

# SOIL POLLUTION EFFECTS ON THE GERMINATION, GROWTH, ROOT GROWTH AND BIOMASS OF *Emilia praetermisa* (Milne-Redh) Asteraceae WITH COW DUNG ENRICHMENT

N.H. Ifediora\*, C.O. Onyebuchi and E.N. Otenyi

Department of Plant Science and Biotechnology, Michael Okpara University of Agriculture, Umudike, Nigeria

\*Corresponding Author Email Address: [ifedioranonyelum@gmail.com](mailto:ifedioranonyelum@gmail.com)

Phone: +2348064328449

## ABSTRACT

The aim of this study was soil pollution effects on the germination, growth, root growth and biomass of *Emilia praetermisa* with cow dung enrichment. Crude oil was varied in 1 %, 3 % and 5 % to the soil while the cow dung levels were constant to amount of soil needed. By 12 weeks, the plants were harvested. Initial heavy metal properties of soil, cow dung and crude oil were analyzed for heavy metal. Growth parameters data were collected, was subjected to descriptive statistics to obtain the means and standard deviations. Analysis of variance analysis was used to compare the difference in the heavy metal properties of soil, cow dung and Crude oil, means of growth data and laboratory analysis. The result showed that the heavy metal in soil, cow dung and crude oil varied significantly ( $P < 0.05$ ). Higher levels of crude oil contaminated soil inhibited the growth parameter of the plant generally. Also, the heavy metal levels in the soil and plant samples was low and mercury, non-existing. Lastly, low levels of heavy metals in the soil and in the plants, tissue indicates the phytoremediation potentials of *E. praetermisa*.

**Keywords:** Cow dung, Crude oil, *Emilia praetermisa*, Growth parameters, Properties, Soil.

## INTRODUCTION

One of the century's major global environmental problems is environmental contamination from crude and other refined petroleum products, which arises from rising oil product demand (Sojini and Ejeromedoghene, 2019). The overuse of petroleum products has increased pollution in the environment (Pinedo *et al.*, 2013). Hydrocarbons, which include saturated, unsaturated, and polycyclic aromatic compounds, make up a large portion of crude petroleum and are regarded as extremely dangerous environmental pollutants (Lundstedt *et al.*, 2007). The world depends heavily on petroleum for energy production, commerce and industry, despite substantial advancements in energy technology and a serious global ecosystem crisis and as a result, petroleum mining and transportation are an inevitable ongoing process (Dudhagara *et al.*, 2016). Because of this, there is always a chance that oil will spill into both terrestrial and aquatic areas (Jin *et al.*, 2019).

The main issue with petroleum hydrocarbons is their toxicity to the environment, people, and plants (Arellano *et al.*, 2017; Yadav *et al.*, 2018). There is proof of the harm that contamination from petroleum and its byproducts causes to bacteria, plants, animals, and ecosystems (Freedman, 2018; Ordinioha and Brisibe, 2013). Crude oil toxicity is also determined by the amount of spilled oil, the amount of residual oil remaining on the site, the ecosystem in the

affected region, the response, recovery, and cleanup timeline (Mohamadi *et al.*, 2016). Various biological organisms, including prokaryotes and eukaryotes, and their life stages react to pollution in different ways (Hao *et al.*, 2017). Crude oil spills undergo a variety of weathering processes, including evaporation, dissolution, dispersion, sedimentation, photo-oxidation, and biodegradation, depending on the climate, hydrodynamics, and geographic location (Acosta-González *et al.*, 2015). Persistent chemicals have a significant impact on a long-term contaminated site, even if they may not be as harmful as a freshly contaminated environment (Jonker *et al.*, 2006).

Furthermore, The genus *Emilia* (Cass.) Cass. comprises ca. 100 species distributed in the tropical and subtropical regions of the world, with the greatest species diversity in East Africa (Beentje and Lansdown, 2018). *Emilia praetermissa* was originally described from Sierra Leone and Nigeria (Eburnlomo, 2012) and was subsequently found in other West Africa countries, including Cameroon, Côte d'Ivoire, Ghana, Guinea, and Liberia. Unlike most *Emilia* species whose chromosome numbers are  $2n = 10$ , *E. praetermissa* has a chromosome number of  $2n = 20$ , suggesting its polyploidy origin (Chung, 2009).

Also, Phytoremediation is an emerging technology that has recently being favored as a good option for the remediation of petroleum hydrocarbons compared to physiochemical methods such as land fill, cap and contain incineration, ozonation and surfactant washing (Agmuthu *et al.*, 2010). Its techniques employ the use of various plants to degrade, extract, contain or immobilize contaminants from soil and water (Waziri *et al.*, 2016). As a means of remediation of soil polluted with these substances, various technologies have been employed among which is phytoremediation (Adedokun and Ataga (2007). For efficient phytoremediation, soil amendment or additives, such as sawdust, peat, waste cotton, organic and inorganic manure, etc. are added to increase micro-organisms activities (Adedokun and Ataga (2007). A soil amendment is any material added to a soil to improve its physical properties, such as water retention, permeability, water infiltration, drainage, aeration and structure (Das and Chandran, 2011). For agriculture to be productive, the agricultural soil that plants depend on must be used sustainably. In Nigeria, soil contamination from crude oil and other petroleum products is currently a serious problem. Therefore, it is impossible to overstate the importance of this study. The aim of the study was to establish the phytoremediation capacity of *E. praetermissa* on crude oil polluted soil with cow dung enrichment.

