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Biodegradation of polycyclic aromatic hydrocarbons in Basrah light crude oil by *Aspergillus terreus*

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Abstract.
This study investigated the ability of the fungus Aspergillus terreus which isolated from highly oil contaminated soil to biodegrade polycyclic aromatic hydrocarbons (PAHs) of Basrah light crude oil, as well as the effect of several parameters on the biodegradation ability of this fungus. The results showed that after 15 days of incubation A. terreus was able to degrade 62.5% of the crude oil and this percentage raises to 85.71% after 30 days of incubation. The results of GC-MC revealed that A. terreus had a good ability to degrade PAHs and the results of residual total PAH compounds was 300.8121 µg/ml after 15 days of incubation and decreased to 220.0279 µg/ml after 30 days of incubation compared with control 605.2314 µg/ml, with the complete degradation of 2 compounds. The results showed that temperature 30°C and pH of 5.5 were the best for degradation process. Finally, the ability of A. terreus to degrade PAH compounds under these optimum conditions was studied. The results showed that after 7 days of incubation the percentage of degradation reached 56.25% and the total PAH concentrations decreased to 231.6025 µg/ml with the complete degradation of two aromatic compounds. After 15 days of incubation the percentage of degradation increased to 71.42% and the concentration of PAHs decreased to 88.1211µg/ml. This study revealed that the fungus A. terreus is a potential tool for the degradation of PAH compounds.
Key words: Biodegradation, Contaminated soils, Light Crude oil, PAHs.

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1. Introduction

Crude oil and its derivatives are one of the most important energy resources all over the world today, due to depletion of light oil resources and increase in energy demand, the primary focus is now shifting towards utilizing heavy oil reserves (Al-Saad *et al*., 2010;Al-Sayegh *et al*., 2015). Crude oil is a complex mixture of a diverse compounds that can be divided into four main classes: saturated hydrocarbons, aromatic hydrocarbons, resins, and asphaltenes (Liu *et al*., 2014). Polycyclic aromatic hydrocarbons (PAHs) are a class of organic pollutants of major concern to the environment and public health because they are genotoxic, mutagenic, carcinogenic, and have strong immunosuppressive qualities, (Abdullatif *et al*., 2016; Al-Dossary *et al*., 2020; Khanpour-Alikelayeh *et al*., 2021). The United States Environmental Protection Agency (US EPA) has already recognized 16 PAHs as priority environmental pollutants based on their abundance and toxicity (Dai *et al*., 2022). They can potentially accumulate in the environment to substantial levels. owing to their nature, PAHs may negatively affect the physicochemical properties of soil, including texture and water holding capacity, they can also migrate into the groundwater and enter the food chain, with severe consequences to living organisms, PAHs are removed from the environment by several methods including biodegradation which is a natural process uses microbial consortium to degrade PAHs in the environment, they use the ability of microorganisms to degrade pollutants and transform them into less toxic products than their parent compounds (Al-Dhabaan *et al*., 2021; Al-Dossary *et al*., 2019; Thacharodi *et al*., 2023). Fungi possess certain advantages over bacteria, owing to the nature of their growth patterns and their resistance to PAHs, as demonstrated by several studies (Hassanshahian *et al*., 2012; Abdel-Shafy and Mansour, 2016; Al-Hawash *et al*., 2018). Filamentous fungi have been readily isolated from oil contaminated soils and have demonstrated their ability to degrade PAHs. *Aspergillus terreus* is one of the most commonly isolated fungi which has

especially owing to the mutagenic or carcinogenic properties of some PAHs (Govarthanan *et al*., 2017; Adenji *et al*., 2019).

recently received more attention as potential tools for degradation of different PAH compounds (Lotfinasabasl *et al*., 2012; Al-Harbi *et al*.,2022).

The present study aimed to determine the ability of the fungus *Aspergillus terreus* which isolated from oil-contaminated soils in Basrah province to degrade PAH compounds of light crude oil and to study the effects of varying temperature and pH on the degradation process.

2. Material and methods

2.1. Samples collection

Ten oil-contaminated soil samples were obtained from the surface layer (5–10 cm) from several oil fields in Basrah province, southern Iraq from September 2021 until November 2021. Soil samples were collected and stored in sterile plastic bags at 4°C until further use (Latha and Kalaivani,2012).

2.2. Chemicals

2.2.1. Crude oil

Light crude oil was purchased from southern oil company (Basra, Iraq), the oil was kept in a cold and dark place after being transferred to the laboratory in a tightly closed sterilized dark bottle.

2.2.2. Media

All chemicals and media were obtained from (Hi-Media company, India).

2.3. Isolation of hydrocarbon degrading fungi

Fungi were isolated from oil-contaminated soil using the dilution method described by Wicklow and Wittingham (1974); 10 g of each soil sample was dissolved in 90 ml distilled water to attain a dilution of 10^{-1} and shaken well for 10 min. A sterile pipette was used to transfer 1 ml of each dilution to a sterile petri dish. For primary isolation of fungi, one type of media used potato dextrose agar (PDA). PDA medium was prepared in accordance with the direction of the manufacturing company (Hi-Media, India). The antibiotic chloramphenicol (250 mg/l) was supplemented to the medium to inhibit bacterial growth. Approximately 15 mL of each sterile medium was added separately to the petri dish containing 1 ml from the diluted sample. The petri dishes were stirred to mix the ingredients well before solidification. Then, the petri dishes were incubated at 25 °C for 7–14 days. Each colony that appeared after incubation was sub-cultured separately on PDA medium. The pure cultures were preserved on PDA medium at 4 °C for further identification.

