weeks	A promising, eco-friendly approach for bioremediation of plastic pollution in oceanic environments.	2. The enzyme's activity is optimal near 35 °C, yet the microalga grows best at lower mesophilic temperatures (21–26 °C), leading to a compromise in conditions.

35091192 (43)	1. The enzyme, named BbPETase, contains an additional N-terminal domain that plays a dual role. It enhances the thermal stability of the enzyme but may hinder its direct binding to the PET substrate. 2. At 30 °C, the enzyme shows activity comparable to that of *Ideonella sakaiensis* PETase (IsPETase), while at 40 °C, BbPETase (especially the full-length form) retains activity for at least three days—resulting in up to a 3.5-fold higher activity than IsPETase.	Polyethylene terephthalate (PET)	Virgin	Daegu, Republic of Korea	Enzymatic degradation	BbPETase	Under controlled conditions, BbPETaseFull exhibited up to a 3.5-fold higher PET hydrolytic activity at 40 °C compared to IsPETase. The engineered BbPETaseNIIG variant showed a 2.8- to 3-fold improvement in activity relative to the wild-type enzyme.	1-3 days	reducing plastic waste and an environment-friendly	1. The additional N-terminal domain, while beneficial for enhancing thermal stability, appears to negatively affect the enzyme's ability to adsorb to the PET substrate.
32345276 (18)	1. The study demonstrates for the first time that PETase—an enzyme originally discovered in *Ideonella sakaiensis*—is functionally expressed in the green alga *Chlamydomonas reinhardtii* (using strain CC-124). PETase expressed in *C. reinhardtii* is active; its catalytic activity was verified by detecting degradation products of PET.	Polyethylene terephthalate (PET)	Virgin	Republic of Korea	Enzymatic degradation	PETase (functionally expressed in *Chlamydomonas reinhardtii*)	1. In the PET powder-9.12 mg of TPA was produced from 30 mg of PET(conversion rate of 35.17%) 2. For PET film-qualitative SEM data support PET degradation (no percentage conversion is provided)	4-week incubation period at 30 °C	Microalgae are advantageous since they do not require organic carbon for growth under photoautotrophic conditions, do not produce endotoxins, and naturally inhabit aquatic environments. This approach could ultimately contribute to reducing plastic accumulation	Relatively slow

38749251 (45)	1. Two novel PET hydrolases were identified: (1) PpPETase: Derived from *Pseudomonas paralcaligenes* MRCP1333, isolated from human feces (2) ScPETase: Derived from Streptomyces calvus DSM 41452. Both enzymes show robust catalytic activity toward different PET substrates, including semicrystalline PET powder (Cry-PET) and low-crystallinity PET films (gf-PET).	Polyethylene terephthalate (PET) (Cry-PET powder and gf-PET film)	Virgin	China	Enzymatic degradation	Wild-type PpPETase (from *Pseudomonas paralcaligenes* MRCP1333) Wild-type ScPETase (from Streptomyces calvus DSM 41452) Engineered variants: PpPETaseY239R/F244G/Y250G, ScPETaseA212C/T249C/N195H/N243K	Engineered variants:1. PpPETaseY239R/F244G/Y250G variant degrades Cry-PET 3.1-fold faster than the wild-type. 2. ScPETaseA212C/T249C/N195H/N243K variant shows a 1.9-fold increase in activity compared to ScPETaseWT. In scale-up degradation tests using a co-expression system (ScPETase-IsMHETase): 1. Cry-PET degradation exceeded 40% conversion, with TPA accounting for 37.7% of the degradation products. 2. gf-PET degradation reached about 6% conversion, with TPA at 25.6%.	after a 30-day incubation at 30 °C some small-molecule substrates (e.g., BHET) were decomposed much faster (e.g., full breakdown of BHET within 24 h for one enzyme screening)	enzymatic PET biodegradation is considered environmentally friendly as it operates under mild conditions (room temperature), avoids harsh chemicals or high energy inputs.	1. The efficiency differs between PET substrates. 2. The co-expression system requires the addition of a second enzyme (IsMHETase) to fully convert intermediate products (mostly MHET) to TPA, which adds complexity.
35628419 (37)	1. A five-member environmental consortium (three *Pseudomonas* spp. and two *Bacillus* spp.) synergistically degrades PET. 2. Discovery of a novel PETase, EstB, which hydrolyzes the PET oligomer BHET and the polymer PET to MHET and TPA.	Polyethylene terephthalate (PET)	Virgin	Reed College and Willamette University, USA	Enzymatic degradation	EstB (novel PETase from *Pseudomonas sp.* strain 9.2) Aldehyde dehydrogenases (predicted role in MHET/EG metabolism) Environmental consortium of *Pseudomonas* and *Bacillus* spp.	PET degradation confirmed via HPLC by release of MHET and TPA after EstB incubation	24-48 days	1. The study highlights a route for sustainable PET waste management: coupling enzyme-based deconstruction with microbial assimilation can reduce environmental PET accumulation and generate value-added bioproducts.	Functional validation is limited to EstB; additional PETases remain uncharacterized.

35382549 (23)	1. The study aimed to overcome the temperature limitation of the mesophilic wild-type IsPETase (with a melting point, Tm, of ~45.1 °C) by engineering more thermostable variants. 2. The findings clearly indicate that increasing the thermostability of IsPETase enhances PET hydrolysis efficiency at higher temperatures (up to 60 °C), which is beneficial because amorphous PET becomes more accessible for enzymatic attack near or above its glass transition temperature.	Polyethylene terephthalate (PET)	Virgin	University of Greifswald, Germany	Enzymatic degradation	IsPETase variants- the triple mutant (TM), TM1(IsPETase TMK95N/F201I), TM3 (which combines K95N/F201I with additional disulfide bridge mutations), and DuraPETase derivatives (e.g., D1)	For PET Nanoparticles: With increasing reaction temperature, product release (sum of MHET, terephthalic acid, and BHET) increased. For example, at 60 °C, the variant TM1 achieved an 8-fold increase in relative activity compared to the wild-type. For Amorphous PET Film: The TM3 variant exhibited one of the highest relative activities, with a reported up to 120-fold increase compared to wild-type IsPETase and a 10- to 20-fold enhancement over the TM.	PET Nanoparticles: 24 hours Amorphous PET Film: 72 hours	By enhancing the thermostability and activity of PETase at higher temperatures, these engineered enzymes offer a promising, environmentally friendly alternative to conventional chemical recycling processes that often involve harsh chemicals and high energy input.	1. The crystallinity and physical aging of PET (especially in film form) can hinder enzymatic access.
29590588 (47)	1. Crystal structure and molecular dynamics (MD) simulations reveal PETase has higher active site flexibility compared to thermophilic PET hydrolases, primarily driven by a novel disulfide bond (C176–C212) near the active site.	Polyethylene terephthalate (PET)	NA	Chile, USA	Enzymatic degradation	PETase from *Ideonella sakaiensis* (main enzyme studied) *Thermobifida fusca* cutinase (TfCut2; used as a comparative reference) Leaf-branch compost cutinase (LCC; also used as a comparative reference enzyme)	PETase demonstrates 5 to 120 times higher depolymerization activity against PET films at 30°C compared to thermophilic PET-degrading enzymes previously tested.	NA	offering an environmentally friendly, energy-efficient alternative	The stability of PETase at higher temperatures is limited; disulfide bonds play a crucial role in maintaining structural and catalytic integrity. Reduction or mutation of these bonds drastically reduces enzyme activity and stability.

29374183 (25)	1. The main focus is to elucidate the structural basis and catalytic mechanism of *Ideonella sakaiensis* PETase (IsPETase) AND finds enzyme-substrate binding and provides engineered variants for enhanced PET degradation. 2. Identified two major degradation steps: (1) Nick generation: Cuts PET chains at internal ester bonds. (2) Terminal digestion: Converts PET ends into monomers.	Polyethylene terephthalate (PET)	Virgin	Republic of Korea	Enzymatic degradation	IsPETase from *Ideonella sakaiensis* IsPETaseR280A	Wild-type IsPETase degraded PET and released- TPA and MHET. R280A variant: Increased degradation efficiency by +22.4% (18h) and +32.4% (36h)	36 hours at 30°C	1. Highlights the potential of mild-temperature, enzyme-driven PET degradation offers a biocatalytic solution over conventional chemical degradation which is energy-intensive and polluting.	IsPETase cannot degrade MHET and also overall slow degradation rate.
38395309 (21)	1. A surface crowding model was developed to explain how enzyme adsorption density affects PET degradation. 2. The enzyme's performance is inhibited at high surface concentrations due to crowding, leading to less productive catalytic conformations. 3. At 55°C, crowding effects are less pronounced, but still exist.	Polyethylene terephthalate (PET)	Recycled PET	MIT, Massachusetts, USA	Enzymatic degradation	IsPETase (from *Ideonella sakaiensis*) Engineered Variants: TS-PETase (thermostable version), TSP-PETase (TS-PETase with T116P mutation), TSP-S238N-PETase and TSP-S238N-S290P-PETase (further thermostabilized mutants)	At 30°C: TSP-PETase showed a 30% increase in maximum product formation rate compared to TS-PETase. At 55°C: TSP-PETase produced 2.4 mM of combined TPA and MHET in 105 minutes at 28.8 g/L PET and 1000 nM enzyme.	Up to 105 minutes	1. The study offers a promising route for scalable enzymatic PET bio recycling. 2. By optimizing enzyme mutations that minimize inhibition from crowding, it enables more effective PET valorization under mild conditions, potentially reducing energy demands compared to chemical recycling.	1. Crowding effects on PET surfaces limit enzyme efficiency, especially at high concentrations. 2. The SC model still requires more robust physical validation for large-scale industrial application.

34499955 (39)	1. This study investigates the synergistic effect of a fungal-derived lytic polysaccharide monooxygenase (PcAA14A) on an engineered PET-degrading enzyme (IsPETaseEHA) from *Ideonella sakaiensis*. 2. It presents insights into PET degradation efficiency enhancement via non-catalytic, protein–protein interaction strategies.	Polyethylene terephthalate (PET)	Virgin PET	Hubei University, Wuhan, China	Enzymatic degradation	Main Enzyme (PET-degrading): IsPETaseEHA (engineered PETase from *Ideonella sakaiensis*) Synergistic Protein(fungal): PcAA14A (LPMO from Pycnoporus coccineus) Catalytically inactive mutant used to confirm mechanism: PcAA14A^H100A^	18 h reaction: 343 µM (IsPETaseEHA alone) → 534 µM (with PcAA14A) → 59.2% increase 5-day reaction: IsPETaseEHA alone: 866 µM With PcAA14A: 1097 µM → ~27.7% increase in total degradation products	upto 5 days	This study offers a novel strategy to boost enzymatic PET degradation by using synergistic protein-protein interactions, avoiding energy-intensive or chemical pre-treatments.	1. PcAA14A does not degrade PET independently and only works in synergy with a PETase. 2. Real-world application needs further validation beyond lab settings.
38833982 (24)	1. This study enhances PET degradation by engineering thermostable IsPETase variants fused with carbohydrate-binding module BaCBM and displaying them on *E. coli* surfaces. 2. Engineered a thermostable IsPETase mutant called HotPETase (HP), and a fusion construct with a carbohydrate-binding module (HLCB).	Polyethylene terephthalate (PET)	Recycled PET	Jining Medical University, Jining, China	Enzymatic	Main enzymes: HotPETase (HP) – thermostable variant of IsPETase HLCB – HP fused with BaCBM (carbohydrate-binding module from *Bacillus anthracis*) D-HLCB – surface-displayed whole-cell *E. coli* expressing HLCB Microorganism: *Escherichia coli* BL21	HLCB: ~25.5% improvement over HP after 50 hours D-HLCB: Maximum 1.03 mM monomeric product yield HP alone: ~271.5 µM monomeric products in 71 hours	HLCB: Prolonged degradation up to ~80 h D-HLCB: Maintained activity for over 100 hours HP: Active PET degradation up to 70 hours	Demonstrates eco-friendly enzymatic PET degradation under mild conditions (around 50°C) and engineered systems offer low-cost, reusable solutions for biocatalytic PET recycling.	1. Despite improved thermostability, D-HLCB showed poor stability at high temperatures (>60°C) due to potential heat-induced cell surface damage.

35630830 (52)	1. This study uses computational approaches—including alanine scanning, site-saturation mutagenesis, and molecular dynamics simulations—to identify PETase variants with enhanced PET-binding and catalytic efficiency.	Polyethylene terephthalate (PET)	NA	Thailand	Enzyme-based biodegradation	Main Enzyme: PETase (from *Ideonella sakaiensis*) Key engineered variants: S238C, Q119F, D112M/S238F, WT PETase	No experimental S238C variant showed improved binding affinity and simulations suggest enhanced catalytic potential, indicating likely improved PET hydrolysis.	NA	PETase mutants like S238C offer promise for enzymatic PET recycling under mild conditions	This was a purely computational study; no lab validation was included.
34044062 (26)	1. It explores a protein engineering strategy by fusing a cellulose-binding domain (CBM) from T. *reesei* to an engineered variant of IsPETase (IsPETaseEHA). 2. Among tested binding domains (CBM, PBM, HFB4), only CBM enhanced PET hydrolysis. 3. The optimal enzyme concentration was 20 µg/mL; higher concentrations led to reduced efficiency.	Polyethylene terephthalate (PET) (amorphous)	Virgin PET	Hubei University, Wuhan, China	Enzyme-based	IsPETaseEHA, IsPETaseEHA_CBM	At 30°C- IsPETaseEHA: 17.9 µM total product IsPETaseEHA_CBM: 30.7 µM (17.4 µM TPA + 13.3 µM MHET) → 71.5% increase At 40°C- IsPETaseEHA: 174.1 µM IsPETaseEHA_CBM: 251.5 µM → 44.5% increase	3 hours	1. Enhanced PET degradation through CBM fusion offers a promising route for low-energy enzymatic recycling. 2. High activity at ambient and moderate temperatures (30–40°C) = environmentally friendly conditions. 3. No toxic chemicals used.	No testing on crystalline PET.
34833182 (40)	1. The primary objective is to improve the activity and thermal stability of IsPETase, a PET-degrading enzyme from *Ideonella sakaiensis*, using rational protein engineering (PROSS algorithm). The study involves designing, expressing, and testing enzyme variants to enhance PET depolymerization under mild conditions.	Polyethylene terephthalate (PET)	Virgin PET	Denmark	Enzyme-based degradation	IsPETase (wild type) Engineered variants: S136EPETase (most impactful), S125RPETase, T270QPETase, and comparison with SonPETase	3.3-fold increase in degradation activity over wild-type at 34 °C (measured by MHET release) Major hydrolysis product: MHET, 5× more than TPA after 24 h	observed over 24 hours	Lower-temperature enzymatic degradation is more sustainable and less energy-intensive than thermal or chemical PET recycling.	1. Other variants (S125RPETase and T270QPETase) showed minimal or no improvements, highlighting the unpredictable nature of protein engineering.