## MATERIALS AND METHOD

The screen house of the Department of Plant Science and Biotechnology, Michael Okpara University of Agriculture, Umudike was location of the study. Within Longitude 07° 34' E, Latitude 05° 29' N and at an elevation of 122 m above sea level was the location of Umudike. (National Root Crop Research Institute, Umudike (NRCRIU), 2022). Samples of used crude oil were collected from Ukwa West Local Government Area, Abia State, Nigeria. Soil samples were collected at the depth of 0 – 15 cm in crop farms around Michael Okpara University of Agriculture, Umudike with the aid of a shovel. Plant samples were collected from the surroundings of Umuahia Township. Having only one factor (crude oil), the research design of this experiment was completely randomized design. Levels of cow dung were constant to amount of soil needed. While the crude oil was varied in 1 %, 3 % and 5 % to the soil and cow dung content. Control soil samples had zero levels of crude oil. Each soil sample were replicated three times.

**Sample preparation of the soil:** Soil samples were sieved with 2 mm sieve and was mixed in the following way; 0 % of 4000 grams of cow dung + 4000 grams of farm soil = Negative control. 10 % of 4000 grams of cow dung + 3600 grams of soil = Positive control. 10 % of 4000 grams of cow dung + 3600 grams of soil + 40 mills of crude oil = 1 %. 10 % of 4000 grams of cow dung + 3600 grams of soil + 120 mills of crude oil = 3 %. 10 % of 4000 grams of cow dung + 3600 grams of soil + 220 mills of crude oil = 5 %. The treated soil samples were permitted to stand for seven days. Subsequently, three plants samples uprooted with the roots, and of same heights were transplanted into each sack bags filled with the treated soil samples. At the end of 12 weeks of transplanting, the plants samples were removed, the soil and plant samples were taken to the laboratory for physical, and chemical constituent evaluation.

**Plant growth parameter measurements:** Growth parameters comprising of germination, leaf length, leaf number, stem girth, root length, root number and biomass was determined.

**Leaf germination (count per replicate):** The number of germinations was done by daily observation of sprouting sample plant seeds per replicates.

**Plant height (cm plant<sup>-1</sup>):** The plant height was measured using a ruler. A meter rule was used to measure upwards starting from base to the tip of plant.

**Leaf length (cm plant<sup>-1</sup>):** The leaf length was measured using a ruler. A meter rule was used to measure upwards starting from base to the tip of plant leaf.

**Leaf number (number plant<sup>-1</sup>):** The leaf number was collected by counting directly the number of leaves per plant.

**Stem girth (cm plant<sup>-1</sup>):** The girth was taken use mechanical Vernier Caliper. Girth was recorded in centimeter per plant.

**Root length (cm plant<sup>-1</sup>):** The root length was taken by placing a

ruler from the rooting point to the tip. Root length was recorded in centimeter per plant.

**Root number (number plant<sup>-1</sup>):** The root number was collected by counting directly the number of roots per plant.

**Biomass (gram plant<sup>-1</sup>):** The stem, root and leaf weight were taken by placing the various parts on a weighing balance. Records were taken in grams

**Analysis of heavy metals:** Bio-available or soluble concentration of heavy metal was determined by Aqua Regia method (Chen and Ma, 2001). Conventional aqua regia digestion was performed in 250 ml glass beakers covered with watch glasses. A well-mixed sample of 0.5000 g was digested in 12 ml of aqua regia on a hotplate for 3 hrs. at 110°C. After evaporation to near dryness, the sample was diluted with 20 ml of 2 % (v/v with H<sub>2</sub>O) nitric acid and transferred into a 100 ml volumetric flask after filtering through Whatman no. 42 filter paper and diluted to 100 ml with DDW. The filtrates were analyzed for Zinc (Zn), Mercury (Hg), Manganese (Mn), Iron (Fe), Lead (Pb), Copper (Cu), Chromium (Cr) and Cadmium (Cd) using atomic absorption spectrophotometer. The values were compared with the widely used normal and critical levels of total concentration of heavy metal for the contaminant limit (c), p index was calculated as the ratio between the heavy metal content in the soil and the toxicity criteria (the tolerable levels).

**Data analysis:** Data collected was subjected to descriptive statistics to obtain the means and standard deviations. Analysis of variance analysis was used to compare the difference in the heavy metal properties of soil, cow dung and Crude oil. Means of growth data and laboratory analysis were subjected to analysis of variance (ANOVA). Statistically significant means at 5 % probability were separated using Duncan multiple range test all the test was done using statistical package for social sciences (SPSS) version 26.

## RESULTS

Initial heavy metal properties of soil, cow dung and crude oil (CRO) is presented in Table 1a. From the result, Zn ranges from 0.06 of CD to 0.01 of CRO. Effects of initial heavy metal properties of soil, cow dung and crude oil (CRO) varied significantly different (P<0.05). Ni ranges from 0.87 of CD to 3.46 of CRO. Initial heavy metal properties of soil, cow dung and crude oil (CRO) varied significantly different (P<0.05). Mo ranges from 6.71 of CD to 22.61 of CRO. Effect of Initial heavy metal properties of soil, cow dung and crude oil (CRO) varied significantly different (P<0.05). Hg ranges from 0.03 of CD to 0.03 Soil, CRO appears negative. Effect of Initial heavy metal properties of soil, cow dung and crude oil (CRO) varied significantly different (P<0.05). Pb has a negative value for all samples. Mn ranges from 0.44 of CD to 0.95 of Soil. Effect of Initial heavy metal properties of soil, cow dung and crude oil (CRO) varied significantly different (P<0.05). Fe ranges from 0.01 of CD to 0.53 of Soil. Effect of Initial heavy metal properties of soil, cow dung and crude oil (CRO) varied significantly different (P<0.05).

