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Effect of Forest-Incubated Composts on Crude-oil Soils for *Zea mays, L.* Cultivation in Delta State, Nigeria

Anselm Enwelem Egwunatum[1*], Emmanuel Uyovbisere[2], Leticia Chisom Umeh[1]

[1]*Department of Forestry and Wildlife, Nnamdi Azikiwe University, Awka, Nigeria.*
[2]*Department of Soil Science, Ahmadu Bello University, Zaria, Nigeria.*

ABSTRACT

This study examined the potential of forests to provide essential composting edaphic conditions that enhance the microbial density and public health for degradation of crude-oil contaminated soil. Legume (L), woodshaving/poultry dropping (WD), cow dung (CD), legume + woodshaving/ poultry dropping (LWD), legume + cow dung (LCD), woodshaving poultry dropping + cow dung (WCD), and legume + gut of ruminant animals (LGA) were incubated at 3, 6 and 8weeks periods in Forest Reserve to produce compost teas and slurries. Crude oil-contaminated soils were then inoculated with compost formulations and soil restorative capacity was evaluated using Zea may, L. seeds in a-8 x 3 x 2 factorial experiment. Collected data of emergence at 1, 2, and 3 weeks after sowing, stand counts at 4 and 8WAS, plant heights, and leaf areas at 10 and 12WAS were subjected to analysis of variance and Pearson correlation matrix. Results showed that total petroleum hydrocarbon decreased with the incubation period for all treatments to 8 weeks. Stand count of maize for the 6weeks incubated compost was LGA (77.5%) > WCD (52.5%) > LCD (45.0%) > LWD (35.0%) > WD (25.0%) > CD (30.0%) > L (25.0%) at 4weeks after sowing with similar trend in plant height and leaf area at 12 WAS. Soil treated with 6weeks incubated compost significantly correlated with plant emergence as PE3-8 (r =0.83), PE6-8 (r = 0.83) as well as plant height HT-3 (r = 0.88) and HT-8 (r=0.93) at 12 WAS.

Keywords: *Compost age, Incubation period, Total petroleum hydrocarbon, Plant emergence, Stand count*

Corresponding author: Anselm Enwelem Egwunatum
e-mail ✉ aegwunatum@unizik.edu.ng
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INTRODUCTION

Compost production has been trailed by efficacy, environmental health and safety issues over the years as a result of its increasing utilitarian value in combating critical challenges in different facet of the human economy (Domingo, 2002; Bunger *et al*., 2007; Kampa & Castanas, 2008; Conza *et al*., 2013; Di Filippo *et al*., 2020). This is because microbial production, proliferation, and progressive multiplication as novel pathways for potencies and sustained activities in biocontrol frontiers have been fraught with the loss of microbes along with methane gas to endanger public health in the immediate environment and increase greenhouse gas in global warming (Luske, 2010; Ziraba, *et al*., 2016; Ma, 2020; Pang *et al.*, 2020).

Microbial density has been reported as crucial for sustained efficacy in biological control with compost application (Mitlal & Singh, 2009; Jodar *et al*., 2017) even though the ability to maintain microbial population during compost utilization has been related to its types, source, and production pathway since nutrient of compost feedstocks constitute the primary base for multiplication of microbes in composts. Unfortunately, most of the nutrients are not effectively mineralized during composting to yield a commensurate population of microbes due to a lack of favorable environmental conditions, particularly temperature and moisture (Barcauskaite *et al*., 2020; Kabasiita *et al*., 2022). The decline in the compost microbial population was observed with a decrease in available nutrients for sustainability. The report has shown that aerated compost production requires sufficient aeration that is often generated artificially to meet the essential supplies of oxygen, moisture, and temperature for optimal performance decomposition of compost feedstocks (Browne *et al,* 2001; Omotayo *et al*., 2012; Fournel, *et al*., 2019; Rahman, *et al*., 2020).

