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Evaluating the Plastic Degrading Potential of Aspergillus Spp. for Sustainable Waste Management

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Abstract

Plastics are synthetic polymers that are widely used in every field of life every day. Along with the increasing use of plastic, the amount of plastic waste produced and accumulated to the environment will also increase. If the plastic waste is not handled properly, it will pollute the environment and threaten many living things including humans. Biodegradation is a promising method for dealing with plastic waste. This method includes many microbes including fungi such as Aspergillus spp. and Penicillium spp. as its biodegradation agents. Some plastics degrading fungi produce many specific enzymes that catalyze the degradation of plastic polymer into simpler and smaller fragments including oligomer, dimer, and monomer through several steps including biodeterioration, depolymerization, assimilation, and mineralization. The fragmented plastic particles are absorbed and used by plastic degrading fungi as their energy and carbon sources. Biodegradation is the appropriate method to overcome the plastics pollution because this method has no side effects as the conventional methods. This paper highlights the critical issue of plastic pollution and the potential for biodegradation as a sustainable and eco-friendly solution to address the growing crisis.

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1. Introduction

Plastic, renowned for its affordability, lightweight properties, water resistance, and versatility, has found extensive application in nearly all aspects of life. Its usage is anticipated to surge annually by approximately 9%, contributing to a global production of 400 million tons per year. Unfortunately, plastics comprise hazardous, non-degradable components detrimental to ecosystems and living organisms. The ubiquitous use of plastic products results in their accumulation in the environment, polluting water bodies, land, and air.

In Western Europe, a substantial 7.4% of municipal solid waste is comprised of plastic, with 65% being polyethylene and polypropylene, 15% polystyrene, 10% PVC, and 5% PET, among other polymers. In 2010, China topped the list as the highest producer of mismanaged plastic waste, accounting for 27.7% of the global total, approximately 8.82 million metric tons per year. Indonesia followed closely at 10.1%, with the Philippines (5.9%), Vietnam (5.8%), Sri Lanka (5%), Thailand (3.2%), Egypt (3%), Malaysia (2.9%), Nigeria (2.7%), Bangladesh (2.5%), and the United States (0.9%) completing the top ten. The production of single-use plastic is highest in North-East Asia (26%), followed by North America (21%), the Middle East (17%), Europe (16%), Asia and the Pacific (12%), Central and South America (4%), and former Soviet countries (3%). The packaging sector dominates the consumption of single-use plastic, accounting for 36%. The building and construction sector uses 16%, textiles 14%, consumer and institutional products 10%, transportation 7%, electronics/electrical 4%, machinery industry 1%, and the remaining 12% is distributed across other sectors.

Plastic waste has emerged as a pervasive contaminant on land and in aquatic environments, necessitating effective waste management solutions. Common methods include landfilling, incineration, and recycling, each of which possesses inherent weaknesses. Landfilling entails long decomposition times, incineration generates toxic emissions, and recycling is often costprohibitive.

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In response, various plastic waste processing techniques have been developed, encompassing energetic, chemical, and biological approaches. Energetic methods employ radiation energies, such as gamma rays, ion beams, electrons, and UV rays. Chemical processing employs specific acids and alkalis. The biological method, biodegradation, stands out as the most promising and environmentally friendly approach to plastic waste management. Biodegradation harnesses microorganisms to break down plastic waste. This paper highlights the critical issue of plastic pollution and the potential for biodegradation as a sustainable and eco-friendly solution to address the growing crisis.

2. Types of Plastic

Plastics can be categorized into two main groups: thermoplastics and thermosets. Thermoplastics are plastics that can be melted and reprocessed, making them recyclable. In contrast, thermosets, also known as thermosetting plastics, cannot be melted due to tightly bound molecular bonds within the plastic polymers.

The four most commonly used plastic polymers in everyday life are:

- 1. High-density polyethylene (HDPE)
- 2. Low-density polyethylene (LDPE)
- 3. Polypropylene (PP)
- 4. Polyethylene terephthalate (PET)

These plastics are frequently used in various applications, with plastic bags being one of the primary sources of plastic pollution.

Polyethylene (PE) is the most widely used plastic, accounting for 29.6% of total daily plastic usage. It is followed by polypropylene (PP) at 18.9%, polyvinyl chloride (PVC) at 10.4%, polyurethane (PUR) at 7.4%, polystyrene (PS) at 7.1%, and PET at 6.9%.

