Original Research

Total Petroleum Hydrocarbon (TPHs) Dissipation through Rhizoremediation by Plant Species

Ali Daryabeigi Zand[1]*, Gholamreza Nabibidhendi[2], Nasser Mehrdadi[2], Ravanbakhsh Shirdam[2], Alireza Mikaeili Tabrizi [3]

[1]Environmental Sciences Research Institute, Shahid Beheshti University, G.C., Evin 1983963113, Tehran, Iran
[2]Faculty of Environment, University of Tehran, Tehran, Iran
[3]Department of Environment, Gorgan University of Agricultural Sciences and Natural Resources, Golestan, Iran

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Abstract

Total petroleum hydrocarbons (TPHs) are one of the most common groups of persistent organic contaminants. Plant-based remediation is a relatively new, efficient and environmentally friendly technology that can be promising for removing many contaminants like hydrocarbon pollutants. The main objectives of the current research were to investigate the phytoremediation efficiency of burningbush, flax, and tall fescue as well as the influence of petroleum hydrocarbons on growth characteristics of these plant species. In order to improve soil condition and study the effect of fertilization on plant growth in oil-contaminated soil, peat fertilizer was applied in a separate treatment. Unfertilized as well as fertilized soil samples were analyzed for TPH removal by GC-FID in different time intervals. All plant species showed promising growth behaviour in highly contaminated soil. A decrease of TPHs was found over the course of the experiment in all treatments. The maximum removal was obtained in flax, in which flax removed 97.9% of the initial TPHs from soil. Results demonstrated that the three studied plant species were effective and promising in removing TPHs from contaminated, aged soil.

Keywords: burningbush, flax, tall fescue, hydrocarbons, phytoremediation, soil

Introduction

During the last decade, concerns about hydrocarbons in the environment have increased considerably. Among them, total petroleum hydrocarbons (TPHs) are of great interest as the accumulation of these compounds in soil might lead to significant risks to humans through different exposure pathways [1]. Soil contamination by petroleum hydrocarbons is not specific to contaminated points and they can move through soil and reach groundwater resources. Relatively high hydrophobicity of petroleum hydrocarbons results in a considerable increase in their ability to accumulate in soil and sediment in comparison to aquatic environments [2].

Petroleum hydrocarbons are one of the most important organic soil pollutants in many parts of Iran, especially near oil refineries and spill sites [3]. The development of methods to remediate soils contaminated with toxic pollutants and other organic residues has been an area of intense research interest for several decades [4, 5]. Although various physical, chemical and biological processes have been employed for effective remediation of contaminated soil, to date, many developing countries like Iran have almost completely relinquished remediation of oil-polluted soils due to the high costs of conventional (physical/chemical) soil remediation methods. The widespread need to remediate soils in areas contaminated with high concentrations of var-

*e-mail: a_d_zand@yahoo.com
a_daryabeigi@sbu.ac.ir

ious organic pollutants like petroleum hydrocarbons encourages interest about environmental friendly remediation technologies.

Plant-based remediation is a relatively new, efficient and environmentally friendly technology that can be promising for removing many contaminants like hydrocarbon pollutants. Synergistic cooperation of plant roots and soil microorganisms promotes the degradation of persistent organic contaminants in phytoremediation. Removal of petroleum hydrocarbons from soil in phytoremediation is often attributed to microorganisms living in the rhizosphere under the influence of plant roots [6, 7]. Microbial communities in planted soils are greater and more active than unplanted soils [8, 9]. Microorganisms in the rhizosphere benefit from the root exudates and plants, in turn, from the metabolic detoxification of potentially toxic compounds brought about by microbial communities. Additionally, microbial populations benefit the plant through recycling and solubilization of mineral nutrients, as well as by supplying vitamins, amino acids, auxins, cytokinins and gibberellins that stimulate plant growth [10].

Many plant species are sensitive to petroleum contaminants [11]. Ninety-six percent reduction of ryegrass biomass after 30 days growth on soil contaminated with 25 g kg^{-1} petroleum hydrocarbons was observed in a phytoremediation study by Tesar et al. [12]. Phytoremediation is a site-specific remediation method; that's why some contradictory results have been reported regarding the efficiency of this technology in removing contaminants from soil [13]. Thus employing native plant species that are tolerant to high concentrations of TPHs in soil is a key factor in the success of phytoremediation.

