



ENVIRONMENTAL IMPACTS OF LONG-TERM USE OF PESTICIDES IN COCOA ECOSYSTEM

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ABSTRACT

The study was conducted to evaluate the impacts of long use of pesticides on soil nutrients, water quality and microbial population within cocoa plantations ecosystem. Soil and water samples were obtained from selected cocoa plantations and uncultivated farmlands in major cocoa producing areas of Cross Rivers State, Nigeria. Samples were analyzed according to standard procedures. Soil pH across the various cocoa plantations ranged between 4.5 and 5.5 while uncultivated farmlands adjacent to the cocoa farms had a pH range of 5.12 to 5.8. On average, the level of residual copper in soils obtained from the cocoa farms was 40 folds of the level of copper in the various uncultivated farmlands. The obtained values for pH, TDS, TSS, EC, DO, hardness, turbidity, nitrates, sulphates and phosphates of the surface and underground waters were within the permissible limit set by World Health Organization (WHO) for potable water. The values obtained for BOD in the surface water samples (2.25 and 2.70) were higher than the permissible level (2.0) set by WHO for portable water. Soil bacterial population was reduced by 38.49, 50.53, 50, 40.98, 41.61 and 45.04% under the selected cocoa plantations in Ajassor, Efraya, Yahunde, Okundi, Ochon and Orimekpang respectively compared to the soil bacterial population in uncultivated farmlands adjacent to the various cocoa farms. Long term use of pesticides increased soil acidity, copper residue accumulation while it reduced soil microbial population.

Keywords: Cocoa, pesticides, black pod, microbial, mirid

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INTRODUCTION

Cocoa, *Theobroma cacao* L., is one of the major cash crops cultivated in the tropical regions of West Africa. The continent of Africa contributes more than 70% of total cocoa output globally. It is the main cash crop that contributes significantly to Nigeria's exchange earnings. A greater proportion of cocoa produced in Nigeria is exported to Europe and Western Nations. Cocoa production is a significant component of rural economy in Nigeria and other cocoa producing countries in West Africa, the Caribbean, South America and Asia as the industry is predominated by small-holder easant farmers who depend on the crop for livelihood and sustenance (Appiah, 2004).

Cocoa cultivation is closely associated with the incidence of pests and diseases. Five major diseases of cocoa include black pod disease, witches' broom, swollen shoot virus, vascular streak dieback and monilia pod rot (Flood *et al.*, 2004). Currently in Nigeria, the two major pests of cocoa with significant destruction and setback for cocoa production are *Phytophthora megakarya* (black pod rot) and *Sahlbergilla singularis* (mirid). *Phytophthora megakarya* is one of the *Phytophthora* species reported as disease causative agent on cocoa and it is regarded as the most virulent of the species. *P. megakarya* is indigenous to West and Central Africa and it has become the main yield limiting factor for cocoa production in affected areas

(Opoku *et al.* 2000). Under high frequency of rainfall, high humidity and low temperature, *P. megakarya* incidence can lead to 100% yield loss.

Among the insect pests of *Theobroma cacao*, the brown mirid, *Sahlbergilla singularis* Haglund is the most harmful in Nigeria (Opeke, 1992). Mirid feed by inserting its mouth parts into the plant and suck the juice. While sucking the plant juice, salivary secretion is injected into the plant tissue which results in plasmolysis of the cell. Cellular lysis results in necrosis, followed by the appearance of lesions (Mariau, 1999). Canker sores develop from the formed lesions due to invasion by cryptogamous parasites causing weakness of the plant part. The combination of tissue necrosis and cryptogamic attack results in wilting of the plant and eventually affects cocoa productivity (Ojelade *et al.*, 2005). The yield of about 30-70% has been attributed to mirid infestation and damage (Idowu, 1989; Ojelade *et al.*, 2005).

Even though, non-chemical methods of pest and disease management are widely recommended in cocoa industry for safety and wholesomeness of the produce, the use of some amounts of chemicals in the form of agrochemicals is unavoidable in the effective management of cocoa farms (Adjinah and Opoku, 2010). To mitigate the devastating impacts of pests and diseases on cocoa production in Nigeria, farmers apply synthetic pesticides as a mean of phyto-sanitation which is currently accepted as the fastest and most potent means of pest control globally. For appropriate, rational and safe use of pesticide to be achieved to sustain cocoa economy in the country, the Federal government of Nigeria through her agency with the sole mandate to conduct research on cocoa (Cocoa Research Institute of Nigeria) has approved several pesticides to be used in cocoa phytosanitation. Among the approved pesticides are: Ridomyl gold (Cu(OH)₂+ Mefenoxam), Ultimax plus (Cu(OH)₂+metalaxyl), Fungoran OH (Cu(OH)₂), Copper Nordox 75WP(Cu(OH)₂), Champ DP (Cu(OH)₂), Actara (Thiamethoxam), Esiom 150 SL (Acetamiprid + Cypermethrin). Even though there are

recommended doses for the various approved pesticides, there are ultimately several challenges associated with pesticides use. These include but not limited to abuse of pesticides use among farmers. Asogwa and Dongo, (2019) attributed the abuse of pesticide use among cocoa farmers to poor farmers' education on pesticide use. Apart from poor farmers' education on pesticide use, Dormon *et al.*, (2007) reported that, consequences of pesticides abuse, misuse and improper application are results of failure of farmers to adopt full research recommendations. Infiltration of banned and unrecompensed pesticides into the markets is one of the challenges with the use of pesticides in the cocoa industry. The entrance of such unapproved chemicals into the markets increases their potential for purchase and use by unsuspected or unlettered cocoa farmers. The chemical nature of the active ingredients of the pesticides determines their persistence and impact on the environment. Most of the fungicides used in controlling black pod disease are copper based. Copper, being a transition metal, is non-biodegradable and does accumulate in soil where it is bound to soil organic matter (Aikpokpodion, 2010). It is reported that only 15% of applied pesticide gets to the target while the rest is distributed within the environment (Aikpokpodion *et al.* 2013). In such a scenario, accumulation of non-biodegradable and persistent pesticide is inevitable. Several of the insecticides used on cocoa pods up till 2000 were either organochlorine or organophosphate which are regarded as persistent pesticides and endocrine disruptors (Aikpokpodion *et al.*, 2012). Residue of organochlorine insecticide DDT (Dichlorodiphenyltrichloroethane) was detected in cocoa beans from three cocoa ecologies in Nigeria (Aikpokpodion *et al.*, 2012) while in another study, Diazinon, an organophosphate insecticide was also detected in cocoa beans from three cocoa ecologies in Nigeria (Aikpokpodion *et al.*, 2012).

