

Investigating chemical-biological degradation of plastic compounds by fungi isolated from industrial plastic waste

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Abstract:

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Introduction: This study aimed to isolate chemical-biological plastic-degrading fungi and investigate their enzymatic biochemical activity.

Materials & methods: A collection of fungal isolates from 30 different plastic compounds collected from industrial waste. *Aspergillus niger*, *Aspergillus flavus*, *Penicillium commune*, *Bipolaris sorokiniana*, *Curvularia eragrostidis* capable of chemical-biological degrading the selected compounds were identified based on PCR. Using lccC and Mnp primers, the gene of Mnp and laccase producing enzyme was identified. SEM was used to determine the surface degradation, porosity, and fragility of polyethylene, and FTIR was used to determine its chemical structure.

Results: *Aspergillus niger*, *Curvularia eragrostidis*, *penicillium commune*, and *Bipolaris sorokiniana* were identified among the industrial plastic waste, they decompose polyethylene more and *Aspergillus flavus* showed the greatest weight loss in the decomposition of industrial plastic waste in the culture medium.

Discussion: The decomposition of plastic waste by microbes in a short time not only solves the problem of waste accumulation but also produces valuable compounds like SCP.

Keywords: Polyethylene; *Aspergillus niger*; *Curvularia eragrostidis*; Laccase; Urease; Lipase

1. Introduction

The word plastic derives from the Greek word *plastikos*, which means it can be molded [1, 2]. The raw materials for plastic production are materials that are high molecular weight polymers made by human hands that are used in various industries (Shimao, 2001). Removing environmental and industrial waste pollution has become a big problem today. Microbes can help in this process [3]. Plastic and heavy metal industrial wastes are the most important biological pollutants. There are two important methods to remove these residues. Chemical method and biological method. In the biological method, microbes help to remove plastic waste using enzymes in a biochemical way. With the help of fungi, including yeasts, biological removal of heavy metals can be done [4]. Plastic materials have been widely used in the production of various industrial

products by humans in the last three decades. The main advantages of plastic are its light weight, flexibility, durability and hardness, as well as its low price [5]. Plastic is defined by the plastic industry Association (SPI) as a large and distinct group of materials whose chemical structure contains carbon, oxygen, nitrogen, hydrogen, and other elements in their chemical structure. Their plastic products are identified by an SPI code at the end of the mold [6]. Plastics are formed by bonding monomers together through chemical bonding. Polyethylene makes up 64% of all plastics [7]. It is a linear hydrocarbon containing long chains. They are ethylene monomers (C₂H₄). The general formula of polyethylene is C_nH_{2n}, where “n” is the number of carbon atoms [8]. The ethylene molecule has a C = C bond. In the polymerization process, the double bond of each one of the monomers is broken, and instead, a simple bond is formed between the carbon atoms of the monomers,

and the product is a large molecule called polyethylene [9]. Polyethylene is widely used in the production of various types. Plastic appliances are used in the kitchen and food industry. Low-density polyethylene (LDPE) in the production of lightweight plastic containers and plastic bags are also used [10]. High-density polyethylene (HDPE) is used in the production of milk and liquid containers and all kinds of kitchen utensils [11]. Various plastic devices such as bendable pipes are made from linear low-density polyethylene (LLDPE) due to their high degree of flexibility [12]. Polyethylene (PE) was first synthesized by chance by the German chemist Hans Von Pechmanv. In 1898 when heating diazomethane, The wax compound synthesized a white form, later called polyethylene [13]. The first method of industrial synthesis of polyethylene was accidentally discovered in 1933 by Azic Navest and Rinoelergison (ICI 14 chemists). The two scientists obtained wax-like substances by heating a mixture of ethylene and benzaldehyde at high pressure. The reason for this reaction was the presence of oxygenated impurities in the devices used, which acted as a polymerization initiator [14]. Accumulation of plastics in nature, while most of them are not biodegradable and increased accumulation in the environment is very dangerous and today is a major environmental problem. Proposed and various solutions proposed, the use of microorganisms in the decomposition of polymer compounds hard to decompose is one of the solutions that have been considered by many researchers today [15] by isolation and identification of *Phylloplane* strain B479 from leaves of plants, showed this ability to degrade commercial polymyoplasty film consisting of polysuccinate (PBS), polybutylene succinate (PBSA), polybutylene terephthalate, quadipate (PBAT), which is on the surface Sterile soil was placed, after 6 days of incubation at 28 °C they lost 99.8% of their weight [14]. H. V. Sowmya et al. (2014) with isolation and identification of *Penicillium simplicissimum* investigation of the degradability of polyethylene concluded that this isolate, with the production of laccase enzymes and Mnp, causes the biodegradation of this plastic compound [16] and *Bjerkandera adusta* TBB-03 strains observed changes in HDPE levels. Raman test showed that the amorphous structure of this compound was destroyed by the production of the enzyme laccase [17]. Biofilm formation by *Pseudomonas aeruginosa* ISJ14 strain was investigated on low-density LDPE polyethylene[18]. Biofilm formation with surface abrasion, Cracks were observed through SEM analysis [18]. In a study by Potrykus Marta et al., showed that polypropylene pretreated with light oxidation due to the formation of carbonyl groups easily is consumed by fungi or bacteria, and its biodegradability is higher [10, 19]. This study is conducted with the aim of biological-chemical removal of plastic compounds such as polyethylene, polypropylene, polyethylene terephthalate, polystyrene and polyamide by Iranian native fungal strains and analyzing the decomposition and enzymes involved in this process.