Polyethylene (PE) is a synthetic plastic characterized by its high molecular weight, complex three-dimensional structure, and hydrophobic properties. It consists of stable polymers formed from ethylene monomers, making it resistant to degradation in nature.

Polypropylene (PP) is another synthetic plastic known for its heat and chemical resistance. This property renders PP resistant to degradation.

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Polyethylene terephthalate (PET) contains two hydroxyl groups (OH) and dicarboxylic aromatic acids with aromatic carbon rings and two carboxyl groups (CO2H). This plastic is synthesized through a two-step chemical process. The presence of aromatic components in PET makes it challenging to naturally degrade in the environment.

Ester bonds within PET polymers contribute to their strong and recalcitrant nature, making them resistant to environmental degradation. However, certain microbial communities have the ability to utilize PET as a carbon and energy source. Extracellular polymeric components produced by these microbes play a crucial role in overcoming the recalcitrant properties of plastics like PET. These components act as biosurfactants that facilitate the interaction between the plastic surface and microbial cell surfaces, aiding in plastic polymer degradation.

Biodegradable plastics and polymers have gained prominence in various industries. Biodegradable polymers are characterized by the presence of amides, esters, or ether bonds. They can be further classified into agro-polymers and bio-polyesters. Examples of biodegradable plastics include polylactic acid (PLA) and polybutylene adipate-coterephthalate. While biodegradable polymers are more easily degraded than nonbiodegradable synthetic polymers, they still possess strong carbon bonds that require months or even years for complete biodegradation in natural environments.

3. The Detrimental Impact of Plastic Waste on the Environment and Potential Biodegradation Solutions

Plastic waste that litters the land is only partially degraded by natural environmental processes, resulting in the release of harmful substances like heavy metals, plasticizers, stabilizers, and plastic dyes into the environment. These pollutants not only contaminate terrestrial environments but are also transported by water, contributing to around 80% of plastic pollution in aquatic ecosystems.

Plastic waste has the ability to drift across different locations, potentially carrying living organisms to new areas and introducing invasive species that can compete with native species. Moreover, the incineration of plastic waste, a common disposal method, generates toxic gases like carbon monoxide (CO) and hydrogen monoxide (HCN), posing a threat to air quality and human health.

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Despite the slow natural degradation of plastics over hundreds or thousands of years, this process results in the release of toxic fragments into the environment, causing various health issues. Additionally, the visual pollution caused by the accumulation of plastic waste diminishes the appeal of tourist destinations, negatively impacting the economies of countries that heavily rely on tourism. Therefore, an effective and environmentally friendly method for processing plastic waste is essential. Numerous studies suggest that using microbial enzymes, such as those produced by fungi, to degrade plastic waste is a promising approach to address plastic pollution.

• **Plastic Waste Biodegradation:** The recycling of plastics has several limitations, particularly when plastics are mixed with other waste materials, making it challenging to separate them by type. Contaminated plastic waste can damage recycling equipment, and the burning of plastics by communities' releases pollutants that harm the environment, including the emission of greenhouse gases.

Biodegradation presents a compelling solution to address these challenges. This process completely breaks down plastic polymers, resulting in the production of microbial biomass as its biological agents. Synthetic polymers, such as polyethylene and polypropylene, serve as potential energy and carbon sources for microorganisms, with polyethylene, for instance, producing energy equivalent to glucose upon complete oxidation.

Nonhydrolyzable synthetic polymers, which contain C-C skeletal bonds, require initial breakdown through redox reactions into simpler compounds before being assimilated by microbial cells. In contrast, hydrolyzable polymers, like polyethylene terephthalate and polyamide, which contain amide and ester bonds, can be directly hydrated by microbial enzymes.

Hydrolysis, the breaking of polymer bonds through water recombination, plays a crucial role in the degradation of semi-crystalline polymers. This process reduces the crystallinity and molecular weight of the polymer, making it more soluble in water and increasing its erosion rate. Moreover, microbial biodegradation involves the formation of biofilms on plastic surfaces, enhancing the degradation process.