39775626 (49)	1. Developed the first large-scale 3D atomistic models of PET, (1) Crystalline PET (cPET) model with 7280 monomers (2) Amorphous PET (aPET) model derived via simulated melting. 2. Simulated PETase (IsPETase) adsorption onto amorphous PET (aPET) and crystalline PET (cPET) and then found stronger and more productive adsorption on aPET, with PET chains entering the active site in some simulations.	Polyethylene terephthalate (PET)	NA	University of Porto, Portugal Forschungszentrum Jülich, Germany RWTH Aachen University, Germany	Enzymatic Adsorption and interaction modeled via molecular dynamics (MD) simulations.	IsPETase from *Ideonella sakaiensis*	NA Simulated substrate access and proximity metrics show aPET is more degradable than cPET.	NA	Supports biocatalytic recycling by revealing the molecular basis for PETase preference toward amorphous PET.	1. No wet-lab validation. 2. Simulations do not replicate full enzymatic hydrolysis or real PET waste.
34587366 (22)	1. Compared wildtype IsPETase with a double-mutant variant (W159H/S238F) for PET hydrolysis and the double mutant showed increased thermal stability.	Polyethylene terephthalate (PET) (amorphous film, crystalline powder, biaxially oriented film)	Virgin	National Renewable Energy Laboratory (NREL), USA & University of Portsmouth, United Kingdom	Enzymatic degradation	Wild-type IsPETase (*Ideonella sakaiensis* PETase) Double mutant IsPETase (W159H/S238F)	Amorphous PET film (2.9% loading): ~0.5–1.5%/h release rate of aromatic products Crystalline PET powder (up to 23.2% loading): conversion improved up to ~5× vs. amorphous PET Highest product release rate: DM enzyme at 40 °C and low substrate loading achieved 2.2 nmol s^{-1} g^{-1}	168 hours	Supports enzymatic recycling of PET, especially with engineered variants like the DM enzyme.	1. Product inhibition by ethylene glycol (EG) and MHET affects degradation efficiency. 2. The WT enzyme is less thermostable and less active at higher temperatures.
34075621 (50)	1. It investigates how PET binds to the IsPETase enzyme and the resulting conformational changes using molecular docking and molecular dynamics (MD) simulations. The goal is to elucidate key molecular interactions and flexible regions for engineering more efficient PET-degrading enzymes.	Polyethylene terephthalate (PET) modeled as MHET-based oligomers	Computational model	Brazil, Spain	Enzymatic	IsPETase (wild type from *Ideonella sakaiensis*)	Simulations show strong binding and proper alignment of PET in catalytic site which confirms productive substrate orientation, especially in subsites I–II	NA	1. Supports bioengineering of IsPETase variants for enhanced PET degradation efficiency. 2. Promotes computational strategies to guide enzyme optimization.	1. No experimental degradation confirmation. 2. Focused only on low-temperature IsPETase.

38095477 (51)	1. This study examines how double mutations (S238F and W159H) in IsPETase from *Ideonella sakaiensis* improve PET degradation by altering the enzyme's structure and dynamics. 2. Structural mutations S238F and W159H reshape IsPETase's active site, turning it from flat/wide to narrower and deeper. 3. The W159H mutation removes a bulky side chain, enlarging the binding cavity and improving PET accommodation.	Polyethylene terephthalate (PET)	Simulated PET model (Used a PET dimer created in silico to mimic realistic binding within the active site)	Quantumzyme LLP, Bangalore, India	Enzymatic (and also All-atom molecular dynamics (MD) simulations, Molecular docking and Free energy landscape)	IsPETase (from *Ideonella sakaiensis*) Mutant studied: S238F/W159H double mutant (DM)	Binding affinity and proximity indicators show better PET binding: PET molecule retention near active site: ~6 PET dimers within 20 Å of DM IsPETase vs. ~2 for WT over 100 ns simulations	NA	1. The study supports rational enzyme engineering for enhanced PET biodegradation under industrial and environmental conditions. 2. Engineering such thermally stable and catalytically enhanced enzymes can improve eco-friendly enzymatic PET recycling pipelines.	1. No real PET substrates, real-time hydrolysis or TPA/EG release measured. 2. Only short PET dimer modeled — doesn't capture full polymer chain behavior.

Table 3. Detailed information on articles related to MHETase & its variants.

PMID No.	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/ Challenges (If any)
32160855 (53)	MHETase gene was successfully constructed in the pUCIDT plasmid.	Polyethylene terephthalate (PET)	Recycled PET and Virgin PET films	Bandung Institute of Technology, Indonesia	Enzyme-based depolymerization	MHETase (from *Ideonella sakaiensis* 201-F6) Recombinant *Escherichia coli* BL21 (DE3) expressing MHETase	NA	NA	more environmentally friendly than incineration and landfill, No need for enzyme purification	1.Further optimization needed for degradation efficiency. 2. Testing on actual PET degradation is yet to be conducted
36587193 (54)	1. The study establishes a yeast whole-cell biocatalyst by expressing MHETase (an enzyme from *Ideonella sakaiensis*) on the surface of *Fusca Saccharomyces cerevisiae*.	MHET (monohydroxy ethyl terephthalate) from PET hydrolysis	Virgin	University of Toronto, Canada	Enzymatic	MHETase from *Ideonella sakaiensis*	Nearly 100% conversion of MHET to TPA	24 hours	1. By using a whole-cell biocatalyst that avoids the need for purified enzymes and harsh chemical conditions, this method can reduce energy consumption and hazardous byproducts	1. The efficiency of MHETase display on the yeast cell surface is only about 16–22% of total MHETase is exposed on the surface.

Table 4. Detailed information on articles related to PETase + MHETase (dual-enzyme systems).

PMID No.	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/ Challenges (If any)
32989159 (55)	1.PET biodegradation through a two-enzyme system involving PETase and MHETase from *Ideonella sakaiensis*. 2.The study investigates the structural and functional properties of MHETase and its synergy with PETase in breaking down PET into its monomeric components, TPA & EG.	Polyethylene terephthalate (PET)	NA	several universities in UK & USA	Enzyme-based depolymerization	PETase and MHETase (from *Ideonella sakaiensis*) PETase + MHETase chimeric fusion proteins	Chimeric PETase–MHETase with 12-residue linker at 0.25 mg PETase/g PET + 0.5 mg MHETase/g PET → highest monomer release from amorphous PET.	after 96 h	sustainable approach, Reduces reliance on chemical recycling and incineration	1. Degradation efficiency depends on substrate accessibility and enzyme stability. 2.The two-enzyme system requires controlled conditions.
38393574 (56)	1. It presents a dual method analytical strategy (bulk absorption and ^{1}H NMR spectroscopy) to quantify and qualify PET degradation products. The research supports enzyme-based PET recycling by improving measurement sensitivity and process monitoring during enzymatic hydrolysis of PET.	Polyethylene terephthalate (PET)	NA	Furtwangler University, Germany	Enzymatic degradation using PETase and MHETase	PETase, MHETase from *Ideonella sakaiensis*	PETase generates ~30 µM MHET, 10 µM TPA, and 2 µM BHET	after 4 hours	supports development of PET recycling strategies should be useful for wastewater bioremediation and industrial enzyme-assisted PET degradation.	No real PET film degradation experiments conducted
37350932 (57)	1. The study identifies surface Tg, enzyme-product interaction, and enzyme inactivation as the main limiting factors in PET degradation.	Polyethylene terephthalate (PET)	Virgin PET	Hereon Institute of Active Polymers, Germany	Enzymatic	IsPETaseTM (S121E/D186H/R280A) IsMHETaseSM (W397A)	Maximum 70% depolymerization with the dual-enzyme system.	in 1 hour at 50°C	1. The study supports development of rapid, surface-optimized enzymatic recycling systems for PET under mild aqueous conditions.	1. No degradation-induced crystallization was observed in PET thin films up to 60°C.

Table 5. Detailed information on articles related to cutinase/LCC & its variants.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/ Challenges (If any)
37594123 (58)	1. The leaf-branch compost cutinase (LCC-ICCG) and its engineered variants (LCC, PelB-LCC, LCC-PBM, PelB-LCC-PBM) showed high PET degradation efficiency, outperforming PETase from *Ideonella sakaiensis*. 2. Engineered enzymes showed enhanced thermostability and surface binding via fusion with binding modules and secretion tags.	Polyethylene terephthalate (PET) Amorphous PET films (AmPET), Extruded PET strands (ExPET), and Recycled PET (RPET)	Recycled	University of Massachusetts Lowell, USA	Enzymatic degradation	LCC-ICCG (leaf-branch compost cutinase, engineered variant) PelB-LCC (LCCICCG fused with PelB secretion signal)	~100% degradation of AmPET and ExPET ~80% degradation of RPET with 200 g/L PET loading in 1-L bioreactor	3 days at 65°C	1. Supports enzymatic PET bio recycling with high efficiency under industrially feasible conditions. 2. Demonstrates potential for replacing thermochemical recycling with scalable bio-based alternatives.	1. Enzyme instability at 65°C beyond 72 h (30% activity loss). 2. Crystallinity increase during degradation hampers reaction continuation.
39653629 (63)	1. It presents a novel enzyme engineering strategy using a fusion protein (cutinase from *Thermobifida fusca* + anchor peptide LCI) to significantly enhance PET degradation efficiency. 2. LCI improved the binding of the enzyme to the PET surface, especially in the early phase of the reaction.	Polyethylene terephthalate (PET)	Virgin PET	Jiangnan University, Wuxi, China	Enzymatic degradation using engineered Tfuc2-LCI fusion protein	Tfuc2 – a thermostable variant of *Thermobifida fusca* cutinase LCI peptide – anchor peptide fused to C-terminal of Tfuc2	Tfuc2-LCI degraded 66.7% of PET substrate Tfuc2 alone degraded ~37%	96 hours at 65°C	1. Potential for more efficient, eco-friendly PET degradation. 2. Reduced need for harsh conditions due to increased binding efficiency	Activity is temperature-dependent (65°C optimal) — may limit environmental applications and stability drops significantly at 70°C.

38944965 (61)	1. It describes the engineering of *E. coli* to express a cutinase from *Ideonella* species (ICCM cutinase) for enhanced PET hydrolysis, focusing on optimizing protein expression, solubility, and enzymatic activity for PET plastic degradation and yielded MHET & TPA. 2. Crystalline PET films showed surface erosion and pore formation, confirmed by SEM.	Polyethylene terephthalate (PET) both film & powder	Virgin PET	School of Life Sciences, Shanghai University, China	Enzymatic hydrolysis	Enzyme: ICCM cutinase (from *Ideonella*) Host microorganism: *Escherichia coli* BL21(DE3), engineered to express the cutinase	PET film degradation yielded ~100% PET degradation	4 days at 60 °C at pH 10	1. The use of *E. coli* and thermostable cutinase at moderate temperatures suggests a sustainable, scalable biocatalytic process. 2. Enables low-energy enzymatic recycling of PET compared to conventional thermo-mechanical methods.	1. Solubility of recombinant ICCM was initially poor — solved with SUMO fusion. 2. PET degradation was limited to surface erosion; full mineralization not achieved.
39843897 (66)	1. It uses computational methods (molecular dynamics, docking, and binding free energy calculations) to analyze the potential of cutinase from *Fusarium oxysporum* (FoCut) for degrading. 2. Molecular dynamics (MD) simulations confirmed that PET remains stably bound in the active site throughout the simulation.	Polyethylene terephthalate (PET)	NA	Brazil	Enzyme-based biodegradation	Cutinase from *Fusarium oxysporum* (FoCut)	NA	NA	1. The strong predicted PET binding suggests FoCut is a promising candidate for PET biodegradation. 2. Cutinases are generally robust, adaptable, and potentially scalable for industrial or environmental PET waste treatment.	Results are based on modeling, not measured PET degradation.

39390488 (60)	1. It evaluates extracellular production of a PET-degrading cutinase (ICCG DAQI variant) by two bacterial systems and examines its performance in enzymatic hydrolysis of PET fibers, including industrial textile PET, with high degradation yields. 2. Cutinase ICCG DAQI, a thermostabilized LCC variant, was expressed extracellularly by C. glutamicum (via signal peptides) and *E. coli* (via leaky membrane release without signal peptides).	PET textile fibers	Recycled	Germany	Enzymatic hydrolysis	ICCG DAQI (LCC variant)	Crude supernatant (3 mg/g PET)- 97.8% TPA and 98.6% EG after 20 h Purified enzyme (1 mg/g PET)- 96.4% TPA and 98.8% EG after 18 h	18-20 hours	1. Demonstrates high-efficiency enzymatic PET recycling using unpurified supernatant, reducing cost and complexity and avoids harmful chemicals and harsh depolymerization processes.	Membrane integrity issues in *E. coli* due to intracellular enzyme overproduction, leading to partial leakage.
35557867 (62)	1. A novel cutinase—named MtCut —derived from the deep sea bacterium Marinactinospora thermotolerans DSM45154 exhibits efficient PET-hydrolyzing activity at ambient temperatures. It catalyzes PET depolymerization in a manner, releasing primarily MHET and TPA, with trace amounts of BHET.	Polyethylene terephthalate (PET)	Virgin	China	Enzymatic	MtCut – novel PET-degrading cutinase from Marinactinospora thermotolerans (deep-sea bacterium)	MtCut achieved nearly complete conversion of PET intermediates (with no MHET detected at high enzyme load in a 72-hour reaction) and displays a 10-fold higher catalytic efficiency toward MHET than IsPETase and ICCG.	60–72 hours	offer a greener, energy-saving alternative	1. PET hydrolysis is influenced by substrate morphology; the enzyme performed better on microparticles.
35039943 (65)	1. An engineered variant of leaf-branch compost cutinase (ICCG) with mutations (Phe243Ile, Asp238Cys, Ser283Cys, Tyr127Gly) exhibits excellent PET-depolymerizing activity at high temperatures (optimal activity at 70 °C).	Polyethylene terephthalate (PET)	Virgin	Republic of Korea	Enzymatic	ICCG (engineered leaf-branch compost cutinase) (produced by *Bacillus subtilis*)	At 37 °C: 3.65% weight loss At 70 °C: 6.99% weight loss	8 days	1. The biocatalytic depolymerization process operates under mild conditions without toxic catalysts or high energy inputs.	The depolymerization efficiency (weight loss of 3.65–6.99% over 8 days) is relatively modest.

32269349 (59)	1. Scientists engineered an optimized PET-degrading enzyme (depolymerase) with significantly enhanced efficiency.	Polyethylene terephthalate (PET)	Recycled PET	France	Enzyme-based depolymerization	LCC-ICCG (engineered thermostable LCC variant)	100% depolymerization of post-consumer PET flakes (20 g)	10 h at 72 °C	1. Highly efficient PET recycling with minimal environmental waste.	1. Requires controlled conditions (72°C) for optimal efficiency, which may not be feasible in natural environments.
31690819 (64)	1. The cutinase TfCut2, when combined with cationic surfactants and specific mutations, enhanced PET degradation efficiency.	Polyethylene terephthalate (PET)	Virgin PET	Keio University, Japan	Enzymatic hydrolysis using *Thermobifida fusca* cutinase (TfCut2)	Wild-type and mutant cutinase enzyme from *Thermobifida fusca* (TfCut2) Mutant variants: G62A, F209A, H129W, G62A/F209A (best performing mutant)	TfCut2 G62A/F209A double mutant + 30 ppm cationic surfactant + lcPET film (3–5% crystallinity) → 90% PET degradation in 24 h and almost complete (~100%) in 30 h	30 hours	1. Mild reaction conditions (40–70°C, pH 7–9) prevent the release of harmful byproducts.	1. Incomplete degradation of high-crystallinity PET (enzyme performance was lower for high-crystallinity PET than for low-crystallinity PET).

