# Effect of Salicylic Acid and Root Inoculation with *P. indica* on Cd Phytoremediation Efficiency and Degradation of Soil Fuel Oil in the Salinity Stress

Amir Hossein Baghaie, Amir Daliri

Department of Soil Science, Arak Branch, Islamic Azad University, Arak, Iran ORCID: Amir Hossein Baghaie: http://orcid.org/0000-0002-8936-1202

## Abstract

**Aim:** This study was done to investigate the effect of salicylic acid (SA) and root inoculation with *Piriformospora indica* on Cd phytoremediation efficiency and degradation of soil fuel oil by sunflower in the salinity stress. **Materials and Methods:** Treatments consisted of soil Cd pollution (0 and 5 mg Cd/kg soil), soil salinity adjusted at the EC equal to 4 and 6 dS/m, and soil pollution to fuel oil at the rates of 0%, 4%, and 8% (W/W). Foliar application of SA at the rates of 0 and 1.5 mmol/l was done 2 weeks after sunflower seedling growth in the presence and absence of *P. indica*. After 60 days, the plant was harvested and soil and plant Cd concentration was measured using atomic absorption spectroscopy. Soil microbial activity and degradation of soil fuel oil were also determined according to the Besaltpour *et al.* method. **Results:** The greatest shoot Cd concentration belonged to the saline soil (6 dS/m) polluted with 10 mg Cd/kg soil and polluted with 8% (W/W) fuel oil, whereas the lowest was observed in the nonsaline soil polluted with 5 mg Cd/kg soil. Increasing soil salinity from 4 to 6 dS/m significantly decreased the soil microbial respiration by 8.3%. Application of 1.5 mmol/l SA significantly increased the degradation of soil fuel oil by 11.3%. **Conclusion:** The interaction effect of *P. indica* and SA had a significant effect on increasing the degradation of soil fuel oil.

Keywords: Fuel oil, Piriformospora indica, phytoremediation, salicylic acid

## INTRODUCTION

Extension of economic activities in recent decades increased the soil contaminated with petroleum products considerably.<sup>[1]</sup> It is possible to remedy the contamination of petroleum hydrocarbon in soils using different chemical and physical approaches such as extraction of gaseous or liquid matter, soil washing, and solidification, and stabilization.<sup>[2]</sup> However, unfortunately, using these methods is very expensive and time-consuming. Therefore, environmental experts are interested to develop some environment-friendly methods such as phytoremediation which are less expensive and use less energy. However, the efficiency of phytoremediation is considerably affected by soil physicochemical characteristics and the contaminant types.<sup>[3]</sup>

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Nowadays, contamination with petroleum compounds, including crude oil, is one of the most important environmental pollutants today that can endanger human health, and removal of such compounds in soil environment seems necessary. Zamani *et al.* investigated the effect of *Piriformospora indica* on the root development of maize (*Zea mays* L.) and remediation of soil contaminated with petroleum hydrocarbons and concluded that inoculation with *P. indica* can enhance phytoremediation efficiency of petroleum hydrocarbons.<sup>[4]</sup> In addition, they mentioned that soil contamination with petroleum hydrocarbons can severely

Address for correspondence: Dr. Amir Hossein Baghaie, Department of Soil Science, Arak Branch, Islamic Azad University, Arak, Iran. E-mail: a-baghaie@iau-arak.ac.ir

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inhibit plant root growth and consequently limit the efficacy of phytoremediation.<sup>[4]</sup> In general, root growth and distribution is an important factor to be considered, especially when the contamination is not evenly distributed in soil, usually the case in the real-world situations. Hence, it is necessary to consider the role of other abiotic stresses such as heavy metals or soil salinity on phytoremediation efficiency.<sup>[5]</sup>

Nowadays, heavy metal pollution is increasing in industrial areas and agricultural soils that can damage human health through different pathways such as the consumption of agricultural products which were grown in contaminated soils. The stable and non-biodegradable properties of heavy metals cause their accumulation in the human body and damage of vital organs such as bones, liver, and kidney.<sup>[6]</sup> Unfortunately, in arid and semi-arid regions, the salinity problem is increasing due to changing climatic conditions. The simultaneous effect of salinity and heavy metals can affect the availability of soil heavy metals or petroleum hydrocarbons; thus, remediation of these soils is very difficult. Since the phytoremediation can remove organic and inorganic compounds from the contaminated soils, finding an appropriate way to increase plant biomass to increase soil heavy metal remediation or degradation of petroleum hydrocarbons in the soils is necessary.<sup>[5]</sup>