**Table 1a:** Initial heavy metal properties of soil, cow dung and crude oil (CRO)

Sample	Zn	Ni	Mo	Hg	Pb	Mn	Fe
Soil	0.01 <sup>b</sup>	1.62 <sup>b</sup>	11.05 <sup>b</sup>	0.03 <sup>b</sup>	-ve	0.95 <sup>a</sup>	0.53 <sup>a</sup>
CD	0.06 <sup>c</sup>	0.87 <sup>c</sup>	6.71 <sup>c</sup>	0.03 <sup>a</sup>	-ve	0.44 <sup>c</sup>	0.01 <sup>c</sup>
CRO	0.01 <sup>a</sup>	3.46 <sup>a</sup>	22.61 <sup>a</sup>	-ve	-ve	0.82 <sup>b</sup>	0.47 <sup>b</sup>
CV	2.1370	0.0356	0.0306	4.2459	-	0.1361	0.12199
LSD	0.00184	0.0023	0.0131	0.0029	-	0.0032	0.0013
Pr(>F)	<0.001***	<0.001***	<0.001***	<0.001***	-	<0.001***	<0.001***

Mean with different superscript alphabet are significantly different (P<0.05). CV (Coefficient of variation), LSD (Least significant difference).

Initial heavy metal properties of soil, Cow dung and crude oil (CRO) is presented in Table 1b. From the result, Cu ranges from 0.08 of CRO to 0.24 of Soil. Effect of Initial heavy metal properties of soil, Cow dung and crude oil (CRO) varied significantly different (P<0.05). Co ranges from 0.01 of Soil to 0.02 CRO. Effect of Initial heavy metal properties of soil, Cow dung and crude oil (CRO) varied significantly different (P<0.05). Cr ranges from 0.02 of CD to 0.09 Soil. Effect of Initial heavy metal properties of soil, Cow dung and crude oil (CRO) varied significantly different (P<0.05). Cd ranges from 0.03 of CRO to 0.03 of Soil and has a -ve for CD.

Effect of Initial heavy metal properties of soil, Cow dung and crude oil (CRO) varied significantly different (P<0.05). Ba ranges from 9.05 of CD to 45.75 of CRO. Effect of Initial heavy metal properties of soil, Cow dung and crude oil (CRO) varied significantly different (P<0.05). B ranges from 0.35 of CD to 1.11 of CRO. Effect of Initial heavy metal properties of soil, Cow dung and crude oil (CRO) varied significantly different (P<0.05). K ranges from 1.14 of CD to 3.15 of CRO. Effect of Initial heavy metal properties of soil, Cow dung and crude oil (CRO) varied significantly different (P<0.05).

**Table 1b:** Initial heavy metal properties of soil, cow dung and crude oil (CRO)

Sample	Cu	Co	Cr	Cd	Ba	B	K
Soil	0.24 <sup>a</sup>	0.01 <sup>c</sup>	0.09 <sup>a</sup>	0.03 <sup>a</sup>	21.74 <sup>b</sup>	0.74 <sup>b</sup>	1.72
CD	0.09 <sup>b</sup>	0.01 <sup>b</sup>	0.02 <sup>b</sup>	-ve	9.05 <sup>c</sup>	0.35 <sup>c</sup>	1.14
CRO	0.08 <sup>c</sup>	0.02 <sup>a</sup>	0.06 <sup>b</sup>	0.03 <sup>b</sup>	45.79 <sup>a</sup>	1.11 <sup>a</sup>	3.15
CV	0.84516	6.1488	1.2227	4.29735	0.00392	0.1362	2.0048
LSD	0.00368	0.0023	0.00225	0.0026	0.0032	0.00318	0.0043
Pr(>F)	<0.001***	0.0023**	<0.001***	<0.001***	<0.001***	<0.001***	<0.001***

Mean with different superscript alphabet are significantly different (P<0.05). CV (Coefficient of variation), LSD (Least significant difference), CD (Cow dung)

Germination count of *E. praetermissa* grown on different levels of crude oil is presented in Table 2. From the result, 4DAP ranges from 0.00 of PC to 1.33 of NC and 1 %. Effect of germination count of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). 5DAP ranges from 0.00 of 3 % and 5 % to 1.33 of NC. Effect of germination count of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). 6DAP ranges from 0.00 of 5 % to 1.33 of NC and 1 %. Effect of germination count of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). 7DAP

ranges from 0.67 of PC, 3 % and 5 % to 1.33 of NC and 1 %. Effect of germination count of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). 8DAP ranges from 0.33 of 5 % to 1.33 of NC and 1 %. Effect of germination count of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). 9DAP ranges from 0.33 of 5 % to 1.33 of NC and 1 %. Effect of germination count of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05).

**Table 2:** Germination count of *E. praetermissa* grown on different levels of crude oil

Treatment	4 DAP	5 DAP	6 DAP	7 DAP	8 DAP	9 DAP
NC	1.33	1.33	1.33	1.33	1.33	1.33
PC	0.33	0.33	0.67	0.67	1.00	1.00
1 %	0.67	0.67	1.33	1.33	1.33	1.33
3 %	0.00	0.00	0.67	0.67	0.67	0.67
5 %	0.00	0.00	0.00	0.67	0.33	0.33
CV	191.663	191.663	116.3687	0.9333	0.9333	110.6567
LSD	1.6272	1.6272	1.6937	1.7576	1.8789	1.8789
Pr(>F)	0.3818 <sup>NS</sup>	0.3818 <sup>NS</sup>	0.4179 <sup>NS</sup>	0.785 <sup>NS</sup>	0.716 <sup>NS</sup>	0.716 <sup>NS</sup>

Mean with different superscript alphabet are significantly different (P<0.05). CV (Coefficient of variation), LSD (Least significant difference), CD (Cow dung), NC (Negative control), PC (Positive control), DAP (Days after planting).