2.4. PAHs degradation by single fungal isolate

The fungal isolate which was used to test its ability to degrade light crude oil was first activated on PDA medium for 7 days. Mineral salts broth medium composition (g

L−1): $NaNO_3$, 0.42; NaCl, 10.0; $MgSO_4.7H_2O$, 0.42; KCl, 0.12; K_2HPO_4, 0.83; Na_2HPO_4, 1.25 was used in the biodegradation experiments. Two plugs were collected using a 5-mm corn borer from the periphery of the selected fungus colony and added to 250-mL conical flasks, each containing 100 ml of mineral salts medium (MSM) (pH 4.5). Light Crude oil (1 ml) was added to each flask as the sole carbon source. The culture flasks were incubated at 25°C in a combined rotary shaker incubator maintained at 115 rpm for 15 and 30 days. Uninoculated medium containing only light crude oil served as a control. All experiments were carried out in triplicate (Okerentugba and Ezeronye, 2003).

2.5. Effect of some physicochemical parameters on the biodegradation process

The effects of some parameters on fungal biodegradation ability were evaluated, according to the procedure described by Hamzah *et al*. (2012) as follows:

2.5.1. Incubation temperature

Two sets of flasks, containing the fungal isolate, the same (MSM) medium as previously described, and heavy crude oil (pH 4.5), were prepared. One set was incubated at 30°C and the other at 35°C, in a combined rotary shaker incubator maintained at 115 rpm for 15 and 30 days. Uninoculated medium containing only light crude oil. served as a control. All the experiments were carried out in triplicate.

2.5.2. pH

The effects of pH on the biodegradation process were evaluated at two different pH values. The pH of the growth medium in one set of flasks was adjusted to 5.5 and the other set to 6.5. Uninoculated medium containing only light crude oil served as a control. The flasks were incubated, in triplicate, at 25°C in a combined rotary shaker incubator maintained at 115 rpm for 15 and 30 days.

2.5.3. Effect of biostimulation on PAHs biodegradation

To evaluate the outcome of biostimulation on the PAHs degradation process by single fungal isolates, 250-ml conical flasks, each containing 100ml of MSM medium, were maintained under the optimal temperature and pH, determined from the previous experiments. Each flask was inoculated with the best fungal candidate. Control flasks containing uninoculated medium with only light crude oil were used. The flasks were set up, in triplicate, for two incubation periods, 7 and 15 days, in a combined rotary shaker incubator maintained at 115 rpm and 30°C.

2.6. Extraction and analysis of PAHs

After the completion of each experiment, the contents in each flask were filtered by GFF filter papers using filtration units, the filtrate was taken and transferred to a separating funnel. Then the filtrate was extracted with 100 ml Chloroform in a separating funnel. with continuous shaking, this process was repeated three times. The liquid fraction was left for a period of time until it settled, yielding two layers: an aqueous layer and an organic layer containing residual light crude oil. The organic layer was transferred to a clean pre-weighed flasks and air dried, then weighed again and the percentage of degrading was measured as follows Outdot (1984):

$$Degradation\% = \frac{\text{mg of crude oil control} - \text{mg of crude oil test}}{\text{mg of crude oil control}} * 100$$

The aromatic fraction was separated by passing the extracted crude oil through a silica gel column. The air-dried samples were redissolved in 5ml of n-hexane and the soluble samples were passed through a silica column. The PAH fraction was separated with 30ml of benzene. To identify PAH compounds, the samples were evaporated to dryness using a rotary evaporator and redissolved in 5ml of the solvent, then injected into gas chromatograph- Mass Spectrometer (GC-Ms).

2.7. Statistical analysis

Minitab version 16 software was used to analyze the results using one-way analysis of variance. Relative least significant difference values were calculated to determine significant differences between the fungal processes. A completely randomized design was used.

3. Results and Discussion

3.1. PAHs biodegradation using *Aspergillus terreus*

Twenty fungal species, belonging to the 9 genera were identified. Eight of these species belonged to the genus *Aspergillus*. The fungus *Aspergillus terreus*, which showed the higher percentage of appearance during the isolation, was selected to find out its efficiency in biodegrading the aromatic compounds of light crude oil in liquid media and in the form of single isolate with two incubation periods of 15 and 30 days. As the fungus showed a good ability to degrade light crude oil, this degrading was observed through some changes, including the good growth of fungal hyphae in the liquid medium and in the form of mycelial attached to the crude oil, and the biomass of the fungal hyphae increased over time. The results of the current study showed that the tested fungus proved its ability to biodegrade light crude oil, in which the percentage of biodegrading of light crude oil recorded 62.5% after 15 days of the incubation period, and it was increased to 85.71% after 30 days of the incubation period (Figure 1). The results of the statistical analysis showed that there were significant differences ($P<0.01$) in the percentage of crude oil degradation of 15 and 30 days.

