



Growth tolerance and remediation potential of six plants in oil-polluted soil

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Abstract

Purpose The aim of this study was to screen plant species for remediation of oil-polluted soil in a semi-arid loess area.

Materials and methods Pot experiments were conducted to test six indigenous plant species: ryegrass (*Lolium perenne*), tall fescue (*Festuca ovina*), wheatgrass (*Agropyron cristatum*), alfalfa (*Medicago sativa*), erect milkvetch (*Astragalus adsurgens*), and caragana (*Caragana korshinskii*). Loessial soil was spiked with crude oil at five pollution levels (0, 0.5, 1, 2, and 4%, w/w). We analyzed plant growth parameters at different stages, soil oxidoreductase activities, and oil degradation rate under different treatments.

Results and discussion Soil pollution by oil negatively affected the germination rate, plant height, and biomass of all six species while inhibiting soil dehydrogenase and catalase activities. These inhibitory effects increased with increasing oil pollution level, with 2% and 1% being the critical levels at which plant growth and soil oxidoreductase activities were significantly inhibited, respectively. Both oxidoreductase activities in the rhizosphere soil were significantly higher than those in the bulk soil, which led to a considerable increase in the degradation rate of total petroleum hydrocarbons (TPHs) in the rhizosphere. The plants showed varying remediation effects in the oil-polluted soil. Erect milkvetch and caragana showed potential for remediation of soil below the 0.5% pollution level, alfalfa for soil below the 1% pollution level, and ryegrass, tall fescue, and wheatgrass for soil below the 4% pollution level. The oil pollution level was negatively correlated with plant height, biomass, oxidoreductase activities, and TPH degradation rate for the six plants ($p < 0.01$), whereas plant biomass was positively correlated with plant height and TPH degradation rate ($p < 0.01$). Dehydrogenase activity was significantly positively correlated with catalase activity ($p < 0.01$), while both of them were positively correlated with TPH degradation rate ($p < 0.05$ and $p < 0.01$, respectively).

Conclusions In conclusion, the six indigenous plants exhibited different tolerances to oil pollution, among which ryegrass, tall fescue, and wheatgrass had the greatest potential for remediation of oil-polluted soil in the loess area.

Keywords Oil pollution · Phytoremediation · Plant biomass · Rhizosphere

Abbreviation

TPH Total petroleum hydrocarbon

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1 Introduction

Oil pollution is a universal problem in oil production and industrial areas (Merkl et al. 2004). There has been increasing demand for oil along with rapid development of the petrochemical industry and the wide application of oil products in various industries. Various degrees of oil dumping or leakage are inevitable in the process of oil extraction, transportation, and refining, causing serious local soil pollution problems (Urum et al. 2006; Lai et al. 2009). Oil pollution blocks soil pores, destroys soil structure, and changes soil water-retention (Wei et al. 2019). Petroleum hydrocarbons also exert toxic effects on soil animals, plants, and microorganisms (Scott and Nelson 2004; Afzal et al. 2011). Polycyclic hydrocarbons are transmitted to humans via the food chain and thus endanger human health (Anderson et al. 2014; Cohen et al. 2014;

Fatima et al. 2015). Therefore, it is necessary to develop efficient, low-cost remediation techniques and explore remediation mechanisms in oil-polluted soils.

Numerous technologies have been developed for remediation of oil-polluted soils, including physical, chemical, and biological methods. However, the physical and chemical methods, such as heating, soil leaching, and solidification, have the disadvantages of high cost, secondary hazards to the environment, and high energy consumption (Kaimi et al. 2007). Phytoremediation is a promising technology that uses plants to clean up contaminated soil (Khan et al. 2013a; Afzal et al. 2014). Compared with physical and chemical methods, phytoremediation is superior because of its low cost, lack of secondary pollution, pollutant degradation (Shehzadi et al. 2015; Fatima et al. 2016, 2017; Rehman et al. 2018), and potential benefits to the landscape (Todd et al. 2016).

Some studies have demonstrated that some plants have a remediation effect on oil-polluted soil. Lin and Mendelssohn (1998) indicated that within a certain range of oil concentration, two marsh plants (*Spartina alterniflora* and *S. patens*) not only survived well but also effectively degraded oil pollutants. Issoufi et al. (2010) found that ryegrass (*Lolium perenne*) and soybean (*Glycine max*) markedly reduced the total petroleum hydrocarbon (TPH) content in a polluted soil. Wiltse et al. (1998) showed that genetically modified alfalfa was tolerant to high concentrations of oil pollution. Euliss et al. (2008) found that oil-polluted soil planted with sedge (*Cyperus rotundus*), switchgrass (*Panicum virgatum*), or gamagrass (*Tripsacum dactyloides*) had considerably less residual TPH content than the non-planted control after 1 year of growth (~70% reduction). Phillips et al. (2009) showed that Altai wildrye (*Leymus angustus*) promoted > 50% TPH degradation in the first growing season.

However, not all environments are suitable for the growth of all plants. Plants can tolerate oil pollution only in conditions that satisfy their habitat requirements and support the healthy growth of the plant. In such conditions, the exudates of plant roots can support the growth of rhizosphere microorganisms which in turn degrade oil and other organic pollutants (Thomas et al. 2017). There are also differences in the harm caused by oil pollution to soils with different textures and chemical properties. Thus, local site conditions must be considered during the development of phytoremediation technologies for oil-polluted soils, and appropriate plants should be selected.

There are abundant oil reserves in northern Shaanxi, one of the major oil industry areas in China. In this area, the environment is highly fragile, with serious water shortage and soil erosion. The oil-polluted soil in this area exceeds 7,081,600 m², and the TPH content in the oil-polluted soil varies between 5000 and 60,000 mg kg⁻¹ (Wang et al. 2017). Soil in this area developed from a loess parent material, and the major soil type is loessial soil. There are few reports on phytoremediation technology for

oil-polluted soils in northern Shaanxi, especially on the remediation effect of indigenous plants.

In this study, we selected six indigenous plants that can grow in the local oil-polluted soil. Pot experiments were conducted to analyze their tolerance to different levels of oil pollution and their remediation effects on oil-polluted soil. Plant species that can adapt to the local environment, tolerate different levels of oil pollution, and have strong remediation effects were selected. This study provides a basis for establishing a phytoremediation system for oil-polluted soil in northern Shaanxi.

