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An overview on role of fungi in systematic plastic degradation

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ABSTRACT

Plastics have largely supplanted natural materials such as paper, wood, and metal due to its cost effectiveness, high flexibility, durability, non-degradability, and fire resistance. Plastic's increased production, global distribution, and long-lasting nature eventually lead to environmental accumulation, posing serious concerns to the environment and biotic health. As a result, in this plastic age, developing appropriate metrics for plastic environmental cleanup could be a pressing concern. Thus, the capacity of biological systems to break down polymers has recently received more attention. It has been shown that different fungal strains consume these plastic polymers as their only source of carbon, converting them into eco-friendly carbon compounds. Various fungal strains, including *Aspergillus nomius, Trichoderma viride, Cephalosporium* sp., *Stagonosporopsis citrulli, Colletotrichum fructicola, Diaporthe italiana,* and others, have been found to successfully and efficiently degrade various plastic polymers. Mechanism of biodegradation includes following steps, that is, biodeterioration, biofragmentation, assimilation, and mineralization. This review mainly focuses on the numerous fungal strains isolated from various sites which engaged in plastic biodegradation, the biodegradation mechanism, and the various assessment methods used to analyze the extent of biodegradation process.

1. INTRODUCTION

Plastics are man-made, non-biodegradable composites which are of substantially petrochemical origin [1]. They come from plants like corn and sugarcane, as well as from natural gas and oil. About 4% of the petroleum produced worldwide is utilized to generate plastic, and an additional 4% is required to fuel the processes used to make plastic [2]. They are simply made up of hydrogen and carbon with some other organic and inorganic materials [3]. Plastics have become universal due to its dual nature as it is a widely used material as well as considered as an environmental contaminant [4]. In the past few decades plastic materials have covered each and every sector of human need. It has replaced the other material such as glass, wood, and metal that were used in varied applications, due to its distinct properties that have created the way for its use in enormous sectors [5]. Low-density polyethylene (LDPE), high-density polyethylene (HDPE), polyethylene terephthalate (PET), polypropylene (PP), polystyrene (PS), polyvinyl chloride (PVC), polycarbonate (PC), and polyurethane (PU) are the most widely used polymers The commonly used plastics are LDPE, HDPE, PET, PP, PS, PVC, PC, PU, etc. [6,7] [Figure 1]. Widespread applications of plastic leads to large scale production which is creating an issue of their disposal and management [4]. Over the course of at least 500 years, about 90% of all plastic created worldwide persists in the environment [8]. The health of the biotic community in both terrestrial and aquatic habitats could be severely threatened by plastic [9-11]. Both the incineration of plastic waste and the dumping of it in landfills produce significant amounts of CO_2 and contribute to global warming [12]. Air, water, and soil pollution result from the environmental implications of dumping so much plastic into the environment [8]. In attempts to develop novel strategies for managing plastic waste, significant research expenditure has explored whether bacteria might utilize the commonly contaminated polymers. By doing so, they could provide a sustainable and eco-friendly alternative to the current extreme usage of plastic [4]. Biodegradation is the most beneficial approach for plastic degradation compared to other methods since it is non-polluting in nature. Multiple environmental parameters and numerous microbial strains are involved in biodegradation [13]. According to reports, the most effective technique to minimize plastic trash in an environmentally acceptable manner is through bioremediation, which uses biological agents such as bacteria, fungi, and algae [14]. Fungi are a varied group of eukaryotic organisms that can act as saprobes, symbionts, and parasites in nearly all aerobic and some anaerobic conditions [15].

