

Biodegradation of polymers

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Exhaustive studies on the degradation of plastics have been carried out in order to overcome the environmental problems associated with synthetic plastic waste. Recent work has included studies of the distribution of synthetic polymer-degrading microorganisms in the environment, the isolation of new microorganisms for biodegradation, the discovery of new degradation enzymes, and the cloning of genes for synthetic polymer-degrading enzymes. Under ambient conditions, polymers are known to undergo degradation, which results in the deterioration of polymer properties, characterized by change in its molecular weight and other physical properties. In this paper mainly the biodegradation of synthetic polymers such as polyethers, polyesters, polycaprolactones, polylactides, polylactic acid, polyurethane, PVA, nylon, polycarbonate, polyimide, polyacrylamide, polyamide, PTFE and ABS have been reviewed. *Pseudomonas* species degrade polyethers, polyesters, PVA, polyimides and PUR effectively. No microorganism has been found to degrade polyethylene without additives such as starch. None of the biodegradable techniques has become mature enough to become a technology yet.

Keywords: biodegradation, polymer, biodegradable polymers

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Introduction

Approximately 140 million tonnes of synthetic polymers are produced worldwide every year. Since polymers are extremely stable, their degradation cycles in the biosphere are limited. In Western Europe alone it is estimated that 7.4% of municipal solid wastes are plastic, which are classified as 65% polyethylene/polypropylene, 15% polystyrene, 10% PVC, 5% polyethylene terephthalate and remaining others. Environmental pollution by synthetic polymers, such as waste plastics and water-soluble synthetic polymers in wastewater has been recognized as a major problem. In view of this, energetic, chemical and biological polymer-degrading techniques have been studied extensively during the last three decades. The energetic agencies can be either thermal or radiant. The radiant energy may be high-energy radiation like gamma rays, ion beams, and electrons or even low energy radiation like ultraviolet (UV) rays. Chemical degradation is caused using certain chemicals like acids and alkalis, etc. Usage of certain microorganisms and enzymes to degrade polymers are classified as the biodegradation method of polymers.

Some types of plastics have been shown to be biodegradable, and their degradation mechanisms have progressively become clearer. Very small variations in the chemical structures of polymer could lead to large changes in their biodegradability. The biodegradability depends on the molecular weight, molecular form and crystallinity. It decreases with increase in molecular weight, while monomers, dimers and repeating units degrade easily. Two categories of enzymes are involved in the process, namely extracellular and intracellular depolymerases. Exoenzymes from the microorganisms first breakdown the complex polymers giving short chains that are small enough to permeate through the cell walls to be utilized as carbon and energy sources. The process is called depolymerization. When the end products are carbon dioxide, water or methane, the process is called mineralization. Different end products are formed depending upon the degradation pathway as shown in Fig. 1.

Polyethers

Polyethylene glycols (PEGs), polypropylene glycols (PPGs) and polytetramethylene glycols come under the class of polyethers and are used in pharmaceuticals, cosmetics, lubricants, inks, and surfactants. *Flavobacterium* sp. and *Pseudomonas* sp. together associate and mineralize PEGs completely

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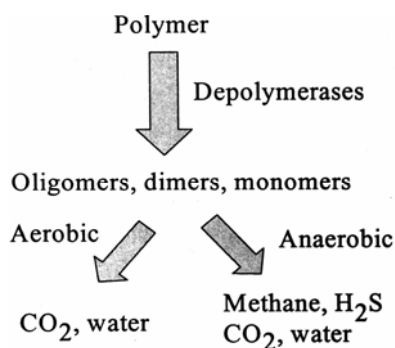


Fig. 1—Reaction pathways during biodegradation of polymers

under aerobic conditions¹. During degradation, the PEG molecules are reduced by one glycol unit at a time after each oxidation cycle. *Pelobacter venetianus* was found to degrade PEG and ethylene glycol under anaerobic conditions².

Polyesters

Polyesters are polymers in which component monomers are bonded via ester linkages. Many kinds of esters occur in nature and the esterase enzymes that degrade them are ubiquitous in living organisms. Ester linkages are generally easy to hydrolyze and hence a number of synthetic polyesters are biodegradable. Therefore, bacterial polyesters (polyhydroxyalkanoates) have been used to make biodegradable plastics. Several polyesters are produced on a semi-commercial scale by a number of companies that make biodegradable plastics. Hydrolytic cleavage of the ester bond in the low molecular weight polyester by the lipase of *Pseudomonas* species has been reported.