2 Materials and methods

2.1 Study area description

The study area was located in Yan'an City, a typical soil area in the Loess Plateau in northern Shaanxi, China (107° 41'–110° 31' E, 35° 31'–37° 31' N). Being situated in the middle reaches of the Yellow River, Yan'an is an undulating region of the Loess Plateau. The terrain is high in the northwest and lower in the southeast, with an average elevation of 1200 m and a total area of 37,037 km². This area belongs to the warm temperate zone, with a semi-humid warm climate. The area has four distinct seasons, abundant sunshine, a large temperature difference between day and night, an average annual frost-free period of 170 days, an average annual temperature of 7.7–10.6 °C, and an average annual sunshine duration of 2300–2700 h. The accumulated temperature ≥ 0 °C is 3878.1 °C, and the average annual precipitation is ~ 500 mm.

2.2 Experimental materials

Six local plant species were used in the experiments: ryegrass (*Lolium perenne*), tall fescue (*Festuca ovina*), wheatgrass (*Agropyron cristatum*), alfalfa (*Medicago sativa*), erect milkvetch (*Astragalus adsurgens*), and caragana (*Caragana korshinskii*). Seeds were collected in wild grasslands in the Baota District of Yan'an City.

The soil used in the experiments was a clean loessial soil obtained from farmland located far away from oil wells, in Baota District, Yan'an City (109° 39.477' E, 36° 19.054' N). At the sampling site, weeds were removed from the soil surface using a shovel and a 0–30-cm soil layer was then collected. The soil sample was transported to the laboratory, air-dried to remove debris, then ground and passed through a 5-mm screen.

Crude oil was extracted from well Wan-107 in the Yanchang Oilfield, Liulin Town, Yan'an. The oil density and dynamic viscosity were 0.839 g cm⁻³ (20 °C) and 4.6 MPa s⁻¹ (50 °C), respectively.

2.3 Determination of soil physicochemical properties

The mechanical composition of the soil was measured using a MS2000 laser particle size analyzer (Malvern Instruments Ltd., Malvern, UK). Soil pH was measured using a 1:2.5 soil-water ratio extraction method and an S220 acidometer (Mettler Toledo Instruments Ltd., Zurich, Switzerland). Organic matter was measured by potassium dichromate oxidation-external heating. Total nitrogen was determined using a semi-micro Kjeldahl method using a UDK129 Kjeldahl nitrogen analyzer (VELP Scientifica, Milan, Italy). Available phosphorus was determined using the sodium bicarbonate-molybdenum antimony colorimetric method with a Lambda 650S UV-vis spectrophotometer (PerkinElmer Corp., Shelton, USA). Available potassium was determined by ammonium acetate extraction-flame spectrophotometry using a M420 flame photometer (Sherwood Scientific Ltd., Cambridge, UK). The basic physicochemical properties of the soil are shown in Table 1.

2.4 Pot experiments

To evaluate the tolerance of plants to oil pollution, pot experiments were conducted using soil treated with different levels of oil. Equal amounts of soil samples were spiked with crude oil at five pollution levels (0, 0.5, 1, 2, and 4%, w/w) and incubated in the dark for a week before pot experiments. Seeds were sown on November 1, 2016, with 40 seeds per pot for each plant and covered by ~1 cm of soil. For each level of oil pollution, there was also a blank control without planting, and each treatment had three replicates. Pots were 20 cm in diameter and 20 cm deep. After sowing, the pots were arranged in random blocks and the positions were changed regularly to maintain good ventilation and light conditions. Water was applied every other day to control soil moisture at about 25%. Seed germination rate was recorded, and plant height was monitored on days 40, 60, and 80. On day 80, plants were harvested and total biomass (fresh weight) was determined. The relative germination rate was defined as the ratio of the germination rate between oil pollution treatment and non-pollution treatment.

After harvest, potted rhizosphere and bulk soils were collected to determine dehydrogenase activity, catalase activity, and TPH content. Soil dehydrogenase activity was determined by the triphenyl tetrazolium chloride method and expressed as

the amount of triphenyl formamidine produced per gram of soil (Casida 1977). Soil catalase activity was determined by potassium permanganate titration and expressed as the amount of potassium permanganate (0.1 mol L^{-1}) consumed per gram of soil (Trasar et al. 1999). The TPH content of soil samples (rhizosphere, bulk, and mixed soils) was determined by infrared spectrometry and the TPH degradation rate of different biological treatments was calculated as follows (Qi et al. 2015):

TPH degradation rate (%)

$$= \frac{\text{Soil TPH content before planting} - \text{Soil TPH content after harvest}}{\text{Soil TPH content before planting}} \times 100 \quad (1)$$

2.5 Statistical analysis

Data are means \pm standard deviation. Statistical analyses were performed using SPSS 18.0 software (SPSS Inc., Chicago, USA). Group means were compared by one-way analysis of variance. Relationships between parameters were assessed using Pearson's correlation. Plots were drawn using Origin 8.5 (OriginLab Corp., Northampton, USA).

3 Results and discussion

3.1 Plant growth parameters

3.1.1 Seed germination

Seed germination is a prerequisite for the reproduction of plants (Erickson et al. 2010). The germination rate in soil under the stress of oil pollution is the primary indicator of whether a plant can be used to remediate oil-polluted soils (Qi et al. 2015). Herein, we first conducted germination tests to determine whether the studied plants had the potential for remediation of oil-polluted soil. At each oil pollution level, the germination rate of herbs (ryegrass, tall fescue, wheatgrass) was generally higher than that of shrubs (erect milkvetch; Table 2), similar to the results of Shan in northern Shaanxi (Shan et al. 2014).

At an oil pollution level of 0.5%, the germination rates of ryegrass and wheatgrass were not affected compared with the

Table 1 Basic physicochemical properties of the experimental soil from northern Shaanxi, China

Soil type	Particle-size distribution (%)			Soil texture	pH	Organic matter (g kg ⁻¹)	Total N (g kg ⁻¹)	Available P (mg kg ⁻¹)	Available K (mg kg ⁻¹)
	Clay (0–2 μm)	Silt (2–50 μm)	Sand (50–2000 μm)						
Loessial soil	10.28	71.44	18.28	Silty loam	8.25	9.78	2.94	12.8	90

Table 2 Seed germination rate of six indigenous plant species in the oil-polluted soil

Pollution level (%)	Germination rate (%)					
	Ryegrass	Tall fescue	Wheatgrass	Alfalfa	Erect milkvetch	Caragana
0	95.0 ± 0.21a	92.5 ± 0.49 ^a	85.0 ± 0.12a	67.5 ± 0.29a	25.0 ± 0.26a	73.3 ± 0.17a
0.5	95.0 ± 0.56a	90.0 ± 0.21b	85.0 ± 0.12a	52.5 ± 0.12b	17.5 ± 0.21b	66.7 ± 0.15b
1	90.0 ± 0.70b	82.5 ± 0.20c	82.5 ± 0.12b	52.5 ± 0.12b	15.0 ± 0.12c	60.0 ± 0.82c
2	85.0 ± 0.12c	70.0 ± 0.26d	80.0 ± 0.21c	37.5 ± 0.10c	12.5 ± 0.10d	53.3 ± 0.25d
4	85.0 ± 0.21c	67.5 ± 0.21e	45.0 ± 0.21d	35.0 ± 0.15d	5.0 ± 0.12e	53.3 ± 0.15e

