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# Effect of Petroleum Crude Oil on Mineral Nutrient Elements, Soil Properties and Bacterial Biomass of the Rhizosphere of Jojoba

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#### Authors' contributions

This work was carried out in collaboration between all authors. Author WMS designed the study, performed the statistical analysis, worte the protocol, and wrote the first draft of the manuscript. Authors GHSAH and RMSAM managed the analysis of the study. Author MAEB managed the microbial analysis and the literature search. All authors read and approved the final manuscript.

Research Article

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#### **ABSTRACT**

**Aims:** This study is to evaluate the effect of petroleum crude oil contaminated soil on the mineral nutrient elements, soil properties and bacterial biomass of the rhizosphere of jojoba plants (*Simmodsia chinensis*).

**Methodology:** A pot experiment was carried out. The soil was treated with different levels of crude oil: 1, 2 and 3% v/w either alone or in combination with inorganic fertilizers.

Results: Malondialdehyde (MDA) concentration increased in jojoba leaves when grown in petroleum oil polluted soil especially at 2% and 3% crude oil. It was noted that, Na, Mg and Ca decreased while K increased in shoots of jojoba. In roots Na and Ca increased however K and Mg decreased with increasing crude oil concentration in the soil. Heavy metals, Cu, Mn, Cd and Pb increased in both shoot and root with increasing crude oil concentration while, Zn decreased comparing with the control. In soil, N and K decreased meanwhile Cu, Fe, Mn and Zn as well as organic matter increased with increasing crude

oil concentration. Soil was free from P while, the addition of inorganic fertilizers improved P content. Bacterial account was significantly increased at the end of the experiment at 1% and 2% crude oil especially after addition of inorganic fertilizers. The electric conductivity and MDA of the leaves increased with increasing crude oil concentration. The addition of inorganic fertilizers to crude oil contaminated soil decreased the electric conductivity and MDA comparing with crude oil only.

**Conclusion:** The observed changes in composition of mineral elements in jojoba plants in the present study could be attributed to the cell injury and disruption in the cell membrane, heavy metal accumulation and toxic nature of the petroleum oil. Also this study has demonstrated that soil contamination with crude oil has a highly significant effect of reducing some mineral element composition of Jojoba plants.

Keywords: Jojoba plant; mineral nutrient elements; soil properties; bacterial biomass.

#### 1. INTRODUCTION

Jojoba (*Simmodsia chinensis*) is found in coastal and cis-montane southern California east to central Arizona and south to Sonora and Baja California [1,2]. Jojoba has very promising scope for cultivation in the desert even in the relatively hot weather. Its nature withstands the hot weather in summer, warm weather in winter, low fertility of soil and low water resources. It needs less than one quarter the amount of water needed for olives with great ability to withstand the high salt in the soil. The payback for planting Jojoba is encouraging. To be used as fuel it needs to be cultivated in huge amounts which are easy in desert land in many countries.

Plants are extremely tolerant to drought [3]. The large seeds have been used locally in North America as a food source by indigenous people [4]. The most noteworthy feature of jojoba is the unusual liquid wax reserved in its seeds. This substance, a fatty acid ester of a long-chain alcohol, is unique in the plant kingdom. It has chemical and rheological properties similar to those of sperm whale oil, making it useful in a host of applications [4]. The oil concentration of the seed at maturity stage ranges from 38 to 62% of seed dry weight [5]. The oil can be obtained by mechanical pressure or by solvent extraction [6]. The unique oxidative stability of the natural jojoba oil, the pleasant feel of the oil on skin and hair, the biodegradability, non petroleum based and renewable product are reasons for its valued use in cosmetic industry [7,8]. The meal remaining after jojoba oil extraction contains 30% protein consisting of 17 amino acids, 7 of which are essential amino acids. The meal becomes suitable for utilization as animal feed and organic fertilizer for soil.