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The biodegradation process of plastic waste follows four stages: biodeterioration, depolymerization, assimilation, and mineralization. Biodeterioration involves the collaborative activities of microorganisms and abiotic factors that break down polymers into simpler compounds. Depolymerization is the secretion of catalytic compounds, such as enzymes and free radicals, by microorganisms to progressively break polymer chains into oligomers, dimers, and monomers. Microbes release exoenzymes during depolymerization, breaking down complex polymers into simpler compounds used as carbon and energy sources. Mineralization, the final stage, produces CO2, H2O, or CH4 through biological activities.

The rate of plastic waste biodegradation depends on various factors, including polymer structure, molecular weight, molecular shape, and crystallinity. Although the distinctions in plastic structures may not be significant, these factors collectively influence the degradation process.

The widespread issue of plastic waste necessitates comprehensive solutions to prevent its detrimental effects on the environment and human health. The biodegradation of plastic waste through the activity of microbial enzymes offers a promising approach to address this problem. By understanding the stages and factors involved in biodegradation, we can work towards more effective and sustainable plastic waste management.

4. Plastic Degrading Fungi

Numerous studies have highlighted the remarkable capability of fungi in the biodegradation of plastics, as summarized in Table 1. Leveraging fungi as bioremediation agents is a highly effective approach to curbing the proliferation of plastic waste in our environment. The rapid growth of fungi across diverse substrates proves to be a significant advantage in plastic biodegradation. Thanks to their swift growth, fungal mycelium can readily proliferate, covering the entire surface of the substrate and subsequently penetrating it to initiate the biodegradation process. Fungi exhibit adaptability to a wide range of environments, including those with extreme conditions such as low pH and arid climates. Fungi strains isolated from soil environments exposed to plastic contamination are particularly valuable for their ability to degrade plastic waste. Some well-documented plastic-degrading fungi include Gliomastix

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sp., Chaetomium sp., Fusarium sp., Mortierella sp., and Paecilomyces sp. Furthermore, indigenous fungi obtained from landfills contaminated with plastics, such as Trichoderma sp., Aspergillus flavus, and Aspergillus niger, demonstrate significant potential as agents for plastic biodegradation.

Table 1: Some plastics degrading fungi

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Numerous studies have reported the existence of polyethylene (PE)-degrading fungi, with Myceliophthora sp. being one of them. This particular fungus produces laccase enzymes that catalyze the degradation of plastic polymers. Laccase is optimally produced at a pH of 5.0 and a temperature of 30°C. The ability of this fungus to grow on a medium containing

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polyethylene is noteworthy, and electron microscopy analysis has shown that the plastic's surface exposed to the fungus is damaged, which is closely linked to the activity of the laccase enzyme.

Two other isolates capable of degrading polyethylene are Aspergillus terreus MANGF1/WL and Aspergillus sydowii PNPF15/TF, both of which were isolated from the rhizosphere of Avicennia marina. Aspergillus terreus MANGF1/WL reduced the weight of plastic samples by 50% at pH 5.0 within 60 days, while Aspergillus sydowii PNPF15/TF reduced the weight of plastic samples by approximately 94% at pH 3.5 within the same incubation period. These reductions in weight were attributed to the activity of depolymerase enzymes that break the polyethylene bonds. Similarly, other Aspergillus species like A. fumigatus and A. oryzae reduced polyethylene weight by 24% and 36%, respectively, indicating their capacity to contribute to plastic degradation.

Additional findings revealed that Aspergillus spp., such as A. niger, A. oryzae, A. japonicus, and Penicillium sp., were successfully isolated from plastic-contaminated dumping sites. Among these, A. oryzae dominated the soil samples. A. oryzae's plastic degradation ability was tested using buried plastic strips, and it was found to degrade the plastics by approximately 20% within 15 days, 24% within 30 days, 26% within 45 days, and 30% within 60 days. Scanning electron microscopy (SEM) analysis revealed that the plastics exhibited microcracks, further confirming the degradation process.

Chaetomium globosum was also identified as a plastic-degrading fungus, with higher degradation rates observed when the plastic was subjected to preliminary treatment with UV light and autoclaving. These treatments reduced the plastic's weight by about 21% within three months when exposed to UV light. However, the degradation of autoclaved plastic using Chaetomium globosum was only 7.5%, and plastic samples that were surface-sterilized before exposure to the fungus degraded by about 5.6% within the same incubation time. Other fungi like Aspergillus niger, A. flavus, and Fusarium sp. AF4 were also reported to damage the plastic surface, thus facilitating the biodegradation process.

