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Full Length Article

Effects of TPH contamination on soil and rice growth: Assessing the efficacy of sodium persulfate as a remediation agent

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ABSTRACT

Soil contamination by total petroleum hydrocarbons (TPH) poses a major risk to soil health and crop productivity. This study aims to investigate the effects of TPH on soil properties and rice growth factors, as well as the potential of sodium persulfate (SP) to mitigate negative impacts. The study includes four treatments: control (no SP or TPH), SP (0.0, 1.50 and 3.0%), TPH (TPH-contaminated soil, 4.0%, 6.0%, and 8.0%), and SP + TPH (SP and TPH levels). Results show that TPH contamination led to reductions in pH, organic matter, nutrient availability, root and shoot growth, chlorophyll content, plant height, grain and biomass yield. SP facilitates the restoration of soil physicochemical properties, mitigates toxicity, and enhances plant growth conditions by promoting both chemical oxidation and microbial degradation of TPH contaminants. Higher doses of SP are required to counteract higher TPH levels. The findings suggest that SP can be an effective amendment for remediating TPH-contaminated soils, but dosage and application should be tailored to site-specific conditions. This study highlights the potential of SP for sustainable management of TPH-contaminated agricultural land, but further work is needed to fully understand its impact on different plant species and to optimize remediation strategies for specific site conditions.

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1. Introduction

Soil pollution caused by TPH is a significant global environmental concern, arising from activities such as oil spills, improper waste disposal, and industrial processes (Bento et al., 2005; Zhang et al., 2021). Soil quality, plant growth, and overall ecosystem health can be significantly compromised by contamination from TPH as evidenced in studies conducted by Afzal et al. (2014) and Megharaj et al. (2011). As such, it is imperative to implement effective and sustainable remediation approaches to mitigate the environmental repercussions of TPH-contaminated soil. Bioremediation, a biological method that employs microorganisms to degrade pollutants, has garnered considerable attention as a cost-effective and eco-friendly approach for TPH remediation (Pérez et al., 2018; Udume et al., 2023). Several methods have been devised to boost the breakdown of TPH in soil through bioremediation. These methods involve the incorporation of various amendments like urea, ammonium sulfate, and NPK (nitrogen, phosphorus, and potassium) fertilizers into the soil, as reported in studies by Udume et al. (2023) and Breedveld and Sparrevik (2000). These amendments can stimulate the growth and activity of native microorganisms in soil, facilitating the degradation of TPH through biodegradation and mineralization pathways (Varjani, 2017; Margesin and Schinner, 1997).

A recent study demonstrated that biochar application (2–4%) improves soil physicochemical properties and mitigates the negative effects of TPH contamination on rice growth, yield, and biomass. Biochar increased soil pH, organic matter, and nutrient availability, enhancing plant height and productivity even under TPH stress. These findings suggest biochar's potential as a sustainable amendment for remediating contaminated soils, though further research on its mechanisms and long-term impacts is needed (Rassaei, 2024a). In a study research by Rahman et al. (2003), they investigated the treatment of sterile and non-sterile soil samples contaminated with sludge oil. They utilized a bacterial consortium (BC), rhamnolipid biosurfactant (RL), and a solution of nitrogen, phosphorus, and potassium (NPK). The study revealed that the soil samples contaminated with 10% sludge exhibited the highest level of degradation for n-alkane. Furthermore, they observed that all the tested additives (BC, NPK, and RL)

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had a significant positive impact on the bioremediation of n-alkane in petroleum sludge. Bioremediation harnesses the natural abilities of microorganisms to eliminate harmful pollutants in soil and water, a persistent issue that poses risks to both plants and animals, including humans (Rassaei, 2024b, 2024c).

To address this challenge, bioremediation techniques are designed to optimize microbial processes through strategic modification of the contaminated environment. One such approach is biostimulation, which involves amending the contaminated soil with organic and inorganic additives. Urea, a common source of nitrogen, has been reported to enhance microbial growth and metabolism, leading to increased TPH degradation in contaminated soil (Zhang et al., 2021). Similarly, ammonium sulfate, a source of nitrogen and sulfur, has been shown to stimulate the activity of hydrocarbon-degrading microorganisms, resulting in enhanced TPH degradation (Gao et al., 2021; Jiang et al., 2023). NPK fertilizers, which provide essential nutrients for plant growth, can indirectly influence TPH bioremediation by promoting plant growth and root exudation, which in turn can stimulate microbial activity in the rhizosphere and enhance TPH degradation (Tangahu et al., 2017). Shahi et al. (2016) found in their study that introducing inorganic nutrients such as carbon, nitrogen, and phosphorus in specific ratios to the environment can significantly enhance the degradation of pollutants, such as petroleum hydrocarbons. Suja et al. (2014) were able to promote the bioremediation of crude oil-contaminated soil by introducing nutrients such as mineral salts and NPK, which stimulated the growth of nearby microbial communities.

The process of bioremediation involves the use of microorganisms to detoxify or remove pollutants, utilizing their diverse metabolic capabilities. This method has been evolving and proven effective in the removal and degradation of various environmental pollutants, including those produced by the petroleum industry (Medina-Bellver et al., 2005). Rassaei (2025a) demonstrated Oleic acid (OA), a monounsaturated fatty acid, has shown promise as a soil amendment for remediating TPH-contaminated soils. OA improves soil properties by increasing pH, organic matter, and nutrient availability, while facilitating TPH reduction through emulsification, solubilization, and enhanced microbial degradation. Its effectiveness is concentration- and time-dependent, highlighting OA's potential as a sustainable amendment to improve soil health and support plant growth in contaminated environments. Bioremediation technology is considered noninvasive and relatively cost-effective (April et al., 2000). Successful oil spill bioremediation relies on establishing and maintaining conditions that promote enhanced oil biodegradation rates in the affected environment. Numerous scientific review articles have addressed the factors influencing the rate of oil biodegradation (Foght and Westlake, 1987; Leahy and Colwell, 1990; Zobell, 1946).

Two main approaches to oil spill bioremediation are bioaugmentation, which involves adding known oil-degrading bacteria to supplement the existing microbial population, and biostimulation, which stimulates the growth of indigenous oil degraders through the addition of nutrients or other growth-limiting cosubstrates. The use of poultry manure as an organic fertilizer in contaminated soil has also been reported (Okolo et al., 2005), showing enhanced biodegradation in its presence alone. Maki et al. (2001) found that photo-oxidation increased the biodegradability of petroleum hydrocarbons by improving their bioavailability and enhancing microbial activities. Biosurfactants, a diverse group of surface-active chemical compounds produced by various microorganisms (Ilori et al., 2005; Muthusamy et al., 2008; Wu et al., 2022), play a role in enhancing the solubilization and removal of contaminants (Bai et al., 1997). Surfactants also promote biodegradation by increasing the bioavailability of pollutants (Lawniczak et al., 2013). Das and Chandran (2011) reported on the bioremediation of oil sludge using biosurfactants. In contrast to biological approaches, physical technologies and chemical oxidation methods offer faster soil contamination remediation. For instance, a soil washing technique with surfactant enhancement successfully eliminated 97% of diesel fuel from a soil column within a 60-h timeframe (Khalladi et al., 2009). Previous research has demonstrated the feasibility of soil washing, while chemical methods, such as hydrogen peroxide (HP), permanganate (PM), ozone, and Fenton reactions, have shown promise for efficient in situ soil remediation with shorter treatment cycles (Chen et al., 2016; Jamialahmadi et al., 2015; Li et al., 2014). Recently, advanced oxidation processes utilizing activated persulfate (PS) have been utilized for soil remediation (Zhang et al., 2015). PS, a potent oxidant, can be stimulated by various activators like transition metals (Me), heat, ultrasound, electrical current, ultraviolet (UV) light, and alkali conditions to generate sulfate free radicals with a redox potential (E0) of 2.6 V (Chen et al., 2015; Gao et al., 2012; Kemp et al., 2013). Research has indicated that *P*S-based oxidation activities have been effective in decomposing various contaminants, including polyaromatic hydrocarbons (PAHs), dyes, halogen-containing compounds, and diesel (Chen et al., 2020; Lominchar et al., 2018; Lin et al., 2017; Silveira et al., 2017; Peng et al., 2020).

However, previous studies have predominantly focused on PS applications in liquid-phase treatment, with limited attention given to soil remediation, especially regarding TPH-contaminated soil and its impact on plant growth factors. In recent years, researchers have begun exploring the removal of TPH using activated PS, primarily with a focus on ferric-activated PS for its cost-effectiveness (Pardo et al., 2015; Oh and Shin, 2014). Some studies have also highlighted base-activated PS for TPH degradation (Lominchar et al., 2018), while others have investigated ultrasound or heat-activated PS for TPH elimination. Lei et al. (2020) utilized ultrasound-activated PS to treat soil contaminated with petroleum hydrocarbons (29 mg kg^{-1} and 100 mg kg^{-1}) and PAHs, achieving an 88.9% TPH removal efficiency.