Jojoba is considered one of the most practical solutions for desert plantation in Egypt, Saudi Arabia and other desert lands. Heat, drought and salt tolerance, lesser possibilities for infection, lesser need for fertilizers, and generous financial income, are certainly the most encouraging goals to plant jojoba in Egypt and Saudi Arabia [9-11].

Petroleum crude oil is a complex mixture of hydrocarbon (aliphatic and aromatic), liquid in its natural state [12] and toxic in nature to different biomass [13]. Oil spillage is a widespread phenomenon and has raised considerable concern on the subject of petroleum oil pollution especially on arable agricultural. Petroleum oil is spilled on soils due to several factors such as pipeline destruction [14.15].

Petroleum oil spillage on soil generally retards plant growth and soil productivity [16-18], reduces aeration by blocking air spaces between soil particles, hence create a condition of an aerobiosis [19] and causes root stress in plants which also reduces leaf growth [20]. The initial reaction of the microorganisms after petroleum oil spillage on the soil is a reduction of activity due to reduced air availability. This has been noted to arise from selective destruction of aerobic bacteria and fungi thus leaving the resistant and adaptive microbial strains to proliferate [21].

The soil composed of aggregate of decaying organic matter (OM); living organisms and weathered mineral materials [22]. The reduction of the growth of *Amaranthus hybridus L*. could be due to reduction of mineral elements in petroleum oil contaminated soil [23]. This could have occurred as a result of reduced availability of mineral elements [24] and increased acidity in the soil [25,26]. Such increase in soil acidity can affect the microbial distribution in the soil, hence reducing their activities in the rhizosphere.

In addition, the availability of heavy metals and micronutrients such as zinc (Zn) is equally affected. It has been reported that a significant increase in heavy metals in soils that were contaminated with automobile waste oil [27]. Zinc as a micronutrient, has been indicated to be beneficial to plants [28]. A number of edaphic factors, including pH, organic matter, effective cation exchange capacity and clay have been indicated to affect Zn availability in soils [29-31]. The petroleum oil contamination can improve soil content with some nutrient elements including Mg, K, P, Na and exhibits a highly significant effect on the chemical composition of maize seeds [32]. Excessive amount of Cd may decrease the uptake of nutrient elements, inhibition of various enzyme activities and induction of oxidative stress including alterations in enzymes of the antioxidant defense system [33]. Lead (Pb), a potentially toxic heavy metal with no known biological function, has attracted more and more considerable attention for its widespread distribution and potential risk to the environment. Pb contamination in soils not only causes the changes of soil microorganism and its activities and resulted in soil fertility deterioration but also directly affects the change of physiological indices and, furthermore, resulted in yield decline [34]. Ultimately, lead enriched in the body of humans through the food chain endangered their health [35]. Soil microbial activity has a great potential as an early and sensitive indicator of stress in soil and has been employed in national and international monitoring programs [36]. The sensitivity of plants to heavy metals depends on an interrelated network of physiological and molecular mechanisms such as (i) uptake and accumulation of metals through binding to extracellular exudates and cell wall constituents; (ii) efflux of heavy metals from cytoplasm to extranuclear compartments including vacuoles; (iii) complexation of heavy metal ions inside the cell by various substances, for example, organic acids, amino acids, phytochelatins, and metallothioneins; (iv) accumulation of osmolytes and osmoprotectants and induction of antioxidative enzymes (v) activation or modification of plant metabolism to allow adequate functioning of metabolic pathways and rapid repair of damaged cell structures [37]. Leakage of electrolytes from plant cells is the earliest effect of stresses [38]. Therefore measurement of the electric conductivity (EC) of tissue diffusates is often used to determine frost injury in plants [39]. Solute leakage may also result from other stresses such as drought [40]. The diffusate conductivity is affected not only by membrane permeability but also by leaching of the apoplast [38]. Adaptation is associated with maintaining osmotic homeostasis by metabolic adjustments that lead to the accumulation of metabolically compatible compounds such as, malondialdehyde (MDA) [41,42].

