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Evaluation of Growth Parameters of Tomato Seeds Planted in Crude Oil Polluted Soil Remediated with Strains of *Bacillus subtilis* and *Pseudomonas aeruginosa*

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Abstract:- The beneficial effects of bioremediation with oil-degrading microorganism, *Pseudomonas aeruginosa* and *Bacillus subtilis*, on tomato seedlings growth in oil-contaminated soil was investigated. The effect of bioremediation was examined with *Pseudomonas aeruginosa* and *Bacillus subtilis* on phytotoxicity, as measured by tomato seed growth. Tomato growth parameters (leaf length, height, seed germination, chlorosis, stunted growth) were evaluated daily for 15days. In the experimental samples that was polluted with crude petroleum and inoculated with *Pseudomonas aeruginosa*, the percentage seed germination was 99% while the seedlings reached a mean height of 66.9±0.20mm with mean leaf length of 8.1±0.10mm, while in the control set-up without the isolate, percentage seed germination, height and leaf length were 39%, 38.6% and 4.4mm respectively. Records from experimental sample inoculated with Bacillus subtilis, percentage seed germination at the end of the 15days monitoring period was 98%, while mean height of the tomato seedlings was 66.0±0.20mm and mean length was 7.9±0.20mm, at the end of the monitoring period was 3.2±0.20mm. Some qualitative morphological observations made on the seedlings were also observed for seedlings. In the untreated control as well as the treated experimental samples (*Pseudomonas aeruginosa* and *Bacillus subtilis*), sprouting of the seeds were rapid and completed in a few days, the seedlings showed steady growth and morphologically normal. In the control soils however, sprouting was sluggish, and growth was stunted, yellowing of the leaves was observed (chlorosis); a number of the seedlings were folded at the tips. These were indications of phytotoxicity resulting from oil pollution. The results of the present research have proven the bioremediation potential of bacterial isolates, for crude oil contamination which also showed significant growth potential for the tomato seeds.

Keywords:- *Phytotoxicity, Pollution, Crude Oil, Pseudomonas aeruginosa, Tomatoes.*

I. INTRODUCTION

Pollution of arable land by industrial effluents, liquid wastes such as crude petroleum, including gaseous petroleum products, are common phenomenon in many countries of the world. This is especially true of oil producing countries (Basumatory *et al.*, 2012; Nwaugo *et al.*, 2007; Brooijmans *et al.*, 2009).

Crude oil contamination is a serious environmental threat for soil and plants growing in it. Contamination of soil with oil results in serious depression of growth of most plants, primarily due to its effects on the physical and chemical properties of soil and soil water properties. Crude oil affects the environment by changing the essential elements of habitat. To overcome this problem, different soil remediation technologies have been developed such as soil washing with surfactants, biological treatment, thermal treatment, etc. Bioremediation has the potential to change the soil physicochemical properties (Brennan *et al.*, 2014), improve the water holding capacities of soil (Evangelou *et al.*, 2014) eventually resulting in increased soil fertility (Mia *et al.*, 2017); with potential beneficial effects on crop productivity, plant establishment, and growth (Brennan *et al.*, 2014).

Bioassays such as measurements of seed germination and early seedling growth have been used to monitor treatment effects and restoration of oil-contaminated sites (Sverdrup *et al.*, 2003). Dorn and Salanitro (2000) found that seed germination and plant growth using corn, wheat, and oats differed from different soils and oil combinations before, during, and after bioremediation. Sayles *et al.* (1999) showed that oil-contaminated soil treated with aerobic biodegradation was less toxic to lettuce and oat root elongation. Hanson *et al.* (2007) reported that *Acinetobacter* sp. A3-treated soil permitted better germination and growth of mungbeans, as evidenced by better plant length, weight, and leaf chlorophyll content. This indicated that *Acinetobacter* sp. A3 was capable of reducing crude oil phytotoxicity through biodegradation.

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Considering the detrimental effects of crude oil pollution on plants and its attendant implications for food security and environmental integrity in oil rich regions, it has become necessary to preserve the available arable land through bioremediation approaches

In this study, we investigated the remediation of crude oil contaminated soil using isolates of *Bacillus subtilis* and *Pseudomonas aeruginosa*. Treated soils were used to study the germination of tomato seedlings and its morphological, and physiological responses were used as indicators for soil toxicity.