As no previous research has examined the impact of SP function as a biosurfactant on TPH-contaminated soil and rice growth factors, the main objective of this study was to investigate the potential of using SP to remediate agricultural soils contaminated with TPH and revitalize rice growth. Specifically, the study aimed to: 1) Evaluate the effects of TPH contamination on soil properties, including pH, organic matter, and nutrient availability. 2) Determine the impacts of TPH on rice growth factors such as root development, shoot growth, chlorophyll content, plant height, grain yield, and biomass production. 3) Examine the ability of SP to degrade TPH contaminants through chemical and biological means and restore soil conditions. 4) Assess the effectiveness of SP in mitigating negative impacts of TPH on rice and improving plant growth conditions. 5) Study the influence of different TPH and SP levels and their interaction on soil remediation efficiency and rice productivity. 6) Investigate other implications of TPH contamination and SP treatment regarding byproduct formation, interactions with soil variables, and impacts on sustainable land management. The overarching goal was to evaluate the potential of SP-assisted bioremediation for TPH-contaminated agricultural soils and highlight its promise and limitations for optimizing contaminated land reuse.

This research presents a unique and innovative approach to the study of bioremediation and its impact on ecotoxicity. This study provides novel insights into the impact of TPH contamination on soil properties and plant growth factors in rice, as well as the effectiveness of SP as a remediation strategy. The research highlights the specific effects of TPH contamination on various

parameters and demonstrates the potential of SP to mitigate these negative impacts. The study also explores the underlying mechanisms of SP in degrading TPH contaminants and improving soil and plant conditions.

2. Method and materials

2.1. Area description

Kamfirouz is located on the Kamfirouz Plain, between 30° 25′ and 30° 10′ N and 52° 20′ to 52° 10′ E, on the eastern and western banks of the Chour River, north of the Doroodzan Dam, and 110 km northwest of Shiraz, Fars-Iran (Rassaei, 2022a). The area covers 11300 ha and has a mean annual precipitation of 580 mm, with the lowest and highest rainfall being 370 and 800 mm, respectively (Rassaei, 2022b, 2022c). Kamfirouz has calcareous soil and is known for its rice farming in Fars, Iran, with the land in the region being allocated to rice farming (Rassaei, 2023a).

2.2. Soil

Soil samples (top soil, depth 0–25 cm) were collected from the Kamfiroz series in Fars province, Iran. The FAO-WRB system identified Calcic Luvisols (LK) as the soil reference groups (Anjos et al., 2015) (World Reference Base for Soil Resources, 2014). The soil samples were passed through a 2 mm sieve, air-dried for 72 h, and stored for analysis (Allen et al., 1974).

The soil samples were analyzed for their texture using the hydrometer method as described by Gee and Bauder (1986). The pH of the soil in a 1:1 mixture with water was determined according to the method of Richards (1969). Additionally, the electrical conductivity (EC) of the soil in the 1:1 suspension extract of soil and water (Gupta, 2000), organic matter (OM) by the Walkley and Black method (Nelson and Sommers, 1982), calcium carbonate equivalent (CCE) by the technique described by Richards (1969) for neutralization using acid was employed, and the 0.005 M DTPA solution was used to extract Mn, Fe, Zn, Cu, and Cd, which were all readily available. The results are presented in Table 1 (Rassaei et al., 2020).

2.3. SP and characterization

The SP were purchased from Sigma-Aldrich Inc. US (Case no. 7775-27-1) with formula $Na_2O_8S_2$, molecular weight of 238.10 g/mol, water solubility of 730 g/l at 25 °C, reagent grade of ≥98%, crystalline form and white color (Schematic representation of SP, Fig. 1). The surface chemistry of SP was analyzed using Fourier transform infrared (FTIR) spectroscopy, this allowed for a comprehensive characterization of the material.

Table 1
Selected chemical properties of soils used. 1- Organic matter, 2- Calcium carbonate equivalent, 3- Cation exchange capacity, 4- Electrical conductivity, 5- None detectable (nd).

Properties	Amount
	Kamfiroz series
Sand (%)	51.8
Silt (%)	25.5
Clay (%)	22.7
Texture	Sandy Clay Loam
OM (%)[1]	3.4
CCE (%)[2]	52.3
CEC (Cmol kg^{-1})[3]	8.34
EC (dS. m^{-1})[4]	0.74
pH (1:1)	7.92
Fe (mg/kg)	3.3
Mn (mg/kg)	5.4
Zn (mg/kg)	0.33
Cu(mg/kg)	0.93
Cd (mg/kg)	nd
Cr (mg/kg)	nd

The Fourier transform infrared spectroscopy (Perkin Elmer, FT-IR Spectrometer, Spectrum Two) was used to analyze the functional groups present in the SP samples. The spectra were collected between the 200–4000/cm ranges and revealed several absorption peaks within this region. For the peak of 573, this corresponds to the stretching vibration of the S–O bond. 601 cm-1: This peak can be attributed to the bending vibration of the S–O bond. For the peak of 734, this corresponds to the bending vibration of the S–O–S bond. For the peak of 1073, this is associated with the stretching vibration of the S=O bond. For the peak of 1191, this indicates the stretching vibration of the C–O bond, which may be present in impurities or additives in the sample. For the peak of 1265, this attributed to the symmetric stretching vibration of the S–O bond. For the peak of 1379, this corresponds to the asymmetric stretching vibration of the S–O bond. For the peak of 1391, this is associated with the symmetric stretching vibration of the O–S–O bond in persulfate ions. For the peak of 1457, this indicates the asymmetric stretching vibration of the O–S–O bond in persulfate ions. For the peak of 2844, this peak corresponds to the stretching vibration of the C–H bond in organic compounds. It suggests the presence of organic impurities or residual organic material in the SP sample. For the peak of 2942, this attributed to the stretching vibration of the C–H bond in organic compounds, similar to the previous peak. For the peak of 2974, this indicates the stretching vibration of the C–H bond in organic compounds, similar to the previous two peaks (Fig. 2).

2.4. Addition of TPH

The volume of diesel fuel needed to add 4%, 6%, and 8% of diesel fuel to 1 kg of soil (Schematic representation of diesel fuel, Fig. 3). For 4% diesel fuel addition: Percentage of diesel fuel = 4% total amount after addition = 1 kg + 4% of 1 kg = 1 kg + 0.04 kg = 1.04 kg (rounded to two decimal places). Volume of diesel fuel needed: percentage = (Amount of diesel fuel added/Total amount after addition) × 100. 4% = (Amount of diesel fuel added/1.04 kg) × 100 Amount of diesel fuel added = 4% × 1.04 kg = 0.0416 kg (rounded to four decimal places). Converting kg to ml: Density of diesel fuel is around 0.83 kg/l (approximate value), 0.0416 kg of diesel fuel is equivalent to 0.0416/0.83 l = 0.05 l = 50 ml (rounded to two decimal places). So, to add 4% diesel fuel to 1 kg of soil, approximately 50 ml of diesel fuel is needed. Similarly, for 6% diesel fuel addition: Percentage of diesel fuel = 6% Total amount after addition = 1 kg + 6% of 1 kg = 1 kg + 0.06 kg = 1.06 kg (rounded to two decimal places). Volume of diesel fuel needed: Percentage = (Amount of diesel fuel added/Total amount after addition) × 100 6% = (Amount of diesel fuel added/1.06 kg) × 100 Amount of diesel fuel added = 6% × 1.06 kg = 0.0636 kg (rounded to four decimal places). Converting kg to ml: 0.0636 kg of diesel fuel is equivalent to 0.0636/0.83 l = 0.077 l = 77 mLl (rounded to two decimal places). So, to add 6% diesel fuel to 1 kg of soil, approximately 77 ml of diesel fuel is

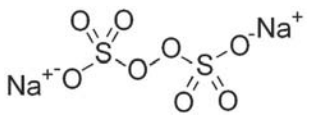


Fig. 1. Schematic representation of Sodium persulfate.

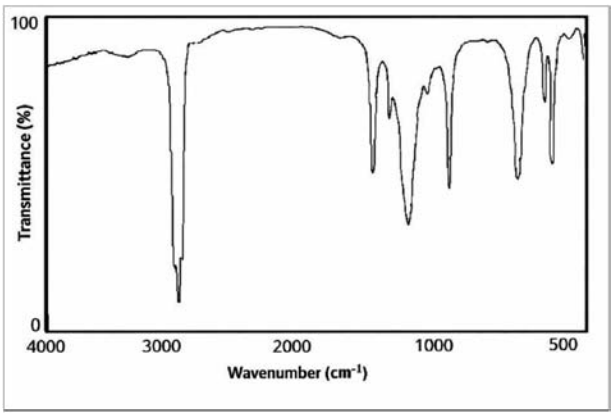


Fig. 2. FTIR transform of Sodium persulfate.

