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Microbial Bioremediation of Plastic and Its Future Aspects

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ABSTRACT

Plastics are polymers commonly used and have a very slow process of degradation. There has been a huge rise in plastic waste accumulation around the globe, which is a major environmental problem. Bioremediation of plastics by microbes has given us the ability in recent years to develop treatment technologies for plastic degradation.The ability of microbes to degrade plastic has been addressed here, and how they can be genetically modified to make the degradation process more efficient. Due to their ability to reduce plastics into simpler compounds, metabolically diverse microorganisms are often used.Various forms of synthetic plastics, such as Polyethylene (PE), Polypropylene (PP), Polyvinyl Chloride (PVC), Polystyrene (PS), Polyurethane (PUR), have been reported for degradation in these organisms. Several forms of microbes involved in degradation, metabolic pathways followed by them, and how these pathways can be manipulated using genetic engineering and gene editing techniques to perform degradation have been discussed here in this study.

KEYWORDS; Bioremediation, Microbial consortium, Degradation, Gene editing tools, Genetic engineering, Metabolic Pathway.

INTRODUCTION

Over the last few decades, pollution due to plastic waste is one of the major concerns across the world. Plastic is made up of high molecular weight synthetic polymers derived from various hydrocarbons and petroleum derivatives **(Ahmed et al., 2018).** Most of the plastic and plastic-based products that we are using are non-biodegradable for example polyvinyl chloride, polyethylene, polystyrene, polypropylene, etc. The fast pace of urbanization has increased the production and usage of plastic in our day to day life due to their use in various industries. Due to improper waste management and littering, it is accumulated in our environment and is threatening the safety of organisms. Approximately 165 million tons of plastic were present in the ocean through Asian countries **(Andrady, 2011) (Mrowiec, 2017)**. The accumulation of plastic polymers in water bodies is one of the most difficult challenges we are currently facing as it is very hazardous for aquatic organisms as well as plants. Through the consumption of these aquatic organisms, this carcinogenic material enters our food chain and affects our health causing cancer, disabilities, or various immunological disorders **(Horton et al., 2017).** Annually lakhs of marine animals like seals, turtles, whales, and various fishes have been reported dead. Even the terrestrial regions and the plants and animals living there are being affected **(Kedzierski et al., 2018)**. Chlorinated plastic is polluting the soil and its texture, it penetrates deep into the soil and contaminates the groundwater and other water resources **(Horton et al., 2017).**

To tackle this concerning situation, scientists and environmentalists are using modern biotechnological techniques to overcome this situation. They are also finding a way to overcome the use of conventional methods of burning and landfilling methods. Also, various other strategies are currently in use for plastic degradation such as chemical treatment, thermal degradation, photodegradation, and other biological procedures. Although there are different techniques and technology to control plastic pollution there's always a need to search for a cost-effective, novel agent to control and degrade plastic by an eco-friendly process.

In the last few decades, genetically modified microbes are advanced, used for tackling environmental issues. It is also accelerating the research interest with the aim for industrial purposes. With the help of genetic engineering tools, scientists are manipulating various bacteria into a tool by enhancing their capabilities or by adding different features to their genome so that they prove themselves useful in the degradation of plastic pollutants. Their biology and the natural consortium has been manipulated to obtain their full potential. For example, *Pseudomonas* species is ubiquitous and is found in both aquatic and terrestrial environments. They have been used for the bioremediation of crude oil, hydrocarbon, and other hydrophobic polymers. Through genetic manipulations, *Pseudomonas* was engineered to oxidize aromatic, aliphatic, polyaromatic compounds. For the complete elimination of plastic polymers, it requires the breakdown of polymers into oligomers and then monomers. Finally, the monomer passes through the cell membrane followed by intracellular metabolism and enzymatic activity.

From this review, we are discussing the concept of the involvement of microbes and their capabilities to degrade the plastic which can help develop new bioremediation strategies, and how all of this can be used in keeping our environment clean and green.

MICROBIAL DEGRADATION OF PLASTIC

Biodegradation of plastic by microbial enzymes involves the breakdown of plastic by biotic and abiotic factors which reduces their molecular weight. For the reduction of plastic waste from the environment, bacteria are suitable as they produce an enzyme that degrades plastic and utilizes it as a substrate. The exposure of the carbonyl group of plastic polymers to UV radiation makes it more vulnerable to microbes **(Leja and Lewandowicz, 2010)**. Many species of microbes have been identified and isolated from various sources such as dumping sites, sewage sludges, landfills, marine waste, which are capable of degrading Polyethylene (PE), Polystyrene (PS), Polyvinyl Chloride (PVC), Polyurethane (PUR), etc. it was found out that they use synthetic homopolymers or heteropolymers as their energy source. They carry out degradation by enzymatic activities and then by cleaving bonds. Mainly they secrete depolymerase- extracellular or intracellular for this process, which cleaves the polymers to generate monomers or short chains that are penetrable. Microbial enzymes such as thermostablelaccase have been reported to degrade PE (Polyethylene) after its two days incubation at 37□C. A hydrolytic enzyme urease can degrade polyurethane by hydrolyzing the linkages between urea and urethane. Depolymerases also carry out degradation **(Gu, 2003)**. Complex polymers undergo hydrolytic cleavage in the cell or the periplasmic space **(Koutny et al., 2006).** Now, these short oligomers are then transported through the cytoplasmic membrane and are used as carbon and energy source by intracellular enzymes **(Shah et al., 2008) (Koutny et al., 2006)**. Further, they enter beta-oxidation or before internalization and may be cleaved by abiotic processes **(Kawai et al., 2004).** In an alternative method, these monomers may undergo sequential degradation which results in the production of metabolites used in the TCA cycle and then enters into carbon metabolism. For example- during the biodegradation of styrene, Pyruvate and Acetaldehyde are obtained as byproducts that enter the TCA cycle or carbon metabolism after further metabolization into phenyl-acetyl CoA. The degradability is also dependent upon crystallinity and molecular weight. The crystalline region takes more time as compared to the amorphous region. However, some strains secrete enzymes for a particular type of polymer eg: *Ideonellasakaiensis* secretes PETase which hydrolyses PET to enter the beta-oxidation pathway **(Joo et al., 2018)**

This process of biological degradation depends upon various factors such as molecular weight, melting temperature, chemical structure, crystallinity, hydrophobicity, etc of plastic **(Okada, 2002).** Molecular weight plays important role in degradation. Higher the molecular weight lesser the degradation potential. Crystallinity is another crucial factor, polymers with amorphous domains are more vulnerable to microbial attack so, the lesser the crystallinity more are the chances of degradation **(Slor et al., 2018)**. Biodeterioration affects the surface of the plastic and brings out changes in its physical, chemical, and mechanical properties. Microbial biofilm formation contributes to the process of deterioration by causing physical and chemical degradation. Their accumulation of plastic depends upon the composition of plastic and environmental factors. The bacteria carry out the degradation process in two ways through a direct mechanism or indirect mechanism **(Shalini and shasikumar, 2015)**. In the direct mechanism, for its nutritional advantage, bacteria operate on the degradation of the plastic, while in the indirect mechanism they produce microbial products that degrade the polymer by biodeterioration, followed by fragmentation, assimilation and mineralization. In addition, all aerobes and anaerobes that release CO_2, H_2O and CO_2, H_2O and CH_4 as products, respectively, follow the indirect approach **(Singh and Sharma, 2008).**