The significance of fungus in the biodegradation of plastic is considerable. The presence of pro-oxidant ions and the secretion of enzymes that aid in degradation, such as proteases, cutinases, and lipases, has been reported to cause effective degradation by fungi. By generating functional groups through oxidation or hydrolysis by

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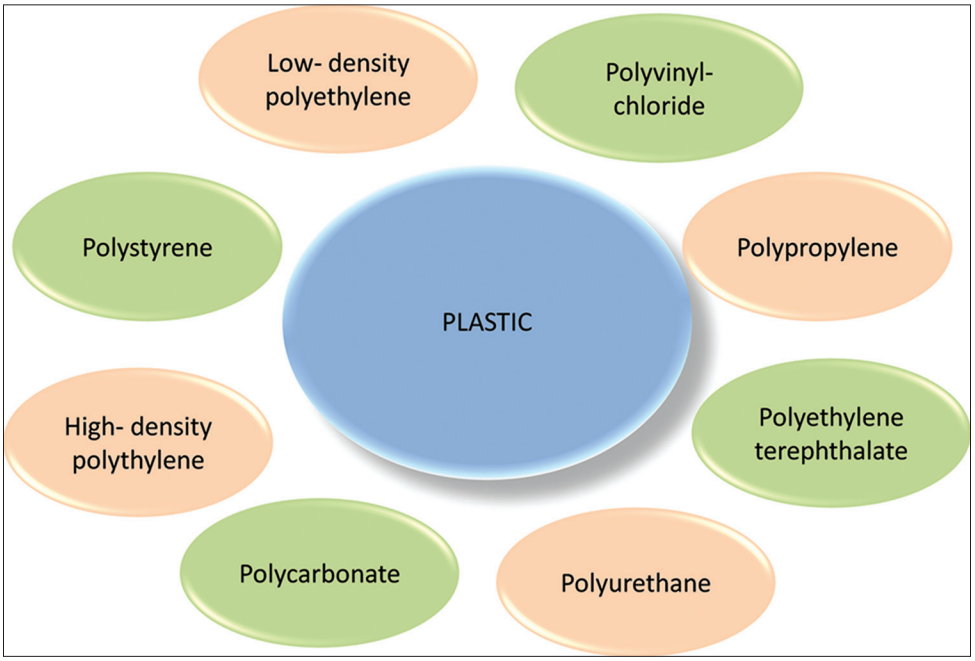


Figure 1: Types of plastic.

the action of enzymes, higher molecular weight compounds can be converted into lower one by making polymers more hydrophilic [13]. Several fungal strains including *Aspergillus clavatus, Trichoderma viride, Aspergillus nomius, Penicillium* sp. have been known to potentially degrading this alarming plastic waste and providing a way to get rid of this plastic waste management issue in an eco-friendly way [16-18]. There is a need to create innovative solutions to both reduce and degrade this waste using a green approach, as the generation of plastic garbage is a problem that affects the entire globe. This review article gives a systematic view at the various fungal strains involved in plastic biodegradation, process of biodegradation, and various assessment techniques involved.

2. BIODEGRADATION

Plastic polymers are a big threat to the entire world and are not biodegradable. It would take decades for these plastics to break down. Biodegradation is the most efficient and ideal method for plastic breakdown due to its non-polluting mechanism, environmental friendliness, and economic viability [13]. Microbes are the intermediaries in a challenging physicochemical process that breaks down polymers into smaller components [19,20]. Complex organic molecules can be biochemically broken down, assimilated, and metabolized by microorganisms, specifically fungi [21,22].

Biodeterioration, biofragmentation, assimilation, and mineralization are a few of the biochemical degradative routes for plastic biodegradation. These procedures all rely on different enzyme functions and bond cleavage [14,23]. Biodeterioration is the chemical and physical activity of microbes that results in a plastic polymer's surface degradation and alteration of its mechanical, physical, and chemical properties [24]. Adhesion and colonization of microbes on the surface of the polymer initiates this first step, the only goal of which is to reduce the resistance and durability of plastic materials. It is frequently required to add hydrophilic functional groups to plastic surfaces to promote microbe adherence because plastics are naturally hydrophobic in nature [25]. The polymers are used by microbes as their main source of carbon as they attach to the polymer's surface and keep multiplying. Next is the depolymerization process known as "biofragmentation," extracellular enzymes and bacterially generated free radicals catalyze the breakdown of biodegraded polymers into smaller pieces [26]. Next step is assimilation in which the biofragmented smaller molecular weight compounds are then transported into the microbial cytoplasm [27,28]. The last step is mineralization, which involves the successful delivery of these plastic derivatives into cells and a sequence of enzymatic reactions that cause them to completely decompose into oxidized metabolites including CO_2, N_2, CH_4, and H_2O [29]. Numerous enzyme activities, including peroxidases, lipases, esterase, cutinase, and laccase, are necessary for the complete mineralization process [30].