Growth response of *E. praetermissa* grown on different levels of crude oil is presented in Table 3. From the result, LL 2WAP ranges from 0.00 of 5 % to 3.10 of NC. Effect of growth response of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). LL 4WAP ranges from 0.00 of 3 % and 5 % to 7.17 of PC. Effect of growth response of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). LL 6WAP ranges from 3.43 of 5 % to 9.60 of PC. Effect of growth response of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). SG 2WAP ranges from 0.00 of 5 % to 0.60 of NC. Effect of growth response of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). SG 4WAP ranges from 0.00 of 3 % and 5 % to 1.03 of PC. Effect of growth response of *E. praetermissa* grown on different levels of crude oil varied significantly different

(P<0.05). SG 6WAP ranges from 0.00 of 5 % to 0.93 of PC. Effect of growth response of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). NL 2WAP ranges from 0.00 of 5 % to 4.67 of NC. Effect of growth response of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). NL 4WAP ranges from 0.00 of 3 % and 5 % to 6.67 of NC. Effect of growth response of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). NL 6WAP ranges from 3.00 of 5 % to 16.33 of PC. Effect of growth response of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). Number of flowers ranges from 0.00 of 1 %, 3 % and 5 % to 41.67 of PC. Effect of growth response of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05).

**Table 3:** Growth response of *E. praetermissa* grown on different levels of crude oil

Treatment	LL 2 WAP	LL 4 WAP	LL 6 WAP	SG 2 WAP	SG 4 WAP	SG 6 WAP	NL 2 WAP	NL 4 WAP	NL 6 WAP	Number of flowers
NC	3.10	3.20 <sup>b</sup>	5.70 <sup>b</sup>	0.60	0.33 <sup>b</sup>	0.60 <sup>ab</sup>	4.67	6.67	11.00	5.67
PC	2.03	7.17 <sup>a</sup>	9.60 <sup>a</sup>	0.53	1.03 <sup>a</sup>	0.93 <sup>a</sup>	2.33	6.33	16.33	41.67
1 %	1.93	1.33 <sup>b</sup>	3.77 <sup>b</sup>	0.57	0.30 <sup>b</sup>	0.57 <sup>b</sup>	3.00	2.33	5.67	0.00
3 %	0.77	0.00 <sup>b</sup>	3.47 <sup>b</sup>	0.33	0.00 <sup>b</sup>	0.33 <sup>bc</sup>	0.67	0.00	3.33	0.00
5 %	0.00	0.00 <sup>b</sup>	3.43 <sup>b</sup>	0.00	0.00 <sup>b</sup>	0.00 <sup>c</sup>	0.00	0.00	3.00	0.00
CV	113.2147	91.6498	30.640	113.2216	112.783	38.987	129.2255	121.1359	80.196	168.5292
LSD	3.2268	3.9016	2.8949	0.8377	0.6839	0.3452	5.0154	6.75828	11.4772	29.02478
Pr(>F)	0.3079 <sup>NS</sup>	0.0107 <sup>*</sup>	0.0033 <sup>**</sup>	0.5077 <sup>NS</sup>	0.0399 <sup>*</sup>	0.0016 <sup>**</sup>	0.3108 <sup>NS</sup>	0.1261 <sup>NS</sup>	0.1131 <sup>NS</sup>	0.0371 <sup>*</sup>

Mean with different superscript alphabet are significantly different (P<0.05). CV (Coefficient of variation), LSD (Least significant difference), CD (Cow dung), NC (Negative control), PC (Positive control), WAP (Weeks after planting) LL (leaf length), SG (Stem girth), NL (Number of leaves).

Root growth and biomass of *E. praetermissa* grown on different levels of crude oil is presented in Table 4. From the result, LLR from 5.77 of 3 % to 16.83 of NC. Effect of root growth and biomass of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). NR ranges from 12.00 of PC to 22.33 of 1 %. Effect of root growth and biomass of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). RW ranges from 0.33 of 1 % to 1.33 of 5 %. Effect of root

growth and biomass of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). SW ranges from 0.00 of 1 %, 3 % and 5 % to 10.33 of PC. Effect of root growth and biomass of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05). LW ranges from 0.00 of 5 % to 9.00 of PC. Effect of root growth and biomass of *E. praetermissa* grown on different levels of crude oil varied significantly different (P<0.05).

**Table 4:** Root growth and biomass of *E. praetermissa* grown on different levels of crude oil

Treatment	LLR	NR	RW	SW	LW
NC	16.83 <sup>a</sup>	16.67	0.67	1.33 <sup>b</sup>	3.00
PC	14.50 <sup>ab</sup>	12.00	1.00	10.33 <sup>a</sup>	9.00
1 %	7.63 <sup>c</sup>	22.33	0.33	0.00 <sup>b</sup>	0.67
3 %	5.77 <sup>c</sup>	13.33	0.67	0.00 <sup>b</sup>	0.67
5 %	7.97 <sup>bc</sup>	15.33	1.33	0.00 <sup>b</sup>	0.00
CV	34.1723	38.3136	102.0621	2.3333	139.9777
LSD	6.5526	11.1060	1.4854	7.0303	6.79085
Pr(>F)	0.01415*	0.3375 <sup>NS</sup>	0.6398 <sup>NS</sup>	0.03236*	0.07358 <sup>NS</sup>

Mean with different superscript alphabet are significantly different (P<0.05). CV (Coefficient of variation), LSD (Least significant difference), CD (Cow dung), NC (Negative control), PC (Positive control), WAP (Weeks after planting) LLR (length of longest root), NR (Number of roots), LW (leaf weight), RW (Root weight), SW (Stem weight), NS (not significant).