Different lowercase letters in a column indicate a significant difference in germination rate between the same plant species at different oil pollution levels ($p < 0.05$)

non-polluted control. With increasing oil concentration in the soil, the germination rate of different plants significantly declined compared with the unpolluted control. The decreases at the pollution level of 4% were 10.0, 25.0, 40.0, 32.5, 20.0, and 20.0% for ryegrass, tall fescue, wheatgrass, alfalfa, erect milkvetch, and caragana, respectively (Table 2). Thus, germination of ryegrass, tall fescue, and caragana was less affected by oil pollution, and their relative germination rates were > 70% at pollution levels 0.5–4%. In contrast, the relative germination rates of wheatgrass and alfalfa were greatly affected by oil pollution and were respectively 52.9 and 51.9% at the 4% pollution level. The relative germination rate of erect milkvetch was most severely affected by oil pollution and was only 20.0% at the 4% pollution level (Table 3). Based on the results of germination tests, ryegrass and tall fescue showed the greatest potential among the plants tested to remediate oil-polluted soil.

Besalatpour et al. (2010) found that the seed germination rate of wheatgrass in oil-polluted soils declined by > 50% compared with that in non-polluted soil. Other studies have shown that the seed germination rates of alfalfa, erect milkvetch, and caragana were considerably reduced when the oil pollution level was > 2% (Shan et al. 2014). The response of plants to oil pollution varies, which results in the differences in seed germination rate of different plants at different levels of pollution (Adam and Duncan 2002). The impact of oil on seed germination may be because seeds are

wrapped in an oil film, which affects their internal physiobiochemical functions, water absorption, and respiration, leading to water shortage and anoxia (Amadi et al. 1993). It is also possible that a high concentration of oil pollutants inhibits the activity of proteases in the seed and thus affects protein decomposition, thereby limiting the nutrient supply (Zhang et al. 2004).

3.1.2 Plant height

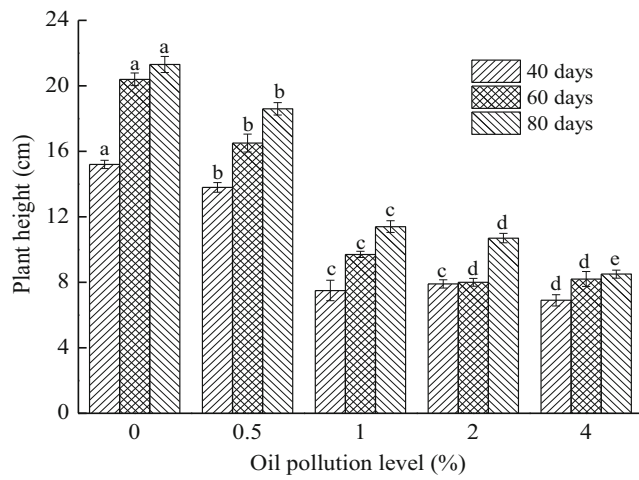
Plant height is a sensitive measure of the tolerance of a plant to oil pollution in the soil (Ogboghodo et al. 2004). In this study, the plant height of the six species was significantly affected by oil pollution of the soil ($p < 0.05$) and showed a downward trend with increasing oil pollution (Fig. 1).

Among the different species, oil pollution showed markedly different inhibitory effects on plant height. Significant growth inhibition was observed for ryegrass, alfalfa, and caragana at the 1% pollution level, and for wheatgrass, tall fescue, and erect milkvetch at the 0.5% pollution level. In the 4% oil-polluted soil, alfalfa and erect milkvetch grew slowly and were almost all dead on day 80. At the 0.5% pollution level, the 80-day plant heights of wheatgrass and tall fescue were 42.3 and 51.8% of the control values (i.e., in unpolluted pots), respectively. With 1% pollution, the heights of ryegrass and caragana were 53.5 and 38.3% of the control values, respectively. At 4% pollution, the heights of ryegrass,

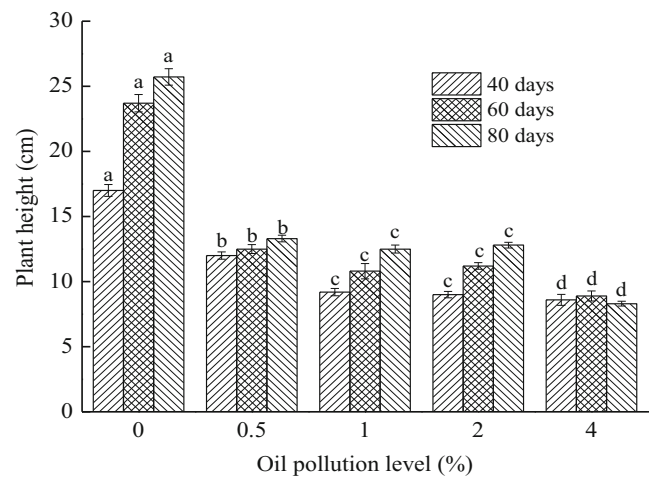
Table 3 Relative germination rate of six indigenous plant species in the oil-polluted soil

Pollution level (%)	Relative germination rate (%)					
	Ryegrass	Tall fescue	Wheatgrass	Alfalfa	Erect milkvetch	Caragana
0.5	100.0	97.3	100.0	77.8	70.0	91.0
1	94.7	89.2	97.1	77.8	60.0	81.9
2	89.5	75.7	94.1	55.6	50.0	72.8
4	89.5	73.0	52.9	51.9	20.0	72.8
Average	93.4	83.8	86.0	65.7	50.0	79.6

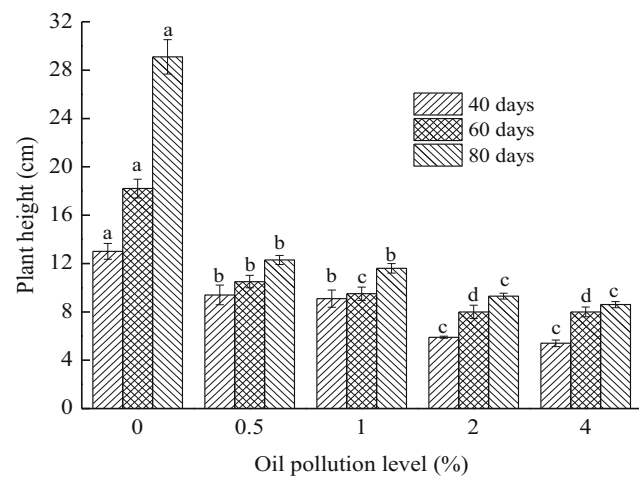
The relative germination rate is the ratio of the germination rate between oil pollution and non-pollution treatments



