Proceedings of the 3rd International Conference on Green Energy, Environment and Sustainable Development (GEESD2022), X. Zhang et al. (Eds.)

doi:10.3233/ATDE220362

Biodegradation of Microplastics in Soil

Yanni YU, Yaning LUAN[1] and Wei DAI[2]
The Key Laboratory for Silviculture and Conservation of Ministry of Education, College of Forestry, Beijing Forestry University, Beijing, China

Abstract. Recently, microplastic pollution has become an environmental issue with widespread concern. As a bigger reservoir of microplastics than the ocean, microplastic pollution borne by soil has an impact on its structure and properties that cannot be ignored. Furthermore, with the help of plants, soil animals and microorganisms, this emerging pollutant will eventually threaten human health. Microplastics are difficult to recycle due to their small particle size, making it hard for traditional methods of plastic pollution management to work. In contrast, biodegradation methods are gaining attention because of its environmental sustainability and wide treatment range. This paper reviews the most popular microplastic biodegradation methods and their influencing factors, aiming to provide ideas and references for the biodegradation of microplastics.

Keywords. Soil, microplastics, biodegradation

1. Introduction

Microplastics, as an emerging pollutant, has attracted much attention worldwide in recent years. It is usually defined as plastic fragments less than 5 mm in diameter and was first introduced by Thompson in 2004 [1]. In addition to microplastics produced for specific applications (e.g., abrasives in toiletries or drug carriers), some hard-to-degrade plastics can gradually crush into smaller particles after entering the environment and cannot be completely removed even after special treatment. People have started the research on microplastics for decades, especially the nature, characteristics and hazards of microplastics in water have been well understood, while the understanding on microplastics in soil is in need of improvement. However, it is suggested that soil may be a larger reservoir with a much higher abundance and input of microplastics than the ocean [2]. Microplastics entering the soil environment, under the influence of their own properties and the plasticizers, stabilizers, and flame retardants contained in them, can affect soil physicochemical properties, soil functions, and biodiversity, and even have compound pollution with other pollutants in the soil environment [3, 4]. In addition, due to the nature of MPs that can easily migrate and transform with the environment, they also have the risk of endangering animal and human health through the food chain.

Current methods for dealing with plastics include recycling, landfilling, thermal, mechanical and biological degradation [5], but microplastics in soil are difficult to

[1] Corresponding Author, Yaning LUAN, The Key Laboratory for Silviculture and Conservation of Ministry of Education, College of Forestry, Beijing Forestry University, Beijing, China; Email: luanyaning@bjfu.edu.cn.
[2] Corresponding Author, Wei DAI, The Key Laboratory for Silviculture and Conservation of Ministry of Education, College of Forestry, Beijing Forestry University, Beijing, China; Email: daiwei1@bjfu.edu.cn.

recycle or enrich, not to mention conventional methods, due to their small size and widespread incorporation everywhere. In contrast, biodegradation methods are currently considered the best way to manage microplastic pollution because they can effectively degrade microplastics in a large area without damaging the surrounding environment and biota. Currently, the most common microplastic biodegradation methods include animal (insect) degradation, microbial degradation and enzyme degradation. However, since the related research is just in its infancy, researchers do not know the specific mechanisms of microplastic degradation by these methods, and there are still many urgent problems to be solved in order to apply them to industrial microplastic treatment. Therefore, this article reviews the hot microplastic biodegradation methods and their influencing factors in recent years, and makes suggestions for the development of biodegradation methods combined with their shortcomings, in order to provide references and ideas for further research directions.

2. Biodegradation Methods of Microplastics in Soil

2.1. Soil Animal-Mediated Microplastic Degradation

Soil animals are essential consumers in the material cycle of ecosystems that can actively assimilate various substances from the environment for their own growth, development and other activities. Studies have shown that some insects (including some invertebrates [6-8] and social insects [9]) are able to chew and feed on plastic products and use them as the sole source of carbon, converting microplastics into CO_2 and H_2O by physical means such as biting, chewing or digesting and a series of biochemical processes [10]. Table 1 summarizes the studies on microplastic degradation by soil animals, and it is easy to see that current studies are focusing on flour beetle, barley pest and waxworm larvae. The degradation of microplastics by these insects is mainly attributed to the gut microbial symbionts. For example, *Aspergillus flavus* is the main cause of microplastic degradation by *G. mellonella* due to its ability to produce Lac and Lac-like multi-copper oxidases [11]; waxworm (*P. interpunctella* larvae) degrading PE films is also achieved by *Bacillus* and *Enterobacteriaceae* in their gut [7].