3. FUNGAL STRAINS INVOLVED IN BIODEGRADATION

The world's fungus species range from 2.2 to 3.8 million from harmless free-living bacteria to dangerous diseases that may survive in a variety of host and environmental niches such soil, water, plants, and animals [31]. Fungi vary in their morphology and can be unicellular, filamentous or dimorphic [32]. They can exist independently or in mutualistic symbiotic relationships or as parasitic pathogens of diverse plants and animals, including humans [33,34]. Numerous fungi inhabit terrestrial, freshwater, and aquatic habitats [35-37].

Numerous fungi species have been found to be able to degrade a wide range of plastic polymers due to their potential to use these synthetic polymers as their primary or only source of energy. In this context, it has been demonstrated that a vast variety of fungi, representing different classes, ecologies, and morphologies, are capable of degrading plastics. Due to its enormous advantage over chemical and physical degradation approaches, the biodegradation of these man-made compounds by microorganisms seems to be one of the important techniques to control the problem of

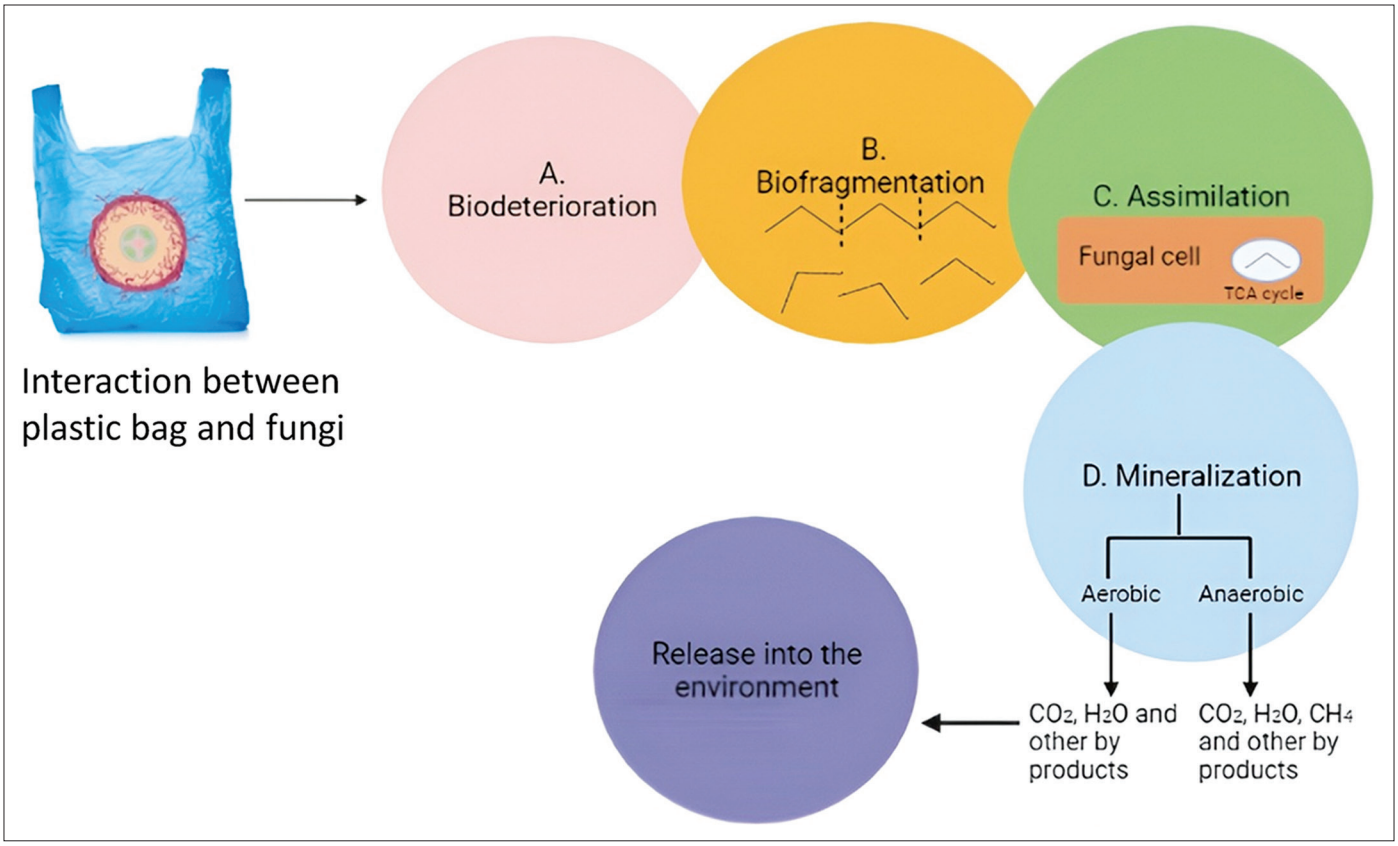


Figure 2: System overview of biodegradation of plastic by fungal community.

plastic waste [38]. Biological degradation is thought to be a more effective and strong solution to this global issue. Biodegradation involves many kinds of plastic degrading microorganisms [39,40]. Many chemicals are transformed into simpler compounds by microorganisms through biochemical processes. An indicator of the biodegradation of plastic polymers is a change in the physical properties of the polymers, such as a reduction in molecular weight, a loss of mechanical strength, or a change in the surface properties of the plastic [29] [Figure 2].

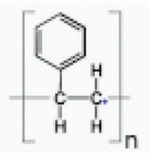
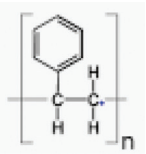
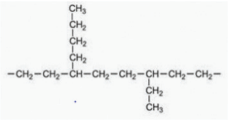
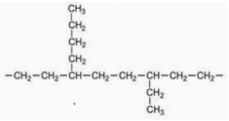
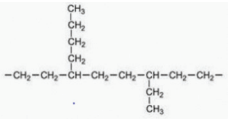
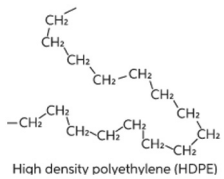
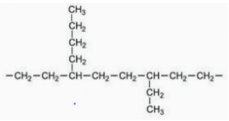