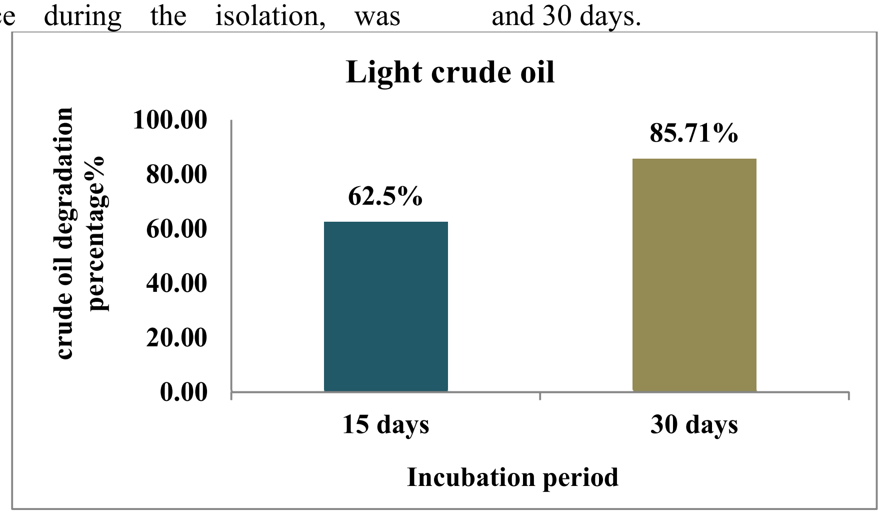

Figure 1: percentage of light crude oil biodegradation by *Aspergillus terreus*

The results of gas chromatography analysis for PAHs compounds extracted from liquid growth cultures showed that the fungus *A. terreus* had a good ability to biodegrade aromatic compounds. The total residual concentrations of aromatic compounds reached 300.812 µg/ml after the end of the 15-day incubation period, and continued to decreased to reach 220.0279µg/ml after the end of the 30-day incubation period. In the present study, a significant number of total PAHs was degraded after 15 and 30 days of incubation compared with the control treatment 605.2314 µg/ml (Table 1& Figure 2).

The results of the statistical analysis showed that there were significant differences ($P<0.01$) in the degradation percentage of PAHs between the two incubation periods of 15 and 30 days and compared with control treatment. Also *A. terreus* was able to completely degrade 2 aromatic compounds (Naphthalene, Fluorene) After 30 days of incubation.

Table 1: Residual concentrations of PAHs for light crude oil in the liquid medium culture of *Aspergillus terreus*

PAHs compounds	Control µg/ml	PAHs compounds	Concentrations µg/ ml/After 15 days	PAHs compounds	Concentrations µg/ml/After 30 days
NAPHTHALENE	14.0912	NAPHTHALENE	4.2109	NAPHTHALENE	0.0000
2-METHYLNAPHTHA	267.1768	2-METHYLNAPHTHA	58.1698	2-METHYLNAPHTHA	49.9172
1-METHYLNAPHTHA	30.3374	1-METHYLNAPHTHA	11.9176	1-METHYLNAPHTHA	8.5857
ACENAPHTHYENE	23.5868	ACENAPHTHYENE	14.0000	ACENAPHTHYENE	13.2114
ACENAPHTHNEN	22.2212	ACENAPHTHNEN	22.0662	ACENAPHTHNEN	17.0213
FLUORENE	63.2133	FLUORENE	0.0000	FLUORENE	0.0000
PHENANTHRENE	32.0006	PHENANTHRENE	35.4997	PHENANTHRENE	23.6992
ANTHRACENE	29.4746	ANTHRACENE	21.4132	ANTHRACENE	16.4264
FLUORANTHENE	32.4746	FLUORANTHENE	11.2936	FLUORANTHENE	9.6211
PYRENE	15.4139	PYRENE	28.4295	PYRENE	20.8044
CHRYSENE	17.8179	CHRYSENE	16.0025	CHRYSENE	12.03209
BENZO(A)ANTHRACENE	6.1380	BENZO(A)ANTHRACENE	9.6660	BENZO(A)ANTHRACENE	8.9251
BENZO(B) FLUORANTHENE	10.0310	BENZO(B) FLUORANTHENE	20.1141	BENZO(B) FLUORANTHENE	13.9512
BENZO(K) FLUORANTHENE	14.4221	BENZO(K) FLUORANTHENE	12.4972	BENZO(K) FLUORANTHENE	5.6656
BENZO(A) PYRENE	.113946	BENZO(A) PYRENE	21.8925	BENZO(A) PYRENE	14.0000
INDENO(1,2,3-CD) PYRENE	15.4374	INDENO(1,2,3-CD) PYRENE	13.6393	INDENO(1,2,3-CD) PYRENE	8.3819
BENZO (G, H, I) PERYLENE	0.0000	BENZO (G, H, I) PERYLENE	0.0000	BENZO (G, H, I) PERYLENE	0.0000
Total	605.2314	Total	300.8121	Total	220.0279

Control 15 days 30 days

Figure 2: **The ability of *Aspergillus terrues* to degrade light crude oil in liquid medium after 15 and 30 days of incubation compared with the control**.

Fungi have a higher tolerance to the toxicity of hydrocarbons owing to their physiology, allowing them to exhibit an effective ability for degradation. The ability of this species to produce more than one type of degradative enzyme in high quantities may increase its potential to degrade PAH compounds, one of the reasons that led to its high ability to biodegrade aromatic compounds is that the fungus *A. terreus* has an active and efficient enzyme system and its high ability to adapt and consume oil compounds (Mohsenzadeh *et al*., 2012; Barnes *et al*., 2018; Hassaine and Bordjiba, 2019). These results are consistent with those of other studies in which fungi especially the genus *Aspergillus* has a great degradation ability for hydrocarbons Al-Hawash *et al*. (2018), Ponnapalli *et al*. (2018), Usman and Tijani. (2020), Al-Dossary *et al*. (2020) and Al-Harbi *et al*. (2022).