The present study has been undertaken to evaluate the effects of petroleum crude oil contaminated soil on the mineral nutrient elements of jojoba and its uptake from heavy metals as well as the changes in soil properties and bacterial biomass.

#### 2. METHODOLOGY

#### 2.1 Materials

Soil was obtained from Dammam zone in the east of Saudi Arabia. The soil samples were air dried, thoroughly mixed and passed through 2 mm sieve to remove gravels and debris. Soil texture was coarse sand. Mechanical analyses were made using serial sieves. The soil pH was 8.07 and its electric conductivity was measured using electric conductivity meter at 25°C (5.19 ds/m).

Petroleum oil is Arabian heavy crude oil obtained from petroleum company Saudi Aramco, Saudi Arabia.

Jojoba seeds are obtained from department of Arid Land Agriculture, Faculty of Meteorology, King Abdul Aziz University, Saudi Arabia.

# 2.2 Experimental Design

The soil was dried in room temperature and sieved through 2 mm mesh. Pot experiments were used to achieve this project. Two main groups of pots were prepared. The dried soil was supplemented with various concentrations of petroleum oil (1, 2 and 3% v/w) then mixed to make homogenized contaminated soil (first group). In the second group, the inorganic fertilizers (75 mg nitrate (NH $_4$ NO $_3$ ) and 30 mg phosphate (KH $_2$ PO $_4$ /kg) were added to the petroleum oil contaminated soils before seeding according to Rosenberg and Ron [43]. Jojoba seeds were surface sterilized with 0.01M HgCl $_2$  for 3 minutes, and washed thoroughly several times with distilled water. Eight uniform sterilized jojoba seeds were planted in each pot. Three replicates for each treatment will be prepared. The pots were kept in the growth chamber and the plants will be subjected to constant 25°C, 60% relative humidity and 14h light /10h dark. The pots were irrigated with water at 2 days intervals.

#### 2.3 Harvesting

At the end of the experiment (85 days) the plants were harvested, and split up into root and shoot systems. Roots were washed carefully with tap water many times then with deionized water three times and dried with filter paper. Shoots and roots were oven dried for determination of elements. Some fresh leaves were used to determine MDA and EC. Soil analysis and total bacterial counts of the rhizosphere were carried out at the beginning and at the end of the experiment.

### 2.4 Lipid Peroxidation

The level of lipid peroxidation was measured by determining the level of MDA. It was determined using the procedure described by Zhang et al. [44]. The plant leaves was homogenized in 1% tri-chloroacetic acid and then centrifuged at 10,000 rpm for 15 min. Supernatant was heated with 0.05 thiobarbituric acid for 30 min at 95°C. The heated

supernatant was then centrifuged at 5000 rpm for 5 min and the absorbance was measured at 532 and 600 nm.

# 2.5 Soil Analysis

The soil samples were air dried and passed through 2mm sieve to remove debris then were analyzed chemically according to Piper; United States salinity Lab. Staff and Jackson [45-47].

#### 2.6 Total Bacterial Viable Count

The number of bacterial cells in the samples was counted using serial dilutions and the spread-plate technique according to Stukus [48].

One gram of the sample was added to 100 ml of sterile distilled water and was shaken for one hour. Tenfold serial dilutions of each sample were prepared and 0.1ml of 10<sup>-5</sup> and 10<sup>-6</sup> dilutions was spread onto nutrient agar plates (three replicates per dilution). The plates were incubated at 30°C for 48h then the colonies were counted.

#### 2.7 Electric Conductivity

Ion leakage was measured as electric conductivity according to Yan et al. [49]. The washed leaves were cut into 1cm pieces and placed in a glass beaker containing 10ml deionized water. The beakers were kept at 30°C for 3h and the conductivity of the solution was measured by a conductivity meter. The same samples were boiled for 10min then cooled to room temperature and their conductivity was measured again. The percentage of electrolyte leakage was calculated as follows:

 $EC (\%) = (C1/C2) \times 100.$ 

Where C1 and C2 are the electrolyte conductivities measured before and after boiling, respectively.