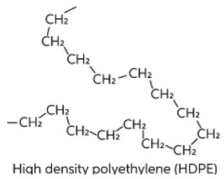
A. clavatus isolated from landfill soil has been found to degrade LDPE [16]. There are numerous *Aspergillus* species, including *Aspergillus terreus, Aspergillus sydowii, Aspergillus tubingensis, Aspergillus fumigates* isolated from Mangrove dumpsite. The coastal environment of the Gulf of Mannar and seawater is known to degrade polyethylene effectively [18,41,42]. Various fungal strains have been discovered so far and showing degradation of different types of plastic in an accomplished way. Various fungal strains isolated from different habitats and degrading different types of plastics have been reported worldwide [Table 1].

4. ASSESSMENT TECHNIQUES

To measure the degree of plastic biodegradation, a variety of assessment techniques have been used. Previously, the gravimetric assessment of polymer weight/mass loss over time when exposed to cultured microbes was the most widely used method for assessing plastic biodegradation [49,50]. Although nowadays, various new assessment methodologies are currently being employed to assess the level of plastic biodegradation. Scanning electron microscopy (SEM), which creates a surface image by illuminating a surface with a high-intensity electron beam and scanning across it. High magnification and hence good resolution are provided by SEM at the nanoscale range. SEM observations are utilized to study and evaluate the colonization of plastic films or particles by microorganisms and at the same time visualizing fractures, trenches, and deformations on the plastic surface [51,52], Which can indicate if the polymer is damaged. SEM has been used in a number of studies to examine fungi on polymers. SEM is a fast technique for observing surface attachment and morphological microstructures [38]. Atomic force microscopy (AFM) is another technique that can be used to identify surface alteration of polymers during degradation [53]. Using this method, topographical changes at the polymer surface, such as the emergence of pits and crevices, the adhesion of microbes to the polymer surface, and an increase in surface roughness, can be directly observed [54].