Biodeterioration is followed by Bio-fragmentation which involves the catalytic action of the microbial enzyme on plastic. Bacteria use oxygenase enzymes to break the plastic polymer. Oxygenases add oxygen molecules into the carbon chain-forming alcohol or peroxy products which are less toxic for the environment **(Pathak, 2017; Dussud and Ghiglione, 2014).** Finally, mineralization and assimilation are carried out. The assimilation process includes the integration of plastic monomers into the microbial cell. Some monomers which are not able to enter the cell are not assimilated **(Kale et al., 2015)**. After assimilation catabolic pathways oxidize the monomers inside the cell and the energy produced by this utilized to form cell biomass **(Singh and Sharma, 2008)**. Secondary metabolites formed are transported outside the cell for further degradation.

PLASTIC DEGRADATION THROUGH SYNTHETIC APPROACH

MICROBIAL CONSORTIUM

The microbial consortium is defined as a species of bacteria that are surviving in a symbiotic relationship, they have shown potential in the bioremediation process. Their functionality has been reprogrammed and organized as per the need for experimentation **(Navarro-Díaz et al., 2016).** Consortium organizes themselves in a manner such that their functions are synchronized, they have proper cellular communication and signaling. They can perform more than one task over a single population **(Villaverde et al., 2017)**. For the total degradation of plastic, its complete mineralization must be executed by several enzymes using different metabolic pathways which can be provided by a consortium.

An example of such a mechanism was observed in a mixed culture of *Lysinibacillusxylanilyticus* and *Aspergillusniger*. These species were isolated from the landfill soils in Tehran and used to degrade Low-Density Polyethylene (LDPE). A comparative study was done between UV and non-UV irradiated LDPE film in the presence and absence of mixed cultures. To monitor the degradation process, cell number, pH, biomass production, carbon, mechanical properties, etc were compared. The results showed that the mineralization percentage of non-UV irradiated LDPE was 7.6% and for UV-radiated LDPE 8.6%. The biodegradation process of LDPE (both UV-radiated and non-UV radiated) was more in the presence of microbes that is 29.5% and 15.8% respectively **(Esmaeili et al, 2013)**. A similar study was done with Polyurethane (PU) using *Bacillus subtilis* and *P. aeruginosa*. The rate of reduction of weight was found to be maximum in PU and Strum test also showed a high level of carbon dioxide due to the high degradation rate **(Shah et al, 2016).**

Although usage of microbial consortium for bioremediation is very challenging due to various factors such as survival rate, co-ordination between the species, etc. but with the advancement in biotechnology various processes are developed using gene editing, pathways engineering which has helped researchers in overcoming these challenges **(Bernstein and Carlson, 2012; Hays et al., 2015; Song et al., 2014)** . They have developed metabolically engineered microbes by modifying them at the genetic level and made them efficient and stable **(Hays et al, 2015).** Their design and development are based on their interactions, cell-cell communication, metabolic activities, etc. Understanding of these above-mentioned factors will guide us towards the development of a novel microbial consortium for plastic degradation **(Jagmann and Philipp, 2014)**. The microbial consortium has various applications in different fields such as medicine, the food industry, and bioremediation.

Table 1: Microbial consortium involved in the degradation of plastic polymer.

Sr. NO.	PLASTIC POLYMER	MICROBIAL CONSORTIUM	REFERENCE
1	Low-density polyethylene (LDPE)	*Microbacterium paraoxydans*and *Pseudomonas aeruginosa*	Rajandas et al, 2012
2	Polylactide	*Cladosporiumsphaerospermum*, *PenicilliumChrysogenum,Rhodotorula mucilaginosa*and *Serratiamarcescens*	Nair et al, 2016
3	Polyurethane (PU)	*Bacillus subtilis* and *Pseudomonas aeruginosa*	Shah et al, 2016
4	Polylactic acid beverage cups	*Actinomadura (*Dairy wastewater sludge) *and Pseudomonas geniculata*	Pattanasuttichonlakul et al, 2018
5	Polyethylene terephthalate	*Pseudomonas chlororaphis*and *Bacillus Cereus*	Vague et al, 2019

GENETIC ENGINEERING AND GENE EDITING TOOLS

Genetic engineering is being used to manipulate the genome of microbial cells in such a way so that it increases their degradation efficiency **(Dvořák et al., 2017**). Genetic manipulation is done by using various methods such as recombinant DNA technology, gene cloning, and gene modification **(Kumar et al., 2019).** Some species are not able to carry out degradation single-handedly. When the polymer is complex such as hydrocarbons or heavy metals, degradation also becomes difficult. The current challenge for scientists is to create a novel pathway for plastic degradation. Recombinant DNA technology has been used to improve the ability of a microbe to degrade plastic waste. With the help of rDNA technology, microbes are being transformed into a suitable host using appropriate vectors and promoters such that they can metabolize these compounds. Along with this, PCR, antisense RNA, site-directed mutagenesis, etc. is also being used **(Kumar et al, 2018).** The main objective of this type of experimentation is to isolate genes involved in degradation pathways, then modify them and incorporate them into hosts like *E. coli* **(Rajakaruna and Taylor-Robinson, 2016)**. For example, with the help of genetic engineering methods, a plastic degrading enzyme manganese-dependent peroxidase has been produced from genetically engineered *Saccharomyces Cerevisiae* and *E. coli.*Laccase enzyme is also produced from a modified strain of *E. coli* BL21 **(Sharma et al, 2018).**