Moreover, various fungi, including Aspergillus niger, A. flavus, A. terreus, A. fumigatus, and Penicillium sp., were obtained from the Red Sea and found to degrade LDPE (low-density

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polyethylene). The weight loss of LDPE samples exposed to these fungi within one month ranged from 16.2% to 43.3%. SEM analysis revealed significant damage to the plastic surfaces compared to the control samples, which remained intact. Similarly, Mucor sp. isolated from soil exposed to polyethylene plastic degraded LDPE by approximately 16% within four weeks in a synthetic medium containing LDPE as the primary carbon source. Other LDPE-degrading fungi, such as Alternaria alternate, Emericella nidulans, Paecilomyces variotii, Penicillium duclauxii, and P. vinaceum, were also identified.

Aspergillus fumigatus exhibited a remarkable ability to degrade HDPE (high-density polyethylene). The fungus reduced the weight of HDPE samples using soil media by 59% within nine months and by 29.1% within two months in a liquid medium. SEM analysis indicated that plastic surfaces exposed to the fungus in soil media exhibited more damage and perforations compared to those in the liquid media. Additionally, Fourier-transform infrared (FTIR) analysis detected the presence of various functional groups in the degraded HDPE, including ketones, aldehydes, and carboxylic acids. Bjerkandera adusta was also reported to degrade HDPE by producing oxidative extracellular enzymes that acted as plastic-degrading agents. Raman analysis further illustrated how the enzymes produced by the fungus degraded the amorphous structure of HDPE.

Certain fungi, such as A. niger, Penicillium oxalicum NS4, and P. chrysogenum, were found to degrade both LDPE and HDPE. A. niger, for instance, reduced the weight of LDPE and HDPE, with different levels of weight reduction observed between submerged and composting methods. Additionally, A. niger was capable of degrading another polymer, PET (polyethylene terephthalate), by approximately 52.94% within one month when grown in Rose Bengal Broth.

Several fungi have been identified as agents for degrading PHB (polyhydroxybutyrate) plastics, including Penicillium, Aspergillus, Fusarium, Alternaria, Trichoderma, A. fumigatus, A. oryzae, and Trichoderma viride. These fungi exhibited varying levels of PHB degradation within seven days when cultivated on solid media containing PHB. The hydrolysis of PHB polymers was catalyzed by depolymerase enzymes produced by these

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fungi, and SEM analysis showed surface perforations on the plastic, which is an initial stage of plastic degradation.

Polyurethane plastic (PUR) degradation was achieved by several fungi, such as Monascus sanguineus, Monascus sp., and Pestalotiopsis microspora, which produced esterase, protease, and lipase enzymes responsible for breaking down the ester bonds of PUR. Additionally, endophytic fungi have been reported to degrade plastics, with Pestalotiopsis isolated from Nephentes ampullaria being able to degrade polyurethane (PUR). The involvement of metallothionein-like proteins in PUR degradation was also suggested. Furthermore, Pestalotiopsis microspora demonstrated the capability to degrade plastics in anaerobic conditions, which is essential for plastic waste degradation in extreme environments.

Fungi have also been identified as agents for degrading polyester-polyurethane (PS-PUR) plastics. Spicaria spp., Aspergillus sonali, A. terreus, A. flavus, A. fumigatus, and Fusarium solani were found to use PS-PUR as a carbon and nitrogen source. In liquid shaking culture, Fusarium solani achieved complete degradation (100%) of the plastic sample within three weeks. Other fungi also exhibited significant levels of PS-PUR degradation under the same conditions. SEM analysis showed signs of plastic degradation, such as changes in color, perforation, and surface damage.

Fungi have proven to be effective agents for the degradation of plastic waste in various environments, including both land and aquatic settings. For instance, Zalerion maritimum, a marine fungus, was found to degrade PE (polyethylene) polymer in aquatic environments.

5. Enzymes Involved in Plastic Biodegradation

Plastic pollution has become a global environmental concern, prompting the search for sustainable methods to address this issue. Microbial biodegradation, particularly by fungi, offers a promising avenue for breaking down plastics. This paper explores the enzymes released by fungi during the biodegradation of plastic polymers and their roles in this process.

