REVIEW

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NABANITA GHOSH[1*], PRIYANKA BHOWMIK[2], PRANAB GIRI[3] AND ARNAB GANGULI[1]

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NABANITA GHOSH[1*], PRIYANKA BHOWMIK[2], PRANAB GIRI[3] AND ARNAB GANGULI[1]

[1]Department of Microbiology, Techno India University, EM-4, EM Block, Sector V, Bidhannagar, Kolkata- 700091, West Bengal
[2]Department of Biological Sciences, Adamas University, P.O. Jagannathpur, Barbaria, Pin-700126
[3]Department of Botany, Dum Dum Motijheel College, Kolkata-700074

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The use of plastics is extensive in today's world. Due to their inherent resistance to degradation, bioaccumulation of plastics in the environment is a major public health issue. The filling of landfill sites and deficiency of degradability have led to land pollution and rising water problems. Various methods are being considered to battle against this major environmental pollutant. Amongst these, the most ecologically acceptable is the use of microorganisms to degrade plastic materials. Bacteria and fungi are important candidates for this role. General studies on the biodegradation of plastics have been approved in order to overcome the environmental difficulties connected with synthetic plastic waste. This review is particularly concerned with the degradation of plastics by soil fungi. This study aims to provide a current comprehensive detail about fungi-mediated degradation of plastics.

Key words: Biodegradation, fungi, fungal enzyme, hydrophobin, microbial degradation, plastic, bioremediation

INTRODUCTION

The modern industrial era has led to the accumulation and release of millions of tons of plastic wastes into aqueous and terrestrial ecosystems, shown in Fig.1. This gargantuan amount of plastic is causing harm to the flora and fauna and to the environment as a whole(Barnes *et al.* 2009). Unfortunately, the best-known method of plastic destruction is to expose it to UV light along with mechanical disruption, land filling and incineration which comes with accost of environmental pollution (Danso *et al*. 2019). Interdisciplinary research and advanced fungal approaches for different plastic waste biodegradation can help to decrease the bad effects of plastic waste pollution in the environment.

This review summarizes the main findings on the fungal degradation of plastics to date. We will try to address currently known fungi to degrade plastic materials, their advantages and drawbacks, their enzymes and the mechanisms of these enzymes.

*Correspondence:naba.9474@gmail.com

Plastic ropes and netting are causing the entanglement of at least 344 species to date(Kühn, *et al.* 2015).Plastic chemicals can undergo significant biomagnifications and are linked to various health problems like developmental impairment, (neurological impairment, growth abnormalities, various hormonal imbalances), cancer, diabetes etc.(Liao and Kannan 2011; Zhang *et al*. 2021).

Plastic disposal methods

Currently, three methods are being used for plastic disposal: burying in landfills, incineration, and recycling(Braun, 2015).

Landfill

Plastic debris in landfills is also a source of a number of secondary pollutants.

Incineration

Polycyclic Aromatic Hydrocarbons (PAHs), Polychlorinated biphenyls (PCBs), heavy metals, toxic carbon- and oxygen-based free radicals, greenhouse gases, especially carbon dioxide, are all produced and released when plastics are incinerated(Fruergaard*et al*. 2010; Luo*et al.* 2005; Noubactep, 2009).

Recycling

Recycling is the most ecologically acceptable plastic disposal methods(Moulton and Kosslyn 2009).

Biodegradation of plastic

The main problem associated with plastic degradation is its bio stability and inherent resistance to degradation. Microbes utilize these chemicals polymers as their nutrients(Abdel-Motaal*et al*. 2014; Muhamad *et al.* 2015). The microbial degradation of plastic polymers is initiated by the microbial enzymes which act in a sequential manner. The steps are i) bio-deterioration (altering the chemical and physical properties of the polymer), ii) bio-fragmentation (polymer breakdown in a simpler form via enzymatic cleavage) and iii) assimilation (uptake of molecules by microo-rganisms) and iv) mineralization (production of oxidized metabolites (CO_2, CH_4, H_2O) after degradation.

Plastic degradation by fungi

Fungi are metabolically diverse. Therefore, they are potential candidate for degradation of plastics (Zeghal *et al.* 2021). They are reported to degrade persistent organic pollutants (POPs) (Singleton, 2001), polycyclic aromatic hydrocarbons (PAHs) (Cerniglia and Sutherland, 2001), BTEX compounds (Buswell, 2001) and pesticides (Pinto *et al.* 2012).

