

# Toxicity Test of Zinc on Contaminated Soil by Petroleum Products (Fuel Oil) around Pertamina Rewulu-Cilacap Pipeline Bantul, Yogyakarta by Using Mung Bean (*Vigna radiata*) and Water Spinach (*Ipomoea aquatica*)

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Received: June 7, 2023

Accepted: April 26, 2024

DOI: 10.22146/ijc.85381

**Abstract:** Toxicity test of zinc on contaminated soil by petroleum products around the Pertamina Rewulu-Cilacap pipeline, Bantul, Yogyakarta by using mung bean (*Vigna radiata*) and water spinach (*Ipomoea aquatica*) have been studied. This study aims to investigate the physico-chemical properties, adsorption, and desorption of soil samples, and the effect of Zn toxicity on the growth of mung bean and water spinach. The physico-chemical properties were evaluated including moisture content, ash content, pH, conductivity, total organic carbon, cation exchange capacity, and heavy metal content. In addition, total petroleum hydrocarbon was analyzed by using GC-MS, and samples were characterized by using FTIR and XRD. The adsorption and desorption capacities of Zn were determined by an atomic absorption spectrophotometer (AAS) measurement. Toxicity test was conducted on the growth of mung bean and water spinach. This study showed that soil sample point III had the highest Zn content at 632.26 mg kg<sup>-1</sup>. Adsorption isotherms of zinc metal followed the Langmuir isotherm model and maximum adsorption occurred at 100 mg kg<sup>-1</sup>. Optimum desorption takes place at the concentration of citrate acid 0.7 mol L<sup>-1</sup> and at pH 3. Toxicity test results revealed that zinc metal at high concentrations was found to be toxic to seedling growth.

**Keywords:** petroleum; zinc; adsorption-desorption; toxicity test

## ■ INTRODUCTION

PT. Pertamina (Persero) Rewulu Fuel Oil Terminal is one of the business units that act as a distribution depot for premium, pertamax, pertalite, and solar types of fuel oil for the Yogyakarta region and parts of Central Java. In recent situations, pipelines are the most reliable and efficient means of transporting fuel oil. Fuel oil distribution through pipelines is relatively fast and the amount can be adjusted flexibly. The cost of operating and maintaining the pipeline is relatively low and the loss rate of fuel oil transported is also relatively small compared to shipping by conventional transportation methods. However, the gaps/weakness of distribution through the pipelines system allowed the risk of leakage due to natural disasters, work accidents, or irresponsible human activities. Several cases of fuel distribution pipeline leaks in Indonesia have occurred, such as what happened at the

PT. Pertamina (Persero) Rewulu Fuel Oil Terminal. During the process of exploration, exploitation, production, transportation, storage, etc., the leakage brings the impact of contamination to the soil [1].

Considering the Law of the Republic of Indonesia number 32 year 2009 about Environmental Protection and Management as well as Government Regulation Number 101 year 2014 regarding the management of hazardous and toxic waste (B3), the problem of soil petroleum-contaminated soil has become a concern in recent years because B3 waste containing high concentrations of hydrocarbons and heavy metals which are toxic resulting in negative effects on water and soil ecosystems, human health, and other natural resources [2]. Physically, heavy metals are not biodegradable and could be persistent for a long time in environmental components such as soil [3].

Zn is one of the essential metals contained in waste fuel oil. Zn metal is needed as a micronutrient that is essential for the human body and other living things. It also plays an important role as an enzyme activator for humans. Zn is also a vital metal for plant growth and is actively absorbed through the roots. The permissible concentration of Zn in plants is around 5–20 mg kg<sup>-1</sup>, therefore, higher Zn concentration is classified as a carcinogen. Although Zn is an essential metal, it can cause death at high concentrations, inhibit growth, and cause reproductive disorders in plants [4].

Indications of heavy metal pollution are not only concentrated in the soil but also in the roots, stems, leaves and even seeds and fruits, so it will be very dangerous if consumed by humans. The period of germination, shoots and roots will decrease with the increasing of metal content in the soil [5]. Research related to Cr toxicity test from polluted soil around the Yogyakarta tannery industry using *Brassica juncea* and *Heliantus anuus* has been conducted by Suherman et al. [6], which showed that chromium has a toxic effect on *B. juncea* and *H. anuus* growth (roots and hypocotyl) in plants decreases with increasing chromium concentration in eluates and synthetic solutions. The soil nature reduces the effect of metal habituation on the growth of *Lepidium sativum*, particularly at low concentrations compared to synthetic solutions. The success of germination depends on various external factors. This indicated that the seed must be supported by internal sensors that possibly send environmental signals into cellular mechanisms that lead to germination [7-9].

This study focused on the toxicity effect of Zn from contaminated soil around the Pertamina pipeline route on seed germination, especially for the growth of mung beans (*Vigna radiata*) and water spinach (*Ipomoea aquatica*). Studies of soil characteristics and adsorption are needed to determine the relationship between soil characteristics and the adsorption capability of contaminated soil. In addition, desorption studies are conducted to investigate the possible distribution of metals in soil. Finally, Zn metal toxicity tests are required to assess the effect of metals on plant growth.

## ■ EXPERIMENTAL SECTION

### Materials

Soil samples were taken from four different sampling points (SP) (20–40 cm depth) based on the range location from the polluted soil point sources around Pertamina Rewulu-Cilacap pipeline Bantul, Yogyakarta. The materials used by pro-analysis qualities from Merck were calcium chloride dihydrate (CaCl<sub>2</sub>·2H<sub>2</sub>O), potassium chloride (KCl), barium chloride dihydrate (BaCl<sub>2</sub>·2H<sub>2</sub>O), magnesium sulphate heptahydrate (MgSO<sub>4</sub>·7H<sub>2</sub>O), hydrofluoric acid (HF) 40%, nitric acid (HNO<sub>3</sub>) 65%, and hydrochloride acid (HCl) 37%. While magnesium standard solution at 1,000 mg L<sup>-1</sup> and zinc standard solution at 1,000 mg L<sup>-1</sup> were from Sigma-Aldrich, and Whatman 42 filter paper was from Cytiva.