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5. Lipases and Esterases:

Lipases and esterases play a crucial role in breaking down ester bonds found in various plastic polymers, including polyesters such as polyethylene terephthalate (PET). These enzymes, often produced by microorganisms like Monascus ruber, catalyze hydrolytic cleavage of ester bonds in plastics.

5.2 Laccases:

Laccases are enzymes commonly found in fungi and play a vital role in the oxidation of phenolic substrates. They have shown promise in degrading plastic materials, including polyethylene, through oxidation processes, ultimately leading to the production of water and carbon dioxide.

5.3 Depolymerases:

Depolymerases, such as polyhydroxyalkanoate (PHA) depolymerases, polyhydroxybutyrate (PHB) depolymerases, and polylactic acid (PLA) depolymerases, target specific plastic polymers. PHA depolymerases are involved in breaking down polyhydroxyalkanoate plastics, while PHB depolymerases degrade polyhydroxybutyrate. PLA depolymerases are essential for the degradation of polylactic acid, recognizing the α -ester bonds in PLA.

5.4 Ureases:

Ureases play a role in plastic biodegradation, particularly in the case of polyurethane (PUR) polymers. These enzymes catalyze the degradation of PUR, contributing to the breakdown of this type of plastic.

5.5 Other Enzymes:

Various other enzymes, including cutinases, proteinases (e.g., proteinase K in PLA degradation), and dehydratases, participate in the breakdown of specific plastic materials. These enzymes demonstrate the diversity of enzymatic pathways involved in plastic biodegradation.

6. Fungal Species and Enzyme Production:

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Different fungal species are known to produce a wide range of enzymes involved in plastic biodegradation. For example, Aspergillus flavus produces laccase, amylase, lignin peroxidase, and manganese peroxidase. Aspergillus niger produces laccase, lignin peroxidase, and manganese peroxidase, while Fusarium graminearum produces amylase, laccase, lignin peroxidase, and manganese peroxidase. These enzymes are essential in breaking down carbon bonds in polyethylene polymers.

Pestalotiopsis microspora, an endophytic fungus, is known for degrading polyurethane plastics. It produces serine hydrolase and polyurethanase. Chaetomium globosum catalyzes the degradation of polyethylene using laccase and manganese peroxidase.

Enzymatic plastic biodegradation, particularly by fungi, offers a sustainable solution to address plastic pollution. The diverse array of enzymes produced by different fungal species allows for the targeted breakdown of various plastic polymers. Understanding these enzymatic processes can guide the development of effective bioremediation strategies and contribute to the reduction of plastic waste in the environment.

7. Conclusions and Future Perspectives

Plastics biodegradation through fungal action holds significant promise as a sustainable solution to the ever-mounting issue of plastic waste. This method is both eco-friendly and safe, contributing positively to environmental conservation. Fungi, equipped with various extracellular and intracellular enzymes, efficiently break down plastic polymers. They utilize these polymers as sources of energy and carbon for their metabolic processes. Implementing this approach has the potential to address the alarming plastic waste accumulation problem.

However, it is important to recognize that the biodegradation of plastic waste is not without its limitations. The process is influenced by various biotic and abiotic factors, as well as the characteristics of the plastic polymers themselves. Moisture, temperature, and pH levels represent key abiotic factors affecting the rate of plastic biodegradation. Biotic factors crucial to the process include the presence of specific enzymes and the hydrophobic nature of plastics. Additionally, the characteristics of the plastic polymers, such as their molecular weight, size, shape, additives, and the presence of biosurfactants, play a significant role. Furthermore, the choice of fungus employed in the biodegradation process has a direct

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impact. Each fungus species exhibits varying effectiveness in degrading specific types of plastic waste, which is associated with their ability to produce the requisite enzymes for plastic decomposition.

Notably, existing research in the realm of plastic biodegradation mainly centers around laboratory-scale experiments and macro plastic waste. Moreover, the reported studies indicate that the biodegradation of plastic waste by microorganisms is a time-consuming process. Therefore, future research and development efforts should focus on enhancing our understanding of fungal metabolism in breaking down plastic waste. This includes delving into metabolic engineering and optimizing processes to bolster the fungal capacity to degrade plastic waste effectively. Addressing these challenges and limitations is imperative to accelerate the progress in the field of plastics biodegradation and reduce the environmental impact of plastic waste.

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