2.2. Microbial-Mediated Microplastic Degradation

Microorganisms can well adapt to almost all environments and have the ability to decompose different organic pollutants, thus being a green and economical means of cleaning up microplastics [17]. In the degradation process, microplastics are used by microorganisms as a substrate for biofilm growth. As the biofilm grows, the structure of the microplastic breaks down and then enzymes secreted by bacteria/fungi break down the microplastic fragments by specific and non-specific actions. When the fragment weight is below 600 kDa, the microplastics are easily taken up by microorganisms in the biofilm during assimilation. Thereafter, the assimilated fragments can be further broken down by enzymes into smaller molecules (CO_2, N_2, CH_4, H_2O, H_2S), which are used by microorganisms as an energy source and finally returned to the atmosphere. Table 2 summarizes the studies on microbial degradation of microplastics, including the species and sources of microorganisms that degrade plastics, as well as the types of plastics. It is no hard to see that the target material is mainly PE while studies on other plastics are relatively few.

Table 1. Animal degradation of soil microplastics.

Animals	Plastic	Degradation effect	Ref.
Tenebrio molitor	PE	49.0±1.4% of the ingested PE was converted to CO_2	[12]
Tenebrio molitor	PS	Degradation rate was 50% per day.	[13]
Z. atratus	PS	Digested 0.58 mg/day/superworm.	[6]
G. mellonella	PP, PE	Degrade 92% of plastic materials, the degradation rate was 1.84 mg/day/worm.	[8]
Achatina fulica	PS	18.5 ± 2.9 mg polystyrene was ingested per snail.	[14]
Zophobas atratus	EPS, LDPE	Within 33 days, the consumption rates of LDPE and PS foams are 58.7 ± 1.8 mg and 61.5 ± 1.6 mg 100 larvae^{-1}d^{-1}.	[15]
Plodia interpunctella	PE	Two bacterial strains isolated from the worm's gut degraded approximately 6.1 ± 0.3% and 10.7 ± 0.2% of the PE films in 60 days.	[7]
Achroia grisella	HDPE	Over an 8-day period, PE consumption increased with an ingestion of 1.83 mg of PE per day per larvae.	[16]

Note: PE is polyethylene, PU is polyurethane, PP is polypropylene, PS is polystyrene, EPS is expanded polystyrene, LDPE is low density polyethylene, HDPE is high density polyethylene.

As one of the most diverse species of organisms in the soil environment, the ability of fungi to degrade microplastics has been widely demonstrated [18]. For example, 27 genera in the phylum Ascomycota (e.g., genus of *Aspergillus* and *Fusarium*), followed by *Basidiomycota* and *Zygomycota*, are associated with microplastic degradation of microplastics [19]. In addition, bacteria like *Bacillus, Pseudomonas, Rhodococcus, Arthrobacter*, *Oscillatoria subbrevis* and *Alcanivorax borkumensis* have also been reported to degrade microplastic [20]. Since the essence of microplastic degradation by insects is the action of their gut microorganisms, the isolation of effective microorganisms from the insect gut is a way to isolate microplastic degradable microorganisms other than directly from the plastic circle.

In terms of degradation efficiency, the results of single culture studies showed that fungi generally had better degradation ability than bacteria for following reasons: the number of enzymes produced by fungi was significantly higher than that of bacteria, and the mycelium of fungi could adhere more firmly to the surface of microplastics and might penetrate into the interior of the particles, furthermore, fungi could reduce the hydrophobicity of microplastics by promoting the formation of chemical bonds such as carbonyl, carboxyl and ester groups in microplastics. However, bacteria require a much less stable external environment than fungi, and therefore should also have a greater degradation potential [21]. Despite the large number of studies devoted to the screening of microplastic-degrading bacteria, the degradation velocities of the screened microorganisms are generally slow, requiring at least 21 days to achieve the desired decomposition [22]. In addition, the degradation effect of single cultures of both fungi and bacteria is generally limited, and the effect can only be enhanced by the synergistic effect between mixed cultures of fungi and bacteria. However, since the interaction mechanism between microorganisms and microplastics is still unclear and the collaboration between different flora is complicated, a lot of research is needed to achieve the goal of achieving the desired degradation effect by adjusting the ratio between strains of bacteria.