Polyhydroxyalkanoates

Polyhydroxybutyrate (PHB) is a naturally occurring polyester that accumulates in bacterial cells as a carbon and energy storage compound. PHB and copolymers containing polyhydroxyalkanoate, PHA (for example 3-hydroxyvalerate) are being used for the manufacture of biodegradable plastics. Several PHA and PHB bacterial *depolymerases* are found to be capable of metabolizing PHB and other PHA polymers. The PHA *depolymerases* are serine hydrolases usually having single substrate-binding domain. Recently, a PHB *depolymerase* with two substrate-binding domain has been reported. The presence of two substrate-binding domain enlarges substrate specificity or enhances adsorption of the enzyme. PHB *depolymerases* are able to degrade all-(*R*) chains, cyclic-(*R*) oligomers, oligolides, and

polymers composed of racemic hydroxybutanoate. The enzymes are generally obtained from the microorganisms like *Alcaligenes faecalis* and *Pseudomonas stutzeri*³.

In order to modify physical properties and retard enzymatic degradation of commercial microbial polyesters like PHA, they are blended with other degradable or non-degradable polymers such as PVA, PMMA, poly (ethylene oxide), PLA, cellulose, PCL, and polystyrene (PS).

Atactic P (R,S-3HB) [atactic poly (R,S-3-hydroxybutyrate)], which does not biodegrade in pure form can undergo enzymatic hydrolysis in a P(R,S-3HB)/PMMA (poly methacrylate) blend, indicating that the enzymatic degradation can be induced by blending with an amorphous non-biodegradable polymer. This is possible because the blend gives P (R-3HB) *depolymerase* more stable binding surface than that provided by the rubbery a-P (R,S-3HB). The *depolymerase* was purified from *A. faecalis*⁴.

Polylcaprolactone

Polylcaprolactone (PCL) is a synthetic polyester that can be degraded by microorganisms and enzymes like lipases and esterases. *Cutinases*, which are obtained from fungal phytopathogens, degrade cutin (the structural polymer of the plant cuticle) and act as PCL depolymerases. The chemical structure of a PCL trimer is similar to that of two cutin monomers, which are inducers of cutinase activity. This knowledge that cutinase can act as a PCL depolymerase has helped in identifying a new system to degrade PCL.

The biodegradability of PCL in the form of blend sheets (for example in polycarbonate-polycaprolactone blend sheets) is much reduced because the packed form of the PCL in the blend sheets protects it from enzymatic digestion⁵. However, the enzymatic degradation can be promoted by means of surface etching by oxygen plasma treatments⁶.

Poly-L-lactide

Poly-L-lactide (PLLA) is a lactic acid-based aliphatic polyester, which is used in medical and packaging applications. It can be degraded both in aerobic and anaerobic conditions. Under thermophilic conditions the latter is faster than the former, probably because lactic acid is more favourable for the anaerobic microorganisms than for aerobic organisms⁷. PLLA is also found to degrade

completely in two weeks in windrow composting.

Polylactic Acid

Polylactic acid (PLA) is found absorbed in animals and humans and hence has been extensively used in medicines. The degradation of the polymer in animals and humans is thought to proceed via non-enzymatic hydrolysis. Several enzymes can degrade the polymer which include *proteinase K*, *pronase* and bromelain. However, few have been characterized with regards to microbial degradation of the polymer³. PLA is also readily degraded in compost. ¹⁴C-labelled PLA was mineralized in compost to CO₂ (about 90% degradation was achieved in 90 days). The contribution of microorganisms to this degradation is still not found. *Actinomycete*, a PLA degrading strain, reduced 100 mg of PLA film by 60% in the first 14 days in liquid culture at 303 K. *Bacillus brevis* is also found to degrade 50 mg of PCL by around 20% in 20 days in liquid culture at 333 K.

Poly (p-dioxanone)

Poly (p-dioxanone) (PPDO) is known as a poly ether-ester and has good tensile strength and flexibility. It is used for bioabsorbable sutures in clinical applications. PPDO is found to be degraded by strains that belong to the α and β subdivision of the class *Proteobacteria* and the class *Actinobacteria*. Degradation leads to the formation of monomeric acids⁸.