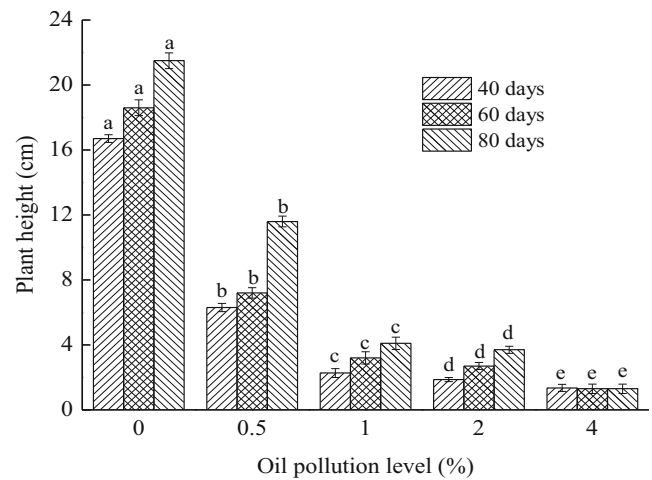
(a) Ryegrass



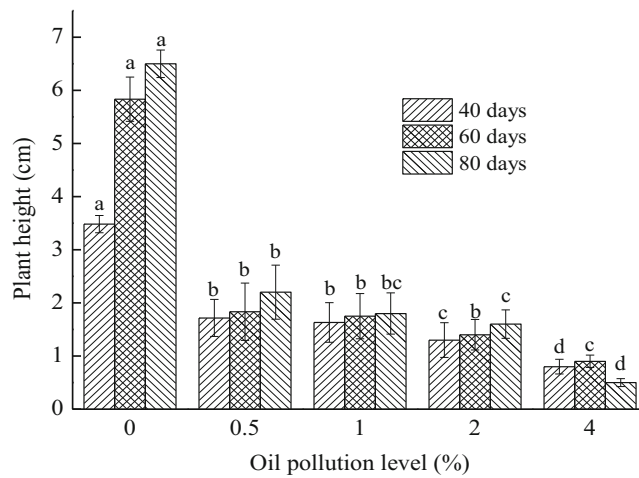
(b) Tall fescue



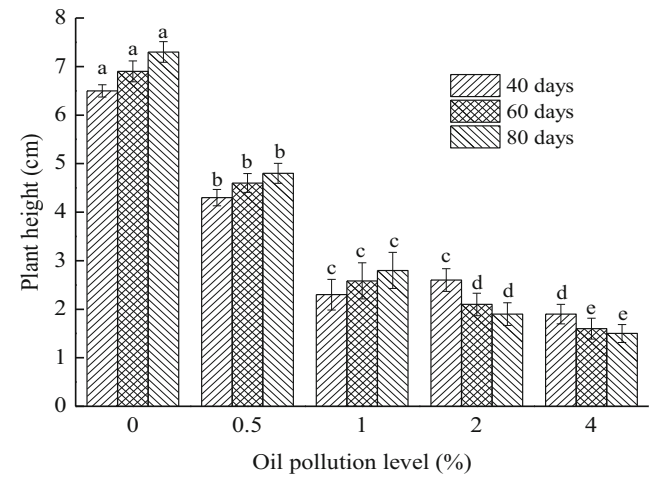
(c) Wheatgrass



(d) Alfalfa



(e) Erect milkvetch



(f) Caragana

Fig. 1 Plant height of the six indigenous species at different growth stages on the oil-polluted soil. **a** Ryegrass. **b** Tall fescue. **c** Wheatgrass. **d** Alfalfa. **e** Erect milkvetch. **f** Caragana. Different lowercase letters

indicate a significant difference in plant height between the same plant species at different oil pollution levels ($p < 0.05$)

wheatgrass, tall fescue, and caragana were 39.9, 29.6, 32.3, and 20.5% of the control values, respectively. These results indicate that ryegrass was the plant least affected by oil pollution. This may be due to greater absorption of petroleum hydrocarbons by the root system of this plant, or the stronger effect of its root exudates on the degradation of petroleum hydrocarbons compared with the other plants.

3.1.3 Plant biomass

Biomass is another important indicator of the tolerance and remediation effect of plants in oil-polluted soil (Baruah and Sarma 1996). After 80 days of growth, the biomass of the six plants decreased with increasing oil pollution level. Compared with the control, <40% biomass reduction was found in ryegrass, tall fescue, and wheatgrass at the 0.5% pollution level and <50% biomass reduction was found in ryegrass and tall fescue at the 1% pollution level. At the 2% pollution level, <50% reduction was only found in ryegrass, while the remaining plants showed a >50% reduction in biomass. At the 4% pollution level, compared with controls, the biomass of ryegrass, tall fescue, wheatgrass, alfalfa, erect milkvetch, and caragana decreased by 69.7, 75.0, 77.8, 96.5, 89.5, and 85.4%, respectively (Fig. 2). These results indicate that a level of oil pollution >2% seriously inhibited biomass accumulation of the plants.

The reductions in plant height and biomass caused by oil pollution can be attributed to the fact that the plant roots are in direct contact with oil pollutants, so low-molecular-weight hydrocarbons enter the plant body causing toxic effects on the plants. High-molecular-weight hydrocarbon oil films can wrap the roots and hinder the absorption of nutrients and water from

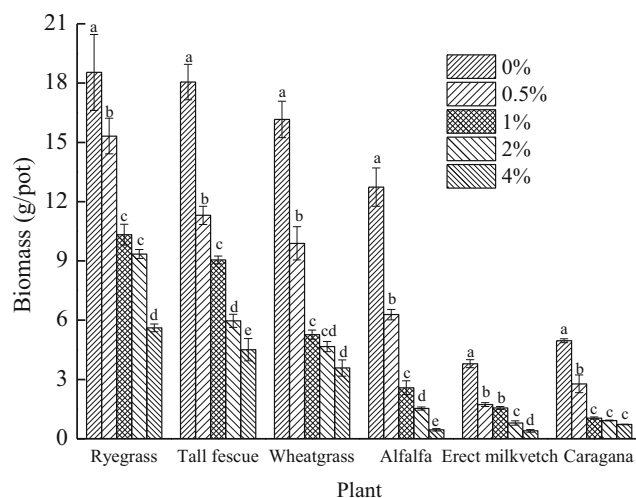


Fig. 2 Biomass of the six indigenous plants after 80 days of growth in the oil-polluted soil. Different lowercase letters indicate a significant difference in biomass between the same plant species at different oil pollution level ($p < 0.05$)

the soil, which in turn affects respiration of the root. The oil may also negatively affect the soil water-holding capacity, which causes a decrease in soil hydrophilicity and water permeability, further hindering plant growth. In addition, oil pollution can cause a sudden increase in soil carbon content, leading to carbon/nitrogen imbalance and affecting plant growth (Xie 1987; Li et al. 1997; Odjegba and Sadiq 2002). Furthermore, volatile petroleum hydrocarbons can lower the concentration of oxygen in the soil air, again affecting root respiration.