*P. indica* is a cultivable root-colonizing entophytic fungus. *P. indica* can enhance the plant growth and biomass production, increase the nutrient uptake by the plant, and improve the resistance of co-cultivated plants to different biotic and abiotic stresses such as salinity or heavy metal contamination. As *P. indica* has been reported to colonize a wide range of plant species under various environmental and nutrient stress conditions, using this method is useful in different conditions. However, due to the simultaneously of several contaminants or stresses, inoculation alone is not an appropriate method. Aslam *et al.* reported that *P. indica* inoculation can improve P-uptake, enhance crop performance, and decrease the negative effects of heavy metals.<sup>[7]</sup> Mohd *et al.* reported that *P. indica* caused the protection of host from heavy metal toxicity. However, they only investigated the role of the heavy metal toxicity.<sup>[8]</sup>

Since plant inoculation alone is not a suitable strategy to reduce the abiotic stress, we should consider an appropriate strategy to help increase plant resistance to stress. It is noteworthy that in arid and semi-arid regions due to the low organic matter, salinity has a greater impact on plant biomass reduction. Previous studies are showing that using plant-promoting growth such as salicylic acid (SA) can help to increase plant resistance to abiotic stresses such as salinity. However, the amount of soil salinity and other chemical properties can determine the phytoremediation efficiency and the amount of degradation of soil petroleum hydrocarbons.<sup>[9]</sup> El-Katon *et al.* investigated the effect of SA on corn productivity and concluded that application timing of SA affects the response of maize (*Z. mays* L.) hybrids to salinity stress.<sup>[10]</sup> To diminish the impact of abiotic stress on plant growth, several plant growth promoters such as glycine betaine and SA have been used at different stages of plant development. However, the SA application efficiency depends on several factors such as its dose and plant species.<sup>[11]</sup>

On the other hand, soil salinity can increase the soil heavy metal availability and thus decrease plant biomass that is a negative point in environmental studies. Raiesi and Sadeghi conducted the interactive effect of salinity and Cd toxicity on soil microbial properties and enzyme activities and concluded that increasing soil salinity has a direct effect on soil Cd concentration and can decrease soil microbial activity.[12] Very few studies have focused on soil biochemical processes and microbial responses to multiple environmental stresses such as salinity, petroleum hydrocarbons, and metal pollution. However, it seems that plant resistance to heavy metal, petroleum hydrocarbons, and salinity has decreased with increasing these parameters alone or in combination. Therefore, it is necessary to study the strategy of increasing plant resistance against several abiotic stresses which have simultaneously a negative influence on a plant. Considering sunflower has a high biomass and heavy metal hyperaccumulators,<sup>[13,14]</sup> its cultivation in organic and inorganic polluted soil can remediate soils from pollutions. However, its remediation efficiency is dependent on many parameters such as soil chemical properties. Thus, this research was done to evaluate the effect of SA and root inoculation with P. indica on sunflower phytoremediation efficiency and degradation of soil petroleum hydrocarbon in a soil treated with cadmium and fuel oil in the salinity stress.

## **MATERIALS AND METHODS**

To investigate the effect of SA and root inoculation with *P. indica* on Cd phytoremediation efficiency by sunflower and degradation of soil petroleum hydrocarbon, a factorial experiment in the layout of randomized completely block design at three replication was design. Treatments (72 treatments) consisted of Cd pollution at the rates of 0 (Cd0) and 5 (Cd5) mg Cd/kg soil, soil salinity adjusted at the EC equal to 4 (S<sub>1</sub>) and 6 (S<sub>2</sub>) dS/m, and soil pollution to fuel oil at the rates of 0 (F0), 4 (F4) and 8 (F8) % (W/W). Foliar application of SA at the rates of 0 (SA<sub>0</sub>) and 1.5 (SA<sub>1.5</sub>) mmol/l was done 2 weeks after sunflower (*Helianthus annuus* L.) seedling growth in the presence (*P. indica* [+]) and absence of *P. indica* (*P. indica* [-]).