II. METHODS

➢ *Study Design*

Experiments to achieve ex-situ bioremediation were set up in sizable plastic containers of equal dimensions. 5% pollution of the freshly obtained garden soil sample was prepared in the laboratory set ups designated as follow:

- Experimental set up (ESU) which contained polluted agricultural soil (5% v/w) and 250ml of the *Pseudomonas aeruginosa* broth at its exponential growth phase evenly dispersed all over the set up.
- A second but similarly designed representing control 1 (CSU 1) which contained the polluted agricultural soil (5%v/w) and 250ml sterile prepared microbiological media (UDBB), but devoid of any microorganism and evenly dispersed in the soil.
- A third also similarly designed representing control 2 (CSU 2) which contained the polluted agricultural soil (5%v/w) and 250ml sterile distilled water (SDW) evenly dispersed all over the soil.

The above designs were set up for both *Pseudomonas aeruginosa* denoted sample A and for *Bacillus subtilis* denoted sample B.

➢ *Testing for Decontamination of the Polluted Soil*

The experimental and control soils were examined nine (9) weeks after bioremediation of the oil pollution treatment process. This was to check the recovery of the agricultural soil from the oil impaction. To achieve this test, 100 Tomato seeds (*Lycopersicon esculantum)* were planted per replicate set up and the results obtained compared to those of a third control (contr 3) which was not treated (untreated and unpolluted agricultural soil). The seeds were watered at weekly intervals and the daily records of**:** Number of germination, their morphology, mean seedling height, mean length of leaves and mean leaf breadth were taken. Observations e.g leaf folds/curls, leaf droppings, chlorosis, stunted growth etc were also made (Akujobi *et al.*, 2011).

III. RESULTS AND DISCUSSION

The percentage seed germination of the tomato seeds (*Lycopesicon esculetum*) and the mean seedling height; The mean leaf length and the mean largest leaf breath of the experimental set ups (ESU) as well as Control 1 (C_1**)**, Control 2 (C_2) and Control 3 (C_3), are presented in Tables 1 to 4 for polluted soils bioremediated with polluted with *Pseudomonas aeruginosa* and *Bacillus subtilis* respectively; for 15days, which marked the end of the monitoring period for planting. 100% percentage seed germination was observed in the untreated control samples (C_3), while the mean height of the seedlings was 71.0±0.20mm. Mean leaf length recorded was 8.9±0.00mm while the largest leaf breath was 3.8±0.20mm. In the experimental samples that was polluted with crude petroleum and inoculated with *Pseudomonas aeruginosa*, the percentage seed germination was 99% while the seedlings reached a mean height of 66.9±0.20mm. The mean leaf length observed for this set up was 8.1±0.10mm, while the mean largest leaf breath was 3.3±0.00mm. For the control set ups of sample A; the records were as follows: percentage seed germination for controls 1and 2 are 39.0% and 35.0% respectively, while the mean height of seedlings for same controls were 38.6±0.00mm for control 1 and 36.8±0.00 for control 2 respectively. The mean leaf length of sample A controls were: 4.4±0.00mm for control 1 and 4.1±0.10 for control 2 respectively, while the largest leaf breath recorded were, 2.5±0.10mm for control 1 and 2.2±0.20mm for control 2 respectively(Tables 1 and 2)

Records from experimental sample B and controls which was crude petroleum polluted agricultural soil, seeded with *Bacillus subtilis* are presented in Table 3 and 4. The record shows the following values: The percentage seed germination at the end of the 15days monitoring period is 98%, while mean height of the tomato seedlings was 66.0±0.20mm ; mean length length recorded was 7.9±0.20mm, while the largest leaf breath at the end of the monitoring period was 3.2±0.20mm.

From the control soils, at the end of the 15day monitoring period, percentage seed germination recorded was 38.0% and 34.0% for B controls 1 and 2 respectively. The mean height of seedlings was 38.0±0.10mm for controls 1 and 36.1±0.20mm for control 2.The mean leaf length for controls 1 and 2 were 3.8±0.40mm and 3.6±0.10mm respectively, while the largest leaf breath were 2.4± 0.00mm for control 1 and 1.9 ±0.00mm for control 2.