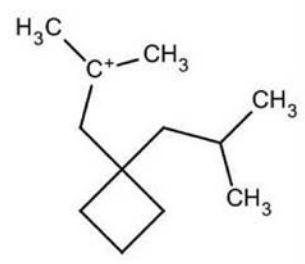


Fig. 3. Schematic representation of diesel fuel.

needed. And for 8% diesel fuel addition: Percentage of diesel fuel = 8% Total amount after addition = 1 kg + 8% of 1 kg = 1 kg + 0.08 kg = 1.08 kg (rounded to two decimal places). Volume of diesel fuel needed: Percentage = (Amount of diesel fuel added/Total amount after addition) × 100 8% = (Amount of diesel fuel added/1.08 kg) × 100 Amount of diesel fuel added = 8% × 1.08 kg = 0.0864 kg (rounded to four decimal places). Converting kg to ml: 0.0864 kg of diesel fuel is equivalent to 0.0864/0.83 l = 0.104 l = 104 mL (rounded to two decimal places). So, to add 8% diesel fuel to 1 kg of soil, approximately 104 mL of diesel fuel is needed. TPH were added to the freshly collected soil at of 0.0, 4.0, 6, and 8.0% weight of the fresh weight of the soil. The TPH was thoroughly mixed into the soil, and SP was applied uniformly to the TPH-treated soil samples at 0.0, 4.0, 6.0 and 8% weight (Rassaei, 2025a, 2025b). To describe the morphology and texture of soil, scanning electron microscopy (SEM, Fig. 4) were utilized before and after adding diesel fuel.

The Fourier transform infrared spectroscopy (Perkin Elmer, FT-IR Spectrometer, Spectrum Two) was used to analyze the functional groups present in the FX samples. The spectra were collected between the 500–4000/cm ranges and revealed several absorption peaks within this region. 1375.14: This peak is likely attributed to the symmetric bending of the $–CH_2-$ group in the aliphatic hydrocarbons present in diesel fuel. 1461.23: This peak is likely attributed to the bending vibration of $–CH_2-$ groups in aliphatic hydrocarbons 2856.14: This peak is likely attributed to the stretching vibration of the asymmetric $–CH_2-$ groups in aliphatic hydrocarbons. 2965.13: This peak is likely attributed to the stretching vibration of the symmetric $–CH_3$ groups in aliphatic hydrocarbons (Fig. 5).

2.5. Addition of SP into the soil

Preparation of a 0.5, 1.50% and 3.0% solution of SP in Crystalline form, with a molecular weight of $Na_2O_8S_2$ of 238.10 g/mol, is needed. The calculation of the amount of SP needed is also required:

0.5% concentration: 0.50% of 1000 g of soil = (0.50/100) × 1000 = 5 g. To convert this to the amount of SP needed, we use the molecular weight: Mass of SP = (5 g × (1 mol/1000 g)) × (238.10 g/mol) = 1.1908 g of SP. Therefore, to achieve a 0.50% concentration, 1.1908 g of SP need to be added to 1000 g' soil.

1.50% concentration: 1.50% of 1000 g of soil = (1.50/100) × 1000 = 15 g. To convert this to the amount of SP needed, we use the molecular weight: Mass of SP = (15 g × (1 mol/1000 g)) × (238.10 g/mol) = 3.5725 g of SP. Therefore, to achieve a 1.50% concentration, 3.5725 g of SP need to be added to 1000 g' soil.

3.0% concentration: 3.0% of 1000 g of soil = (3.0/100) × 1000 = 30 g. Mass of SP = (30 g × (1 mol/1000 g)) × (238.10 g/mol) = 7.143 g of SP. To achieve a 3.0% concentration, 7.143 g of SP need to be added to 1000 g of soil. It is important to consider the water solubility of SP (730 g/l at 25 °C) and the application method to ensure proper dissolution and distribution in the soil therefore,

To incorporate SP into the soil, these general steps can be followed:

(1) Measure the desired amount: Based on the calculations provided earlier, determine the amount of SP needed to achieve the desired concentration (1.50 and 3.0%).
(2) Dissolve SP: SP is highly soluble in water, so dissolve it in a suitable volume of water. Considering the water solubility of SP, which is 730 g/l at 25 °C, gradually add the calculated amount of SP to a container with water and stir until it is fully dissolved.
(3) Prepare the soil: Prepare the sandy clay loam soil by loosening it and removing any large debris or clumps, this will ensure better mixing and distribution of the SP solution.
(4) Apply the solution: Pour the SP solution evenly over the soil surface. The amount of solution should be sufficient to moisten the soil thoroughly, but avoid excessive runoff, the soil should be damp but not waterlogged.
(5) Mix the soil: After applying the solution, used a mixer to mix the soil and solution thoroughly to ensure that the SP solution is evenly distributed throughout the soil profile.
(6) Allow for reaction time: SP requires time to react with contaminants in the soil (15 days).

2.6. Plantation

Base fertilizers were applied to the soil three days prior to rice transplanting, including urea at 120 kg N ha^{-1}, double superphosphate at 90 kg P_2O_5 ha^{-1}, and potassium chloride (KCl) at 90 kg K_2O ha^{-1}. Thirty-day-old healthy rice seedlings (*Oryza sativa* L., cv. Lanjan), approximately 15–20 cm in height, were transplanted into the prepared soil to ensure uniform growth conditions (Rassaei et al., 2020a; Rassaei, 2023a). For the first 20 days after transplanting, the water was continuously maintained at the soil surface to support seedling establishment. From day 20 until the full grain production stage (approximately 100 days after transplanting), the water level was raised to approximately 5 cm

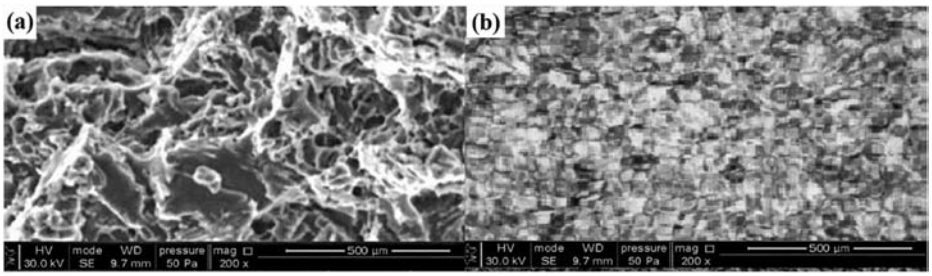


Fig. 4. SEM of Soil before (a) and after adding diesel fuel (b).

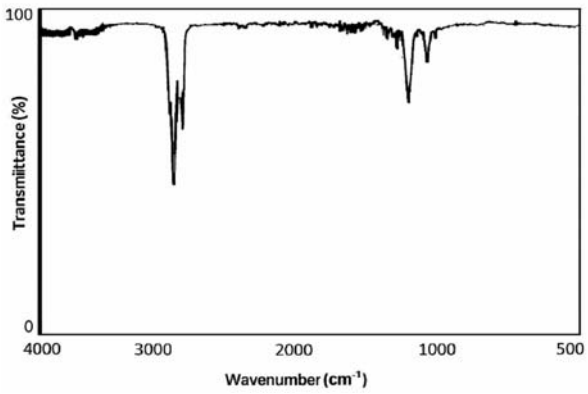


Fig. 5. FTIR transform of diesel fuel.

above the soil surface and maintained at this level. After this stage, irrigation was stopped to allow water to naturally evaporate for 48 h in preparation for harvest (Rassaei, 2023b, 2025c, 2025d).

2.7. Bioremediation study

The bioremediation study involved conducting laboratory analyses on soil samples collected from polluted areas that were amended with various treatments. The samples collected at different time points, such as 2 weeks, 4 weeks, and 8 weeks, were designated as weeks 2, 4, and 8, respectively. The experimental design series was denoted as 2–8 weeks. The effectiveness of bioremediation in reducing petroleum hydrocarbon contamination was monitored by measuring the rate of clean-up of TPH.

2.8. TPH quantification

The total petroleum hydrocarbon (TPH) content was determined using EPA8015 and the extraction of the petroleum hydrocarbon was conducted using cold extraction method with dichloromethane. In brief, a clean 50 mL extraction bottle was weighed with 3.0 g of the polluted sample, and 1 mL of 60 mg of dichlorooctadecane surrogate standard was added. The mixture was shaken for 5 h in a shaking water bath, and the extract was allowed to settle for at least 1 h. The sample was then filtered with cotton wool and sodium sulfate was added to a clean beaker washed with methylene chloride. The residues were washed with 20 mL of extraction solvent, filtered through a funnel, and transferred to a vial for analysis using gas chromatography (GC).

The procedure was carried out iteratively until a solution without any color was achieved. To each of the concentrated extracts, 4 mL of cyclohexane was added. The extract solution was then concentrated to 1 mL, and the extract volume was adjusted to about 2 mL with cyclohexane. Silica gel was activated by heating overshot at 130 °C. A slurry of 10 g of activated silica gel in methylene chloride was prepared and placed into a chromatographer column (autosampler) with a 10 mm internal diameter. A light tapping was applied to the column in order to settle the silica gel and extract the methylene chloride. Anhydrous sodium sulfate (2 mL) was added on top of the silica bed, and the column was preeluted with 40 mL of pentane at a rate of 2 mL/min. Before exposing the sodium sulfate layer to air, the eluate was discarded. Then, an extra 2 mL of cyclohexane was used to finalize the transfer of the 2 mL cyclohexane extract into the column. Further, 25 mL of pentane was added just before exposing the sodium sulfate layer to air, and elution was continued until the fraction was collected.