3.2. Optimization of PAHs degradation using *Aspergillus terreus*

The fungus *A. terreus* was chosen to investigate the effects of temperature and pH, on the biodegradation process of PAHs, for a period of 15 and 30 days. One of the most important parameters that can affect fungal growth is temperature (Delille *et al*., 2004). In the present study, *A. terreus* grew over a range of temperatures, 25–35°C, with the optimal temperature for growth and degradation being 30°C. At 30°C, the total concentration of PAH compounds decreased considerably from an initial concentration of 605.2314 µg/ml for control to 232.1224 µg/ml after 15 days and also continued to decreased to 182.6342 µg/ml after 30 days of incubation period (Table 2). The percentage of degradation represents 57.14% at 30°C after 15 days and raises to 85.71% after 30 days of incubation (Figure 3). The temperature 35°C effect negatively on the degradation process, in which the percentage of degradation for light crude oil recorded 14.28% after 15 days of incubation and 42.85% after 30 days, also the residual total of PAHs recorded 416.9232µg/ml after 15 days and decreased to 284.1441 after 30 days of incubation.This finding is consistent with that of several studies Teerapatsakul *et al*.(2016), Al-Hawash *et al*. (2018) , Al-Dossary *et al*. (2020), which reported that the optimum temperature for fungal growth during the oil degradation processes was 30°C. This may be because of an increase in fungal growth, which potentially improved energy efficiency in the system by facilitating contact between the cell surfaces and the energy source. However, at higher temperatures, this process may slow down, perhaps owing to the deformation of cell surfaces available as sites for substrate attachment (Salam *et al*., 2011; Al-Asheh and Duvnjak, 2018; DSouza *et al*., 2021).

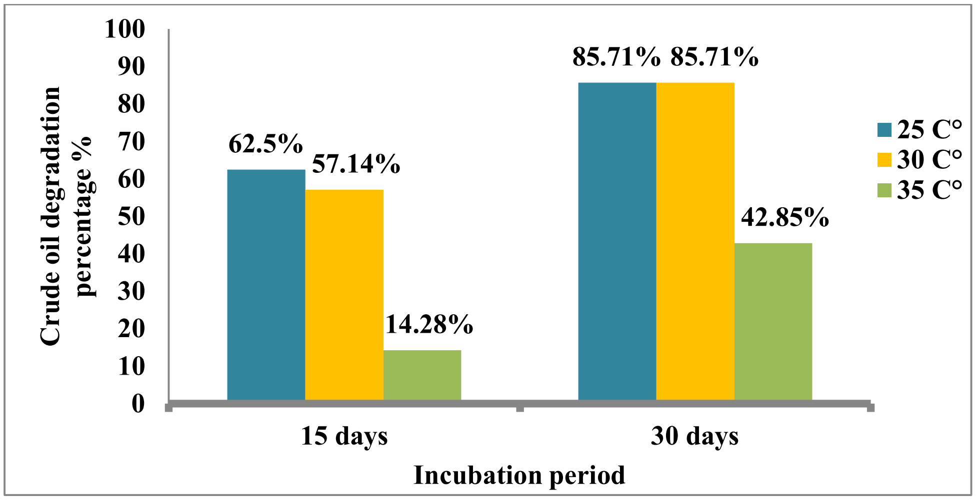

Figure 3: Percentage of biodegrading of light crude oil under different temperatures by *Aspergillus terreus*

Table 2: Effect of temperature at 30°C on biodegradation of PAHs by *Aspergillus terreus*

PAHs compounds	Control µg/ml	PAHs compounds	Concentrations µg/ ml/After 15 days	PAHs compounds	Concentrations µg/ml/After 30 days
NAPHTHALENE	14.0912	NAPHTHALENE	0.0000	NAPHTHALENE	0.0000
2-METHYLNAPHTHA	267.1768	2-METHYLNAPHTHA	48.4438	2-METHYLNAPHTHA	41.5670
1-METHYLNAPHTHA	30.3374	1-METHYLNAPHTHA	6.5096	1-METHYLNAPHTHA	6.1281
ACENAPHTHYENE	23.5868	ACENAPHTHYENE	17.3355	ACENAPHTHYENE	16.1453
ACENAPHTHNEN	22.2212	ACENAPHTHNEN	21.5990	ACENAPHTHNEN	20.8837
FLUORENE	63.2133	FLUORENE	0.0000	FLUORENE	0.0000
PHENANTHRENE	32.0006	PHENANTHRENE	31.6871	PHENANTHRENE	30.4226
ANTHRACENE	29.4746	ANTHRACENE	24.6792	ANTHRACENE	22.6843
FLUORANTHENE	32.4746	FLUORANTHENE	10.3557	FLUORANTHENE	8.2473
PYRENE	15.4139	PYRENE	14.2189	PYRENE	9.0204
CHRYSENE	17.8179	CHRYSENE	8.9602	CHRYSENE	6.9739
BENZO(A)ANTHRACENE	6.1380	BENZO(A)ANTHRACENE	6.3508	BENZO(A)ANTHRACENE	4.8113
BENZO(B) FLUORANTHENE	10.0310	BENZO(B) FLUORANTHENE	10.8774	BENZO(B) FLUORANTHENE	0.3546
BENZO(K) FLUORANTHENE	14.4221	BENZO(K) FLUORANTHENE	12.7511	BENZO(K) FLUORANTHENE	0.9509
BENZO(A) PYRENE	.113946	BENZO(A) PYRENE	8.6712	BENZO(A) PYRENE	7.3788
INDENO(1,2,3-CD) PYRENE	15.4374	INDENO(1,2,3-CD) PYRENE	9.6829	INDENO(1,2,3-CD) PYRENE	7.0660
BENZO (G, H, I) PERYLENE	0.0000	BENZO (G, H, I) PERYLENE	0.0000	BENZO (G, H, I) PERYLENE	0.0000
Total	605.2314	Total	232.1224	Total	182.6342