Delta State Ministry of Environment (2018) reported approximately 2.11 million barrels of crude oil spilled in the Delta State environment as a result of sabotage, failed pumps, corrosion of pipelines, and careless loading of aged pipelines for distribution. Significantly, crude oil spill affects soil structure as it constitutes blockades in the micro and macro pores to displace soil air and lubricate the internal pore linings to resist natural binding properties of clay minerals present essential for optimal biophysical and chemical functioning of the soil, with consequent effect on the water holding capacity of the soil (Phillip & Roberts, 1989). Soils contaminated with petroleum hydrocarbon inhibit microbial and non-hydrocarbon substrate utilization, ATP production, dehydration activity, and nitrogen fixation besides short-term increase in biomass concerning microbial population (Pfaender & Buckley, 1986). The inhibition of critical biological activities such as nitrification and ammonification reactions for productive agriculture has also been observed in hydrocarbon-contaminated soil (Ayade, 2003) with a general increase in carbon-nitrogen ratio.

The existing practice of chemical application of surfactants has been reported as ineffective as a result of inconsistency, uneconomical, unattractive, and unaffordable to poor resource practitioners as a viable remediation approach (Adebusoye *et al.*, 2007; Muthusamy *et al.*, 2008; Brooijmans *et al.*, 2009; Saum *et al*., 2018) which accounted for a significant reduction in the average farm size and available scarce arable land, especially for Zea, *may* and Cassava production in crude oil spills communities (Ogisi *et al*., 2006). Compost materials rich in microbes have been suggested and employed at various times and climes to initiate, increase and induce bio-control actions against deterrent complications in different segments of the bio-physical environment including agriculture (Ermolaev *et al.,* 2019; Sharma *et al*., 2019).

Tropical forests are a notably rich source of microbial varieties and a wide range of ecosystem engineers that detoxifies and modifies leafy, woody as well as non-woody organic components on the forest floor to enrich upper soil profile conditions. High microbial activities related to increased porosity, enhanced water-holding capacity and anti-thermal properties of root exudates on soil particles are indicators of a resilient environment that provide support for other biological programs (Daverey & Pakshirajan, 2009).

This study, therefore, leverages the potential influence of the edaphic biota of a tropical forest as a safe system for nutrient mineralization, infusion, and enrichment as an alternative functional source for composting feedstock since the ingenuity of compost products has been reportedly linked to its source as aerated and non-aerated with the environment at pollution risk while soliciting for microbial activities. It is in line with this, that agricultural wastes known for a wide range of microbial constituents (Sayara *et al*., 2020) were examined as potential compost feedstocks for the generation of specialized spectral microbes that could degrade crude oil-contaminated soil for the cultivation of *Zea may* which is an extensively cultivated cereal in all ecological zones of Nigeria, including crude-oil spill-prone communities in Delta State.

MATERIALS AND METHODS

Description of study area

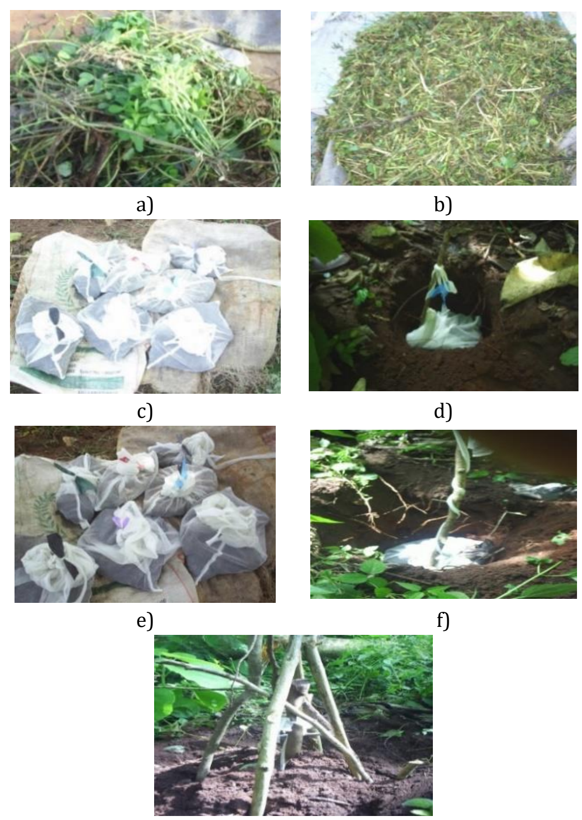
The sampling site of the soil used for the research project was Orogun in Ughelli North Local Government of Delta State. Orogun has the coordinate points of latitudes 06^0 09′ N and longitude 05^0 39′ E and lies in a tropical climate influenced by the movement of Inter-tidal discontinuity (ITD).