Fourier-transform infrared spectroscopy (FTIR) is used to identify functional groups contained in polymer films. FTIR spectrum detects and semi-quantifies changes in initial polymer arrangement, such as the addition of carbonyl groups during oxidation [55,56]. The crystallinity of polymer films was measured using X-ray diffraction examination. It was carried out using an X-ray diffractometer. By examining the diffraction patterns produced by polymer films, the structure of such films was discovered [57]. Due to their high water content, hydrophilic surfaces have higher surface energies and yield smaller contact angles with water. As a result, polar functional groups that develop in polymers as a result of environmental degradation cause the contact angle to decrease. The rate of disintegration is further accelerated by increased hydrophilicity because it encourages

Table 1: Various fungal strains showing plastic biodegradation.

Substrate	Sample location	Fungal strains	Weight loss (%)	Assessment techniques	References
PSc 	NCIM	*Mucor* sp.	1.81±0.13	Weight reduction measurement, FTIR, SEM, TGA–DTG, GC-MS and GPC	[43]
PS 	NCIM	*Cephalosporium* sp.	2.17±0.16	Weight reduction measurement, FTIR, SEM, TGA–DTG, GC-MS and GPC	[43]
LDPE 	Sewage disposal ground	*A. nomius*	4.9	Determination of Dry Weight of Residual LDPE, AFM, GC-MS and FTIR	[44]
LDPE 	Landfill soil	*T. viride*	5.13	Weight loss and tensile strength analyses	[17]
LDPE 	Sewage disposal ground	*Streptomyces* sp.,	5.2	Determination of dry weight of residual LDPE, AFM, GC-MS and FTIR	[44]
HDPE 	Marine environmental site dumped with plastic waste	*A. tubingensis*	6.02±0.2	dry weight of the residual HDPE, FTIR analysis, fungal cell surface hydrophobicity and SEM analysis	[42]
LDPE 	Landfill soil	*A. nomius*	6.63	Weight loss and tensile strength analyses	[17]
HDPE 	Marine environmental site dumped with plastic waste	*A. flavus*	8.51±0.1	Dry weight of the residual HDPE, FTIR analysis, fungal cell surface hydrophobicity and SEM analysis	[42]

(*Contd*...)

Table 1: (*Continued*).

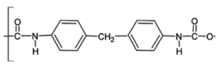
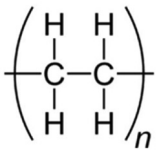
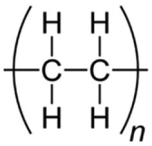
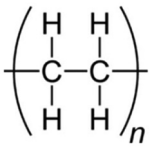
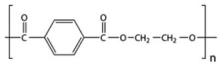
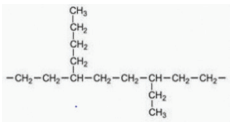
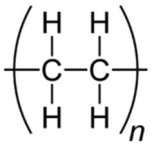
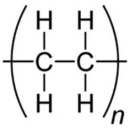
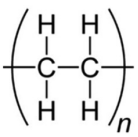
Substrate	Sample location	Fungal strains	Weight loss (%)	Assessment techniques	References
PU 	Solid waste-dumping site	*Aspergillus* sp.	15–20	SEM, FTIR and DSC measurement	[45]
PE 	Seawater	*A. niger*	19.5	Weight loss, tensile Strength, SEM and FTIR	[18]
PE 	Seawater	*A. fumigatus*	20.5	Weight loss, tensile Strength, SEM and FTIR	[18]
PE 	Seawater	*A. terreus*	21.8	Weight loss, tensile Strength, SEM and FTIR	[18]
PET 	Solid waste litter site	*Aspergillus* sp.	22	Weight loss, FTIR, SEM and XRD	[46]
LDPE 	Landfill soil	*A. clavatus*	35	Weight loss, CO_2 evolution measured by Strum test, infrared spectra and morphological changes measured by SEM and AFM analysis	[16]
PE 	Mangrove Dumpsite	*A. sydowii*	37.94	Weight loss, tensile strength, SEM and FTIR	[41]
PE 	Dumpsite soil	*T. harzianum*	40.	SEM, FTIR, NMR analyses and enzymatic assay	[47]
PE 	Mangrove Dumpsite	*A. terreus*	41.82	Weight loss, tensile Strength, SEM and FTIR	[41]