Fungi favor acidic conditions for growth (Rousk *et al*., 2009). In this experiment the optimum pH was 5.5, with a percentage of degradation reached to 62.5% after 15 days and continued to raises to 85.71% after 30 days of incubation for the heavy crude oil. The total PAHs concentration decreasing considerably to 435.939µg/ml after 15 days and decreased to 228.0279µg/ml after 30 days of incubation (Table 3 & Figure 4). While pH 6.5 affect negatively on the degradation process, the percentage of degrading of light crude oil recorded 28.75% after 15 days and decreased to 28.57% after 30 days, also the total PAHs concentrations recorded 512.0867µg/ml after 15 days of incubation and decreased to 330.5147 µg/ml after 30 days, probably because an increase in hydroxyl ions in the culture media which inhibited the adsorption of PAHs to cation-binding sites (Jalali *et al*., 2002).

Similar results were reported by Hamzah *et al*. (2012), Al-Dossary *et al*. (2020) who illustrated that culture conditions directly affected microorganism growth and the degradation process. There were significant differences (*P* <0.01) in the PAHs concentrations recorded under conditions of varying temperatures and pH and between 15 and 30 days of incubation period.

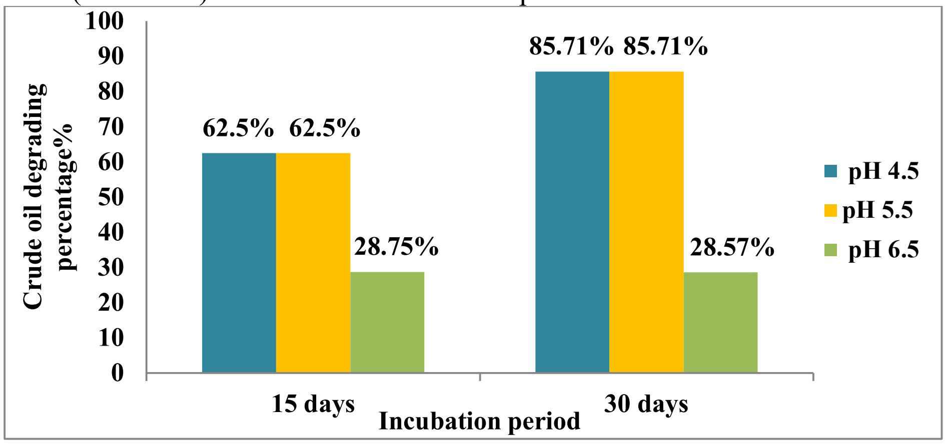

Figure 4: Percentage of biodegrading of light crude oil under different pH value by *Aspergillus terre*

Table 3: Effect of potential of Hydrogen ions at the pH 5.5 on biodegradation of PAHs by *Aspergillus terreus*

PAHs compounds	Control µg/ml	PAHs compounds	Concentrations µg/ ml/After 15 days	PAHs compounds	Concentrations µg/ ml/After 30 days
NAPHTHALENE	14.0912	NAPHTHALENE	12.6649	NAPHTHALENE	0.0000
2-METHYLNAPHTHA	267.1768	2-METHYLNAPHTHA	202.5626	2-METHYLNAPHTHA	98.6451
1-METHYLNAPHTHA	30.3374	1-METHYLNAPHTHA	26.7850	1-METHYLNAPHTHA	24.5437
ACENAPHTHYENE	23.5868	ACENAPHTHYENE	28.1652	ACENAPHTHYENE	18.7508
ACENAPHTHNEN	22.2212	ACENAPHTHNEN	18.7594	ACENAPHTHNEN	0.0000
FLUORENE	63.2133	FLUORENE	0.0000	FLUORENE	0.0000
PHENANTHRENE	32.0006	PHENANTHRENE	35.7733	PHENANTHRENE	16.9543
ANTHRACENE	29.4746	ANTHRACENE	18.0786	ANTHRACENE	9.1398
FLUORANTHENE	32.4746	FLUORANTHENE	22.0451	FLUORANTHENE	7.7489
PYRENE	15.4139	PYRENE	9.4939	PYRENE	6.9814
CHRYSENE	17.8179	CHRYSENE	17.1340	CHRYSENE	8.2892
BENZO(A)ANTHRACENE	6.1380	BENZO(A)ANTHRACENE	6.2480	BENZO(A)ANTHRACENE	4.1470
BENZO(B) FLUORANTHENE	10.0310	BENZO(B) FLUORANTHENE	9.4310	BENZO(B) FLUORANTHENE	8.4347
BENZO(K) FLUORANTHENE	14.4221	BENZO(K) FLUORANTHENE	2.6806	BENZO(K) FLUORANTHENE	0.9425
BENZO(A) PYRENE	.113946	BENZO(A) PYRENE	11.3835	BENZO(A) PYRENE	7.3896
INDENO(1,2,3-CD) PYRENE	15.4374	INDENO(1,2,3-CD) PYRENE	14.7339	INDENO(1,2,3-CD) PYRENE	8.0609
BENZO (G, H, I) PERYLENE	0.0000	BENZO (G, H, I) PERYLENE	0.0000	BENZO (G, H, I) PERYLENE	0.0000
Total	605.2314	Total	435.939	Total	228.0279