Table 6. Detailed information on articles related to mix enzymatic approaches.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
27214855 (67)	1. The study explores a dual enzyme system for PET degradation, combining LC-cutinase and carboxylesterase (TfCa) which leads to continuous MHET hydrolysis, improving PET breakdown.	Polyethylene terephthalate (PET)	NA	University of Leipzig, Germany	Enzymatic degradation	TfCut2 – polyester hydrolase from *Thermobifida fusca* KW3 LC-cutinase (LCC) – metagenome-derived PET hydrolase TfCa – carboxylesterase from *Thermobifida fusca* KW3, immobilized on SulfoLink resin for enhanced stability Dual enzyme combination = TfCut2 or LCC + immobilized TfCa	Dual enzyme system with LCC + immobilized TfCa at 30 µg/mL → ~48% PET film degradation in 24 h, 2.4× higher product release than TfCut2 alone.	24 hours	1. The study demonstrates the potential of enzyme-based PET degradation as an alternative to conventional chemical recycling. 2. The dual enzyme system enhances the breakdown of PET, reducing the accumulation of plastic waste.	1. PET degradation was not complete, and further optimization is needed. 2. The presence of MHET as an intermediate initially inhibits the hydrolysis process.
39357610 (68)	1. A novel thermophilic hydrolase named BMHETase was identified that exhibits dual activity—degrading both BHET and MHET, which are key intermediates in PET biodegradation. BMHETase demonstrated significantly higher BHET degradation activity than other.	Polyethylene terephthalate (PET)	Recycled Postconsumer PET (pcPET) film derived from a commercial PET container	China	Enzymatic degradation	BMHETase and the engineered mutant BMHETase6M LCC-ICCG (additional Enzyme)	BMHETase 6M has an 8-fold higher activity for MHET degradation and a 2-fold increase for BHET compared to the wild type. combined with LCC-ICCG, BMHETase 6M achieved complete degradation	incubated for 24 hours at 55°C and pH 8.0	By developing BMHETase6M, the engineered enzyme enables completer and more efficient depolymerization of PET, which can enhance recycling processes and reduce the accumulation of PET waste.	Although BMHETase6M shows greatly enhanced activity, its MHET degradation activity is still relatively weak at higher temperatures compared to its BHET degradation.

							of residual MHET, only TPA remained			
34705547 (69)	1. The study demonstrates that polyester hydrolases from Antarctic bacteria—specifically, Mors1 from Moraxella *sp.* strain TA144 and OaCut from *Oleispira* antarctica RB-8—are active at moderate temperatures (25°C) and can hydrolyze both aliphatic & aromatic polyesters.	Polyethylene terephthalate (PET) - aromatic polyester polycaprolactone (PCL) - aliphatic polyester	Virgin Commercial PET Film	Chile, Germany	Enzymatic hydrolysis	enzymes: 1. Mors1 (from Moraxella *sp.* strain TA144) 2. OaCut (from *Oleispira* antarctica RB-8)	amorphous PET films - Mors1 achieved approximately 0.59% weight loss after 24 hours at 25°C, increasing to about 2.5% after 10 days.	10 days at 25 °C	the use of such psychrophilic enzymes for PET degradation could contribute to a more sustainable management of plastic waste.	1. The degradation rates of PET at moderate temperatures (25°C) remain low compared to those achieved by thermophilic enzymes operating near PET's glass transition temperature.
35753492 (74)	1. A novel enzyme complex (termed PC enzyme complex) was developed by assembling chimeric PETase (cPETase) and chimeric Candida antarctica lipase B (cCALB) on a scaffolding protein equipped with a carbohydrate-binding module (CBM3).	Polyethylene terephthalate (PET)	Environmental waste	Republic of Korea	Enzymatic degradation	PC enzyme complex = cPETase (chimeric PETase from *Ideonella sakaiensis*) + cCALB (chimeric Candida antarctica lipase B) + CBM3-scaffold assembled via cohesin–dockerin interactions	For highly crystalline PET: The enzyme complex increased TPA release by approximately 6.5-fold compared to the individual enzymes. The PET film exhibited a 12% weight loss overall, with a reduction rate of about	crystalline PET and BHET - at 30 °C for 4 days waste PET bottles - from 1 to 7 days at 30 °C.	the study emphasizes that eliminating the need for pretreatment (a costly and energy-intensive step) may lower processing costs and environmental impact in PET recycling/upcycling applications.	The study is conducted under laboratory conditions, and additional work is necessary to optimize enzyme production and integration into large-scale PET recycling processes.

							1.7% per day.			
35717313 (71)	1. The study aimed to create a whole-cell biocatalyst by engineering *Pseudomonas putida* to (i) metabolize PET monomers as sole carbon sources and (ii) express PET hydrolases extracellularly.	Polyethylene terephthalate (PET)	Virgin Commercial PET Film	USA	Enzymatic degradation	Engineered strain: *Pseudomonas putida* KT2440 PET hydrolases tested: LCC (leaf compost cutinase) HiC (*Humicola insolens* cutinase) IsPETase (from *Ideonella sakaiensis*)	Approximately 1.5% weight loss in 6 days	6 days	while engineered PET depolymerization is feasible, the low degradation efficiency and fitness costs highlight challenges that must be overcome.	1. The engineered strains showed lower PET hydrolase expression and PET depolymerization efficiency in minimal medium compared to nutrient-rich LB medium.
34085821 (76)	1. The paper uses multiscale QM/MM molecular dynamics simulations to unravel the detailed catalytic mechanism by which enzymes break the ester bonds in polyethylene terephthalate (PET).	Polyethylene terephthalate (PET)	Virgin	Spain	Enzymatic degradation (via a catalytic mechanism simulated by QM/MM methods)	PETase LCC-ICCG variant	The catalytic process shows a rate-limiting free energy barrier in the range of approximately 18.9–21.1 kcal·mol^{-1} depending on the enzyme model and substrate length.	NA	A promising eco-friendly alternative to conventional mechanical and chemical recycling.	1. The study uses oligomer models (dimer and trimer of MHET) to represent PET. These models do not capture the full complexity of long-chain polymers or the heterogeneous crystallinity seen in industrial PET.

35452981 (70)	1. The novel polyesterase from Moraxella *sp.* (MoPE) degrades both amorphous and semi-crystalline forms of PET.	Polyethylene terephthalate (PET)	Virgin	Athens, Greece	Enzymatic degradation	MoPE (a polyesterase from Moraxella *sp.* isolated from Antarctic Sea water) FoFaeC (a feruloyl esterase from *Fusarium oxysporum* belonging to the tannase family)	For semi-crystalline PET powder = ~1.3 µM products are released; For amorphous PET powder = 3.5–6.6 times higher product release than the semi-crystalline PET powder and PET film, respectively ; When synergy with FoFaeC enhances TPA release by= ~1.4–1.5-fold.	Approximately 3 days	By releasing terephthalic acid (TPA) in a controlled manner, the process offers the potential to recover monomers that can be repurposed into new polymers or value-added products.	1. Lower degradation efficiency is observed on PET films compared to powders due to a reduced reaction surface area. 2. The enzyme works more efficiently on amorphous PET than on highly crystalline PET; water-induced plasticization is needed to make crystalline PET accessible.
37110762 (75)	1. The PETase enzyme (BbPETaseCD) from Burkholderiales bacterium was improved through the rational design of disulfide bridges using computational tools.	Polyethylene terephthalate (PET)	Recycled PET	Tianjin University, China	Enzyme-based biodegradation	Main enzyme: BbPETaseCD (from Burkholderiales bacterium) Engineered variant: N364C/D418C, P207C/D280C, A209C/R283C, P207C/D280C/N364C/D418C, A209C/R283C/N364C/D418C	N364C/D418C variant showed ~11-fold improvement in PET degradation over WT BbPETaseCD	over 14 days at 40°C	1. Positive environmental implication as enzyme degradation of PET is more sustainable compared to high-temperature chemical processes.	1. Only the N364C/D418C variant had good expression and stability; other variants showed poor soluble expression and possible misfolding.

38703683 (72)	1. The paper introduces a novel bacterial enzyme cascade reaction system (BECRS) to degrade PET microplastics and films efficiently.	Polyethylene terephthalate (PET)	Virgin PET	Nanjing Tech University, China	Enzyme-based biodegradation via whole-cell biocatalysis	FAST-PETase – engineered PET hydrolase with high activity CBM3 (Carbohydrate-Binding Module 3) – enhances PET surface binding MHETase – intracellularly expressed to remove inhibitory MHET Together forming BECRS = FAST-PETase + CBM3 surface display + intracellular MHETase	PET film (amorphous): Up to 3437 ± 148 μg/(d·cm²) degradation rate, 14.6 mM total product release Crystalline PET MPs: 21.40% degrade (10 μm MPs), 16.09% degrade (100 μm MPs), Over 90% of products = TPA	7 days for both	1. BECRS shows high promise as a sustainable PET biodegradation method which operates under mild conditions, efficiently converts PET to TPA, robust against environmental variation.	1. Engineered system tested only in controlled lab/simulated gut conditions — real-world validation still needed. 2. Focused on degradation to TPA — recovery or reuse of TPA not addressed

37804764 (73)	1. Particle size and crystallinity significantly affected PET hydrolysis: smaller particles and amorphous PET degraded faster. 2. C-terminal position of PETase in co-display fusion proteins yields better PET hydrolysis than N-terminal.	Polyethylene terephthalate (PET)	Virgin PET & real world recycled bottles	Northeast Agricultural University, Heilongjiang, China	enzyme-based biodegradation	FAST-PETase, MHETase (co-display enhanced yield), IsPETase, LCC-ICCG, PE-H (carboxylic ester hydrolase from *Pseudomonas aestusnigri*) Best surface-display combination: pGSA-FAST-PETase	71.3% weight loss in 24h for 200-mesh PET powder at 50 °C using FAST-PETase surface display Up to 84.6% weight loss when co-displaying FAST-PETase and MHETase Particle size effect: 50 mesh: 34.1% 2000 mesh: 68.4% PET bottle hydrolysis: Neck (low crystallinity): 52.9% Base: 31.4% Body (high crystallinity): 10.2%	24 hours	1. This study shows viable, scalable, and energy-efficient PET recycling at moderate temperatures (50 °C). 2. No harsh chemicals involved, using biological display on *E. coli*. 3. Effective on real-world PET bottles, especially lower crystallinity parts. 4. Could reduce landfill PET waste if optimized for mass deployment.	At high temperatures, enzyme loses activity quickly (40% drop at 60 °C)

Table 7. Detailed information on articles related to others enzymatic approaches.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
39187447 (77)	1. A PET hydrolase gene was identified in Streptomyces spp. via genome mining. 2. Results show the enzyme can potentially act in a multi-enzyme PET-degradation cascade.	BHET	NA	Nigeria	Enzymatic degradation	Enzyme: Putative PET hydrolase	No direct PET degradation. BHET- releasing MHET & TPA	24 hours at pH 8 & 37 °C	1. The enzyme shows potential for biocatalytic recycling of PET waste via intermediate breakdown (BHET). 2. The enzyme works at mesophilic temperatures, making it suitable for mild industrial/bioremediation conditions.	1. PET polymer was not directly tested. 2. Structural model was based on in silico prediction, not crystallized.
39561546 (78)	1. The paper explores polyesterase activity, including activity toward polyethylene terephthalate (PET), across a range of carboxylesterases (26 family) in family IV, and compares it with known PETases. It evaluates their ability to hydrolyze PET and other polyesters using HPLC, agarose clearing assays, and biochemical characterizations.	Polyethylene terephthalate (PET)	Virgin	Bangor University (UK), University of Toronto (Canada) CSIC, Madrid, Spain	Enzymatic	IS12 (benchmark from family IV), GEN0105 (main impactful, high PET activity), CBA10055 (thermostable, moderate activity), MGS-MesE1, BHR01264, Node152, Node186 (ancestral variants) Comparators: IsPETase, FAST-PETase, HotPETase, TfCut1, TfCut2, LCC-WCCG	NA	12-hour reaction time for HPLC-based degradation assays at 60–70°C	Demonstrates new classes of enzymes (family IV carboxylesterases) capable of PET degradation, expanding the toolbox for enzymatic plastic recycling.	1. Most enzymes in family IV showed weak to moderate activity on actual amorphous PET compared to high activity on model substrates (3PET). 2. No full degradation of PET was achieved by most family IV enzymes. 3. GEN0105 showed the highest PET degradation among the family IV group, but still less effective than benchmark PETases.

38406590 (79)	1. PET30, a cold-active esterase from *Kaistella jeonii*, can bind and degrade PET, though with lower catalytic efficiency than IsPETase.	Polyethylene terephthalate (PET)	NA	Brazil, Germany	Enzymatic degradation	PET30 (cold-active esterase from *Kaistella jeonii*) IsPETase used for comparison only	NA	NA	PET30 may serve as a candidate for bioremediation due to its cold-active nature.	1. Lower catalytic efficiency compared to IsPETase. 2. The enzyme likely represents an evolutionary precursor to more efficient PET-degrading enzymes. 3. Results are entirely computational; no in vitro or in vivo validation.
35850328 (80)	1. The engineered yeast strain (AJD2 pAD PET_IS) overexpresses PETase (an enzyme originally from *Ideonella sakaiensis*) which actively degrades PET films and powder.	Polyethylene terephthalate (PET) powder and film	Virgin	Wrocław, Poland	Enzymatic degradation	PETase, the engineered yeast strain *Yarrowia lipolytica* (strain AJD2 pAD PET_IS)	*Y. lipolytica* PETase strain + 1 % olive oil → 53.1% PET film weight loss 44.7% without supplementation(oil)	7-10 days	1. Using an engineered GRAS (Generally Recognized as Safe) yeast to produce PETase enables biodegradation under mild conditions (ambient temperature, moderate pH) without the need for high-energy inputs. 2. Reducing energy consumption and plastic waste while yielding reusable monomers.	1. Certain inorganic salt additions (e.g., $MnSO_4$ and $CuSO_4$) that enhanced degradation in small-scale experiments did not translate well during scale-up; in some cases, they reduced the degradation rate. 2. The overall efficiency is highly dependent on the medium composition.
35981419 (81)	1. The crystal structure of Ple629 was solved in complex with a PET monomer analogue (MHETa), revealing its unique substrate-binding mode compared to other PET hydrolases.	Polyethylene terephthalate (PET)	Virgin PET	China	Enzymatic degradation	Ple629 Enzyme	D226A/S279A: 5.5-fold improvement in PET hydrolysis over wild-type	24 hours	1. These findings provide a structural and mechanistic basis for further enzyme engineering toward efficient biotechnological recycling of PET.	While the study successfully improved activity and thermostability by targeted mutagenesis, it also revealed that increased thermostability alone does not guarantee superior PET hydrolysis.

Table 8. Detailed information on articles related to *Ideonella sakaiensis*.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
34260304 (82)	1.The ΔPETase mutant lost its ability to degrade PET completely, proving PETase is the primary enzyme responsible for PET degradation. 2. The ΔMHETase mutant showed reduced but still partial PET degradation, indicating that MHETase helps in assimilation.	Polyethylene terephthalate (PET)	NA	Nara Institute of Science and Technology, Japan	microorganism based degradation	Microorganism/Bacterium: *Ideonella sakaiensis* Enzymes: PETase (PET hydrolase) MHETase (MHET hydrolase)	1. The wild-type strain degraded approximately 8% of the PET film weight. 2. The ΔMHETase mutant showed 20% of the degradation capacity of the wild-type strain. 3. The ΔPETase mutant showed 0% degradation, meaning PETase is essential for PET breakdown.	10 days	potential applications in bioconversion and plastic waste management.	1. The study analyze PET degradation only under controlled lab conditions. 2. The efficiency of degradation is still low (only 8% weight loss in 10 days).
34620982 (84)	1. *Ideonella sakaiensis* 201-F6 can not only degrade PET but also convert it into poly(hydroxyalkanoate) (PHA), a biodegradable plastic.	Polyethylene terephthalate (PET)	Virgin PET granules	Nara Institute of Science and Technology, Japan	Microbial degradation using (PET is first broken down enzymatically and assimilated into bacterial metabolism)	*Ideonella sakaiensis* 201-F6	7.9 g/L of PET degraded (~79%) 9.0% of PET carbon converted into PHA.	6 days	1. Reduces PET pollution by converting it into biodegradable PHA.	Only 9.0% of PET carbon is converted into PHA, meaning most PET is metabolized into other compounds (e.g., CO_2).

39110086 (85)	1. The paper's primary goal is to uncover the transcriptional regulation mechanisms that govern the expression of key PET degradation genes in *Ideonella sakaiensis*.	Polyethylene terephthalate (PET)	Virgin	Japan	microbial metabolism	*Ideonella sakaiensis*	NA	NA	The work improves our understanding of how *Ideonella sakaiensis* regulates key enzymes for PET degradation.	The study is limited to dissecting the transcriptional regulation of PET degradation genes; while it identifies MRP as a key activator for MHETase and TPA metabolic operon, it does not clarify the full regulatory network.
36103351 (83)	1. The study demonstrates that *Ideonella sakaiensis* is not only capable of aerobic PET degradation but is also a facultative anaerobe that ferments PET under oxygen-limited conditions.	Polyethylene terephthalate (PET)	Environmental	Cambridge, UK	Microbial fermentation under anaerobic conditions	*Ideonella sakaiensis*	75% of PET film degraded after 30 days in the anaerobic fermentation setup. Overall PET-to-acetate/ethanol conversion yield of approximately 15 mol%.	30 days	The process offers a sustainable route for PET upcycling by converting plastic waste into valuable chemicals (acetate and ethanol) or even electricity (when co-cultured with G. sulfurreducens) .	Low conversion rate and Slow Degradation rate.