#### 2.8 Estimation of Mineral Elements

Estimation of cations was determined according to Champan and Pratt [50]. The concentrations of metals in the vegetation and soil samples were determined using Atomic Absorption Spectro-photometry (Varian SpectrAA 220 FS, Varian, Palo Alto, CA, USA).

# 2.9 Statistical Analysis

The data were analyzed statistically by one-way ANOVA. Values in the tables indicate the mean values ± SD based on independent three determinations (n =3). Least significant difference (L.S.D) test was used to assess the differences between treatments.

#### 3. RESULTS AND DISCUSSION

MDA concentration increased in the leaves of jojoba plants grown in the petroleum oil polluted soil with increasing concentration of petroleum oil in the soil as shown in Table 1.

This increment was significant at 2% and 3% crude oil in the soil. Addition of inorganic fertilizers to crude oil contaminated soil showed a significant increase in MDA at 3% however it still less than MDA content at 3% petroleum oil without fertilizer. It has been reported that jojoba plants grown in crude oil contaminated soil was exposed to drought stress [51]. As recorded in Table 2, the contamination of soil by crude oil either in absence or presence of inorganic fertilizers leads to increase heavy metal (Cu, Fe, Mn and Zn) contents. The plants exposed to heavy metal stress with special regards to copper and zinc at 2% crude oil and for iron and manganese at 2% crude oil supplemented with inorganic fertilizers. All these stresses lead to increment of MDA. Our results are in agreement with previous reports where MDA increased under soil heavy metal pollution in different plant species [52-54]. There were no phosphorus in the tested soil at all concentrations of crude oil, however, the addition of inorganic fertilizers improve the phosphorus content of the soil. Potassium ions content in general decreased with increasing crude oil concentration in the soil. The reduction in K content may be due to nutrient immobilization consequent to the formation of complexes in the soil after degradation and uptake [32]. For organic matter, it was increased with increasing crude oil either alone or in combination with inorganic fertilizers. The increment in the total carbon content of the soil with increasing concentration of crude oil may be attributed to the high amount of carbon in the oil. This could have been converted to soil organic carbon. Similar findings have been reported by Benka-Coker and Ekundayo [55]. This observation also agrees with the findings of Ekundayo and Obuekwe [56] who noted increment of organic carbon content of oil polluted soils in Southern Nigeria. It may also be related to slow decomposition rate of the amendment by soil organisms science contamination of the soil with crude oil might have resulted in poor soil aeration. The decrease in total nitrogen with increasing of crude oil levels may be due to temporal immobilization of this nutrient by microbes resulting in an increment in bacterial population as shown in Table 3.

Table 1. Effect of petroleum oil concentration and inorganic fertilizers (IF) on MDA in leaves of jojoba plant

Patroloum oil concentration (9/)	MDA (mg FW <sup>-1</sup> ) in leaves grown in soil with					
Petroleum oil concentration (%)	Petroleum oil only	Petroleum oil + IF				
(0) Control	11.23 ± 0.65	10.73 ± 0.88				
1	13.03 ±1.02	08.98 ±1.55				
2	16.07** ± 1.66	11.53 ± 1.47				
3	24.26** ± 2.31	16.33**± 1.12				
L.S.D. at 5%	2.46	2.46				
L.S.D. at 1%	3.39	3.39				

Values are given as mean  $\pm$  SD, (\*) significant difference at P=0.05 and (\*\*) highly significant difference at P=0.01

Table 2. Effect of petroleum oil (PO) concentrattion and inorganic fertilizers (IF) on chemical soil analysis of jojoba plant