Polyurethane

Polyurethane (PUR) produced by the diisocyanate poly addition process is the characteristic chain link of urethane bond (Fig. 2). Growth of microorganisms could not be supported by PUR and so the biodegradation was also found incomplete. PUR degradation proceeded in a selective manner, with the amorphous regions being degraded prior to the crystalline regions. Also, PUR's with long repeating units and hydrolytic groups would be less likely to pack into high crystalline regions as normal polyurethane, and these polymers were more accessible to biodegradation.

Biodegradation of PUR

The PUR *depolymerases* of the microorganisms have not been examined in detail, although due to the presence of ester linkage most degradation is carried out by *esterases*. *Comamonas acidovorans* TB-35 utilizes a polyester PUR containing polydiethylene glycol adipate as the sole source of carbon but not the polyether PUR. *C. acidovorans* can completely

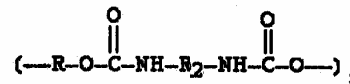


Fig. 2—Structure of polyurethane

degrade 50 mg of PUR dissolved in mineral salt medium at 303 K. The products of degradation were adipic acid and diethylene glycol. Absence of any metabolites confirms that urethane linkage is not cleaved.

The degradation of PUR is inhibited by the presence of a detergent that does not inhibit the hydrolysis of a water-soluble ester compound, suggesting that degradation proceeds via a two-step mechanism namely,

1. Hydrophobic adsorption to the polymer surface, followed by
2. Hydrolysis of the ester bond of PUR.

The gene cloned for the PUR degrading enzyme encodes a protein of 548 amino acid residues. The deduced protein sequence contains a signal sequence, the lipase box and catalytic triad, and three hydrophobic domains, which play role in the hydrophobic adsorption of the enzyme to the polymer surface. It was found that two proteolytic enzymes, *papain* and *urease* degraded medical polyester PUR⁹. Bacteria like *Corynebacterium* sp. and *Pseudomonas aeruginosa* could degrade PUR in the presence of basal media¹⁰. However, none of the isolates grew on PUR alone. Physical tests of the degraded polyester PUR revealed marked decrease in tensile strength and elongation for each isolate. Several fungi are observed to grow on PUR surfaces and especially *Curvularia senegalensis* was observed to have a higher PUR-degrading activity.

Although cross-linking was considered to inhibit degradation, *papain* was found to diffuse through the film and break the structural integrity by hydrolyzing the urethane and urea linkage producing free amine and hydroxyl group. Polyester PUR degradation by *porcine pancreatic elastase* was ten times faster than its activity against polyether PUR.

Polyvinyl Alcohol

Polyvinyl alcohol (PVA) is a vinyl polymer joined by only carbon-carbon linkages. The linkage is the same as those of typical plastics, such as polyethylene, polypropylene and polystyrene, and

water-soluble polymers, such as polyacrylamide and polyacrylic acid. Among the vinyl polymers produced industrially, PVA is the only one known to be mineralized by microorganisms. PVA is water-soluble and biodegradable and hence, used to make water-soluble and biodegradable carriers, which may be useful in the manufacture of delivery systems for chemicals such as fertilizers, pesticides and herbicides.

PVA is completely degraded and utilized by a bacterial strain, *Pseudomonas* O-3, as a sole source of carbon and energy. However, PVA-degrading microorganisms are not ubiquitous within the environment. Almost all of the degrading strains belong to the genus *Pseudomonas* although some do belong to other genera¹¹. Generally, the carbon-carbon linkage of PVA is degraded either by the enzymes dehydrogenase or oxidase, which is further degraded by the action of hydrolase or aldolase to form simple compounds.

Amongst the PVA-degrading bacteria reported so far, a few strains showed no requirement for pyroloquinoline quinone (PQQ). From a PVA-utilizing mixed culture, *Pseudomonas* sp. VM15C and *P. putida* VM15A were isolated. Their symbiosis is based on a syntrophic interaction. VM15C is a PVA-degrading strain that degrades and metabolizes PVA, while VM15A excretes a growth factor that VM15C requires for PVA utilization (Fig. 3).