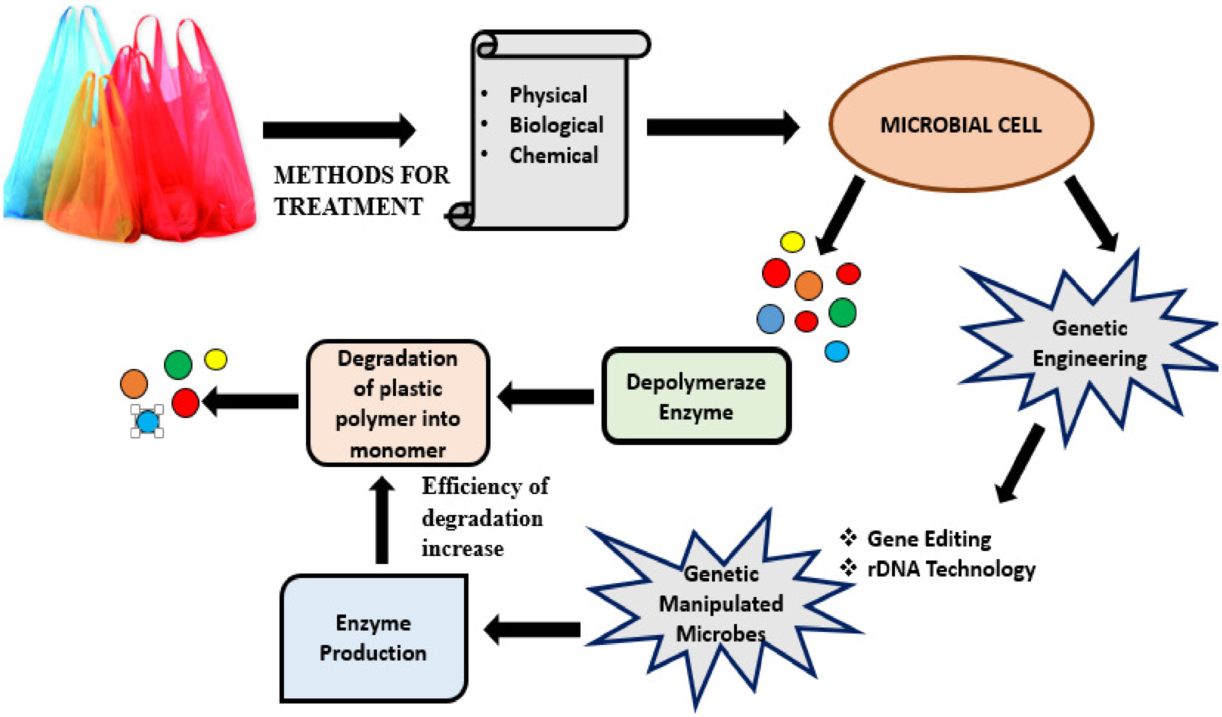

Figure 1: Microbial bioremediation of plastic using genetic engineering.

To carry out complete degradation, genes are complemented by different bacterial species that are genetically engineered. These strains are manipulated in such a way that they are present in the transporter that cooperates with the genes of the degradation pathway to transport and degrade the molecule inside the cell. In the process of PET degradation by *Escherichia coli*. It synthesizes an enzyme LC-cutinase which degrades the plastic polymer and yields terephthalate and ethylene glycol as principal monomers **(iGEM, 2016)**. Further another strain of bacteria *Commamonas testosterone* degrades tetraphthalate into protocatechuate which is then utilized as a nutrient source by *P.putida*(**Jimenez et al, 2002**). As far as ethylene glycol is concerned, it is mineralized by *E.Coli* into CO_2 and H_2O. The bacterial strain is now modified and complemented with other degradation pathway genes to conduct degradation effectively **(iGEM, 2016)**. The plastic polymer is split outside the cell into normal cells, but a changed cell has transporters linked to the degradation pathway for transporting and degrading the plastic polymer within the cell **(iGEM, 2016).**

In the case of polyurethane (PUR) degradation, polyurethane esterase cleaves PUR and produces ethylene glycol which diffuses across the bacterial membrane **(Kang et al., 2011).** A genetically mutated, osmotically inducible gene in *E.coli*, osmY encodes osmotic inducible protein Y, which fuses and exports PUR esterase outside the cell **(Bokinsky et al, 2011; Kang et al., 2011).** The engineered strain also consists of a second plasmid operon composed of glycoaldehyde dehydrogenase (aldA) and glycoaldehydereductase, which allows the central metabolite of the bacteria to use ethylene glycol. Hence, the complete degradation process is being carried out in a single species which makes the bacteria self-sufficient and it is also able to utilize PUR as a nutrient source to convert plastic polymer into bacterial biomass **(Boronat et al., 1983).** Thus, more PUR is degraded in this way and more bioremediation is being done.

ANALYSIS OF PLASTIC DEGRADATION

Plastic degradation can be analyzed by observing the carbon dioxide gas evolution, weight loss percentage, etc(**Shah et al., 2008**). Various tests are performed to determine the rate in plastic composition and its degradation. For example- to measure the carbon dioxide level after the exposure of the plastic polymer to bacteria in activated sludge, an analysis named respirometricanalysis is carried out. For the comparison of loss of weight percentage spectroscopic techniques such as Nuclear Magnetic Resonance (NMR), X-Ray Diffractometer (XRD), Scanning Electron Microscope (SEM), Fourier Transform Infrared Spectroscopy (FTIR) has been used. In the weight loss percentage process, microbes adhere to polymer surface and polymer degradation leads to a loss in weight. For degradation analysis Strum test is done which observes the evolution of metabolic carbon dioxide during the growth period of bacteria (**Gajendiran et al., 2016**). Another analysis is done on the basis of changes observed in tensile strength and color of plastics during degradation **(Dang et al., 2018).**

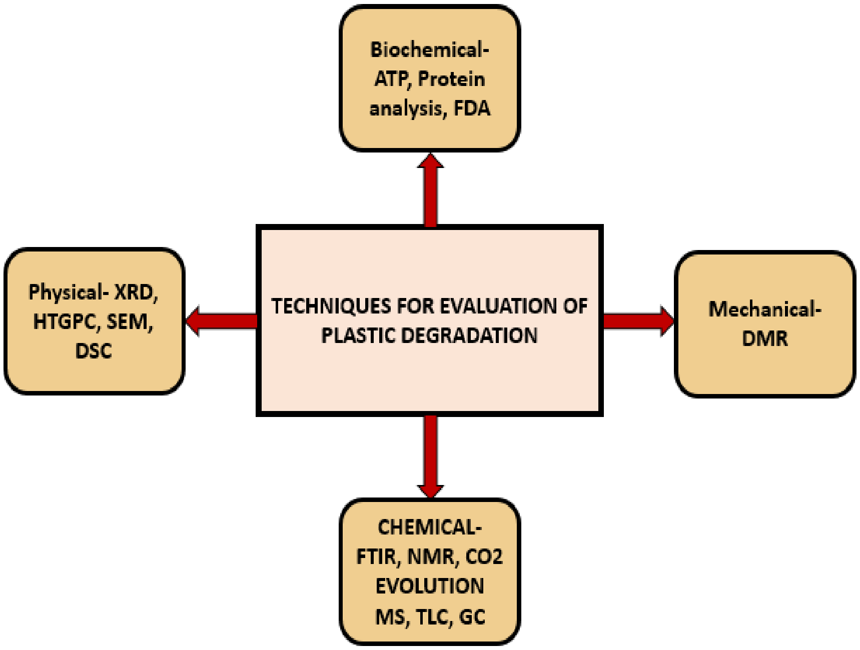

Figure 2: Techniques used for the analysis of plastic degradation.