Treatments	N (%)	P (PPM)	K (PPM)	OM (%)	Metal ions c	C/N			
					Cu	Fe	Mn	Zn	<del>.</del>
Control	14.0±1.3	0.0±0.0	50.2±4.2	0.1±0.03	0.17±0.11	0.93±0.25	0.09±0.03	0.17±0.04	0.01±0.00
PO (1%)	7.0±1.1 **	$0.0 \pm 0.0$	55.7±3.9	1.4±0.20*	0.16±0.12	0.50±0.38	0.11±0.07	0.15±0.02	0.20±0.04*
PO (2%)	5.6±0.8**	$0.0 \pm 0.0$	44.3±3.6	2.2±0.71**	0.50±0.22**	0.76±0.44	0.36±0.13**	0.33±0.14	0.39±0.12**
PO(3%)	9.8±1.2**	$0.0 \pm 0.0$	39.6±2.3**	3.4±0.60**	0.17±0.07	0.78±0.23	0.21±0.07	0.17±0.09	0.35±0.11**
Control+IF	26.6±2.7**	2.7±0.5**	71.6±5.7**	$0.0 \pm 0.0$	0.16±0.09	1.05±0.67	0.13±0.06	0.17±0.06	$0.0 \pm 0.0$
PO (1%)+IF	12.6±0.9	1.8±0.3**	53.1±4.8	1.3±0.45*	0.21±0.14	0.89±0.43	0.30±0.11*	0.20±0.12	0.10±0.03
PO (2%)+IF	8.4±1.4**	0.7±0.1*	51.8±2.7	2.3±0.30**	0.24±0.14	1.00±0.53	0.40±0.14**	0.25±0.16	0.27±0.09**
PO (3%)+IF	8.4±1.9**	1.6±0.7**	45.5±5.1	4.3±1.1**	0.15±0.05	0.72±0.27	0.16±0.02	0.14±0.04	0.51±0.23**
L.S.D at 5%	2.642	0.561	7.239	0.948	0.219	0.734	0.155	0.164	0.182
L.S.D at 1%	3.641	0.773	9.975	1.308	0.302	1.011	0.213	0.226	0.250

Values are given as mean ± SD, (\*) significant difference at P=0.05 and (\*\*) highly significant difference at P=0.01

C/N ratio is narrowed while this ratio increased with increasing crude oil concentration and this was reflected on bacterial population biomass (Table 3). C/N ratio recorded higher percentage in oil contaminated soil than in soil contaminated with inorganic fertilizers except at 3%. An earlier report by Jobson et al. [57] stated that oil spills on land resulted in an imbalance in the carbon: nitrogen ratio, which, if greater than 17:1 in soils resulted in net immobilization of the nutrients by microbes leading to loss of soil fertility. As clear in Table 3, the bacterial count at the end of the experiment, for control, there was a non significant difference with the beginning of the experiment, followed by a drastic increase at 1% and 2% crude oil either alone or in combination with inorganic fertilizers. The high value was recorded at 1% oil in combination with inorganic fertilizers. When an oil spill occurs, the result is a large increase in carbon and this also stimulates the growth of the already present oil degrading microorganisms [58,59]. However, these microorganisms are limited in the amount of growth and remediation that can occur by the amount of available nitrogen and phosphorous by adding these supplemental nutrients in the proper concentrations, the hydrocarbon degrading microbes are capable of achieving their maximum rate of pollutant uptake. The ability of microorganisms for petroleum oil degradation depends on a number of factors which include temperature, sunlight irradiation, pH, viscosity of the oil, coarseness of the soil, the level of the oil in the environment and in adequate mineral nutrient, especially nitrogen and phosphorous often limits the growth of hydrocarbon utilizing microorganisms [60].

Table 3. Effect of petroleum oil (PO) concentration and inorganic fertilizers (IF) on bacterial count in root rhizosphere of jojoba plant

Treatments	No of bacteria × 10 <sup>7</sup> ( CFU/g sand)							
	At the beginning of experiment	At the end of experiment						
Control	3.1 ± 2.082	3.3 ± 4.359						
PO (1%)	3.8 ± 3.000 *	7.5 ± 3.512 **						
PO (2%)	4.0 ± 3.512 **	7.5 ± 5.000 **						
PO (3%)	4.3 ± 2.517 **	$3.9 \pm 3.000$						
Control +IF	$3.4 \pm 4.000$	$3.4 \pm 4.509$						
PO (1%) +IF	4.4 ± 3.606 **	27.2 ± 6.658 **						
PO (2%) +IF	4.4 ± 4.726 **	15.7 ± 6.658 **						
PO (3%) +IF	$3.5 \pm 4.509$	5.2. ± 2.309 **						
L.S.D. at 5%	0.66	0.82						
L.S.D. at 1%	0.86	1.13						