2. Methods and materials

2.1 Sample collection

Based on Cochran's formula, the number of samples was determined to be 30 [20]. A total of 30 samples of plastic containers, including disposable plastic cups, plastic bottles, and nylon from around Tehran, were collected and immediately sent to the laboratory. Sterile pliers and zippers were used to collect samples. Industrial plastic waste was obtained from food production factories.

2.2 Isolation and purification of fungi-consuming compounds

To isolate and purify plastic-degrading fungi, pieces of plastic separated from the environment are chopped into 1×1 cm dimensions and added to the Potato dextrose agar (PDA) medium (g/L). Ingredients of the medium include glucose (20), cooked potato (300), Agar (15) and Saburo dextrose agar (SDA) (g/L): Glucose (40), Meat peptone (5) Peptone (g/L): Glucose (40), Meat peptone (5) is Peptone. Then the plates were placed in an incubator at 28 °C for 3-7 days. The colony of saprophytic fungi appears with different colors on the culture medium. Each colony was cultivated separately on a plate containing PDA medium, with point culture methods. After 3–7 days of incubation of plates in the incubator, the colonies were isolated separately at a temperature of 28 °C. Furthermore, chloramphenicol (50 µg/mL) was used to prevent bacterial growth. After repeated cultivation and the formation of pure colonies, the isolates were examined macroscopically and microscopically. Lactophenol catechin blue was used to stain the culture slide. After preparing the standard fungal suspension in M9 saline base medium containing as per g/l: Na₂HPO₄.2H₂O (72.2), KH₂PO₄ (30), NaCl (5), NH₄Cl (5) were cultured and the effect of decomposition of compounds in the presence of 1% they were considered as the only source of carbon [21]. Standard suspension was used for all experiments. Culture media were prepared by Merck Company (Darmstadt, Germany). To preparation of standard suspension size was adjusted to between 1.0×10^6 and 5.0×10^6 spores/mL by microscopic enumeration with a cell-counting Neubauer Chamber [22].

2.3 Selecting the desired isolates

For the selection of an isolate, 250 mL erlenmeyer flasks, 10 mL of culture medium should be at least M9 accompanied by chloramphenicol (to prevent bacterial growth), as well as 600 µL from fungus suspension and 0.1 g of polyethylene and other plastic compounds and granules prepared by Zar Shimi Company in a sterile manner by flame. After closing the lid of the Erlenmeyer flasks with cotton and foil under sterile conditions, the Erlenmeyer flasks were placed in a shaker incubator at a temperature of 28 °C and 120 rpm. Then the growth curve of fungi was read at time intervals of 0, 24, 48 and 72 hours and the optical absorbance (OD) of the samples was read. The isolates that had better growth were considered selected isolates (Safarzadeh and Moghimi, 2019).