Heavy metal properties of soil twelve weeks after planting of *E. praetermissa* is presented in Table 5. From the result, Cd ranges from 0.17 of NC and 5 % to 0.35 of 1 %. Effect of heavy metal properties of soil twelve weeks after planting varied significantly different (P<0.05). Cr ranges from 0.09 of NC to 0.15 of PC. Effect of heavy metal properties of soil twelve weeks after planting varied significantly different (P<0.05). Pb ranges from 0.00 of 3 % to 0.15

of PC and has a -ve result for the rest of the treatments. Hg ranges from 0.09 of NC and PC to 0.18 of 1 %. Effect of heavy metal properties of soil twelve weeks after planting varied significantly different (P<0.05). Zn ranges from 0.07 of PC to 0.29 of 1 %. Effect of heavy metal properties of soil twelve weeks after planting varied significantly different (P<0.05).

**Table 5:** Heavy metal properties of soil treated with crude oil twelve weeks after planting of *E. praetermissa*

Treatment	Cd	Cr	Pb	Hg	Zn
NC	0.17 <sup>b</sup>	0.09 <sup>a</sup>	-ve	0.09 <sup>d</sup>	0.13 <sup>d</sup>
PC	0.13 <sup>a</sup>	0.15 <sup>a</sup>	0.15 <sup>b</sup>	0.09 <sup>a</sup>	0.07 <sup>a</sup>
1 %	0.35 <sup>a</sup>	0.10 <sup>d</sup>	-ve	0.18 <sup>a</sup>	0.29 <sup>a</sup>
3 %	0.14 <sup>d</sup>	0.13 <sup>c</sup>	0.00 <sup>c</sup>	0.11 <sup>c</sup>	0.18 <sup>b</sup>
5 %	0.17 <sup>c</sup>	0.14 <sup>b</sup>	-ve	0.11 <sup>b</sup>	0.15 <sup>c</sup>
CV	0.3673	0.5726	0.8063	0.60695	0.4232
LSD	0.0018	0.0018	0.0018	0.0018	0.0018
Pr(>F)	<0.001***	<0.001***	<0.001***	<0.001***	<0.001***

Mean with different superscript alphabet are significantly different (P<0.05). CV (Coefficient of variation), LSD (Least significant difference), Cd (Cadmium), Cr (Chromium), Pb (lead), Hg (Mercury) Zn (Zinc).

Heavy metal properties of *E. praetermissa* grown on different levels of crude oil twelve weeks after planting is presented in Table 6. From the result, Cd ranges from 0.10 of NC to 0.49 of NC. Effect of heavy metal properties of *E. praetermissa* grown on different levels of crude oil twelve weeks after planting varied significantly different (P<0.05). Cr ranges from 0.04 of NC and 1 % to 0.69 of PC. Effect of heavy metal properties of *E. praetermissa* grown on different levels of crude oil twelve weeks after planting varied significantly different (P<0.05). Pb has a value 1.03 and appears -ve for the rest

of the treatments. Effect of heavy metal properties of *E. praetermissa* grown on different levels of crude oil twelve weeks after planting varied significantly different (P<0.05). Hg ranges from 0.05 of NC and 1 % to 0.29 of 5 %. Effect of heavy metal properties of *E. praetermissa* grown on different levels of crude oil twelve weeks after planting varied significantly different (P<0.05). Zn ranges from 0.12 of 1 % to 0.37 of 5 %. Effect of heavy metal properties of *E. praetermissa* grown on different levels of crude oil twelve weeks after planting varied significantly different (P<0.05).

**Table 6:** Heavy metal properties of *E. praetermissa* grown on different levels of crude oil twelve weeks after planting

Treatment	Cd	Cr	Pb	Hg	Zn
NC	0.10 <sup>a</sup>	0.04 <sup>d</sup>	-ve	0.05 <sup>d</sup>	0.13 <sup>a</sup>
PC	0.15 <sup>c</sup>	0.69 <sup>c</sup>	-ve	0.11 <sup>b</sup>	0.15 <sup>c</sup>
1 %	0.12 <sup>d</sup>	0.04 <sup>d</sup>	-ve	0.05 <sup>d</sup>	0.12 <sup>a</sup>
3 %	0.19 <sup>b</sup>	0.07 <sup>c</sup>	-ve	0.11 <sup>b</sup>	0.19 <sup>b</sup>
5 %	0.49 <sup>a</sup>	0.24 <sup>b</sup>	1.03	0.29 <sup>a</sup>	0.37 <sup>a</sup>
CV	0.33849	0.32691	-	0.5744	0.37002
LSD	0.0018	0.0018	-	0.0018	0.0018
Pr(>F)	<0.001***	<0.001***	-	<0.001***	<0.001***

Mean with different superscript alphabet are significantly different (P<0.05). CV (Coefficient of variation), LSD (Least significant difference), Cd (Cadmium), Cr (Chromium), Pb (lead), Hg (Mercury) Zn (Zinc).

## DISCUSSION

The findings of this contemporary study showed that among all the three-growth media studied, soil had higher Cu, Co, and Cr, which may be as a result of human activities or anthropogenic activities around the site of collection. This result is supported by Ifediora *et al.* (2023a) who reported in their study, presences of most heavy metals in soil and hydrocarbon such as oil. Hence, the need for phytoremediation. This also, disagrees with the work of Merkl *et al.* (2012) who observed higher chemical properties in soil before contamination than those in the crude oil. The result showed that the heavy metal in soil, cow dung and crude oil varied significantly (P<0.05). The cow dung had less contribution to the heavy metal levels in the growth media when compared to crude oil and soil. This may be as a result of the long-established potential of crude oil to contaminate the soil with heavy metals. This is supported by Ifediora *et al.* (2023b) who also, in their study confirmed that spent engine oil contaminates the soil with toxic heavy metals.