### Instrumentation

The apparatuses in this research are analytical balance (Mettler Toledo AB54-S), digital balance sheet (ACIS AD 300i), shaker (Marius Instrumenten), Muffle furnace (Fischer model 184), and oven (Kirin). The instrumentation used are an X-ray diffractometer (XRD, Shimadzu model XRD-6000), atomic absorption spectrophotometer (AAS, Perkin Elmer 3110), Fourier transform infrared spectrophotometer (FTIR, Thermo Nicolet iS10), gas chromatography-mass spectrometry (GC-MS, Shimadzu), and hot plate-stirrer (Thermolyne Cimarec 1).

### Procedure

#### **Physico-chemical properties evaluation of the soil samples**

For the physico-chemical assessment of the soil samples, the procedures were started from the drying step of the 10–15 g samples at 105 °C for 24 h. The water content of soil samples was determined by the ISO 11465 (1993). Ash content of soil samples by DIN EN 14775: 2004. pH of the soil samples was measured using the ISO 10390 method. Furthermore, organic carbon content was evaluated based on ASTM D 2974. Cation exchange capacity (CEC) is determined based on DIN ISO 11260.

The electrical conductivity was measured by DIN CEN/TS 15937. Samples characterization by FTIR was conducted to determine functional groups, while the X-ray diffractometer instrument was used to study the mineral content in soil samples. Data retrieval is repeated three times for standard deviation consideration.

#### **Determination of total petroleum hydrocarbons (TPH)**

As much as 10 g of soil samples were extracted with 10 mL of *n*-pentane. After the addition of *n*-pentane, the sample is cornered for 15 min using an ultrasonic device and allowed to settle for 30–60 min at room temperature. Two additional extractions, each with 20 mL of *n*-pentane, were performed in the same way. The 1 mL of organic extract was then transferred into vials and analyzed with GC-MS.

#### **Metal determination**

Metal content is measured based on the method from British Standards European Norm (BS-EN) 13656 (2020) where 1 g of soil sample was extracted with the addition of 4 mL of 40% HF solution and was shaken for 24 h. Then, the mixture was added with 12 mL of aqua regia and again 24 h shaking process. The filtrate is separated, which is then calculated for metal level with AAS. Data retrieval is repeated three times to consider the standard deviation.

#### **Adsorption studies with concentration variations**

Soil sample (1 g) was put into a polyethylene vial, and 15 mL of Zn(II) solution was added with concentration variations of 5, 10, 25, 50, 75, 100, 150, and 200 mol L<sup>-1</sup>. This mixture is shaken for 24 h and then separated by filtration. The remaining Zn(II) concentration in the filtrate is measured by AAS instruments. Data collection is repeated three times to calculate the standard deviation. The study was conducted at a room temperature (around 27 °C) and a chamber pressure of ±1 atm.

#### **Desorption study with variation in citric acid concentration**

A study on the optimum Zn desorption with variations in citric acid concentration was initiated with 15 mL of citric acid solution with 0.0, 0.1, 0.3, 0.5, 0.7, and 0.9 mol L<sup>-1</sup> concentrations which were added to the 1 g of soil sample. The mixture was shaken for 24 h and then

centrifuged at 5000 rpm for 15 min. The supernatant was separated, and the remaining Zn(II) concentration in the supernatant was measured with the AAS instrument. Repetition in data collection is carried out three times to calculate the standard deviation.

#### **Determination of optimum pH**

pH soil sample solution was evaluated by the addition of 15 mL of Millipore water in a polyethylene bottle. Furthermore, the pH of the solution is adjusted to pH variations of 3, 5, 7, and 9. The mixture is shaken for 24 h and then centrifuged at 5000 rpm speed for 15 min. The supernatant is separated, and the remaining Zn(II) concentration in the supernatants is evaluated with the AAS instrument. Repetition is carried out three times to calculate the standard deviation.

## ■ RESULTS AND DISCUSSION

The soil samples evaluated in this study were from around the Pertamina Cilacap-Rewulu pipeline, Bantul-Yogyakarta, as shown in Fig. 1. Soil sampling is carried out in the topsoil layer which is the topmost soil layer with a depth of 5–30 cm from the earth's surface. Four soil SP are achieved including point I, which is unpolluted soil. The study of physico-chemical properties of soil includes several parameters such as water content, pH, ash level, electrical conductivity, total organic carbon, CEC, and heavy metal concentration.

The results of the characterization of physico-chemical properties in soil samples are presented in Table 1. It is known that the water content in the four



**Fig 1.** Map of soil sampling location

**Table 1.** Physico-chemical properties and metal content of the soil samples around Pertamina Rewulu-Cilacap pipeline

Parameters	Sampling point I	Sampling point II	Sampling point III	Sampling point IV
Water content (%)				
a. 105 °C	29.95	30.07	33.11	31.92
b. Room temperature drying	24.89	23.58	26.16	25.40
pH				
a. H <sub>2</sub> O	6.94	7.65	7.86	7.80
b. KCl	6.23	7.07	7.06	7.00
c. CaCl <sub>2</sub>	6.64	7.24	7.21	7.17
Ash content (%)	87.35	90.08	91.64	91.21
Electrical conductivity ( $\mu\text{S cm}^{-1}$ )	146.30	197.30	371.00	354.70
Total organic carbon ( $\text{mg g}^{-1}$ )	42.18	45.45	57.06	56.14
Cation exchange capacity ( $\text{meq}/100 \text{ g}$ )	0.78	0.98	1.08	1.06
Zn total ( $\text{mg kg}^{-1}$ )	163.70	519.37	632.26	620.54

samples is different, and the highest water level is in point III soil samples. The difference in water content is caused by the ability of the soil to store different water. The ability of soil to store water is closely related to the surface area of the land. The greater the land surface area, the greater the water storage capacity of the soil. In addition, soil pH evaluation at the medium of H<sub>2</sub>O, KCl, and CaCl<sub>2</sub> solution is used to obtain a pH that is close to natural conditions and pH values caused by the existence of other cations.