Degradation of polyethylene (PE)

Polyethylene (PE) is the most abundant non degradable plastic waste. PE has an extensive C-C backbone and therefore very resistant to hydrolysis.A study found that *Fusarium oxysporum, Fusarium falciforme* and *Purpureocillum lilacinum* can grow very well on PE films and can use PE as their sole source of carbon (Spina *et al*. 2021).PE degradation occurs initially by changes of the superficial properties by abiotic (eg.UV) or biotically by enzymatic activities. Then it is broken down into smaller units which are then utilised by the fungus for its metabolism and converted into CO_2, methane and water.Other reports showed that *Cladosporium cladosporioides* was able to degrade PE.N. asteroids. *Mortierella alpina* and *Gordonia* were shown to form vigorous biofilm on the PE surface by extensive mycelium formation (Bonhomme *et al*. 2003).

Degradation of polyethylene terephthalate (PET)

Aromatic terephthalate ring in PET is the main reason of its lack of biodegradability. Several fungi have been reported to degrade PET. *Penicillium restrictum* and *P. simplicissimum* were found to produce lipase enzymes and were found to reduce PET biomass by about 3%(Sim *et al.* 2015).*Aspergillus oryzae, Aspergillus nidulans* (Adýgüzel and Tunçer 2017), *Penicillium citrinum* (Liebminger *et al.* 2007), *Humicola insolens* (Araújo *et al.* 2007), *Fusarium solani* (Alisch-Mark *et al.* 2006), *Fusarium solanipisi* (Vertommen *et al.* 2005), and *Fusarium oxysporum* (Dimarogona *et al.* 2015),etc have shown to produce cutinase enzyme which can degrade PET. *Penicillium citrinum* polyesterase was found to be effective against PET (Liebminger *et al.* 2007).

Degradation of polyurathane (PU)

Polyurathane (PU) is vulnerable to biodegradation. *Nectria gliocladioides, Penicillium ochrochloron* and *Geomyces pannorum* were found to degrade PU by forming biofilms(Brunner *et al.* 2018).In another study, *Cladosporium cladosporioides* complex, including *C. pseudocladosporioides, C. tenuis-simum, C. asperulatum*, and *C. monteci-llanum*, and two other fungi namely *Aspergillus fumigatus* and *Penicillium chrysogenum* were found to degrade polyether polyurethane foams (Brunner *et al.* 2018).*Geomyces pannorum* and a *Phoma* sp. were also reported to cause PU degradation.

Degradation of poly hydroxy butyrate (PHB)

PHB is a thermoplastic produced by certain bacteria as inclusion bodies. Fungi from Deuteromycetes resembling species of *Penicllium* and *Aspergillus*, Ascomyecetes are reported to degrade PHB (Lee *et al*. 2005).Lichens which are composite organisms are also reported to degrade PHB (Lee *et al.* 2005).

Degradation of poly lactic acid (PLA)

Polylactic acid is mainly used in waste disposal bags and containers. *Thermomyces lanuginosus* and *A.*

fumigatus were reported to cause significant degradation to PLA(Karamanlioglu *et al.* 2014). In another study white rot fungi *Phanerochaete chrysosporium* has been reported to biodegrade PLA. Significant reduction in the average molecular weight of the PLA sample was also observed by this fungal action (Stoleru *et al*. 2017).Other predominant fungi able to digest PLA are *Fusarium moniliforme, Penicillium roqueforti*, *Tritirachium album, Aspergillus fumigatus*, and *Thermomyces laniginosa* (Grishchuk *et al.* 2011).

Degradation of poly vinyl alcohol (PVA)

PVA is a water-soluble polymer and widely been used in adhesives, films, paper coating etc. *Galactomyces geotrichum, Trichosporon laibachii, Fimetariella rabenhorsti, Fusarium oxysporum* and *G. geotrichum* were reported to utilise PVA as sole source of carbon and can degrade it.In another study, rhizosphere-associated fungus *Penicillium brevicompactum* OVR-5 had been reported to degrade PVA(Mohamed *et al.* 2022).A novel fungus *Eutypella* had been demonstrated to degrade PVA (Deng *et al*. 2019).

Degradation of poly vinyl chloride (PVC)

PVC is recalcitrant and that's why a serious environmental threat. It has been recently reported that members of order Xylariales, class Ascomycetaecan utilise PVC as sole source of carbon. *Penicillium* and *Aspergillus* also possess this activity.Another study had shown that soil fungi like *Phanerochaete chrysosporium* PV1, *Lentinus tigrinus* PV2, *Aspergillus niger* PV3, and *Aspergillus sydowii* PV4 can biodegrade PVC(Ali *et al.* 2014).