Some qualitative morphological observations made on the seedlings were also recorded in Table 5 for sample A soils treated with *Pseudomonas aeruginosa*, and Table 6 for sample B soils treated with *Bacillus subtilis*. In the untreated control (C_3) as well as the treated experimental samples A and B, sprouting of the seeds were rapid and completed in a few days, the seedlings showed steady growth and morphologically normal. In the control soils however, sprouting was sluggish, and when they did sprout, growth was stunted, yellowing of the leaves was observed (chlorosis); A number of the seedlings were folded at the tips while their stems were pale and etiolated. These were indications of phytotoxicity resulting from oil. A number of the control stems with leaves felled and dropped on their sides.

Table 1 Mean percentage seed germination (%) and height of seedlings (mm) of tomato seedlings (*Lycopersicon esculetum*) planted in crude petroleum polluted agricultural soil after bioremediation using Pseudomonas *aeruginosa* [SAMPLES A]

Mean Percentage Seed Germination (%)					Mean Height Of Seedling (mm)			
Time in Days	**Exptl**	**Contr 1**	**Contr 2**	**Contr 3**	**Exptl**	**Contr 1**	**Contr 2**	**Contr 3**
0	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
1	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
2	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
3	4.0±0.50	0.0±0.00	0.0±0.00	5.0±0.50	5.0±0.50	0.0±0.00	0.0±0.00	3.8±0.10
4	20.0±0.30	2.0±0.50	1.0±0.50	25.0±0.50	4.6±0.00	1.2±0.10	1.0±0.10	6.2±0.10
5	25.0±0.20	8.0±0.50	3.0±0.00	30.0±0.00	10.2±0.00	3.4±0.10	2.1±0.00	14.8±0.00
6	32.0±0.10	8.0±0.50	9.0±0.00	42.0±0.50	20.2±0.00	6.1±0.10	5.9±0.10	22.6±0.10
7	45.0±0.50	23.0±0.00	22.0±0.50	68.0±0.00	28.0±0.10	8.6±0.10	6.1±0.10	30.0±0.10
8	58.0±0.30	26.0±0.50	24.0±0.00	82.0±0.50	34.2±0.10	14.6±0.10	12.5±0.00	38.8±0.30
9	62.0±0.20	32.0±0.00	26.0±0.00	96.0±0.00	40.4±0.10	20.1±0.00	18.8±0.10	46.9±0.40
10	71.0±0.20	34.0±0.50	29.0±0.00	100.0±0.00	46.5±0.00	34.9±0.00	21.5±0.10	50.4±0.10
11	86.0±0.10	36.0±0.50	30.0±0.50	100.0±0.00	52.8±0.00	28.0±0.00	22.4±0.00	58.8±0.20
12	92.0±0.00	38.0±0.50	32.0±0.50	100.0±0.00	59.4±0.10	30.1±0.00	28.2±0.10	62.5±0.00
13	97.0±0.00	38.0±0.50	32.0±0.50	100.0±0.00	62.0±0.20	32.2±0.10	31.1±0.10	62.6±0.00
14	99.0±0.10	38.0±0.00	33.0±0.00	100.0±0.00	48.5±0.10	36.8±0.20	33.4±0.20	68.1±0.10
15	99.0±0.00	38.0±0.00	33.0±0.00	100.0±0.00	66.9±0.20	38.0±0.00	36.9±0.00	69.0±0.00

Table 2 Mean leaf length (mm) and largest leaf breath (mm) of tomato seedlings (*Lycopersicon esculetum*) planted in crude petroleum polluted agricultural soil after bioremediation using Pseudomonas *aeruginosa* [Sample A]