Next, 20 mL of the sample extract was poured into a separating funnel, and 10 mL of dichloromethane was added. The mixture was shaken for approximately 10 min, and the organic layer was allowed to separate. Before injecting the extract into the GC, the extraction mixture was passed through a filter paper containing 5 g of sodium sulfate into a vial. The analysis of the extract was performed using an HP Agilest 6890 GC (Agilent Technologies, UK) equipped with a flame atomization detector, capillary column (15×0.25 nm), when using an autosampler and a film thickness of 0.25 mm. The injection temperature and volume were set at 350 °C and 1 mL, respectively. Helium was used as the carrier gas at a flow rate of 2 mL/min, and hydrogen was applied during combustion in the flame ionization detector (FID). The column was initially held at 40 °C for 2 min. Saari et al. (2007) established a methodology that involved determining TPH values by multiplying the raw data from the FID graph or table with the dilution factor used for each sample.

2.9. Treatments

A greenhouse experiment was conducted in a typical rice paddy soil in the Kamfirouz Plain to track the treatments. The treatments included a control group with no TPH or SP, a group with only TPH, a group with only SP, and a group with both TPH and SP. The research was carried out utilizing a randomized complete block design (RCBD) with three replications. The treatments included three levels of SP (0.0, 1.50, and 3.0 percent weight) and four levels of TPH (0.0, 4.0, 6, and 8.0 percent weight). Soil samples were analyzed before and after the TPH levels for some physical chemical properties like pH, EC, SOM, and pH, available nutrient (N, P, and K). Soil nitrogen availability was determined using the alkali diffusion method with 1.0 mol l^{-1} NaOH. Available phosphorus was measured using the molybdenum antimony anti-colorimetric method with 0.5 mol l^{-1} $NaHCO_3$. Soil potassium availability was determined using the flame photometric method with 1.0 mol l^{-1} NH_4OAC. The experiment was performed in 36 plastic pots, with each pot containing 6 kg of

soil. Prior to planting rice, soil samples were placed in growth chambers for 15 days with a 5 cm layer of deionized water on top to create a waterlogged condition (Rassaei et al., 2020).

2.10. Rice growth and yield measurements

Plant height with T ruler, chlorophyll index with chlorophyll meter (Konica Minolta Chlorophyll Meter Spad-502 Plus, Japan), leaf area using surface measuring device (Portable Leaf Area Meter AM350, UK) were determined. The rice plant and its roots were harvested and after washing, they were placed at 60 °C. After 36 h, the dry weight of shoots and roots was measured using a digital scale (LTP, China) and then were grinded by soil grinder device (FZ-102, China), and then ground by a soil grinder device (FZ-102, China). For examine rice productivity the quantity of grains, were taken per treatment.

2.11. Data analysis

Data analysis was conducted using SPSS Version 16.0, a statistical software package developed by IBM in the United States. Repeated measures ANOVA and Multivariate tests were performed to check the effect of treatments. The means were compared by Duncan's tests to see the statistical significance of the results as a result of treatments with SP, TPH, and their interaction. A statistical significance level of <0.05 was indicated by the P value.

3. Results and discussion

3.1. Effect of SP and TPH and their interaction on soil-physiochemical properties of TPH contaminated soil

Based on the data in Table 2, the pH of the soil decreased with increasing TPH contamination, from 7.92 in the control to 7.10 with 8.0% TPH. Addition of SP alone also slightly decreased pH. EC decreased with higher TPH contamination and also with addition of SP, both alone and combined with TPH. The highest reduction in EC was seen with SP 3.0% combined with 8.0% TPH. Organic matter content decreased with higher TPH contamination. Addition of SP alone did not significantly change organic matter, but SP combined with TPH further decreased organic matter levels. Available N, P and K all decreased with increasing TPH contamination. Addition of SP alone increased N and K levels, while SP combined with TPH gave mixed results for nutrient availability. Higher SP dose gave higher nutrient levels. TPH contamination acidified the soil by decreasing pH. SP addition had a similar effect, likely due to production of sulfate and other acids during decomposition. Reduction in EC with SP addition can be attributed to decomposition of contaminants and conversion of soluble compounds to insoluble forms, removing ions from solution. Decrease in organic matter with TPH and further decrease with SP can be explained by oxidation of organic compounds by SP. Reduction in available nutrients with TPH is likely due to immobilization by contaminants. SP addition helps restore some nutrient availability by decomposing contaminants, though effect depends on SP dose and TPH level.

In summary, the results show that TPH contamination acidifies soil and reduces organic matter and nutrient availability, while addition of SP helps remediate these effects to some extent by decomposition of the contaminants. The extent of remediation depends on the SP dose and TPH level. When TPH contaminants like oil and grease degrade in soil, they release organic acids. These organic acids donate hydrogen ions (H^+) to the soil solution, increasing the H^+ concentration and acidifying the soil. This decreases soil pH. Common organic acids released include naphthenic acids and fatty acids from the degraded TPH. The organic acids can also displace base cations like calcium (Ca^{2+}) and magnesium (Mg^{2+}) from cation exchange sites on soil particles. This loss of base cations further contributes to soil acidification and lower pH. The TPH contaminants bind to and immobilize ions that contribute to electrical conductivity (EC), including nitrate (NO_3^-), phosphate (PO_4^{3-}), and base cations like Ca^{2+} and Mg^{2+}. By removing these ions and charged particles from the soil solution, TPH contamination lowers EC. Additionally, the TPH contaminants can alter soil properties in ways that decrease ion mobility even if ions are still present. For example, they can lower soil aeration and water content, limiting ion movement and diffusion. As the TPH degrades, some soluble soil compounds are converted to insoluble forms, removing ions from solution and further lowering EC.

SP acts as an oxidizing agent that can decompose the TPH contaminants in soil. By breaking down the contaminants, SP releases nutrients and ions that were immobilized by the TPH, helping to restore EC levels to some extent. SP counteracts the mechanisms through which TPH releases organic acids and displaces base

Table 2
DMRT table of effect of Sodium persulfate (SP) on soil-physiochemical properties of total petroleum hydrocarbon TPH contaminated soil. Where, control = NO added TPH nor Sodium persulfate, TPH 4.0% = 4.0% contaminated soil with TPH, TPH 6.0% = 6.0% contaminated soil with TPH, TPH 8% = 8.0% contaminated soil with TPH, Sodium persulfate 1.50%, adding only 1.50% Sodium persulfate, Sodium persulfate 1.50%* TPH 4.0% = Added 1.50% Sodium persulfate to Contaminated soil with 4.0% TPH, Sodium persulfate 1.50%* TPH 6.0% = Added 1.50% Sodium persulfate to Contaminated soil with 6.0% TPH, Sodium persulfate 1.50%* TPH 8% = Added 1.50% Sodium persulfate to Contaminated soil with 8.0% TPH, Sodium persulfate 3.0%, adding only 3.0% Sodium persulfate, Sodium persulfate 3.0%* TPH 4.0% = Added 3.0 % Sodium persulfate to Contaminated soil with 4.0% TPH, Sodium persulfate 3.0%* TPH 6.0% = Added 3.0% Sodium persulfate to Contaminated soil with 6.0% TPH, Sodium persulfate 3.0%* TPH 8.0% = Added 3.0% Sodium persulfate to Contaminated soil with 8.0% TPH.

Treatment	Mean pH	Mean EC (dSm^{-1})	Mean Organic matter (%)	Mean available nutrient		
				N (%)	P (%)	K (%)
Control	7.92 a	0.74 a	3.40 a	0.56 a	2.21 a	0.63 a
TPH 4.0%	7.55 b	0.68 ab	3.04 b	0.43 b	2.12 ab	0.31 b
TPH 6.0%	7.14 c	0.63 b	2.81 c	0.34 c	1.98 bc	0.22 c
TPH 8.0%	7.10 c	0.60 c	2.70 c	0.30 c	1.90 c	0.20 c
SP 1.50%	7.35 b	0.66 ab	3.05 b	0.45 ab	2.05 ab	0.40 b
SP 1.50%* TPH 4.0%	7.10 c	0.62 b	2.88 b	0.39 b	1.98 bc	0.28 c
SP 1.50%* TPH 6.0%	7.02 c	0.59 c	2.79 b	0.35 bc	1.93 c	0.25 c
SP 1.50%* TPH 8.0%	7.00 c	0.57 c	2.68 c	0.33 c	1.85 c	0.22 c
SP 3.0%	7.26b	0.67 ab	3.12 b	0.49 a	2.10 ab	0.51 a
SP 3.0%* TPH 4.0%	7.05 c	0.63 b	2.94 b	0.42 b	1.98 bc	0.36 c
SP 3.0%* TPH 6.0%	7.00 c	0.60 c	2.83 b	0.37 bc	1.93 c	0.32 c
SP 3.0%* TPH 8.0%	6.97 c	0.58 c	2.74 c	0.35 c	1.85 c	0.29 c

Significance is shown by letter in each column, with different letters indicating significant differences ($p < .05$) between treatments within the same column.

cations, thus helping to mitigate soil acidification and raise pH levels. However, the extent of remediation depends on how much of the TPH contaminants the SP can decompose. Higher doses of SP and lower levels of TPH contamination allow for more complete decomposition and better remediation of soil properties. SP releases immobilized nutrients and ions. SP acts as a strong oxidizing agent that can decompose the large hydrocarbon molecules in TPH contaminants like oil and grease. As SP oxidizes and breaks down the TPH contaminants, it frees up any nutrients and ions that were immobilized by binding to the TPH molecules. This includes releasing nitrates, phosphates, and base cations that were immobilized and not contributing to the soil EC. Releasing these nutrients and ions that were previously bound up in the TPH contaminants help restore some of the EC levels in the soil solution.