Plant species were selected for this study based on the need to develop a highly fibrous root density. Burningbush (*Kochia scoparia* (L.) Schard) is a common plant species in many parts of Asia, including Iran. Burningbush develops a large root system and has a branched fibrous root system, which may contribute to its successful phytoremediation potential. In addition, flax (*Linum usitatissumum* L.), which had been used by Adam and Duncan (2002) had shown promising tolerance and germination rate in hydrocarbon-polluted soil [14]. In addition, tall fescue (*Festuca arundinacea*) is a robust, broadleaf grass that develops deep and extensive root systems and spreads by rhizomes. Initial growth may be slow, but this plant species grows vigorously once established. Tall fescue has been shown to reduce hydrocarbons in previous studies [15].

The main objectives of the present study were:

(1) to evaluate the phytoremediation capability of burning bush (*Kochia scoparia* (L.) Schard), flax (*Linum usitatissumum* L.) and tall fescue (*Festuca arundinacea*) and

(2) to investigate the effect of TPHs on growth parameters of the three mentioned plant species including germination, shoot height and biomass, and root length and biomass.

Our current research used burningbush and flax were used for the first time in the history of phytoremediation studies. Furthermore, since nutrient additions to soil through fertilization may also increase the plant biomass and thus promote pollutant removal as suggested by some authors [16, 17], the influence of peat fertilizer upon plant growth and phytoremediation performance was also evaluated.

Table 1. Physical and chemical characteristics of the soil used in the current study.

Parameter	Value	Analytical method
Clay (%)*	28	Hydrometer measurement
Silt (%)	32	Hydrometer measurement
Sand (%)	26	Hydrometer measurement
Gravel (%)	14	Sieve
Organic matter (%)	4.57	Walkley-Black
Organic C (%)	2.65	-
Soil pH	7.6	1:1 soil/water slurry
Electrical Conductivity (dS/m)	3.02	1:2 soil/water slurry
Total N (%)	0.12	Kjeldahl
Phosphorus (mg/kg)	34.2	Olsen

Materials and Methods

Soil was provided from contaminated lands around an Oil Refinery of Tehran. The soil was sieved through a 10 mm sieve and mixed thoroughly. In most studies soil is sieved by a 2 mm sieve which, according to AASHTO and Massachusetts's Institute of Technology standards is the boundary limit between sand and gravel particles [18]. However, this leads to a considerable loss of coarse grain portion of real soil and lack of accordance between real soil from the contaminated site and soil used in the phytoremediation experiment. Some physical and chemical properties of the experimental soil are presented in Table 1. Phosphorus was measured by Olsen P extracting solution (0.5*M* $NaHCO_3$, pH 8.5); total nitrogen by Kjeldahl digestion; pH was analyzed by glass electrode using a 1:1 soil:water ratio; and EC EC was measured by conductivity meter in a soil-water extract (1:2 soil:water ratio) [19-21].

After a relatively homogeneous mixture of soil was obtained, the soil was weighed and transferred to PVC pots (1.5 kg of soil per pot). Increases in soil electric conductivity affect the plant growth; nevertheless, most plants are not significantly impacted until the electrical conductivity is greater than 4 decisiemens per meters [22]. The soil used in this research has an electrical conductivity of 3.02 decisiemens per meters (Table 1). In addition, the soil's contamination by TPHs results in a decrease of a plant's nitrogen absorption and increase in the C/N ratio. In the contaminated soil used in this study the C/N ratio was not very high (approximately 22). According to Xu and Johnson (1997), when C/N ratio is under 25, petroleum hydrocarbon degradation and removal may be enhanced compared to the higher values of C/N ratio [23].

In order to study the effect of fertilization on plant growth in hydrocarbon-polluted soil as well as phytoremediation efficiency, peat fertilizer was used. Characteristics of the utilized peat fertilizer were as follows: pH = 5.5, total nitrogen = 1.1 percent, existing phosphorus = 32.7 mg/kg, potassium = 2,280 mg/kg, and organic carbon = 30.9 percent. The soil composition in the pots was as follows:

T1: clean soil of lands surrounding Oil Refinery of Tehran without any kinds of contamination background (control soil);

T2: contaminated soil (80%) + clean soil (20%);

T3: soil used in treatment T2 (80%) + peat fertilizer (20%).