2.4 Qualitative and molecular study of enzymatic activity of the obtained isolates

In this study, qualitative and molecular methods were used to evaluate the enzymatic activity of the strains. Isolates, Slate urea agar medium, Malt agar with Magnesium chloride substrate, Peptone agar with substrate Tween 80, Glucose-yeast agar with Alpha naphthol substrate, and Skim milk medium according to the following method, the enzymatic activities of urease, Mnp, Lipase, Laccase and Protease of isolates were examined respectively [24]. For urea activity, a slant urea agar culture medium was used [25], to prepare, 24 g of Urea medium. Base agar whose components include (g/L): Peptone (1), Dextrose (1), Sodium chloride (5), Monosodium phosphate (2), Phenol rejection (0.012), agar (15) in 950 cc of distilled water at 121 °C for 15 minutes autoclaved, Urea due to sensitivity to high temperature and decomposition and inactivation, after ambient temperature reached 50 °C, 50 mL of 40% Urea was filtered through a 0.22-micron membrane filter inside the medium [26]. Evaluation of peptone agar lipase enzyme activity with Twin 80 substrate whose components include (g/L): Peptone (10), Calcium chloride (0.1), Sodium chloride (5), agar (15) before sterilization and after temperature the environment reached 50 to 55 °C. 10 mL Twin 80 with a membrane filter of 0.45 microns to the medium was filtered [27]. To evaluate the lactase enzyme from a glucose-yeast medium with alpha naphthol substrate including Yeast extract (5), Dextrose (5), and agar (5) after sterilizing the medium of 0.05 g of the alpha compound, yeast extract (5), Dextrose (5), agar (5) after sterilizing the medium 0.05 g of alpha compound naphthol was added to the medium. Skim milk culture medium was used to evaluate protease enzyme activity. 100 g of ready-made skim milk powder was added to 500 mL of distilled water and dissolved separately 20 g of ready-made nutrient agar powder (g/L): Peptone (5), yeast extract (1.5), Meat extract (1.5), Sodium chloride (5), agar (15) were prepared. Both cultures were autoclaved separately and added together at a temperature of 50 to 55 degrees Celsius next to the flame [28]. Qualitative evaluation of Mnp enzymes Lipase, laccase and protease based on the halo created by the activity of isolates and his release activity based on the change in the color of the culture

medium was evaluated from purple to yellow. To evaluate the presence of the gene producing Mnp and laccase c enzymes by molecular method, Mnp (Manganese peroxidase) and lccC (laccase C) primers were used according to research by Li et al. For molecular identification, isolates purified on agar sabouraud dextrose medium are separated from the culture medium and assisted. Liquid nitrogen was frozen and crushed to break down the cells and their DNA physically It was extracted and isolated using the Gene Pioneer Transfer kit. In the next step, its PCR was primed Mnp, ICCc was performed [23]. The product was taken on gel electrophoresis. PCR results and gel electrophoresis showed that *Aspergillus niger* laccase (+) and its genomic region is 2609 bp, and *Aspergillus flavus* is Mnp (+) and the genomic region is 560 bp. The sequence of Mnp and ICCc primers in (Table. 1) is shown.

2.5 Identifying the selected isolate

For microscopic observation of reproductive and vegetative structures of fungi by slide culture technique and staining, Cotton blue lactophenol was used. Fungi are based on microscopic and macroscopic shapes, including color. Colonies were identified on the surface and back of the plate. For molecular identification of purified isolates, isolate on sabouraud dextrose agar medium on culture medium and with the help of liquid nitrogen, frozen and crushed to physically break down the cell and their DNA with the help of the gene transfer Pioneer kit extracted and separated. Next, PCR was performed by primers: ITS1, and ITS2. Sequences of ITS1 and ITS2 primers are shown in Table 2. The results were taken on gel electrophoresis. The bands in the region 560 bp genome were observed by amplifying the extracted genomic region. Korea PCR product was sent. After determining the sequence of gene gate and BLAST at the NCBI site, it was identified using MEGA7 software to identify fungal phylogenetic trees [29].

2.6 The qualitative study of the decomposition activity of plastic compounds by selected isolates

Examine selected isolates for 5 enzymes of urease, Manganese oxidase, lipase, laccase and proteases. The decomposition of isolates in Erlenmeyer flasks containing 10 mL

Table 1. The primer sequences of Mnp and ICCc genes [23].

| primerName | Sequences (5' -> 3') | MW | OD |
|--------------|-----------------------------|------|----|
| Mnp Forward | ACTAGTATGGCCTTCAAGTCCCTCA | 650 | 5 |
| Mnp Reverse | CCATGGTTATGCAGGGCCGTTGAAC | 650 | 5 |
| ICCc Forward | CACCATGCTGCGTTCTTCTTTCTTCTC | 2609 | 5 |
| lccC Reverse | GGAGAGCCTACGAGTGTTCCTACTGG | 2609 | 5 |

Table 2. ITS1, ITS2 primer sequences [23].

| Name | Sequences (5' -> 3') | MW | OD |
|------|----------------------|-----|----|
| ITS1 | TCCGTAGGTGAACCTGCGG | 560 | 5 |
| ITS2 | GCTGCGTTCTTCATCGATGC | 560 | 5 |

of M9 medium with 600 μL suspension was the standard for fungi and 1% plastic compounds were the sole carbon source. Isolates that have growth were chosen more favourably.

