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Impact of Drifts Resulting From Pesticide Application on Soil Microorganisms around Waste Receptacle in Port Harcourt

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Abstract— The aim of this study was to assess the impact of drift resulting from pesticide application on soil microorganisms around waste receptacle in Port Harcourt. The results showed that mean Total Heterotrophic Bacteria counts reduced after the application of pesticides (3.82×10^8cfu/g) when compared to soils of the control (2.7×10^9cfu/g) and before (1.02×10^9cfu/g) the application of pesticides. The mean Total Heterotrophic Fungi counts also reduced after the application of pesticides (6.8×10^5cfu/g) when compared to soils of the control (2.1×10^6cfu/g) and before (2.8×10^6cfu/g) the application of pesticides. Identification of bacterial isolates indicated that *Staphylococus aureus*, *Bacillus subtilis*, *Bacillus megaterium*, *Pseudomonas sp*. and *Micrococcus sp.*, had the highest percentage frequency occurrence of 15.4% while *Bacillus cereus*, *Bacillus lichenformis* and *Escherichia coli* had the lowest percentage frequency of 7.7%. After the application of pesticides, Bacillus *cereus*, *Bacillus lichenformis* and *Escherichia coli* which were present in soils before the application were absent. Fungal isolates such as *Aspergillus niger, Penicillium sp, Fusarium siculi, and Aspergillus fumigatus* all had frequency occurrence of 18.2% respectively while *Aspergillus nidulas, Microsporium canis* and *Yeast* had frequency occurrence of 9.1% receptively. *Aspergillus nidulas*, *Microsporium canis* and *Yeast* were also absent in soils after the application of pesticides. It was recommended that proper surveillance during pesticide application should be carried out to avoid drift to non-target organism and that concentrations of such pesticides should be taken into account to avoid reduction in the number of microorganism in soils because of the vital roles they play in maintaining soil health.

Index Terms— bacteria, drifts, fungi, human health hazards, pesticide application, Port Harcourt, waste receptacle,

I. Introduction

Pesticides are chemicals that destroy, kill, restrain or prevent life forms that are of nuisance value in the environment [1]. The benefits of pesticide are numerous in crop and pest management such as application in waste receptacles and agricultural fields. However, they can also impact negatively on the environment causing pollution if use is not properly controlled [2]. Pesticides drive away pests or prevent their development and are toxic in nature. Many of the pesticides are broad spectrum in nature and are referred to as biocides.

Pesticide drift involves the application of pesticides to produce droplets which can either remain suspended in the air and or may be carried away by wind from the intended target area. Pesticide drift can also be defined “the physical movement of pesticides through the air at the time of pesticide application or soon thereafter from the target site to any non-or-off target site” [2].

Drifts may occur during the application of droplets thereby causing droplets or residues to migrate from the intended target site hence causes some chemicals to turn into vapour which may either move away from the target site of application, thereby, causing unwarranted exposure to people, animals, plants and property and the entire environment.

When pesticides are applied and drift occurs it can

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pose a lot of health hazards to people and pets because these sprays and residues drift to nearby areas such as homes, schools, and playground including wildlife and plants which are also at risk, as a result, drift affects natural areas and water sources.

Human health hazards emanating from the application of pesticides and its resultant residue drifts involves short – term impacts and long-term impacts. Impacts from short-term includes; headaches and nausea while impact from long term which is also known as chronic impact includes; cancer, reproductive harm, and endocrine disruption. The effect from chronic health impact may occur many years later even when the exposure of pesticide was minimal in the environment or result from the pesticide residues which we ingest through our food and water thereby, producing negative impacts [3].

However, this study sought to investigate the impact of drift resulting from pesticide application on soil microorganisms found around waste receptacle in Port Harcourt.

II. Statement of Problem

The methods of collection and disposal of solid waste in most developing cities like Nigeria, is highly inadequate [4], [5]. Waste receptacles have been known to harbour pests, microorganisms and also alter the aesthetics of the environment. These pests and microorganisms enter into nearby homes, thus creating the need for disinfection and management of pest around the waste receptacles. When pesticides are applied around the waste receptacle, it leads to residues of pesticides drifting from the target area to nearby environments such as soil thereby affecting soil microorganisms which are indicators of soil health. These microorganisms are responsible for maintain the physiological status of the soil and when altered can lead to reduction in the functioning of soil. However, there is need to minimize drift resulting from pesticide application.