Table 9. Detailed information on articles related to *Bacillus strains*.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
39379655 (90)	1. To explore the plastic-degrading potential of culturable bacteria isolated from the gut of Zophobas morio fed on different synthetic polymers, including PET. 2.The degradation effect was enhanced by abiotic pretreatment, though polymer surface morphology was less affected than chemical composition.	Polyethylene terephthalate (PET)	Virgin PET	Ferdowsi University of Mashhad, Iran	Microorganism-based degradation	Strain P9: *Bacillus safensis* Total of 15 strains isolated; P9 showed the highest PET degradation	Without pretreatment: 1.1% PET film weight loss (28 days) With thermal (80 °C, 10 days) or sulfuric acid pretreatment: up to 1.9% weight loss Approximate 1.7× improvement with sulfuric acid	28 days	Suggests promising use of superworm-derived bacteria in integrated plastic waste management	1. PET degradation efficiency is still low (~2%), even after pretreatment. 2. The study did not identify or quantify specific degradation products or enzymes
38003625 (88)	1. A new PET-degrading strain, *Bacillus safensis* YX8, was isolated from PET waste and The strain degraded PET nanoparticles, producing TPA, MHET, and BHET. 2. Extracellular enzymes produced by strain YX8 were capable of PET hydrolysis, forming a clear zone on a polycaprolactone (PCL) plate.	Polyethylene terephthalate (PET)	Recycled PET	Nanjing Agricultural University, China	Microbial degradation	*Bacillus safensis* YX8 (Bacterium)	PET nanoparticles (low crystallinity) incubated with strain YX8 → degraded into TPA (4. µM), MHET (3.3 µM), BHET (0.4 µM)	15 days at 45°C for PET nanoparticle degradation 2 months for PET film degradation, but with very low efficiency	1. Contributes to sustainable PET biodegradation methods by identifying new PET-degrading bacterial strains. 2. Potential use of *Bacillus safensis* in microbial PET treatment systems.	1. Strain YX8 showed low efficiency in degrading PET film, which has a higher crystallinity. 2. Extracellular enzymes successfully degraded PET nanoparticles but had limited impact on high-crystallinity PET.

32333346 (89)	1. Bioprospecting identified *Bacillus muralis* as the most efficient PET-degrading strain. 2. PET from commercial water bottles was incubated with isolates; visible surface damage (cracks, erosion, pinholes) was observed via SEM. and FTIR showed chemical changes on PET after microbial treatment, notably in C=O and C–O bonds, indicating polymer breakdown.	Polyethylene terephthalate (PET)	Environmental	Veracruz State, Mexico	Microbial degradation (single strain bioprospecting method)	*Bacillus muralis*	PET film showed surface cracks, cavities, erosion, and pinholes after incubation with *Bacillus muralis*	after 3 days	Offer sustainable, site-adapted bioremediation strategies.	1. No quantitative PET degradation metrics and PET degradation only confirmed visually/analytically, not at a mass-conversion level. 2. Laboratory-only validation – real-world environmental performance of these strains still untested.
35003008 (86)	1. The study engineered artificial microbial consortia to improve the biodegradation of PET under ambient conditions. 2. Adding *Rhodococcus jostii* to form a three-species consortium further relieved inhibition by terephthalic acid (TPA) and increased degradation efficiency to 31.2% within 7 days.	Polyethylene terephthalate (PET)	Virgin	Tianjin University, Tianjin, China	Microbial degradation using an artificial microbial consortium	Engineered *Bacillus subtilis* (specifically the Bs-PETase strain that secretes PETase)	Up to 31.2% PET film weight loss within 7 days by three-species consortium (Bs_PETase + Bs_MHETase + R. *jostii*) the two-species consortium achieved 13.6%, and the four-species consortium reached 23.2%.	7 days	by incorporating bacteria that can metabolize toxic intermediates (TPA and EG), the consortium minimizes environmental pollution by converting these compounds further into CO_2 and water through the tricarboxylic acid cycle.	1. Accumulation of degradation products such as TPA and EG can inhibit PETase activity and bacterial growth. 2. Balancing the growth and activity of different species in the consortium is challenging—for example, *Pseudomonas putida* tends to overgrow in nutrient-rich media, which requires careful medium selection.

33850642 (87)	1. The study verifies that inoculation of compost soil with *Bacillus strains* (B. *subtilis* and B. *cereus*) accelerates the biodegradation of both PLA and PET films. 2. The combined presence of these *Bacillus strains* with Miscanthus (Miscanthus × giganteus) further enhances degradation.	Polyethylene terephthalate (PET) Polylactic acid (PLA)	Virgin	Poland	by bacterial strains	*Bacillus subtilis*, *Bacillus cereus*	No PET film weight loss was reported PET + B. *cereus* + Miscanthus → slight chemical degradation (carbonyl index drop by 14%)	over a period of 6 months under greenhouse conditions	1. The use of *Bacillus* strains (which produce robust endospores) and Miscanthus (a plant with low soil requirements and high biomass yield) suggests a sustainable strategy for converting contaminated soils into biologically and economically productive areas.	1. The study was conducted under controlled compost/greenhouse conditions; further research is needed to verify effectiveness under field conditions. 2. While the study provides several indicators (carbonyl index, tensile strength, weight loss, gas permeability), the exact correlation to complete mineralization (i.e., conversion to CO_2, CH_4, water, and biomass) is not quantified.

Table 10. Detailed information on articles related to *Comamonas testosteroni*.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
35293270 (91)	1. It presents a bio-chemical system using alkali-resistant *Comamonas* testosterone F6 and surfactants (especially Tween 20) to enhance PET microplastic degradation in alkaline medium.	Polyethylene terephthalate (PET)	Virgin PET	Tiangong University, China	Microbial degradation	Alkali-resistant *Comamonas* testosterone strain F6 (whole-cell biocatalyst)	*Comamonas* testosteroni F6 + No surfactant → 8.04% *Comamonas* testosteroni F6 + Tween 20 → 11.04% *Comamonas* testosteroni F6 + Tween 80 → 9.83%	in 30 days	1. Demonstrates a biocompatible method to degrade microplastics. 2. Enhances PET biodegradation without using high-energy inputs. 3. Potentially applicable in wastewater treatment and eco-textiles.	1. Requires pre-processed PET microplastics (1 mm) — limited applicability to intact PET. 2. Enzyme composition and degradation pathway details are not fully elucidated. 3. System effectiveness without surfactant is limited.
30961251 (92)	1. Alkaline whole-cell biocatalysis (strain F6) was more effective than neutral (F4) or alkali-only conditions. 2. Whole cell biocatalysis avoids product inhibition, as cells metabolize TA and related products.	Polyethylene terephthalate (PET)	Virgin PET	Tianjin Polytechnic University, China	Microorganism-based – using bacterial whole-cell biocatalysts	Organisms: *Comamonas testosteroni* F4 (neutral strain), *Comamonas testosteroni* F6 (alkali-resistant engineered strain)	PET particle size reduced from 7.3 µm to 1.58 µm (alkaline F6) and 2.63 µm (neutral F4) in 48 hours TA concentration peaked, then decreased, indicating microbial uptake of PET monomers	over 48 hours	1. Supports eco-friendly PET degradation using whole cells instead of purified enzymes. 2. Alkaline conditions enhance PET hydrolysis, while simultaneous microbial assimilation avoids toxic accumulation of monomers.	1. PET degradation was partial, not full mineralization. 2. Reaction limited by PET crystallinity and availability of extracellular enzyme expression. 3. Results specific to lab conditions with prepared micro-PET, not field or real-world plastics.

34057546 (93)	1. *Comamonas testosteroni* strain 3a2 was isolated from petrochemical wastewater and shown to degrade terephthalic acid (TPA)—a direct monomer of PET—as part of the degradation pathway of para-toluic acid (pTol).	Terephthalate	NA	Izmir, Turkey	Microorganism-based degradation using an aerobic bacterial strain (*Comamonas testosteroni* 3a2), relying on enzymatic oxidation.	Microorganism: *Comamonas testosteroni* strain 3a2 (Main impactful organism) Enzymes (detected and expressed): TPADO, TADO, PDO	Complete degradation of 107 ± 3 mg/L of TPA in 12 hours	12 hours	1. Efficient mineralization of TPA could prevent its environmental accumulation after enzymatic or hydrolytic PET depolymerization which suggests value in bioaugmentation strategies for PET waste treatment systems where PETase/MHETase enzymes liberate TPA.	1. PET polymer was not directly used or degraded in this study. 2. TPA degradation was tested in controlled aqueous cultures, so real-world environmental effectiveness is unverified.
39708916 (94)	1. Engineered *Comamonas testosteroni* RW31 to express PET-degrading enzymes (FAST-PETase & IsMHETase). 2. Biofilm formation on PET surfaces was observed, potentially enhancing degradation.	Polyethylene terephthalate (PET)	Virgin	Centro de Investigaciones Biológicas Margarita Salas (CIB-CSIC), Spain	Microbial degradation	Microorganism: *Comamonas testosteroni* RW31 (genetically engineered strain) Enzymes: FAST-PETase & IsMHETase (heterologously expressed)	In vitro: 37.1% PET weight loss In vivo: 0.83% PET weight loss	7 days (in vitro degradation with purified enzymes at 40°C) 30 days (in vivo degradation by *Comamonas testosteroni* RW31 in culture medium)	Supports upcycling of PET waste into PHB, a biodegradable bioplastic.	1. Low in vivo PET degradation efficiency (0.83% weight loss in 30 days). 2. PHB production yield is still relatively low compared to other bacterial strains.

39708756 (95)	1. *Comamonas testosteroni* F4 was used as a whole-cell biocatalyst to degrade PET. 2. Surfactants Span-80 and TRE significantly enhanced biofilm formation, enzyme secretion, and PET degradation efficiency.	Polyethylene terephthalate (PET)	Virgin	Tiangong University, Tianjin, China	Whole cell biodegradation using bacterium	Bacterium: *Comamonas testosteroni* F4	Control (no surfactant): 2.23% after 4 weeks Span-80 surfactant: 2.78% in 2 weeks, 3.51% in 4 weeks PQ-7 (cationic surfactant): 3.67% in 4 weeks SDS (anionic surfactant): 1.04% in 2 weeks Renewal of bacterial cultures (RBC) + Span-80: 5.67%	Tracked over 4 weeks	1. Avoids chemical solvents, high temperatures, or incineration. 2. Biodegradable surfactants (Span-80 and TRE) are non-toxic and metabolizable by bacteria, enhancing sustainability.	Degradation efficiency still relatively low (5.67% in 4 weeks)

Table 11. Detailed information on articles related to *Rhodococcus species*.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
40192096 (96)	1. The bacterium was immobilized in kappa carrageenan cryobeads to improve degradation efficiency and stability. 3. Repeated freeze-thaw cycles significantly enhanced the mechanical strength of the immobilized system.	terephthalic acid (TPA)	NA	IIT, Delhi, India	Microbial	*Rhodococcus erythropolis* MTCC 3951	100% degradation of 5 mM TPA KC beads with 5 freeze-thaw cycles (Set 3) → reusable for 10 cycles (~92% removal in cycle 10)	14 hours	1. Promotes sustainable bioremediation of TPA, a PET degradation intermediate. 2. Immobilized system reduces secondary pollution and allows for reuse.	Focuses only on TPA, not full PET polymer degradation and also real-world PET waste (contaminants, additives, crystallinity) not tested. 2. Slight decrease in efficiency after extended reuse cycles (10+) due to structural weakening and microbial viability loss.
37794223 (97)	1. The study focuses on TPA, which is a monomeric breakdown product of PET. 2. This is the first report of this bacterial strain degrading such a high TPA concentration.	Terephthalate	Environmental and Industrial recycled	India	Microorganism-based	Microorganism: *Rhodococcus erythropolis* MTCC 3951 Enzyme: Protocatechuate 3,4-dioxygenase (P34O)	5 mM TPA: fully degraded in 10 h 120 mM TPA (≈20 g/L): completely degraded in 84 h (High efficiency)	5 mM: 10 hours 120 mM: 84 hours	Demonstrates removal of TPA from real industrial wastewater	1. Not applicable to PET polymer; the study only covers TPA. 2. Performance in mixed pollution environments with other plastic components remains unknown.

39706601 (98)	1. *Rhodococcus rhodochrous* IITR131 was identified as an efficient PET-degrading bacterium. 2. The study analyzed the biochemical and molecular pathways responsible for PET degradation.	Polyethylene terephthalate (PET)	Virgin PET	NA	Microbial degradation	*Rhodococcus rhodochrous* IITR131	PET film (0.1 mm thickness) incubated with IITR131: 4.9% weight loss after 15 days 8.3% weight loss after 30 days 19.7% weight loss (0.1 mm → 0.081 mm) after 60 days	60 days	1. Demonstrates the potential of *Rhodococcus rhodochrous* IITR131 for PET biodegradation. 2. Suggests biotechnological applications for PET waste management and bio recycling.	NA

Table 12. Detailed information on articles related to *Pseudomonas species*.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
39701409 (99)	Not direct PET degradation but focuses on PET-derived monomer metabolism 1. Engineered strains grew faster on TPA but showed reduced efficiency on EG utilization. 2. Demonstrated the potential of ALE for optimizing microbial PET upcycling strategies.	Polyethylene terephthalate (PET) (studied through its degradation product, terephthalic acid, TPA)	Chemically deconstructed PET waste (TPA and ethylene glycol) (Recycled)	USA, Denmark	Microbial degradation	Microorganism: *Pseudomonas putida* KT2440 (engineered strain)	Growth rate on TPA improved by 4.1-fold after ALE Growth rate on TPA + EG improved by 3.5-fold	NA	1. Potential to convert PET-derived TPA into high-value biochemicals. 2. Supports the development of biocatalysts for sustainable plastic recycling.	Engineered strains performed worse on ethylene glycol (EG), requiring further optimization.
829485 (100)	1. The study investigated bacterial degradation of phthalate esters and their hydrolyzed forms (phthalate anions) where terephthalate (the aromatic core of PET) was biologically oxidized by pseudomonads and other aerobic bacteria. 2. Pathway= Ester → monoester → phthalate → 4,5-dihydroxyphthalate → protocatechuate → ring-cleavage.	Terephthalate anion	Synthetic terephthalate and phthalate esters	University of Miami School of Medicine, USA	Microbial degradation using aerobic pure bacterial cultures	Bacteria: *Pseudomonas fluorescens*, *Pseudomonas testosteroni*, *Micrococcus sp.* Enzymes: Phthalate 4,5-dioxygenase, 4,5-dihydroxyphthalate decarboxylase, Protocatechuate 3,4-dioxygenase	P. *testosteroni* grew on terephthalate within ~24–48 h lag, then utilized them completely.	7 days	1. Demonstrated that terephthalate (PET monomer) is readily biodegradable by soil and aquatic bacteria. 2. Suggests PET biodegradation may be possible in multi-step systems, where hydrolysis is followed by aromatic ring degradation	1. Focused on small molecule esters and terephthalate, not high-molecular-weight PET. 2. Results are foundational but require modern enzymatic or polymer-degrading validation

Table 13. Detailed information on articles related to other single species.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
38459363 (102)	1. *Brucella* intermedia IITR130 was isolated from a contaminated lake ecosystem.	Polyethylene terephthalate (PET)	Virgin	India	Microbial biodegradation	*Brucella* intermedia IITR130	26.06% weight loss of the PET sheet after 60 days Calculated half-life of PET degradation: 137.8 days	60 days (26.06%)	1. A microbial approach to PET degradation that is environmentally friendly compared to conventional thermal or chemical recycling methods.	1. Although the strain degrades 26.06% of the PET sheet in 60 days, the overall rate is relatively slow (half-life of 137.8 days). 2. The efficiency of degradation may be limited by the physical and chemical properties of PET, such as its crystallinity and smooth surface, which could restrict enzyme accessibility.
34678385 (103)	1. Serratia plymuthica IV-11-34 showed clear respiratory activity and CO_2 release when cultured with PET films.	Polyethylene terephthalate (PET)	Virgin PET	Nicolaus Copernicus University, Poland	Microbial degradation	Serratia plymuthica IV-11-34 (isolated from willow rhizosphere at landfill site)	~7% biodegradation (measured by CO_2 evolution)	40 days	1. Shows eco-compatible biofilm-based PET degradation using a Rhizospheric bacterium. 2. Suggests potential for bioremediation of PET-contaminated soils.	1. PET degradation rate is low (7%), much slower than for PLA. 2. No detection of degradation intermediates (TPA/MHET). 3. Inference of degradation is based on indirect measurements (CO_2, gene expression)

38944007 (101)	1. The paper describes the isolation of *Gordonia sp.* CN2K, a bacterium capable of utilizing PET as the sole carbon and energy source. It demonstrates PET degradation through metabolite analysis, enzyme activity assays, and structural changes of PET sheets under SEM.	Polyethylene terephthalate (PET)	Virgin	Kalaburagi, Karnataka, India	Microbial degradation	bacterium: *Gordonia sp.* CN2K enzymes: Esterase, PCA 3,4-dioxygenase, Catechol 1,2-dioxygenase	40.43% weight loss of PET	45 days under shaking culture in mineral salts medium.	1. The study provides eco friendly and non-toxic biological PET degradation without secondary pollutants. 2. Conversion of endocrine-disrupting intermediate TPA into protocatechuate (PCA), and then into TCA-cycle metabolites, ensures complete mineralization of PET.	Mixed thermophilic microbial community from seed sludge +N4 Dominant phyla: Thermotogae (42.9%) & Firmicutes (49.6%) Potential PET-associated degraders: Tepidanaerobacter syntrophicus, Clostridium_sensu_stricto_7
34320761 (105)	1. PET was included in acidogenic fermentation of food waste along with HDPE, PS, and PP. 2. PET underwent limited surface erosion and morphological change, indicating some biodegradation.	Polyethylene terephthalate (PET) Also tested: HDPE, PS, PP	Virgin PET	National University of Singapore (NUS), Singapore	Microbial biodegradation	Mixed thermophilic microbial community from seed sludge Dominant phyla: Thermotogae (42.9%) & Firmicutes (49.6%) Potential PET-associated degraders: Tepidanaerobacter syntrophicus, Clostridium_sensu_stricto_7	PET showed ~4.1% weight loss	25 days	NA	1. PET degradation was very limited (~4%), consistent with other anaerobic settings. 2. No chemical analysis of PET degradation byproducts (ex., MHET, TPA).