2.7 The quantitative study of the decomposition activity of plastic compounds by selected isolates

To quantitatively investigate the decomposition activity of isolates as described in the environment-containing Erlenmeyer M9 culture was prepared with 600 μL fungal suspensions and 1% plastic compounds and Erlenmeyer incubators. Shakers were placed at 28 °C at 120 rpm. Optical absorption (OD) of the samples in the range periods of 0, 24, 48, and 72 hours with a wavelength of 600 nm were taken. Separators with more favourable growth were elected [30].

2.8 Cultivation of selected strains in a culture medium containing polyethylene sheets and other plastic compounds

Culture media containing polyethylene and other plastic compounds were used for the cultivation of selected strains, as only a certain size plate of compounds was initially provided for the carbon source [31]. Then 10 mL of M9 medium was sterilized at 121 °C for 20 minutes after Cooling, 1 \times 1 cm polyethylene plates and other sterilized compounds by the flame with sterile pliers. Added 600 L of standard fungal suspension to the medium. Using M9 culture medium and fungal suspension in a shaker incubator, they were heated for 15, 30, and 45 days at 28 °C and 120 rpm. The isolates were adapted to the environment and polyethylene and other plastic compounds consumed only the carbon source and decomposed [32].

2.9 Measure the weight loss of polyethylene and other compounds using a digital scale

To investigate the weight loss of the plates of the compounds, a digital scale was used. Erlenmeyer was removed containing the culture medium, the fungal isolates were removed, then deionized water and sterilized on paper. The sterile decanter filter and funnel were washed, and after complete drying of the samples with a digital scale, the weight of the treated plates was measured, and their weight loss compared to the initial weight was reported as a percentage [33].

2.10 A Survey of surface changes (creating holes and cracks) created using electron microscopy (SEM)

Biodegradation is a superficial and apparent decomposition that alters materials' chemical, physical and mechanical properties. To investigate polyethylene sheets and other compounds, they are first cultured from Erlenmeyer containing the medium and fungal isolates were removed. For culture, fungal isolates were removed. To stabilize, the isolates on the plates were immersed in 25% glutaraldehyde for 2 hours and then rinsed with distilled water in alcohol. 25% were placed to complete dehydration. Dried plates on the slide containing glue the carbon were transferred to the Saba Sensor Laboratory and photographed with an FEI model electron microscope Quantum 2000 was performed [34, 35].

2.11 Investigation of chemical changes caused by biodegradation using FTIR infrared spectroscopy technique

Microorganisms such as fungi cause chemical changes and breakdown by affecting polymer compounds. There are different links in them. Bond failure, chemical bond deformation and grouping of the new function are biodegradable. The culture medium and fungal isolates were washed with sterile distilled water. After complete drying, the plates were ground into a powder and mixed with a softener (mineral oil). Powder and oil slurries were rubbed on a sheet of potassium bromide and sandwiched with another sheet of potassium bromide. After completing the mentioned steps, the various bond graphs with the FTIR (infrared spectrometer) model Perkin Elmer model Spectrum 1 were taken [36].

3. Results

3.1 Collecting samples and examining fungal isolates capable of producing enzymes

At this stage, 30 different plastic compounds were collected from plastic waste landfills. Their ability to grow in environments containing different plastic compounds was measured. Isolates were able to grow and adapt to the compounds and isolates selected that were able to grow in this environment were not removed. After 3-7 days of incubation of plates in an incubator at 28 °C, colonies were formed in different media and cultures of fungi. In Figure 1 A, different colonies in PDA, SDA culture media has been shown. To observe the microscopic reproductive and vegetative structures of the fungus, the slide culture technique was carried out (Figure 1 B).

Purified colonies of fungi and their reproductive and vegetative structures and microscopic images in Figures 2 A, and B it has been shown.