The rainfall pattern is bimodal, with peak periods in July and September, and an annual average of between 1600 – 2000mm. the average mean annual temperature is between 25 – 29^{o}C and the mean between the coldest and warmest period is about 4 - 5^{o}C. Relative humidity is high for most of the year with a mean relative humidity of 77% (Nigeria Meteorological Service Station, 2019).

Compost production

The various compost types were prepared under natural conditions at existing air and soil temperature and pressure conditions of the composting site at the Ogwashi-Uku Forest Reserve. The condition of the composting site was quite natural. The forest trees were mostly secondary, with tap roots and the forest floor nearly bare of grasses due to significant canopy cover. This canopy cover also helped to conserve humidity and prevented likely excessive heat from direct sunlight. The average environmental temperature of the composting site was 26 – 28^{o}C. Topography was undulating and the reserve was covered by red sandy soil with a thin humic layer on the surface. The prepared compost feed-stocks were loaded in perforated 2 mm mesh size cotton bags in three replications per compost type, amounting to 21 litter bags. This was to forestall any possibility of interrupting the decomposition time sequence of three, six- and eight weeks during the harvesting of the compost. Furthermore, each of the 2 mm mesh size cotton bags was double glazed with 2 mm mesh size plastic net bags, labeled, and buried separately in 0.50m x 0.50m x 0.50m root-free pits in deep-forested soil at 3m apart to allow for active decomposition as well as for microbial supply under the environmental conditions. The composting site was then fenced off from public entry and the possibility of soil compacting by straying grazing animals.

Composts were carefully harvested at ages 3, 6, and 8 weeks respectively to produce compost slurries by weighing 3 kg of each compost type into several containers and then mixed with 4litres of tap water. These were allowed to incubate for 7 days at room temperature and stirred occasionally every two days. Compost teas were generated by filtering through a 2 mm mesh size before storage at 4^{o}C and allowed to equilibrate with screen house temperature before application on crude oil contaminated soils.



a) b) c) d) e) f) g)

Figure 1. Compost feedstock and composting method in Ogwashi-Uku lowland rainforest Reserve.

Contamination procedure

Six kilograms of soil were weighed out respectively into seventy-two polythene pots, with drainage holes at the bottom, plugged with cotton wools to retain the soil. Soil samples were then contaminated with 140mls of crude oil at the rate of 23.33 ml/kg and then left for 1 week to allow for maximum reaction. Twenty grams of compost slurries were then weighed and mixed with the contaminated potted soils. Then 15ml of compost teas of the respective formulations were repeatedly applied to the contaminated soil at 2 weeks intervals, on days 7 and 21 of the month for a 9months period. Pots were watered regularly to keep the soil at field moisture capacity throughout the experiment. The control treatment was crude oil-contaminated potting soil at the same rate but was treated with distilled water.

Determination of TPH

Soil samples at three and nine months after application of the respective incubated compost formulations were analyzed for total petroleum hydrocarbon (TPH) using the Ultra-Violet (UV) visible spectrophotometer to determine the extent of degradation after the application of the various compost treatments. The wavelengths obtained were used to photometrically determine the concentration of crude oil (TPH) in each sample.