The use of CaCl<sub>2</sub> 0.01 and KCl 1.00 mol L<sup>-1</sup> salt solution is due to these two cations having the highest cation content in soil solution [10]. The presence of K<sup>+</sup> and Ca<sup>2+</sup> cations in salt solutions added to the soil suspension will cause more H<sup>+</sup> ions to be released in the solution so that the pH value decreases. This is possibly because the release of cations will replace H<sup>+</sup> ions in the soil so that H<sup>+</sup> ions will be pushed into the soil solution, which causes the soil concentration to be close to the concentration in its natural conditions. This statement is in accordance with the pH results at the four SP where the pH value with the H<sub>2</sub>O extractor is higher than that of the KCl and CaCl<sub>2</sub> extractors. In Table 1 (IV), the order of pH values of the four sample points from high to low are SP III > IV > II > I. pH values will determine the adsorption and desorption processes as well as the chemical speciation of heavy metals in the soil.

Ash content in all four samples as shown in Table 1, seems to have variable values. Of the four samples, SP III

had the highest ash content of 91.64%. Ash level indicates the amount of mineral content and other inorganic matter in the soil, including heavy metals. Measured ash content is an inorganic material that cannot be burned during the ashing process so it remains in the form of ash. By the ashing process, all organic compounds may have evaporated due to the presence of heat energy that is able to destruct soil samples and evaporate organic compounds in them. The residual inorganic materials in the ashing process are used to bond Zn metal in the soil when organic matter is low.

Soil could deliver electricity through metal ions dissolved in the soil. Electrical conductivity by the soil occurs because of the free salt content contained in soil water and exchangeable ion content on the surface of soil solid particles [11]. Water in soil can also solve ions that can be exchanged with other ions. Therefore, the electrical conductivity will increase as the salt concentration in the solution increases. This relationship is closely related to the results of the study in Table 1. where the moisture content value is commensurate to the electrical conductivity value in the sample, and the highest electrical conductivity value is at the SP III, followed by SP IV and then further down in SP II and I.

Total organic carbon indicates the amount of carbon atoms in an organic compound. The increase in

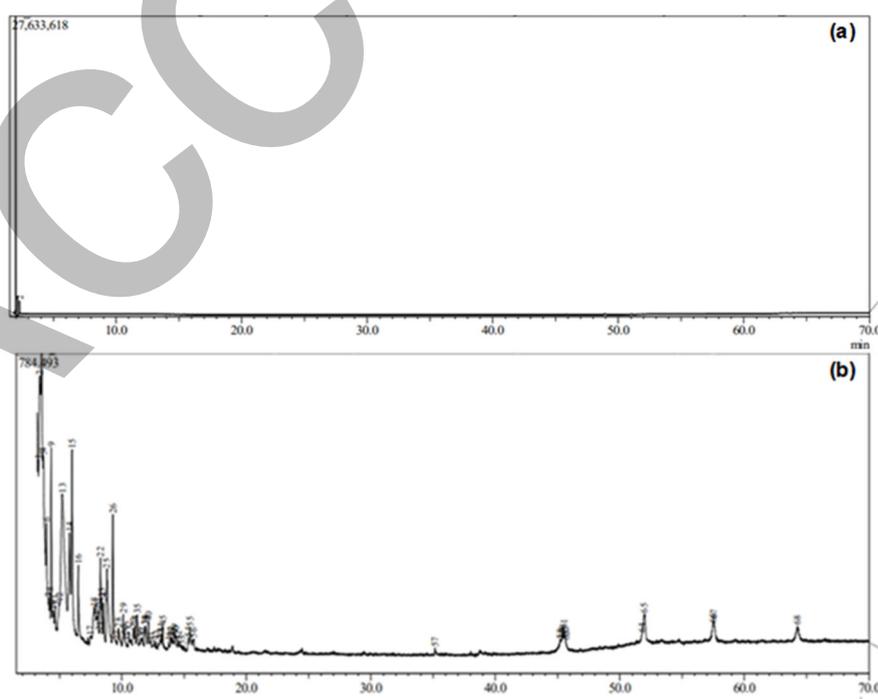
soil organic carbon is caused by soil organic matter that decomposes to produce organic acids, so organic matter is added to the soil, which can increase soil organic carbon levels. Organic matter itself will increase the ability of the soil to hold water so that the soil water content will be higher [12]. Soil organic matter also has functional groups that can contribute a negative charge to the soil. Negative charges can exchange cations in the soil so that the CEC also increases [13]. This is shown in Table 1 that the total organic carbon content in the sample is closely proportional to the moisture content and cation exchange capacity. Organic carbon levels in SP III had the highest value of  $57.06 \text{ mg g}^{-1}$ , followed by SP IV, II and I.

High levels of organic carbon in the soil indicate that more soil organic matter results in a high dominance of negative charges in the soil so that it is proportional to the number of cations that can be bound by the soil. This is in accordance with the results obtained in Table 1, whereas the CEC value is proportional to the TOC value. The CEC value is highest in SP III, while SP I has the lowest value because it is far from polluting sources. The more negative charge, the more cations or metals can be exchanged, so the high CEC will increase the metal content in the soil.