38360576 (104)	1. Developed P. pantotrophus DSM 2944 into a SynBio chassis capable of degrading PET monomers. 2. Wild type could metabolize EG (ethylene glycol) natively amd engineered to express tph operon (TphA1A2A3B, PcaK, PcaR) to degrade TPA.	PET monomers: TA and EG	Virgin	RWTH Aachen University, Germany	microbial biodegradation	Organism: Paracoccus pantotrophus DSM 2944 Genes/enzymes: TphA1, A2, A3: terephthalate dioxygenase complex, TphB, PcaK, PcaR	EG: fully consumed within 30 h TA: ~20% consumption (4 mM from 20 mM in same period)	30 hours	1. Demonstrates microbial platform for bioconversion of PET monomers to biomass, supporting circular bioeconomy. 2. No toxicity or environmental persistence studies included.	1. PET polymer itself was not tested. 2. TA degradation was partial, indicating transport/permeability issues. 3. Focused on proof-of-concept for metabolic engineering, not complete PET mineralization.

Table 14. Detailed information on articles related to defined co-cultures & mixed isolates.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
32510669 (106)	1. The strain first converted BHET into MHET, then into TPA. 2. A new esterase gene (EstB) was cloned and expressed, encoding the enzyme EstB, which hydrolyzes BHET into MHET.	BHET	NA	Zhejiang University of Technology, China	Microbial degradation (whole-cell biocatalysis) and enzymatic hydrolysis (EstB)	Microorganism: Enterobacter *sp.* HY1 Enzyme: EstB esterase	BHET degradation: 80.8% of 1000 mg/L BHET	within 120 hours	1. The strain can help in eliminating toxic PET intermediates (BHET, MHET, TPA). 2. May help in complete PET mineralization in microbial consortia or engineered bioreactors	The study does not prove direct PET degradation.
38219583 (107)	1. The research explores the strain's ability to degrade 11 different toxic allelochemicals and phthalate esters, including TPA (terephthalic acid), using two dioxygenase enzymes: TphA1A2A3 (terephthalate 1,2-dioxygenase) and OphA1A2 (phthalate 4,5-dioxygenase).	Terephthalic acid (TPA)	NA	Wuhan, China	Microbial driven enzymatic degradation	Bacterium: Pigmentiphaga kullae CHJ604 Enzymes: TphA1A2A3 (terephthalate 1,2-dioxygenase), OphA1A2 (phthalate 4,5-dioxygenase)	Complete degradation of 332.26 mg/L TPA	5 days	1. This bacterium can detoxify agricultural soils polluted with plastic-related xenobiotics like TPA and phthalates. 2. Enzyme broad substrate specificity makes it promising for bioremediation of PET-degraded products.	No PET polymer tested directly and functional in monomer degradation, but polymer breakdown would need combined PETase pre-treatment.

39615570 (108)	1. The work strongly highlights the construction of microbial consortia that could degrade PET along with other polymers, emphasizing the synergistic microbial interactions and enzyme complementation strategy.	Polyethylene terephthalate (PET)	Both virgin and recycled PET	University of Almeria, Spain	Microbial (fungi + bacteria-based consortia with enzymatic synergy)	Bacteria: *Bacillus* spp. (*subtilis* RBM2), *Pseudomonas aeruginosa* RBM21 Fungi: *Fusarium oxysporum* RHM1, Scedosporium apiospermum RHM14	Consortium bb: Bs-Psa (B. *subtilis* RBM2 + P. *aeruginosa* RBM21) → highest growth (10^9 CFU/mL) on PET-V & PET-R	14 days	1. Using microbes adapted to harsh composting conditions ensures robust enzyme activity. 2. Potential to reduce PET accumulation in agricultural waste environments, if scaled.	1. PET degradation is not quantified. 2. Synergistic effects complex; not all consortia showed improved degradation (e.g., adding F. oxysporum to some consortia reduced plastic range).
37327543 (109)	1. PET microplastics were found in reservoir sediments and showed size reduction in the presence of microorganisms under elevated hydrostatic pressure.	Polyethylene terephthalate (PET)	Environmental	Shaanxi Province, China	Microbial degradation	*Rhodococcus*, Flavobacterium, Bradyrhizobium, tphA3 gene	0.7 MPa hydrostatic pressure → 366.62 µm PET microplastic size reduction (13.66% size reduction)	30 days at 0.7 MPa pressure	Demonstrates that deep reservoir sediments can facilitate PET biodegradation through pressure-driven microbial activity.	1. PET degradation is partial and slow and Only PET particle size was reduced—not full depolymerization. 2. PET-degrading genes (like tphA3) are present but low in abundance, limiting efficiency.

38809402 (110)	1. Five *Comamonas testosteroni* strains (3APTOL, 3ABBK, 2B, 3A1, C8) were tested for their ability to degrade TPA and all strains completely degraded 100 mg/L TPA within 6 to 8 hours, depending on the strain.	Terephthalate	Virgin	Denizli, Turkey	Microorganism-based (Using bacterial degradation under aerobic fermentation conditions)	Microorganism: *Comamonas testosteroni* strains: 3APTOL, 3ABBK, 2B, 3A1, C8 Enzymes (genes encoding them): tphA2, tphA3, pmdA, pmdB, ophA2	100% degradation of 100 mg/L TPA Strain C8: 100% in 6 hours Other strains: 100% in 8 hours	6-8 hours	1. Although PET polymer wasn't tested directly, this study indicates the efficient microbial degradation of TPA, an essential intermediate in PET breakdown.	1. The study does not address PET polymer degradation. 2. Strain-specific differences in gene presence (e.g., lack of pmdB in strain C8) suggest incomplete understanding of alternative degradation pathways.
32290598 (111)	1. Identification of PET hydrolase candidate genes belonging to the superfamily of alpha/beta-hydrolases, indicating metabolic potential for PET degradation. 2. Discovery of enzyme-coding sequences potentially capable of PET hydrolysis.	Polyethylene terephthalate (PET)	Environmental	Apatlaco River, Morelos, Mexico	Microorganism-based biodegradation (environmental)	Enzymes: Alpha/beta hydrolases (mainly impactful) Microorganisms: Flavobacterium aquicola, Bordetella flabilis, Flavobacteriia bacterium	NA	NA	Natural microbial communities from polluted environments (such as Apatlaco River) can degrade PET and other plastic pollutants, thus contributing significantly to bioremediation and reducing plastic pollution.	No quantitative measurements provided regarding the exact efficiency or rate of PET degradation.

38002377 (114)	1. A two-step chemo-microbial degradation process was developed: Step 1: PET was depolymerized chemically using an orange peel ash (OPA) catalyst in glycolysis, yielding primarily bis(2-hydroxyethyl) terephthalate (BHET). Step 2: A microbial consortium (comprising three *Pseudomonas* and two *Bacillus* species) biologically degraded BHET completely into PET monomers (terephthalic acid (TPA) and ethylene glycol (EG)).	Polyethylene terephthalate (PET)	Recycled	Reed College, Portland, USA	Chemical (glycolysis) depolymerization using an OPA biomass-waste catalyst Microbial consortium degradation of chemically depolymerized BHET products	Microorganisms- Three *Pseudomonas species*, Two *Bacillus* species	Chemical Depolymerization Step (Glycolysis): 1.PET to BHET conversion efficiency: 92% yield 2.Complete PET conversion was achieved chemically. Microbial Degradation Step (Consortium): 1. BHET biodegradation: 19% (2 weeks), 35% (10 weeks) 2. Total combined chemical + microbial degradation (over 10 weeks): 33% without hydrolysis ~56.5% in 2 weeks including hydrolysis	Chemical step- 1.5 hours Microbial degradation- 10 weeks (35%)	Utilizes biomass-derived catalyst (OPA), which is environmentally friendly, sustainable, and recyclable.	1. Complete degradation (100% efficiency) was not achieved within the tested timeframe (max 35% after 10 weeks). 2. The presence of impurities (from biomass catalyst) potentially impacts biodegradation efficiency or product recovery processes.

36228720 (115)	1. Several microbial strains were isolated from plastic waste–polluted sediments. The study focused on both individual isolates and a microbial consortium (MC).	Polyethylene terephthalate (PET)	Environmental waste	India	Microbial biodegradation (in vivo)	Combination of Sarcina aurantiaca (TB3) + *Bacillus subtilis* (TB8) + *Aspergillus flavus* (STF1) + *Aspergillus niger* (STF2)	Sarcina aurantiaca (TB3) → ~18% *Bacillus subtilis* (TB8) → ~20% *Aspergillus flavus* (STF1) → ~22% *Aspergillus niger* (STF2) → ~25% Microbial consortium (MC) → 28.78%	over 60 days	The microbial consortium degrades PET in vivo without posing additional health or environmental risks.	1. Slow Degradation Rate. 2. The biodegradation process was conducted in controlled laboratory media (BHM and MSM), which may differ from environmental conditions. 3. In vivo Complexity: – The microbial degradation in a natural setting may be influenced by many additional factors such as competition, environmental fluctuations, and bioavailability of PET.
37688172 (112)	1. Two bacterial strains isolated from insect guts (from Hymenoptera: Xylocopa appendiculata and Eumenes decorates) were shown to degrade PET. 2. Morphological changes in PET films (e.g., erosion, scratches, and roughening) were confirmed via scanning electron microscopy (SEM).	Polyethylene terephthalate (PET)	Virgin-Commercial PET film and PET powder	Korea	microbial metabolism	Microorganisms: Xanthomonas *sp.* HY-74, *Bacillus sp.* HY-75	PET film weight loss: HY-74: 1.57 ± 0.21% HY-75: 1.42 ± 0.46%	PET film degradation was measured over 6 weeks PET powder degradation was assessed after 4 weeks	NA	1. The degradation efficiency is relatively low over the tested period. 2. The specific enzymes and metabolic pathways responsible for PET breakdown remain unidentified.

35460002 (116)	1. Three rhizospheric bacterial isolates were identified as Priestia aryabhattai VT 3.12, *Bacillus pseudomycoides* VT 3.15, and *Bacillus pumilus* VT 3.16. 2. Biofilm formation was observed on PET, which facilitated degradation.	Polyethylene terephthalate (PET)	Environmental samples (PET collected from plastic waste-contaminated soil)	Jalandhar, Punjab, India	Microbial degradation using rhizobacteria	Microorganisms identified: 1. Priestia aryabhattai VT 3.12 2. *Bacillus pseudomycoides* VT 3.15 3. *Bacillus pumilus* VT 3.16	PET sheet degradation: 40% (VT 3.12) 36% (VT 3.15) 32% (VT 3.16) PET powder degradation: 69% (VT 3.12) 66% (VT 3.15) 64% (VT 3.16)	PET sheets-28 days PET powder-18 days	1. Rhizobacteria show strong potential for PET degradation in soil environments, making them suitable for eco-friendly plastic waste management. 2. Suggests the possibility of biotechnological applications in sustainable PET waste recycling.	1. Only lab-scale experiments were conducted, requiring further testing in real-world environmental conditions. 2. PET degradation rates vary based on physical form—powder degrades much faster than PET sheets.
38519709 (117)	1. PET degradation was enhanced by enzymatic activities (laccase and esterase), biofilm formation, and bacterial colonization.. 2. BS5 was the most efficient PET degrader, producing 17.11 U/mL of laccase and 1.1 U/mL of esterase, leading to 4.44% PET weight loss in 4 weeks.	Polyethylene terephthalate (PET) (along with LDPE and PS)	Virgin PET granules	Malaysia	Microbial degradation using bacterial isolates	Microorganisms: Dermacoccus *sp.* MR5 Corynebacterium *sp.* MR10 *Bacillus sp.* BS5 (best PET degrader) Priestia *sp.* TL1 Enzymes Produced: Laccase, Esterase	4.44% weight loss (BS5 strain) Laccase production: 17.11 U/mL Esterase production: 1.1 U/mL	4 weeks	1. UV pretreatment + bacterial degradation could improve PET biorecycling strategies. 2. The process could help in plastic waste management and pollution reduction.	1. PET degradation efficiency (4.44%) is still low. 2. Some bacterial strains (Dermacoccus *sp.* MR5) showed reduced growth and enzyme production after 3 weeks, indicating possible nutrient depletion or inhibitory byproducts.

33361127 (113)	1. The screening targeted bacteria capable of breaking down PET as a sole carbon source. 2. The full consortium had a shorter doubling time and higher efficiency compared to individual bacterial strains.	Polyethylene terephthalate (PET)	Environmental Sample (PET debris from petroleum-contaminated soils)	Texas, USA	Microbial degradation	*Bacillus thuringiensis* strain C15 (strain 9.1) *Bacillus albus* strain PFYN01 (strain 13.1) *Pseudomonas sp.* B10 (strain 9.2) *Pseudomonas sp.* SWI36 (strain 10) *Pseudomonas sp.* SWI36 (strain 13.2)	Full consortium caused ~3% PET weight loss after 8 weeks Complete conversion of BHET to TPA and EG confirmed	8 weeks at 30°C	The study suggests bioaugmentation with bacterial consortia as a strategy for PET degradation.	Slow degradation compared to chemical methods.

Table 15. Detailed information on articles related to environmental aspects.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
28964604 (118)	1. Eight bacterial strains were isolated from mangrove sediments across Peninsular Malaysia. Among them, *Bacillus cereus* and *Bacillus gottheilii* demonstrated the ability to grow on and degrade PET microplastics. 2. UV-treated PET particles were used as the sole carbon source in mineral salt medium for 40 days.	Polyethylene terephthalate (PET)	Virgin PET	six sites across Peninsular Malaysia,	Microorganism-based biodegradation	*Bacillus cereus*, *Bacillus gottheilii*	*Bacillus cereus*: 6.6% weight loss of PET *Bacillus gottheilii*: 3.0% weight loss of PET	40 days incubation period	1. Isolated native bacteria from mangroves capable of degrading PET which indicates bioremediation potential for plastic-contaminated coastal soils and sediments. 2. PET degradation by *Bacillus* species could help reduce microplastic persistence, especially in tropical marine ecosystems.	Degradation of PET is slow and partial.