3.2 Investigation of enzymatic activities of fungal isolates

Activity qualitative enzymatic tests of urease, Mnp, lipase, laccase, and protease were performed for isolates and based on the intensity of color and halo created, and was evaluated

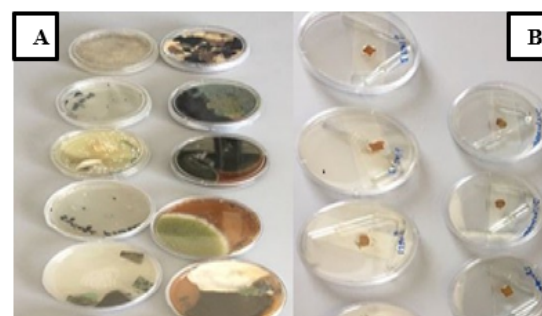


Figure 1. A: Colonies on plates containing PDA and SDA medium from plastic container contamination, B: Samples of slide culture method.

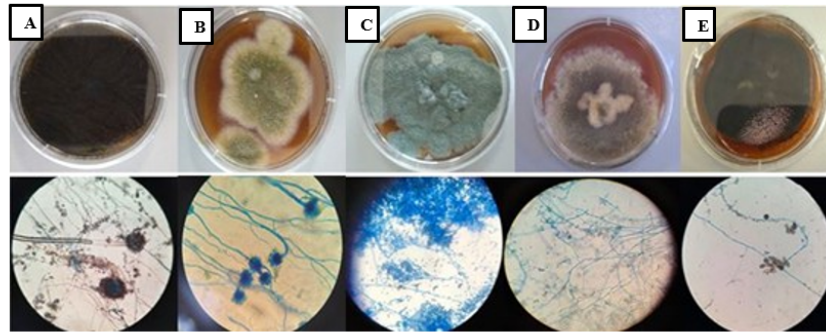


Figure 2. Colonies purified from container contamination and microscopic appearance and structure of fungi with ×40 magnification A: *Aspergillus niger*, B: *Aspergillus flavus*, C: *Penicillium Commune*, D: *Bipolaris sorokiniana*, E: *Curvularia eragrostidis*.

(Figure 3 and Figure 4). Based on the results of the Table 3 *Aspergillus niger* produced all urease, Mnp, lipase, laccase and protease enzymes. Qualitative results of the presence of enzymes in the studied isolates are shown in Table 3.

3.3 Identification of the selected isolate

The 560 bp region of the ITS gene was amplified by PCR. Laccase and manganese peroxidase gene expression, which are among the most important plastic degrading enzymes, were investigated in superior strains. The results can be seen in the figure 5.

To confirm the genus and species of strains containing plastic degrading enzymes, PCR was performed. Figure 6 shows the gel electrophoresis product.

3.4 Investigation of biodegradation of plastic compounds

To investigate the biodegradation of polyethylene and other compounds by fungal isolates from liquid and base media, M9 salt was used. Observation of turbidity and growth of fungal mycelium indicates the ability to consume compounds as the only carbon source in this environment was the small activity of the isolates by taking the light ab-

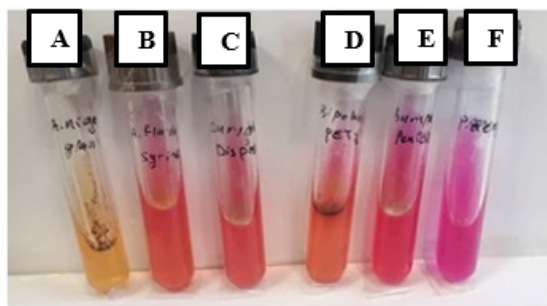


Figure 3. Qualitative study of urease enzyme in isolated strains. The strains are arranged from left to right as follows: A: *Aspergillus niger*, B: *Aspergillus flavus*, C: *Curvularia eragrostidis*, D: *Bipolaris sorokiniana*, E: *Penicillium commune*, F: *Pseudomonas aeruginosa*.

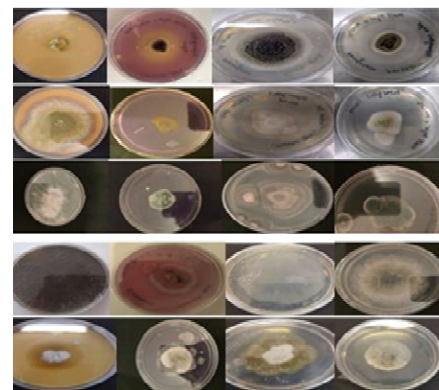


Figure 4. Qualitative study: Mnp enzyme, lipase, laccase, protease A: *Aspergillus niger*, B: *Aspergillus flavus*, C: *Penicillium commune*, D: *Bipolaris sorokiniana*, E: *Curvularia eragrostidis*.