The germination rate result of *E. praetermissa* showed that germination occurred after four days of initiation. The negative control sample had more germination rate than the other samples across the time of observation. This may be connected to reduction in water imbibition by the seed, caused by films of crude oil. Higher levels of crude oil contaminated soil inhibited germination. This result is confirmed by Riskuwa-Shehu *et al.* (2017) who stated that generally, low germination rates were observed in the six-legume species they screened. The reason might also be the toxicity of the crude oil, which caused the reduction in germination rate. In support to this observation, Vwioko and Fashemi (2005), noted a reduction in germination rate which could be as a result of coating of oil on seed surface, thereby affecting physiological functions within the seed. Marques *et al.* (2010) made a similar observation and concluded that one of the plant responses to the presence of oil is the delay and/or reduction in germination rate.

The leaf length of the plant generally was affected by the crude oil levels. This may be as a result of increased contamination level of the crude oil, which lead to decrease in length of leaf. The findings of the present study agreed with Nwankwo (2014), who observed that levels of crude oil contamination have different levels of inhibition of growth. Merkl *et al.* (2004), supported this by stating that oil contamination (3 %) can cause low growth stimulation as observed for shoot length of *Mimosa camporum*. Sagaya *et al.*, (2023) stated that the Statistical analysis of the vegetative growth showed that there was significant difference in all the vegetative parameter studied. The effect of the spent engine oil was more obvious on the contaminated soil-plant than those of the control.

Similar observation was observed in the stem girth, hence, the effect of difference in levels of crude oil were significantly different (P<0.05). This may be as a result of increased contamination level of crude oil, which lead to decrease in stem girth. The findings of the present study agreed with Nwankwo (2014), who in her study noticed that the results showed a similar trend in growth trial. At 5% crude oil level (Soil 5 %), the growth inhibition was 86 % when compared to the uncontaminated soil and 79.5 % inhibition at 7.5 % crude oil level (Soil 7.5 %). Idowu and Fayinminnu (2016), also agrees with the findings by stating that increased contamination of soil with spent engine lubricating oil (3 and 6 % levels of contamination) had observable adverse effects on the growth parameters.

The number of leaves was affected by the crude oil levels, which could be as a result of physiological adaptations to reducing environmental conditions which is concentration dependent. The findings of the study agreed with Sagaya *et al.*, (2023) who stated that the plant height, number of leaves and leaf area of the plants were affected by the spent engine oil and the results tend toward the same way as the concentrations of the oil increased, the vegetative parameter reduced. Adekunle *et al.* (2010) used maize (*Zea mays* L.), cowpea (*Vigna unguiculata*) and spinach (*Amaranthus hybridus*) to measure crude oil resistance by investigating such indices as seed germination, plant height and leaf number. They concluded that of the three crops used except for *Z. mays*, the other two crops recorded 100 % inhibition in seed germination.

Flowering occurred only in the untreated samples which could be as a result of physiological adaptations to reducing environmental conditions. Nwankwo (2007), agreed with the result by stating that possibly, the oxygen depleted environment caused by the presence of the oil and use of other electron acceptors such as nitrates led to reducing environmental conditions and probably affected plants' use of nutrients and their physical development. Sagaya *et al.*, (2023) agreed by observing that the significant percentage differences observed in the yield parameters such as length, dry weight, fresh weight and number of fruits could be attributed to the high level of oil in soil and hence the uptake of ions and nutrients carried out by the roots becomes difficult. Adedokun and Ataga (2007), disagrees with this by stating that improvement in growth was also recorded in amendments of diesel and spent engine oil polluted soils with cotton and saw dust when compared with non-amended polluted soils.

The root number was discovered to be higher in the controlled

samples. The reduction in root number in plants exposed to crude oil contamination is associated with the surface blocking caused by the crude oil against uptake of nutrient and water. Merkl *et al.* (2012) had reported that shoot length and root length of eight tropical plant were lower in soil plant with 3 % and 5 % crude oil contamination. Ifediora *et al.* (2022) reported also that plants of the treatment with more contamination had widened cell spaces in the cortex that were virtually always present where death of parenchyma cells was observed. Similar observation was been made by Anyasi and Atagana (2018) in a report on profiling of plants at petroleum contaminated site for phytoremediation

Similarly, biomass of the plant at harvest had higher weight in controlled sample. However, this ability of *E. praetermissa* may be attributed to the remediation potentials of the plant to toxic soil. This finding agrees with the observation cited by Merkl *et al.* (2004), that crude oil contamination (3 %) can cause growth stimulation in the shoot biomass of *Saprochaete capitata* possibly due to a hormonally influenced stress response. Nwankwo (2007), disagreed with the result by stating that the variation in germination, growth and biomass yield of plants on unpolluted soils showed that soil quality can affect productivity. For example, the soil obtained from the Niger Delta did not sustain growth of tomato seeds after the initial germination in comparison to soil from Bardsey, UK. Adedokun and Ataga (2007), supported by stating that the growth parameters: plant height, leaves and biomass were comparatively low in all the polluted soils compared to the control, with no pollutant.

Furthermore, the heavy metal was observed to be less in the control samples, hence the crude oil contamination was responsible for higher heavy metal residue in the soil. Toxic metals like lead was not observe in most of the soil samples. However, higher metals observed in crude oil contaminated soil showed remediation capacity of the plant studied which may be as a result of the organic enrichment present. This finding agrees with the observation cited by Oguche *et al.*, (2022), who wrote that lemon grass is suitable for phytoremediation due to its ability for hyper-cumulative of these heavy metals which are bioavailable for uptake. Marchand (2017), supported by saying that during a 5-month greenhouse trial, the efficiency of *M. sativa* singly and in combination with compost have been used for the degradation of PHC and the phytostabilization or extraction of TE. *M. sativa* cultivated in this contaminated soil without compost did not lead to the rhizodegradation of high molecular weight PAH and the removal of soil Co, As, and Pb. Udom and Nuga (2015), agrees by noting that the reduction in heavy in soil treated with hydrocarbons over time of planting have been reported in the study of the biodegradation of petroleum hydrocarbons in a tropical ultisol using legume plants and organic manure.