The saline soil was collected from Roudasht agricultural research station, Isfahan, Iran. The initial soil EC was 6 dS/m and selected as a saline soil. To prepare the nonsaline soil, the saline soil was washed with deionized water until the soil drainage EC was decreased<sup>[15]</sup> to 4 dS/m (S<sub>1</sub>). The selected physicochemical properties of studied soil are shown in Table 1.

According to the mentioned treatments, the soil was polluted with the mentioned Cd levels and incubated for 2 weeks to equilibrium. After that, the soil was polluted with fuel oil at the rates of 0%, 4%, and 8% (W/W) and incubated for 2 weeks to equilibrium.

Three days after seed germination on filter paper, the sunflower seedlings were transferred into 5 kg plastic pots (two seedlings in each pot) and filled with the treated soil. Two fungal plugs of 10 mm in diameter were placed at a distance of 1 cm below the sunflower seedlings in the soil at sowing time. *P. indica* was obtained from the soil biology of water and soil research institute. *P. indica* was cultured in Petri dishes on a modified Kafer medium.<sup>[16]</sup>

Two weeks after plant germination, the SA foliar application was done. After 60 days of the experiment, plants were harvested and plant Cd concentration<sup>[17]</sup> was measured using atomic absorption spectroscopy (AAS). Soil microbial respiration was measured as evolved  $CO_2$ . For this purpose, three replicate soil samples of each treatment were incubated for 3 days at 26°C in 250-ml glass containers closed with rubber stoppers. The evolving  $CO_2$  was trapped in NaOH solution, and the excess in alkali was then titrated with HCl. Three glass containers with NaOH but without soil were also used as controls.<sup>[18]</sup>

The statistical analysis was done using SAS software V. 9.1. The least significant difference test was used to determine the differences between the means. The 95% (P = 0.05) probability value was considered to determining the significant difference.

## RESULTS

The greatest soil Cd concentration has belonged to soil with the greatest level of soil salinity and petroleum hydrocarbons in the presence of *P. indica*, whereas the lowest has belonged to the nonsaline soil with the lowest

Table 1: Selected some physicochemical properties ofsoil studied

Characteristic	Unit	Amount
Sand	%	80
Silt	%	10
Clay	%	20
pН	-	7.1
EC	dS/m	6
OC	%	0.1
Total Pb	mg/kg	ND
Total Zn	mg/kg	55
Total Cd	mg/kg	ND

ND: Not detectable by atomic absorption spectroscopy

amount of soil fuel oil in the absence of *P. indica*. Based on the results of this study, soil salinity from 4 to 6 dS/m significantly increased the soil Cd concentration by12.2% that is a negative point in environmental pollution [Table 2]. The soil Cd concentration in non-Cd polluted was not detectable by AAS.

The greatest shoot Cd concentration belonged to the saline soil (6 dS/m) polluted with 10 mg Cd/kg soil and polluted with 8% (W/W) fuel oil, whereas the lowest was observed in the nonsaline soil polluted with 5 mg Cd/kg soil [Table 3]. The shoot Cd concentration in non-Cd polluted was not detectable by AAS. Increasing soil salinity has increased the shoot Cd concentration, as increasing soil salinity from 4 to 6 dS/m significantly increased the shoot Cd concentration by 5.5% in the Cd-polluted soil (5 mg Cd/kg soil)-treated soil that received 8% (W/W) fuel oil. However, the plant biomass was decreased by 3% [Table 4]. Increasing soil pollution to fuel soil significantly increased and decreased the shoot Cd concentration and plant biomass, respectively.

The greatest soil microbial respiration has belonged to the soil with the lowest soil salinity and 8% (W/W) fuel oil, whereas the lowest was observed in the soil without receiving any fuel oil and the greatest level of soil salinity [Table 5]. Increasing soil Cd concentration and soil salinity significantly decreased the soil microbial respiration. Based on the results of this study, increasing soil salinity from 4 to 6 dS/m significantly decreased the soil microbial respiration by 8.3%. However, increasing soil pollution to fuel oil significantly increased the soil microbial respiration, as a significant increase by 18% was observed, when the soil pollution to fuel was increased from 4% to 8% (W/W).