Considering the age-long culture of cocoa cultivation in Nigeria, the repeated use of persistent and non-biodegradable pesticides has the capacity to contaminate various

components of the environment which includes water, air and soil ecosystem. The health quality of the various environmental components has direct or indirect impacts on the people who live within the environment. If the soil within the environment is contaminated with pesticides residues, the contaminants could gain access to surface and underground water through leaching and run-off. This will ultimately lower the quality of water consumed by the inhabitants of the community. The consumption of such water could lead to several health issues. Currently in Nigeria, there is little or no information on the long-term use of pesticides and their impacts on soil nutrients, surface and underground water quality and microbial population. The study was therefore, conducted to evaluate the impacts of long-term use of pesticides on soil microbial population, soil chemical properties, surface and groundwater quality within selected communities around cocoa plantations that have received continuous pesticides application more than a decade.

MATERIALS AND METHODS

Soil sampling for microbial population count

Soil samples were collected in selected cocoa farms in Ajassor, Yaunde, Efraya, Mfum, Orimekpan and Ochon in Cross Rivers State, Nigeria. Soil sampling was done with soil auger at the depth of 0-10cm for microbial population. The cocoa plantations were selected for soil sampling on the condition that they had received continuous pesticides application for the control of black pod disease and insect pests' infestation for fifteen years and above. The collected samples were kept in labeled transparent polythene bags and later transferred into an ice bag prior to their transportation to the laboratory for microbiological studies.

Microbial population: Serial dilution method for bacterial count

1 gram of each soil sample was introduced into a test tube containing 10ml of distilled water with dilution factor 10^{-1} . The mixture was well shaken and 1ml was taken from the first test tube and to a second test tube with 9ml of distilled water whose dilution factor became 10^{-2} . Another 1ml was thereafter taken from the test tube of 10^{-2} dilution factor into another test

tube with 9ml distilled water to obtain 10^{-3} dilution factor. In a similar manner, an aliquot of 1ml was taken and added to the rest test tubes to obtain dilution factors of 10^{-4} , 10^{-5} , 10^{-6} , 10^{-7} , 10^{-8} , 10^{-9} and 10^{-10} (Sonu, 2015). From the test tube with soil sample of 10^{-10} dilution factor, 1ml was taken and transferred to prepared 15ml nutrient agar media in petri-plates and incubated for 24 hours.

Biochemical Tests

Carbohydrate fermentation

The principle of carbohydrate fermentation test is based on the ability of certain bacteria to cleave different carbohydrates depending on the enzymes they possess. This is usually detected by the evolution of CO_2 in liquid ammonia or color change of pH indicator due to acid production. The sugar fermentation test was done using Hugh and Liefson's basal medium. 1gram of sugar medium was measured in 10ml distilled water for the preparation of Hugh and Liefson's basal medium. 10ml of the basal medium was measured in different McCartney bottles and 0.1ml of different sugar solution was measured into the bottles and thoroughly shaken. The bottles and the contents were sterilized at 121°C for 15minutes in an autoclave. 3 drops of bromocresol purple was added to the sterile medium and an inverted Durham tube was placed in each bottle and inoculated with pure culture of the organisms and incubated at 37°C for 2-7 days. Medium was examined each day to observe color change. The production of acid was determined by change in color from purple to yellow while the production of gas was determined by the accumulation of gas in the inverted Durham tube.

Catalase test

Catalase test was carried out by placing a young culture of the organism on a clean microscope slide followed by the addition of 3% hydrogen peroxide with a wire loop. The production of white effervescence froth indicated that the organism was catalase positive while the absence of effervescence froth was an indication that, the organism was catalase negative.

Citrate test

The pure culture of organism was inoculated in a test tube of Koser citrate medium and incubated for 96 hours at 35°C.

Sulphide Indole motility test

The multiple test was used to simultaneously test for indole sulphide and motility in the organisms. The medium was stabbed with sterile needle containing isolates to a depth of 5mm. medium was then incubated for 48hours. The medium was observed for cloudiness (motility), release of dark precipitate (H₂S production) while the appearance of a dark red color after the addition of 2-3 drops of Kovac reagent depicts positive indole test.

The Methyl red Voges-Proskauer test was used to distinguish coliform from aerogenes which are similar in media. MRVP broth medium was prepared, and the broth was inoculated at 35°C for 24hours and then 48-72hours.

The oxidase test was carried out by soaking a piece of filter paper in oxidase reagent solution. Fresh young culture was scrapped with a glass rod and rubbed on filter paper. A blue coloration showed positive oxidase test.

The urease test was carried out by inoculating urea medium heavily with the culture and thereafter incubated for 24 hours at 35°C. The change in color from yellow to pink showed the presence of urease.

Water Analysis

Water sample collection was made in Ajassor, Efraya, Mfum, Yaunde, Orimekpan and Ochon within Etung, Ikom, Boki and Obubra local government area of Cross River State. Water samples were collected from well and streams within the environment of existing cocoa plantations in the various communities. The samples were collected in plastic bottles pre-treated with 0.1M nitric acid and well rinsed with deionised water to prevent sample contamination. The container and samples were placed under cold condition in an ice bag.

Soil pH, electrical conductivity, temperature, total dissolved solids, turbidity, total suspended solids, dissolved oxygen, biochemical oxygen demand, hardness, nitrate, sulphates, phosphates, Cu, Fe and Pb were determined according to the official standard procedure of AOAC (1998)

Soil Sampling and Chemical Analysis

Soil samples were collected with soil auger at a depth of 0-20cm. The samples were air-dried under room temperature and sieved with a 2mm sieve. Soil pH was determined with pH meter in 1:1 soil/water ratio in de-ionized water. Available phosphorus was determined using Bray and Kurtz method (Bray & Kurtz, 1945), organic carbon was determined with Walkley and Black method (1934), Exchangeable bases were extracted with 1N ammonium acetate solution while micronutrients were extracted with 0.1M HCl. Buck Scientific 210 Atomic absorption spectrophotometer was used to quantify the exchangeable bases and micronutrients while Flame photometer was used for the determination of Na and K.