2.3. Enzyme-Mediated Microplastic Degradation

The mechanism of plastic biodegradation involves various biochemical reactions and the ones mediated by microorganisms is essentially triggered by enzyme [31]. Enzymes that degrade plastics can be classified into intracellular and extracellular enzymes according to their location of action. Intracellular enzymes such as esterase and lipases catalyze the hydrolysis of ester bonds in polymeric esters [32], for example, esterase degrade PE, PET, and PVC [33] and lipases degrade PET and PU [34]; extracellular enzymes degrade complex polymers by breaking them down into oligomers, dimers, or monomers that can be absorbed into the body by microorganisms and then further broken down by intracellular enzymes [5]. So far, the types of enzymes identified as having degrading effects on microplastics include proteases, lipases, keratases, insect laccase, manganese peroxidase, lignin peroxidase, and alkane hydroxylase [35].

Table 2. Microbial degradation of soil microplastics.

Microorganisms	**Microbial sources**	**Plastic**	**Degradation effect**	**Ref.**
Bacillus bacterial	Mangrove environments	PE, PET, PP, PS	Degrade a mixture of various plastics	[23]
Ideonella sakaiensis 201-F6	Natural microbial communities exposed to PET	PET	Produces two enzymes that convert PET efficiently into its two environmentally benign monomers	[24]
Zalerion maritimum	Marine environment	PE	Decrease the mass and size of the pellets.	[25]
Bacillus subtilis	Nutrient medium	PE	Lost 9.26% weight in 30 days	[26]
Kocuria palustris M16, *Bacillus pumilus* M27, *Bacillus subtilis* H1584	Arabian sea	PE	Lost 1%, 1.5% and 1.75% weight after 30 days	[27]
Pestalotiopsis microspora	Ecuadorian rainforest	PU	Degraded the polyester diol portion and produced degradation metabolites that used as carbon source to mineralize the polymer.	[28]
Aspergillus terreus, *Aspergillus sydowii*	Rhizosphere soil of Avicennia marina	PE	Most efficient polythene deteriorating fungal	[29]
C. pseudocladosporioides	Mineral medium	PU	Degraded up to 87% after 14 days	[30]

Research on enzymatic degradation of microplastics has just started, and the number of studies is small. The types of enzymes studied at present are mainly extracellular enzymes, and the reaction types are mainly PET hydrolysis, i.e., PET is gradually hydrolyzed under the action of enzymes, then dehydrogenated and oxidized, and finally becomes small molecules to be absorbed and utilized by microorganisms. The degradation process of other types of polymers is similar to PET, only that the enzymes involved in the reaction may be different. However, their specific types and mechanisms of action are not yet clear, and need to be explored in depth in combination with genomics and other means.

3. Influencing Factors of Biodegradation

The microbial biodegradation process consists of three steps of microbial activity: firstly, changes in the physical, chemical and mechanical properties of microplastics inside or on the surface caused by microbial growth; secondly, the transformation of microplastics into oligomers and monomers through microbial activity; and finally microorganisms obtain the necessary energy, carbon and nutrients from the microplastic fragments through assimilation, converting plastic carbon into CO_2, H_2O and biomass [36]. In this process, the biodegradation of microplastics is governed by multiple factors, the most critical of which include the properties of the polymer (polymer chains, crystallinity, chemical structure, type of functional groups, molecular weight, complexity of additives and polymer formulations, etc. [36]), as well as the type of organism and means of pretreatment. For example, the degradation rate of lower density polyethylene chains is generally greater than that of higher density polyethylene chains. Microorganisms are selective for the degradation of different polymers and the enzymes secreted by certain microorganisms may not be applicable to the target plastic. For example, under the same conditions, *Bacillus subtilis* inoculated alone in nutrient solution degraded PS with only 20.0% weight loss compared to 74.59% weight loss of PET; *Pseudomonas aeruginosa* degraded PS with 5.0% weight loss while PET showed no weight loss [37]. In addition, since microorganisms and enzymes play a dominant role in the biodegradation process, some environmental factors affecting their metabolic processes also influence the degradation of microplastics, such as temperature, humidity, and light [38].

4. Research Outlook

Microplastics from all kinds of sources accumulate in the soil and undergo further physical, chemical and biological interactions to migrate and transform and even enter the food chain to threaten human health. Compared with other methods, biodegradation has greater advantages in terms of both efficiency and sustainability, and thus it can be further developed as the most suitable method for microplastic pollution management. However, since the sources and distribution of soil microplastics, as well as their own and load toxicity effects, are not sufficiently studied to provide a guiding reference for soil microplastics biodegradation measures, future research should focus on the following aspects: identifying more soil animals, microorganisms and enzymes with good degradation functions for soil microplastics; genetic engineering should be used to modify the species that better meet the degradation requirements; research on the biodegradation mechanism should be strengthened to understand the synergistic or inhibitory effects among the mixed cultures, so as to provide reference for adjusting the ratio of cultures to achieve the desired effect.