Also, the heavy metal levels in the plant samples was low and were non-existing in some toxic metals like mercury. This may be as a result of *E. praetermissa* capacity to take up heavy metals because, phytoremediation of contaminated sites was found to be dependent upon the soil amendments used as they render contaminants unavailable for uptake. Marchand (2017), had reported that in addition to the difficulty related to the use of aged co-contaminated soils and the complexity of these pollutants, soil characteristics such as the high content of organic matter are favoring the sorption to soil particles. Kaartinen, *et al.*, (2013)

made related observation by showing that the compost amendment significantly increased the water content of *M. sativa* shoots resulting in higher plant vigor, plant growth and survival. Also, they stated that compost application into the studied soil may have indirectly promoted plant growth through soil TE reactions with mineral components of the compost, the formation of stable complexes with organic ligands, nutrient supply and microbial inoculation. Similar findings have been made by Nwichi *et al.* (2015). Finally, Messou *et al.* (2013) in their study of plants diversity and phytoaccumulators identification on the Akouedo landfill mentioned *E. praetermissa* as a tropical plant involve in phytoaccumulators.

### Conclusion

The focus of this study was on the understanding and the improving of remediation technologies for the local treatment of polluted land using *E. praetermissa*, without need of transportation of the pollutants through the city. The findings indicated that soil, crude oil and cow dung are natural sources of heavy metals. The seed germination rate, root growth and biomass were observed to significantly differ ( $P < 0.05$ ) along the treatment. Higher crude oil treated samples negatively affected the germination rate, growth and flowering properties of *E. praetermissa*. Low levels of heavy metals in the soil after growth period and availability of heavy metal in the plant tissue indicates the phytoremediation potentials of *E. praetermissa*. Thus, phytoremediation is a promising and sustainable method for reclaiming soils contaminated with these toxic metals. Further studies should assess the biosafety of the crude samples on the plant to mitigate against possible toxic impact to animals feeding on the plant.

### REFERENCES

- Acosta-González, A., Martirani-von Abercron, S. M., Rosselló-Móra, R., Wittich, R. M. and Marqués, S. (2015). The effect of oil spills on the bacterial diversity and catabolic function in coastal sediments: a case study on the Prestige oil spill. *Environmental Science and Pollution Research*, 22: 15200-15214.
- Adedokun, O. M. and Ataga, A. E. (2007). Effects of amendments and bioaugmentation of soil polluted with crude oil, automotive gasoline oil, and spent engine oil on the growth of cowpea (*Vigna unguiculata* L. Walp). *Scientific Research and Essay*, 2 (5): 147-149.
- Adekunle, I. M. (2011). Bioremediation of soils contaminated with Nigerian petroleum products using composted municipal wastes. *Bioremediation Journal*, 15: 230 - 241.
- Agamuthu, P., Abioye, P. O. and Abduiaziz, R. (2010). Phytoremediation of soil contaminated with used lubricating oil using *Jathropa curcas*. *Journal of Hazardous Materials*, 179(1-3): 891-4.
- Anyasi, R. O. and Atagana, H. I. (2018). Profiling of plants at petroleum contaminated site for phytoremediation. *International Journal of Phytoremediation*, 20(4): 352–361.
- Arellano, P., Tansey, K., Balzter, H. and Tellkamp, M. (2017). Plant family-specific impacts of petroleum pollution on biodiversity and leaf chlorophyll content in the Amazon rainforest of Ecuador. *PloS one*, 12(1): e0169867.
- Beentje, H. J. and Lansdown, R. V. (2018). *Cyperus compressus*. The IUCN red list of threatened species 2018: e.T164257A120149831. <https://doi.org/10.2305/IUCN.UK.2018->