BACTERIA INVOLVED IN BIODEGRADATION OF SYNTHETIC PLASTIC

1.PE (Polyethylene) – It is a long-chain polymer that is made up of ethylene monomers and is very stable. So it is very difficult to degrade high molecular weight PE. Biodegradation of PE is carried out by two mechanisms that are hydroxy-biodegradation and oxo-biodegradation. Physiochemical pre-treatments such as UV radiation **(Albertsson et al., 1987; Albertsson and Karlsson, 1988, 1990), thermo-oxidation (Lee et al., 1991),** and chemical oxidizing agents **(Brown et al., 1974)** are needed to degrade long-chain PE to carry out the microbial degradation of PE. These pre-treatments are carried out along with the formation of low molecular weight products to depolymerize long-chain PE **(Albertsson et al., 1995, 1998; Erlandsson et al, 1998; Hakkarainen and Albertsson, 2004)**. At the beginning of the degradation process, UV light is used as an activator to activate inert material. After its exposure to UV light, it is also treated with nitric acid, and then it is exposed to the microbial treatment. Several strains have been isolated which has the ability to utilize non-treated PE as a carbon source. For instance, some species namely *Serratiamarcescens, Pharmidiumlucidum* and *Oscillatoriasubbrevis* have been reported as the cause of weight loss of un-pretreated PE during incubation **(Azeko et al., 2015), (Sarmah and Rout, 2018).**

Table 2: Bacteria associated with PE degradation.

Sr. no	Strain	Source	Weight loss percentage of PE	Reference
1.	*Rhodococcusruber*	Soil of disposal site	4	Orr et al, 2004
2.	*Arthrobactersp*	Plastic waste dumpsites	12-15	Balasubramanian et al, 2010
3.	*Phormidiumlucidum*	Domestic sewage water	42	Sarmah and Rout 2018
4.	*Pseudomonas sp.*	Waste dumping soil	5	Tribedi and sil, 2013
5.	*Enterobacterasburiae*	Gut of waxworm	6-11	Yang et al, 2014
6.	*Serratiamarcescens*	Ground soil	36	Azeko et al, 2015
7.	*Pseudomonas sp.*	Soil	-	Yoon et al, 2012
8.	*Bacillus subtilis*	Marine water	1.75	Harshvardhan and Jha, 2013

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2.PS (Polystyrene) – It is a type of synthetic plastic that is being used in the making of disposable cups, packaging materials, electronic items, laboratory wares, etc. It is lightweight plastic and has very good thermal insulating capacity along with stiffness. During pre-treatments such as thermal or chemical treatments, it releases products such as styrene, benzene, toluene, etc which are further degraded by microbes. PS-degrading species of bacteria have been isolated from soil- *Xanthomonas sp., Sphingobacterium sp.,* and *Bacillus sp. Actinomyces*and *Rhodococcusruber* utilizes PS as its sole carbon source for its growth **(Mor and Sivan ,2008).** PS degrading capability was also found in a broader range of mealworms or super worms **(Yang et al., 2018).** Many bacteria have been isolated from the gut of mealworms and super worms that eat plastic. *Agrobacterbeijerinckii*, a lignin-degrading bacterium, secretes the enzyme hydroquinone peroxidase that depolymerizes PS in the presence of non-aqueous into a lower molecular product (**Nakamiya et al., 1997).**

Table 3: Bacteria associated with PS degradation.

Sr. no.	Strain	Source	Weight loss percentage of PS	Reference
1.	*Xanthomonas sp.; Bacillus sp. Sphingobacterium sp.;*	Field soil	40-56	Elsaku et al, 2003
2.	*Rhodococcusruber*	Soil of disposal site	0.8	Mor and Sivan, 2008
3.	*Bacillus sp, Microbacterium sp. Paenibacillusurinalis, Pseudomonas aeruginosa*	Soil buried expanded PS film	-	Atiq et al, 2010
4.	*Exiguobacterium sp.*	Mealworm's gut	7.5	Yang et al, 2015

3.PP (Polypropylene) - Polypropylene is a type of thermoplastic. It is used in the making of plastic moldings, stationery items, packaging materials, etc. During degradation, it is first exposed to UV radiation from sunlight or it can also be oxidized at high temperatures. During the degradation process, a decrease in its viscosity and formation of new groups such as carbonyl group and carboxyl group has been observed. Many soil microbiotas have been tested for their potential to be able to degrade PP. A mesophillic strain *Stenotropomonaspanacihum* isolated from municipal solid waste was able to degrade both low molecular weight and high molecular weight PP with a biodegradability of 12.7%-20.3% in terms of CO□ **(Jeon and Kim, 2016).** However, PP also requires other pre-treatments including thermo-oxidation which helps to facilitate the microbial degradation of PP.

Table 4: Bacteria associated with PP degradation.

Sr. no.	Strain	Source	Weight loss percentage of PP	Reference
1.	*Bacillus flexus, Pseudomonas stutzeri, Bacillus subtilis*	Plastic-dumping site	-	Arkatkar et al, 2010
2.	*Engyodontium album, Phanerochaetechrysosporium*	Plastic-dumping site	4-5	Jeyakumar et al, 2013
3.	*Stenotrophomonaspanacihumi*	Soil of waste storage yard	-	Jeon and Kim, 2016
4.	*Aneurinibacillusaneurinilyticus;Brevibacillusbrevis,Brevibacillusagri; Brevibacillus sp.;*	Landfills and sewage	22.8-27.0	Skariyachan et al, 2018
5.	*Bacillus sp. Rhodococcus sp.*	Mangrove environments	4-6.4	Auta et al, 2018

4.PUR (Polyurethane) – PUR is a thermosetting polymer that does not melt when heated. Due to this property, it is used in the manufacturing of various products such as insulation panels, wheels, and tires, synthetic fibers, elevators, skateboards, etc. High energy UV radiation is used for its degradation which disturbs its chemical structure, before its treatment with microbes. PUR plastic has been reported to be degraded by various bacterial species. From an infected catherer, a strain of *Staphylococcus epidermis* was isolated which utilizes polyether PUR as a nutrient source in the absence of any organic nutrients **(Jansen et al., 1991)**. A bacterium *Comamonasacidovorans* secretes PUR degrading esterase **(Allen et al., 1999),** protease is purified from *Pseudomonas fluorescens***(Vega et al., 1999)** and also three esterases from *Pseudomonas chlororaphis* they have the capacity to emulsify polyester PUR **(Howard et al., 1999; Ruiz et al., 1999).**

Table 5: Bacteria associated with PUR degradation.