Determination of metal levels in the soil is very important because the presence of metals will affect metal

desorption and metal toxicity in the soil. In Table 1 some metals analyzed from SP III have higher metal content when compared to SP I, II and IV. Soil samples at SP III Zn metal content were higher at  $632.26 \text{ mg kg}^{-1}$  followed by samples at SP IV at  $620.54 \text{ mg kg}^{-1}$ , then SP III samples at  $519.37 \text{ mg kg}^{-1}$  and SP I at  $163.70 \text{ mg kg}^{-1}$ .

The GC-MS chromatogram results, as shown in Fig. 2. Fig. 2(a) belongs to SP I, which is an unpolluted soil sample, and no hydrocarbon results are obtained, while SP II, III and IV samples in Fig. 2(b-d) that have been identified based on several peaks of the area (%) are the highest obtained hydrocarbon compounds that are dominant in soil samples, namely  $\text{C}_8\text{H}_{10}$  and  $\text{C}_9\text{H}_{12}$  with the highest concentrations found in SP III samples with 39% of  $\text{C}_8\text{H}_{10}$  and for  $\text{C}_9\text{H}_{12}$  was 12.44%, then SP IV, namely  $\text{C}_8\text{H}_{10}$  by 31.16%,  $\text{C}_9\text{H}_{12}$  by 12.06% and followed by SP II, namely  $\text{C}_8\text{H}_{10}$  by 27.56%,  $\text{C}_9\text{H}_{12}$  by 9.52%. Hydrocarbon compounds are volatile organic compounds, and it is clear from the results obtained that the average hydrocarbon compounds detected at a low retention time of 5.20–9.27 min. These volatile organic compounds cannot be removed from soil structure because of their relatively high hydrophilic properties, and they usually move several kilometers from the pollutant supply [14].



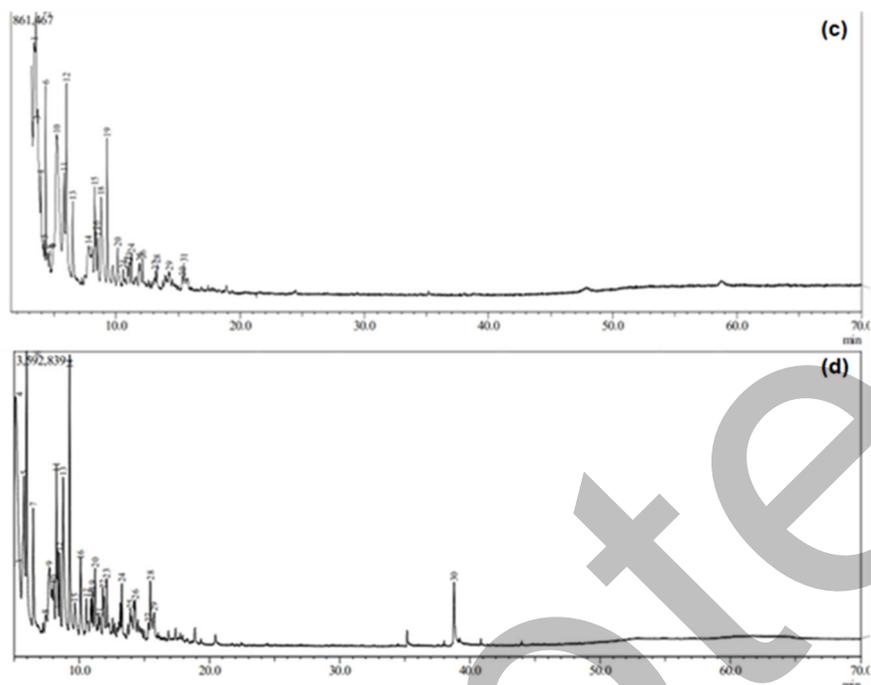


Fig 2. Chromatogram of the sampling point (a) I, (b) II, (c) III, and (d) IV

FTIR spectra from the four samples in Fig. 3 showed that all samples have functional groups at relatively the same wavenumber. The wavenumber at SP I, II, III and IV of  $3449\text{ cm}^{-1}$  related to the absorption region for O–H from Si–OH and Al–OH, reinforced by the presence of wavenumbers at  $1034\text{ cm}^{-1}$  from all four samples showing absorption for Si–O and Al–O extended vibrations. There is also a typical absorption of hydrocarbons, namely at wavenumbers  $2932\text{--}2924\text{ cm}^{-1}$ , which shows the extended vibrational functional group C–H. SP III and IV at wavenumbers  $1427\text{ cm}^{-1}$  showed C–H bend vibrations. The absorption band at wavelength  $1636\text{ cm}^{-1}$  at SP I, II, III and IV is the bending vibrational absorption region –OH of  $\text{H}_2\text{O}$ . In SP I and II, there were absorptions at the wavenumbers of  $756$  and  $748\text{ cm}^{-1}$ , indicating absorption regions for Si–O extended vibrations derived from quartz and silica [15–16]. In addition, in SP I, there was absorption at wavenumber  $532\text{ cm}^{-1}$  and SP II, III, IV absorption at wavenumber  $540\text{ cm}^{-1}$ , which showed Si–O–Al bending vibration [17]. Bending vibrations of Si–O and Si–O–Si are shown at wavenumber  $471$  and  $463\text{ cm}^{-1}$  in SP I and II while at wavenumber  $478\text{ cm}^{-1}$  in SP III and IV. The results of the analysis on the four samples showed the content of essential groups that function as metal binders

in the soil and from these vibrations, it can be concluded that metal ions interact with the soil surface not only via silanol groups but also siloxane groups.