III. Objectives of the Study

1. To enumerate the number of soil microorganisms around waste receptacle.
2. Characterize the possible bacteria and fungi in soils around the waste receptacle before and after the introduction or application of pesticide.

IV. Description of Study Area

The locations of the waste receptacle and the co-ordinates using the GPS are given in Figure 1. The waste receptacle is located at Market Junction by Ikwerre Road in Mile IV Diobu in Obio/Akpor LGA and the geographical positioning system is between latitudes 4°82ˈ71"N and longitudes 6°98ˈ14"E for the waste receptacle and latitudes 4°82ˈ69"N and longitudes 6.98ˈ99"E for the control.

The waste receptacle is situated close to markets, churches, schools, borehole, restaurant and shops. The vegetation consist mainly grasslands and surrounded by surface waters with drainage system/gutters that drives the leachates into the surface water.

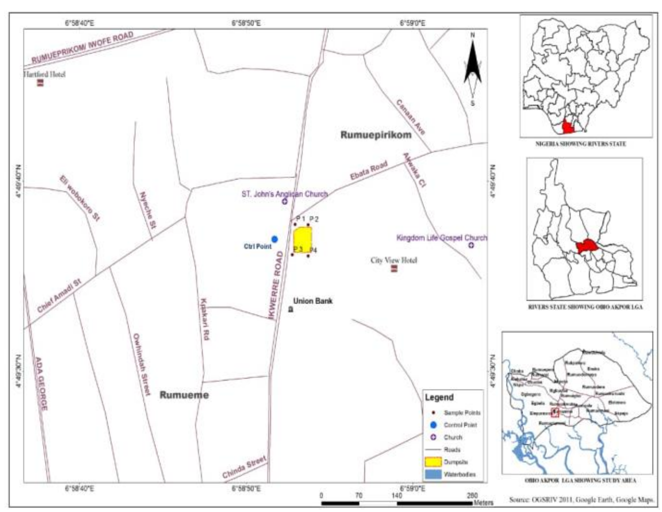

Figure 1: Map showing Port Harcourt as the study Area

V. Review of Past Literatures

[6] observed that pesticides may adversely affect the proliferation of beneficial soil microbes and their associated biotransformation process in the soil and inactivate nitrogen fixing and phosphorous- solubilizing microorganism. They can also reduce activities of soil enzymes which are key indicators of soil health.

[7] also discovered that any alteration in the activities of soil microorganisms due to applied pesticides eventually leads to the disturbance in soil ecosystem and loss of soil fertility.

[8] discovered that pesticides application may also inhibit or kill certain group of microorganisms and outnumber other groups by releasing them from the competition.

VI. MATERIALS AND METHODOLOGY

A. Sample Type, Collection and Preparation

Samples were collected from the soil around the waste receptacle before application of pesticides. Samples were then collected 14 days after the application of pesticides to enable the percolation of pesticides into the soil and Control samples were obtained 50m away from the waste receptacle where the pesticides were applied. Soil samples were obtained from different depths of 0 – 15cm, 15 – 30 cm, 30 – 45cm and 45 – 60cm using a hand auger from four different points around the waste receptacle. Samples were labelled A, B, C, and D while the control was CP1, CP2, CP3 and CP4. Using a hand-auger, approximately 1 inch of the top of the surface soil was discarded as this represented materials collected before penetration of the sample depth [9]. The same sample depths of soils from four different points around the waste receptacle were homogenized to form a composite soil mixture and were labelled P1, P2, P3 and P4 while that of the control was not homogenized. P1 represented homogenized soils from sample depths 0-15cm from the four different points (A, B, C and D); P2 represented homogenized soils from sample depths 15-30cm from the four different points (A, B, C and D); P3 represented homogenized soils from sample depths 30-45cm from the four different points (A, B, C and D) and P4 represented homogenized soils from sample depths 45-60cm from the four different points (A, B, C and D). A total of eight samples of soils (P1, P2, P3, P4, CP1, CP2, CP3 and CP4) were transferred directly into well labelled sterilize glass bottle containers and capped secured tightly to minimize contamination and was taken to the laboratory for analyses.

B. Analytical Methods

i. *Microbiological Analysis:* Heterotrophic plate count was performed using spread plate technique. Aliquots of 0.1ml of selected dilutions were inoculated separately on duplicate Nutrient Agar plates. The inoculated plates were incubated at 37°C for 24 hours and colony forming units counted thereafter; taking cognizance of the dilution factor. Unit of measurement was colony forming units (cfu) per gram of soil. For Fungi, aliquots of 0.1ml from the serial dilutions were transferred onto sterile dry potato dextrose agar plates and spread evenly using a sterile spreader and incubated in an incubator at 25ºC for 5 days. Fungal colonies of all the dilutions were enumerated and tabulated. The fungal counts were performed by Misra and Miles drop plate method. Each dilution was plated in triplicate and the experiment was repeated 3x.