3.3. Biostimulation of PAHs degradation

The biostimulation process involved the adjustment of environmental conditions to stimulate the growth and enzymatic activity of the existing microorganisms. The optimum conditions, that is, 30°C and pH 5.5, were used together to study their effect on PAHs degradation by *Aspergillus terreus*. The results showed that after 7 days of incubation period the degradation percentage were 56.25% and the total PAHs concentrations decreased to 231.6025µg/ml. A high biodegradation efficiency was achieved with biostimulation (Figure 5), where total PAH compounds concentrations decreased to 231.6025 µg/ml in just 7 days which is less than the concentration of the residual PAHs by the same fungus in 15 days without biostimulation. This suggests that subjecting the fungi to the optimum temperature and pH resulted in higher enzymatic activity and growth, with a higher number of fungal mycelia coming in contact with the heavy crude oil, thereby enhancing the degradation process more readily (Garapati and Mishra, 2012; Zeneli *et al*.,2019; Al-Dossary *et al*., 2020; Sayed *et al*., 2022). The results of the current study were consistent with those of several earlier studies Garapati and Mishra, (2012), Kuappi *et al*. (2011), Wu *et al*. (2016). A higher degradation efficiency was observed after 15 days of incubation, with the PAH concentrations decreasing to 88.1211µg/ml (Table 4), with a degradation percentage of 71.42% (Figure 5). There were significant differences ($P < 0.01$) in the degradation percentage between the fungus in this experiment and that in the previous experiment without biostimulation. Several reports state that the degradation process was maximized once optimal conditions for growth were achieved Govarthanan *et al*. (2017), Wu *et al*. (2016), Al-Dossary *et al*. (2020).

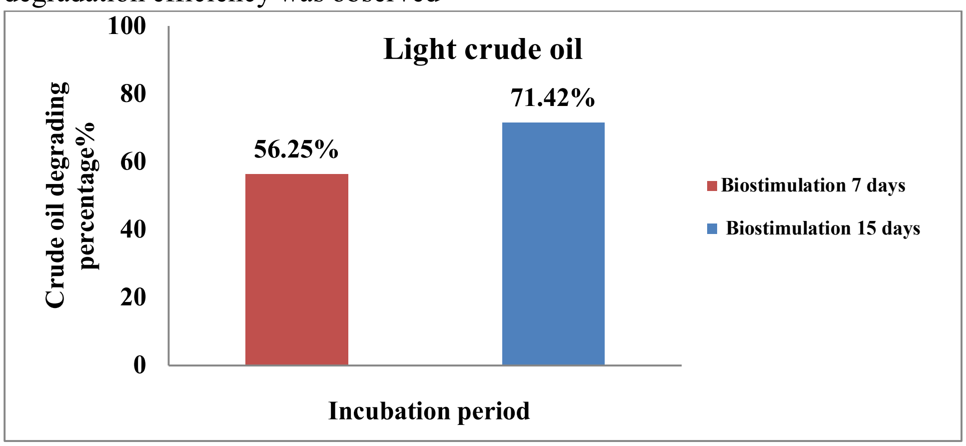

Figure 5: Percentage of biodegrading for light crude oil under biostimulation by *Aspergillus terreus*

Table 4: Effect of biostimulation on biodegradation of PAHs by *Aspergillus terreus*

PAHs compounds	Control µg/ml	PAHs compounds	Concentrations µg/ ml/After 7 days	PAHs compounds	Concentrations µg/ ml/After 15 days
NAPHTHALENE	14.0912	NAPHTHALENE	11.1860	NAPHTHALENE	0.0000
2-METHYLNAPHTHA	267.1768	2-METHYLNAPHTHA	39.0718	2-METHYLNAPHTHA	17.6396
1-METHYLNAPHTHA	30.3374	1-METHYLNAPHTHA	13.0409	1-METHYLNAPHTHA	2.4113
ACENAPHTHYENE	23.5868	ACENAPHTHYENE	27.8668	ACENAPHTHYENE	11.4432
ACENAPHTHNEN	22.2212	ACENAPHTHNEN	21.5079	ACENAPHTHNEN	2.0756
FLUORENE	63.2133	FLUORENE	13.7005	FLUORENE	5.3009
PHENANTHRENE	32.0006	PHENANTHRENE	29.2526	PHENANTHRENE	3.8614
ANTHRACENE	29.4746	ANTHRACENE	3.3988	ANTHRACENE	0.0000
FLUORANTHENE	32.4746	FLUORANTHENE	7.5819	FLUORANTHENE	5.0614
PYRENE	15.4139	PYRENE	13.8913	PYRENE	11.3882
CHRYSENE	17.8179	CHRYSENE	15.1687	CHRYSENE	8.2750
BENZO(A)ANTHRACENE	6.1380	BENZO(A)ANTHRACENE	5.1108	BENZO(A)ANTHRACENE	2.6951
BENZO(B) FLUORANTHENE	10.0310	BENZO(B) FLUORANTHENE	5.5669	BENZO(B) FLUORANTHENE	4.0641
BENZO(K) FLUORANTHENE	14.4221	BENZO(K) FLUORANTHENE	0.3555	BENZO(K) FLUORANTHENE	1.4204
BENZO(A) PYRENE	.113946	BENZO(A) PYRENE	11.0665	BENZO(A) PYRENE	10.2448
INDENO(1,2,3-CD) PYRENE	15.4374	INDENO(1,2,3-CD) PYRENE	13.8356	INDENO(1,2,3-CD) PYRENE	2.2401
BENZO (G, H, I) PERYLENE	0.0000	BENZO (G, H, I) PERYLENE	0.0000	BENZO (G, H, I) PERYLENE	0.0000
Total	605.2314	Total	231.6025	Total	88.1211

4. Conclusion

The fungus *Aspergillus terreus* exhibited a high degradation potential under laboratory conditions, with the ability to decreased the total PAHs of light crude oil reached to 88.1211µg/ml after 15 days of incubation. Biostimulation under the optimum temperature and pH, conditions stimulated the degradation process to a higher degree. In addition, the incubation period also affected the biodegradation process, with approximately 71.42% of light crude oil being degraded after 15 days of incubation. Our study indicated that *A. terreus* is a potential biodegrading agent that can be utilized in the bioremediation of PAHs in the environment.