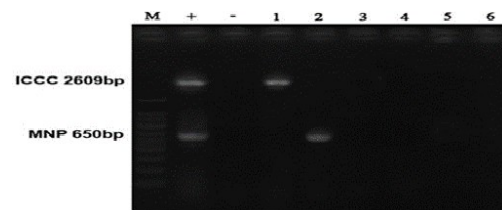


Figure 5. Molecular study of laccase and Mnp gene, *Aspergillus niger* laccase (+), *Aspergillus flavus* Mnp (+). lccC: laccase gene; MNP: Mnp.

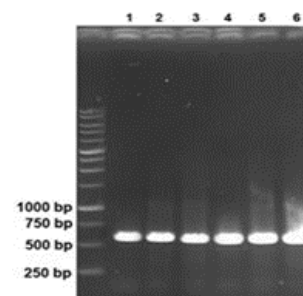


Figure 6. Gel electrophoresis PCR products (fungal strains).

Table 3. Investigation of enzymatic activity and degradation of plastic compounds by selected isolates.

| Name of fungi | Enzymes studied | | | | |
|--------------------------------|-----------------|-----|--------|---------|----------|
| | Urease | Mnp | Lipase | Laccase | Protease |
| <i>Aspergillus niger</i> | + | + | + | + | + |
| <i>Aspergillus flavus</i> | - | + | + | + | + |
| <i>Penicillium commune</i> | - | + | + | - | - |
| <i>Bipolaris sorokiniana</i> | + | + | - | - | - |
| <i>Curvularia eragrostidis</i> | - | + | + | - | + |

sorption of the samples and increasing the growth rate of isolates in the environment at different time intervals was determined.

3.5 Measure the remaining dry weight by weight loss percentage

The remaining plates were removed from the M9 salt base and washed. They were rinsed with a 2% SDS (sodium dodecyl sulfate) solution and then with deionized distilled water. Complete drying of the plates in weight loss percentage was calculated by the following formula (Table 4).

$$\text{Weight loss percentage (\%)} = \frac{\text{initial weight} - \text{final weight}}{\text{initial weight}} \times 100$$

3.6 Examination of surface changes created using electron microscopy (SEM)

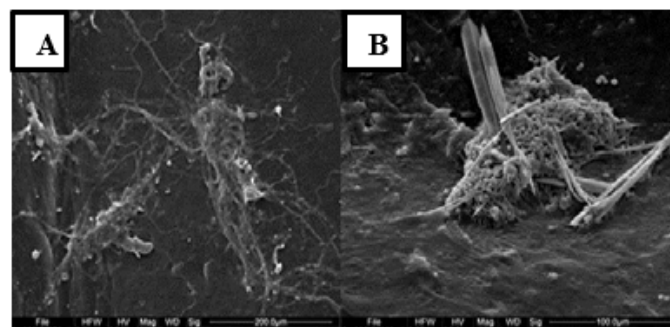
To study superficial and physical changes such as cracks and cavities as well as the formation of fungal mycelium electron microscopy was used on polyethylene and other compounds. Obtained images, badge provides surface changes as well as the formation of biofilm structures on the surface of polyethylene, and other compounds (Figure 7).

3.7 Investigation of chemical changes caused by biodegradation using FTIR infrared spectroscopy technique

Microorganisms such as fungi and bacteria cause chemical changes and break bonds are differentiated into polymeric compounds to investigate chemical changes caused by the FTIR technique. Fourier Transform Infrared Spectroscopy was used. This study examines the Chemical changes,

Table 4. Percentage of weight loss of compounds after 45 days of incubation with a suspension of fungal isolates.