It is generally believed that the greater the biomass formed by plant growth, the better the remediation effect (Shi 2013). With regard to plant biomass alone, ryegrass and tall fescue showed the potential to remediate soil below 1% oil pollution in the present study. The biomass of plants is closely related to the characteristics of the plant species; the high biomass of ryegrass and tall fescue in soil below the 1% pollution level might be attributed to their large tiller number per plant.

3.2 Soil oxidoreductase activities

Zhou (1987) indicated that the degradation of petroleum in soil is mainly attributed to the role of oxidoreductases, and the most important ones are dehydrogenase and catalase. At lower oil pollution levels, the dehydrogenase and catalase activities in the rhizosphere, bulk soil, and non-planted control increased with increasing oil concentration (Figs. 3 and 4). However, when the oil pollution level exceeded 1%, soil oxidoreductase activities decreased rapidly with increasing oil concentration. Thus, 1% was the critical level for the change in the activity of the two oxidoreductases. At the 4% oil pollution level, soil oxidoreductase activities were significantly lower compared with the non-planted control. These results indicate that a low level of oil pollution enhanced soil oxidoreductase activities, while a higher pollution level inhibited the oxidoreductase activities.

At different oil pollution levels, both oxidoreductase activities were the largest in the soil planted with ryegrass, followed by tall fescue; the lowest oxidoreductase activities were found for caragana. The activities of the two oxidoreductases at each oil pollution level were in the order rhizosphere > bulk soil > non-planted control, indicating that the plant's rhizosphere was more conducive to the degradation of petroleum pollutants. On the one hand, the significant increase in the rhizosphere oxidoreductase activities may be due to the adaptation of enzyme-secreting microorganisms to the stress environment; on the other hand, oil pollution caused an increase in soil available carbon, which could enhance soil microbial activities and thus lead to an increase of enzyme secretion. In addition, as the plants grew, their root exudates increased, which also enhanced soil microbial activities and consequently increased soil oxidoreductase activities.

When the oil pollution level was less than 2%, the dehydrogenase activity in the rhizosphere soil was significantly

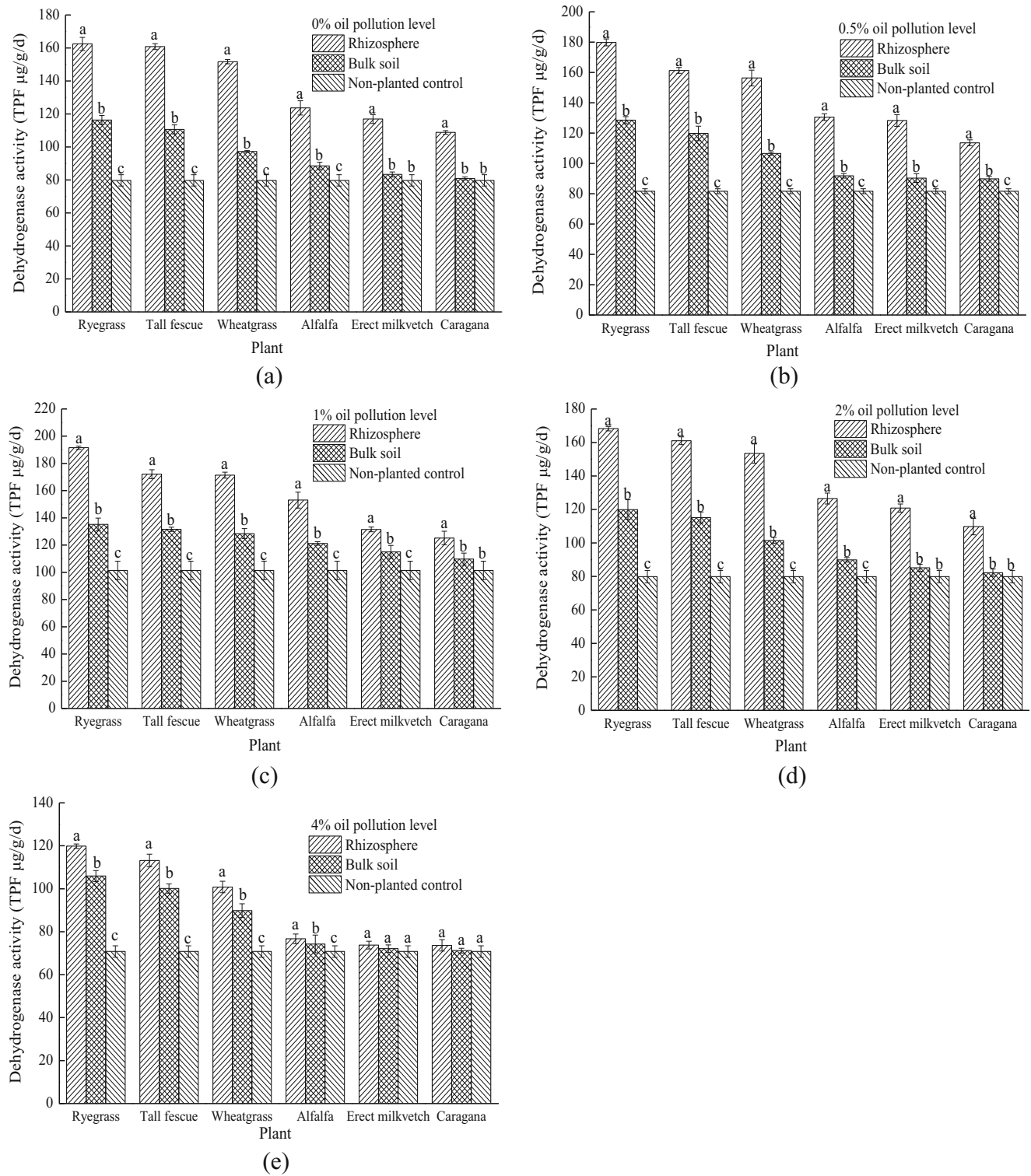


Fig. 3 Effects of oil pollution on soil dehydrogenase activity in rhizosphere, bulk soil, and the non-planted control. **a** 0% oil pollution level. **b** 0.5% oil pollution level. **c** 1% oil pollution level. **d** 2% oil pollution level. **e** 4% oil pollution level. Different lowercase letters

indicate a significant difference in dehydrogenase activity of the same plant species among rhizosphere, bulk soil, and the non-planted control ($p < 0.05$)

higher than that in the bulk soil and the non-planted control for the same plant. A similar trend was observed in soil catalase

activity when the oil pollution level was less than 1%. Thus, the oil degradation ability of the rhizosphere soil appeared to