Sr. no.	Strain	Source	Weight loss percentage of PUR	Reference
1.	*Corynebacterium sp., Pseudomonas aeruginosa*	Soil	1.2-17.7	Kay et al, 1991
2.	*Bacillus sp.*	Soil	4	Li et al, 1998
3.	*Pseudomonas fluorescens*	Soil	ND	Howard and blake, 1998
4.	*Arthrobactersp.Bacillus sp.* *Pseudomonas sp.* *Micrococcus sp.* *Corynebacterium sp.*	Soil	22	Shah et al, 2008
5.	*Bacillus safensis*	Cedar wood	7	Nakkabi et al, 2015
6.	*Staphylococcus epidermidis*	An intravenous catheter	30	Janseen et al, 1991
7.	*Chaetomiumglobosum*	Soil	-	Darby and Kaplan, 1968
8.	*Pseudomonas chlororaphis*	Soil	-	Howard et al., 1999
9.	*Alicycliphilus*sp.	Decomposed soft foam	-	Oceguera-Cervantes et al., 2007

5. PVC (Polyvinyl Chloride)- It is a thermoplastic polymer and it is very rigid and flexible. It can resist abrasion and chemicals and has low moisture absorption capacity. It is mostly used in the production of pipes, electrical wires, floor coverings, water-resistant items of clothing, etc. PVC with low molecular weight is degraded using microbes. PVC contains the highest proportion of plasticizers which is susceptible to microbial attack. Various bacterial strains isolated from garden soil, waste disposal site, and marine environment have been reported to degrade PVC. However, key enzymes that can be used to degrade PVC are yet not discovered.

Table 6: Bacteria associated with PVC degradation.

Sr. No	Strain	Source	Weight loss percentage of PVC	Reference
1.	*Bacillus* sp.	Marine	0.26	Kumari et al., 2019
2.	*Pseudomonas citronellolis*	Soil	13	Giacomucci et al., 2019
3.	*Acanthopleurobacterpedis*; *Bacillus cereus*; *Pseudomonas otitidis*; *Bacillus aerius*;	Plastic disposal sites	-	Anwar et al., 2016
4.	*Mycobacterium* sp.	Garden soil	-	Nakamiya et al., 2005
5.	*Chryseomicrobiumimtechense*;	Landfill leachate	-	Latorre et al., 2012

CONCLUSION

Plastics pose a great environmental challenge and it is very difficult to solve this issue. Compared to other physical and chemical approaches, the use of microbes to solve this problem is a better strategy. Various numbers of plastic-degrading bacteria have been isolated from the soil, as described above, and enzymes have been obtained from them. However, these enzymes are not capable of contributing very effectively to plastic breakdown.Therefore, attempts are being made to find many more enzymes and also to discover novel mechanisms of degradation that can be genetically engineered to produce better results. Bacteria have introduced both natural and engineered mechanisms of plastic degradation. Study on different strains of bacteria is also being carried out and their capacity for degradation is being tested. The use of genetic engineering has been of great benefit in modifying and developing the ability of bacteria to do this job. On the other hand, the development of these microbes to enhance the activity of the enzymes they secrete will be beneficial and will also improve the efficiency of enzymatic degradation. It is also possible to manipulate microbial metabolic pathways so that whole bacteria can serve as a plastic-degrading factory.All of this plastic biodegradation research has increased our awareness of these degrading microbes, their plastic enzymatic behaviour, how their genes are involved, and the structure of polymers for biodegradation. This knowledge has proved to be useful for these microbes' molecular evolution and reproduction.Our enhanced technologies can also help to establish innovative biotechnological approaches for the disposal of waste. Microbes thus serve as a reliable and eco-friendly plastic remediation tool.

REFERENCES

Ahmed, T., Shahid, M., Azeem, F., Rasul, I., Shah, A.A., Noman, M., Muhammad, S., et al., 2018. Biodegradation of plastics: current scenario and future prospects for environmental safety. Environ. Sci. Pollut. Res. 1–12.

Albertsson, A. C., Andersson, S. O., and Karlsson, S. (1987). The mechanism of biodegradation of polyethylene. *Polym.Degrad. Stab.* 18, 73–87. doi: 10.1016/ 0141-3910(87)90084-x

Albertsson, A. C., Barenstedt, C., Karlsson, S., and Lindberg, T. (1995). Degradation product pattern and morphology changes as means to diffffferentiateabiotically and biotically aged degradable polyethylene. *Polymer* 36, 3075–3083.doi: 10.1016/0032-3861(95)97868-g

Albertsson, A. C., Erlandsson, B., Hakkarainen, M., and Karlsson, S. (1998). Molecular weight changes and polymeric matrix changes correlated with the formation of degradation products in Biodegraded Polyethylene. *J. Environ. Polym.Degrad.* 6, 187–195.

Allen, A. B., Hilliard, N. P., and Howard, G. T. (1999).Purification and characterization of a soluble polyurethane degrading enzyme from *Comamonasacidovorans.Int. Biodeter. Biodegr.*43, 37–41.

Andrady, A.L., 2011. Microplastics in the marine environment. Mar. Pollut. Bull. 62 (8), 1596–1605.

Anwar, M. S., Kapri, A., Chaudhry, V., Mishra, A., Ansari, M. W., Souche, Y., et al. (2016). Response of indigenously developed bacterial consortia in progressive degradation of polyvinyl chloride.*Protoplasma*253, 1023–1032. doi: 10.1007/ s00709-015-0855-9

Arkatkar, A., Juwarkar, A. A., Bhaduri, S., Uppara, P. V., and Doble, M. (2010). Growth of Pseudomonas and Bacillus biofilms on pretreated polypropylene surface. Int. Biodeter. Biodegr. 64, 530–536. doi: 10.1016/j.ibiod.2010.06.002

Atiq, N., Garba, A., Ali, M. I., Andleeb, S., Khan, N. A., and Robson, G. D. (2010).Isolation and identification of polystyrene biodegrading bacteria from soil. Afr. J. Microbiol. Res. 4, 1537–1541.

Auta, H. S., Emenike, C. U., Jayanthi, B., and Fauziah, S. H. (2018). Growth kinetics and biodeterioration of polypropylene microplastics by Bacillus sp. and Rhodococcus sp. isolated from mangrove sediment. Mar. Pollut. Bull. 127, 15–21. doi: 10.1016/j.marpolbul.2017.11.036

Azeko, S. T., Etuk-Udo, G. A., Odusanya, O. S., Malatesta, K., Anuku, N., and Soboyejo, W. O. (2015). Biodegradation of linear low density polyethylene by Serratiamarcescens subsp. marcescens and its cell free extracts. Waste Biomass Valor. 6, 1047–1057. doi: 10.1007/s12649-015-9421-0

Balasubramanian, V., Natarajan, K., Hemambika, B., Ramesh, N., Sumathi, C. S., Kottaimuthu, R., et al. (2010). High–density polyethylene (HDPE)- degrading potential bacteria from marine ecosystem of Gulf of Mannar, India. Lett. Appl. Microbiol. 51, 205–211. doi: 10.1111/j.1472-765X.2010. 02883.