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References

Abdullatif, S.A.; Al-Khatib, F.M. and Al-Adhub, A. (2016). The fluctuations in concentrations, origins and sources of Polycyclic Aromatic Hydrocarbons (PAHs) in water (dissolved and particulate fractions) of the Shatt al Arab River and its branches/Basra-Iraq. IJIET., 6(3):552-555.

Abdel-Shafy, H. I. and Mansour, M. S. (2016). A review on polycyclic aromatic hydrocarbons: source, environmental impact, effect on human health and remediation. Egypt. J. Pet., 25(1): 107-123.

Adeniji, A.; Okoh, O. and Okoh, A. (2019). Distribution pattern and health risk assessment of polycyclic aromatic hydrocarbons in the water and sediment of Algoa bay, South Africa. Environ. geochemist. Health., 41(3): 1303-1320.

Al-Dhabaan, F. A. (2021). Isolation and identification of crude oil-degrading yeast strains from Khafji oil field, Saudi Arabia. Saudi J. Biol. Sci., 28(10): 5786-5792.

Al-Saad, H. T.; Al-khatib, F.M. and Al-Adhab, A.A. (2010). Determination the concentration and origin of poly cyclic aromatic hydrocarbon compounds (PAHs) in water and sediments of AL–Howaiza marsh. Basrah J. Sci., 28(2). 146-154.

Al-Dossary, M. A.; Abood, S. A. and AL-Saad, H. T. (2019). Biodegradation of crude oil using *Aspergillus* species. Biodegradation., 9(4):2224-3208.

Al-Dossary, M. A.; Abood, S. A. and Al-Saad, H. T. (2020). Factors affecting polycyclic aromatic hydrocarbon biodegradation by *Aspergillus flavus*. Rem. J., 30(4): 17-25.

Al-Harbi, N. K.; Alzaban, M. I.; Albarakaty, F. M.; Abd El-Aziz, A. R.; AlRokban, A. H. and Mahmoud, M. A. (2022). Transcriptome Profiling Reveals Differential Gene Expression of Laccase Genes in *Aspergillus terreus* KC462061 during Biodegradation of Crude Oil. Biol, 11(4), 564.

Al-Hawash, A. B.; Dragh, M. A.; Li, S.; Alhujaily, A.; Abbood, H. A.; Zhang, X. and Ma, F. (2018). Principles of microbial degradation of petroleum hydrocarbons in the environment. Egypt. J. Aquat. Res., 44(2): 71-76.

Al-Sayegh, A.; Al-Wahaibi, Y.; Al-Bahry, S.; A. Elshafie, A.; Al-Bemani, A. and Joshi, S. (2015). Microbial enhanced heavy crude oil recovery through biodegradation using bacterial isolates from an Omani oil field. Microb., 14, 141.

Al-Asheh, S.; and Duvnjak, Z. (2018). Adsorption of cooper and chromium by *Aspergillus carbonarius*. Biotechnology Progress, 11, 638–642. Barnes, N.M.; Khodse, V.B.; Lotlikar, N.P.; Meena, R.M. and Damare, S.R. (2018). Bioremediation potential of hydrocarbon-utilizing fungi from select marine niches of India. 3 Biotech, 8, 21.

Dai, C.; Han, Y.; Duan, Y.; Lai, X.; Fu, R.; Liu, S. and Zhou, L. (2022). Review on the contamination and remediation of polycyclic aromatic hydrocarbons (PAHs) in coastal soil and sediments. Environ. Res., 205:112423.

Delille, D.; Coulon, F. and Pelletier, E. (2004). Effects of temperature warming during a bioremediation study of natural and nutrient amended hydrocarbon‐contaminated sub-Antarctic soils. Cold Reg Sci Technol., 40: 61–70.

DSouza, G. C.; Sheriff, R. S.; Ullanat, V.; Shrikrishna, A.; Joshi, A. V.; Hiremath, L. and Entoori, K. (2021). Fungal biodegradation of low-density polyethylene using consortium of *Aspergillus* species under controlled conditions. Heliyon., 7(5): 1-7.

Garapati, V. K. and Mishra, S. (2012). Hydrocarbon degradation using fungal isolate: Nutrients optimized by combined grey relational analysis. Int. j. eng. res. appl., 2: 390–399.

Govarthanan, M.; Fuzisawa, S.; Hosogai, T. and Chang, Y.C. (2017). Biodegradation of aliphatic and aromatic hydrocarbons using the filamentous fungus *Penicillium* sp. CHY‐2 and characterization of its manganese peroxidase activity. RSC Advances., 34: 20716–20723.

Hamzah, A.; Manikan, V. and Abd Aziz, N.A.F. (2017). Biodegradation of tapis crude oil using consortium of bacteria and fungi: Optimization of crude oil concentration and duration of incubation by response surface methodology. Sains Malays., 46: 43-50.

Hamzah, A.; AbuZarin, M.; Abdulhamid, A.; Omar, O. and Senaf, S. (2012). Optimal physical and nutrient parameters for growth of *Trichoderma viren* UKMP-IM for heavy crude oil degradation. J. Biosci. Biotechnol., 41(1): 71-79.