Although the PQQ supplier, VM15A does not degrade or utilize PVA directly in the symbiotic mixed culture. It is thought to utilize some unidentified metabolites excreted from the VM15C cells growing on PVA. Thus, if PQQ growth factor is added to the medium, the PVA-degrading strain can

grow on PVA in a pure culture. In the natural environment, however, in which PVA-degrading bacteria depend on other strains for PQQ, the amount excreted by other bacteria is a critical factor for PVA degradation. It has also been observed that a concentration of 10g/l of PQQ is sufficient for the strain to grow at maximum rate.

Nylon

High molecular weight nylon-66 membrane was degraded significantly by lignin degrading white rot fungi grown under ligninolytic conditions with limited glucose or ammonium tartrate¹². The characteristics of a nylon-degrading enzyme purified from a culture supernatant of white rot fungal strain IZU-154 were identical to those of *manganese peroxidase*, but the reaction mechanism for nylon degradation differed significantly from *manganese peroxidase*. The enzyme could also degrade nylon-6 fibres. The nylon was degraded to soluble oligomers by drastic and regular erosion.

A thermophilic strain capable of degrading nylon 12 was isolated from 100 soil samples by enrichment culture technique at 60°C. At this temperature, the strain not only grew on nylon 12, but also reduced the molecular weight of the polymer. The strain was identified as a neighbouring species to *Bacillus pallidus*. It has an optimum growth temperature of around 60°C and was also found to degrade nylon 6 as well as nylon 12, but not nylon-66¹³.

Polyethylene

Polyethylenes of low density are used widely as films in packaging industry. They pose serious problem with respect to biodegradability because of their slow rate of degradation under natural

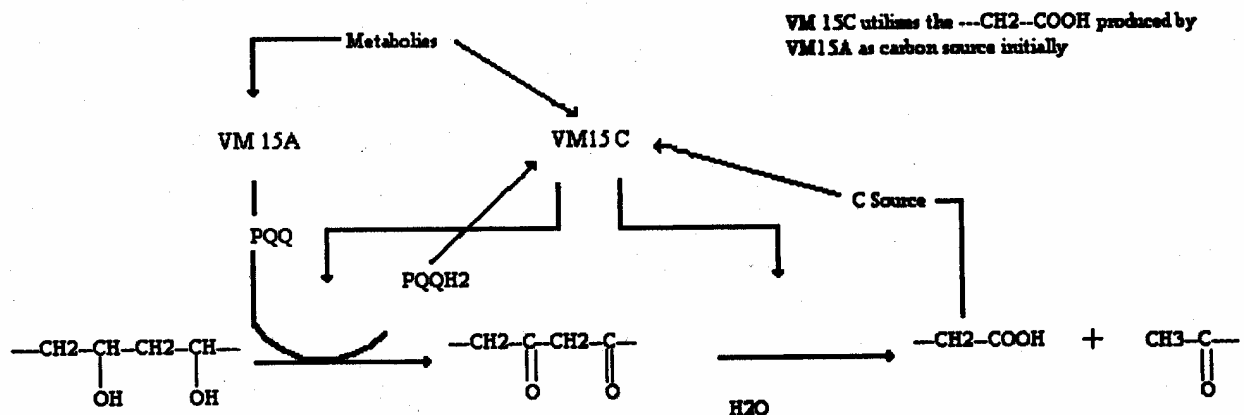


Fig. 3—PVA biodegradation pathway

conditions. They also pose problems to the environment, fresh water and animals. Extracellular *Streptomyces* sp. cultures were found to degrade starch blended PE.

Phanerochaete chrysosporium has also been found to degrade starch blended LDPE in soil. A decrease of more than 50% reduction in elongation was observed when the soil is inoculated with the organism as against a decrease of only 12% with uninoculated soil system¹⁴.

High molecular weight polyethylene is also degraded by lignin-degrading fungi under nitrogen-limited or carbon-limited conditions, and by manganese peroxidase. Fungi like *Mucor rouxii* NRRL 1835 and *Aspergillus flavus* and several strains of *Streptomyces* are capable of degrading polyethylene containing 6% starch. Degradation was monitored from the changes in the mechanical properties like tensile strength and elongation¹⁵. Biodegradability rate of blends of LDPE and rice or potato starch was enhanced when the starch content exceeded 10% (w/w). No microorganism or bacterium has been found so far that could degrade PE without additives¹⁶.