XRD analysis was carried out to determine the type of constituent minerals found in soil samples around the Pertamina Rewulu-Cilacap pipeline. Based on Fig. 4, the minerals contained in SP I are albite, kaolinite, quartz, quartz, and calcite at  $2\theta$ :  $21.81$ ,  $23.51$ ,  $27.62$ ,  $27.87$  and  $29.66^\circ$ . SP II has the content of kaolinite, albite, mullite,

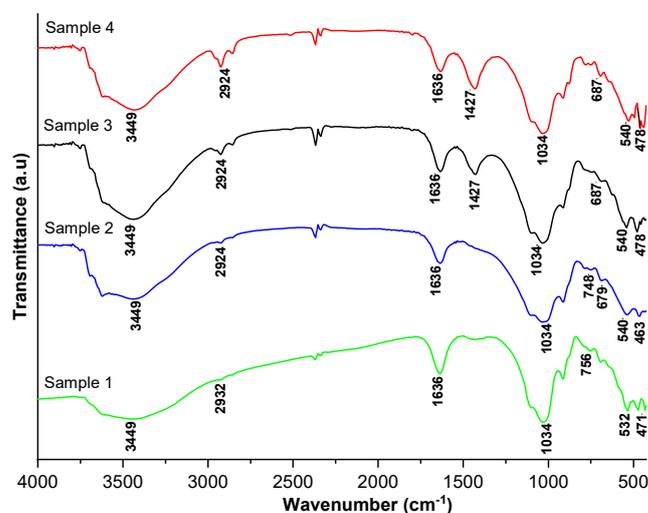


Fig 3. FTIR spectra of soil samples

quartz, and quartz at  $2\theta$ : 19.78, 21.90, 24.99, 27.74 and 27.99°. SP III has mineral contents of albite, kaolinite, quartz, calcite, and montmorillonite at  $2\theta$  of 21.79, 23.55, 27.64, 29.31, and 30.22°. SP IV had mineral content, namely albite, kaolinite, quartz, calcite, and montmorillonite at  $2\theta$ : 21.86, 23.60, 27.86, 29.35, and 30.82°. All four samples had similar clay minerals. Clay minerals are one of the soil mineral contents that affect the presence of metals and the ability of the soil to adsorb and desorb metals in the soil.

The adsorption capacity of soil samples to Zn was studied by observing the relationship of variations in Zn solution concentration in each soil sample to determine the ability of each soil sample to absorb Zn metal in the soil. The adsorption study of concentration variations was carried out by making variations in the concentration of standard Zn(II) solution as adsorbate and soil samples as adsorbents. The adsorption results of the four soil samples are shown in Fig. 5. It shows that the higher the concentration of Zn solution, the more Zn is adsorbed, but after maximum adsorption is reached, it will decrease to a concentration of 150 mg L<sup>-1</sup>. Zn adsorption in SP I, II, III and IV reached optimum at a standard solution concentration of Zn 100 mg L<sup>-1</sup>. Zn adsorbed at maximum state varies. Each amounted to 591.846, 780.459, 926.429, and 904.171 mg kg<sup>-1</sup>. At high enough concentrations, adsorbate has occupied all available active sites in the soil so that the adsorption process will terminate [18]. The decrease in adsorption capacity after reaching maximum conditions is due to the number of active sites used to bind to Zn that have been fully filled or have reached equilibrium.

Table 2 shows that the Freundlich isotherm model is a suitable adsorption isotherm in the Zn adsorption process because it has a high R<sup>2</sup> value close to the value

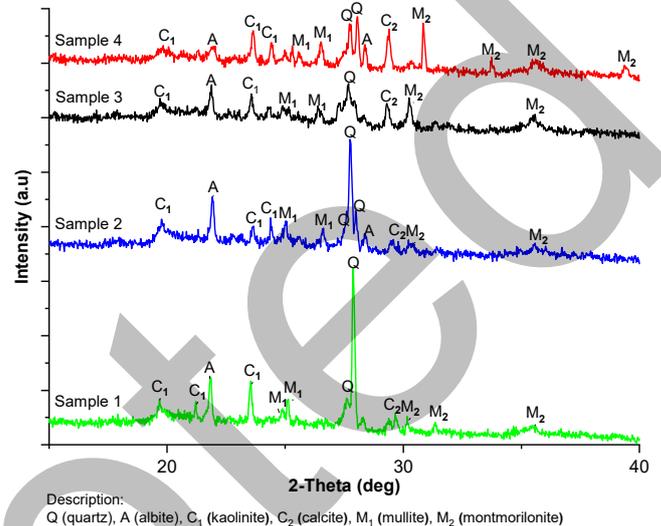


Fig 4. XRD spectra of soil samples

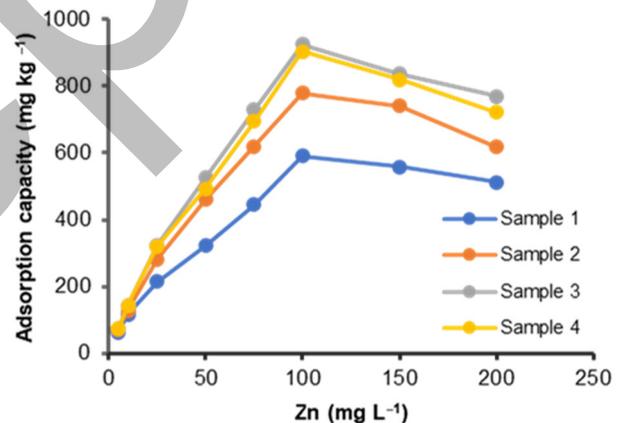


Fig 5. Zn concentration's effect on the adsorption rate of soil samples

**Table 2.** Isothermic parameters for adsorption of Zn metal on soil around the Pertamina Rewulu-Cilacap pipeline

Parameter	Sampling point I	Sampling point II	Sampling point III	Sampling point IV
	Isotherm Langmuir			
R <sup>2</sup>	0.8770	0.9325	0.9288	0.9064
q <sub>max</sub> (mmol kg <sup>-1</sup> )	0.0096	0.0013	0.0139	0.0139
K <sub>L</sub> (L mol <sup>-1</sup> )	5,347.8000	71,428.6000	13,513.5000	12,820.5000
E (kJ mol <sup>-1</sup> )	20.8900	27.8800	23.7200	23.5900
Isotherm Freundlich				
R <sup>2</sup>	0.9889	0.9933	0.9966	0.9912
n	2.0600	2.1400	2.4400	2.6400
K <sub>f</sub>	71.0700	119.3700	192.7700	196.3500