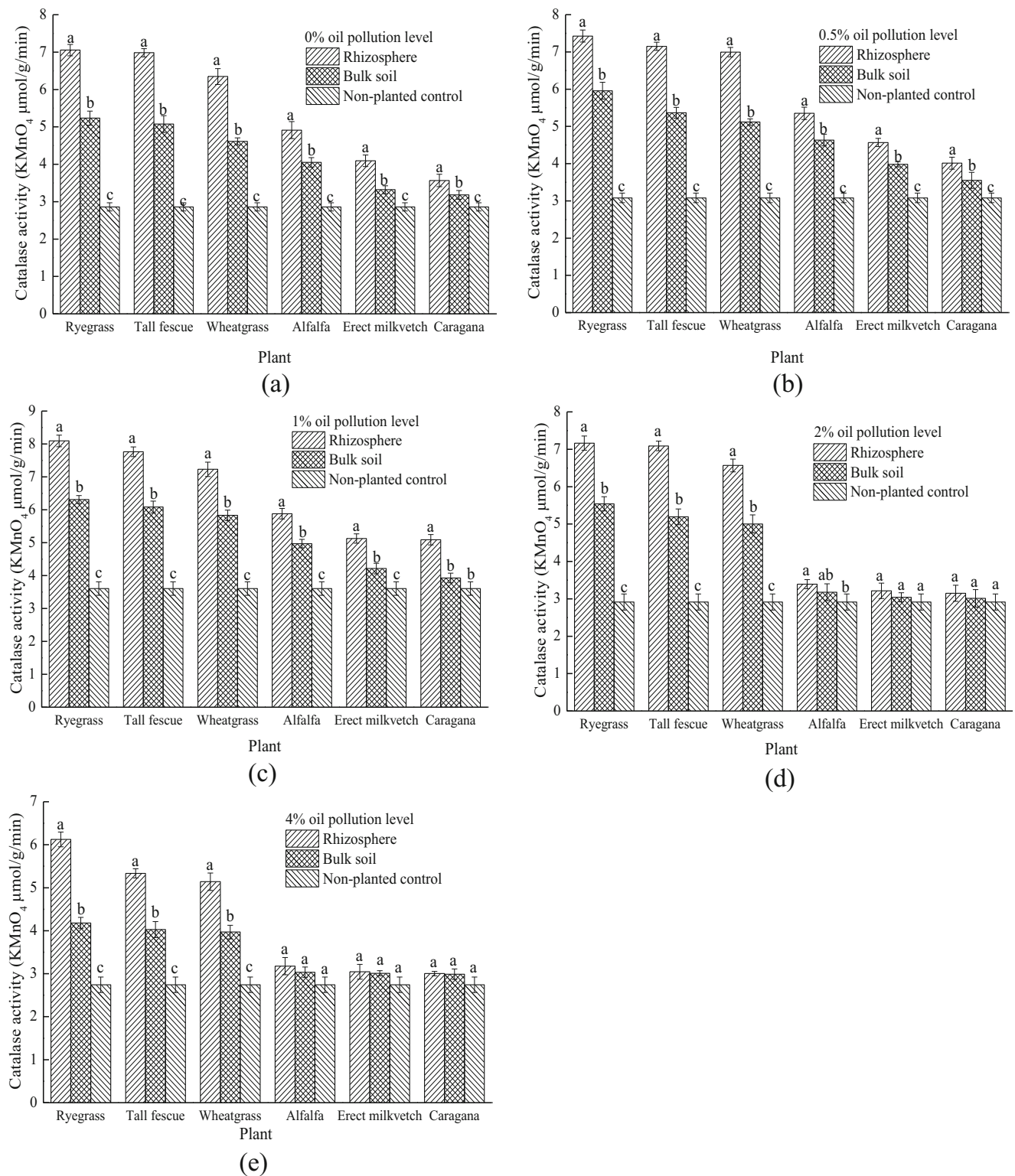


Fig. 4 Effects of oil pollution on soil catalase activity in rhizosphere, bulk soil, and the non-planted control. **a** 0% oil pollution level. **b** 0.5% oil pollution level. **c** 1% oil pollution level. **d** 2% oil pollution level. **e** 4% oil

pollution level. Different lowercase letters indicate a significant difference in catalase activity of the same plant species among rhizosphere, bulk soil, and the non-planted control ($p < 0.05$)

be the largest compared with that of the bulk soil and the non-planted control. At the 4% oil pollution level, soil dehydrogenase activity was not significantly affected by planting of erect

milkvetch or caragana, while the catalase activity was not affected by planting of alfalfa, erect milkvetch, or caragana. These results indicate that, except for alfalfa, erect milkvetch,

and caragana, planting of ryegrass, tall fescue, or wheatgrass may considerably promote the degradation of petroleum hydrocarbons at various pollution levels. Among the plants tested, ryegrass was deemed the best option for the remediation of oil pollution.

3.3 Hydrocarbons degradation

At each pollution level, more TPHs were degraded in the rhizosphere than in bulk soil, and there was a significant difference in the degradation rate of TPHs between the rhizosphere and bulk soil of each plant (Fig. 5). At the 0.5% and 1% pollution levels, the average TPH degradation rate of rhizosphere soil for the six plants was 1.9 times that of bulk soil. In the rhizosphere soil, ryegrass and tall fescue had the highest degradation rate, both of which showed significant differences compared with other plants. At the 2% and 4% pollution

levels, the average TPH degradation rate of rhizosphere soil for the six plants was 1.8 and 1.7 times that of bulk soil, respectively. Compared with other plants, ryegrass, tall fescue, and wheatgrass resulted in the highest TPH degradation rate in the rhizosphere. To some extent, the level of soil oxidoreductase activity reflects the soil’s ability to degrade petroleum pollutants (Zhou 1987). Therefore, an enhancement of dehydrogenase and catalase activities may be one of the main reasons for the increase of TPH degradation rate in the rhizosphere compared with bulk soil.