Germination experiment

The *Zea may* seeds that passed the viability test by the Flotation method were selected for use in the germination experiment at four maize (*Zea may*) seeds per germination troughs in 12cm^2 enforcement. Percentage emergence was assessed after one, two, and three weeks of sowing and calculated. The stand count, which is an indication of the progressive efficacy of compost on contaminated soil after the initial emergence of Zea may, was monitored throughout the growth using plant heights and leaf areas at 10 and 12 weeks after sowing (WAS) respectively. Maize (*Zea mays*) has a strong exhausting effect on soils as a high feeder (Jaliya *et al*., 2006) and is therefore a good test crop for soil fertility, especially for crude oil contaminated soils.

Average leaf widths of three representative leaves per plant were also taken at 10 and 12WAS from the top, middle, and base of the plant. Length from the basal point of the leaf to the tip of the leaflet and width of the widest part of the leaf was measured with a rule. Leaf area was computed from the length and breadth measurements of the longest leaf per plant and multiplied by a correction factor of 0.75. The leaf area per plant at 10 and 12WAS was obtained by multiplying the leaflet area by the number of leaves.

Experimental design and analyses

The experiment involved eight (8) treatments that comprised seven compost formulations and control on the stand counts, synthesized from three (3) incubation ages for growth assessment under two (2) periods of 10 and 12 weeks after sowing using plant heights and leaf areas, arranged in a completely randomized design. Thus, the experiment was carried out in 8 x 3 x 2 factorial in a complete randomized design. Data collected were subjected to analyses of variance (ANOVA) and significant means separated by the Duncan Multiple Range Test (DMRT) at a 5% level of probability.

RESULTS AND DISCUSSION

Effect of compost on TPH

The effects of various lowland rainforest incubated compost formulation types and ages on the degradation of crude oil-contaminated soil are shown **(Figures 2 and 3)**. There were significant differences (p>0.05) in the reductions by the 3, 6, and 8weeks incubation composts. The legume + gut of ruminant animal (LGA) compost type were consistent for the 3 weeks and 6 weeks incubation periods and efficient in reducing the total petroleum hydrocarbon of the contaminated soils, even further than was achieved at 3 months reaction periods. For the 8 weeks of incubated composts, the legume + gut of ruminant animal (LGA) compost stimulated the highest percentage reduction in total petroleum hydrocarbon of 67.9%, consistent with the trend at 3 months.

There was a decline in the efficacies of compost formulations after 6 weeks of incubation at both 3 and 9 months evaluation periods depicted by a high percentage reduction of total petroleum hydrocarbon.

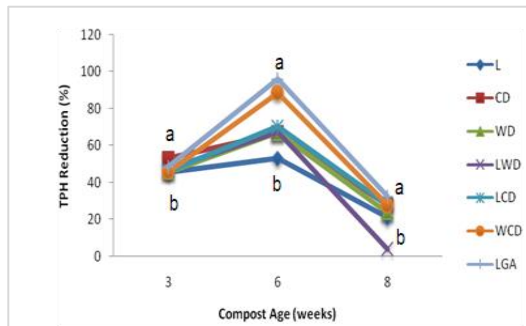


Figure 2. Comparative reduction in TPH of contaminated soils as influenced by lowland rainforest incubated compost formulations at 3 months (Means of the same age with the same alphabet are not significantly different at a 5% level of probability).

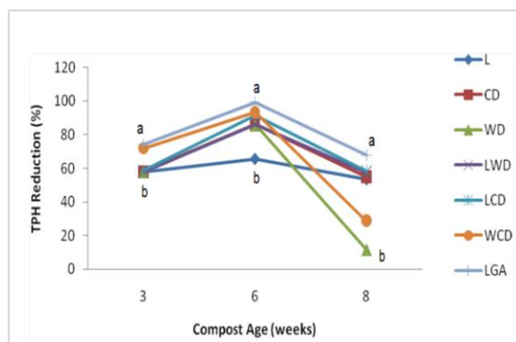


Figure 3. Comparative reductions in TPH of contaminated soils as influenced by lowland rainforest incubated compost formulations at 9 months (Means in the same age with the same alphabet is not significantly different at a 5% level of probability).