of 1, which is 0.9889, 0.9933, 0.9966, and 0.9912. The Freundlich isotherm equation model shows that adsorption takes place on heterogeneous surfaces and active sites with different adsorption energies [19]. This is in accordance with the results of soil characterization by using FTIR, in which two kinds of active sites, namely silica and alumina, appeared so that it is possible to unevenly distribute active sites on the soil surface.

Unlike Langmuir isotherms, the energy in the Freundlich model is different on each surface. The value of "n" in the Freundlich equation in all four samples is 2.06, 2.14, 2.44 and 2.64. The  $n > 1$  situation occurred due to the distribution of surface sites or a factor that causes a decrease in adsorbent-adsorbate interaction with an increase in surface density and n values in the range of 1–10, indicating that adsorption takes place well and through favorable physical interaction [20].

Desorption capacity is carried out with the aim of determining the release of Zn metal from soil samples using citric acid of various concentrations. The result of Zn metal desorption using citric acid is shown in Fig. 5. Fig. 6 shows that the higher the concentration of citric acid solution as the solution medium, the higher the Zn ions will be desorbed to their maximum point. The desorption results of sample points I, II, III, and IV reached the optimum point at a citric acid concentration of  $0.70 \text{ mol L}^{-1}$  with a desorbed Zn metal concentration of 159.52, respectively; 493.78, 625.07, and 602.64  $\text{mg kg}^{-1}$ . This is in accordance with the statement that low molecular weight organic acids (LMWOA), with more and more groups, have a higher ability to absorb heavy metals [21]. The higher the concentration of citric acid solution, the more citric acid molecules are in the solution so more Zn metal will interact with citric acid. This causes the release of Zn metal from the soil sample. Soil that interacts with citric acid causes  $\text{H}^+$  ions from citric acid to replace Zn metal in the form of  $\text{Zn}^{2+}$  in the soil, resulting in ion exchange. After that,  $\text{Zn}^{2+}$  ions that have been released into the citric acid solution will form a stable complex with citric ions.

Based on Zn ion speciation at various pH, it was revealed that the pH value is a key parameter that affects metal ion speciation [22]. The solvent used is water to

determine the optimum pH. The use of  $\text{H}_2\text{O}$  aims to make more and more  $\text{H}^+$  ions decomposed in solution. pH has an important role in the binding of metals in the soil. Fig. 7 shows that desorption takes place more optimally in acidic conditions, namely at pH 3. At low pH, more Zn metal will be desorbed because in the acidic state, Zn metal is in cationic form, so it is easy to leach. Increasing soil pH will decrease the positive charge in the soil and reduce the desorption of Zn, which will cause the hydrolysis of metal cations to increase.

Toxicity test on mung beans (*V. radiata*) and water spinach (*I. aquatica*) using an eluates solution containing desorbed metals at a variation of pH 7 (neutral). In toxicity tests with *V. radiata*, as seen in Fig. 8(a) and 8(b), root and hypocotyl length are influenced by the presence of Zn metal in the desorption eluate, and