The greatest degradation of soil fuel oil has belonged to the nonsaline, no Cd-polluted soil that contains 8% (W/W) fuel oil under cultivation of *P. indica*-inoculated plant which received SA foliar application [Table 6]. Regardless of soil salinity or Cd pollution, SA foliar application significantly increased the soil fuel oil degradation. Application of 1.5 mmol/l SA significantly increased the degradation of soil fuel oil by 11.3%, whereas the soil fuel degradation was decreased when the soil pollution to Cd increased.

Table 2:	Table 2: Effect of treatments on soil Cd concentration (mg/kg soil)											
Fuel oil (%)	Cd concentration (mg/kg soil)	tion <i>P. indica</i> (+)					P. indica (—)					
		S <sub>1</sub> SA <sub>0</sub>	<b>S</b> <sub>1</sub> <b>SA</b> <sub>1.5</sub>	S <sub>2</sub> SA <sub>0</sub>	<b>S</b> <sub>2</sub> <b>SA</b> <sub>1.5</sub>	S <sub>1</sub> SA <sub>0</sub>	<b>S</b> <sub>1</sub> <b>SA</b> <sub>1.5</sub>	S <sub>2</sub> SA <sub>0</sub>	<b>S</b> <sub>2</sub> <b>SA</b> <sub>1.5</sub>			
F0	Cd0	ND	ND	ND	ND	ND	ND	ND	ND			
	Cd5	4.00p	4.11o	4.25m	4.39h	3.72r	3.92q	3.92q	4.19n			
F4	Cd0	ND	ND	ND	ND	ND	ND	ND	ND			
	Cd5	4.271	4.36i	4.51e	4.62c	4.11o	4.00p	4.33j	4.42g			
F8	Cd0	ND	ND	ND	ND	ND	ND	ND	ND			
	Cd5	4.44f	4.54d	4.65b	4.72a	4.31k	4.44f	4.51e	4.62c			

Means with the similar letters are not significant (P=0.05), ND: Not detectable. P. indica: Piriformospora indica

Fuel oil (%)	Cd concentration	P. indica (+)				P. indica (—)			
	(mg/kg soil)	S <sub>1</sub> SA <sub>0</sub>	<b>S</b> <sub>1</sub> <b>SA</b> <sub>1.5</sub>	S <sub>2</sub> SA <sub>0</sub>	<b>S</b> <sub>2</sub> <b>SA</b> <sub>1.5</sub>	S <sub>1</sub> SA <sub>0</sub>	<b>S</b> <sub>1</sub> <b>SA</b> <sub>1.5</sub>	S <sub>2</sub> SA <sub>0</sub>	<b>S</b> <sub>2</sub> <b>SA</b> <sub>1.5</sub>
F0	Cd0	ND	ND	ND	ND	ND	ND	ND	ND
	Cd5	3.22t	3.39r	3.48q	3.680	3.12u	3.22t	3.37s	3.54p
F4	Cd0	ND	ND	ND	ND	ND	ND	ND	ND
	Cd5	4.00k	4.11h	4.12g	4.39b	3.77n	4.00k	3.891	4.00k
F8	Cd0	ND	ND	ND	ND	ND	ND	ND	ND
	Cd5	4.10i	4.17f	4.31d	4.61a	3.85m	4.09j	4.18e	4.34c

Means with the similar letters are not significant (P=0.05), ND: Not detectable by AAS, AAS: Atomic absorption spectroscopy, P. indica: Piriformospora indica