Germination at 1week after sowing

Results of compost formulations incubated over the periods showed significant differences ($p \leq 0.05$) in the percentage emergence of *Zea may* **(Figure 4)**. The compost formulation from the 3weeks incubated woodshaving/poultry dropping + cow dung (WCD) recorded the highest emergence (7.50%) at 1week after sowing. The 6weeks woodshaving poultry dropping + cow dung (WCD), legume + gut of ruminant animal (LGA), and legume + woodshaving/poultry dropping (LWD) showed the highest emergence of 17.5%. There was no germination by Legume formulation with the 6 weeks materials 1 week after sowing. There was no emergence with the 8weeks composts and the control was treated with distilled water.

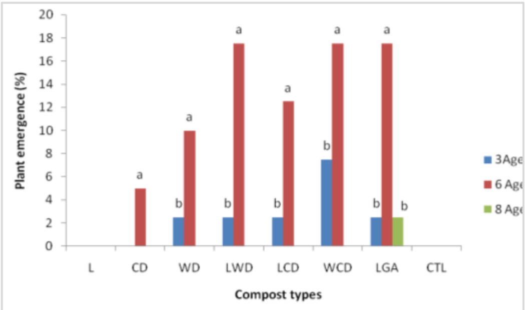


Figure 4. Effects of compost formulations on the emergence of maize at 1 week after sowing (Means in the same treatment with the same alphabet are not significantly different at a 5% level of probability using DMRT)

Germination at 2 weeks after sowing

The 3-week incubated compost revealed that the legume + gut of ruminant animals (LGA) had the highest emergence of 12.50% and the legume (L) was 2.50% **(Figure 5)**. The *Zea may* emerge with 6 weeks of compost ranging from 7.50 - 30%. Woodshaving/poultry dropping + cow dung (WCD) recorded the highest emergence (30%) and legume (L) the least. Besides the performance, all other formulations were not significantly different across compost formulations. There was no emergence of *Zea may* in the legume (L), cow dung (CD), and woodshaving/poultry dropping (WD) with the 8weeks incubated composts as only the Legume + cow dung (LCD) induced the highest emergence of 5%.

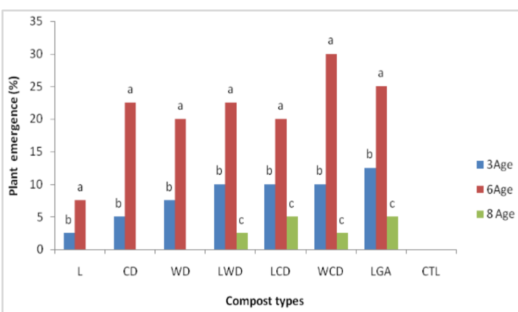


Figure 5. Effects of compost formulations on the emergence of maize at 2 weeks after sowing (Means in the same treatment with the same alphabet is not significantly different at a 5% level of probability using DMRT)

Germination at 3 weeks after sowing

The legume + gut of ruminant animal (LGA) and cow dung (CD) of the 3weeks incubated compost recorded the highest mean emergence (17.50%) at 3 weeks after sowing **(Figure 6)**. The LCD and LWD recorded 15% emergence respectively while the least percentage emergence (10%) was shown by legume (L) and woodshaving/poultry dropping (WD). The control treatment did not support any emergence at 3weeks after sowing. However, the percentage emergence of the 6weeks incubated composts was LGA (47.50%) > WCD (37.50%) while the 8weeks incubated compost formulation values ranged from 2.5 – 10%.