Mean Leaf Length (mm)					Mean Largest Leaf Breath (mm)			
Time in days	**Exptl**	**Contr 1**	**Contr 2**	**Contr 3**	**Exptl**	**Contr 1**	**Contr 2**	**Contr 3**
0	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
2	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
3	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
4	0.0±0.00	0.0±0.00	0.0±0.00	2.6±0.10	1.2±0.10	0.0±0.00	0.0±0.00	1.4±0.20
5	2.0±0.10	1.0±0.10	0.0±0.0	3.0±0.10	1.2±0.10	0.1±0.10	0.2±0.10	1.6±0.10
6	2.9±0.30	1.3±0.00	1.0±0.10	3.6±0.10	1.5±0.30	0.6±0.40	0.4±0.20	1.0±0.10
7	0.40	1.6±0.20	1.2±0.20	4.0±0.10	1.6±0.10	1.2±0.10	0.6±0.10	1.9±0.20
8	4.6±0.10	2.0±0.20	1.7±0.30	4.0±0.30	1.8±0.20	1.3±0.00	1.2±0.20	2.0±0.10
9	5.2±0.20	2.2±0.20	2.0±0.00	4.5±0.20	2.1±0.20	1.3±0.00	1.2±0.20	2.4±0.30
10	5.9±0.30	2.6±0.10	2.1±0.10	5.0±0.40	2.2±0.30	1.5±0.10	1.3±0.10	2.7±0.20
11	6.5±0.10	3.2±0.20	2.3±0.30	5.8±0.10	2.4±0.40	1.7±0.30	1.4±0.10	2.9±0.10
12	7.1±0.10	3.6±0.40	2.5±0.30	6.8±0.20	2.6±0.20	1.9±0.10	1.5±0.20	3.0±0.00
13	7.2±0.20	3.6±0.10	2.8±0.30	7.9±0.00	2.8±0.00	2.2±0.10	1.6±0.10	3.3±0.20
14	7.4±0.20	3.6±0.10	2.8±0.20	8.0±0.10	3.0±0.10	2.3±0.20	1.8±0.10	3.5±0.40
15	7.8±0.10	3.8±0.10	2.9±0.10	8.0±0.10	3.1±0.10	2.4±0.10	1.9±0.00	3.8±0.20
16	8.0±0.10	3.8±0.10	2.9±0.10	8.1±0.00	3.2±0.00	2.5±0.10	2.2±0.20	3.8±0.20

Table 3 Mean percentage seed germination (%) and height of seedlings (mm) of tomato seedlings (*Lycopersicon esculetum*) planted in crude petroleum polluted agricultural soil after bioremediation using Bacillus *subtilus* [SAMPLES B]

Mean Percentage Seed Germination (%)					Mean Height of Seedling (mm)			
Time in days	**Exptl**	**Contr 1**	**Contr 2**	**Contr 3**	**Exptl**	**Contr 1**	**Contr 2**	**Contr 3**
0	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
1	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
2	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
3	1.0±0.20	0.0±0.00	0.0±0.00	5.0±0.10	2.0±0.00	1.0±0.20	0.0±0.00	3.8±0.50
4	3.0±0.30	0.0±0.00	1.0±0.20	25. 0±0.40	3.2±0.50	1.1±0.10	1.1±0.40	6.2±0.20
5	26.0±0.20	6.0±0.10	1.0±0.10	30.0±0.10	6.6±0.30	2.4±0.30	1.9±0.40	14.8±0.40
6	37.0±0.10	20.0±0.10	8.0±0.30	42.0±0.30	16.9±0.20	5.5±0.20	4.0±0.20	22.6±0.00
7	40.0±0.00	23.0±0.30	20.0±0.30	68.0±0.20	26.4±0.10	8.6±0.40	6.2±0.30	30.0±0.10
8	50.0±0.50	27.0±0.40	22.0±0.20	82.0±0.10	30.5±0.00	14.0±0.30	12.9±0.10	38.8±0.20
9	68.0±0.10	30.0±0.20	28.0±0.10	96.0±0.30	36.2±0.40	16.4±0.20	15.0±0.40	46.9±0.40

10	78.0±0.10	34.0±0.30	29.0±0.10	100.0±0.00	42.1±0.30	20.5±0.30	18.9±0.20	50.4±0.30
11	82.0±0.10	36.0±0.10	30.0±0.10	100.0±0.00	50.5±0.10	26.2±0.10	26.4±0.00	58.8±0.10
12	88.0±0.40	38.0±0.30	33.0±0.40	100.0±0.00	55.9±0.50	30.1±0.00	27.0±0.50	62.5±0.30
13	96.0±0.30	38.0±0.00	33.0±0.20	100.0±0.00	60.1±0.30	33.0±0.20	30.1±0.40	65.6±0.20
14	98.0±0.10	38.0±0.00	34.0±0.50	100.0±0.00	62.0±0.00	36.6±0.30	34.2±0.10	68.1±0.10
15	98.0±0.10	39.0±0.20	34.0±0.00	100.0±0.00	66.2±0.20	39.8±0.10	36.3±0.20	71.0±0.20

Table 4 Mean leaf length (mm) and largest leaf breath (mm) of tomato seedlings (*Lycopersicon esculetum)* planted in crude petroleum polluted agricultural soil after bioremediation using *Bacillus subtilus* [SAMPLE B]