## Table 4: Effect of treatments on plant biomass (g)

Fuel oil (%)	Cd concentration	P. indica (+)				P. indica (-)			
	(mg/kg soil)	S <sub>1</sub> SA <sub>0</sub>	S <sub>1</sub> SA <sub>1.5</sub>	S <sub>2</sub> SA <sub>0</sub>	<b>S</b> <sub>2</sub> <b>SA</b> <sub>1.5</sub>	S <sub>1</sub> SA <sub>0</sub>	S₁SA <sub>1.5</sub>	S <sub>2</sub> SA <sub>0</sub>	<b>S</b> <sub>2</sub> <b>SA</b> <sub>1.5</sub>
F0	Cd0	6.4c	6.7a	6.2e	6.4c	6.1f	6.3d	5.8i	6.1f
	Cd5	6.3d	6.5b	6.1f	6.2e	6.0g	6.2e	5.7j	6.0g
F4	Cd0	6.1f	6.2e	5.7j	5.9h	5.7j	5.8i	5.4m	5.7j
	Cd5	5.6k	5.8i	5.3n	5.4m	5.3n	5.4m	5.0p	5.3n
F8	Cd0	5.20	5.51	4.9q	5.0p	4.8r	5.0p	4.5t	4.8r
	Cd5	5.0p	5.3n	4.5t	4.8r	4.5t	4.6s	4.0v	4.2u

Means with the similar letters are not significant (P=0.05). P. indica: Piriformospora indica

Fuel	Cd concentration	P. indica (+)					P. indica (—)			
oil (%)	(mg/kg soil)	S <sub>1</sub> SA <sub>0</sub>	<b>S</b> <sub>1</sub> <b>SA</b> <sub>1.5</sub>	S <sub>2</sub> SA <sub>0</sub>	<b>S</b> <sub>2</sub> <b>SA</b> <sub>1.5</sub>	S <sub>1</sub> SA <sub>0</sub>	S <sub>1</sub> SA <sub>1.5</sub>	S <sub>2</sub> SA <sub>0</sub>	<b>S</b> <sub>2</sub> <b>SA</b> <sub>1.5</sub>	
F0	Cd0	8.39p	8.450	8.12u	8.31r	8.22t	8.31r	8.15u	8.26s	
	Cd5	8.22t	8.34q	8.00v	8.12u	8.12u	8.25s	8.00v	8.13u	
F4	Cd0	10.11g	10.25e	9.92j	10.08h	10.00i	10.11g	9.811	9.88k	
	Cd5	10.00i	10.19f	9.76m	9.811	9.92j	10.00i	9.68n	9.76m	
F8	Cd0	10.44b	10.59a	10.31d	10.39c	10.31d	10.31d	10.19f	10.22e	
	Cd5	10.31d	10.37c	10.12g	10.18f	10.17f	10.22e	10.00i	10.11g	

Means with the similar letters are not significant (P=0.05). P. indica: Piriformospora indica

Table 6:	Table 6: Effect of treatments on soil fuel oil degradation (%)										
Fuel oil (%)	Cd concentration (mg/kg soil)	on <i>P. indica</i> (+)					P. indica (—)				
		S <sub>1</sub> SA <sub>0</sub>	<b>S</b> <sub>1</sub> <b>SA</b> <sub>1.5</sub>	S <sub>2</sub> SA <sub>0</sub>	<b>S</b> <sub>2</sub> <b>SA</b> <sub>1.5</sub>	S <sub>1</sub> SA <sub>0</sub>	<b>S</b> <sub>1</sub> <b>SA</b> <sub>1.5</sub>	S <sub>2</sub> SA <sub>0</sub>	<b>S</b> <sub>2</sub> <b>SA</b> <sub>1.5</sub>		
F0	Cd0	ND	ND	ND	ND	ND	ND	ND	ND		
	Cd5	ND	ND	ND	ND	ND	ND	ND	ND		
F4	Cd0	55.4k	57.3j	52.6m	53.11	52.5m	55.2k	49.6p	47.3q		
	Cd5	57.9j	55.1k	50.1o	51.4n	49.5p	52.5m	42.2s	45.4r		
F8	Cd0	66.9c	70.4a	63.7f	65.9d	64.1f	68.5b	62.3g	64.2f		
	Cd5	64.3f	68.6b	60.2h	63.6f	62.1g	64.1f	59.4i	62.6g		

Means with the similar letters are not significant (P=0.05), ND: Not detectable, P. indica: Piriformospora indica