Degradation of polycaprolactone

Aspergillus, Fusarium, Penicillium and *Parengy odontium* had been observed to degrade poly caprolactone.

Enzymatic system of fungi

Fungi have some intracellular and extracellular enzymes which give them the ability to degrade polymers (Table1). The intracellular enzymatic system helps to detoxify and fungal adaptation (Jeon and Kim, 2016). This system is formed by peroxidases and transferases in the Cytochrome P450 family (CYP), which is involved in the oxidation and conjugation process(Schwartz *et al.* 2018).The extracellular enzymatic system is formed by the hydrolytic system and the unspecific oxidative system. The hydrolytic system produces hydrolases that help in polysaccharide degradation and the oxidative system helps to complex structure breakdown like lignin degradation (Sánchez 2009).

Fungal cutinase in plastic degradation

Cutinaseis a member of the serine esterase family hydrolases. Fungal cutinases are proven to be an excellent enzyme for the biodegradation of plastic polymers e.g. PET. cutinases from *Aspergillus oryzae, Aspergillus nidulans* (Bermúdez-García *et al*. 2017),*Penicillium citrinum* (Liebminger *et al.* 2007), *Humicola insolens* (Ronkvist *et al*. 2009), *Fusarium solani* (Alisch-Mark *et al*. 2006; Tournier *et al.* 2020), *Fusarium solani pisi* and *Fusarium oxysporum* were reported to be PET degraders.Hu *et al*. (2016) had shown that *Fusarium solani* cutinase when over expressed in *Pichia pastoris* was able to degrade PBS plastics (Hu *et al*. 2016).Cutinases can degrade other types of plastic as well (Masaki *et al.* 2005). *Penicillium citrinum* (Liebminger *et al.* 2007), *Pichia pastoris* (Shi*et al*. 2019). *Aspergillus oryzae*, *Humicola insolens* were also found to produce cutinase enzymes involved in plastic polymer degradation.

Fungal lipase in plastic degradation

Lipases are esterases that catalyse lipids. *Rhizopus delemer, Candida Antarctica* (Vertommen *et al.* 2005) *Thermomyces lanuginosus* (Eberl*et al.* 2009),*Candida rugosa* were reported to degrade plastic. These fungi can degrade poly (butylene succinate-co-hexamethylene succinate) co-polymer (Pereira *et al.* 2001). Lipase from *Rhizopus delemer* was able to degrade polyurethane (Tokiwa *et al.* 2009).

Fungal esterases in plastic degradation

Esterase, derived from *Xepiculopsis graminea*, and *Penicillium griseofulvum* were reported to degrade polyurethan. Fungal esterases also degrade polycaprolactone polymers (Ganesh *et al.*2017). *Monascus ruber, M. sanguineus* have been shown to degrade polyurethane and are prolific esterase producers (El-Morsy*et al*. 2017).

Fungal laccases in plastic degradation

Laccase is another enzyme which showed potential to degrade plastics (Mehra *et al*. 2018). These are multi copper enzymes. Marine fungal community also produce these enzymes. e.g. *Peniophora* sp. showed transcription of laccase enzymes (Otero *et al.* 2017). The plastic degradation by these enzymes is multi step pathways. Laccases transfers electrons from organic substrates to molecular oxygen. *Cochliobolus* sp., isolated from plastic dumped soils was found to degrade polyvinyl chloride (PVC).

Fungal peroxidases in plastic degradation

Peroxidases are enzymes which are involved in oxidation reduction reactions.Fungal peroxidises fall under- 1) lignin peroxidases (LiP), 2) manganese peroxidases (MnP), and 3) versatile peroxidases (VP) (Hofrichter and Ullrich 2006). Reports of effective polymer degradation by fungal peroxidises showed degradation of polyethylene e.g.*Phanerochaete chrysosporium*, and *Trametes versicolor* peroxidases. *Fusarium graminearum* showed polyethylene degradation by producing peroxidase (Ganesh *et al.* 2017).*Aspergillus flavus, Aspergillus niger, Fusarium graminearum* were also reported to produce several peroxidases, which can degrade various biopolymers. Lignin peroxides are produced by *Aspergillus niger*and *Aspergillus flavus.*

Fungal depolymerase in plastic degradation

Polyhydroxybutyrate (PHB) can be degraded by PHB depolymerise isolated from *Aspergillus fumigates* (Jung*et al*. 2018) and *Fusarium solani* (Shivakumar, 2013).These enzymes degrade the ester bonds in PHB and break them into smaller mono (3 hydroxybutyrate) units and finally into water and carbon dioxide (Ojha *et al.* 2017). In another study, *Penicillium pinophilum* (ATCC 9644) had been shown to produce extracellular depolymerise(Panagiotidou *et al*. 2014).Most of these enzymes are most active in alkaline pH while depolymerases of *P. pickettii* and of *P. funiculosum* have pH optima at 5.5 or 6.