37838714 (125)	1. The research explores microbial community-level degradation of chemically deconstructed PET (DCPET) and its monomers. 2. *Rhodococcus* (generalist) and Hydrogenophaga (specialist) were key organisms in PET and monomer degradation.	Chemically deconstructed PET	Chemically deconstructed PET	Michigan Technological University, USA	Microbial community-mediated degradation	bacteria: *Rhodococcus* spp. (generalist, dominant in most conditions), Hydrogenophaga spp. (specialist, terephthalate degradation), Paracoccus, *Pseudomonas*, Shinella, Achromobacter, Variovorax, Mesorhizobium Enzymes mentioned by function (gene-level): Terephthalate 1,2-dioxygenase, Mono(2-hydroxyethyl) terephthalate hydrolase, Protocatechuate and benzoate pathway enzymes	NA microbial growth (OD600) increased substantially (up to ~1.6 OD units) on DCPET and terephthalamide, showing significant substrate utilization.	136 hours	Demonstrates feasibility of mixed community PET biodegradation, aiding biotechnological upcycling of unsorted plastic waste.	No direct enzymatic characterization and no quantification of monomer breakdown or conversion

37421171 (119)	1. Multiple deep-sea marine bacteria (from Alphaproteobacteria, Gammaproteobacteria, and Actinobacteria) can degrade PET. Degradation was performed under simulated marine conditions, confirming activity in deep-sea environments. 2. Genomic analysis revealed PETase-like and MHETase-like genes, suggesting enzymatic pathways similar to those in *Ideonella sakaiensis*.	Polyethylene terephthalate (PET)	Environmental	Deep-sea sediments from the Western Pacific Ocean	Microorganism-based enzymatic degradation	Marinobacter sediminum BC31_3_A1 Marinobacter gudaonensis BC06_2_A6 *Thalassospira* xiamenensis BC02_2_A1 *Nocardioides marinus* BC14_2_R3	PET film weight loss: N. marinus BC14_2_R3 → 1.8% ±0.2% M. sediminum BC31_3_A1 → 1.3% ±0.2% M. gudaonensis BC06_2_A6 → 1.2% ±0.07% T. xiamenensis BC02_2_A1 → 1.3% ±0.2%	30 days	1. Marine bacteria with PET-degrading ability may play a role in the natural attenuation of plastic pollution in deep-sea environments. 2. Non-toxic, low-molecular-weight intermediates (TPA, MHET) indicate potential for safe environmental application.	1. PET was not fully mineralized; accumulation of intermediates (e.g., MHET) was noted. 2. Degradation rate is relatively slow under natural conditions.
36356186 (121)	1. Isolated 78 thermophilic bacteria from two Peruvian hot springs: El Tragadero and Quilcate. 2. 11 bacterial isolates demonstrated PET hydrolysis ability, producing visible halos on PET-agar plates. 3. Brevi*Bacillus* borstelensis showed PET degradation activity, previously known mainly for polyethylene degradation.	Polyethylene terephthalate (PET)	Environmental	El Tragadero and Quilcate hot springs, Peru	Microorganism-based biodegradation	Thermophilic PET-hydrolyzing bacterial strains: *Bacillus oceanisediminis* (ETP1), *Bacillus licheniformis* (ETP2, QP3, QP6, QP12, QP15, QP19, QP22, QP28), Brevi*Bacillus* borstelensis (QP8), Tistrella mobilis (QP27)	11 isolates formed visible clear halos around colonies, indicating PET hydrolysis Best isolates (largest halo formation): *Bacillus oceanisediminis* & B. *licheniformis*	2 days PET hydrolysis observed at 55–60°C over 24–48 hours post-inoculation.	1. Isolated strains could be employed for bioremediation of plastic pollution, particularly in high-temperature environments.	1. No quantification of PET degradation. 2. Enzymes responsible for PET degradation were not purified or characterized. 3. PET degradation demonstrated only under lab conditions, not in situ hot spring environmental trials.

40252025 (120)	1. Isolated deep-sea marine strain *Pseudomonas chengduensis* BC1815 from Pacific Ocean sediments and demonstrated PET film biodegradation and EG assimilation by BC1815 under laboratory conditions. 2. Identified and characterized a novel PET esterase (OF113_10420) anchored to the cell membrane.	Polyethylene terephthalate (PET)	Virgin	China (Pacific Ocean)	Microorganism-based enzymatic degradation	*Pseudomonas chengduensis* BC1815 PET esterase (OF113_10420)	PET weight reduced by 1.43% after 3 weeks whole-cell biocatalyst (OD600 = 1.0) on PET film at pH 9.5 = releasing 6.2 µM MHET BHET degradation (whole cells, 5 h): 78.3% converted to MHET (best at pH 8.0) MHET degradation (whole cells, 24 h): ~3.7% converted to TPA (best at pH 5.5)	3 weeks	1. Surface-anchored PET esterase minimizes enzyme loss to the ocean, favoring localized biodegradation. 2. Finding marine bacteria capable of EG metabolism is critical because EG pollution is increasingly recognized alongside TPA.	1. Relatively low PET degradation percentage (only ~1.43%) under lab conditions over weeks. 2. The PET esterase mainly produced MHET under neutral conditions; full mineralization limited. 3. Does not investigate cocultures combining TPA and EG degraders for full PET mineralization.
38734049 (123)	1. Bacterial consortia capable of degrading PET were isolated from 10 out of 19 deep-sea sediment samples collected from the Pacific Ocean (depths: 4862–5396 m).	Polyethylene terephthalate (PET)	Virgin	Eastern Central Pacific Ocean	Microbial degradation	Main Bacteria-*Alcanivorax sp.* A02-7, *Pseudomonas sp.* A09-2 Others-*Thalassospira*, *Nitratireductor*, *Nocardioides*, Muricauda, Owenweeksia, Alteromonas, Dietzia	PET weight loss observed= 1.8% to 16.2% (16.2% highest BC14 dominated by *Alcanivorax*, *Pseudomonas*, *Nitratireductor*, *Thalassospira* among 10 consortia)	over 60 days	1. Suggests that deep-sea bacteria like *Alcanivorax* and *Pseudomonas* can biodegrade PET even in harsh conditions.	1. Low degradation rate (max 16.2% in 60 days) compared to engineered systems. 2. Lack of in-situ validation; results are based on simulated conditions.

36330933 (124)	1. FTIR-ATR analyses revealed the emergence of new absorption peaks (around 1650 cm^{-1}, indicative of vinyl or carbonyl groups) and a reduction in peaks associated with the original polymer structure.	Biobased polyethylene terephthalate (BPET) Low-density polyethylene (LDPE)	Virgin	Marine caves, South Portugal	Microorganism	most effective-I18, recovered from the tunicate Didemnum *sp.* Others (nominal key role)-Halodesulfovibrio, *Pseudomonas*, and Tepidibacter	NA not any specific quantitative data	45 days	useful for marine microplastic pollution	Just early stage of biodegradation, not complete mineralization.
34646260 (122)	1. The marine *Bacillus* species AIIW2 was shown to degrade commercially available PET film.	Polyethylene terephthalate (PET) film	recycled PET	NA	Microorganism	Marine *Bacillus* species AIIW2 (KU877334)	PET film weight loss increased from 0.42% after 15 days to 1.93% after 90 days.	a period of 90 days	Marine *Bacillus* species AIIW2 is capable of initiating PET degradation and mineralization, which could contribute to long-term bioremediation strategies in marine environments.	1. PET degradation (1.93% weight loss in 90 days) remains low. 2. The controlled laboratory setup (using standard PET film, defined media, and constant temperature) does not fully replicate the complex conditions found in natural marine environments.

Table 16. Detailed information on articles related to other microbial-only approach.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
39135878 (126)	1. The study uses metagenomic analysis of porcine gut microbiota to identify the presence, diversity, and potential function of genes and enzymes involved in degrading PET. 2. Proposed a hypothetical PET degradation pathway involving oxidoreductases, hydrolases, ligases, isomerases, and laccases.	Polyethylene terephthalate (PET)	NA	Sichuan Agricultural University, Chengdu, China	Microbial biodegradation inferred through macro genomic functional gene profiling	porcine gut microbial community (Clostridium, Lacto*Bacillus*, Ruminococcus); MGene_174 carboxylesterase (novel PETase-like enzyme);	No direct PET weight loss experiments Identified 94 functional MP degradation genes, with PET as one of the top 3 targets (with PE & DEHP). Proposed that PET would be degraded in the gut.	NA	1. Suggests pigs and similar gut microbiota may play a role in reducing microplastic accumulation, including PET, in food animals and ecosystems. 2. Enriched PET-degrading microbes (e.g., Lacto*Bacillus*) could be harnessed for environmental or industrial plastic bioremediation.	1. No actual PET polymer degradation performed. 2. Pathway reconstruction based on bioinformatics inference, not wet-lab enzyme testing.

Table 17. Detailed information on articles related to combined approaches (enzyme + microorganism).

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
26965627 (127)	1. *Ideonella sakaiensis* 201-F6, a novel bacterium that uses PET as a primary energy and carbon source. 2. The bacterium secretes two enzymes: PETase: Hydrolyzes PET into MHET. MHETase: MHET into TPA and EG.	Polyethylene terephthalate (PET)	Environmental sample (from a PET-contaminated site)	Japan	Enzyme-based microorganism degradation	Bacterium: *Ideonella sakaiensis* 201-F6 Enzymes: PETase, MHETase	Almost complete PET hydrolysis	6 weeks at 30°C	1. The monomers TPA and EG are harmless to the environment and can be reused in plastic production. 2. The bacterium decomposes PET into non-toxic monomers (TPA and EG), which can be recycled.	1. Slow degradation rate takes weeks to degrade PET. 2. Not highly efficient in natural conditions: Works optimally in a controlled lab environment.
33579403 (128)	1. Microbial consortium no. 46 was discovered at a PET bottle recycling site, which degraded PET at room temperature.	Polyethylene terephthalate (PET)	Environmental sample (PET waste from a recycling site)	Japan	Enzyme-based microbial degradation	Bacterium: *Ideonella sakaiensis* 201-F6 Enzymes: PETase, MHETase	1. Amorphous PET was significantly degraded in lab conditions 2. Crystalline PET is more resistant to degradation.	6 weeks at 30°C	Degradation products (TPA & EG) are non-toxic and reusable, making this method eco-friendly.	1. Slow degradation rate 2. Less effective on crystalline PET
32343496 (132)	1. A thermophilic anaerobic bacterium, *Clostridium thermocellum*, was genetically engineered to secrete a PET-degrading enzyme (LCC - leaf-branch compost cutinase).	Polyethylene terephthalate (PET)	Commercial PET films (likely virgin)	Qingdao Institute of Bioenergy and Bioprocess Technology, Chinese Academy of Sciences, China & Greifswald University, Germany	Microbial-enzyme degradation	*Clostridium thermocellum* (engineered) Cutinase (LCC) Enzyme	Approximately 62% of PET film	over 14 days	promising biotechnological solution for PET bioremediation	1. MHET accumulation inhibits degradation, suggesting the need for additional MHET-hydrolyzing enzymes in the system.

33865980 (138)	1. Developed a bio-upcycling process for PET waste, integrating enzymatic hydrolysis and microbial conversion. 2. Used thermostable Leaf-Branch Compost Cutinase (LCC) to hydrolyze PET into monomers.	Polyethylene terephthalate (PET)	Recycled and environmental PET samples	RWTH Aachen University, Germany University College Dublin, Ireland Leipzig University, Germany Strasbourg University, France Bioplastech Ltd. (Industry partner), Ireland	1. Enzymatic degradation using Leaf-Branch Compost Cutinase (LCC). 2. Microbial degradation using engineered *Pseudomonas umsongensis* GO16 to metabolize TA and EG into bioplastics.	Leaf-Branch Compost Cutinase (LCC) *Pseudomonas umsongensis* GO16	~47% PET depolymerization	10–24 hours	1. Contributes to a circular economy by converting PET into valuable biodegradable materials (PHA & bio-PU). 2. Reduces plastic waste by replacing fossil-fuel-derived plastics.	1. LCC enzyme activity declines over time due to prolonged high-temperature exposure. 2. Engineered *Pseudomonas umsongensis* GO16 required ~45 days of evolution to efficiently metabolize EG.
33468707 (144)	1. The study compiles a database of plastic-degrading microorganisms and examines their distribution across microbial taxa. A total of 16,170 putative plastic degradation orthologs were identified across 6,000 microbial species. PET was one of the most documented plastics, with 8,233 putative PET-degrading genes found in microbial genomes.	Polyethylene terephthalate (PET) also polyhydroxybutyrate (PHB), polylactic acid (PLA), and polycaprolactone (PCL)	various environmental sources (including soil, plastic waste dumping sites, compost, and marine environments)	Global analysis	Microbial degradation (by using bacteria and fungi) Also enzymatic mechanisms	Microorganisms: Actinobacteria, Proteobacteria, *Pseudomonas species*, *Ideonella sakaiensis* Enzymes: PETase from *Ideonella sakaiensis*, cutinases, esterases	NA	NA	1. The study provides insights into the evolutionary potential of plastic biodegradation and its applications for bioremediation. 2. The global database can help identify new microbial candidates for biotechnological applications in plastic waste management.	PET degradation is still inefficient, particularly for high-crystallinity PET (most studies use amorphous PET).

36474116 (131)	1. Two PET-degrading bacteria, *Pseudomonas nitroreducens* S8 and *Pseudomonas monteilii* S17, were isolated. The combined bacterial culture demonstrated higher colonization and biofilm formation on PET surfaces, leading to enhanced degradation.	Polyethylene terephthalate (PET)	Recycled PET waste bottles and Virgin Amorphous PET film	China	Microbial degradation (bacterial) combined with enzymatic degradation (cutinase ICCG	Microorganisms: *Pseudomonas nitroreducens* S8 *Pseudomonas monteilii* S17 Enzyme: Engineered cutinase ICCG, expressed in *E. coli*	PET pretreated by combined P. *nitroreducens* S8 + P. *monteilii* S17 for 7 days, followed by ICCG cutinase hydrolysis → 94.5% degradation	6 days	1. The study proposes a scalable microbial-enzymatic PET degradation approach, reducing PET accumulation in the environment. 2. Biodegradation converts PET into TPA, BHET, and MHET, which are further metabolized, minimizing toxic accumulation.	1. While bacterial biofilm formation enhances PET degradation, it requires further optimization to improve efficiency. 2. Complete mineralization of PET degradation products was not achieved, as some monomers accumulated.
37714010 (135)	1. The study demonstrates a one-step biological process for upcycling PET oligomers—by-products from glycolysis-based chemical recycling—into polyhydroxyalkanoates (PHA), a biodegradable plastic.	PET oligomers	Recycled	Qingdao, China	Enzymatic depolymerization via microbial co-cultivation	LCC-ICCG (This PET hydrolase variant, secreted by *E. coli* BL21 (DE3)-LCC-ICCG, is the key catalyst for depolymerizing PET oligomers.)	The co-culture produced PHA with a yield of 1.10 g/L and a conversion rate of 0.11 g PHA per g of PET oligomer	72 hours	It avoids the quality loss typical of mechanical recycling and leverages biological methods for sustainable plastic upcycling.	1. The extracellular concentration of LCC-ICCG reached around 50 mg/L, which might require improvement for industrial scalability. 2. Maintaining optimal population ratios between the two engineered strains is critical.