The initial concentration of TPHs in the soil used in treatment T2 was 34,358±1,633 mg/kg (almost 3.5 percent by weight), which demonstrates a rather high level of contamination in soil. A natural attenuation treatment (without plant) in which petroleum hydrocarbons were naturally attenuated was also considered.

Burningbush, flax, and tall fescue (*Festuca arundinacea*) were cultivated over a four-month period in a greenhouse. The seeds were planted at 1.5-2.0 cm depth of the surface soil in each pot in the following quantities: 40 for burningbush, and 2 grams for flax and tall fescue. The pots were placed inside the greenhouse under sunlight. The temperature was between 21°C and 33°C. Monitoring of plant growth was done every 10 days. The pots were watered twice a week to maintain a constant and sufficient moisture level and to minimize the generation of leachate. PVC pans were placed under each pot to collect leachate. Leached water was collected and included in the next watering to avoid petroleum hydrocarbons loss. However, Hutchinson et al. (2001) showed that only 0.02% of the TPH in aged soil was leached from the pots with irrigation water. Germination rate in the initial weeks was studied by counting the number of grown seeds or surface density observation. Shoot height also was measured and monitored. Destructive pots were destroyed after 30, 60, and 120 days. For this purpose, first the plants were carefully removed from their soil and carefully washed with running water. Then using a ruler, root length and shoot height were measured. In order to measure dry biomass, plants were placed in an oven at 70°C for 48 hours and then weighed. Soil samples were taken by a core sampler (inner diameter = 10 mm) from the whole height of the pots every month.

For TPH analysis, soil samples were air dried at room temperature and passed through a 2 mm sieve. The samples were stored at 4°C prior to extraction and analysis. Ultrasonic extraction was performed using dichloromethane solvent. Ten cc of dichloromethane was added to about 5 grams of contaminated soil and then it was placed in an ultrasonic water bath for three minutes at room temperature. All of these operations were repeated three times [24]. Then the obtained extracts were concentrated to 1 ml under a gentle stream of nitrogen gas. Two µL of the sample was injected into a gas chromatograph UNICAM 610 series equipped with a flame ionization detector (FID). The column used for analysis was DB-5 with 30 m length, 0.25 mm internal diameter and 0.2 µm thickness of film.

Table 2. Measurement results of root length, root biomass, and shoot biomass for destructive pots.

Plant species	Parameter	Time (day)	Soil treatment		
			T1	T2	T3
Burningbush	Root length (cm)	30	17	10 (-41)[¥]	16 (-6)
		60	38	18 (-53)	29 (-24)
		120	47	23 (-51)	37 (-21)
	Root biomass (gr)	30	5.0	2.1 (-58)	4.2 (-16)
		60	10.8	3.9 (-64)	7.9 (-27)
		120	16.2	6.7 (-59)	15.4 (-49)
	Shoot biomass (gr)	30	3.1	1.7 (-45)	2.5 (-19)
		60	6.0	3.8 (-37)	5.3 (-12)
		120	11.0	7.1 (-35)	13.1 (19)
Flax	Root length (cm)	30	20	18 (-10)	21 (-5)
		60	36	34 (-5)	40 (-11)
		120	40	35 (-12)	42 (5)
	Root biomass (gr)	30	10.1	12 (19)	17.2 (70)
		60	20.8	24.9 (20)	30.7 (47)
		120	24.8	30.3 (22)	35.1 (41)
	Shoot biomass (gr)	30	4.2	3.6 (-28)	3.3 (-21)
		60	8	7.6 (-5)	7.3 (-9)
		120	10.8	9.7 (-10)	9.9 (-8)
Tall fescue	Root length (cm)	30	20	16 (-20)	15 (-25)
		60	38	33 (-13)	31 (-18)
		120	42	37 (-12)	33 (-21)
	Root biomass (gr)	30	30.3	29.0 (-4)	26.2 (-14)
		60	58.3	57.7 (-1)	49.2 (-16)
		120	69.0	69.2 (0.3)	58.4 (-15)
	Shoot biomass (gr)	30	10.3	9.0 (-13)	9.5 (-8)
		60	22.8	20.2 (-11)	21.9 (-4)
		120	29.2	27.9 (-4)	24.6 (-16)

[¥] Values in parentheses represent changes in comparison with control (%).