RESULTS

Result of microbial population in soil samples collected from the selected cocoa plantations and uncultivated farmland adjacent to the plantation in Ajassor, Efraya, Yaunde, Okundi, Ochon and Orimekpan is presented in Table 1. The obtained results showed a 38.49% reduction in the microbial population in the soil sample collected from Ajassor cocoa plantation when compared with the adjacent uncultivated farmland. In a similar manner, there was a 50.53, 50, 40.98, 41.61 and 45.04% reduction in microbial population in soil samples obtained from cocoa plantations in Efraya, Yaunde, Okundi, Ochon and Orimekpan respectively compared with the uncultivated farmlands adjacent to the various cocoa plantations.

Table 1: Variation in soil microbial population

Community	Cocoa farm	Adjacent Uncultivated bush	% reduction in microbial population
Ajassor	$1.55 \times 10^6 \pm 2.65 \times 10^4^c$	$2.52 \times 10^6 \pm 5 \times 10^3^f$	38.49
Efraya	$5.64 \times 10^5 \pm 7.8 \times 10^3^a$	$1.14 \times 10^6 \pm 1.14 \times 10^4^a$	50.53
Yaunde	$5.9 \times 10^5 \pm 1 \times 10^3^a$	$1.18 \times 10^6 \pm 6.8 \times 10^3^b$	50
Okundi	$1.24 \times 10^6 \pm 2.65 \times 10^3^d$	$2.05 \times 10^6 \pm 3.04 \times 10^4^e$	40.98
Ochon	$8.8 \times 10^5 \pm 6.08 \times 10^3^c$	$1.37 \times 10^6 \pm 5.0 \times 10^3^d$	41.61
Orimekpang	$6.87 \times 10^5 \pm 7.8 \times 10^3^b$	$1.25 \times 10^6 \pm 1.0 \times 10^4^c$	45.04

Means on the same column with different superscripts are statistically significant ($p < 0.05$)

The result of physical characterization of 21 isolates of bacteria obtained from the experimental soil samples is presented in Table 2. From the result, 11 of the isolates were circular in shape, 6 were filamentous while 4 of the isolates were irregular. The margin of the organisms shows that 11 isolates were entire, 6 were filliform and 4 were undulate. The elevation of the isolates showed that 20 of the isolates were flat while 1 was raised. The

opacity of the isolates indicated that 19 were translucent while 2 were opaque. Under color characterization, 15 of the isolates were creamy while 6 were whitish. The texture of the 21 isolates showed they were all butyrous. The cell shape showed that 20 of the isolates were rod-like while 1 isolate was coccoid. Under Gram reaction, 11 of the isolates were gram positive while 10 were gram negative.

Table 2: Physical characterization of Bacteria

Colony characteristics	Physical characterization		
Form	Circular (11)	Filamentous (6)	Irregular (4)
Margin	Entire (11)	Filliform (6)	Undulate (4)
Elevation	Flat (20)	Raised (1)	
Opacity	Translucent (19)	Opaque (2)	
Color	Cream (15)	Whitish (4)	
Texture	Butyrous (21)		
Cell shape	Rod (20)	Coccoid (1)	
Gram reaction	Positive (11)	Negative (10)	

Results of biochemical characterization of the bacterial isolates are shown in Table 3. Result of the test showed that, all the 21 isolates were negative to methyl red test. Under Voges prosekauer test, 12 of the isolates were positive while 9 were negative. While all the isolates were negative to urease test, all the isolates were positive to catalase test. 16 of the isolates were positive to citrate while 5 were negative. 10 of the isolates were positive to sulphide test while 11 were negative to the test. Indole test showed that all the isolates were negative. 16 isolates were positive to motility test while 5 were negative. 12 isolates were positive to oxidase while 9 were negative. Glucose test showed that 11 of the isolates were acid producing, 8 produced acid and gas while 2 did

not respond to glucose test. 13 isolates did not respond to lactose test while 8 isolates produced gas when treated with lactose. Under mannitol test, 19 isolates produced gas while 2 isolates did not respond to the test. 20 isolates produced acid when treated with galactose while 1 isolate did not respond. 4 isolates did not respond to sucrose test while 17 isolates produced gas. 8 isolates produced gas under arabinose test while 13 showed no change. 10 of the isolates produced acid under maltose test while 11 isolates showed no change under maltose test. Table 4 shows the list of all possible bacteria in soil samples obtained from the various cocoa plantations and uncultivated farmlands based on the physical and biochemical characterization.

Table 3: Biochemical characterization of Bacteria

Test	Biochemical characterization	
Methyl red	Positive (0)	Negative (21)
Voges Prosekauer	Positive (12)	Negative (9)
Urease	Positive (10)	Negative (21)
Catalase	Positive (21)	Negative (0)
Citrate	Positive (16)	Negative (5)
Sulphide	Positive (10)	Negative (11)
Indole	Positive (0)	Negative (21)
Motility	Positive (16)	Negative (5)
Oxidase	Positive (12)	Negative (9)
Glucose	Acid produced (11)	Acid and gas (8) No change (2)
Lactose	No change (13)	Acid produced (8)
Mannitol	Acid produced (19)	No change (2)
Galactose	Acid produced (20)	No change (1)
Sucrose	No change (4)	Acid produced (17)
Arabinose	Acid produced (8)	No change (13)
Maltose	Acid produced (10)	No change (11)

Table 4: Possible Bacteria in soil samples based on physical and biochemical characterization

S/No	Possible organisms
1	<i>Bacillus amyloliquefaciens</i>
2	<i>Bacillus endophyticus</i>
3	<i>Bacillus pumilus</i>
4	<i>Athrobacter nicotianae</i>
5	<i>Bacillus fumarioli</i>
6	<i>Bacillus sporothermodurans</i>
7	<i>Microbacterium gubbeenense</i>
8	<i>Bacillus safensis</i>
9	<i>Athrobacter mysorens</i>
10	<i>Candidimonas humi</i>
11	<i>Achromobacter insolitus</i>
12	<i>Nesterenkonia halobia</i>
13	<i>Bacillus trypoxylicola</i>
14	<i>Burkholderia ubonensis</i>
15	<i>Pseudomonas chlororaphis</i>
16	<i>Burkholderia thailandensis</i>
17	<i>Pseudomonas indica</i>
18	<i>Pseudomonas spp</i>
19	<i>Pseudomonas spp</i>
20	<i>Pseudomonas spp</i>
21	<i>Pseudomonas chlororaphis</i>

Impact of long-term pesticide use on soil chemical properties

The result of the chemical properties of soil samples obtained from selected cocoa plantations in Ajassor, Efraya, Yaunde, Okundi, Orimekpan and Ochon and the respective uncultivated farmland adjacent to the farms is presented in Table 5. The soil pH of the

cocoa farms ranged from 4.5 – 5.5 while the uncultivated farmland had pH range of 5.12-5.80. Among the cocoa plantations, the selected cocoa plantation in Okundi had the lowest value of soil pH while that of Yaunde had the highest pH value. Table 5 shows that, apart from the soil sample from cocoa plantation in Ajassor and Okundi, the soil organic carbon in all other cocoa plantations had lower values of soil organic matter compared to the uncultivated farmland adjacent to the plantations.