	Mean leaf length (mm)				**Mean largest leaf breath (mm)**			
Time in days	**Exptl**	**Contr 1**	**Contr 2**	**Contr 3**	**Exptl**	**Contr 1**	**Contr 2**	**Contr 3**
0	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
1	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
2	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
3	0.0±0.00	1.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	1.4±0.20
4	1.6±0.10	1.0±0.10	0.0±0.00	3.0±0.10	1.1±0.50	0.1±0.10	0.15±0.10	1.6±0.50
5	2.5±0.40	1.1±0.20	1.0±0.20	3.6±0.30	1.3±0.30	0.4±0.30	0.4±0.10	1.6±0.40
6	3.1±0.20	1.3±0.00	1.2±0.10	4.0±0.40	1.5±0.10	1.0±0.30	0.4±0.10	1.9±0.20
7	4.4±0.00	1.8±0.10	1.5±0.00	4.0±0.40	1.7±0.20	1.2±0.40	0.8±0.10	2.0±0.30
8	4.9±0.00	1.9±0.20	1.9±.040	4.5±0.30	2.1±10	1.2±0.40	1.1±0.20	2.4±0.40
9	5.8±0.10	2.4±0.10	1.9±0.40	5.0±0.10	2.3±0.30	1.4±0.20	1.2±0.10	2.7±0.30
10	6.4±0.20	3.3±0.40	2.4±0.30	5.8±0.30	2.3±0.30	1.6±0.30	1.4±0.30	2.9±0.20
11	6.4±0.10	3.6±0.10	2.6±0.10	6.8±0.10	2.5±0.40	1.85±0.20	1.6±0.40	3.0±0.20
12	7.3±0.20	37.1±0.10	2.8±0.00	7.9±0.00	2.8±0.50	1.3±0.40	1.7±0.10	3.3±0.30
13	7.3±0.00	4.4±0.40	2.9±0.10	8.0±0.10	2.7±0.20	2.3±0.10	1.8±040	3.5±0.10
14	7.9±0.20	4.4±0.40	2.9±0.10	8.0±0.10	3.2±0.30	2.4±0.10	1.9±0.10	3.8±0.00
15	7.9±0.20	4.4±0.40	3.0±0.10	8.1±0.10	3.2±0.20	2.4±0.00	1.9±0.00	3.8±0.10

Table 5 Some morphological observation made on the tomatoes seedlings (*Lycopersicon esculentum*) on polluted agricultural soil after bioremediation using Pseudomonas *aeruginosa*

Days	- Qualitative morphological properties of tomato seedlings (*Lycopersicon esculetum*)																			
	Stunted growth				Chlorosis of leaves				Chlorosis/etiolation of stems				Leaf folding				Leaf dropping			
	Exptal	Contr1	Contr2	Contr 3	Exptal	Contr1	Contr2	Contr3	Exptal	Contr1	Contr2	Contr3	Exptal	Contr1	Contr2	Contr3	Exptal	Contr1	Contr2	Contr3
0	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	—	NG	NG	NG	NG	NG	NG	NG
3	—	NG	NG	—	—	—	NG	—	—	—	NG	—	—	—	NG	—	—	—	NG	—
6	—	+	+	—	—	—	++	—	—	+	+	—	—	—	—	—	—	+	NG	—
9	—	+	++	—	—	—	++	—	—	+	++	—	—	+	++	—	—	+	++	—
12	—	++	+++	—	—	—	++	—	—	+	++	—	—	++	++	—	—	++	++	—
15	—	++	+++	—	—	—	++	—	—	+	++	—	—	++	++	—	—	++	++	—

- *Legend*

✓ NG = No germination seen yet
✓ — = Absent
✓ + = Present
✓ ++ = Higher percentage present
✓ +++= Highest percentage present

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Table 6 Some Morphological Observations Made on the Tomato Seedlings (*Lycopersicon Esculetum*) Planted on Crude Petroleum Polluted Soil After Biomediation Using *Bacillus Subtilus* [SAMPLE B]