| Fungi | Plastic type and weight loss percentage | | | | | | | |
|--------------------------------|---|-----|-----|-----|------------|-------------|-------------|--------------|
| | PE | PP | PET | PS | Nylon (PA) | PP (granul) | PS (granul) | PET (granul) |
| <i>Aspergillus niger</i> | 17.6 | 5.3 | 5.3 | 5.3 | 10 | 11.11 | 12.3 | 11.11 |
| <i>Aspergillus flavus</i> | 3.1 | 5.3 | 3.1 | 4.2 | 42 | 33.3 | 42 | 11.11 |
| <i>Penicillium commune</i> | 43 | 6.4 | 5.2 | 3.1 | 3 | 6.4 | 5.2 | 11.11 |
| <i>Bipolaris sorokiniana</i> | 25 | 5.3 | 3.1 | 5.3 | 11.11 | 11.11 | 25 | 11.11 |
| <i>Curvularia eragrostidis</i> | 43 | 2.1 | 2.1 | 11 | 25 | 2.1 | 3.1 | 2.1 |
| <i>Pseudomonas aeruginosa</i> | 12.4 | 3 | 3.6 | 5.2 | 5.2 | 11.11 | 11.11 | 4.1 |

**Figure 7.** Electron microscope images of a sample treated with A. *Aspergillus niger* biofilm, B. *Curvularia eragrostidis* after 45 days incubation, has a hole created in the surface, Magnification: 1000 K, resolution: 100 μ m.

bond failure and formation of new bonds in polypropylene and polyethylene compounds under fungal treatment. *Aspergillus niger* and *Curvularia eragrostidis* were evaluated for 45 days (Figure 8).

In polypropylene specimens treated with *Curvularia eragrostidis*, peaks related to C-H groups in the range of 603 cm^{-1} , C-H flexural 703 cm^{-1} , C = C 1646 cm^{-1} were removed. Group couriers O-H 3254 cm^{-1} and C = O 1722 cm^{-1} were formed in the treated samples. The peak of C-H aldehyde group was reduced in the treated polypropylene sample. The peak of S = O 1367 cm^{-1} , and 1462 cm^{-1} groups was increased in the treated polypropylene sample. In samples of propylene treated with fungi *Aspergillus niger*. Peaks related to groups C-X 603 cm^{-1} , C = C 1645 cm^{-1} were removed. Peak intensity increased in areas C-H 2917 cm^{-1} and 2346 cm^{-1} . In general, with attention to polypropylene samples treated with *Aspergillus niger*, *Curvularia eragrostidis* undergoes a chemical reaction that leads to the formation and removal of the index peak in the sample polypropylene (Figure 8A).

In polyethylene samples treated with fungus *C.eragrostidis* peak of bending C-H decreases at 728 cm^{-1} and peaks at C = O aldehyde and ester at 1728 cm^{-1} Increased. In treated specimens of *Aspergillus niger*, the C-H tensile peak de-

creased in 725 cm^{-1} area, C-H bending peak in the 1464 cm^{-1} area decreased, and O-H peak Carboxylic acid in the area of 2646 cm^{-1} shows a sharp decrease and a decrease in the peak in the area of 1728 cm^{-1} Is on interaction. Comparison between C-O and O-H groups between control polyethylene and polyethylene under treatment. The mentioned fungus can be concluded that no chemical reaction has occurred and only mixed the face has been on physical interaction (Figure 8B).

4. Discussion and conclusion

There are many studies in the field of biodegradation on fungal strains. Glen et al. (2021) examined the impact of the consortium *Aspergillus niger* (RVBT12), *A. flavus* (RV-BT43), *A. oryzae* (RV-BT117) in CBD medium instead of sucrose in the carbon medium of the substituted LDPE compounds was examined. After 40 days of incubation, SEM and FTIR techniques were used to prove biodegradation using. In another study, Manisha et al. (2019) isolated 109 different fungal isolates from 12 separate sites on the west coast of India, after 60 days Incubation of polyethylene compounds with isolates in sabouraud dextrose agar medium, isolates that most weight loss and tensile strength in polyethylene composition