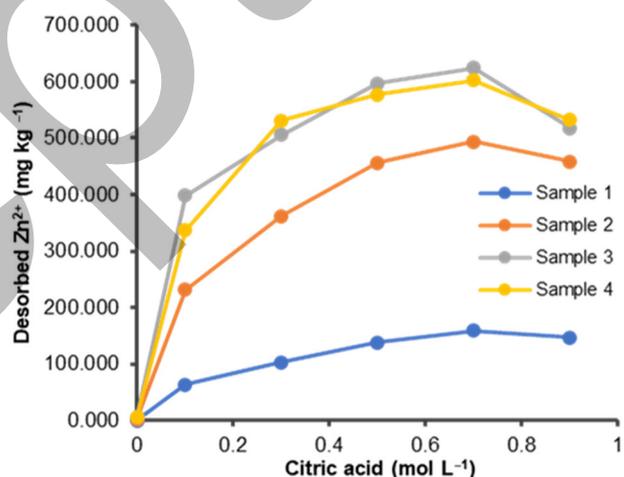


Fig 6. Effect of citric acid concentration on soil sample desorption

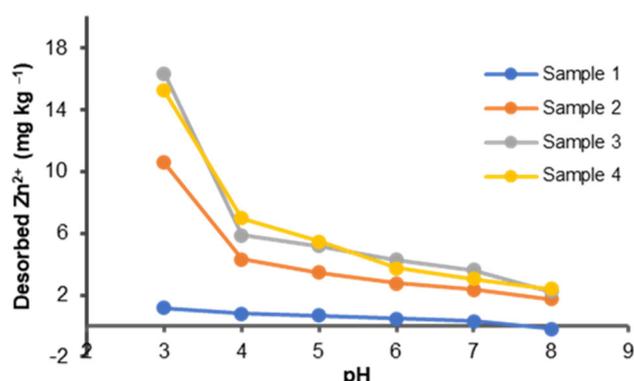


Fig 7. pH variation effect on the Zn desorption from the soil samples

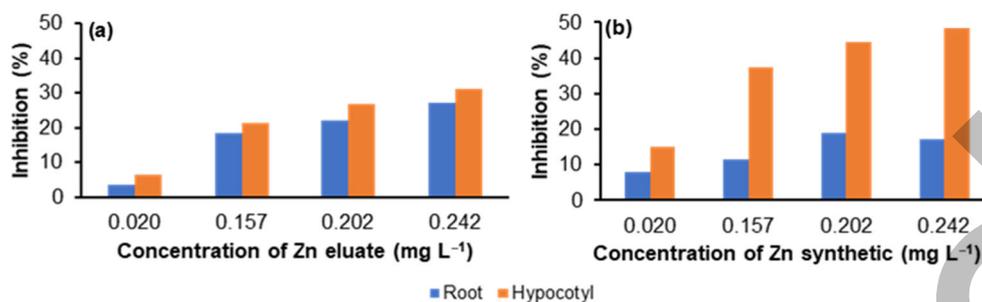


Fig 8. Toxicity evaluation of (a) Zn eluates and (b) Zn synthetic solution to the *V. radiata* (at pH neutral)

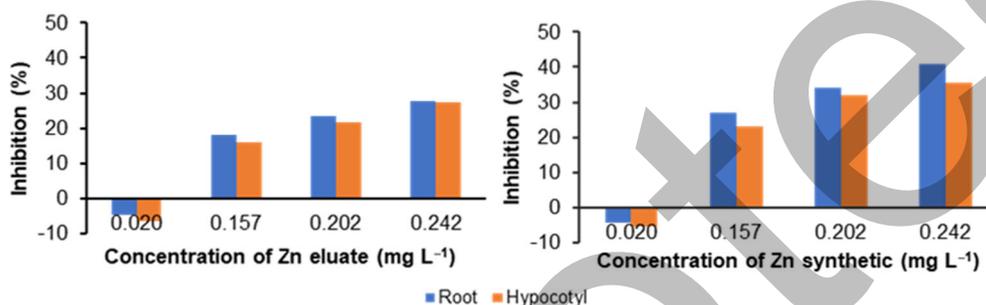


Fig 9. Toxicity evaluation of (a) Zn eluates and (b) Zn synthetic solution to the *I. aquatica* (at pH neutral)

there is an immediate relationship between the level of inhibition of the growth response of *V. radiata* with increasing concentrations of Zn metal, so the hypocotyl from *V. radiata* gives a sensitive response against metal Zn when compared to the root. Toxicity test results on water spinach (*I. aquatica*), as shown in Fig. 9(a) and 9(b) in both eluate solutions and standard solutions Zn 0.020 mg L<sup>-1</sup> show negative inhibition values. A negative inhibition value indicates the lower metal toxicity at that concentration, even in this concentration range, metal ions support the growth of *I. aquatica*. These phenomena are possibly due to eluate solution containing dissolved organic matter that can support the growth of water spinach seeds [18], and the toxicity test results show a sensitive response to root compared to hypocotyl. This phenomenon due to Zn toxicity inhibits root cell elongation or cell cycle extension so that it will inhibit root growth, while roots that are in direct contact with the medium containing metal ions are affected by root shortening and inability to adsorb water from the medium [23].

The results of toxicity tests on *V. radiata* and *I. aquatica* showed that the low Zn concentrations in eluates do not inhibit root growth and hypocotyl in seedlings of *V. radiata* and *I. aquatica*, so it is proven that the higher

the Zn concentration, the higher the toxic effect on root growth and hypocotyl and vice versa. In other words, the severity of the plant growth response is influenced by the high concentration of Zn metal, so Zn metal has a toxic effect on root growth and hypocotyl from *V. radiata* and *I. aquatica* seeds.

## CONCLUSION

The petroleum-contaminated soil containing zinc and hydrocarbon concentration corresponded to the physico-chemical properties of soil samples. SP II, III, and IV are the most polluted soil sampling locations compared to samples at SP I, which are unpolluted soil samples. It is also known that maximum adsorption occurred at a concentration of 100 mg kg<sup>-1</sup> and the adsorption isotherms of Zn into soil samples followed the Freundlich isotherm model. Furthermore, the toxicity test exhibited that Zn has a toxic effect on the growth of *V. radiata* and *I. aquatica*. In addition, the Zn ions seem to be more toxic to *V. radiata* than *I. aquatica*.

## ACKNOWLEDGMENTS

The author thanks the Ministry of Health of the Republic Indonesia for the assistance with education costs through the DPTK (*Direktorat Penyediaan Tenaga*

*Kesehatan*) scholarship with Decree Number KH. 01.07/V/18753/2021. Also, thank you to Universitas Gadjah Mada for supporting these activities through the final project recognition (RTA) research scheme 2023 with contract number 5075/UN1.P.II/Dit-Lit/PT.01.01/2023.

#### ■ CONFLICT OF INTEREST

We declare there are no conflicts of interest regarding the publication of this manuscript.

#### ■ AUTHOR CONTRIBUTIONS

Linda Johana Latumahina served as a researcher, data collector, and drafting the manuscript. Yosua Tanzil served for writing the report and conducting revisions. Suyanta as a research co-supervisor, checks revision results, provides suggestions for improvements. Suherman served as a research's supervisor, finalize the manuscript, and prepare the final report.