SP counters organic acid release and cation displacement. As SP decomposes TPH contaminants, it breaks the bonds holding together the organic acids that were contributing to soil acidification. This reduces the amount of organic acids released during TPH degradation, countering one of the mechanisms by which TPH lowered soil pH. SP can also oxidize and decompose compounds that displace base cations from soil exchange sites, thereby mitigating cation displacement and soil acidification caused by TPH. These actions help SP increase the soil pH and counter some of the soil acidification caused by TPH contamination. Degree of remediation depends on SP dose and TPH level. The extent to which SP can remediate the negative effects of TPH contamination depends on how much of the TPH it can decompose. Higher doses of SP mean it can oxidize and break down a larger portion of the TPH contaminants. This allows it to more effectively release immobilized nutrients, reduce organic acid release, and displace fewer base cations. Lower levels of TPH contamination also make it easier for SP to fully decompose the TPH present, improving its remediation effectiveness.

3.2. Effect of SP on TPH removal from soil over weeks

Table 3 and Fig. 6 shows removal and remained TPH over times. Application of SP effectively reduced the amount of TPH remaining in the contaminated soils over time. Higher doses of SP (3.0% vs. 1.50%) and longer incubation periods (8 weeks vs 2 weeks) led to greater TPH removal at each initial contamination level (4%, 6% and 8% diesel fuel). For example, after 8 weeks the mean TPH remaining in the 4.0% contaminated soil was 10.6 g/kg with 1.5% SP and 9.6 g/kg with 3.0% SP, representing reductions of 26% and 32% respectively from the initial TPH level. Likewise, 8.0% contaminated soil had 20.4 g/kg TPH left with 1.50% SP and 18.4 g/kg with 3.0% SP after 8 weeks, which are reductions of 22% and 27% from the original concentration. At a given SP dose and incubation time, higher initial TPH concentrations resulted in more TPH remaining, though the absolute reductions tended to be greater at higher contamination levels.

In general, the differences between treatments widened over time, indicating that SP continued to degrade TPH throughout the 8-week period. The results demonstrate that SP is effective for remediating diesel-contaminated soil, with degradation effectiveness increasing at higher doses and longer exposure times. However, higher doses may be required to achieve the same level of TPH removal in more heavily contaminated soils. Other soil properties and crop growth responses would need to be evaluated to optimize a balanced remediation strategy. Overall, the data show that SP has good potential as an in situ remediation option for diesel-contaminated agricultural soils, though more research is warranted to fully understand its effects and develop practical recommendations.

There are several reasons why applying SP resulted in the observed reductions in TPH in the contaminated soils: (1) SP dissociates into sulfate radicals (SO_4^-) in water or moist soil, these sulfate radicals are strong oxidizing agents that can react with and break down hydrocarbon molecules like those in diesel fuel. (2) The sulfate radicals react primarily by abstracting hydrogen atoms from the hydrocarbon chains, this disrupts the molecular structure and leads to decomposition of the longer-chain TPH compounds into shorter-chain or mineralized end products. (3) The rate of TPH degradation depends on the concentration of sulfate radicals, which in turn is influenced by the dose of SP applied, higher doses produce more sulfate radicals and result in greater TPH reduction. (4) The exposure time also affects degradation, the longer the soil is exposed to sulfate radicals from SP, the more TPH decomposition can occur, this explains the increasing TPH removal from week 2 to week 8. (5) The initial TPH concentration also influences the degradation process, soils with higher TPH levels provide more hydrocarbon substrates for the sulfate radicals reaction, however, the radicals may be saturated or scavenged at very high contaminant concentrations. (6) Other factors like soil moisture, pH, and temperature can also impact the generation and reactivity of sulfate radicals, but within this experiment, the SP dose and incubation time appear to be the primary drivers of differences in TPH reduction between treatments. In summary, the degradation of TPH by SP occurs through reactions between sulfate radicals and

Table 3
DMRT table of effect of Sodium persulfate (SP) on TPH removal from soil in weeks. Where, TPH 4.0% = 4.0% contaminated soil with TPH, TPH 6.0% = 6% contaminated soil with TPH, TPH 8.0% = 8.0% contaminated soil with TPH, Sodium persulfate 1.50%*TPH 4.0% = Added 1.50% Sodium persulfate to Contaminated soil with 4.0% TPH, Sodium persulfate 1.50%*TPH 6.0% = Added 1.50% Sodium persulfate to Contaminated soil with 6.0% TPH, Sodium persulfate 1.50%*TPH 8.0% = Added 1.50% Sodium persulfate to Contaminated soil with 8.0% TPH, Sodium persulfate 3.0%* TPH 4.0% = Added 3.0% Sodium persulfate to Contaminated soil with 4.0% TPH, Sodium persulfate 3.0%* TPH 6.0% = Added 3.0% Sodium persulfate to Contaminated soil with 6.0% TPH, Sodium persulfate 3.0%* TPH 8% = Added 3.0% Sodium persulfate to Contaminated soil with 8.0% TPH.

Treatment	Mean remained TPH g/kg	Mean remained TPH g/kg	Mean remained TPH g/kg
	Incubation times		
	week 2	week 4	week 8
TPH 4.0%	14.2 a	12.0 a	11.2 a
TPH 6.0%	19.8 b	18.0 b	16.8 b
TPH 8.0%	26.4 c	24.0 c	22.4 c
SP 1.50%* TPH 4.0%	13.0 ab	11.4 ab	10.6 ab
SP 1.50%* TPH 6.0%	17.6 bc	16.2 bc	15.2 bc
SP 1.50%* TPH 8.0%	24.0 cd	22.0 cd	20.4 cd
SP 3.0%* TPH 4.0%	12.0 b	10.4 b	9.6 b
SP 3.0%* TPH 6.0%	16.0 c	14.4 c	13.2 c
SP 3.0%* TPH 8.0%	22.0 d	20.0 d	18.4 d

Significance is shown by letter in each column, with different letters indicating significant differences ($p < .05$) between treatments within the same column.

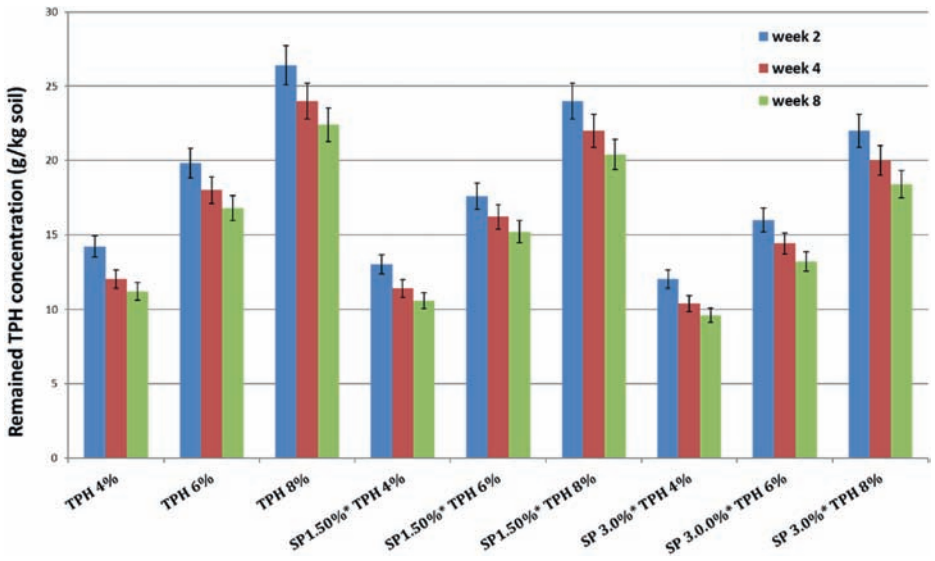


Fig. 6. Remained TPH in soil after treatments in 2, 4, and 8 weeks (Error bars for the selected charts series at 5% value).

hydrocarbon molecules, the concentration and exposure time of these reactive species determine the extent of degradation, as reflected in the results.

3.3. Effect of SP and TPH and their interaction on rice growth factors

3.3.1. Effect of SP on rice growth factors

The effects of SP on various variables related to rice growth were analyzed using an analysis of variance (ANOVA). The ANOVA results for root dry weight, shoot dry weight, chlorophyll content, plant height, grain yield, and biomass yield are presented below:

Root Dry Weight: The ANOVA results showed that the differences in root dry weight between groups were not statistically significant ($F = 1.614$, $p = .214$), suggesting that the different concentrations of SP did not have a significant effect on root dry weight.

Shoot Dry Weight: It is similar to root dry weight, the differences in shoot dry weight between groups were not statistically significant ($F = 1.795$, $p = .182$), the concentrations of SP did not have a significant impact on shoot dry weight.