Acknowledgements

This study was supported by the National Natural Science Foundation of China (Grant No. 32001197) and Fundamental Research Funds for the Central Universities (Grant No. 2021ZY14).

References

[1] Thompson CR, Olsen Y, Mitchell RP, Davis A, Rowland SJ, John AWG, McGonigle D, Russell AE. Lost at sea: Where is all the plastic? Science. 2004;304(5672):838-838.

[2] Nizzetto L, Futter M, Langaas S. Are agricultural soils dumps for microplastics of urban origin? Environmental Science & Technology. 2016;50(20):10777-10779.

[3] Rillig MC. Microplastic in terrestrial ecosystems and the soil? Environmental Science & Technology 2012;46(12):6453-6454.

[4] de Souza Machado AA, Kloas W, Zarfl C, Hempel S, Rillig MC. Microplastics as an emerging threat to terrestrial ecosystems. Glob. Chang. Biol. 2018;24(4:1405-1416.

[5] Amobonye A, Bhagwat P, Singh S, Pillai S. Plastic biodegradation: Frontline microbes and their enzymes. The Science of the Total Environment. 2021;759:143536.
[6] Yang Y, Wang J, Xia M. Biodegradation and mineralization of polystyrene by plastic-eating superworms Zophobas atratus. Science of the Total Environment. 2020;708:135233.
[7] Yang J, Yang Y, Wu WM, Zhao J, Jiang L. Evidence of Polyethylene biodegradation by bacterial strains from the guts of plastic-eating waxworms. Environmental Science & Technology. 2014;48(23):13776-13784.
[8] Bombelli P, Howe CJ, Bertocchini F. Polyethylene bio-degradation by caterpillars of the wax moth Galleria mellonella. Current Biology. 2017;27(8):R292-R293.
[9] Dawson AL, Kawaguchi S, King CK, Townsend KA, King R, Huston WM, Bengtson Nash SM. Turning microplastics into nanoplastics through digestive fragmentation by Antarctic krill. Nature Communications. 2018;9(1):1001.
[10] Danso D, Chow J, Streit WR. Plastics: Environmental and biotechnological perspectives on microbial degradation. Applied and Environmental Microbiology. 2019;85(19).
[11] Zhang J, Gao D, Li Q, Zhao Y, Li L, Lin H, Bi Q, Zhao Y. Biodegradation of polyethylene microplastic particles by the fungus Aspergillus flavus from the guts of wax moth Galleria mellonella. Science of the Total Environment. 2020;704:135931.
[12] Brandon AM, Gao SH, Tian R, Ning D, Yang SS, Zhou J, Wu WM, Criddle CS. Biodegradation of polyethylene and plastic mixtures in mealworms (larvae of tenebrio molitor) and effects on the gut microbiome. Environmental Science & Technology. 2018;52(11):6526-6533.
[13] Yang Y, Yang J, Wu WM, Zhao J, Song Y, Gao L, Yang R, Jiang L. Biodegradation and mineralization of polystyrene by plastic-eating mealworms: Part 1. Chemical and physical characterization and isotopic tests. Environmental Science & Technology. 2015;49(20):12080-12086.
[14] Song Y, Qiu R, Hu J, Li X, Zhang X, Chen Y, Wu WM, He D. Biodegradation and disintegration of expanded polystyrene by land snails Achatina fulica. Science of the Total Environment. 2020;746:141289.
[15] Peng BY, Li Y, Fan R, Chen Z, Chen J, Brandon AM, Criddle CS, Zhang Y, Wu WM. Biodegradation of low-density polyethylene and polystyrene in superworms, larvae of Zophobas atratus (Coleoptera: Tenebrionidae): Broad and limited extent depolymerization. Environmental Pollution. 2020;266:115206.
[16] Kundungal H, Gangarapu M, Sarangapani S, Patchaiyappan A, Devipriya SP. Efficient biodegradation of polyethylene (HDPE) waste by the plastic-eating lesser waxworm (Achroia grisella). Environmental Science and Pollution Research. 2019;26(18):18509-18519.
[17] Singh B, Singh K. Microbial degradation of herbicides. Critical Reviews in Microbiology. 2016;42(2):245-61.
[18] Sánchez C. Fungal potential for the degradation of petroleum-based polymers: An overview of macro- and microplastics biodegradation. Biotechnology Advances. 2020;40:107501.
[19] Zhang X, Li Y, Ouyang D, Lei J, Tan Q, Xie L, Li Z, Liu T, Xiao Y, Farooq TH, Wu X, Chen L, Yan W. Systematical review of interactions between microplastics and microorganisms in the soil environment. Journal of Hazardous Materials. 2021;418:126288.
[20] Ru J, Huo Y, Yang Y. Microbial degradation and valorization of plastic wastes. Frontiers in Microbiology. 2020;11:442.
[21] Li L, Luo Y, Li R, Zhou Q, Peijnenburg WJGM, Yin N, Yang J, Tu C, Zhang Y. Effective uptake of submicrometre plastics by crop plants via a crack-entry mode. Nature Sustainability. 2020;3(11):929-937.
[22] Du H, Xie Y, Wang J. Microplastic degradation methods and corresponding degradation mechanism: Research status and future perspectives. Journal of Hazardous Materials. 2021;418:126377.
[23] Auta HS, Emenike CU, Fauziah SH. Screening of bacillus strains isolated from mangrove ecosystems in Peninsular Malaysia for microplastic degradation. Environmental Pollution. 2017;231:1552-1559.
[24] Yoshida S, Hiraga K, Takehana T, Taniguchi I, Yamaji H, Maeda Y, Toyohara K, Miyamoto K, Kimura Y, Oda K. A bacterium that degrades and assimilates poly (ethylene terephthalate). Science. 2016;351(6278):1196-1199.
[25] Paço K, Duarte JP, da Costa PSM, Santos R, Pereira ME, Pereira AC, Freitas AC, Duarte TAP. Rocha-Santos, biodegradation of polyethylene microplastics by the marine fungus Zalerion maritimum. Science of the Total Environment. 2017;586:10-15.
[26] Vimala PP, Mathew L. Biodegradation of polyethylene using bacillus subtilis. Procedia Technology. 2016;24:232-239.
[27] Harshvardhan K, Jha B. Biodegradation of low-density polyethylene by marine bacteria from pelagic waters, Arabian Sea, India. Marine Pollution Bulletin. 2013;77(1):100-106.
[28] Shah Z, Krumholz L, Aktas DF, Hasan F, Khattak M, Shah AA. Degradation of polyester polyurethane by a newly isolated soil bacterium, Bacillus subtilis strain MZA-75. Biodegradation. 2013;24(6):865-77.