Phytoremediation effects were evaluated using the degradation rate of TPHs in the mixture of rhizosphere and bulk soils after 80 days of planting (Fig. 6). Due to volatilization, photolysis, and microbial activity in the soil, the non-planted control showed TPH degradation at various oil pollution levels (degradation rates 14.2–18.0%). All six plants promoted the degradation of petroleum pollutants in the soil

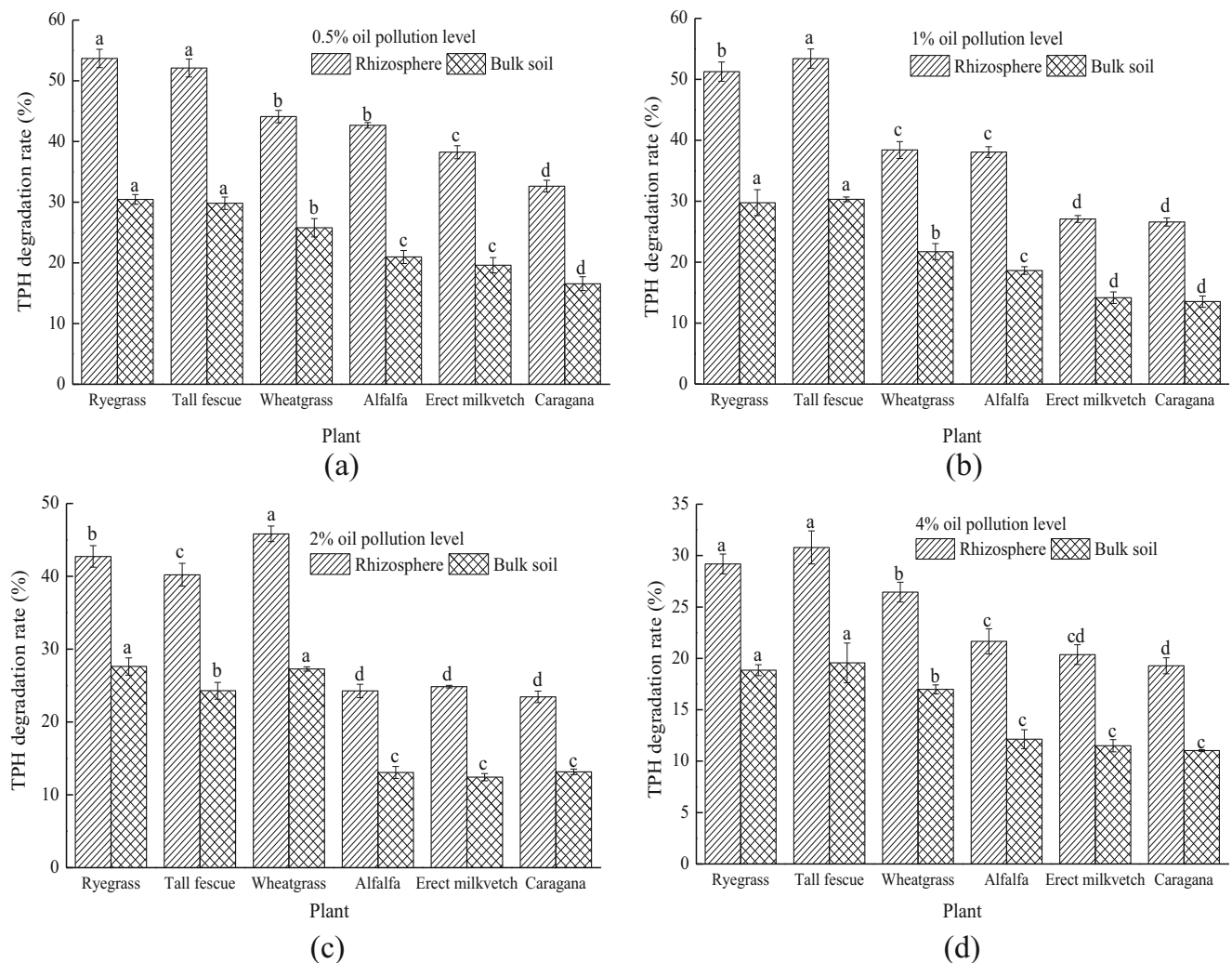
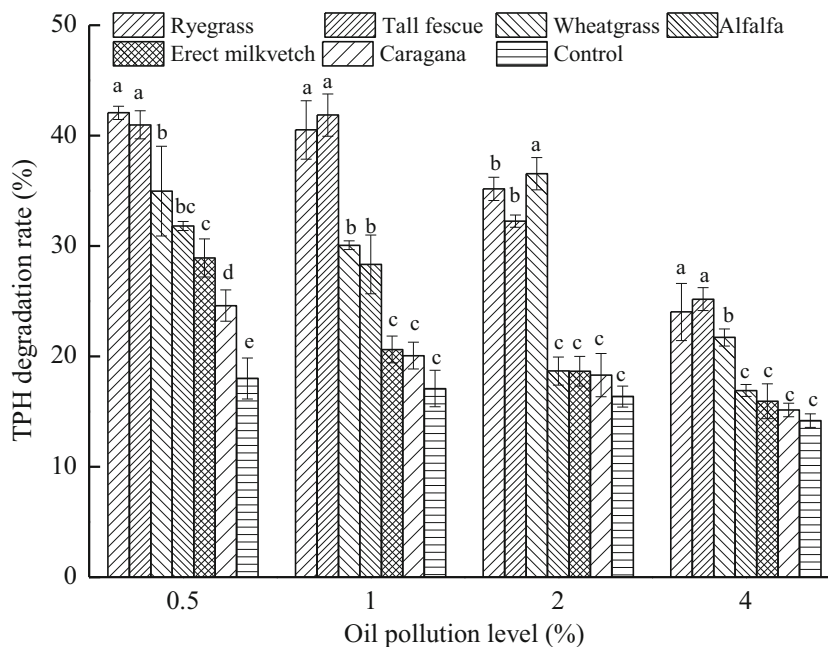


Fig. 5 Degradation rate of total petroleum hydrocarbons (TPHs) by the six ingenious plants in the rhizosphere and bulk soil. **a** 0.5% oil pollution level. **b** 1% oil pollution level. **c** 2% oil pollution level. **d** 4% oil pollution

level. Different lowercase letters indicate a significant difference at the same oil pollution level among different plants ($p < 0.05$)

Fig. 6 Degradation rate of total petroleum hydrocarbons (TPHs) by the six ingenious plants in the oil-polluted soil. Different lowercase letters indicate a significant difference at the same oil pollution level among different plants ($p < 0.05$)



compared with the non-planted control. In general, the degradation effects of different plants were in the order ryegrass > tall fescue > wheatgrass > alfalfa > erect milkvetch > caragana. As the concentration of oil pollutants increased, the degradation rate of TPHs showed a decreasing trend.