Hydrophobins produced by fungi: role in plastic degradation

Hydrophobins are small molecular weight amphipathic proteins secreted by fungi and show an idiosyncratic conservation of eight cysteine residues.It had been shown that degradation of polyester polybutylene succinate-coadipate (PBSA) by *Aspergillus oryzae* was enhanced by the pre incubation with the hydrophobin protein RolA (Terauchi *et al*. 2020).The hydrophobin was actually absorbed to the PBSA surface and then recruit the cutinase which actually degrade the polyester. In another study, *Humicola insolens*cutinase (HiC) in presence of hydrophobin HFB4 showed more than 2.5-times higher amounts of PET degrading activity compared to HiC alone. Fusion of *Trichoderma* hydrophobins to cutinase from *Thermobifida cellulosilytica* resulted in the increase in PET hydrolysis (Ribitsch *et al*. 2015; Ribitsch and Guebitz, 2021).

Factors affecting biodegradation

Biodegradation of plastic is controlled by several factors which include chemical structure of the polymers, surface properties, molecular weight, miscibility of polymer's constituent and the phase structure of plastic polymers and presence of hydrolysed and oxidized compounds.

Limitations of studying fungal communities

Fungal potential as a plastic degrader is less studied and less understood because of different factors that influence molecular studies of fungi. Nucleic acid extraction and manipulation of fungi is difficult as fungal cell walls are made up of chitin which makes them difficult to lyse (Park *et al*. 2018; Wu*et al*. 2005).Another problem relies in the less availability of information about the marker genes e.g. 18s rRNA. Around 50% of the fungal species don't have any DNA sequence information in public

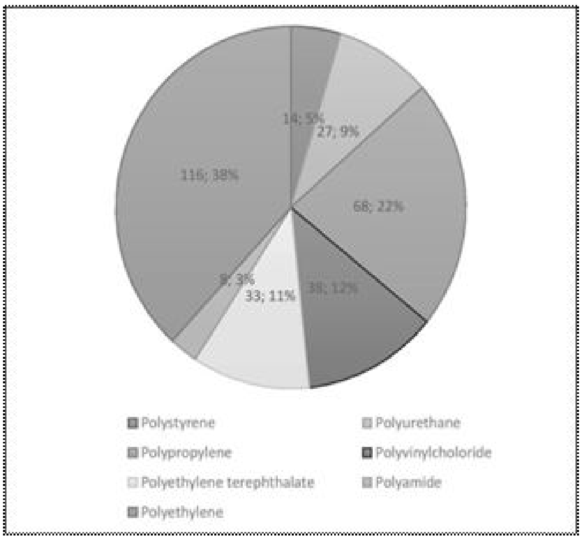

Fig. 1 : Global production of polymers in 2016

Table 1: Fungal strains depicting the biodegradation of plastic

Fungi	Enzyme	Plastics	Reference
Aspergillus fumigatus	Polyhydroxybutyrate depolymerase	PHB, PBS, PES	Jung *et al*. 2018 Liu *et al*. 2019
	Bifunctional lipase-cutinase	PCL	
Fusarium solani	Cutinase	PBS PCL	Hu *et al*. 2016 Shimao, 2001
Candida rugosa	Lipase	PLA/cellulose nanocrystal composites	Hegyesi *et al*.2019
Tritirachium album	Proteinase K	PBS	Hu *et al*. 2018
Ideonella sakaiensis	PETase and MHETase	PET	Knott *et al*. 2020
Thermobifida cellulosilytica	Proteinase K, Protease, Esterase and Lipase	PLA	Gigli *et al*. 2019
Thermomyces insolens	Cutinase	PET	Zimmermann and Billig, 2010

databases till date(Xu 2016).Culturing of fungal sample is time consuming. Moreover as different developmental stage of a single fungal species differs, it further complicates the identification process(Horton *et al*. 2021; Xu 2016).

CONCLUSION

Biological degradation of plastics by microbes is an attractive option and ensures efficient disposal of plastic wastes.Finding plastic-active fungal enzymes and implementing them in the degradation of plastic waste and production of true biopolymers would significantly reduce global burden of plasticwaste.

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