- 2.RLTS.T164257A1201498 31.en. (Accessed in 22/1/24).
- Chen, M. and Ma, L. Q. (2001). Comparison of three aqua regia digestion methods for twenty Florida soils. *Soil Science Society of American Journal*, 65(2):491-9.
- Chung, K. F., Ku, S. M., Kono, Y. and Peng, C. I. (2009). *Emilia praetermissa* Milne-Redh. (Asteraceae)-a misidentified alien species in Northern Taiwan. *Taiwania*, 54(4): 385-390.
- Das, N. and Chandran, P. (2011). Microbial degradation of petroleum products contaminants: an overview. *Biotechnology Research International*, 941810.
- Dudhagara, D. R., Rajpara, R. K., Bhatt, J. K., Gosai, H. B., Sachaniya, B. K. and Dave, B. P. (2016). Distribution, sources and ecological risk assessment of PAHs in historically contaminated surface sediments at Bhavnagar coast, Gujarat, India. *Environmental pollution*, 213: 338-346.
- Ebunlomo, A. O., Odetola, A. O., Bamidele, O., Egwurugwu, J. N., Maduka, S. and Anopue, J. (2012). Effects of *Emilia praetermissa* leaf extract on the haematological and biochemical parameters of stress induced ulcerated Wistar rats. *African Journal of Biochemistry Research*, 6(14): 185-189.
- Freedman, B. (2018). *Environmental science*. Canada: Dalhousie University Libraries Digital Editions.
- Hao, C., Wei, P., Pei, L., Du, Z., Zhang, Y., Lu, Y. and Dong, H. (2017). Significant seasonal variations of microbial community in an acid mine drainage lake in Anhui Province, China. *Environmental Pollution*, 223: 507-516.
- Iadow, O. D. and Fayinminnu, O. O. (2016). Total petroleum hydrocarbon and heavy metal remediation with *Jatropha curcas* L. seedlings grown on spent oil polluted soil. *Nature and Science*, 14(4): 14-20.
- Ifediora, N. H., Edeoga, H. O., Garuba, O. and Obi, O. M. (2022). Phytotoxicity study on the effects of waste engine oil on the anatomy of *Sataria barbata* (Lam.) Kunth and *Brachiaria deflexia* (Schummach.) Ce Hubb. Ex Robyns. *African Scientist*, 22(1):1595- 6881.
- Ifediora, N. H., Rowland, C. G. and Lawrence, B. N. (2023a). Heavy metal concentration and physicochemical changes of soil treated with spent engine oil and organic manure (poultry) after 12 weeks of growing *Cyperus compressus* L. *African Scientist*, 24(3):1595-6881.
- Ifediora, N., Oti, V. and Adaji, A. (2023b). Changes in physicochemical and heavy metal properties of soil treated with crude oil and poultry manure after 12 weeks of growing *Phyllanthus urinaria*. *BIU Journal of Basic and Applied Sciences*, 8(1): 36-48.
- Jin, J., Wang, H., Jing, Y., Liu, M., Wang, D., Li, Y. and Bao, M. (2019). An efficient and environmental-friendly dispersant based on the synergy of amphiphilic surfactants for oil spill remediation. *Chemosphere*, 215: 241-247.
- Jonker, M. T., Brils, J. M., Sinke, A. J., Murk, A. J. and Koelmans, A. A. (2006). Weathering and toxicity of marine sediments contaminated with oils and polycyclic aromatic hydrocarbons. *Environmental Toxicology and Chemistry: An International Journal*, 25(5): 1345-1353.
- Kaartinen, T., Sormunen, K. and Rintala, J. (2013). Case study on sampling, processing and characterization of landfilled municipal solid waste in the view of landfill mining, *Journal of Cleaner Production*, 55: 56-66.
- Lundstedt, S., White, P. A., Lemieux, C. L., Lynes, K. D., Lambert, I. B., Öberg, L. and Tysklind, M. (2007). Sources, fate, and toxic hazards of oxygenated polycyclic aromatic hydrocarbons (PAHs) at PAH-contaminated sites. *AMBIO: A Journal of the Human Environment*, 36(6): 475-485.
- Marchand, C. (2017). Phytoremediation of soil contaminated with petroleum hydrocarbons and trace elements. PhD Thesis, Linnaeus University, Kalmar. pp 1-87.
- Marques, M., Rosa, G. S., Aguiar, C. R. C., Correia, S. M. and Carvalho, E. M. (2010). Seedling emergence and biomass growth of oleaginous and other tropical species in oil contaminated soil. *The Open Waste Management Journal*: 3(1).
- Messou, A., Coulibaly, L., Doumbia, L. and Gourene, G. (2013). Plants diversity and phytoaccumulators identification on the Akouedo landfill (Abidjan, Côte d'Ivoire). *African Journal of Biotechnology*, 12(3).
- Mohamadi, B., Liu, F. and Xie, Z. (2016). Oil spill influence on vegetation in Nigeria and its determinants. *Polish Journal of Environmental Studies*, 25(6).
- National Root Crops Research Institute, Umudike (NRCRIU): Geographical data. (2022).
- Nwaichi, E. O., Frac, M., Nwoha, P. A. and Eragbor, P. (2015). Enhanced phytoremediation of crude oil-polluted soil by four plant species: effect of inorganic and organic bioaugmentation. *International Journal of Phytoremediation*, 17(12): 1253-1261.
- Nwankwo, C. A. (2014). Compost to reduce oil contamination in soils. PhD Thesis, The University of Leeds, School of Civil Engineering. pp. 1-262.
- Oguche, J. E. Agbo, S. A., Oyegoke, T. and Emeniru, D. C. (2022). Phytoremediation of used engine oil contaminated soil using lemon grass. *International Journal of Engineering Science and Application*, 6(4): 95-102.
- Ordinoha, B. and Brisibe, S. (2013). The human health implications of crude oil spills in the Niger Delta, Nigeria: An interpretation of published studies. *Nigerian medical journal: journal of the Nigeria Medical Association*, 54(1): 10.
- Pinedo, J., Ibáñez, R., Lijzen, J. P. A. and Irabien, A. (2013). Assessment of soil pollution based on total petroleum hydrocarbons and individual oil substances. *Journal of Environmental Management*, 130: 72-79.
- Riskuwa-Shehu, M. L., Ijah, U. J. J., Manga, S. B. and Bilbis, L. S. (2017). Evaluation of the use of legumes for biodegradation of petroleum hydrocarbons in soil. *International Journal of Environmental Science and Technology*, 14(10): 2205-2214.
- Sagaya, A., Abdulrahman, A. A., Oluwanisola, P. O. and Tsoho, S. B. (2023). Effects of soil pollution on the germination, growth, fruiting and leaf anatomy of *Abelmoschus caillei* (A Chev.) Stevels Malvaceae. *Science World Journal*, 18(1): 64-70.
- Sojinu, S. O. and Ejeromedoghene, O. (2019). Environmental challenges associated with processing of heavy crude oils. *Processing of Heavy Crude Oils*, 241.
- Waziri, M., Abdullahi, U., Audu, A. A. and Kalimullah (2016). Phytoremediation potentials of selected plants in industrially contaminated soils. *International Journal of Environmental Science and Development*, (7):10.
- Yadav, K. K., Gupta, N., Kumar, A., Reece, L. M., Singh, N., Rezaia, S. and Khan, S. A. (2018). Mechanistic understanding and holistic approach of phytoremediation: A review on application and prospects. *Ecological Engineering*, 120: 274-298.