Bernstein, H.C., Carlson, R.P., 2012. Microbial consortia engineering for cellular factories: in vitro to in silico systems. Comput.Struct.Biotechnol. J. 3, 201210017.

Bokinsky, G., Peralta-Yahya, P.P., George, A., Holmes, B.M., Steen, E.J., Dietrich, J., Lee, T.S., Tullman-Ercek, D., Voigt, C.A., Simmons, B.A. and Keasling, J.D. (2011). Synthesis of three advanced biofuels from ionic liquid-pretreated switchgrass using engineered Escherichia coli. *Proceedings of the National Academy of Sciences*, 108: 19949-19954. https:// doi.org/10.1073/pnas.1106958108

Boronat, A., Caballero, E. and Aguilar, J. (1983).Experimental evolution of a metabolic pathway for ethylene glycol utilization by Escherichia coli. Journal of Bacteriology, 153: 134-139

Cooper, D.A. and Corcoran, P.L. (2010).Effects of mechanical and chemical processes on the degradation of plastic beach debris on the island of Kauai, Hawaii. Marine Pollution Bulletin, 60: 650-654. https://doi.org/10.1016/ j.marpolbul.2009.12.026

Das, M.P. and Kumar, S.A.N.T.O.S.H. (2013). Influence of cell surface hydrophobicity in colonization and biofilm formation on LDPE biodegradation. International Journal Pharmacy and Pharmaceutical Sciences, 5: 690-694. ISSN- 0975-1491

Darby, R. T., and Kaplan, A. M. (1968).Fungal susceptibility of polyurethanes.*Appl. Microbiol.* 16, 900–905. doi: 10.1128/aem.16.6.900-905.1968

Dussud, C., Ghiglione, J.F., 2014. Bacterial degradation of synthetic plastics. In: CIESM Workshop Monogr., No. 46.

Dvořák, P., Nikel, P.I., Damborský, J., de Lorenzo, V., 2017. Bioremediation 3.0: Engineering pollutant-removing bacteria in the times of systemic biology. Biotech. Adv. 35, 845–866.

Eisaku, O., Linn, K., Takeshi, E., Taneaki, O., and Yoshinobu, I. (2003). Isolation and characterization of polystyrene degrading microorganisms for zero emission treatment of expanded polystyrene. Environ. Eng. Res. 40, 373–379.

Erlandsson, B., Karlsson, S., and Albertsson, A. C. (1998).Correlation between molar mass changes and CO2 evolution from biodegraded 14C-labeled ethylene-vinyl alcohol copolymer and ethylene polymers.*ActaPolym.*49, 363– 370.doi: 10.1002/(sici)1521-4044(199807)49:7<363::aid-apol363>3.0.co;2-u

Esmaeili, A., Pourbabaee, A.A., Alikhani, H.A., Shabani, F., Esmaeili, E., 2013. Biodegradation of low-density polyethylene (LDPE) by mixed culture of *Lysinibacillusxylanilyticus*and *Aspergillusniger*in soil. PLoS One 8 (9), e71720.

Gajendiran, A., Krishnamoorthy, S., Abraham, J., 2016. Microbial degradation of low-density polyethylene (LDPE) by *Aspergillusclavatus*strain JASK1 isolated from landfill soil. 3. Biotech 6 (1), 52.

Gewert, B., Plassmann, M.M. and MacLeod, M. (2015).Pathways for degradation of plastic polymers floating in the marine environment. Environmental Science: Processes & Impacts, 17: 1513-1521. https://doi.org/10.1039/C5EM00207A.

Giacomucci, L., Raddadi, N., Soccio, M., Lotti, N., and Fava, F. (2019).Polyvinyl chloride biodegradation by *Pseudomonas citronellolis*and *Bacillus flflexus*.*New Biotechnol.*52, 35–41. doi: 10.1016/j.nbt.2019.04.005

Gu, J.D. (2003). Microbiological deterioration and degradation of synthetic polymeric materials: recent research advances. International Biodeterioration& Biodegradation, 52: 69-91. https://doi.org/10.1016/S0964-8305(02)00177-4.

Hakkarainen, M., and Albertsson, A. C. (2004). “Environmental degradation of polyethylene,” in *Long Term Properties of Polyolefifins*, ed. A. C. Albertsson (Berlin: Springer), 177–200. doi: 10.1007/b13523

Harshvardhan, K., and Jha, B. (2013).Biodegradation of low–density polyethylene by marine bacteria from pelagic waters, Arabian Sea, India.*Mar. Pollut. Bull.* 77, 100–106. doi: 10.1016/j.marpolbul.2013.10.025

Hays, S.G., Patrick, W.G., Ziesack, M., Oxman, N., Silver, P.A., 2015. Better together: engineering and application of microbial symbioses. Curr.Opin.Biotechnol. 36, 40–49.

Horton, A.A., Walton, A., Spurgeon, D.J., Lahive, E., Svendsen, C., 2017. Microplastics in freshwater and terrestrial environments: Evaluating the current understanding to identify the knowledge gaps and future research priorities. Sci. Total Environ. 586, 127–141.

Howard, G. T., and Blake, R. C. (1998).Growth of Pseudomonas fluorescens on a polyester–polyurethane and the purification and characterization of a polyurethanase–protease enzyme. Int. Biodeterior. Biodegrad. 42, 213–220. doi: 10.1016/s0964-8305(98)00051-1

Howard, G. T., Ruiz, C., and Hilliard, N. P. (1999).Growth of *Pseudomonas chlororaphis*on a polyester–polyurethane and the purification and characterization of a polyurethanase–esterase enzyme.*Int. Biodeterior. Biodegrad.*43, 7–12. doi: 10.1016/s0964-8305(98)00057-2

International Genetically Engineered Machine Competition (iGEM) (2016). *Plasticure.*Ben-Gurion University of the Negev.*SaheliGhosh et al. (2019)*197 http://2016.igem.org/Team:BGU_ISRAEL/Description

Jagmann, N., Philipp, B., 2014. Reprint of design of synthetic microbial communities for biotechnological production processes. J. Biotech. 192, 293–301.