36385587 (129)	1. The study investigated the biodegradation of two PET variants (a colored PET (PETcol) and a transparent PET (PETtra)) derived from food packaging containers using *Ideonella sakaiensis* (ISAK).	Polyethylene terephthalate (PET)	virgin	Innsbruck, Austria	Microorganism-driven enzymatic biodegradation	*Ideonella sakaiensis* degrades PET through its extracellular enzymes [PETase and MHETase] during its growth in liquid culture.	Transparent PET (PETtra): upto 96% Colored PET (PETcol): about 63.9% (±17.5%)	Transparent PET (PETtra): around 4 weeks Colored PET (PETcol): about 7 weeks	1. The demonstrated rapid biodegradation—especially of PETtra—by *Ideonella sakaiensis* suggests that biotechnological approaches can dramatically shorten the natural lifetime of PET.	1. Different PET variants show markedly different degradation rates; suggesting that factors like crystallinity, color additives, and possibly surface properties significantly influence the efficiency.
34864034 (143)	1. A novel stepwise screening and verification (SSV) strategy was developed to isolate bacteria that can use PET as a sole carbon source. Four bacterial strains were obtained, among which *Stenotrophomonas pavanii* JWG-G1 was highlighted.	Polyethylene terephthalate (PET)	Virgin PET	Wuxi, China	Combined method – microbial pretreatment followed by enzymatic degradation	*Stenotrophomonas pavanii* JWG-G1 *Thermobifida fusca* cutinase (TfC)	PET films pretreated with S. *pavanii* JWG-G1 achieved approximately 91.4% weight loss, which is 2.73 fold increased to non-pretreated PET films.	14 days	By combining microbial pretreatment with enzymatic hydrolysis, the process may offer an energy-efficient and sustainable alternative to chemical or thermal treatments, thereby reducing secondary pollution and lowering energy consumption.	1. The isolated strain S. *pavanii* JWG-G1, when used alone, showed very weak PET degradation (only 0.7% weight loss in 7 days). 2. The integration of microbial pretreatment with enzymatic degradation requires careful optimization of conditions for industrial scalability.
39992123 (149)	1. A novel screening method using PET-dependent bioactivity assays to search through hundreds of chemical/nutrient conditions for those that enhance the biodegradation of PET by Piscinibacter *sakaiensis*.	Polyethylene terephthalate (PET)	Virgin	Houston, Texas, USA	enzyme-dependent microbial activity	Piscinibacter *sakaiensis* (with its key enzyme IsPETase)	P. *sakaiensis* in 0.39% gm802 + 0.125% ethylene glycol → ~2× faster amorphous PET degradation over 5 weeks compared to control, reaching ~40–50% weight loss	5 weeks	By converting PET waste into microbial biomass—with byproducts limited to CO_2 and water—this method could reduce the accumulation of PET waste and mitigate the widespread issues of micro- and nanoplastic pollution.	only for amorphous PET. Some conditions showed notable variation in degradation efficiency between replicates, indicating challenges in reproducibility and the need for further optimization.

33752005 (145)	1. Identified genes r+B83esponsible for degradation of PET and other plastics from landfill soil, leachate, and compost using shotgun metagenomics.	Polyethylene terephthalate (PET)	Environmental	Pirana landfill site, Ahmedabad, Gujarat, India	Microorganism-driven enzymatic degradation	Enzymes- PETase, MHETase, Cutinases Microorganisms Identified (PET-degraders)- *Thermobifida fusca*, *Pseudomonas mendocina*, Nocardia *sp.*	NA	NA	The study confirms the presence of natural microbial populations with genes capable of degrading PET in real landfill environments, suggesting that native microbiomes can contribute to PET degradation.	The study only reports genetic potential, not actual PET degradation rates and no experimental validation of enzyme activity or PET breakdown was done.
39629164 (140)	1. Engineered yeast Yarrowia *lipolytica* (expressing PETase) was co-cultivated with *Pseudomonas stutzeri* (engineered to produce PHB) to couple PET depolymerization with PHB production in one step.	Polyethylene terephthalate (PET)	Virgin	Qingdao, China	enzyme-mediated microbial degradation	PETase (from *Ideonella sakaiensis*) expressed in *Yarrowia lipolytica* *Pseudomonas stutzeri* (TPA-degrading strain with phbCAB genes for PHB synthesis)	~3% of PET (0.31 g/L TPA from 10 g/L PET) was depolymerized in the co-culture	9.5 days	This one-step upcycling approach could help reduce PET waste by converting it into a biodegradable bioplastic (PHB)	PET depolymerization was very slow and inefficient.
33404203 (148)	1. P. *umsongensis* GO16 can utilize both monomers of PET (TA, EG)-as sole carbon and energy sources.	Polyethylene terephthalate (PET) (Monomers {TA and EG} used to simulate enzymatic PET hydrolysis)	Recycled	University College Dublin, Ireland, and University of Surrey, UK	Microbial driven enzymatic degradation	Microorganism: *Pseudomonas umsongensis* GO16 (primary PET monomer-degrading strain) Enzymes: TphA1, TphA2, TphA3 (Terephthalate 1,2-dioxygenase complex), TphB, TphK	NO PET polymer degradation, it focuses on the genome analysis of *Pseudomonas umsongensis* GO16, which is capable of metabolizing PET monomers (terephthalic acid & ethylene glycol).	NA	1. Demonstrates potential for biological upcycling of PET-derived waste into biodegradable plastics (PHA). 2. GO16 can valorize PET hydrolysates instead of complete mineralization to CO_2, helping preserve carbon value in new biomaterials.	1. No direct enzymatic hydrolysis of PET plastic tested; only PET monomers were used. 2. PHA yield is still low (9% CDW).

39557102 (136)	1. Developed a biological two-step process: (1) Enzymatic hydrolysis of micronized post-consumer PET using PES-H1 hydrolase. (2) Microbial upcycling of PET monomers (TA and EG).	Polyethylene terephthalate (PET)	Recycled	RWTH Aachen University and University of Greifswald, Germany Nanjing Tech University, China	Enzyme-based hydrolysis followed by microbial conversion	Enzyme: PES-H1 (a thermostable polyester hydrolase) Microorganism: *Pseudomonas putida* KT2440 PET2bio	This process demonstrates a two-step biocatalytic PET upcycling, but the enzyme and microbe did not act on PET polymer.	within 72 hours	This process uses waste PET as feedstock, contributing to carbon circularity, reduced plastic waste accumulation and replacement of fossil-based chemicals.	High phosphate buffer concentrations required for enzymatic hydrolysis inhibit microbial growth — necessitating hydrolysate dilution.
38040305 (134)	1. It explores the combined effects of a PET hydrolase enzyme (WCCG-a quadruple mutant of LCC cutinase which breaks ester bonds chemically) and plastic-degrading microbial communities (especially those in the plastisphere, especially during the thermophilic composting phase) during high-temperature composting.	Polyethylene terephthalate (PET)	Virgin	Shanghai, China	Enzymatic + Microbial composting	Microbes (from compost & plastisphere): natural compost microbiota enriched with *Bacillus*, Acinetobacter, Cladosporium, Aspergillus, Penicillium, and other thermophilic plastic-degrading bacteria/fungi. Enzyme: WCCG (quadruple mutant of leaf-branch compost cutinase LCC: F243W/D238C/S283C/Y127G, expressed in *E. coli*)	Without WCCG enzyme: PET-MPs reduced by 26% With WCCG enzyme: PET-MPs reduced by 35%	50 days	1. Composting method is low-cost, scalable, and environmentally friendly. 2. Encourages biofilm formation and long-term colonization of degraders.	1. Toxic breakdown products (e.g., antimony, phthalates) may affect microbial balance. 2. Incomplete PET degradation (~35% max in 50 days). 3. Compost maturity compromised by WCCG addition.
39360733 (142)	1. PET pellets and films underwent biofragmentation into nanoplastics and monomers (e.g., TPA) when incubated with *C. testosteroni* KF-1.	Polyethylene terephthalate (PET)	Virgin	USA	Enzymatic/Microbial Biodegradation	Microorganism: *Comamonas testosteroni* KF-1 Main Enzyme: CtesDRAFT_PD1902 (hydrolase) (not homologous to IsPETase but shows PETase-like structural binding motifs)	biofragmentation + BHET release 56%	upto 42 days	1. Demonstrates that *Comamonas testosteroni* from wastewater environments naturally produces PET-hydrolyzing enzymes.	1.PET degradation was partial and more effective on pellets (rougher surface) than films.

39206086 (139)	1. Developed engineered *Pseudomonas sp.* JY-Q strain displaying PETase and MHETase on its surface via Signal (OprF)-cOmpA anchoring system.	Polyethylene terephthalate (PET)	Virgin	Zhejiang University of Technology, China	Engineered microorganism-based enzymatic degradation	Microbe: *Pseudomonas sp.* JY-Q Enzymes (surface-displayed): FASTPETase-MHETase chimerase, non-tandem PETase + MHETase display variants also tested	Tandem FASTPETase-MHETase surface display → complete BHET hydrolysis in 72 h; PET films showed visible erosion after 28 days	PET film incubation: 28 days Complete BHET degradation: within 72 hours	1. Offers a safe, biologically based strategy to treat combined PET and PAE pollutants. 2. The use of whole-cell biocatalysts (JY-Q) reduces need for enzyme extraction/purification.	1. Optimization still required for enzyme expression levels, display system efficiency, long-chain PAE (DEHP) degradation was lower (~45%) compared to short-chain (DBP) PAEs.
36246227 (130)	1. A novel hydrophobic cell surface display (HCSD) system that immobilizes PETase on the surface of *E. coli* to efficiently degrade PET. 2. Enhanced degradation performance was observed due to improved contact angle and increased hydrophilicity of PET films after HCSD treatment.	Polyethylene terephthalate (PET)	Virgin	Beijing, China	Enzyme-based microbial whole-cell catalysis	Microbe: *Escherichia coli* BL21(DE3) (engineered for surface display) Enzyme: HCSD-PETase (PETase from *I. sakaiensis* fused with hydrophobin HFBII + FadL membrane anchor)	52% more TPA production over 7 days compared to free PETase.	7 days	1. The HCSD system improves PET degradation efficiency while avoiding enzyme purification, reducing biocatalyst cost. 2. Whole-cell immobilization strategy reduces waste, enhances reusability, and supports industrial application under moderate conditions (40°C).	1. Initial enzyme expression is lower compared to free PETase, leading to slower early-stage degradation.
38630153 (146)	The research investigates microbial enzymes and pathways involved in the degradation of PET, using shotgun metagenomics of pond sediment samples.	Polyethylene terephthalate (PET)	Environmental	Afelele Pond, Nigeria	Microbial enzymatic degradation	Enzymes: PETase, MHETase, PET hydrolase Microorganisms: Sphingobium *sp.*, Pseudomonadota members, Cesiribacter *sp.*, Novosphingobium *sp.*	NA	NA	1. The presence of naturally evolved PET-degrading enzymes and microbes indicates the environmental adaptation of microbiota to persistent plastics.	PET degradation is not experimentally quantified.

16607524 (147)	1. The strains PP4 and PPD degrade terephthalate (a PET monomer), isophthalate, and phthalate via specific dioxygenases.	Terephthalate	Virgin	IIT, Bombay, India	Enzyme-based microbial degradation	Microorganisms: *Pseudomonas aeruginosa* PP4, *Pseudomonas sp.* PPD, Acinetobacter lwoffii ISP4 Enzymes: Terephthalate dioxygenase (in PP4 and PPD), Phthalate dioxygenase, Isophthalate dioxygenase, Protocatechuate 3,4-dioxygenase (P3,4DO)	Only phthalate isomers (not PET) degraded. Terephthalate degraded fastest (doubling time ~3–4 h), isophthalate slower (~7 h).	20-24 hours	The study implies these soil bacteria could be useful in biodegradation of aromatic plastic monomers.	1. The study does not include PET polymer degradation. 2. The work is conducted under controlled lab conditions with defined media and purified substrates.
39461526 (137)	1. Engineered S. *pavanii* expressing DuraPETase significantly improved PET microplastic degradation at 30 °C. 2. Biofilm formation on PET surfaces by S. *pavanii* increased local enzyme concentration and improved degradation efficiency.	Polyethylene terephthalate (PET)	Recycled	Jiangnan University, China	Engineered microorganism-based enzymatic biodegradation using biofilm-assisted whole-cell catalysis	DuraPETase (engineered PETase variant), Est_B (novel endogenous BHETase from S. *pavanii*), Microorganism: Engineered *Stenotrophomonas pavanii* JWG-G1	38.04 μM total product release (TPA, MHET, BHET) from PET microplastics in 30 days. Est_B alone achieved 100% BHET degradation (0.23 mg/mL in 4 hours at 30 °C).	30 days 4 hours for BHET degradation by Est_B	1. Study emphasizes ambient-temperature degradation and bioreactor applicability, which reduces energy input. 2. PET degradation occurred in varied water environments (tap, lake, seawater), showing practical field potential.	1. Complete biodegradation of all by-products (e.g., EG) was not confirmed. 2. Study was conducted under controlled lab conditions; natural environmental complexity might reduce effectiveness.
39190796 (141)	1. The core is to Quantify and compare abiotic photoaging and biotic biodegradation processes under sediment–water interface conditions.	Polyethylene terephthalate (PET) (alongside PP, PLA)	Virgin	Zhejiang University, China	Both enzymatic/microbial (biotic) and photoaging (abiotic)	*Rhodococcus*, Cupriavidus, Sphingomonas, *Bacillus*, Arthrobacter	Carbonyl Index (CI) of PET increased to 5.35 under light–sterile conditions.	90 days	Reveals PET undergoes surface oxidation and generates potentially toxic degradation products (e.g., terephthalic acid, low-MW aromatics).	1. No kinetic degradation data (% conversion or hydrolysis). 2. Not full degradation — only early-stage aging and oxidation quantified chemically.

39904161 (133)	1. *Clostridium thermocellum* was genetically engineered to secrete Leaf-Branch Compost Cutinase (LCC) for PET degradation. 2. A whole-cell PET depolymerization process was optimized in a 1-L bioreactor.	Polyethylene terephthalate (PET)	Post-consumer PET (recycled PET bottles were pretreated before degradation)	Qingdao Institute of Bioenergy and Bioprocess Technology & Nanjing Tech University, China Greifswald University, Germany	Biological degradation using an engineered microorganism	Microbe: Clostridium *thermocellum* DSM1313 (engineered thermophilic whole-cell biocatalyst) Enzyme: LCC F243I (engineered variant of leaf-branch compost cutinase)	96.70%	10 days	The approach avoids harsh chemical conditions.	1. pH decrease during the degradation process affected enzyme activity and required external pH control to maintain stability. 2. PET crystallinity impacted the degradation rate—lower crystallinity samples degraded more efficiently

Table 18. Detailed information on articles related to combined approaches (chemical + biological).

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
37061790 (150)	1. Ammonium hydroxide treatment effectively depolymerizes PET into soluble aromatic monomers.	Polyethylene terephthalate (PET)	Recycled PET pellets	Michigan Technological University, USA	A coupled process- Chemical deconstruction, Biological degradation	Chemical pretreatment: Ammonium hydroxide (NH_4OH) aminolysis/hydrolysis of PET at 240 °C for 1 h Microbial consortia: LS1_Calumet, LS2_Calumet (compost-derived) and EB2_Mackinac, EB9_Mackinac (sediment-derived)	approximately 99.8% monomers were degraded.	7 days	1. By using a chemical pretreatment that also supplies nutrients (nitrogen and phosphorus), the need for expensive, defined media is eliminated.	1. The chemical deconstruction produces a mixture of compounds, and one of these (terephthalamide) may inhibit microbial activity. 2. Variability in degradation efficiency was observed among different microbial consortia.
37248049 (153)	1. The review highlights hybrid approaches that combine chemical depolymerization (or enzymatic hydrolysis) with biological transformation, offering an alternative to cost-prohibitive mechanical recycling.	Polyethylene terephthalate (PET)	Recycled/Waste	USA	Microbial biotransformation with chemical	Chemical pretreatment + PETase + MHETase (dual enzyme system) expressed in *Pseudomonas putida* KT2440	nearly 100% PET-derived BHET conversion to TPA + EG	NA	The microbial biotransformation process is a sustainable alternative to conventional recycling-it works under mild, ambient conditions, reducing energy use and greenhouse gas emissions.	1. The current performance of PET-hydrolyzing enzymes may not yet be sufficient for widespread industrial use. 2. Successfully integrating chemical recycling (or enzymatic hydrolysis) with biological upcycling requires the careful coordination of multi-step processes, from depolymerization to metabolic conversion.