Hassaine, A. and Bordjiba, O. (2019). Removal of hydrocarbons from liquid media by *Aspergillus niger* van Tieghem. Acta Ecol. Sin., 39(4): 300-305.

Hassanshahian, M.; Emtiazi, G. and Cappello, S. (2012). Isolation and characterization of crude oil‐degrading bacteria from the Persian Gulf and the Caspian Sea. Mar. Pollut. Bull., 64: 7–12.

Jalali, R.; Ghafourian, H.; Asef, Y.; Davarpanah, S. and Sepehr, S. (2002). Removal and recovery of lead using non‐living biomass of marine algae. J. Hazard. Mater., 92: 253-262.

Khanpour-Alikelayeh, E.; Partovinia, A.; Talebi, A. and Kermanian, H. (2021). Enhanced biodegradation of light crude oil by immobilized Bacillus *licheniformis* in fabricated alginate beads through electrospray technique. Environ. Monit. Assess., 193(6): 328.

Kuappi, S.; Sinkkonen, A. T. and Romantschuk, M. (2011). Enhancing bioremediation of diesel‐fuel contaminated soil in a boreal climate; Comparison of biostimulation and bioaugmentation. Int. biodeterior. biodegrad., 65: 390-399.

Latha R, Kalaivani R. 2012. Bacterial degradation of crude oil gravimetric analysis. Pel. Rese. Lib. 3(5): 2789-2795.

Liu, H.;Yao, J.;Yuan, Z.; Shang, Y.; H. Chen, H.;Wang, F.; Masakorala, K.;Yu, C.; Cai, M.;Blake, R.E. and Choi, M.M.F. (2014). Isolation and characterization of crude-oil-degrading bacteria from oil-water mixture in Dagang oilfield. Int. Biodeterior. Biodegrad., 87: 52-59.

Lotfinasabasl, S.; Gunale, V. R. and Rajurkar, N. S. (2012). Assessment of petroleum hydrocarbon degradation from soil and tarball by fungi. Biosci. Discov., 3(2), 186-192.

Mohsenzadeh, F.; Rad, A. C. and Akbari, M. (2012). Evaluation of oil removal efficiency and enzymatic activity in some fungal strains for bioremediation of petroleum - polluted soils. IJEHSE., 9: 26–34.

Okerentugba, P. and Ezeronye, O. (2003). Petroleum degrading potentials of single and mixed microbial cultures isolated from rivers and refinery effluent in Nigeria. Afri. J. Biotechnol., 2(9): 288-292.

Oudot, J. (1984). Rates of microbial degradation of petroleum compounds as determined by computerized capillary gas chromatography and computerized mass spectrometry. Mar. Environ. Rese., 13: 277-302.

Ponnapalli, M. G.; Sura, M. B.; Sudhakar, R.; Govindarajalu, G. and Sijwali, P. S. (2018). Biotransformation of Artemisinin to 14 - hydroxydeoxyartemisinin: C - 14 hydroxylation by *Aspergillus flavus*. J. Agric. Food Chem., 66: 10490-10495.

Rousk, J.; Brookes, P. C. and Baath, E. (2009). Contrasting soil pH effects on fungal and bacterial growth suggest functional redundancy in carbon mineralization. Appl. Environ. Microbiol., 75: 1589-1596.

Salam, L.B.; Obayori, O.S.; Akashoro, O.S. and Okogie, G.O. (2011). Biodegradation of bonny light crude oil by bacteria isolated from contaminated soil. Int. J. Agric. Biol., 13: 245-250.

Satti, S.M.; Shah, A.A.; Marsh, T.L. and Auras, R. (2018). Biodegradation of poly lactic acid in soil microcosms at ambient temperature: Evaluation of natural attenuation, bio augmentation and bio - stimulation. J. Polym. Environ., 26: 3848-3857.

Sayed, K.; Baloo, L.; Kutty, S.R.B.; Al Madhoun, W.; Kankia, M. U.; Jagaba, A. H. and Singa, P. K. (2022). Optimization of palm oil mill effluent final discharge as biostimulant for biodegradation of tapis light crude petroleum oil in seawater. J. Sea Res., 188: 102268.

Thacharodi, A.; Hassan, S.; Singh, T.; Mandal, R.; Khan, H. A.; Hussain, M. A. and Pugazhendhi, A. (2023). Bioremediation of polycyclic aromatic hydrocarbons: An updated microbiological review. Chemosphere., 328: 138498.

UNEP (United Nation Environmental program). (1989). Comparative toxicity test of water accommodated fraction of oils and oil dispersants to marine organisms. Reference methods for marine pollution No. 454, 21P.

Usman, N.; Atta, H. I. and Tijjani, M. B. (2020). Biodegradation studies of benzene, toluene, ethylbenzene and xylene (BTEX) compounds by *Gliocladium* sp. and *Aspergillus terreus*. J. App. Sci. Environ Manage., 24(6):1063-1069.

Wicklow, D. T. and Wittingham, C. (1974). Soil micro fungal changes among the profiles of disturbed conifer hard wood forest. Ecology., 55: 3-16.

Wu, M.; Dick, W.A.,;Li, W.; Wang, X.; Yang, Q.; Wang, T. and Chen, L. (2016). Bioaugmentation and biostimulation of hydrocarbon degradation and the microbial community in a petroleum - contaminated soil. Int. biodeterior. Biodegrade., 107: 158-164.

Zeneli, A.; Kastanaki, E.; Simantiraki, F. and Gidarakos, E. (2019). Monitoring the biodegradation of TPH and PAHs in refinery solid waste by biostimulation and bioaugmentation. J. Environ. Chem. Eng. 7(3): 1-7.