Characterization and identification of bacteria isolates was performed by Gram stain, spore staining, motility test, catalase test, citrate utilization test, indole production test, hydrogen sulphide production test, urease test, oxidative test, methyl red and voges-proskaeur test, triple sugar iron test and sugar oxidation fermentation test.

VII. RESULTS AND DISCUSSION

The results of the effect of pesticide application and its drift at waste receptacle in Port Harcourt are presented in Tables

1 – 3.

Table 1

Microbial Counts of Isolates from Soil Treated with pesticides in Port Harcourt

Application	Sample Depths	THB (cfu/g)	Mean (cfu/g)	THF (cfu/g)	Mean (cfu/g)
CONTROL	CP1 (0 – 15cm)	2.83×10^9	**2.7×10^9**	2.4×10^6	**2.1×10^6**
	CP2 (15 – 30cm)	2.63×10^9		1.8×10^6	
BEFORE	BP1 (0 – 15cm)	1.28×10^9	**1.02×10^9**	1.9×10^6	**2.8×10^6**
	BP2 (15 – 30cm)	0.55×10^9		2.6×10^6	
	BP3 (30 – 45cm)	2.08×10^9		6.0×10^6	
	BP4 (45 – 60cm)	0.20×10^9		1.8×10^6	
AFTER	AP1 (0 – 15cm)	4.1×10^8	**3.82×10^8**	1.0×10^6	**6.8×10^5**
	AP2 (15 – 30cm)	3.7×10^8		0.4×10^6	
	AP3 (30 – 45cm)	4.6×10^8		0.6×10^6	
	AP4 (45 – 60cm)	2.9×10^8		0.7×10^6	

KEY:

CP1- Control at 0-15cm soil depth

CP2- Control at 15-30cm soil depth

CP3- Control at 30-45cm soil depth

CP4- Control at 45-60cm soil depth

BP1- Before pesticide application at 0-15cm soil depth

BP2- Before pesticide application at 15-30cm soil depth

BP3- Before pesticide application at 30-45cm soil depth

BP4- Before pesticide application at 45-60cm soil depth

AP1- After pesticide application at 0-15cm soil depth

AP2- After pesticide application at 15-30cm soil depth.

AP3- After pesticide application at 30-45cm soil depth.

AP4- After pesticide application at 45-60cm soil depth.

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A. Total Heterotrophic Bacteria (THB)

The mean Total Heterotrophic Bacteria (THB) counts in soils before application of pesticide show that 1.02×10^9cfu/g was obtained before application while 3.82×10^8cfu/g was recorded after application. The control was 2.7×10^9cfu/g and this was higher than the value obtained before and after application (Table 4.1). This showed that bacterial count reduced comparatively. This is to say that pesticides affect microbial biomass. However, from this study, pesticides altered the soil microbial population, both qualitatively by reducing the top soil quality and quantitatively by reducing the number of microorganisms and this may in turn affect soil health and physiological status of their habitat. This result corroborates with [10] who observed that the application of pesticides inhibits or kill certain group of microorganisms. Furthermore, [11] showed in their study that application of ½ litre and 1 litre of pesticide also led to a major decline in the bacterial population that may lead to a decline in the soil fertility of farmland. Majority of microorganisms that are found in these waste receptacles derived their nutritional requirement from the waste; hence, accounts for the high bacterial growth profile [12]. High bacterial count recorded occurred before the application of pesticides and reduced after the application of pesticide. The microbial biomass is considered as an indicator of soil health [12] and a useful indicator of the improvement or the degradation of grounds [13].

B. Total Heterotrophic Fungi (THF)

The mean Total Heterotrophic Fungi (THF) counts in soils before application of pesticide show that 2.8×10^6cfu/g was obtained, 6.8×10^5cfu/g was recorded after application and 2.1×10^6cfu/g was for the control (Table 4.1). There was a reduction in Total Heterotrophic Fungi, however, this was as a result of pesticides that were applied around waste receptacles which caused a reduction in Total Heterotrophic Fungi thereby affecting the microbial biomass as well as affected the soil health. This finding is in line with [11] that conducted a study on effect of pesticide on soil microbial spectrum and observed that the number of fungi species present in the sampled soils reduced drastically due to injection of half litre (1/2) of pesticide, hence *Heruncola grisea* and *Alternonia terins* which are usually present in the soil in little quantity were totally extinct when compare with the control samples.