39104625 (151)	1. The paper investigates microbial utilization of chemically deconstructed PET (CDPET) as a carbon source and explores strategies for controlling biofilm formation during PET monomer upcycling to valuable products like single-cell protein.	Polyethylene terephthalate (PET)	Recycled	Michigan Technological University and Idaho National Laboratory, USA	Chemical + Microbial biodegradation	Chemical deconstruction: Ammonium hydroxide-based hydrolysis at 260°C for 10 min Microbial Consortium-LS1_Calumet, *Rhodococcus sp.*, Paracoccus *sp.*	NH_4OH-deconstructed PET + LS1_Calumet consortium → ~1–2 g/L TPA	48 hours	Supports upcycling of PET waste into valuable products (e.g., single-cell protein).	1. Incomplete monomer degradation= TPA monoamide and terephthalamide not metabolized. 2. Low solubility of some products (Especially under certain pH).
37699265 (152)	1. PET samples pretreated with choline chloride: glycerol DES showed increased hydrophilicity and biofilm formation, facilitating microbial colonization. 2. DES coating enhanced PET degradation, resulting in significant weight loss compared to untreated PET.	Polyethylene terephthalate (PET)	Virgin	Thailand	Combined: Chemical pretreatment using DES (Choline chloride: glycerol 1:2) & Microbial degradation using a plastic-degrading bacterial consortium	Chemical pretreatment: Deep eutectic solvent (DES) coating (Choline chloride: glycerol, 1:2 molar ratio) to increase PET surface hydrophilicity and improve bacterial adhesion. *Achromobacter xylosoxidans* A9, Chitinophaga jiangningensis EA02, *Nocardioides zeae* EA12	PET weight loss in lab composting: With DES + Bioaugmentation= 22–23% Without DES but With Bioaugmentation= 23% Natural Attenuation (No Bioaugmentation) = 10% In pilot-scale composting: DES-coated PET with bioaugmentation= 25% Without bioaugmentation= ~21% Without DES= 5–6%	Lab-scale composting: 42 days Pilot-scale composting: 65 days	1. It demonstrates a practical, eco-friendly strategy to improve PET degradation using a combination of green chemical pretreatment (DES) and microbial bioaugmentation. 2. Low energy requirement for DES coating (ambient temperature); hence, scalable and suitable for municipal composting facilities.	1. PET degradation, although improved, remains moderate (max 25%) even with combined DES and bioaugmentation. 2. Molecular weight change of PET was not measured, suggesting limited depolymerization.

Table 19. Detailed information on articles related to other combined approaches.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
37788986 (154)	1. The paper centers on enhancing the activity of PET-degrading enzymes through two main strategies: (1) Identification of Novel Enzymes: It reports the discovery and initial characterization of two novel enzymes (Dh3 and Dh5) capable of degrading PET. (2) Biofilm Modulation: It innovates by demonstrating that increasing bacterial biofilm formation can potentiate enzyme activity, thereby enhancing PET degradation.	Polyethylene terephthalate (PET)	NA	Brunel University, UK	Enzymatic degradation coupled with Biofilm Modulation and Assays	Dh3 (from Alkalilimnicola ehrlichii, ~68% identity to PET2), Dh5 (from Aquabacterium *sp.*, ~54% identity to PET12), Thc_Cut2	PET breakdown products (TPA and MHET) - at 230–260 nm	5 days of incubation at 37 °C	By improving the efficiency of PET-degrading enzymes through biofilm modulation, this work offers a promising route toward developing scalable bioreactor systems for plastic degradation.	1. Low Native Efficiency. 2. The study relies on spectrophotometric and weight loss assays, which, while indicative, do not provide full-scale degradation kinetics.
17142403 (155)	1. *Rhodococcus sp.* RHA1 degrades TPA using a bifurcated catabolic pathway. 2. This is the first report of a bifurcated pathway for terephthalate, indicating sophisticated metabolic capacity.	Terephthalate	Virgin	University of British Columbia, Canada	Enzymatic degradation via aerobic microbial metabolism using dioxygenases and β-ketoadipate pathway enzymes	Organism: *Rhodococcus sp.* strain RHA1	Terephthalate was completely utilized as a sole carbon source in aerobic culture (not quantified as %)	NA	The discovery of aerobic bifurcated terephthalate degradation helps understand how PET-derived monomers may be mineralized in the environment once released.	PET polymer degradation was not addressed and only focused exclusively on terephthalate degradation, a downstream PET hydrolysis product.

Table 20. Detailed information on articles related to unconventional methods (marine).

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
34154652 (156)	1. The work provides evidence that PET biodegradation occurs in the marine plastisphere and that different PET forms (crystallinity, weathering) influence the degradation dynamics. 2. The study used a multi-OMIC approach (proteogenomic, metabolomics, and predicted metagenomics) to examine the biodegradation of PET by marine microbial communities.	Polyethylene terephthalate (PET) as, PET powder (crystallinity >40%), Amorphous PET films, Weathered PET powder	Virgin PET	NA	Biodegradation mediated by marine microorganisms via enzyme-driven hydrolysis	Thioclava *sp.* BHET1 *Bacillus sp.* BHET2	Thioclava *sp.* BHET1: 22-fold increase in BHET accumulation relative to controls *Bacillus sp.* BHET2: 2.6-fold increase in BHET accumulation relative to controls	6 weeks	1. The study suggests that even though PET is highly recalcitrant—especially in its crystalline form—marine microbial communities possess the enzymatic toolkit to initiate PET biodegradation.	1. The enzymatic pathway in *Bacillus sp.* BHET2 remains unclear, suggesting the need for further biochemical characterization. 2. The efficiency of degradation is affected by the physical state of PET (e.g., high crystallinity limits chain accessibility).
36494435 (157)	1. Engineered Vibrio natriegens Vmax to degrade PET under high-salinity marine conditions. 2. Identified salt-induced gene promoters (P1, P2-1, P2-2) to enhance enzyme production for PET degradation. 3. Whole-cell and crude enzyme systems showed significant PET degradation activity.	Polyethylene terephthalate (PET)	virgin	Shanghai Jiao Tong University & Chinese Academy of Sciences, China	Genetically engineered Vibrio natriegens expressing PET hydrolases	Microorganism: Vibrio natriegens Vmax (engineered marine bacterium) Enzymes: PETase, MHETase, LCC, TfCa (various PET hydrolases used in different constructs)	Crude enzyme system: MPP: BHET → 127 mg/L, MHET → 14 mg/L in 24–60 h Whole-cell system: Lower than crude enzymes; max BHET/MHET ~10–11 mg/L in 8 days	8 days for whole-cell degradation of PET 120 hours (~5 days) for crude enzyme degradation	1. Demonstrates the feasibility of PET biodegradation in marine environments. 2. Engineered strains can be immobilized using chitin-binding proteins, reducing biosecurity risks.	1.Degradation efficiency is still lower than optimal for large-scale applications. 2. Biosecurity concerns with the release of engineered bacteria in the environment.

34381429 (158)	1. The enzyme Mle046 is a marine MHETase-like enzyme that degrades MHET. 2. It exhibits broad activity over a range of temperatures (from ~5°C to 60°C) and pH values (optimal at pH 8–9).	PET and PBAT	NA	Germany	Enzyme-based degradation	Mle046 (a marine MHETase-like enzyme)	92% degradation of MHET (800 µM)	Approximately 21 minutes at 30°C	NA	NA
32836129 (159)	1. PET biofilms were successfully formed by marine bacteria enriched using hydrocarbons (tetradecane, diesel, and naphthalene/phenanthrene) and the most effective PET degradation was observed in microcosms with diesel- and tetradecane-acclimated consortia. 2. SEM showed significant morphological damage on PET.	Polyethylene terephthalate (PET)	Virgin PET	Italy	Microbial biodegradation using marine hydrocarbon-degrading bacterial consortia	Microorganisms: *Alcanivorax*, Cycloclasticus, *Thalassospira*	NA No direct % PET	45 days of incubation in marine microbial enrichment microcosms	Suggests that natural marine microbial consortia can initiate PET degradation	1. PET degradation was partial and surface-limited; no evidence of full mineralization. 2. Potential risk of increased microplastic generation from degradation products. 3. The study was conducted in microcosms; real-world marine conditions may vary.
32117139 (160)	1. The PE-H enzyme, from *Pseudomonas aestusnigri*, was identified as a type IIa PET hydrolase. 2. It degrades BHET and amorphous PET (PETa) to MHET, but not TA. 3. Wild-type PE-H was inactive against commercial crystalline PET (PETb). 4. Mutant Y250S showed improved degradation activity, including hydrolysis of PETb (albeit limited).	Polyethylene terephthalate (PET) Tested substrates: BHET, PETa (amorphous), PETb (crystalline commercial PET bottle)	Virgin	Germany	Enzymatic degradation	Enzyme: PE-H (wild-type and Y250S variant) Organism: *Pseudomonas aestusnigri* Recombinant expression in *E. coli* BL21(DE3)	PETa (amorphous): WT PE-H released ~4.2 mg/L MHET 48 h PETb (crystalline): Only Y250S variant showed detectable activity (5.4 mg/L MHET) Activity on PETb was ~45× lower than on PETa	48 hours at 30°C	1. Demonstrates potential for low-temperature PET biodegradation, beneficial in marine or mesophilic environments	1. Wild-type enzyme cannot degrade crystalline PET 2. Degradation yields (even for mutant) are low compared to PETase from *I. sakaiensis*. 3. No full PET mineralization; only partial hydrolysis to MHET. 4. No release of terephthalic acid (TA) observed.

Table 21. Detailed information on articles related to unconventional methods (larvae).

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
37276692 (161)	1. The study demonstrated that Tenebrio molitor (mealworm) larvae can rapidly biodegrade commercial PET resin powders. 2. Sequencing of 16S rRNA gene amplicons showed that the gut microbial community shifts upon PET ingestion. Dominant genera associated with PET degradation included *Spiroplasma sp.* (41.63%), *Dysgonomonas sp.* (2.4%), and Hafnia-*Obesumbacterium sp.* (6.5%).	Polyethylene terephthalate (PET)	virgin	China	microorganism-driven biodegradation (via the insect gut ecosystem).	*Spiroplasma sp.*, *Dysgonomonas sp.*, and Hafnia-*Obesumbacterium sp.*	Mass Reduction: The ingested PET experienced an average mass reduction of 71.03% (PET-1) and 73.28% (PET-2) after passing through the digestive tract over a 36-day feeding trial. PET Half-life: 3.36 hours for PET-1 and 4.32 hours for PET-2	approximately 12–15 hours	The high degradation efficiency and short PET half-life in the larval gut suggest that this biological approach may help prevent the formation of persistent micro- and nanoplastics and also alternative to conventional recycling methods.	1. Nutritional Deficiency: PET does not provide the essential nutrients required for optimal larval growth. 2. Complex Interactions: There is inherent complexity in deciphering the relative contributions of the host's digestive enzymes versus those produced by the gut microbiota.

38219578 (162)	1. Under antibiotic suppression of gut microbes, PET depolymerization continued at levels statistically similar to unsuppressed conditions. This indicates that the host's digestive system can depolymerize PET independently, with the gut microbiome playing a synergistic role by further degrading intermediates and providing additional nitrogen. 2. The combined metabolomic, transcriptomic, and microbiome analyses suggest that multiple functional metabolic pathways are activated, ensuring rapid PET biodegradation.	Polyethylene terephthalate (PET)	Virgin	China	Other (Insect-mediated biodegradation via T. molitor larvae) (Instead of using an isolated enzyme or microbial culture, the biodegradation is carried out by feeding PET to the larvae and analyzing the degradation products.)	Tenebrio molitor	In feeding experiments over 36 days- 73.3% PET1100 (low MW): >95.7% mass reduction PET27100 (medium MW): 86.0% mass reduction PET63500 (high MW): 74.0% mass reduction	13 days for PET1100 15 days for PET27100 16 days for PET63500	The rapid degradation of PET by T. molitor larvae suggests a promising, eco-friendly approach to mitigating PET pollution, with degradation efficiencies far surpassing those of many microbial systems.	1. Degradation efficiency declines with increasing molecular weight of PET. 2. T. molitor larvae fed exclusively on PET receive inadequate nutrients, which affects larval growth and survival. Supplementary nutrition (e.g., wheat bran) is often required. 3. Challenges related to maintaining large insect populations, processing waste insect biomas.

Table 22. Detailed information on articles related to other unconventional methods.

PMID	Key Findings	Type of Plastic Used	Source of Plastic	Location of study	Degradation Method	Used Enzyme/ Microorganism	Degradation Efficiency	Time Required For Degradation	Environmental Impact Assessment	Limitation/Challenges (If any)
34902688 (163)	1. In-situ bioremediation of mangrove soil contaminated with PET and PS microplastics was achieved using an indigenous microbial consortium.	Polyethylene terephthalate (PET) and polystyrene (PS)	Virgin	Sementa mangrove, Malaysia	Microbial biodegradation (via indigenous microbial consortium and bioaugmentation) under natural mangrove conditions	*Bacillus gottheilii*	For PET microplastics, the control soil- 16.4% weight loss the bioaugmented (treated) soil- 18% weight loss	90 days	Degradation of PET microplastics not only decreases the plastic weight but also alters the soil's elemental composition (increased Si, S, Fe; decreased C, O, Na, Mg, Al, Cl, and K), implying potential improvements in soil health and ecosystem function.	1. Environmental factors such as pH fluctuations, tidal variations, salinity, and nutrient availability influenced the degradation outcomes and microbial growth. 2. The overall degradation rates are low and more time consuming.
38101010 (164)	1. The study evaluated the chemical and physical modifications, as well as the microbial community (plastisphere), on different materials—including PET—after 43 days of immersion in Lake Maggiore.	Polyethylene terephthalate (PET)	Virgin	Lake Maggiore, Italy	Environmental degradation	PETase, MHETase genus *Pseudomonas*, *Bacillus*	thermal analyses (DSC and TGA) indicate that molecular-level changes occurred, suggesting an incipient stage of degradation.	43 days	The persistence of PET in freshwater systems, as evidenced by minimal macroscopic degradation despite measurable molecular alterations, underscores its long-term environmental durability.	1. PET exhibited high durability with no significant visible degradation in so long time (43 days).
33916472 (167)	This research presents an innovative method of transforming post-consumer PET bottle waste (NO PET biodegradation occurred) into	Polyethylene terephthalate (PET)	Recycled	Federal University of São Carlos (UFSCar), Brazil	NA	NA	NA (But 100% reuse of PET waste into high-performance filters)	NA	1. This study presents a sustainable alternative to PET waste disposal, turning recycled bottles into valuable air filtration materials. 2. Electrospinning process uses toxic	1. Does not address PET biodegradation or breakdown into monomers like TPA. 2. Performance depends on precise process control (fiber diameter, thickness, porosity).

	electrospun nanofiber membranes for high-efficiency air filtration applications.								solvents (TFA, DCM), raising environmental/health concerns.	
35017590 (166)	1. It provides the first in experimental evidence suggesting PET microplastics may undergo structural changes and partial degradation in the human gastrointestinal tract, particularly due to interactions with colonic microbiota.	Polyethylene terephthalate (PET)	Virgin PET	Institute of Food Science Research (CIAL), CSIC-UAM, Madrid, Spain	Simulated gastrointestinal digestion (INFOGEST model) Followed by dynamic colonic fermentation (simgi® simulator)	NA	structural changes were documented: Amorphization of PET MPs (via Raman shifts), Surface biofilms formed after colonic fermentation, Loss of crystalline structure and roughened surface	72 hours	PET may undergo partial biodegradation under gut-like conditions, implying risks for both gut function and MP bioavailability	1. PET degradation is partial and not mineralization. 2. No enzymatic pathways identified; degradation inferred from morphology and Raman shifts. 3. Used pure PET without processing additives — real-world MPs are more complex.
33444995 (165)	1. Developed Lensless Digital Holographic Microscopy (LDHM- new imaging method) as a real-time monitoring tool for PET degradation. 3. Used PETase enzyme produced by a marine diatom (Phaeodactylum tricornutum) to degrade PETG film.	Polyethylene Terephthalate Glycol (PETG) (a PET copolymer, more amorphous than PET)	Commercial PETG film (industrial sample)	University of Marburg, Germany	Enzyme-based degradation	NA	43 days of observation (mostly in first 11 days) in a saltwater-based system	1. Supports the potential for marine bioremediation of PET pollution using engineered microalgae. 2. PET degradation occurred under moderate conditions (30–37°C, pH 8.0, saltwater environment).	1. PETG is more amorphous than regular PET, making it easier to degrade. Results may not fully translate to highly crystalline PET. 2. LDHM is effective for surface changes but does not quantify complete PET degradation.	NA