Values are given as mean ± SD, (\*) significant difference at P=0.05 and (\*\*) highly significant difference at P=0.01

Electric conductivity of the leaves after growth of jojoba seedling in petroleum oil contaminated soil increased significantly with increasing crude oil concentration in the soil either alone or in combination with inorganic fertilizers as shown in Table 4. This is in agreement with those obtained by Caol et al. [61] who showed an increase in the relative conductivity, injury index MDA and proline levels in the leaves of the oil palm seedlings when compared with the control under low temperature and drought stress. The application of water stress markedly increased the electrolyte leakage of snapdragon plants [62,63]. Moreover, In *Arabidopsis*, it has been established that one of the immediate effects of osmotic stress, hyperosmotic or hypo-osmotic, is the change in the electric properties of the plasma membrane [64]. These changes occur within 1 min, whereas expression of AtMEKK1, a component of a mitogen-activated protein kinase cascade responsible for

activating osmoresponses, occurs within about 5 min [65]. In the case of hyperosmotic shock, these rapid changes are followed by turgor recovery, a process that takes about 30 min to complete, and is caused by large changes in the net ion fluxes across the plasma membrane [66].

Table 4. Effect of Petroleum oil (PO) concentration and inorganic fertilizers (IF) on electric conductivity (EC) in leaves of jojoba plant

Treatments	EC <sub>1</sub>	EC <sub>2</sub>	EC <sub>1</sub> / EC <sub>2</sub> (%)
Control	42.03 ± 3.42	346.76± 37.21	12.12 ± 0.31
PO (1%)	66.35± 4.67 **	389.3± 40.53	17.04 ± 0.92**
PO (2%)	65.80± 2.96 **	343.0± 22.75	19.18 ± 0.41**
PO (3%)	121.00± 6.37**	461.0± 32.94**	26.24 ± 0.49**
Control +IF	56.96 ± 4.86 **	428.6± 25.67**	13.28 ± 0.34*
PO (1%) +IF	49.96± 3.67 **	294.5± 16.31*	16.96 ± 0.31**
PO (2%) +IF	57.96± 5.32 *	296.5± 27.15*	19.54 ± 0.01**
PO (3%) +IF	56.16± 2.57 **	274.6± 19.75**	20.45 ± 0.35**
L.S.D. at 5%	7.62	50.03	0.81
L.S.D. at 1%	10.49	68.93	1.12

Values are given as mean ± SD, (\*) significant difference at P=0.05 and (\*\*) highly significant difference at P=0.01

The changes in the ionic contents of K, Na, Ca and Mg were recorded in both roots and shoots in Table 5. There is an observed increase in Na content in root of jojoba plant with increasing crude oil concentration either alone or in combination with inorganic fertilizers (two treatments). Na content in case of contamination with crude oil only was more than that of soil contamination with crude oil and inorganic fertilizers. Meanwhile there was a decrease in Na content in shoots of jojoba plants with increasing crude oil concentration at two treatments. This indicates that, Na was accumulated and retained in roots, and prevented from translocation to shoots. For potassium, a reverse situation was obtained, where the translocation of K to shoots was higher than the accumulation in the roots. In general, the addition of inorganic fertilizers improved the K content in both roots and shoots of jojoba plants. For magnesium, there was a significant decrease in its content in both shoots and roots with the increment of crude oil at two treatments. Mg content in treatments of crude oil with inorganic fertilizers was more than that with crude oil only. For Ca, its content was increased in the roots however it significantly decreased in roots and shoots with increasing crude oil concentration at two treatments. Accumulation of Cu, Mn, Cd and Pb was high at 2% in roots, while Zn content decreased with increasing crude oil concentration as shown in Table 6. Translocation of Cu, Mn, Zn, Cd and Pb recorded a high content at 3% crude oil. In this respect, there are some reports stated that, crude oil in soil makes the soil condition unsatisfactory for plant growth due to the reduction in the level of available plant nutrient or a rise in toxic levels of certain elements such as iron and zinc [67,68]. The results obtained by John et al. [69] suggest that roots of Brassica juncea L. are efficient barriers to Cd and Pb translocation to the above ground plant parts. The uptake of metals by plant is coupled to a chemiosmotic process across the membrane of intact root cells [70]. It has been shown that Pb is unevenly distributed in roots, where different root tissues act as barriers to apoplastic and symplastic Pb transport and hence Pb transport to shoot gets restricted. Although tolerance and root to shoot metal transport are often negatively correlated, and tolerance is often associated with enhanced metal retention in roots, it does not necessarily mean that increased root retention itself could be the cause of tolerance [71].