Table 2: Biochemical Characteristics of Bacterial Isolates from Soil samples treated with Pesticide

Isolates	Colonial/morphology	Gram Reaction	Cell Morphology	Oxidase	Catalase	Methyl Red	Vogues Proskauer	Indole	Citrate	Triple Sugar Iron	Motility	Sucrose	Glucose	Fructose	Lactose	Probable Identity	F	F%
1	Gold-yellow, Small, Moist, Opaque round colony	+	Cocci	+	+	+	+	+	_	A	+	_	A	A	A	*Staphylococcus aureus*	2	15.4
2	Milk color, irregular, Opaque, undulated colony	+	Rod	-	+	+	+	_	+	A	+	_	A	_	_	*Bacillus subtilis*	2	15.4
3	Light green, Round, Moist, Opaque colony	-	Rod	+	+	+	_	_	_	_	+	_	_	_	_	*Pseudomonas sp.*	2	15.4
4	Yellow small, round, opaque, colony	+	Cocci	+	+	+	_	+	+	_	+	_	A	A	A	*Micrococcus sp.*	2	15.4
5	Milk color, irregular, opaque, rough surface, large colony	+	Rod	+	+	+	_	+	+	_	+	A	A	A	_	*Bacillus cereus*	1	7.7
6	Milk color, large, flat edged, moist, Opaque colony	+	Rod	_	+	+	_	+	_	_	+	_	A	A	_	*Bacillus megaterium*	2	15.4
7	Milk color, dry surface, filamentous, raised, opaque colony	+	Rod	+	+	+	+	_	_	_	+	A	A	A	A	*Bacillus lichenformis*	1	7.7
8	Large, Circular, convex, greyish white, moist, smooth and opaque colony	-	Rod	+	+	+	_	+	+	A	_	_	A	A	A	*Escherichia coli*	1	7.7

Key:

- = negative F= Frequency of occurrence

+ = positive

A = Acid

C. Biochemical Characteristics of Bacterial Isolates

The bacterial isolates that were identified from soils treated with pesticides around waste receptacle were *Staphylococus aureus*, *Bacillus subtilis*, *Bacillus megaterium*, *Pseudomonas sp*, *Micrococcus sp.* (Table 4.2). *Bacillus cereus*, *Bacillus lichenformis* and *Escherichia coli* were only present in soil samples before application but were not regular after application of pesticides on the soils (Table 4.2). Table 4.2 showed the distribution and percentage occurrence of bacteria isolates and this revealed that *Staphylococus aureus*, *Bacillus subtilis*, *Bacillus megaterium*, *Pseudomonas sp*, *Micrococcus sp.* all had frequency occurrence of 15.4% respectively. *Bacillus cereus*, *Bacillus lichenformis* and *Escherichia coli* had frequency occurrence of 7.7% receptively. Gram positive and gram negative bacteria isolates were identified from soils before and after application of pesticides around waste receptacle. The predominant bacterial isolates include the following: *Staphylococus aureus*, *Bacillus subtilis*, *Bacillus megaterium*, *Pseudomonas sp*. and *Micrococcus sp.*, which had the highest percentage frequency occurrence of 15.4% while *Bacillus cereus*, *Bacillus lichenformis* and *Escherichia coli* had the lowest percentage frequency of 7.7%. After the application of pesticides, microorganisms such as *Bacillus cereus*, *Bacillus lichenformis* and *Escherichia coli* were not present in soils but were present before the application of pesticides and these microbes produce enzymes like DNase, Hyluronidase, staphylokinase, staphylolysin, streptokinsase among others that help degrade waste materials at receptacle sites [14]. This is to say that pesticide application reduces the capacity of the soil microbes in degrading waste. The observation is in line with that of [15] who identified the presence of *Bacillus, Staphyloccocus and Klebsiella* from a waste dumpsite located at Eagle Island, River state. The presence of these identified organisms in the study site is a thing of great concern as these bacteria have been associated with a number of public health problems [16]. Also the findings corroborates with [6] who observed that pesticides can also reduce activities of soil enzymes which are key indicators of soil health.