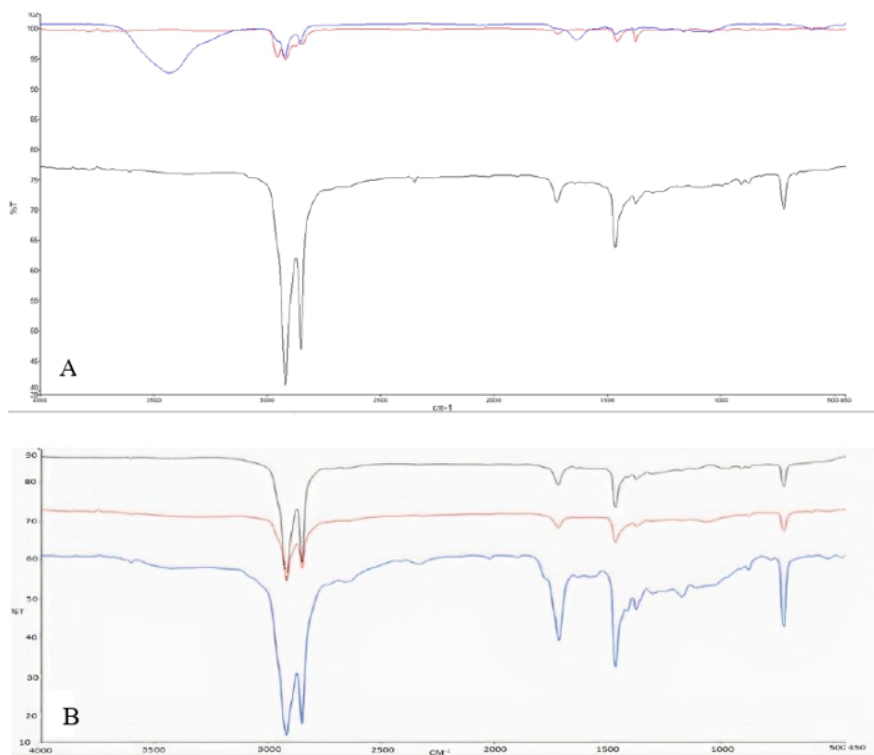


Figure 8. A. FTIR curve for syringe samples (polypropylene) under 45-day treatment with *Aspergillus niger*, *Curvularia eragrostidis* fungal suspension blue graphs for control samples (without fungal suspension), red graphs for samples treated with *Curvularia eragrostidis*, black graphs of samples treated with suspension *Aspergillus niger* After 45 days of incubation. B. FTIR curve for polyethylene samples under 45-day treatment with fungal suspension *Aspergillus niger*, *Curvularia eragrostidis*.

were selected. Isolates selected are: *Aspergillus terreus* / MANGF1, *Aspergillus sydowii* / PNP15. Use of SEM Imaging Electron Microscope FTIR Analysis of Physical and Chemical decomposition spectroscopy isolates were examined. A study was performed by , during which fungus *Pleurotus ostreatus* was isolated due to its ability to produce the enzymes laccase, Mnp and lignin peroxidase are parsers of LDPE pages [24]. Experiment by Anudurga et al. (2016) Fungal strain *Aspergillus clavatus* was isolated from a landfill. After 90 days of incubation in saline medium and the fungal strain, 35% weight loss of the compounds was reported. Shimao et al. (2001) showed *Phanerochaete chrysosporium*, *Fusarium*, *Amycolaptosis* sp, and bacteria *Pseudomonas stutzeri* are capable of producing compounds by producing the enzymes' Mnp, cutinase and serine hydrolase that decompose polyethylene, polylactic acid, polycaproatone, polyhydroxy alkanooate.

In this study, several fungal strains were isolated from landfills and their ability to produce extracellular enzymes including lactase, Mnp, lipase, protease, and urease was investigated. *Aspergillus niger*, *Aspergillus flavus*, *Bipolaris sorokiniana*, and *Curvularia eragrostidis* were used to study the biodegradation of polyethylene compounds, polyethylene terephthalate, polypropylene, polystyrene, and polyamide for the first time in Iran. This research can help solve the problem of accumulation and pollution in the environment by producing intermediate products such as alcohol, ketones, aldehydes, ethers, oxides and esters, which are used in industry, medicine and agriculture. On the other hand, many of these fungi are microflora of the same wastes, and due to their biocompatibility, not only is there no need to provide special conditions for plastic decomposition, but they have a high resistance to toxic compounds in the wastes. The widespread production of fungi decomposing enzymes is the most important parameter for the success of fungi in decomposing waste and turning them into useful compounds such as Scp in future research.

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Author contributions

Conceptualization: [M.Larypoor]; Methodology: [M.Larypoor and F.Rahmati]; Formal analysis and investigation: [M. Larypoor and Elham Taghinia]; Writing - original draft preparation: [M.Larypoor and Elham Taghinia]; Writing - review and editing: [M.Larypoor]; Supervision: [M. Larypoor].

Availability of data and materials

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Competing Interests

The authors have no conflicting interests to declare.

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