#### ■ REFERENCES

- [1] Ajona, M., and Vasanthi, P., 2021, Bioremediation of petroleum contaminated soils – A review, *Mater. Today: Proc.*, 45, 7117–7122.
- [2] Yuniati, M.D., 2018, Bioremediation of petroleum-contaminated soil: A review, *IOP Conf. Ser.: Earth Environ. Sci.*, 118 (1), 012063.
- [3] Briffa, J., Sinagra, E., and Blundell, R., 2020, Heavy metal pollution in the environment and their toxicological effects on humans, *Heliyon*, 6 (9), e04691.
- [4] Khan, M.U., Muhammad, S., Malik, R.N., Khan, S.A., and Tariq, M., 2016, Heavy metals potential health risk assessment through consumption of wastewater irrigated wild plants: A case study, *Hum. Ecol. Risk Assess.: Int. J.*, 22 (1), 141–152.
- [5] Singh, D., Sharma, N.L., Singh, C.K., Yerramilli, V., Narayan, R., Sarkar, S.K., and Singh, I., 2021, Chromium(VI)-induced alterations in physico-chemical parameters, yield, and yield characteristics in two cultivars of Mungbean (*Vigna radiata* L.), *Front. Plant Sci.*, 12, 735129.
- [6] Suherman, S., Akmarina, W., Sutiawan, W., Aprilita, N.H., Mudasir, M., and Morita, K., 2019, Toxicity test of desorbed chromium from polluted soil around tannery industry in Yogyakarta-Indonesia by using *Brassica juncea* and *Helianthus annuus* L., *Molekul*, 14 (2), 157–164.
- [7] Lyubenova, M., Dineva, S., Cala, K., Dinich, B., and Boteva, S., 2019, Ecotoxicity of purified industrial wastewater, *Environ. Ecol. Res.*, 7 (4), 209–219.
- [8] Smolinska, B., and Rowe, S., 2015, The potential of *Lepidium sativum* L. for phytoextraction of Hg-contaminated soil assisted by thiosulphate, *J. Soils Sediments*, 15 (2), 393–400.
- [9] Bożym, M., Król, A., and Mizerna, K., 2021, Leachate and contact test with *Lepidium sativum* L. to assess the phytotoxicity of waste, *Int. J. Environ. Sci. Technol.*, 18 (7), 1975–1990.
- [10] Kome, G.K., Enang, R.K., Yerima, B.P.K., and Lontsi, M.G.R., 2018, Models relating soil pH measurements in H<sub>2</sub>O, KCl and CaCl<sub>2</sub> for volcanic ash soils of Cameroon, *Geoderma Reg.*, 14, e00185.
- [11] Astari, S.A., Mudasir, M., and Suherman, S., 2024, Soil toxicity around the textile industry in Bantul using spinach seeds (*Amaranthus gangeticus*) and bean sprouts (*Phaseolus aureus*), *Molekul*, 19 (1), 1–7.
- [12] Libohova, Z., Seybold, C., Wysocki, D., Wills, S., Schoeneberger, P., Williams, C., Lindbo, D., Stott, D., and Owens, P.R., 2018, Reevaluating the effects of soil organic matter and other properties on available water-holding capacity using the National Cooperative Soil Survey Characterization Database, *J. Soil Water Conserv.*, 73 (4), 411–421.
- [13] Singh Yadav, S.P., Bhandari, S., Bhatta, D., Poudel, A., Bhattarai, S., Yadav, P., Ghimire, N., Paudel, P., Paudel, P., Shrestha, J., and Oli, B., 2023, Biochar application: A sustainable approach to improve soil health, *J. Agric. Food Res.*, 11, 100498.
- [14] Muir, B., Sobczyk, M., and Bajda, T., 2021, Fundamental features of mesoporous functional materials influencing the efficiency of removal of VOCs from aqueous systems: A review, *Sci. Total Environ.*, 784, 147121.
- [15] Katti, K.S., Sikdar, D., Katti, D.R., Ghosh, P., and Verma, D., 2006, Molecular interactions in intercalated organically modified clay and clay-

- polycaprolactam nanocomposites: Experiments and modeling, *Polymer*, 47 (1), 403–414.
- [16] Amarasinghe, P.M., Katti, K.S., and Katti, D.R., 2008, Molecular hydraulic properties of montmorillonite: A polarized fourier transform infrared spectroscopic study, *Appl. Spectrosc.*, 62 (12), 1303–1313.
- [17] Hospodarova, V., Singovszka, E., and Stevulova, N., 2018, Characterization of cellulosic fibers by FTIR spectroscopy for their further implementation to building materials, *Am. J. Anal. Chem.*, 9 (6), 303–310.
- [18] Suherman, S., Schmidt, C., Kolb, M., Zachmann, D., and Bahadir, M., 2013, Partitioning of copper and lead between solid and dissolved organic matter in a humus-rich soil of the Harz Mountains (Germany) and ecotoxicity test with *Lepidium sativum*, *Fresenius Environ. Bull.*, 22 (2), 318–327.
- [19] Lv, N., Wang, X., Peng, S., Zhang, H., and Luo, L., 2018, Study of the kinetics and equilibrium of the adsorption of oils onto hydrophobic jute fiber modified via the sol-gel method, *Int. J. Environ. Res. Public Health*, 15 (5), 969.
- [20] Desta, M.B., 2013, Batch sorption experiments: Langmuir and Freundlich isotherm studies for the adsorption of textile metal ions onto teff straw (*Eragrostis tef*) agricultural waste, *J. Thermodyn.*, 2013 (1), 375830.
- [21] Jing, Y.D., He, Z.L., and Yang, X.E., 2007, Effects of pH, organic acids, and competitive cations on mercury desorption in soils, *Chemosphere*, 69 (10), 1662–1669.
- [22] Huang, J., Yuan, F., Zeng, G., Li, X., Gu, Y., Shi, L., Liu, W., and Shi, Y., 2017, Influence of pH on heavy metal speciation and removal from wastewater using micellar-enhanced ultrafiltration, *Chemosphere*, 137, 199–206.
- [23] Pang, Y.L., Quek, Y.Y., Lim, S., and Shuit, S.H., 2023, Review on phytoremediation potential of floating aquatic plants for heavy metals: A promising approach, *Sustainability*, 15 (2), 1290.