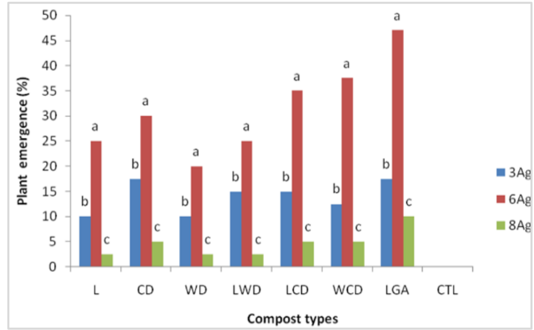


Figure 6. Effects of compost formulations on the emergence of *Zea may* at 3 weeks after sowing (Means in the same treatment with the same alphabet are not significantly different at a 5% level of probability using DMRT)

Stand count at 4 weeks after sowing

The stand count was 15-22.5% and 2.50-15.0% for the 3-and 8-week compost formulations respectively at 4 weeks after sowing **(Figure 7)**. Stand count was 25-77.5% for the 6weeks compost formulation with legume + gut of ruminant animal (LGA) > 52.5% Woodshaving /poultry dropping + cow dung (WCD) > 45% legume + cow dung (LCD).

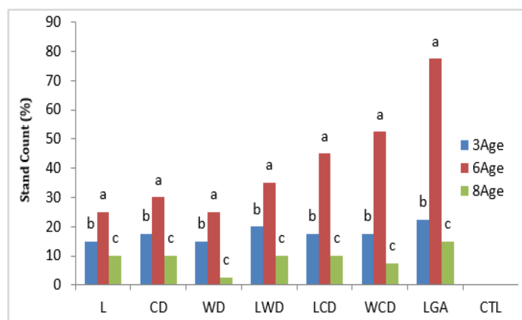


Figure 7. Effects of compost formulations on stand count of maize at 4 weeks after sowing (Means in the same treatment with the same alphabets is not significantly different at a 5% level of probability using DMRT)

Stand count at 8 weeks after sowing

Results showed that the stand count of *Zea may* with 3-and 8-week compost formulations was 15 – 30% and 10 – 37 % respectively while the 6 weeks materials ranged from 42% to 100% at 8 weeks after sowing **(Figure 8)**. Legume +gut of ruminant animal (100%) > woodshaving/poultry dropping + cow dung (97%) > legume + cow dung (92%) > Woodshaving/poultry dropping (75%) > legume + woodshaving/poultry dropping (72%) > cow dung (65%) > Legume (L) with 42%. The LGA and WCD significantly induced 100% emergence and growth of the *Zea may* be cropped on the remediated soils.

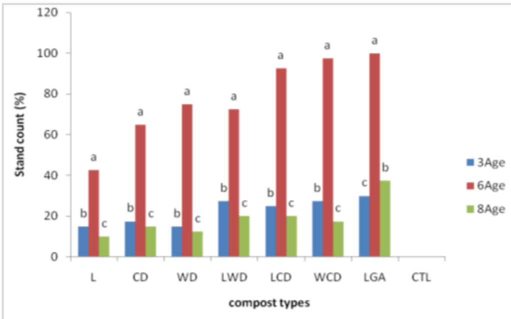


Figure 8. Effects of compost formulations on stand count of *Zea may* at 8 weeks after sowing (Means in the same treatment with the same alphabet are not significantly different at 5% level of probability using DMRT).

Correlation matrix of agronomic parameters and soil physical properties

The correlation matrix of selected agronomic parameters with the physical properties of compost-remediated soil is shown **(Table 1)**. Bulk density enhanced all the agronomic parameters measured.