Jansen, B., Schumacher-Perdreau, F., Peters, G., and Pulverer, G. (1991).Evidence for degradation of synthetic polyurethanes by Staphylococcus epidermidis.Zentralbl.Bakteriol. 276, 36–45. doi: 10.1016/s0934-8840(11)80216-1

Jeon, H. J., and Kim, M. N. (2016a).Comparison of the functional characterization between alkane monooxygenases for low-molecular-weight polyethylene biodegradation. Int. Biodeter. Biodegr. 114, 202–208. doi: 10.1016/j.ibiod.2016. 06.012

Jeyakumar, D., Chirsteen, J., and Doble, M. (2013). Synergistic effects of pretreatment and blending on fungi mediated biodegradation of polypropylenes. Bioresour. Technol. 148, 78–85. doi: 10.1016/j.biortech. 2013.08.074

Jimenez, J.I., Minambres, B., Garcia, J.L. and Diaz, E. (2002).Genomic analysis of the aromatic catabolic pathways from *Pseudomonas putida*KT2440.*Environmental Microbiology*, 4: 824-841. https://doi.org/10.1046/j.1462-2920.2002.00370.x

Joo, S., Cho, I.J., Seo, H., Son, H.F., Sagong, H.Y., Shin, T.J., Choi, S.Y., Lee, S.Y. and Kim, K.J. (2018). Structural insight into molecular mechanism of poly (ethylene terephthalate) degradation.*Nature Communications*, 9: 382.https://doi.org/10.1038/s41467-018-02881-1.

Kale, S.K., Deshmukh, A.G., Dudhare, M.S., Patil, V.B., 2015. Microbial degradation of plastic: a review. J. Biochem. Technol. 6 (2), 952–961.

Kang, C.H., Oh, K.H., Lee, M.H., Oh, T.K., Kim, B.H. and Yoon, J.H. (2011).A novel family VII esterase with industrial potential from compost metagenomic library.*Microbial Cell Factories*, 10: 41. https://doi.org/10.1186/1475-2859-10-41

Kawai, F., Watanabe, M., Shibata, M., Yokoyama, S., Sudate, Y. and Hayashi, S., (2004). Comparative study on biodegradability of polyethylene wax by bacteria and fungi. Polymer Degradation and Stability, 86: 05-114. https://doi.org/10.1016/ j.polymdegradstab.2004.03.015.

Kay, M. J., Morton, L. H. G., and Prince, E. L. (1991).Bacterial degradation of polyester polyurethane. Int. Biodeter. Biodegr. 27, 205–222. doi: 10.1016/0265- 3036(91)90012-

Kedzierski, M., d'Almeida, M., Magueresse, A., Le Grand, A., Duval, H., César, G., Sire, O., Bruzaud, S., Le Tilly, V., 2018.Threat of plastic ageing in marine environment. Adsorption/desorption of micropollutants. Mar. Pollut. Bull. 127, 684–694.

Koutny, M., Lemaire, J. and Delort, A.M. (2006). Biodegradation of polyethylene films with prooxidant additives. Chemosphere, 64: 1243-1252. https://doi.org/10.1016/j.chemosphere.2005.12.060

Kumar, M., Jaiswal, S., Sodhi, K.K., Shree, P., Singh, D.K., Agrawal, P.K., Shukla, P., 2019. Antibiotics bioremediation: Perspectives on its ecotoxicity and resistance. Environ. Int. 124, 448–461.

Kumar, N.M., Muthukumaran, C., Sharmila, G., Gurunathan, B., 2018.Genetically modified organisms and its impact on the enhancement of bioremediation. In: Bioremediation: Applications for Environmental Protection and Management. Springer, Singapore, pp. 53–76.

Kumari, A., Chaudhary, D. R., and Jha, B. (2019). Destabilization of polyethylene and polyvinylchloride structure by marine bacterial strain. *Environ. Sci. Pollut. Res. Int.* 26, 1507–1516. doi: 10.1007/s11356-018-3465-1

Latorre, I., Hwang, S., and Montalvo-Rodriguez, R. (2012). Isolation and molecular identifification of landfifill bacteria capable of growing on di-(2-ethylhexyl) phthalate and deteriorating PVC materials.*J. Environ. Health Sci. Part A* 47, 2254–2262. doi: 10.1080/10934529.2012.707549

Lee, B., Pometto, A. L., Fratzke, A., and Bailey, T. B. (1991). Biodegradation of degradable plastic polyethylene by *Phanerochaete*and *Streptomyces* species. *Appl. Environ. Microbiol.*57, 678–685. doi: 10.1128/aem.57.3.678-685.1991
Leja, K. and Lewandowicz, G. (2010).Polymer Biodegradation and Biodegradable Polymers-a Review. Polish Journal of Environmental Studies, 19:255-266.

Li, W. J., Jayakody, L. N., Franden, M. A., Wehrmann, M., Daun, T., Hauer, B., et al. (2019). Laboratory evolution reveals the metabolic and regulatory basis of ethylene glycol metabolism by Pseudomonas putida KT2440. Environ. Microbiol. 21, 3669–3682. doi: 10.1111/1462-2920.14703

Mahalakshmi, V. and Siddiq, S.A. (2015).Enhanced biodegradation of polyethylene by development of a consortium.Pelagia Research Library, Advances in Applied Science Research. 6:183-189. ISSN: 0976-8610.

Mor, R., and Sivan, A. (2008). Biofilm formation and partial biodegradation of polystyrene by the actinomyceteRhodococcusruber.Biodegradation 19, 851– 858.doi: 10.1007/s10532-008-9188-0

Mrowiec, Bozena, 2017. Plastic pollutants in water environment.OchronaSrodowiska i Zasobów Nat. 28, http://dx.doi.org/10.1515/oszn-2017-0030.

Nakamiya, K., Hashimoto, S., Ito, H., Edmonds, J. S., Yasuhara, A., and Morita, M. (2005). Microbial treatment of bis (2-ethylhexyl) phthalate in polyvinyl chloride with isolated bacteria.*J. Biosci. Bioeng.*99, 115–119. doi: 10.1263/jbb.99.115

Nakamiya, K., Ooi, T., and Kinoshita, S. (1997).Non-heme hydroquinone peroxidase from *Azotobacterbeijerinckii*HM121.*J. Ferment. Bioeng.*84, 14–21. doi: 10.1016/s0922-338x(97)82780-8

Nair, N.R., Sekhar, V.C., Nampoothiri, K.M., 2016. Augmentation of a microbial consortium for enhanced polylactide (PLA) degradation.Indian J. Microbiol. 56 (1), 59–63.

Nakkabi, A., Sadiki, M., Fahim, M., Ittobane, N., Saad, I. K., Barkai, H., et al. (2015a). Biodegradation of poly(ester urethane)s by Bacillus subtilis. Int. J. Environ. Res. 9, 157–162

Navarro-Díaz, M., Valdez-Vazquez, I., Escalante, A.E., 2016. Ecological perspectives of hydrogen fermentation by microbial consortia: What we have learned and the way forward. Int. J. Hydrogen Energy 41, 17297–17308.

Oceguera-Cervantes, A., Carrillo-García, A., López, N., Bolaños-Nuñez, S., Cruz-Gómez, M. J., Wacher, C., et al. (2007). Characterization of the polyurethanolytic activity of two *Alicycliphilus*sp. strains able to degrade polyurethane and n-methylpyrrolidone. *Appl. Environ. Microbiol.*73, 6214– 6223.doi: 10.1128/aem.01230-07

Okada, M., 2002.Chemical syntheses of biodegradable polymers.Prog.Polym. Sci. 27, 87–133.