Time in days	Qualitative morphological of tomato seedlings																			
	Stunted growth				Chlorosis of leaves				Chlorosis/etiolation of stem				Leaf folding				Leaf droping			
	Ex ptl	Cont r1	Cont r2	Cont r3	Ex ptl	Cont r1	Cont r2	Cont r3	Ex ptl	Cont r1	Cont r2	Cont r3	Ex ptl	Cont r1	Cont r2	Cont r3	Ex ptl	Cont r1	Cont r2	Cont r3
0	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	-	NG	NG	NG	NG	NG	NG	NG
3	-	NG	NG	-	-	NG	NG	-	-	-	NG	-	-	-	NG	-	-	-	NG	-
6	-	NG	NG	-	-	+	++	-	-	+	+	-	-	-	-	-	-	+	NG	-
9	-	+	+	-	-	++	++	-	-	+	++	-	-	+	++	-	-	+	++	-
12	-	++	++	-	-	++	++	-	-	++	++	-	-	++	+++	-	-	++	++	-
15	-	++	+++	-	-	++	++	-	-	+++	+++	-	-	+++	+++	-	-	+++	+++	-

- *Legend*

✓ NG = No germination seen yet
✓ - = Absent
✓ + = Present
✓ ++ = Higher percentage present
✓ +++= Highest percentage present

IV. DISCUSSIONS

Crude oil contamination affects the agriculture sector particularly due to soil toxicity and reduced plant growth. Crude oil causes environmental risks in the soil ecological system, by inhibition of plant growth, damage to soil structure, disturbance of soil water quality (Han *et al.*, 2016).

The present research was designed to observe the growth response of tomato seedlings in crude oil polluted soil samples remediated with isolates of *Pseudomonas aeruginosa* and *Bacillus subtilis*. It was found that these methods are not only beneficial to remediate the soil but also capable to overcome the toxic nature of crude oil as evidenced by the improved growth performance of the seedlings in comparison with the unpolluted soil. Phytotoxicity study showed that leaf breath, heights, seed germination and leaves lengths of the plants were significantly reduced in the samples planted in control soil relative to the experimental and unpolluted soils. The percentage increase recorded for seed germination and plant height in both experimental soils (*P. aeruginosa* and *B. subtilis* remediated) might be due to an increase in nutrient uptake by the plant and a decrease in crude oil uptake (Mosa *et al.*, 2016). Increased microbial activities resulting in reduced accumulation of crude oil residues made these nutrients available in the state that they were easily assimilated by the test plant. This resulted to the better plant yield indices observed, in terms of percentage seed germination; plant height and plant leaf breath parameters in the experimental soil samples. From the results obtained, the recovery effects were better in the experimental and unpolluted soil samples in contrast to the controls. The result on plant height agrees with previous findings of Ikhajiagbe and Anoliefo (2011) on the significant reduction of plant growth occasioned by oil pollution. Ojimba and Iyagba (2012) reported the decreased output of horticultural crops in crude oil polluted farms as compared with the unpolluted farms.

Various studies have reported the role of these remediation techniques in the improvement of plant growth (Shahid *et al.*, 2017; Laird *et al.*, 2010). Our results also relate with Laird *et al.* (2010), where fresh weight and dry weight increased because of remediation of the soil. Hence it could be inferred that the isolates used in this study may reduce the leaching losses of nutrients by causing retention of nutrients in the soil.

Disturbances of major physiological processes within plants systems due to hydrocarbon contamination are common; and manifest in the forms of morphological, pigment, chlorotic and necrotic foliar patterns alterations. In this study, only tomato seeds grown in experimental soil and unpolluted soil didn't exhibit stunted growth, chlorosis and leaf folding/dropping. For plants grown in contaminated soil, the parameters were not observed completely. This finding is not without precedence as some couples of investigations claimed similar results (Chuku *et al.*, 2018; Okonokhua *et al.*, 2007). Studies by Kekere *et al.* (2011) reported that there was no crop yield recorded in cowpea grown in contaminated soil due to plant mortality. They also observed that crude oil contamination at all intensities resulted in a significant reduction in leaf number when compared with the control treatment in a concentration-dependent manner. Our results correlate with Ali *et al.* (2017), who reported that crude oil adversely affects the plant-water-soil relationship, resulting in chlorosis. The relative plants' heights and the number of leaves were worst affected especially in plants grown in crude oil tainted soils where absence of leaves was observed at the end of the experiments.

V. CONCLUSION

The results of the present research have proven the bioremediation potential of bacterial isolates, for crude oil contamination which also showed significant growth potential for the tomato plants. It enhanced the morphological, physiological, and biochemical parameters of the plant. The *P. aeruginosa* bioaugmented soil showed better indices than *B subtilis;* this again points to the higher potentials of *Pseudomonas aeruginosa* than *Bacillus Subtilis.* From the above observations, the bioremediation process increased the fertility index of the crude petroleum polluted agricultural soils.

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