Polycarbonate

Bisphenol-A polycarbonate is a widely used polymer because of its excellent physical properties such as transparency, high tensile strength, impact resistance, rigidity and water resistance. Polycarbonate (PC) gets its name from the carbonate groups in its backbone chain. The transparency of PC (up to 88% transmittance of visible light) is exploited in such applications as aircraft light housings and dials, windscreens for cars, safety helmets, contact lenses, etc. Panels of PC are used as enclosures for growing plants since it allows diffusion of light and reduces plant sunburn resulting in more production.

Biodegradation of Polycarbonate

PC is stable to bioorganism attack. PCL sheets are known to degrade in lipase AK, but when they are blended with PC they become less biodegradable⁵. PC being hydrophobic probably suppresses the biodegradation, but that alone does not fully explain the observed behaviour.

Several authors have described enzymatic degradation of aliphatic polycarbonate (polyethylene carbonate, PEC). No degradation of PEC with molecular weight of 300-450 kDa in hydrolytic enzymes including lipase, esterase, lysozyme,

chymotrypsin, trypsin, papain, pepsin, collagenase, pronase and pronase E were observed indicating that hydrolytic mechanisms based on hydrolases or aqueous conditions were excluded for biodegradation of PEC¹⁷.

Polyimide

Polyimides find application in electronic and packaging industries. These polymers possess high strength and resistance to degradation. Fungi like *Aspergillus versicolor*, *Cladosporium cladosporioides* and *Chaetomium* sp. were found to degrade this polymer. Whereas bacteria like *Acinetobacter johnsonii*, *Agrobacterium radiobacter*, *Alcaligenes denitricans*, *C. acidovorans*, *Pseudomonas* sp. and *Vibrio anguillarum* were not effective in biodegrading this polymer.

Fibre-reinforced Polymeric Composite

Fibre-reinforced polymeric composite materials (FRPCMs) are materials important in aerospace and aviation industries. A fungal mixture consisting of *A. versicolor*, *C. cladosporioides*, and *Chaetomium* sp. and a mixed culture of bacteria including a sulphate-reducing bacterium were found to grow on this composite material. Only the fungi mixture could cause deterioration detectable over more than 350 days¹⁸.

Polyacrylamide

Polyacrylamides are water-soluble synthetic linear polymers made of acrylamide or the combination of acrylamide and acrylic acid. Polyacrylamide finds applications in pulp and paper production, agriculture, food processing, and mining, while its major use is as a flocculant in industrial water as well as municipal water and wastewater treatment.

Biodegradation of Acrylamide

Acrylamide is readily biodegraded by microorganisms in soil and water systems under aerobic conditions by deamination to acrylic acid and ammonia that are utilized as carbon and nitrogen sources. *Pseudomonas stutzeri*, *Rhodococcus* spp., *Xanthomonas* spp. and mixed cultures have demonstrated degrading abilities under aerobic conditions in numerous studies¹⁹. However, few studies have addressed the possibility of anaerobic acrylamide biodegradation²⁰.

Polyamide

Polyamide-6 (PA-6) is a widely used engineering

material. Oxidative degradation of PA-6 membranes was found by lignolytic white rot fungus IZU-154. *A. niger* mediated degradation of polyamides based on tartaric acid and hexamethylenediamine and *Corynebacterium aurantiacum* mediated degradation of ϵ -caprolactam, as well as its oligomers has been reported^{12,21}. Lignolytic fungus, *Phanerochaete chrysosporium* is also found to degrade PA-6²². Degradation of the polymer was observed through the decrease in the average molecular mass (50% after 3 months), as well as in the physical damage to the fibres visible under a scanning electron microscope.

Polytetrafluoroethylene

Polytetrafluoroethylene (PTFE) in the form of micro powders are used as additives in inks, coatings and lubricants. PTFE is classified as a polymer that undergoes main chain scission by irradiation. Ionizing radiation was capable of decreasing the molecular weight of PTFE. Low-pressure plasmas were also found useful for altering surface properties of polymer materials like PTFE to satisfy the demand of certain applications without altering the bulk properties.

Acrylonitrile Butadiene Styrene

Wood boring pholads, *Martesia striata*, damage ABS pipes by drilling holes through the pipes, which were used for 5 years for transportation of hypochlorite. They were found to have hole with sizes ranging from 3 to 19 mm². Unlike other boring animals (e.g. *Lithophaga*), *M. striata*, penetration is not assisted by chemical dissolution of the substrates and, therefore, attack is not limited to calcium carbonate substrates²³.