Magnesium content of the soil samples from the various cocoa plantations ranged from 0.28 – 1.78cmol/kg while the uncultivated farmlands had magnesium ranging from 0.27 – 1.73cmol/kg. Cocoa plantation in Ajassor had the least Mg content while the soil sample from Yaunde had the highest Mg content. Soil analysis shows that; the various cocoa plantation was higher in Mg content than their counterpart uncultivated farmlands. Table 5 Shows that the levels of calcium in soil samples from all the cocoa plantations were lower than those of the uncultivated farmlands adjacent to the plantations. The reduction of calcium was due to high demand of calcium by the tree crop. The nitrogen content of soil samples from the cocoa plantations ranged from 0.12 – 0.28% while the N content of the uncultivated farmlands adjacent to the plantations ranged between 0.14 and 0.36%. Copper residues in the soil samples from the various cocoa plantations

ranged from 16.27 – 50.06mg/kg while the level of copper in uncultivated farmlands adjacent to the plantations ranged from 0.4 – 1.32mg/kg. Under the cocoa plantation in Ajassor, the level of copper residue in soil was 40 folds of the amount of copper in the uncultivated farmland adjacent to the plantation. In a similar manner, the level of copper in Efraya cocoa plantation was 43 folds of the copper in the soil sample from the uncultivated farmland in the same neighborhood. In addition, the copper residue in soil samples obtained from cocoa plantation in Yaunde, Orimekpan, Ochon and Okundi was 42, 32, 40 and 31 folds of the copper in the respective uncultivated farmlands adjacent to the various cocoa plantations.

Water Quality in cocoa growing communities

The results of water analysis obtained from cocoa producing communities in Mfum, Ajassor, Yaunde, Efraya, Orimekpan and Ochon is presented in Table 6. The pH of the water samples ranged from 6.40 – 7.20 with an average value of 6.56. The result showed that, stream water from Ajassor, Yaunde, Efraya, Orimekpan and Ochon were slightly acidic while the well water from Mfum was alkaline. Among the stream water samples, the sample from Yaunde was the most acidic while Ochon sample was the least acidic.

The total dissolved solids (TDS) in the water samples ranged from 20.24 to 80.40mg/L with a mean value of 30.20 mg/L. Water sample from Ajassor stream had the least value of total dissolved solids while the well water from Mfum had the highest total dissolved solids. Total suspended solids (TSS) in the analyzed water samples ranged from 0.50 – 0.61mg/L with an average value of 0.50mg/L. The least

value of total suspended solids was recorded for water sample from Ochon while the stream water from Efraya had the highest TSS value. However, there was no detectable suspended solid in the well water from Mfum.

Electrical conductivity of the water samples ranged between 6.0 and 15 μ S/cm with a mean value of 7.48 μ S/cm. Water sample from Ajassor stream had the least value of electrical conductivity while the sample obtained from the well water in Mfum had the highest value. Dissolved oxygen (DO) in the water samples ranged from 1.20 - 3.26 mg/L with an average value of 2.69mg/L. The water sample from the well had the least value while the water obtained from Yaunde stream had the highest value.

Biochemical oxygen demand (BOD) in the evaluated water samples ranged between 0.50. and 2.70 mg/L with an average value of 2.25mg/L. The sample from the well had the lowest value while the water sample from Efraya stream had the highest value.

Total hardness of the water samples ranged from 15.0-40.0mg/L with a mean value of 20.84mg/L. The water sample from Efraya stream had the lowest value of hardness while the well water from Mfum had the highest value.

The concentration of copper in the various water samples ranged from 0.01 – 0.04mg/kg with an average value of 0.02mg/kg. The lowest value of copper was found in water samples from Yaunde and Orimekpan while the highest value was recorded in sample from the well water.

Table 5: Chemical properties of soil samples from cocoa plantations and uncultivated farmlands