Table 1. Correlation matrix of selected agronomic parameters with physical properties of crude oil contaminated soils remediated with compost formulation of 3-, 6- and 8-week incubation periods

Variables BD3	BD6	BD8	PO3	PO6	PO8	PE33	PE63	PE83	HT3	HT6	HT8	LA3	LA6	LA8
BD3	0.09ns	0.04ns	0.19ns	0.07ns	0.01ns	-0.30ns	-0.41ns	-0.33ns	-0.21ns	-0.40ns	-0.12ns	-0.23ns	-0.20ns	-0.31ns
BD6		0.73*	-0.35ns	-0.82*	-0.82*	-0.76*	-0.83*	-0.83*	-0.95**	-0.72*	-0.94**	-0.76*	-0.83*	-0.91**
BD8			-0.32ns	-0.95**	-0.96**	-0.81*	-0.85**	-0.55ns	-0.87**	-0.68ns	-0.87*	-0.93**	-0.76*	-0.88**
PO3				0.25ns	0.24ns	0.07ns	0.31ns	0.18ns	0.37ns	0.10ns	0.21ns	0.31ns	0.23ns	0.34ns
PO6					0.99**	0.83*	0.83*	0.60ns	0.88**	0.64ns	0.93**	0.90**	0.77*	0.89**
PO8						0.84*	0.86*	0.62ns	0.90**	0.66ns	0.94**	0.92**	0.78*	0.90**
PE38							0.93**	0.87**	0.85**	0.93*	0.84**	0.94**	0.94**	0.88**
PE68								0.83*	0.92**	0.86**	0.87**	0.95**	0.87**	0.96**
PE88									0.81*	0.91*	0.75*	0.75*	0.94*	0.81*
HT3										0.79*	0.97**	0.90**	0.89**	0.98**
HT6											0.71*	0.82*	0.94**	0.79*
HT8												0.87**	0.84**	0.96**
LA3													0.88*	0.94*
LA6														0.88*

Note: BD3, BD6 & BD8 =Bulk Density of the 3, 6 & 8weeks incubation periods; PO3, PO6 & PO8 =Porosity of the 3, 6 & 8weeks incubation periods; PE3, PE63 & PE83
=Plant Emergence of 3, 6 & 8weeks incubation periods at 3 weeks after sowing; HT3, HT6 & HT8 = Plant Heights of 3, 6 & 8 weeks incubated compost at 12 weeks after sowing;
LA3, LA6 & LA8 =Leaf Areas of 3, 6 & 8 weeks incubated compost at 12 weeks after sowing.
= Significant at 0.05 level of probability   **= Highly significant at 0.05 level of probability   ns=Not significant at 0.05 level of probability

There was no significant correlation between the agronomic material and the soil bulk density treated with 3 weeks of incubated compost, unlike the 6 weeks of compost. All the agronomic parameters under investigation significantly correlated with the bulk density of the 6 weeks incubated materials.

Notably, the bulk density of the 6weeks incubated materials (BD6) was highly correlated with plant heights of *Zea may* at 12 weeks after sowing. The soil treated with 6weeks incubated composts were equally significantly correlate with plant emergence as PE38 (r =0.83*) and PE68 (r = 0.83*) as well as plant height HT3 (r = 0.88**) and HT8 (r=0.93*) at 12 weeks after sowing. It also significantly correlated with the leaf areas of *Zea may* on the bio-remediated soils with the 3-, 6- and 8-weeks incubated materials. However, properties that did not significantly correlate with others showed that they vary independently, with the negative and positive signs associated with such properties and parameters suggesting the direction of such effects under investigation.

The 6weeks LGA compost formulation demonstrated high potential for degradation of contaminated soils for the emergence of maize at 2 weeks after sowing compared to its 3- and 8-week types. However, LGA, LWD, and WCD exhibited the same potential for the emergence of maize at 1 week after sowing which narrowed to LWD and LGA by 2 weeks after sowing, and eventually dove-tailed to just LGA with 47.5% emergence by 3 weeks after sowing. This may not be unrelated to the pH of the 6weeks incubated compost formulations that all tended toward alkalinity, especially that of LGA. This finding agrees with Hassan *et al.* (2006) that soil reaction plays an important role in the solubility of nutrient elements which probably were locked up by contaminated crude oil molecules but became available for the growth of *Zea may* through microbial degradation. These released nutrients increased the sustainability of *Zea mays* soil toxicity while enhancing stand count (Pasha *et al*., 2020).