[29] Sangale MK, Shahnawaz M, Ade AB. Potential of fungi isolated from the dumping sites mangrove rhizosphere soil to degrade polythene. Scientific Reports. 2019;9(1):5390.
[30] Álvarez-Barragán J, Domínguez-Malfavón L, Vargas-Suárez M, González-Hernández R, Aguilar-Osorio G, Loza-Tavera H. Biodegradative activities of selected environmental fungi on a polyester polyurethane varnish and polyether polyurethane foams. Applied and Environmental Microbiology. 2016;82(17):5225-5235.
[31] Yuan J, Ma J, Sun Y, Zhou T, Zhao Y, Yu F. Microbial degradation and other environmental aspects of microplastics/plastics. Science of the Total Environment. 2020;715:136968.
[32] Roohi, Bano K, Kuddus M, Zaheer MR, Zia QF, Khan MF, Ashraf GM, Gupta A, Aliev G. Microbial enzymatic degradation of biodegradable plastics. Current Pharmaceutical Biotechnology. 2017;18(5): 429-440.
[33] Orr IG, Hadar Y, Sivan A. Colonization, biofilm formation and biodegradation of polyethylene by a strain of Rhodococcus ruber. Applied Microbiology and Biotechnology. 2004;65(1):97-104.
[34] Howard GT. Biodegradation of polyurethane: A review. International Biodeterioration & Biodegradation. 2002;49(4):245-252.
[35] Ahmed T, Shahid M, Azeem F, Rasul I, Shah AA, Noman M, Hameed A, Manzoor N, Manzoor I, Muhammad S. Biodegradation of plastics: current scenario and future prospects for environmental safety. Environmental Science and Pollution Research. 2018;25(8):7287-7298.
[36] Emadian SM, Onay TT, Demirel B. Biodegradation of bioplastics in natural environments. Waste Management. 2017;59:526-536.
[37] Asmita K, Shubhamsingh T, Tejashree S. Isolation of plastic degrading micro-organisms from soil samples collected at various locations in Mumbai, India. International Research Journal of Environment Sciences. 2015;4(3):77-85.
[38] Sharma S, Basu S, Shetti NP, Nadagouda MN, Aminabhavi TM. Microplastics in the environment: Occurrence, perils, and eradication. Chemical Engineering Journal. 2021;408:127317.