The injector and FID detector temperatures were adjusted at 280°C and 340°C, respectively. Initial column temperature was adjusted at 50°C for 5 minutes, and then increased to 250°C with 10°C/min slope and remained at 250°C for 40 minutes (a few of the soil samples were analyzed with a gas chromatograph HP 5890 series equipped with FID in another laboratory). The difference between soil treatments was tested by one-way ANOVA. Significance level was considered at 0.05. All statistical analyses were performed using the Statistical Package for Social Sciences (SPSS) software 10.0 for Window (SPSS Inc., IL, USA).

Table 3. TPH removal rates in different treatments (mg/kg/day).

Treatment	Plant species	Time interval (day)			
		0-30	30-60	60-90	90-120
T2 (without fertilizer)	Burningbush	461.8	272.8	159.0	16.6
	Flax	411.9	466.3	173.0	69.7
	Tall fescue	473.8	189.7	307.5	133.7
T3 (with fertilizer)	Burningbush	399.1	225.8	177.5	58.0
	Flax	356.8	206.2	-103.4	252.6
	Tall fescue	439.6	155.5	135.2	96.1
Natural Attenuation	-	194.6	104.9	106.8	39.6

Results

All plant species employed in the current phytoremediation study showed promising behaviour in petroleum hydrocarbon-contaminated soil. However, oil pollution depressed plant growth to some extent. Final seedling emergence of studied plants and shoot height monitoring results are presented in Figs. 1 and 2, respectively. Root length, root biomass, and shoot biomass measurement results for destructive pot are presented in Table 2. In addition, Figs. 3 to 5 show residual amounts of TPHs in soil.

Burningbush and flax species that were employed for the first time in the history of phytoremediation of oil-contaminated soil, showed promising behaviour in highly contaminated soil. Germination of burningbush was clearly visible on day 7. Seedling emergence of burningbush and tall fescue was not depressed by the presence of petroleum hydrocarbons in soil (Fig. 1). However, contaminated treatments (T2 and T3) reached their maximum germination rate later than control treatment. A reduction of germination by 23% was found for flax. Peat fertilizer didn't have significant influence on germination rate.

Growth depression was observed in all three tested plant species. Shoot heights were lower in contaminated soil than in uncontaminated soil. A remarkable reduction of shoot height by the presence of petroleum hydrocarbons was observed for burningbush. Oil contamination couldn't influence shoot height of tall fescue significantly (P>0.05). Concerning the peat fertilizer effect in contaminated soil, it increased the shoot height of the three studied plants during the whole experiment. While peat fertilizer positively affected burningbush and flax growth, it didn't have considerable influence on shoot height of tall fescue. In most cases, plant growth was diminished after 90 days cultivation, probably due to nutrient depletion in confined soil of pots.

Table 2 shows that maximum root length was achieved in tall fescue. Tall fescue (T2) also had the greatest root and shoot biomass among the studied plant species (Table 2). At the end of the experiment root lengths of burningbush, flax and tall fescue were decreased by 51%, 12%, and 12%,

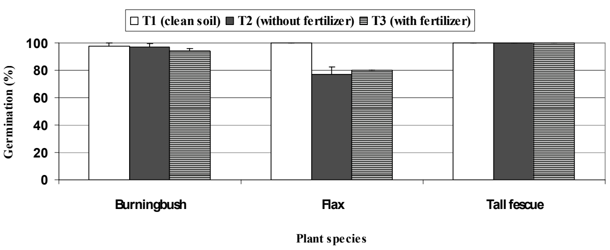

Fig. 1. Germination of plant species in different treatments.