The distribution and frequency of occurrence of bacteria also revealed that pathogenic bacteria such as *Staphylococus aureus*, *Bacillus subtilis*, *Bacillus megaterium*, *Pseudomonas sp*, *Micrococcus sp.*, *Bacillus cereus*, *Bacillus lichenformis* and *Escherichia coli* present in the waste dumpsite as observed in this study was not surprising, as it concurs with that [17] who reported to have identified the presence of coliforms, faecal coliforms and pathogens such as *Escherichia coli, Pseudomonas spp* and Salmonella from samples collected close to sewage sites.

Table 3

Morphological Characteristics of fungi from soil treated with pesticides

Isolates Codes	Morphological Characteristics	Microscopic Characteristics	Probable Organisms	F	F%
1.	Black cotton growth, white boarder with dark brown reverse side.	Septate hyphae, road head conidia and spores	*Aspergillius niger*	2	18.2
2.	Green cotton radial growth with white boarder and dark brown reverse	Non septate branding hyphae and chain-like conidia. No spores	*Penicillium sp.*	2	18.2
3.	Brown cotton growth with brown reverse	Septate hyphae, round head conidia spores	*Aspergillius nidulas*	1	9.1
4.	White fluffy colony with yellow reverse side	Branding septate hyphae, no conidia head, no spores.	*Fusarium siculi*	2	18.2
5.	White fluffy growth with white reverse side	Septate branding hyphae, no conidial head, spores present	*Microsporium canis*	1	9.1
6.	Green colony with white boarder ad dark brown reverse side	Septate hyphae, columnar head, spores present. Vesicle present.	*Aspergillus fumigatus*	2	18.2
7.	Mucoid, milk colour opaque, elevated round colony	Coccoid shape	Yeast	1	9.1

Key: F= Frequency of occurrence

D. Biochemical Characteristics of Fungal Isolates

The fungal isolates that were identified from soils treated with pesticides around waste receptacle were *Aspergillus niger, Penicillium sp, Fusarium siculi, and Aspergillus fumigatus* (Table 4.3). *Aspergillus nidulas, Microsporium canis* and *Yeast* were only present in soil samples before the application of pesticides but were not regular after application of pesticides on the soils (Table 4.3). The distribution and percentage occurrence of fungal isolates shown in Table 4.3 revealed that *Aspergillus niger, Penicillium sp, Fusarium siculi, and Aspergillus fumigatus* all had frequency occurrence of 18.2% respectively. *Aspergillus nidulas, Microsporium canis* and *Yeast* had frequency occurrence of 9.1% receptively. The most predominant fungal isolates that were identified from soils before and after the application of pesticides around waste receptacle were *Aspergillus niger, Penicillium sp., Fusarium siculi* and *Aspergillus fumigatus.* After the application of pesticides, microorganisms such as *Aspergillus nidulas*, *Microsporium canis* and *Yeast* were not present in the soils but were present in soils before the application of pesticides. Fungi are important decomposers in the soil food web and help bind physically soil particles together thereby creating a condition of stable aggregate that increases water infiltration and soil water holding capacity [12]. A reduction of fungal isolates may affect decomposition of organic matter and also cause a reduction in the physical binding of soil particles together. *Aspergillus niger, Penicillium sp, Fusarium siculi, and Aspergillus fumigatus* all had frequency occurrence of 18.2% respectively while *Aspergillus nidulas, Microsporium canis* and *Yeast* had frequency occurrence of 9.1% receptively. This is in line with [18] that discovered that the high fungi counts observed in the control soil and soils before application of pesticides may result from complex substrates of plant origin that are present in the soil suitable for agriculture.

VIII. CONCLUSION

Pesticides have been widely applied in the management of pests around waste receptacles in Port Harcourt. During the application of pesticides drifts may occur which adversely affects the ecosystem and also affects non-target organisms. This study effectively analysed the impact of drifts resulting from application of pesticides around waste receptacles. The study revealed that there were reductions in total heterotrophic bacteria and total heterotrophic fungi counts after the application of pesticides when compared to soils that were not treated with pesticides. This reduction may affect soil health around waste receptacles which may affect soil health. This is because the absence of microorganisms in such ecosystem may disrupt nitrification and denitrification.

Based on the findings of this study, the following recommendations were made:

1. A proper surveillance should be carried out to avoid drift to non-target organism. This is because it can cause instant death on persons around such environments.
2. Concentrations of such pesticides should be taken into account to avoid reduction in the number of microorganism in soils because of the vital roles they play in maintaining soil health.

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