Orr, I. G., Hadar, Y., and Sivan, A. (2004).Colonization, biofilm formation and biodegradation of polyethylene by strain of Rhodococcusruber. Appl. Microbiol. Biotechnol. 65, 97–104.

Pathak, V.M. and Navneet (2017). Review on the current status of polymer degradation: a microbial approach. Bioresources and Bioprocessing, 4: 15. https://doi.org/10.1186/s40643-017-0145-9. Poonam, K., Rajababu, V., Yogeshwari, J. and Patel, H. (2013). Diversity of plastic degrading microorganisms and their appraisal on biodegradable plastic. Applied Ecology and Environmental Research, 11: 441-449. ISSN 1785 0037.

Pattanasuttichonlakul, W., Sombatsompop, N., Prapagdee, B., 2018. Accelerating biodegradation of PLA using microbial consortium from dairy wastewater sludge combined with PLA-degrading bacterium. Int. BiodeteriorationBiodegrad. 132, 74–83.

Rajakaruna, S.S., Taylor-Robinson, A.W., 2016.Application of recombinant DNA technology (genetically modified organisms) to the advancement of agriculture, medicine, bioremediation and biotechnology industries. J. Appl. Biotechnol. Bioeng.1, 00013.

Rajandas, H., Parimannan, S., Sathasivam, K., Ravichandran, M., Yin, L.S., 2012. A novel FTIR-ATR spectroscopy based technique for the estimation of low-density polyethylene biodegradation. Polymer Testing 31 (8), 1094–1099.

Ruiz, C., and Howard, G. T. (1999).Nucleotide sequencing of a polyurethanase gene (pulA) from *Pseudomonas flfluorescens.Int. Biodeter. Biodegr.*44, 127–131. doi: 10.1016/s0964-8305(99)00074-8

Sarmah, P., and Rout, J. (2018). Efficient biodegradation of low-density polyethylene by cyanobacteria isolated from submerged polyethylene surface in domestic sewage water. Environ. Sci. Pollut. Res. Int. 25, 33508–33520. doi: 10.1007/s11356-018-3079-7

Seneviratne, G., Tennakoon, N.S., Weerasekara, M.L., Nandaseha, K.A. (2006): Polyethylene biodegradation by a developed Penicillium-Bacillus biofilm. Current Science, 90: 20-21.

Shah, A. A., Hasan, F., Akhter, J. I., Hameed, A., and Ahmed, S. (2008). Degradation of polyurethane by novel bacterial consortium isolated from soil. Ann. Microbiol. 58, 381–386. doi: 10.1007/bf03175532

Shah, Z., Gulzar, M., Hasan, F., Shah, A.A., 2016. Degradation of polyester polyurethane by an indigenously developed consortium of Pseudomonas and Bacillus species isolated from soil. Polym.Degrad. Stab. 134, 349–356.

Shalini, R. and Sasikumar, C. (2015).Biodegradation of Low Density Polythene Materials Using Microbial Consortium–An Overview.*International Journal of Pharmaceutical and Chemical Sciences*, 4: 507-514. ISSN: 2277-5005

Sharma, B., Dangi, A.K., Shukla, P., 2018. Contemporary enzyme based technologies for bioremediation: a review. J. Environ. Manag. 210, 10–22.

Singh, B. and Sharma, N. (2008).Mechanistic implications of plastic degradation.*Polymer Degradation and Stability*, 93: 561-584. https://doi.org/10.1016/j.polymdegradstab.2007.11.008

Skariyachan, S., Patil, A. A., Shankar, A., Manjunath, M., Bachappanavar, N., and Kiran, S. (2018). Enhanced polymer degradation of polyethylene and polypropylene by novel thermophilic consortia of Brevibacillussps.andAneurinibacillus sp. screened from waste management landfills and sewage treatment plants. Polym.Degrad. Stab. 149, 52–68. doi: 10.1016/j. polymdegradstab.2018.01.018

Slor, G., Papo, N., Hananel, U., Amir, R.J., 2018. Tuning the molecular weight of polymeric amphiphiles as a tool to access micelles with a wide range of enzymatic degradation rates. Chem. Commun..

Song, H., Ding, M.Z., Jia, X.Q., Ma, Q., Yuan, Y.J., 2014. Synthetic microbial consortia: from systematic analysis to construction and applications. Chem. Soc. Rev. 43, 6954–6981.

Tribedi, P., and Sil, A. K. (2013).Low-density polyethylene degradation by Pseudomonas sp, AKS2 biofilm. Environ. Sci. Pollut. Res. Int. 20, 4146–4153.doi: 10.1007/s11356-012-1378-y

Upreti, M.C. and Srivastava, R.B. (2003).A potential Aspergillus species for biodegradation of polymeric materials. Current Science, 84: 1399-1402. https://doi.org/10.2307/24108383. Urbanek, A.K., Rymowicz, W. and Mirończuk, A.M. (2018).Degradation of plastics and plastic-degrading bacteria in cold marine habitats. Applied Microbiology and Biotechnology, 102: 7669-7678. https://doi.org/10.1007/s00253-018-9195-y.

Vague, M., Chan, G., Roberts, C., Swartz, N.A., Mellies, J.L., 2019. Pseudomonas isolates degrade and form biofilms on polyethylene terephthalate (PET) plastic. bioRxiv. 647321.

Villaverde, J., Rubio-Bellido, M., Merchán, F., Morillo, E., 2017. Bioremediation of diuron contaminated soils by a novel degrading microbial consortium. J. Environ. Manag. 188, 379–386.

Vega, R. E., Main, T., and Howard, G. T. (1999).Cloning and expression in *Escherichia coli* of a polyurethane-degrading enzyme from *Pseudomonas flfluorescens.Int. Biodeter. Biodegr.*43, 49–55. doi: 10.1016/s0964-8305(98)
00068-7

Yang, J., Yang, Y., Wu, W. M., Zhao, J., and Jiang, L. (2014).Evidence of polyethylene biodegradation by bacterial strains from the guts of plastic-eating waxworms. Environ. Sci. Technol. 48, 13776–13784. doi: 10.1021/es504038a

Yang, Y., Chen, J., Wu, W. M., Zhao, J., and Yang, J. (2015a). Complete genome sequence of Bacillus sp. YP1, a polyethylene-degrading bacterium from waxworm's gut. J. Biotechnol. 200, 77–78. doi: 10.1016/j.jbiotec.2015.02.034

Yoon, M. G., Jeon, H. J., and Kim, M. N. (2012). Biodegradation of polyethylene by a soil bacterium and *AlkB*cloned recombinant cell. *J. Bioremediat. Biodegrad.*3:145.