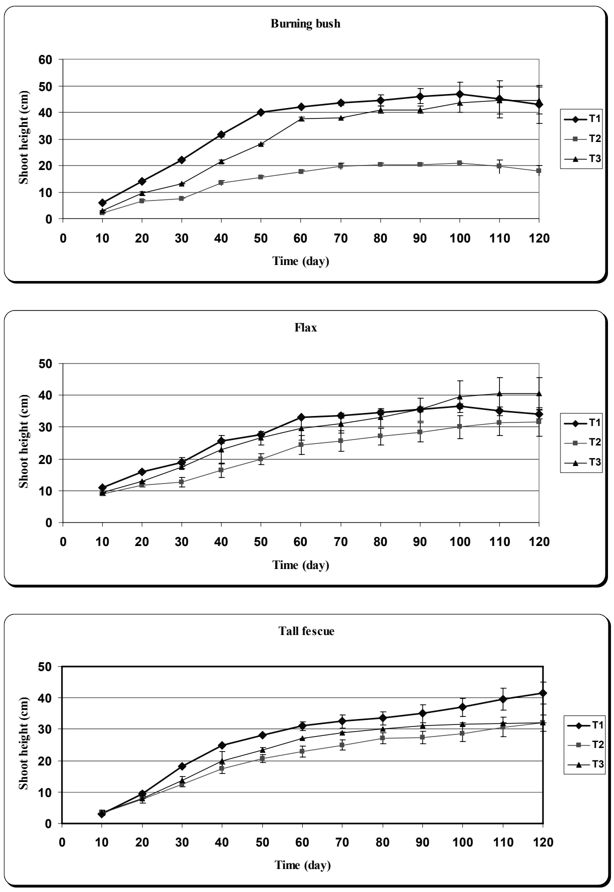

Fig. 2. Shoot height monitoring during phytoremediation.

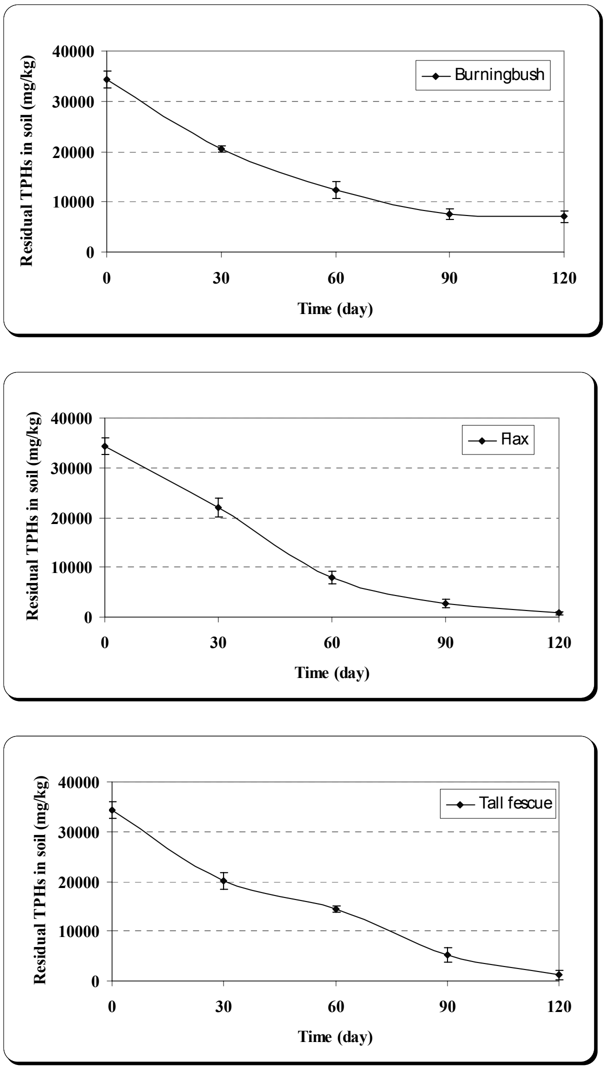

Fig. 3. Residual amounts of total petroleum hydrocarbons (TPHs) in the rhizosphere of burningbush, flax, and tall fescue in treatment 2 (without fertilizer).

respectively in treatment 2, compared with plants cultivated in clean soil. Petroleum hydrocarbon pollution reduced root biomass of burningbush by 59%, while it didn't adversely affect root biomass of tall fescue and flax. Shoot biomass of burningbush, flax, and tall fescue were decreased by 35%, 10%, and 4%, respectively, in unfertilized treatment compared with control treatment. In some cases peat fertilizer increased plant biomass compared to control treatment (e.g. burningbush shoot biomass). Peat fertilizer could reduce the adverse effect of hydrocarbons on burningbush and flax growth, likely due to improvement of soil nutrient conditions. Soil amendment by peat could positively affect tall fescue growth. One probable reason is that organic fertilizers like peat may act as a physical barrier limiting roots from access to water, thereby adversely affecting plant growth.