## DISCUSSION

Based on the results of this study, increasing soil salinity significantly increased the soil Cd concentration that may be related to the role of soil salinity on soil Cd solubility.<sup>[19]</sup> Raiesi and Sadeghi investigated the effect of soil salinity on soil Cd concentration and concluded that NaCl salinity can enhance the solubility and mobility of toxic metals and thus their bioavailability in metal-polluted soils by affecting metal chemistry, speciation, and distribution of heavy metals.<sup>[20]</sup> Increasing soil heavy metal availability in saline soils is predominantly related to the formation heavy metals metal complexes with soil inorganic ligands.<sup>[21]</sup> Accordingly, the greatest soil Cd concentration belonged to the soil polluted with the greatest level of salinity. However, increasing soil salinity concentration has an adverse effect on plant biomass [Table 4]

and thereby can decrease phytoremediation efficiency [Table 3]. The important point is that as the concentration of soil petroleum hydrocarbons increases, the soil Cd availability also increases which may be attributed to the role of petroleum hydrocarbon compounds in increasing soil salinity and consequently increasing soil Cd solubility. On the other hand, the functional group of petroleum hydrocarbon compounds has a negative charge<sup>[22]</sup> which may increase the soil Cd availability.

Plant inoculation with *P. indica* significantly increased the plant resistance to abiotic stresses such as heavy metal or salinity, and thus, plant Cd phytoremediation can increase [Table 3]. Based on the results of this study, plant inoculation with *P. indica* significantly increased plant biomass in Cd-polluted soil (5 mg Cd/kg soil) by 12.2% with the soil salinity equal to 6 dS/m. Das *et al.* reported that inoculation with *P. indica* can lead to early flowering, higher biomass, and altered secondary metabolites of the medicinal plant.<sup>[23]</sup> Shahabivand *et al.* also reported that the cadmium toxicity in *H. annuus* can be modulated by endosymbiotic fungus (*P. indica*).<sup>[24]</sup> It is mentioned that *P. indica* is easily cultivable, lacks host specificity, and interacts with many different plant species and this can help diminish the negative effects of abiotic stresses.<sup>[25]</sup>

Based on the results of this study, soil pollution to fuel oil acts as a carbon source for microorganism rather than toxicity, as increasing the soil pollution to fuel oil from 0% to 8% (W/W) significantly increased the soil microbial respiration by 14.6%. However, soil salinity and Cd had an adverse effect on soil microbial respiration. Jurelevicius *et al.* investigated the bacterial community response to petroleum hydrocarbon and concluded that in many times, soil petroleum hydrocarbons can act as a carbon source for soil microbial activities that are similar to our work.<sup>[26]</sup> *Acinetobacter* sp. was found to be capable of utilizing n-alkanes of chain length C10–C40 as a sole source of carbon.<sup>[27]</sup> In addition, *Pseudomonas* are the best-known bacteria capable of utilizing hydrocarbons as carbon and energy sources and producing biosurfactants.<sup>[28]</sup>

Due to the decrease in plant resistance to abiotic stresses such as heavy metals, using SA as a plant promoting-growth agent can enhance soil microbial activities and consequently increase the degradation of soil fuel oil. Accordingly, SA foliar application (1.5 mmol/kg soil) significantly increased the soil microbial respiration of saline soil (6 dS/m) that was polluted with 5 mg Cd/kg soil and 4% (W/W) fuel oil suggesting the promoting role of SA on plant biomass and consequently plant root exudate. Cheng et al. investigated the relation between root exudate quantity and soil biogeochemical processes and concluded that increasing plant root exudate can increase soil microbial activity.<sup>[29]</sup> Baumert et al. reported that plant root exudate can help to increase soil microbial activity and thereby increase soil microbial respiration.<sup>[30]</sup> Due to the importance of plant root exudate as a surrogate for soluble rhizodeposits, it is worth to test the effect of these compounds on soil process in future studies.

With increasing soil pollution to fuel oil, the soil fuel degradation was increased which may be attributed to the role of fuel oil as a carbon source for soil microorganisms to degradation of soil petroleum hydrocarbons.<sup>[28]</sup> However, a higher level of soil petroleum hydrocarbon has toxicity effects. Besalatpour et al. investigated the land-farming process effects on biochemical properties of petroleum-contaminated soils and concluded that the increase in microbial biomass in contaminated with petroleum hydrocarbons may be resulted from the utilization of oil hydrocarbon compounds by microorganisms as a carbon source<sup>[18]</sup> that is similar to our results. However, they did not consider the role of other physicochemical properties such as soil salinity or heavy metal pollution in their results. Based on the results of our studies, increasing soil pollution to heavy metal or soil salinity significantly decreased the degradation of fuel oil in the soil. Accordingly, increasing soil salinity from 4 to 6 dS/m significantly decreased the degradation of soil fuel oil by 15.3%.