Chlorophyll Content: The ANOVA results indicated that the differences in chlorophyll content between groups were not statistically significant ($F = 2.185$, $p = .128$), suggesting that the different concentrations of SP did not have a significant effect on chlorophyll content.

Plant Height: The differences in plant height between groups were not statistically significant ($F = 2.752$, $p = .078$), the concentrations of SP did not have a significant impact on plant height.

Grain Yield: The ANOVA results showed that the differences in grain yield between groups were not statistically significant ($F = 2.333$, $p = .113$), indicating that the different concentrations of SP did not have a significant effect on grain yield.

Biomass Yield: It is similar to grain yield, the differences in biomass yield between groups were not statistically significant ($F = 2.424$, $p = .104$). The concentrations of SP did not have a significant impact on biomass yield. Based on the ANOVA results (table was not providing because it was not significant), it appears that the different concentrations of SP did not have a significant effect on the variables related to rice growth. The lack of statistical significance suggests that the addition of SP did not lead to noticeable differences in root dry weight, shoot dry weight, chlorophyll content, plant height, grain and biomass yield.

The effects of SP on rice growth factors can vary depending on various factors, including specific experimental conditions, concentration of SP, duration of treatment, soil characteristics, and presence of contaminants. SP is primarily used as an oxidizing agent for soil remediation purposes, targeting organic contaminants such as hydrocarbons. While SP has the potential to promote the degradation of contaminants and improve soil conditions, its direct effects on rice growth factors may not be significant. SP is typically applied to the soil to address contamination issues rather than as a growth-promoting agent for crops like rice.

3.3.2. Effect of TPH rice growth factors

The results of the ANOVA for the effect of TPH on the variables are shown in Table 4. The ANOVA results indicate that the addition of TPH to the soil has a significant effect on all of the examined variables: root dry weight, shoot dry weight, chlorophyll content, plant height, grain yield, and biomass. The p-values for all variables are less than 0.001, indicating highly significant differences between the TPH levels. For root dry weight, shoot dry weight, chlorophyll content, and biomass, there is a clear increasing trend in the variability (sum of squares) between groups as TPH levels increase. This suggests that higher TPH concentrations lead to greater variation and reduce performance in these variables. The variability (sum of squares) within groups remains relatively low, indicating consistency within each TPH level group. Similarly, for plant height and grain yield, the variability between groups increases with higher TPH levels, indicating a negative impact on these variables. The variability within groups remains relatively low, suggesting consistent measurements within each TPH level group. These results suggest that TPH contamination negatively affects rice growth factors, including root and shoot development, chlorophyll content, plant height, grain yield, and biomass accumulation. Higher TPH levels in the soil lead to reduced growth and productivity of the rice plants. The negative effects observed on rice growth factors in the presence of TPH contamination can be attributed to several factors:

(1) **Toxicity:** TPH are complex mixtures of petroleum-derived compounds, including polycyclic aromatic hydrocarbons

Table 4
ANOVA table between TPH and rice growth factors. The table presents the type III sum of squares, degrees of freedom (df), mean square, F-value and significance (p-value) for each dependent variable when TPH was treated as the fixed factor.

		Sum of Squares	df	Mean Square	F	Sig.
Root dry weight	Between Groups	4.997	3	1.666	81.383	.000
	Within Groups	.655	32	.020		
	Total	5.652	35			
Shoot dry weight	Between Groups	22.580	3	7.527	45.530	.000
	Within Groups	5.290	32	.165		
	Total	27.869	35			
Chlorophyll	Between Groups	539.897	3	179.966	44.662	.000
	Within Groups	128.945	32	4.030		
	Total	668.842	35			
Plant height	Between Groups	6258.908	3	2086.303	78.312	.000
	Within Groups	852.509	32	26.641		
	Total	7111.417	35			
Grain	Between Groups	21.074	3	7.025	136.637	.000
	Within Groups	1.645	32	.051		
	Total	22.719	35			
Biomass	Between Groups	59.799	3	19.933	137.858	.000
	Within Groups	4.627	32	.145		
	Total	64.425	35			

(PAHs), which can disrupt plant physiology and metabolism. These compounds may interfere with nutrient uptake, inhibit enzymatic activities, and damage cellular structures, ultimately reducing biomass production (Li et al., 2025; Li et al., 2023). PAHs have been reported to impair photosynthesis, alter membrane integrity, and induce DNA damage in plants, contributing to stunted growth at higher concentrations (Chen et al., 2020).

(2) **Altered Soil Physical Properties:** High levels of TPH can modify soil texture, structure, and water-holding capacity. The formation of hydrophobic layers around soil particles can reduce water infiltration and availability, causing water stress and limiting root development (Chen et al., 2020).

(3) **Nutrient Imbalance:** Petroleum hydrocarbons may disrupt nutrient cycling and microbial activity, resulting in deficiencies or toxicities of essential elements, such as nitrogen, phosphorus, and potassium, these imbalances can hinder root and shoot growth, chlorophyll synthesis, and overall plant performance (Li et al., 2023; Wang et al., 2022).

(4) **Oxidative Stress:** Metabolism of TPH in soil or by plants can generate reactive oxygen species (ROS), leading to oxidative damage of plant cells and tissues. Elevated ROS levels can impair photosynthesis, respiration, and other metabolic processes, ultimately limiting growth and productivity (Wang et al., 2022).

(5) **Interactions with Soil Microorganisms:** TPH contamination can alter the composition and function of soil microbial communities, which are essential for nutrient cycling, organic matter decomposition, and plant growth promotion. Changes in microbial activity may further exacerbate nutrient limitations and affect plant health (Sah et al., 2022).

(6) **Hormonal Disruption:** TPH may interfere with plant hormone signaling, such as auxins and gibberellins, reducing cell division and elongation. Additionally, altered membrane permeability can restrict water and nutrient uptake, imposing further constraints on growth (Huang et al., 2019)

It is important to note that the specific mechanisms underlying the negative effects of TPH contamination on rice growth factors may vary depending on the specific characteristics of the petroleum hydrocarbons, soil conditions, and plant species involved. Conducting further research and investigations into these mechanisms can provide more insights into the specific pathways through which TPH exerts its detrimental effects on rice plants. So in summary, the various hydrocarbon compounds within TPH are likely phytotoxic and can interfere with multiple plant growth mechanisms through membrane disruption, nutrient deficiency, hormonal imbalance, and metabolic toxicity. These inhibitory effects on growth appear to increase in a concentration-dependent manner, leading to progressively as TPH concentrations rise from 4.0% to 6.0%–8.0%. TPH acts as an effective growth inhibitor for rice plants, however, the inhibitory effects increase in a concentration-dependent manner, with the highest concentrations of 8.0% producing the most significant reductions in plant height.

3.3.3. Effect of SP on TPH levels and its effectiveness to improve rice growth factors

Table 5 shows that application of SP significantly increases root and shoot dry weight (Fig. 7), chlorophyll content (Fig. 8), plant height (Fig. 9), grain and biomass yield of rice (Fig. 10), compared to contaminated soil without any amendment. Root dry weight, shoot dry weight, chlorophyll content, plant height and grain yield increased with increasing concentration of SP. The highest values were obtained with 3.0% SP. SP acts as an oxidizing agent that chemically oxidizes the contaminants and breaks them into smaller, less toxic or nontoxic compounds. It releases oxygen which helps aerobic microbial populations to degrade the contaminants. It also improves soil properties like porosity, aeration, aggregation and nutrient availability which have positive impact on plant growth. Addition of SP significantly increased root and shoot growth of rice which indicated that SP decreased the phytotoxic effect of TPH and increased availability of nutrients to the plant. Increase in chlorophyll content and plant height also suggested that SP decreased stress and improved growth of rice. Higher grain yield with increasing concentration of SP indicated reduction in TPH toxicity and availability of more nutrients for grain filling.

However, it is important to note that Figs. 7–10 show a slight decrease in chlorophyll content, plant height, and root/shoot dry weight under the combined treatment of 4% TPH with 3% SP compared to 1.5% SP. This suggests that at higher SP concentrations and certain TPH levels, SP may induce secondary stress effects on rice plants. Such effects could be due to soil chemical changes, including pH reduction or increased oxidative stress, which can

Table 5
Tests of between Sodium persulfate (SP) and TPH on ric growth factors. The table presents the type III sum of squares, degrees of freedom (df), mean square, F-value, significance level and R^2 values when tested for the interaction between SP and TPH as fixed factors on the rice growth factors. a. R Squared = .987 (Adjusted R Squared = 0.981); b. R Squared = 0.944 (Adjusted R Squared = 0.918); c. R Squared = 0.955 (Adjusted R Squared = 0.935); d. R Squared = 0.991 (Adjusted R Squared = 0.986); e. R Squared = 0.997 (Adjusted R Squared = 0.996); f. R Squared = 0.999 (Adjusted R Squared = 0.998).