Table 5. Effect of petroleum oil (PO) concentration and inorganic fertilizers (IF) on Na, K, Mg and Ca contents (mg g<sup>-1</sup> DW) in roots and shoots of jojoba plant

Treatments	Inorganic cation contents (mg g <sup>-1</sup> DW)									
	Na		K		Mg		Ca			
	Roots	Shoots	Roots	Shoots	Roots	Shoots	Roots	Shoots		
Control	16.6±2.3	20.4±2.1	18.5±1.7	20.0±2.3	20.2±2.0	47.0±3.8	13.1±1.2	20.5±2.0		
1PO (1%)	13.2±1.4	21.4±2.2	16.4±1.9	23.9±3.5	16.6*±2.4	38.5**±1.9	13.5±0.7	18.7±1.6		
PO (2%)	19.6±2.4	15.2**±2.0	11.5**±2.1	29.7**±1.5	13.7**±1.5	37.6**±2.7	20.0**±1.8	10.2**±1.6		
PO (3%)	34.3**±1.8	13.9**±2.7	9.3**±0.89	58.8**±2.9	9.8**±1.6	16.1**±0.9	20.8**±2.9	9.2**±0.90		
Control +IF	15.7±1.6	18.4±1.5	22.3**±2.4	21.6±1.9	21.3±2.2	50.1±1.8	25.2**±2.1	23.9**±1.5		
PO (1%) +IF	13.9±2.1	20.8±1.8	33.1**±2.1	29.5**±2.1	20.9±2.7	46.7±1.5	32.2**±2.0	17.2*±2.2		
PO (2%) +IF	16.6±2.4	20.2±3.0	20.8±2.6	35.5**±2.6	19.7±2.5	44.4±2.4	20.4**±1.7	11.4**±1.2		
PO (3%) +IF	26.1**±2.03	12.9**±1.3	15.6±1.7	61.0**±2.5	15.8*±1.6	26.5**±1.6	29.1**±2.4	13.6**±1.5		
L.S.D. at 5%	3.5	3.7	3.4	4.3	3.6	3.9	3.4	2.7		
L.S.D. at 1%	4.8	5.1	4.7	5.9	5.0	5.3	4.7	3.8		

Values are given as mean ± SD, (\*) significant difference at P=0.05 and (\*\*) highly significant difference at P=0.01

Table 6. Effect of petroleum oil concentration (PO) and inorganic fertilizers (IF) on heavy metals contents (mg g<sup>-1</sup> DW) in roots and shoots of jojoba plant