Regardless of the adverse effects of soil salinity on soil microbial activities, increasing soil salinity to more than 4 dS/m can decrease plant biomass and thereby has an adverse effect on the amount of plant root exudate. Based on the results of this study, increasing soil salinity from 4 to 6 dS/m significantly decreased the plant biomass and soil fuel oil degradation by 11.1% and 13.2%, respectively. In general, salt can affect plant biomass and germination and its density, and in the most serious cases, leading to generalized plant death, limiting nutrient absorption, and reducing the quality of the available water.<sup>[31]</sup> It should be noted that the interaction between heavy metal and salinity has negative effects on plant growth, especially in petroleum hydrocarbon-contaminated soils, and these plant stresses can reduce the phytoremediation or fuel oil degradation efficiency.

Based on the results of this study, the interaction effects of SA foliar application and P. indica had a significant effect on increasing fuel oil degradation and diminish the negative effects of heavy metals or soil salinity. Accordingly, plant inoculation with P. indica significantly increased the soil fuel oil degradation by 14.8% in the saline soil (EC equal to 6 dS/m) that was polluted with 10 mg Cd/kg soil, whereas interaction effects of SA foliar application and plant inoculation with P. indica increased the degradation of soil fuel oil by 16.3%. In can be concluded that plant inoculation with P. indica or SA foliar application can diminish the abiotic stresses such as heavy metals or soil salinity and thereby increase soil fuel oil degradation [Table 6]. Abdelaziz et al. reported that P. indica enhances the plant growth and yields under salinity stress, opening up a window of opportunity for its application in desert agriculture<sup>[32]</sup> that is similar to our study. In general, symbiosis with beneficial soil microorganisms is a natural mechanism that might offer a quicker, cost-efficient, and eco-friendly solution to mitigate salinity stress.<sup>[32]</sup> Yun et al. reported that the negative effects of salinity on corn seedlings were alleviated after inoculated plant with P. indica that may be related to improving stomatal conductance and lower K+ efflux

from roots and increased shoot K concentration relative to noninoculated plants under high salinity.<sup>[33]</sup> However, the role of SA foliar application on increasing plant resistance to abiotic stress and its benefit role on increasing soil fuel oil degradation cannot be ignored.

Application of 1.5 mmol/l SA indirectly increased the soil fuel degradation by 14.1% in the soil under cultivation of sunflower inoculated with *P. indica*. Jini *et al.* reported that the SA application can moderate the negative effects of salinity stress by changes in the plant physiological mechanisms.<sup>[34]</sup> Kiarostemi *et al.* concluded that SA application can increase the peroxidase, polyphenol oxidase, and superoxide dismutase activities and consequently increase the plant resistance to abiotic stresses in salt-sensitive plants.<sup>[35]</sup> However, in their research, they only mentioned the role of SA on salinity stress. Therefore, it is necessary to consider the interaction effects of SA and *P. indica* that is the main purpose of this research.

## CONCLUSION

Plant inoculation with P. indica has a positive role on these parameters. In addition, abiotic stress such as heavy metal or salinity stress had adverse effects on degradation of petroleum hydrocarbons in soil. However, the role of other physicochemical properties on degradation of soil fuel oil cannot be ignored. The greatest degradation of soil fuel oil belonged to the polluted soil with 8% (W/W) fuel oil under cultivation of inoculated sunflower with P. indica and receiving SA foliar application. Since salinity increases the soil heavy metal availability, it is necessary to investigate the effect of other heavy metals on the biodegradation of soil petroleum hydrocarbons in future studies. In addition, it is also necessary to point out the role of SA and P. indica application on the changes in antioxidant enzyme activities. In addition, it is needed to investigate the role of other plant growth-promoting compounds such as gibberellic acid on the amount of soil petroleum degradation in future studies.

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#### **Conflicts of interest**

There are no conflicts of interest.

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