Source	Dependent Variable	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	Root dry weight	6.738[a]	11	.613	169.189	.000
	Shoot dry weight	33.381[b]	11	3.035	36.781	.000
	Chlorophyll	843.825[c]	11	76.711	46.787	.000
	Plant height	9128.199[d]	11	829.836	230.973	.000
	Grain	25.468[e]	11	2.315	777.298	.000
	Biomass	71.859[f]	11	6.533	2.104E3	.000
Intercept	Root dry weight	202.130	1	202.130	5.583E4	.000
	Shoot dry weight	998.204	1	998.204	1.210E4	.000
	Chlorophyll	24825.190	1	24825.190	1.514E4	.000
	Plant height	224133.290	1	224133.290	6.238E4	.000
	Grain	585.699	1	585.699	1.966E5	.000
	Biomass	1676.943	1	1676.943	5.402E5	.000
TPH * SP	Root dry weight	.298	6	.050	13.717	.000
	Shoot dry weight	1.397	6	.233	2.822	.032
	Chlorophyll	27.942	6	4.657	2.840	.031
	Plant height	140.700	6	23.450	6.527	.000
	Grain	.461	6	.077	25.791	.000
	Biomass	1.331	6	.222	71.457	.000
Error	Root dry weight	.087	24	.004		
	Shoot dry weight	1.980	24	.083		
	Chlorophyll	39.350	24	1.640		
	Plant height	86.227	24	3.593		
	Grain	.071	24	.003		
	Biomass	.075	24	.003		
Total	Root dry weight	208.954	36			
	Shoot dry weight	1033.565	36			
	Chlorophyll	25708.365	36			
	Plant height	233347.716	36			
	Grain	611.238	36			
	Biomass	1748.877	36			
Corrected Total	Root dry weight	6.825	35			
	Shoot dry weight	35.361	35			
	Chlorophyll	883.175	35			
	Plant height	9214.426	35			
	Grain	25.539	35			
	Biomass	71.934	35			

transiently inhibit plant growth. Moreover, excessive SP might disrupt beneficial soil microbial communities responsible for nutrient cycling and plant health. Therefore, while SP generally improves rice growth by degrading TPH and improving soil conditions, there appears to be an optimal SP concentration threshold beyond which its benefits may plateau or slightly decline. This highlights the need to carefully balance SP dose and contamination level for effective remediation without phytotoxicity. Further research is recommended to define these optimal concentrations and better understand the complex interactions between SP, TPH contamination, soil biology, and rice growth.

There are several reasons why SP amendment improved plant growth in TPH contaminated soil: (1) SP acts as an oxidizing agent that chemically oxidizes the TPH contaminants and breaks them into smaller, less toxic compounds, this reduces the toxicity and phytotoxic effect of TPH which allows better plant growth (Wu et al., 2016). (2) SP releases oxygen during decomposition that helps aerobic microorganisms to actively degrade the TPH contaminants, this reduces the TPH concentration in soil and mitigates its harmful effects on plants (Dai and Liu, 2025). (3) SP improves soil properties like porosity, aeration, aggregation and nutrient availability, this creates a more favorable environment for root growth and nutrient uptake by plants (Liu et al., 2023). (4) The oxidation products of TPH and SP are less toxic and some may be used as nutrients by plants and microbes, this additional nutrient source benefits plant growth (Wang et al., 2022). (5) SP improves plant growth primarily through mitigating soil toxicity and enhancing soil quality rather than directly affecting plant physiological processes (Richardson et al., 2011). (6) The heat generated by SP oxidation of TPH may enhance microbial activity and nutrient availability in soil, this could stimulate plant growth (Richardson et al., 2011).

In summary, SP facilitates TPH degradation, reduces toxicity, enhances soil properties, supplies additional nutrients and may directly affect plant physiology. All these factors contribute to better plant growth in TPH contaminated soil after SP amendment. In conclusion, the results demonstrate that SP can be used as an amendment for remediation of TPH contaminated soil, it enhances microbial degradation of TPH, improves soil properties and reduces toxicity which results in better growth and yield of rice. The study suggests that 3% concentration of SP can be used for effective remediation of TPH contaminated soil.

Petroleum hydrocarbon contamination of soil is a significant global environmental issue that has widespread consequences. Soil pollution resulting from oil and fuel spills is particularly extensive and environmentally damaging, posing threats to human health and ecosystems, especially in cold regions (Hewelke et al., 2018). The presence of refinery products deteriorates the biochemical and physicochemical properties of the soil, thereby impeding the growth and development of plants (Gospodarek et al., 2021). Contamination with petroleum-derived substances leads to water and oxygen deficits, as well as a shortage of available forms of nitrogen and phosphorus, causing significant changes in soil properties (Gospodarek et al., 2020). The contamination of soil with petroleum

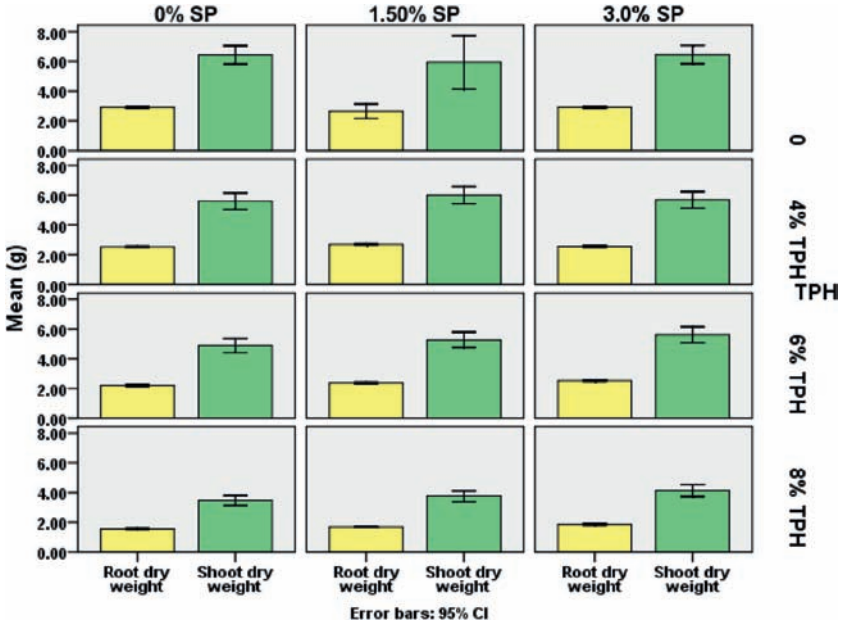


Fig. 7. Using TPH levels (0, 4, 6, and 8% w/w and Sodium persulfate levels 0, 1.50, and 3.0 % w/w) on rice root and shoot dry weight (g).

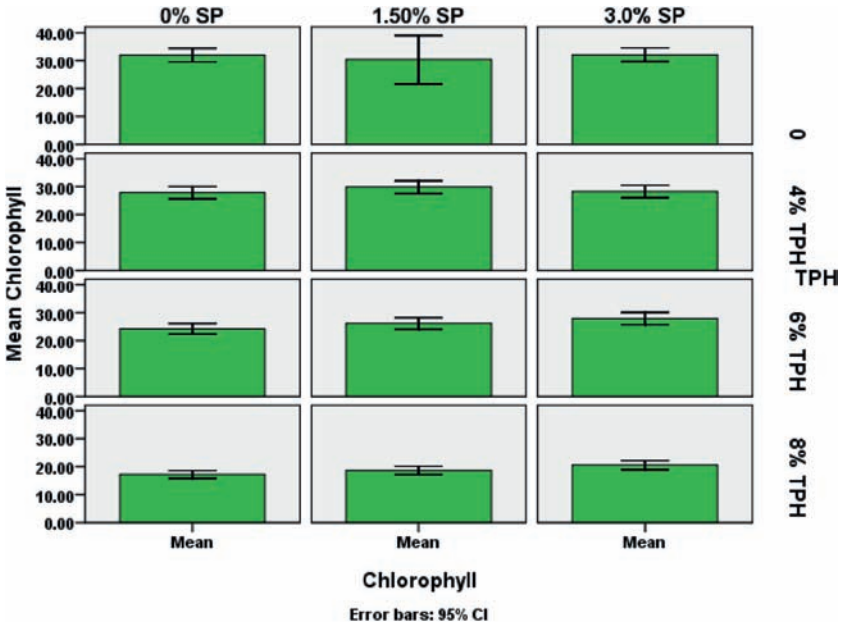


Fig. 8. Using TPH levels (0, 4, 6, and 8% w/w and Sodium persulfate levels 0, 1.50, and 3.0 % w/w) on rice chlorophyll.

hydrocarbons has far-reaching implications, including organic pollution of underground water, which hampers its use and results in economic losses, environmental problems, and decreased agricultural productivity (Sayed et al., 2021). The toxicity of petroleum hydrocarbons poses a vulnerable situation for microorganisms, plants, animals, and humans. Soil enzymes, which play a crucial role in soil biochemical reactions, are adversely affected by petroleum hydrocarbons (Lee et al., 2020). Oil spills create conditions that render essential nutrients like nitrogen and oxygen unavailable for plant growth, impacting plant health (Oyem and Oyem, 2013). Different levels of crude oil contamination have been shown to significantly reduce plant growth, as measured by plant height, fresh weight, and leaf area, with the degree of effect being proportional to the contamination levels. Crude oil pollution also has adverse effects on soil fertility and plant production, as it can impede plant growth and even lead to plant death by forming physical barriers and coating the roots (Kekere et al., 2011). Diesel oil exhibits a more pronounced inhibitory effect on nitrification compared to petrol (Kucharski et al., 2010). Moreover, diesel oil can cause chronic or acute effects in plants, interfering with their water relations (Bona et al., 2011).