Figs. 3-5 show that TPH concentrations decreased in all treatments. Significant influence of studied plants on petroleum hydrocarbon removal at different sampling times was obtained ($P<0.05$). All three plant species caused significantly higher petroleum hydrocarbon dissipation compared to unplanted soil ($P<0.05$). Natural attenuation could reduce TPH level in soil by 38.9% at the end of the experiment. The highest phytoremediation efficiency obtained for flax in which plant presence could reduce TPH levels was 59% in comparison with unplanted treatment. Peat fertilizer did not show a positive and significant role in phytoremediation efficiency of studied plant species. TPHs dissipation by flax was decreased in presence of peat fertilizer (18.8% reduction).

In addition, a control treatment for fertilized treatment was also considered. Natural attenuation of T2 (unfertilized treatment) and T3 (fertilized treatment) didn't make a significant difference. The residual concentrations of TPHs in soil on days 30, 60, 90, and 120 were 21,865, 19,241, 16,551, and 15,126 mg/kg, respectively, for T3 control treatment (natural attenuation of fertilized soil with peat). The obtained results showed that the two control treatments (with and without fertilizer) had almost the same behaviour. Natural attenuation could finally reduce TPH levels in soils by 38.9% and 44% for unfertilized and fertilized treat-

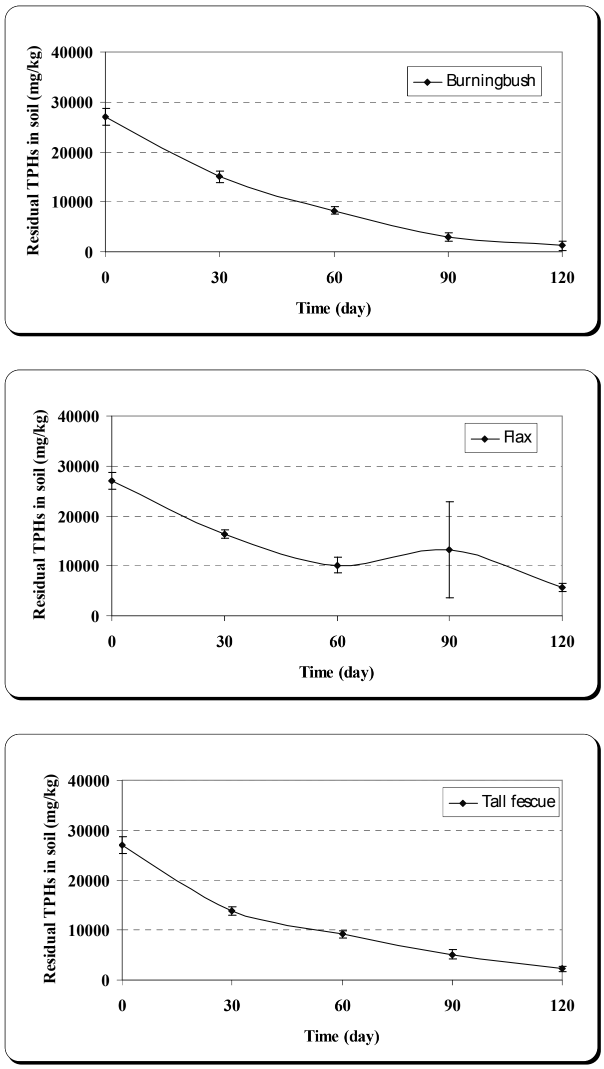

Fig. 4. Residual amounts of total petroleum hydrocarbons (TPHs) in the rhizosphere of burningbush, flax, and tall fescue in treatment 3 (with fertilizer).

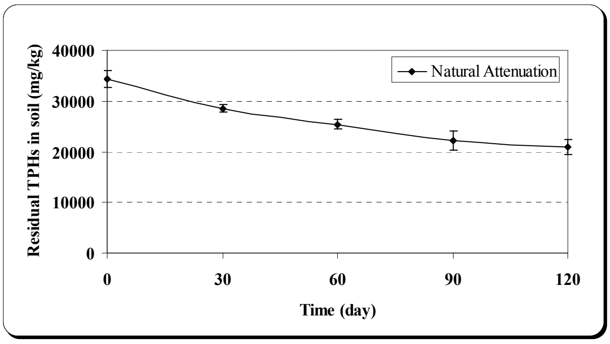

Fig. 5. Natural attenuation of total petroleum hydrocarbons (TPHs).

ments, respectively. The considerable effect of nutrient addition thorough fertilization on natural attenuation of TPHs was observed in the last month in which peat increased TPH dissipation by 3.2%. It seems that biostimulation can not remove TPHs from aged soils in an unplanted treatment.