At the 0.5% pollution level, all six plants showed significant degradation effects compared with the non-planted control. The greatest degradation rate was observed for ryegrass and tall fescue, ~2.3 times higher than the degradation in the control. The lowest rate was found for caragana, 1.4 times that in the control. At the 1% pollution level, tall fescue, ryegrass, wheatgrass, and alfalfa showed significant degradation effects, respectively 2.5, 2.4, 1.8, and 1.7 times that in the control. At the 2 and 4% pollution levels, significant degradation effects were observed for wheatgrass, ryegrass, and tall fescue; their degradation rates at 2% pollution were respectively 2.2, 2.2, and 2.0 times that in the control, and the rates at the 4% pollution level were 1.5, 1.7, and 1.8 times that in the control.

Plants can promote the degradation of oil pollutants in soil via direct absorption of petroleum hydrocarbons by the roots. Moreover, root exudates may enhance the degradation of petroleum hydrocarbons and organic matter mineralization in the rhizosphere (root–soil interface) by changing the soil microbial community (Afzal et al. 2012; Xie et al. 2012; Khan et al. 2013a, 2013b). Therefore, the key to success in applying phytoremediation technology to oil-polluted soils is to ensure that the plant roots grow well. Ryegrass, tall fescue, and wheatgrass all have developed fibrous roots, in which the large root surface provides sufficient space for plant nutrient absorption and microbial growth. This may be the main reason for the relatively high

degradation rates of petroleum hydrocarbons in the soil by these three plants.

3.4 Correlation analysis

To further explore the effect of oil pollution on plant growth and the remediation of oil pollution by the plants, we analyzed the correlation between oil pollution level, plant growth parameters, soil oxidoreductase activities, and TPH degradation rate. For all six plants, oil pollution level was significantly and negatively correlated with plant height ($r = -0.722$ to -0.912), biomass ($r = -0.764$ to -0.947), soil oxidoreductase activities ($r = -0.713$ to -0.947), and TPH degradation rate ($r = -0.729$ to -0.974 ; p values < 0.01) (Table 4). Specifically, plant height and biomass at different growth stages significantly declined with increasing oil pollution level, which eventually led to a reduction in the degradation of oil pollutants by the plants.

There was a significant positive correlation between plant height and biomass at harvest ($r = 0.778$ – 0.982), between plant biomass and TPH degradation rate ($r = 0.729$ – 0.915), and between dehydrogenase and catalase activities ($r = 0.817$ – 0.989) for all six plant species (p values < 0.01 ; Table 4). This indicates that the greater the biomass, the better the plant's remediation effect on the oil-polluted soil. Meanwhile, there were significant positive correlations between soil oxidoreductase activities and TPH degradation rate ($r = 0.615$ – 0.935 for dehydrogenase, $r = 0.614$ – 0.924 for catalase; p values < 0.05 ; Table 4). This indicates that the stronger the oxidoreductase activity, the higher the degradation rate of TPHs. For phytoremediation of oil-polluted soils, the ideal plant seeds should have strong vitality under

Table 4 Correlation analysis of oil pollution level, plant growth parameters, soil oxidoreductase activities, and total petroleum hydrocarbon (TPH) degradation rate for six indigenous plants in the oil-polluted soil

Plant	Index	Oil pollution level	Plant height	80-day biomass	Dehydrogenase	Catalase
Ryegrass	Plant height	− 0.722**				
	80-day biomass	− 0.908**	0.929**			
	Dehydrogenase	− 0.947**	0.477	0.739**		
	Catalase	− 0.917**	0.419	0.687*	0.985**	
	TPH degradation rate	− 0.974**	0.658*	0.858**	0.935**	0.899**
Tall fescue	Plant height	− 0.912**				
	80-day biomass	− 0.922**	0.846**			
	Dehydrogenase	− 0.910**	0.720**	0.722**		
	Catalase	− 0.892**	0.696*	0.705*	0.989**	
	TPH degradation rate	− 0.956**	0.779**	0.915**	0.902**	0.908**
Wheatgrass	Plant height	− 0.842**				
	80-day biomass	− 0.764**	0.805**			
	Dehydrogenase	− 0.881**	0.678*	0.401		
	Catalase	− 0.927**	0.706*	0.485	0.988**	
	TPH degradation rate	− 0.729**	0.436	0.729**	0.616*	0.663*
Alfalfa	Plant height	− 0.785**				
	80-day biomass	− 0.826**	0.982**			
	Dehydrogenase	− 0.839**	0.335	0.419		
	Catalase	− 0.837**	0.608*	0.691*	0.817**	1
	TPH degradation rate	− 0.880**	0.822**	0.890**	0.673*	0.924**
Erect milkvetch	Plant height	− 0.788**				
	80-day biomass	− 0.947**	0.778**			
	Dehydrogenase	− 0.911**	0.898**	0.858**		
	Catalase	− 0.817**	0.777**	0.914**	0.825**	
	TPH degradation rate	− 0.807**	0.442	0.827**	0.629*	0.614*
Caragana	Plant height	− 0.819**				
	80-day biomass	− 0.783**	0.930**			
	Dehydrogenase	− 0.879**	0.501	0.304		
	Catalase	− 0.713**	0.497	0.297	0.860**	
	TPH degradation rate	− 0.876**	0.914**	0.843**	0.615*	0.673*

* $p < 0.05$, and ** $p < 0.01$

pollution stress, and their germination rate should be unaffected or little affected by the pollutants. Our correlation analysis results also showed that plant biomass and oxidoreductase activity were indicators for determining phytoremediation effects and can be used in the selection of remediation plants based on local environmental conditions.

4 Conclusions

In this study, we determined the growth tolerance of six indigenous plants from northern Shaanxi, China, to oil pollution in loess soil, the responses of oxidoreductase activities in the rhizosphere and bulk soil, and the recovery of oil-polluted soil planted with different plants. We found that oil pollution

negatively affected seed germination, plant height, and biomass formation of all six species while inhibiting soil dehydrogenase and catalase activities. These inhibitory effects on plant growth and soil oxidoreductase activities increased with increasing pollution level. The oxidoreductase activities in the rhizosphere soil were significantly higher than those in the bulk soil, thus leading to a significant increase in the degradation rate of TPHs in the rhizosphere. For phytoremediation of the oil-polluted loessial soil, ryegrass, tall fescue, and wheatgrass showed greater potential than alfalfa, erect milkvetch, and caragana. In the practice of phytoremediation, we must first evaluate the soil pollution level, and, when selecting plants, we should consider the natural environmental conditions in the polluted area and the cost of management to choose high-biomass plants to maximize the remediation benefits.

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