Persulfate has gained significant popularity as an alternative oxidant for the remediation of organic contaminants. It possesses advantages such as high aqueous solubility, stability at room temperature, cost-effectiveness, and benign end products (Liu et al., 2014). Unlike H_2O_2, persulfate exhibits slower kinetic disappearance, which can be advantageous as it allows the oxidant

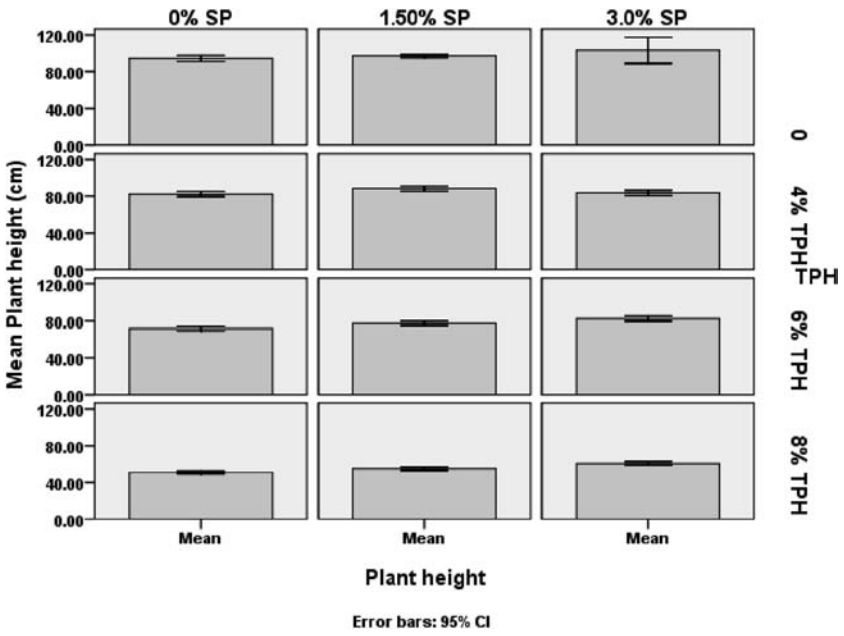


Fig. 9. Using TPH levels (0, 4, 6, and 8% w/w and Sodium persulfate levels 0, 1.50, and 3.0 % w/w) on rice height.

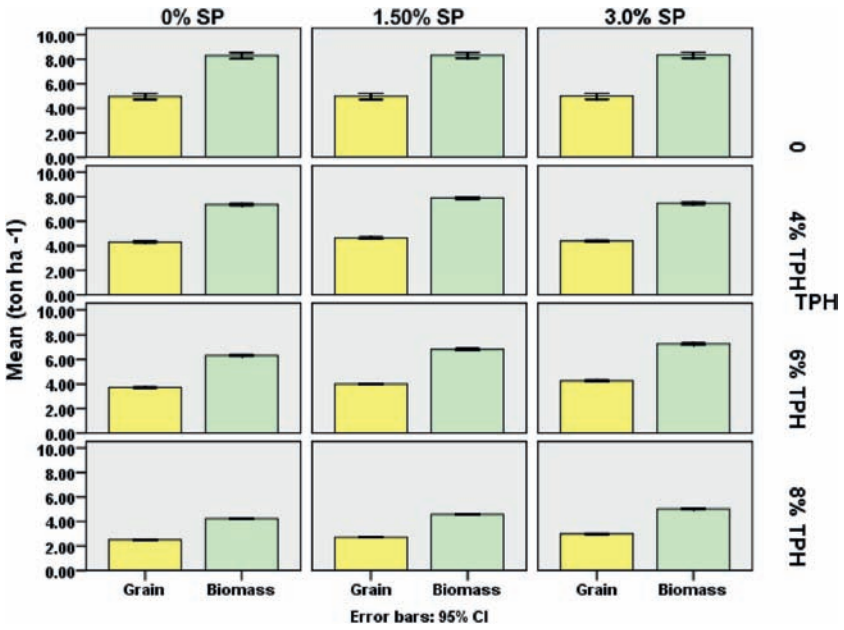


Fig. 10. Using TPH levels (0, 4, 6, and 8% w/w and Sodium persulfate levels 0, 1.50, and 3.0 % w/w) on rice grain and biomass yield in tonha^{-1}.

to reach the source zone (Liu et al., 2014; Osgerby, 2006). The sulfate (SO4•−) radicals produced by persulfate are more stable than hydroxyl radicals (HO•) and can be transported over longer distances in the sub-surface, persisting for weeks (Huang et al., 2002). Additionally, sulfate radicals have a lower affinity for natural soil organics (Brown and Robinson, 2004; Usman et al., 2012). However, unreacted persulfate molecules may remain in the soil, potentially reducing the pH and adversely affecting the soil microbial community and subsequent biodegradation processes (Chen et al., 2015). Chemical oxidation treatments can influence bacterial populations and alter redox conditions, but they can also enhance biodegradation by increasing substrate contaminant bioavailability and biodegradability, providing oxygen to microbial communities and releasing nutrients in some cases, thereby improving bioremediation (Agarwal and Liu, 2015; Sutton et al., 2011). The findings of this study indicate that pretreatment of soil contaminated with total TPH using chemical oxidants enhances biodegradation. In the control microcosm, the decrease in TPH concentration is attributed to the washing away of TPH components during water spraying onto the soil surface. Persulfate, known for its lack of heat production or precipitation during reaction, poses fewer risks to in situ microorganisms and has high solubility, leading to a wider radius of influence (ITRC, 2005; Yen et al., 2011). Ultrasound-assisted transmission of persulfate into the slurry results in a more even distribution of the persulfate solution within the contaminated soil, aiding desorption to some

extent. This facilitate easier contact between the generated sulfate radicals ($SO_4^{\bullet-}$) and the petroleum hydrocarbons in the slurry (Doraiswamy and Thompson, 1999). The feasibility of TPH removal using persulfate alone indicates that persulfate can be activated thermally and ultrasonically. The production of hydroxyl ($OH^{\bullet}$) radicals can promote the removal of organic compounds (Lebik-Elhadi et al., 2020). To identify the radicals contributing more to TPH degradation, two scavengers, methanol (MeOH) and tertiary butanol (TBA), are introduced into the system. Both alcohols are considered effective scavengers for radical quenching, although they react differently (Qi et al., 2015; Zhang et al., 2020).

4. Conclusion

The results of this study demonstrate that TPH contamination negatively impacts soil properties and plant growth factors in rice, including reductions in pH, organic matter, nutrient availability, root and shoot growth, chlorophyll content, plant height, grain yield, and biomass. Amendment of contaminated soil with SP effectively mitigates adverse effects by serving as a strong oxidizing agent that chemically degrades TPHs into less toxic compounds. It also provides oxygen to stimulate microbial degradation of remaining contaminants. By remediating the contamination, SP is able to restore soil properties, reduce toxicity, and improve plant growth conditions, though higher doses are required to counteract higher TPH levels. The findings suggest several important implications:

(1) TPH contamination poses major risks to soil health and crop productivity, remediation measures should be taken, especially in agricultural areas where food production is a priority. (2) SP can be an effective amendment for remediating TPH-contaminated soils, it provides chemical and biological means of degrading petroleum hydrocarbons to improve soil and plant conditions, higher doses may be required for higher contaminant concentrations, so SP levels would need to be optimized based on initial TPH levels. (3) SP may directly or indirectly influence plant growth through changes in hormone levels, membrane permeability, and other physiological factors, more researches are needed to determine the extent of these effects, especially for different plant species, SP not significantly impacts rice growth factors in uncontaminated soil. (4) Other factors like soil type, moisture, temperature, and microbial activity levels may also affect the efficiency of TPH remediation using SP, additional studies should explore how these variables interact to influence contaminant degradation and restoration of soil and plant health. (5) The byproducts of TPH and SP degradation may provide nutrient sources for plants and microbes, but they could also have residual toxic effects if SP treatment is insufficient, future research should aim to identify these byproducts and their potential impacts. (6) SP treatment may need to be combined with other remediation strategies, such as bioaugmentation or biostimulation of microbial communities, to maximize TPH degradation, integrated remediation approaches could speed up restoration of contaminated sites. (7) The effects of TPH contamination and SP treatment should be evaluated for other crops to determine how results may vary based on plant species and their interactions with soil properties and microbial communities.

In summary, these findings demonstrate the promise of SP for remediating TPH-contaminated agricultural soils to revitalize crop productivity. However, SP dosage and application must be tailored to site-specific conditions. Additional researches are still needed to optimize SP treatment strategies, understand impacts on different plant species, explore integrated remediation approaches, and evaluate the effects of byproducts and other variables that could influence the success of remediation efforts. With further work, SP has the potential to become an important tool for sustainable management of TPH-contaminated land.

CRediT authorship contribution statement

Farzad Rassaei: Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Software, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization.

Availability of data and materials

The datasets during and/or analyzed during the current study are available from the corresponding author upon reasonable request.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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