Community	pH	Ca	Mg	Na	K	ECEC	Exc. acidity	Org. C	N	P	Zn	Cu	Mn	Fe	
		Cmol/kg							%			Mg/kg			
Ajassor cocoa farm	4.7±0.9 ^c	0.28±0.1 ^a	0.28±0.1 ^a	0.02±0.01 ^a	0.16±0.2 ^b	0.74±0.2 ^a	0.15±0.1 ^b	1.50±0.2 ^b	0.120±0.05 ^a	8.42±1 ^a	14.17±1 ^a	50.06±5 ^d	51.55±4 ^c	108.42±16 ^{ab}	
Ajassor uncultivated	5.39±1.1 ^g	0.31±0.2 ^{ab}	0.27±0.2 ^a	0.016±0.01 ^a	0.10±0.1 ^a	0.70±0.3 ^a	0.16±0.04 ^b	1.40±0.1 ^b	0.14±0.06 ^a	7.91±1 ^a	10.97±2 ^a	1.25±0.7 ^a	34.93±3 ^b	77.55±10 ^a	
Efraya cocoa farm	4.8±0.7 ^d	3.01±0.8 ^g	1.19±0.6 ^d	0.11±0.01 ^b	0.22±0.1 ^{bc}	4.53±1.2 ^c	0.13±0.12 ^b	2.07±0.6 ^c	0.35±0.1 ^c	7.57±1 ^a	40.52±5 ^{bc}	40.53±3 ^c	230±15 ^c	150.4±13 ^c	
Efraya uncultivated	5.7±1.2 ⁱ	4.8±1.2 ^h	1.11±0.6 ^d	0.09±0.01 ^b	0.10±0.1 ^a	6.10±1.4 ^{de}	0.11±0.1 ^b	2.68±1 ^{cd}	0.16±0.02 ^a	10.89±2 ^b	35.19±3 ^b	0.93±0.1 ^a	314±13 ^f	148.25±12 ^c	
Yaunde cocoa farm	5.5±0.8 ^h	1.93±0.3 ^d	1.78±0.8 ^e	1.93±0.1 ^e	0.24±0.09 ^c	5.88±1.2 ^d	0.25±0.13 ^c	2.01±0.6 ^c	0.28±0.05 ^b	10.98±2 ^b	53.03±5 ^d	21.05±3 ^b	345±16 ^f	95.43±9 ^a	
Yaunde uncultivated	5.8±1.3 ^j	2.02±0.5 ^{de}	1.73±0.7 ^e	1.89±0.2 ^e	0.21±0.1 ^{bc}	5.85±0.9 ^d	0.24±0.09 ^c	2.50±1 ^{cd}	0.32±0.09 ^c	12.00±1 ^c	48.37±4 ^c	0.50±0.1 ^a	287±12 ^e	82.04±13 ^a	
Orimekpan cocoa farm	4.6±0.5 ^b	0.54±0.3 ^c	0.32±0.1 ^a	0.54±0.1 ^d	0.10±0.03 ^a	1.50±0.4 ^b	0.17±0.05 ^b	1.28±0.7 ^{ab}	0.14±0.06 ^a	14.06±2 ^d	48.16±4 ^c	38.87±4 ^c	25.55±2 ^a	182±3 ^{cd}	
Orimekpan uncultivated	5.32±1 ^g	0.59±0.2 ^c	0.30±0.2 ^a	0.52±0.2 ^d	0.08±0.01 ^a	1.49±0.6 ^b	0.16±0.03 ^b	1.30±0.5 ^{ab}	0.16±0.04 ^a	17.12±2 ^e	45.00±4 ^c	1.20±0.8 ^a	19.24±2 ^a	168±16 ^c	
Ochon cocoa farm	4.9±1.2 ^e	2.7±0.7 ^f	0.85±0.1 ^c	2.7±1.1 ^f	0.20±0.1 ^{bc}	6.45±1.2 ^e	0.05±0.01 ^a	2.18±0.8 ^c	0.33±0.05 ^c	14.46±2 ^d	12.90±1 ^a	16.27±2 ^b	174.61±11 ^d	204±14 ^d	
Ochon uncultivated	5.48±1.1 ^h	2.14±0.5 ^e	0.83±0.4 ^c	2.66±0.9 ^f	0.17±0.02 ^b	5.8±1.0 ^d	0.03±0.01 ^a	2.85±1 ^d	0.36±0.08 ^c	16.42±2 ^e	11.02±1 ^a	0.40±0.1 ^a	152.63±14 ^d	190±23 ^d	
Okundi cocoa farm	4.5±1.1 ^a	0.34±0.1 ^{ab}	0.51±0.2 ^b	0.34±0.1 ^c	0.21±0.1 ^{bc}	1.40±0.21 ^b	0.30±0.1 ^c	1.33±0.8 ^{ab}	0.18±0.04 ^{ab}	10.73±1 ^b	55.47±5 ^d	41.62±3 ^c	383.71±14 ^g	120±12 ^b	
Okundi uncultivated	5.12±1.4 ^f	0.39±0.1 ^b	0.45±0.3 ^b	0.33±0.1 ^c	0.20±0.1 ^{bc}	1.37±0.65 ^b	0.27±0.08 ^c	1.04±0.4 ^a	0.21±0.1 ^b	9.84±1 ^{ab}	53.13±6 ^d	1.32±0.8 ^a	342.32±12 ^f	98.00±6 ^a	

Means on the same column with different superscripts are statistically significant ($p < 0.05$)

Table 6: Chemical properties of water samples

Parameter	pH	TDS	TSS	DO	BOD	Hardness	NO ₃ ²⁻	SO ₄ ²⁻	PO ₄ ³⁻	Cu	Fe	Pb	EC	Turbidity
		(mg/L)											(µS/cm)	(NTU)
Mfum well	7.20±1.1 ^d	80.40±4.2 ^c	BDL	1.20±0.1 ^a	0.50±0.1 ^a	40.00±4 ^b	0.03±0.02 ^a	BDL	0.96±0.02 ^b	0.04±0.02 ^c	0.07±0.03 ^b	BDL	15.00±3 ^d	0.20±0.04 ^a
Ajassor stream	6.5±0.8 ^b	20.24±2.6 ^a	0.59±0.02 ^b	3.12±0.2 ^c	2.50±0.9 ^b	18.20±2 ^a	0.048±0.03 ^a	0.62±0.1 ^b	0.90±0.02 ^b	0.02±0.01 ^b	0.025±0.02 ^a	BDL	6.00±1 ^a	1.60±0.06 ^{bc}
Yaunde stream	6.42±0.7 ^a	25.02±2.3 ^{ab}	0.58±0.04 ^{ab}	3.26±0.7 ^c	2.58±0.5 ^b	16.00±3 ^a	0.046±0.06 ^a	0.41±0.4 ^a	0.95±0.01 ^b	0.01±0.01 ^a	0.032±0.04 ^a	BDL	8.00±1.2 ^c	1.40±0.02 ^b
Efraya stream	6.4±0.4 ^a	27.00±1.9 ^{ab}	0.61±0.07 ^b	2.50±0.9 ^b	2.70±1 ^b	15.00±2 ^a	0.05±0.02 ^a	0.83±0.3 ^c	0.60±0.4 ^a	0.02±0.01 ^b	0.032±0.01 ^a	BDL	8.10±2 ^c	1.84±0.04 ^c
Orimekpan stream	6.56±0.7 ^b	30.20±2.1 ^b	0.54±0.02 ^a	2.86±1 ^b	2.64±1 ^b	16.64±4 ^a	0.062±0.03 ^b	0.40±0.2 ^a	0.85±0.03 ^{ab}	0.01±0.01 ^a	0.028±0.02 ^a	BDL	7.26±1.7 ^b	1.30±0.06 ^b
Ochon stream	6.72±0.4 ^c	26.14±1.6 ^{ab}	0.50±0.05 ^a	3.20±1.2 ^c	2.59±0.9 ^b	19.20±3 ^a	0.045±0.01 ^a	0.52±0.1 ^{ab}	0.98±0.01 ^b	0.02±0.01 ^b	0.03±0.01 ^a	BDL	7.48±1.5 ^b	1.02±0.03 ^b
Mean	6.63	34.83	0.56	2.69	2.25	20.84	0.05	0.56	0.87	0.02	0.04	0	8.64	1.23
WHO limit (2004)	6.00 - 9.00	500	20	4.0 – 6.00	2	120	45	250	5	1	0.1	0.05	200	25