Rubber

Two major problems concerning rubber are the wastage of valuable rubber and the disposal of waste tyres leading to environmental pollution. Two major approaches to solve this problem are the recycle and reuse of used and waste rubber, and the reclaim of rubber raw materials. The reuse of used rubber includes using scrap rubber as fuel source. This is achieved by selective oxidation process. Physical reclaiming of rubber involves breaking down the used/scrap material through mechanical, thermo-mechanical, microwave or ultra sound and then mixing it with the virgin material during compounding²⁵.

Biodegradation process for reclaiming of rubber

Biological attack of natural rubber latex is quite

facile. But addition of sulphur and numerous other ingredients to rubber reduces biological attack. A recent approach was to utilize a chemolithotrope bacterium in aqueous suspension for attacking powder elastomers on the surface only, so that after mixing with virgin rubber diffusion of soluble polymer chains is facilitated and bonding during vulcanization becomes again possible.

Devulcanized scrap rubber can be obtained by holding the comminuted scrap rubber in a bacterial suspension of chemolithotropic microorganisms with a supply of air until elemental sulphur or sulphuric acid is separated²⁵. This process can reclaim rubber and sulphur in a simplified manner.

The biodegradation of the *cis*-1, 4-polyisoprene chain was achieved by a bacterium belonging to the genus *Nocardia* led to considerable weight loss of different soft type NR-vulcanizates. The microbial desulphurization or devulcanization of particle surfaces was investigated in order to increase the possibility of producing high quality rubber products containing a larger percentage of recycled rubber. Old tyres with 1.6% sulphur, were treated with different species of *Thiobacillus ferrooxidans*, *T. thiooxidans*, *T. thioparus* in shake flasks and in a laboratory reactor. The sulphur oxidation depends to a large extent on the particle size. The best results were obtained with *T. thioparus* with a particle size of 100-200 μ m. 4.7% of the total sulphur of the rubber powder was oxidized to sulphate within 40 days³.

The reclaimed rubbers can be used in the preparation of thermoplastic elastomers. Ground rubber tyres can be used properly with thermoplastics and thereby minimizing their disposal problem to a considerable extent.

Biodegradable Polymers

Since most of the polymers are resistant to degradation, research over the past couple of decades has focused on developing biodegradable polymers, which are degraded and catabolized ultimately to carbon dioxide and water by bacteria, fungi under natural environment. During the degradation process they should not generate any harmful substances. These polymers can be classified into three major categories (1) polyesters produced by microorganisms (2) natural polysaccharides and other biopolymers like starch and (3) synthetic polymers like aliphatic polymers (eg, poly ϵ -caprolactone, poly L-lactide and poly butylenesuccinate, etc. are

commercially produced).

Another approach towards achieving biodegradability has been through the addition of biodegradable groups into the main chain during the preparation of industrial polymers prepared by free radical copolymerization. Use of ethylene bis(mercaptoacetate) as a chain transfer agent during the copolymerization of styrene and MMA, preparation of copolymers of vinylic monomers with cyclic comonomers containing the biodegradable functions like ketene acetal, cyclic disulphides are two such approaches²⁶.

Conclusion

Polymers are generally resistant towards degradation and most of the research has been towards developing technologies for recovery and reuse of the waste. Physico-chemical methods of degradation have been more successful than biological means. Nevertheless, the studies on the biodegradation of plastics have increased our knowledge of degrading microbes, enzymes and their genes, and polymer structures for biodegradation. This knowledge has also been useful for the design of biodegradable polymers and for the molecular evolution and breeding of degradation enzymes and microbes. Biodegradation of polymers such as polyethers, polyesters, polycaprolactones, polylactides, polylactic acid, polyurethane, PVA, nylon, polycarbonate, polyimide, polyacrylamide, polyamide, PTFE and ABS have been reviewed. *Pseudomonas* species degrade polyethers, polyesters, PVA, polyimides and PUR effectively. Degradation of several of these polymers proceed through adsorption of the organism to the polymer surface followed by hydrolytic cleavage, are also discussed in this review. Enzymatic degradation of nylon proceeds through drastic erosion. No microorganism has been found to degrade polyethylene without additives such as starch. The future lies in the polymers that will degrade in the environment, but to degrade the tough plastics and polymers that have already been manufactured and are being manufactured, new enzymes and organisms need to be designed.

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