TPH removal rates at different time intervals is also presented in Table 3. The highest removal rate was 473.8 mg/kg/day, obtained for tall fescue (T2). TPH dissipation in all treatments (planted and unplanted) was higher in the first 60 days than the second 60 days. The lowest TPH reduction rates were found in the last month. For instance, the TPH removal rate by burningbush was 461.8 mg/kg/day in the first 30 days, while it decreased to 16.6 mg/kg/day in the last 30 days (around 96% reduction in removal rate).

Discussion

Germination is one of the most important stages in plant establishment. Sensitivity of germination as well as initial growth steps of plant species can influence phytoremediation efficiency. Some studies have suggested a link between poor germination and subsequent poor growth in hydrocarbon-contaminated soil [25]. In this study, petroleum hydrocarbon pollution did not have a significant adverse effect on germination of studied plant species ($P>0.05$); however, the subsequent growth was depressed significantly in most cases by petroleum hydrocarbon pollution ($P<0.05$). In the current study, delays in seedling emergence were recorded in some cases.

Peat fertilizer couldn't increase seedling emergence significantly ($P>0.05$). Germination of tall fescue reached up to 100% in all treatments, which demonstrates the tolerance of this plant to the presence of hydrocarbons in soil. Generally, seedling emergence was not considerably lower in contaminated soil than clean soil. This is due to the fact that the soil used in this study was aged soil. Aged soils contain fewer compounds toxic to plants (e.g. low molecular weight hydrocarbons). Seedling emergence can be inhibited or delayed by toxic oil effects. Salanitro et al. reported seedling emergence reduction of corn, wheat, and oat in soil contaminated with heavy crude oil [26]. A reduction of germination by 30% to 90% for some native species of Mexico in petroleum-polluted soil was also observed by Gallego-Martinez et al. [27]. Oil components can enter into the seed and disturb metabolic reactions or even kill the embryo [14]. The soil used in this study was aged soil and mainly contained high molecular weight hydrocarbons. Therefore, a probable cause of germination delay of studied plants can be the water repellent property of hydrocarbons. Hydrocarbons may act as a physical barrier preventing seeds from access to water and oxygen or delaying their access [14].

Although fertilizer use may not have an important impact on plant tolerance or sensitivity to petroleum contamination, it can have a positive effect on plant growth even in contaminated soils through biostimulation. Considerable reduction of plant biomass as well as root length by the presence of petroleum hydrocarbons was found in some cases. For example, the presence of petroleum hydrocarbons in soil could finally reduce root length, shoot and root biomass of burningbush by 51, 35, and 59 percent, respectively. Chaineau et al. reported a growth rate reduction of beans and wheat by more than 80 percent [25]. Gallego-Martinez et al. also found a reduction of biomass for three plant species [27]. Inhibition of plant growth can be caused by toxic effects of petroleum hydrocarbons. Small molecules of hydrocarbons can enter and pass cell membranes, leading to reduced membrane integrity or even to death of the cell [28]. Plant height and shoot biomass are good indicators of plant health; however, greater shoot biomass measurements are not necessarily indicative of enhanced remediation efficiency [29]. Greater root biomass is likely to be associated with more extensive root exploration of the soil and, subsequently, higher microbial biomass and activity.

On-site observations also showed that tall fescue posses the most extensive and dense root system among studied plants. Burningbush and flax also had a relatively dense root system that has been reflected in their root biomass. With regard to their root system and also their remarkable tolerance in petroleum-contaminated soil, it seems that these three plant species may be a promising in phytoremediation of TPH-contaminated soils. Generally the adverse effect of petroleum hydrocarbon on plant growth parameters was not high in the current study compared to some other research. A high reduction of biomass of *B. brizantha* and *P. maximum* by 85% and 99%, respectively, compared to the control, was found in soil contaminated with light crude oil, which has a large fraction of small molecular compounds [28]. The difference between the results of this study and related research can originate from the fact that aged soils have fewer low molecular weight hydrocarbons leading to less toxic effects on plant tissues.