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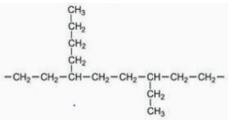
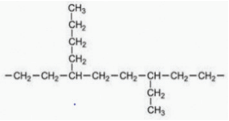
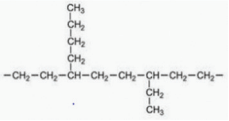
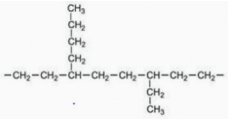
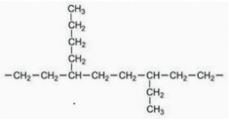
Table 1: (*Continued*).

Substrate	Sample location	Fungal strains	Weight loss (%)	Assessment techniques	References
LDPE 	Sea water samples	*Penicillium* sp.	43.4	Weight reduction and SEM	[18]
LDPE 	Culture Collection of the Institute of Excellence in Fungal Research	*D. italiana*	43.90	Weight loss, tensile strength, FTIR, SEM and GC-MS	[48]
LDPE 	Culture Collection of the Institute of Excellence in Fungal Research	*S. citrulli*	45.12	Weight loss, tensile strength, FTIR, SEM and GC-MS	[48]
LDPE 	Culture Collection of the Institute of Excellence in Fungal Research	*T. jaczewskii*	46.34	Weight loss, tensile strength, FTIR, SEM and GC-MS	[48]
LDPE 	Culture Collection of the Institute of Excellence in Fungal Research	*C. fructicola*	48.78	Weight loss, tensile strength, FTIR, SEM and GC-MS	[48]

PS: Polystyrene, PE: Polyethylene, LDPE: Low-density PE, PET: PE terephthalate, HDPE: High-density PE, PU: Polyurethane, NCIM: National Collection of Industrial Microorganism, FTIR: Fourier-transform infrared spectroscopy, SEM: Scanning electron microscopy, GC-MS: Gas chromatography mass spectrometry, AFM: Atomic force microscopy, DSC: Differential scanning calorimetry, GPC: Gel-permeation chromatography, *A. nomius*: *Aspergillus nomius*, *T. viride*: *Trichoderma viride*, *A. tubingensis*: *Aspergillus tubingensis*, *A. flavus*: *Aspergillus flavus*, *A. niger*: *Aspergillus niger*, *A. fumigatus*: *Aspergillus fumigatus*, *A. terreus*: *Aspergillus terreus*, *A. clavatus*: *Aspergillus clavatus*, *A. sydowii*: *Aspergillus sydowii*, *T. harzianum*: *Trichoderma harzianum*, *D. italiana*: *Diaporthe italiana*, *S. citrulli*: *Stagonosporopsis citrulli*, *T. jaczewskii*: *Thyrostroma jaczewskii*, *C. fructicola*: *Colletotrichum fructicola*, TGA–DTG: Thermogravimetric analysis-derivative thermogravimetry, NRM: Nuclear magnetic resonance, XRD: X-ray diffraction.

microbe adhesion to the polymer surface [58]. Differential scanning calorimetry is a method for carrying out thermal analysis (DSC). Its analysis provides capacity to track phase transitions, solid state transformations, and thermodynamic parameters during controlled sample heating and cooling. DSC analysis can be used to measure a variety of characteristics, including the glass transition temperature, crystallization temperature, melting temperature, polymer crystallinity percentage, specific heat capacity, transformation enthalpy, and many others [59]. Evolution of CO_2 is typically taken into account as a sign of biological decay [14,56]. By monitoring the CO_2 released during biotic or abiotic mineralization in a controlled setting, the rate of polymer degradation can be estimated [61-63].