Key: TDS = Total dissolved solids ; TSS =Total soluble solids ; DO =Dissolved oxygen ; BOD =Biological oxygen demand ; EC =Electrical conductivity ;

Means on the same column with different superscripts are statistically significant ($p < 0.05$)

BDL: Below detection limit

DISCUSSION

Microbial population

The reduction in microbial population observed in soil samples from cocoa plantations was the consequence of long-term use of pesticides within the cocoa plantations. In a typical cocoa plantation, there are three main types of pesticides used by farmers. One is the use of copper-based (CuO, Cu(OH)₂) fungicides for the control of black pod disease which includes but not limited to Metalaxyl, Mandipropamid, Mefenoxan, Pyraclostrobin, Dimethomorph, and Ametoctradin; the second type of pesticide is insecticides for the control of mirid which includes the class of neonicotinoids (Thiamethoxam, Acetamiprid, Cypermethrin, Thiacloprid and Imidacloprid); Pyrethroids (Deltamethrin) while the third category of pesticide is herbicide which includes Glyphosate. In the past, organochlorine and organophosphates were intensely used by cocoa farmers for the control of mirid (*Sahlbergella singularis*). Though the use of the insecticides had long been banned in Nigeria, some farmers may still be using them considering their affordability and the continuous production of the chemicals by manufacturing companies. Taking everything into account, the various classes or categories of pesticides used in cocoa phytosanitation, are all synthetic agrochemicals which are either moderately or highly toxic to soil microorganisms. The soil matrix is the recipient of at least 85% of the volume of every pesticide used in Cocoa plantations (Aikpokpodion et al. 2010). Whenever farmers use agrochemicals on the farm, non-target effect of such agronomic activity is mainly on soil biota (Wang et al., 2006).

The use of agrochemicals subject soil microorganisms to toxicological stress as non-target. More than 95% of applied herbicides and 98% insecticides get to soil microorganisms as non-target (Miller, 2004). It has been reported that, application of insecticides affects the growth, survival and functionality of symbiotic *rhizobial* association with roots of nitrogen fixing plants resulting in N-fixation alteration (Niewiadomska, 2004). The growth and population of *azotobacter* has been hampered significantly because of carbofuran, malathion,

parathion application (Pandey and Singh, 2004). Application of chloripyrifos in agricultural soil has been reported to have had negative impacts on the activities of *Pseudomonas fluorescences*, *Bacillus subtilis*, *Mycobacterium phlei*, *Trichoderma harzianum*, *Penicillium expansum* and *Fusarium oxysporum* (Virag et al. 2007). Synthetic agrochemicals generally impart toxicological effects on soil microorganisms due to the ease with which the active ingredients in pesticides permeate the cells of biota. Some pesticides are also lipophilic which make them accumulate in the lipid portion of the soil organism and exert chronic toxicity. Solubility of pesticides' active ingredients in water also enhances their percolation under gravity within the soil system thereby increasing the coverage of impact. Insecticides such as Cypermethrin, Chlorfluzuron and phoxim have the history of imparting inhibitory effects on soil microorganism even at recommended dose (Amirkhanov et al., 1994). Aikpokpodion et al. (2010) reported the contamination of cocoa plantations with residues of pesticides used in phytosanitation among Nigerian cocoa farmers.

The repeated use of complex chemicals in plant protection inevitably destroys the microbial life that is needed to maintain a healthy soil ecosystem (Shang et al, 2019). Apart from the annual nutrients mining by cocoa trees, the reduction in microbial population in cocoa plantations due to pesticides' toxicological effects may be a contributory factor responsible for the decline in soil fertility in most cocoa soils in Nigeria. The negative impacts of long-term use of pesticides on microbial population in cocoa ecosystem is evident as shown by the biochemical characterization of soil bacteria isolates in the examined soil samples.

The results of biochemical characterization suggest that, in Ajassor cocoa plantation, the prominent bacteria bacterial were *Burkholderia thailandensis*, *Pseudomonas indica*, *Pseudomonas Spp*, *pseudomonas chlororaphis* while *Candidimonas humi*, *Achromobacter insolitus*, *Nesterenkoria halobia*, *Bacillus trypoxylicola*, *Burkholderia ubonensis* and *Pseudomonas chlororapis* were predominant in

the uncultivated farmland adjacent to cocoa farms. Considering the predominant bacteria in each of the two locations, it was observed that, there were some specific bacteria present in the uncultivated farmland that were not found in the cocoa plantation. One of the predominant bacteria in the uncultivated farmland found missing in soil from cocoa plantation possibly due to extinction or depopulation by pesticides was *Bacillus tryoxylicola*. *Bacillus spp* can produce variety of biochemical materials as metabolites with antibacterial and bacteriostatic activities such as subtilin, antibacterial protein, organic acids which can inhibit the growth and reproductive activities of pathogenic bacteria. The secreted substances from the organisms have the capacity to destroy bacterial structure and kill pathogens. When beneficial microorganisms are destroyed as a result of long term use of pesticides, the ultimate consequence on soil ecosystem could be grave. Extinction or depopulation of beneficial soil microorganisms could have adverse effects on nutrients mineralization, alteration of organic matter turnover in soil, nitrogen fixation in soil and loss of soil biodiversity (Baxter *et al.*, 2008; Eisenhauer *et al.* 2009; Mader *et al.*, 2002).

Similar observation was made in the biochemical characterization of soil bacteria in soil samples obtained from the selected cocoa plantation in Efraya. In the cocoa plantation, *Bacillus amyloliquefaciens*, *Bacillus endophyticus*, *Bacillus pumilus*, *Athrobacter nicotianae*, *Bacillus furarioli* were predominant while *Bacillus sporothermodurans*, *Microbacterium gubbeenense*, *Nacillus safensis* and *Athrobacter mysorens* were predominant in the uncultivated farmland adjacent to cocoa farm. The report of Onwona-Kwakye *et al.*, (2020) also reported a decrease in bacteria population and loss of biodiversity in soils of rice plantations treated with pesticides.