Treatments	Heavy metals concentration (mg g-1 DW)									
	Cu		Mn		Zn		Cd		Pb	
	Roots	Shoots	Roots	Shoots	Roots	Shoots	Roots	Shoots	Roots	Shoots
Control	0.138±0.012	0.020±0.002	0.284±0.008	0.081±0.003	0.404±0.021	0.098±0.004	0.056±0.005	0.024±0.004	0.020±0.002	0.090±0.010
PO ( <b>1</b> %)	0.125*±0.006	0.045**±0.004	0.590**±0.018	0.111**±0.010	0.396±0.023	0.161**±0.012	0.041**±0.003	0.035**±0.003	0.024*±0.003	0.106*±0.007
PO (2%)	0.164**±0.005	0.090**±0.007	0.723**±0.005	0.128**±0.005	0.334**±0.024	0.211**±0.003	0.066*±0.005	0.049**±0.004	0.036**±0.005	0.158**±0.005
PO (3%)	0.154**±0.005	0.129**±0.006	0.518**±0.009	0.259**±0.004	0.268**±0.019	0.305**±0.008	0.098**±0.011	0.061**±0.002	0.028**±0.003	0.154**±0.011
Control+IF	0.140±0.011	0.016±0.002	0.364**±0.002	0.104**±0.009	0.400±0.019	0.110±0.004	0.055*±0.006	0.019*±0.001	0.020±0.002	0.082±0.006
PO (1%) +IF	0.198**±0.008	0.041**±0.003	0.552**±0.002	0.153**±0.005	0.509**±0.016	0.198±0.009	0.035**±0.002	0.031**±0.003	0.021±0.002	0.095±0.003
PO (2%) +IF	0.175**±0.011	0.066**±0.005	0.635**±0.008	0.291**±0.008	0.421±0.021	0.239**±0.012	0.068**±0.003	0.051**±0.004	0.030**±0.003	0.150**±0.015
PO (3%) +IF	0.160**±0.002	0.098**±0.007	0.488**±0.012	0.358**±0.021	0.381±0.005	0.305**±0.014	0.089**±0.002	0.056**±0.003	0.014**±0.001	0.143**±0.006
L.S.D. at 5%	0.014	0.008	0.020	0.017	0.034	0.016	0.009	0.005	0.004	0.015
L.S.D. at 1%	0.019	0.011	0.028	0.023	0.047	0.022	0.012	0.008	0.006	0.021

Values are given as mean ± SD, (\*) significant difference at P=0.05 and (\*\*) highly significant difference at P=0.01

Furthermore, the translocation of Cd from root to shoot is an important factor affecting accumulation of this metal in aerial tissues of *Brassica juncea*. Our results are in confirmation with that of Zhu et al. [72] who also observed higher accumulation of Cd into the roots of *B. juncea* as compared to above ground parts by over expressing gammaglutamylecyteine synthetase. Cd accumulation in stem and inactivation in root cells are probably related to its binding in cell walls, compartmentalization in vacuoles and complexation with metal binding proteins and peptides, especially phytochelatin and metallothioneins [73]. These processes are strategies employed by plants, at least in part, to face unavoidable stress conditions.

Plants are highly susceptible to oil exposure and this may kill them within a few weeks to several months. There are several vegetal species that are capable of growing in soils polluted with hydrocarbons and they participate in their degradation through the rhizosphere. Part of the root which favors the growth of several microorganisms [74] and increases biomass and microbial activity, accelerating degradation processes [75].

#### 4. CONCLUSIONS

Jojoba is a promising plant for growth in desert. It tolerates heat, drought and salts. Also, its oil is used in pharmaceutical industry.

In this study, the effect of petroleum crude oil contaminated soil on the growth; nutrient elements content and heavy metal uptake of jojoba were studied. The changes in soil properties and the rhizosphere bacterial biomass were also studied.

In the present study, Na, Mg and Ca ions decreased in shoots of jojoba planted in crude oil contaminated soil while, K ion increased. In contrast, Na and Ca ions increased while K and Mg decreased in the jojoba roots. Also, Cu, Mn, Cd and Pb accumulated in jojoba plant while, Zn ion concentration decreased.

In the crude oil contaminated soil, Na and K decreased however, Cu, Fe, Mn, Zn and organic matter increased in comparison with non contaminated soil. Also, Bacterial biomass was significantly increased at the end of the experiment.

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#### **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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