TPH concentrations decreased in all treatments. Planted treatments showed significantly higher petroleum hydrocarbon dissipation compared to unplanted treatment. Since the most important mechanism of phytoremediation is based on the stimulation of soil microorganisms, it can be assumed that higher root biomass, as obtained for tall fescue in this study, means a larger rhizosphere for the microbial population and it is correlated with a higher degradation of hydrocarbons in soil [30]. Phytoremediation efficiency of flax and tall fescue was very close. Flax showed the best phytoremediation capability in oil-contaminated soil. Quantity and quality of the flax exudates is likely the most important reason for its high phytoremediation efficiency. However, further research should be conducted to address interactions among flax exudates, microorganisms, and hydrocarbons. Phytoremediation potential of burningbush was lower than flax and tall fescue, probably due to the interaction between released enzymes and hydrocarbon degrading bacteria. Peat fertilizer improved phytoremediation efficiency of burningbush. This may be attributed to the positive effect of used organic fertilizer on soil enzymatic activities, probably due to the higher microbial biomass produced in the presence of peat. However, peat fertilizer didn't have a significant influence on phytoremediation efficiency of tall fescue ($P>0.05$), and decreased flax phytoremediation efficiency considerably. Reducing organic fertilizer content in soil or utilization of inorganic amendments may alter the influence of fertilizer on flax phytoremediation efficiency. Relatively high reduction of TPHs in the rhizosphere of surveyed plants may be attributed to suitable plant growth in hydrocarbon-contaminated soil used in the current study. Reduced plant height and biomass production may be considered as a basis for unsuccessful phytoremediation. When evaluating plant species for phytoremediation, the decrease in plant growth, and especially root biomass, should be as low as possible [30], as obtained in the current research.

Studying the removal rate shows that TPH removal in the first half of the experiment were in general lower than the second half of the experiments. In the second half of the experiment, the TPH content in soil dropped considerably. TPH removal in the rhizosphere of plants was not significant after the 90^{th} day ($P>0.05$). Phytoremediation efficiency decrease in the second half of the experiment may be attributed to plant growth reduction, especially after the 90^{th} day. In addition, Merkl et al. suggest that degrading microorganisms are stimulated in their growth and activity by root exudates, which vary with plant age and nutritional status [30]. Quantitative and qualitative alteration of plant exudates with plant age may influence the population and activity of hydrocarbon degrading microorganisms in the rhizosphere, thereby influence the phytoremediation efficiency and rate with time. Non-appreciable TPH removal was also observed in a phytoremediation study by Escalante-Espinosa et al. [10] from 120 to 180 days of culture in planted and unplanted treatments. Hutchinson et al. and Escalante-Espinosa et al. found that the maximum phytoremediation rate is reached at the first stage of culture (up to 60 days) [10, 16], as we observed in the current study.

Conclusion

Based on the obtained results, all three studied plants are promising species for phytoremediation of aged, petroleum hydrocarbon-contaminated soils. However, petroleum hydrocarbon contamination depressed growth of the studied plants. Flax and burningbush, which had rarely been used in phytoremediation experiments in order to remove TPHs from soil, showed promising behaviour in oil-contaminated soil as well as acceptable phytoremediation efficiency in this research. They can be introduced as phytoremediator plant species. Tall fescue showed the best root biomass production and caused high hydrocarbon dissipation compared to unplanted soil, suggesting that greater root biomass is likely to be associated with higher microbial population and activity in the rhizosphere and, subsequently, higher phytoremediation efficiency. Peat fertilizer had a positive effect on plant growth in some cases, but it couldn't enhance phytoremediation efficiency significantly. Flax, burningbush, and tall fescue are well-known and easily accessible plant species in many countries. According to the results, soil remediation with these plant species can be a promising way to manage oil-polluted sites in semi-arid regions. The investigation of the microbial population in the rhizosphere, as well as studying the influence of other organic and inorganic fertilizers on hydrocarbon removal through biostimulation, would increase understandings about plant-based remediation of hydrocarbon-contaminated soils. The use of vegetation as a feasible remediation approach for soils contaminated with petroleum hydrocarbons may become attractive in Iran as a developing country because it is inexpensive and requires minimum maintenance and management.

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