The study further showed that high soil acidity often associated with crude oil contamination was corrected to a point that enhanced the increased uptake of nutrient elements especially phosphorous which may have increased the growth and development of *Zea may*. But beyond a point, vegetative growth of *Zea may* be drastically reduced as it translated to decreased uptake of several nutrient elements, especially phosphorus. This is in agreement with the trend of the percentage emergence of *Zea may,* especially the 8weeks incubated compost to further authenticate that incubating compost formulations beyond 6 weeks diminished the potency of formulations to degrade crude oil molecules for favorable germination and stand count of *Zea may*. This may have equally accounted for the poor performance of the 8weeks incubated compost formulations as there was a drastic reduction in the soil potential to support maize emergence.

The results showed a gradual increase in the relationship between age and decomposition which seemed to reach a maximum at 6 weeks of incubation. So that despite the P-availability in the soil as reflected in L, CD, WD, LWD, LGA, LCD, and WCD of the 8weeks incubated compost formulations, the various P-adsorption capacities may have been counteracted by the low pH status for plant uptake (Isirimah *et al.*, 2010) as a result of the inefficacies of compost formulations at this incubation period. Consequently, the efficacies at 8 weeks incubation were probably inert since a certain level of P in the formulations appeared not maintained.

The 6weeks incubated compost formulations had the highest stand count while the legume showed the least stand count. The LCD and WCD doubled up with LGA while WD, legume + LWD, and CD showed similarity in the stand count of *Zea may*. These differences in performance even within the same incubation period may be due to the selective response of contaminated soils to the different microbial make-up as well as the density of compost formulations is not only degrading the hydrocarbons, but also acting as an organic source of fertilizer (Landgraf & Klose, 2002) in association with the bulk density of the crude oil contaminated soils as influenced by the respective compost formulations, both in age and type.

The LGA of 6 weeks incubated compost with the least bulk density as well as highest porosity had the highest degradation efficiency compared to the control with bulk density and porosity respectively. These observed differences in the bulk density may not be unconnected with the differences in the capacity to aggregate the soil particles which was best for the legume + gut of ruminant animals at 6 weeks incubation and least for the control treatment application.

The plant height and leaf areas of 6weeks compost formulations showed a similar pattern with a progressive increase in plant height of *Zea may* as well as the leaf areas for the LGA and WCD. These increments may be due to the gradual buildup of soil nutrients after remediation with compost formulations probably as a result of the wider microbial spectrum and population of these dual source compost feedstocks with the animal organic microbes. This may not be unrelated to the likely higher microbial density in the guts of ruminant animals compared to cow dung (Barcauskaite *et al*., 2020). However, at 8 weeks after sowing, only the control treatment failed to support the emergence of *Zea may*. This indicated that the water applied could not elicit degradation of the contaminated crude oil molecule unlike the various compost formulations, especially the 6weeks incubated compost.

CONCLUSION

Forest incubated compost formulations had significant effects on the degradation of crude-oil contaminated soil with the potential to reduce public health risks and provide better-composting pathways. The 6weeks compost formulations had superior degradation efficacies with Legume + gut of ruminant animal (LGA) compost formulations as highest degradation efficacy of crude oil molecule for the highest plant emergence and stand count of *Zea may* cultivation in pursuit of land reclamation. Furthermore, the study highlighted the efficacy of animal-plant multiple-source compost over either animal or plant single-source organic-based compost formulation types even at the same optimal 6weeks incubation period in the bio-remediation of crude oil contaminated soils for *Zea may* growth and development.

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ETHICS STATEMENT: None

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