Soil analysis

A critical look at the pH values of soil samples obtained from the uncultivated farmland showed that the pH values obtained in each location was higher than those of the cocoa plantations. The lower pH of cocoa plantations

compared with the uncultivated farmland is an indication of higher soil acidity in cocoa farms compared with the uncultivated soils. The increased acidity must have been triggered by the long-term application of CuSO_4 fungicides in the various cocoa farms in the control of black pod disease. It has been established that only 15% of applied pesticides gets to the target while the rest ends up in the soil matrix (Leonilla 2002, Aikpokpodion *et al.*, 2012). When CuSO_4 is applied, the residue of the compound gets dissociated into Cu^{2+} and SO_4^{2-} when in contact with water in the soil. In a reversible reaction, the SO_4^{2-} formed combines with water molecule to form HSO_4^- which is a weak base. The weak base could combine with other water molecules to produce more impact on soil acidity. Different agrochemicals can affect soil pH in diverse ways depending on the chemistry of the active ingredients of the pesticides. Stanley *et al.* (2013) reported a decrease in soil pH two weeks after field application of atrazine and paraquat. The report of Azeez *et al.*, (2021) which evaluated the effects of fungicides spray on nutrient content of soil of cocoa growing areas of Southwestern Nigeria showed that, 57, 33 and 33% of the evaluated cocoa plantations had soil pH lower than those of adjacent arable farms.

Soil pH is a vital property that controls a lot of activities within the soil system. pH generally controls the bioavailability of plant nutrients. At low soil pH, availability of many of the macronutrient is reduced. Conversely, at low soil pH many micronutrients are readily bioavailable. At low pH, the availability of exchangeable bases which are largely required by plants will be reduced in soil solution due to their dislodgment from exchangeable sites by hydrogen whose ionic population is favored at low pH. Low soil pH can also have negative impact on the type of element that becomes available to plant. When soil pH drops below 5.50, aluminium becomes readily available to plants and soil microbes. Because most plants are not aluminium tolerant, they could experience aluminium toxicity which may affect growth and yield (Imadi *et al.*, 2016). Increased acidity in soil is always accompanied by a shift in the type of soil microorganisms and

their activities. When this occurs, there will be a significant decrease in organic matter decomposition which can lead to immobilization of basic nutrients with less available nutrients for plant uptake. (Tarah et al., 2017). It is expected that the built-up and recycling of leaf litter fall under cocoa plantations should significantly impart the organic matter content of the soils compared to the uncultivated farmland. However, the situation as shown by the obtained data on soil organic carbon suggests that the low soil pH might have either impaired the activities of the soil microbes responsible for the breakdown of organic matter or caused a shift in the microbial population within the soil which caused accumulation of plant residues with little decomposition and mineralization.

The lower level of calcium in the cocoa plantations compared to uncultivated farmlands could limit the capacity of the soil to neutralize the acidic effect of hydrogen ions in the soil. Under normal soil conditions, exchangeable bases like Ca and Mg react with soluble and active H^+ and Al^{3+} in soil to neutralize their effects of soil acidity. In a situation of low Ca^{2+} in soil, the capacity to neutralize hydrogen and aluminum acidity becomes weakened which eventually keeps the soil in acidic condition. The observed higher concentration of magnesium in cocoa plantations as compared with uncultivated farmland was similarly reported by Azeez et al., (2021) on cocoa plantations in southwestern Nigeria. In a similar manner, potassium concentration in the cocoa plantations was higher than the content of uncultivated farmland. The higher Mg and K in cocoa plantations in relation to uncultivated farmlands might be due to higher mineralization of Mg and K in the various plantations. It could also be a result of fertilizer application by the farmers. Though, it is well known that cocoa farmers in Nigeria do not use fertilizers to replenish the nutrients lost through cocoa pod harvest, some of the farmers occasionally use inorganic fertilizers whenever fund is available to make fertilizer purchase. On the other hand, a few of them use organic fertilizers in the form of compost and manure.

All the investigated cocoa plantations except the one in Ajassor had higher exchangeable acidity than the corresponding uncultivated farmlands. Exchangeable acidity is the combination of acidity caused by H^+ and Al^{3+} in soil. Since, there are no anthropogenic activities associated with agronomic practices among Nigerian farmers that introduce aluminum into the soil, it is safe to state that, activities that led to soil acidity invariable determined the exchangeable acidity in cocoa plantations. The values of nitrogen in soil samples obtained from cocoa plantations in all the communities were lower than those obtained in their counterpart uncultivated farmlands. Besides the uptake of nitrogen by cocoa plants, the long-term application of pesticides in the various cocoa plantations is one of the key factors that led to the reduction in nitrogen status of the soils. Application of synthetic chemicals have detrimental effects on microorganisms on a general note. Prolonged application of insecticides inhibits both the process of nitrification and the microbes involved (Gundi et al., 2005). It has been established through scientific studies that, application of chlorpyrifos, monocrotophos, lindane, dichlorvos, endosulfan at concentration of 0.02 to 10 times of the field recommended dose reduced the biochemical process of nitrification and denitrification in agricultural soils (Madhaiyan et al., 2006). Insecticides generally have negative effects on soil microorganisms that are vital to nitrogen transformation in soils. The severity of their impacts on the organism depends on the class of the insecticides and the rates of their application (Das and Mukherjee, 2000).

The levels of copper in cocoa plantations were much higher than the values obtained in samples from the uncultivated farmlands. The significant disparity between the residue of Cu in cocoa plantations and the uncultivated adjacent farmlands is a consequence of the long-term use of copper-based fungicides in controlling the menace of black pod disease of cocoa. Severity of black pod disease caused by *Phytophthora megarkarya* in Cross River is occasioned by the high annual rainfall in the region. *Phytophthora* thrives in an environment

of high humidity and moderate temperature. Frequent precipitation with high humidity keeps the environment conducive for the growth and spread of the disease. If black pod disease infestation is not controlled, there could be a 100% loss of cocoa pods (Opoku *et al.*, 2000). This is the main reason for the aggressive use of Cu-based fungicides by the farmer to prevent the loss of investment associated with commercial cocoa cultivation. Zinc, manganese and Iron concentrations in soil samples obtained from cocoa plantations were higher in values than the values obtained for same micronutrients in their counterpart uncultivated farmlands. Micronutrient bioavailability in soil solution increases with increased soil acidity. The higher content of Zn, Mn and Fe in the cocoa farms might be a result of higher acidity in the plantations compared to the uncultivated farmlands.