The oligomeric fraction produced during polymer breakdown or the detection of low-molecular-weight metabolites can also be studied using gas chromatography (GC) with flame ionization detection (GC-FID) or mass spectrometry (GC-MS) [64-66]. Another chromatographic method utilized for the examination of complex oligomeric mixtures created during biodegradation is LC-MS [66]. Gel-permeation chromatography (GPC) is used to quantify molar mass and molecular weight shifts [67,68]. High-performance liquid chromatography (HPLC) is also used to identify certain homologues of low-molecular-weight polymers [69].

5. APPLICATIONS

In general, fungi are more effective in degrading polymers than bacteria because they can stick to the hydrophobic surfaces of polymers, produce extracellular enzymes that target insoluble fibers, and endure challenging growth environments [24,70,71]. Fungi produce a wide range of enzymes that have the potential to break down the chemical bonds of the plastic polymers [72]. According to a study, enzymes called laccases from fungi like *Aspergillus flavus* and *Pleurotus ostreatus* significantly degraded polyethylene [73,74]. According to another study, PS can be broken down by an extracellular esterase from *Lentinus tigrinus* [75]. Serine and cysteine hydrolase from *Pestalotiopsis microspore* was shown in another investigation to be active in degrading PU [76]. The breakdown of lignin is frequently correlated with the enzymes

manganese peroxidase (MnP) and lignin peroxidase (LiP) [77]. Several compounds converted into oxidized or polymerized products by these enzymes, which catalyze oxidation-reduction reactions [78]. Numerous fungi species, including *Humicola insolens*, *Penicillium citrinum*, and *Fusarium oxysporum*, produce enzymes such as polyesterase, cutinase, and hydrolase that are effective PET degraders [79-81]. Another study showed that *F. solani* was converted to terephthalic acid (TPA) from a hydrolyzed PET collection through a cutinase [82].

6. CONCLUSION

Several fungal microbes have the ability to breakdown the plastic polymer under the right conditions. The capacity of various fungal species can cause chemical and physical changes in various plastic polymers such as LDPE, HDPE, PET, PS, and others. Biodeterioration (Adhesion and colonization of microbes on the surface), biofragmentation (fragmentation of plastic polymer into monomer), assimilation (transport into microbial cytoplasm), and mineralization (enzymatic reactions that result in the breakdown into various oxidized metabolites such as CO_2, N_2, CH_4, and H_2O) are the four stages of biodegradation of plastic by fungal strains. Microbiological activities that use enzymes including peroxidases, lipases, esterase, cutinase, and laccase to catalyze plastic breakdown are growing as an environmental sustainable alternative to physicochemical depolymerization. Biodegradation, on the other hand, has a significant downside in terms of rate of degradation. Future research will put a lot of emphasis on the biological systems or biodegradation mechanism that are accountable for the detected chemical and physical change. With more efficient degradation on plastic polymer, it is important to identify novel fungal strains. The primary goal of upcoming study will be to understand and identify the biological systems that appear to be causing the apparent chemical and physical damage. Only few enzymes of a fungal origin have been associated to polymer breakdown thus far. However, these enzymes' biochemical and structural features have received little attention. These details are necessary to comprehend the mechanisms underlying the biodegradation of resistant polymers. This knowledge will be useful for creating novel biodegradable plastic polymers, designing microbial cell factories with improved breakdown efficiency, and genetically altering enzymes through protein engineering. The goal is to isolate and discover potent microbial consortia directly from plastic contamination locations, as well as purify potent microbial enzymes for commercial usage and increase catalytic activity through genetic engineering.

7. AUTHORS' CONTRIBUTIONS

All authors made substantial contributions to conception and design, acquisition of data, or analysis and interpretation of data; took part in drafting the article or revising it critically for important intellectual content; agreed to submit to the current journal; gave final approval of the version to be published; and agreed to be accountable for all aspects of the work. All the authors are eligible to be an author as per the International Committee of Medical Journal Editors (ICMJE) requirements/guidelines.

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All authors declare that they have no conflict of interest.

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This article does not contain any studies with human participants or animals performed by any of the author.

11. DATA AVAILABILITY

Data sharing is not applicable to this article as no datasets were generated or analyzed during the current study.

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