Surface and groundwater analysis

The alkalinity of the well water sample was an indication of carbonate rock beneath the well which supplied the water with calcium carbonate (CaCO_3). The values obtained for pH in all the water samples fall within the acceptable limit of the World Health Organization (6.0 -9.0) for drinking water. A pH range of 5.70 – 7.00 was reported by Ezenweani and Ezeweani (2019) for drinking water in Ugbowo community in Benin City, Nigeria while a pH range of 4.49 – 5.45 was reported by Okereke *et al.*, (2022) for samples of drinking water from different parts of Abia State, Nigeria. The highest value of dissolved solids in well water compared to the stream water samples was expected due to the solubility of certain mineral substances within the geologic materials which eventually found their way into the underground water. Leaching of solid materials from underground parent rocks under the influence of hydrolysis was also a reason for higher value of total dissolved solids in the well water. Below the ground level carbonate rocks such as limestone (CaCO_3) is one of the geologic rocks susceptible to gradual dissolution by groundwater. In the report of Chinedu *et al.*, (2011), a total dissolved solid of 100, 20, 30 and 20 mg/L was reported for Borehole, bottle sachet, rain and river water

respectively in Sango Ota, Nigeria. In another report on water quality, Okereke *et al.*, (2022) reported a range of 5.77 – 213mg/L of total dissolved solids in twenty-four (24) communities within Abia state, Nigeria. In the context of water quality, total dissolved solids refer to the combination of all inorganic salts, mainly calcium, magnesium, potassium, sodium, chlorides, bicarbonates, sulphates and organic matter that dissolve in water (Youcai, 2017).

Total suspended solid (TSS) is the dry weight of suspended particles which are not soluble in water. Total dissolved solid and total dissolved solids (TDS) are related in terms of analytical procedure. While TSS is the amount of insoluble substances trapped by separating filter, TDS is the total amount of dissolved substances which passes through the separating filter. The dissolved solids are generally recovered from the liquid after evaporation. The range of TSS obtained in the study is higher than the range (0.00 – 0.30mg/L) reported by Ezeweani and Ezeweani, (2019) for drinking water in communities within Ugbowo, Benin-City, Nigeria. On the other hand, Sharon *et al.*, (2011) reported a much higher values and range for different water samples in Sango Ota, Nigeria. The obtained TSS values in the investigated water samples are much below the permissible limit (20mg/L) for TSS as recommended by the World Health Organization standard.

The higher value obtained for electrical conductivity in the well water in comparison with surface water is not unconnected with the geological materials present in the underground water samples in the form of dissolved cations and anions. Conductivity is a measure of water's capacity to pass electrical flow. The potential of water to cause electrical flow is related to the concentration of ions in the water. The conductive ionic species are derived from dissolved salts and inorganic chemical species such as hydrogen carbonates, chlorides, sulphates, calcium, sodium, potassium and magnesium. A critical study of the result shows that the well water was higher in calcium, Cu, Fe, dissolved solids and phosphates compared

with samples from the various streams. The chemical species within the sample from well water produced higher current compared to samples from the various streams with lower concentration of ionic species. A range of 5.0 - 25 μ S/cm was reported by Ezeweani and Ezeweani (2019) from the analysis of drinking water in Benin City, Nigeria while Shalon et al., (2011) reported a range of 21.00 – 149.10 μ S/cm for water samples in Sang-ota Southwestern, Nigeria. In a separate report, Okereke et al., (2022) reported a range of 0.00-0.10 μ S/cm for electrical conductivity of water samples from 21 communities in Abia State, Nigeria.

Dissolved oxygen is an important parameter that determines water quality as most aquatic organisms depend on the oxygen in water to survive (Mohan et al., 2021). If the dissolved oxygen falls below 5mg/L, aquatic life becomes stressed and undergoes hypotoxic conditions which could lead to mass mortality. The low concentration of dissolved oxygen in the well water was the result of the low interface between the surface of water in the well and the surrounding air on soil surface. On the other hand, streams or rivers with rapidly moving water contain more dissolved oxygen than stagnant water due to the constant interface between surface water and the surrounding air. The DO values obtained in the studied water samples are below the range of DO (4.0-6.0mg/L) set by the World health organization for portable water.

Biochemical oxygen demand is the amount of oxygen consumed by bacteria and other microbes during the decomposition of organic matter under aerobic conditions. Specifically, it is the measurement of the quantity of oxygen required to remove waste organic matter from water during decomposition by aerobic bacteria. The higher the value of BOD, the greater the level of contamination of water with organic matter. This explains why the BOD value in the sample from well water was much lower than the values obtained in samples from the various streams that constantly receive diverse dead organic matter as they flow along their various paths. In most of the streams investigated, some of the inhabitants of the

communities where the streams are located have their bath, wash clothes and on some occasions deposit dead animals in the streams. The discharge of various forms of organic pollutants in the water bodies may be responsible for higher values of BOD in the various stream water than the permissible level (2mg/L) set by the World Health Organization. However, it is worthy of note to state that well water in general is not precluded from the possibility of water contamination with organic matter. If a well of water is left uncovered, it becomes susceptible to water contaminants of organic or inorganic nature. In the report of Shalon et al., (2011), a BOD value of 3.40mg/L was recorded for Iju river in Ogun State, Nigeria compared to BOD values reported for borehole, chlorinated swimming pool, tap water and sachet water which were less than 1.04 mg/L.

The hardness of water reflects high mineral contents. Hardness of water is formed when water percolates deposits of dolomite, gypsum, or limestone which are mainly compounds of calcium and magnesium carbonates, bicarbonates and sulphides. The higher value of hardness obtained in the well water was an indication of the deposition of the carbonates, bicarbonates, or sulphates of calcium /magnesium or both in the underground water. The hardness of the well water was however, below the permissible limit (120mg/L) set by WHO. The higher value of copper residue in the well water suggests fractional mobility of a proportion of accumulated copper residue in soil to the surrounding well. Most cocoa plantations in Nigeria are contaminated with copper residue due to long term application of copper-based fungicides (Aikpokpodion et al., 2010).

CONCLUSION

The scientific investigation carried out on the environmental impact of long-term use of pesticides in cocoa plantation was found to have had specific adverse effects on soil biota and soil health. Long term use of pesticides reduced microbial population in treated cocoa plantations. The continuous use of agrochemicals led to the inhibition of beneficial

bacteria species in cocoa plantations. The long-term use of pesticides in cocoa plantations reduced the level of Nitrogen and organic carbon in cocoa plantations while soil acidity was increased in cocoa plantations because of the long-term use of pesticides. Accumulation

of copper was high in cocoa plantations due to the long-term use of copper-based fungicides in the control of black pod disease. The long-term use of pesticides in cocoa plantations did not have significant effects on the quality of water samples evaluated.

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