Determination of Pesticide Residues in Edible Crops and Soil from University of Agriculture Makurdi Farm Nigeria

Part 1 in the series of pesticide residues

Peter O. Onuwa†**, Ishaq S. Eneji, Adams U. Itodo and Rufus Sha'Ato**

Corresponding Author**: peteronuwa@yahoo.com,**

Department of Chemistry, Federal University of Agriculture Makurdi, Benue State, Nigeria.

Abstract

Aims: To extract pesticide residues in some edible crops and soils from farm lands so as to determine their concentration in order to assess their safety status

Study Design: Solid-Phase extraction technique was used to extract pesticides content mainly of organochlorine class from some edible crop plants and soils as well as determining some main physicochemical parameters of the soils.

Place and Duration of Research: University of Agriculture, Makurdi commercial crop farms from in the month of July, 2015.

Methodology: Pesticide residues were extracted from edible crops (*Daucus carota*, *Capsicum Anuum*, *Telfairia occidentalis*, *Solanum lycopersicum*, *Amaranthus Hybridus*, *Solanum macrocarpon*) and soils using dispersive solid-phase extraction method. The extracts were analysed using GC-MS technique

Results: The mean concentration (mg/kg) of pesticides in soil samples were 12.1 ± 0.1, 0.09 ± 0.02, 0.12 ± 0.02, 14.9 ± 0.2 and 5.05 ± 0.2 for butachlor, aldrin, dieldrin, pendimethalin and propanil, respectively. Pesticides concentration in vegetables (µg/kg) generally ranged from 0.001 ± 0 - 1.64 ± 0.6, the highest value being cypermethrin and the least alpha-HCH. Their concentrations in each sample ranged in this order; *D. carota* (1.62 ± 0.01- 0.001 ± 0); *C. Anuum* (1.63 ± 0.7 - 0.002 ± 0.001); *T. occidentalis* (1.64 ± 0.6 - 0.001 ± 0.001); *S. lycopersicum* (0.84 ± 0.7 - 0.002 ± 0.01); *A. Hybridus* (1.61 ± 0.5 - 0.001 ± 0); *S. macrocarpon* (1.62 ± 0.01 - 0.001 ± 0), respectively.

Conclusion: Cypermethrin showed the highest concentration values in all vegetable samples while alpa-HCH has the least. Although all samples showed values within WHO maximum residue limits (MRLs) for spices, indicating that they are transiently safe, but it is necessary to have prolonged seasonal monitoring.

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Keywords: Edible crops, Organochlorine, residues, soil, solid phase extraction

1. Introduction

Much Concerns have been raised about risks of pesticide residues in food as most pesticides show a high degree of toxicity, especially in developing countries like Nigeria, where the European Union places a ban on some agricultural commodities for some times now. Food items banned from Europe till June 2016 include beans, sesame seeds, melon seeds, dried fish and meat, peanut chips and palm oil [1]**.** According to the European Food Safety Authority, the prohibited beans were found to have concentration of dichlorvos ranging from 0.03 to 4.6 mg/kg, where as the acceptable maximum residue limit is 0.01mg/kg [1]**.** Pesticides are formulated to eliminate certain organisms but they also generate some risks within the environment. Human health is often threaten by the presence of pesticides in vegetables and soils. Some of the pesticides of interest include cypermethrin (α-cyano-3-phenoxybenzyl-2,2-dimethyl-3-(2,2-dichlorovinyl)cyclopropanecarbo- ylate), butachlor, aldrin, dieldrin, pendimethalin and propanil and alpa-HCH. Cypermethrin belongs to a class of pesticides known as synthetic pyrethroid and it is generally used in combating insects in commercial agricultural practice. It is also used in consumer products for subsistence purposes [2]. Ectoparasites of cattle, sheep, and poultry are sufficiently controlled by the application of cypermethrin [3]. It is applicable in veterinary medicine for effective control of ticks on dogs (Somasani, 2014)· It acts as a neurotoxin in insects. It can be broken down with ease on soil and plants but its effects can be felt for some weeks if it is applied to indoor inert surfaces [2].When it is exposed to sunlight, water and oxygen, its rate of decomposition is facilitated. Cypermethrin is a very toxic pesticide to fish, bees and aquatic insects. Cypermethrin has relatively low dermal toxicity or when ingested. High exposure to it causes nausea, headache, muscle weakness, salivation, shortness of breath and seizures [4]. Measurement of metabolites of the urine serves as means of monitoring how much workers are exposed to the pesticide, while quantitation of cypermethrin present in blood or plasma may be confirmed by excessive dosage [5]. Aldrin as an organochlorine insecticide was generally available until the 1970s, when its use was sanctioned in most countries. However, it is still in use till now especially in some developing countries. Before aldrin was phased out, it was seriously deployed as a pesticide for treating seed and soil. Aldrin and related "cyclodiene" pesticides were well known as persistent organic pollutants [6]. Dieldrin is an organochloride originally produced as an insecticide in 1948. Dieldrin is similar to aldrin, which reacts further to form dieldrin [7]. Alpha-Hexachlorocyclohexane (α-HCH) is also an organochloride which is one of the isomers of hexachlorocyclohexane (HCH) [3]. It results from the production of lindane (γ-HCH) and it is still contained in commercial grade lindane which is used as insecticide. Lindane. United States has stopped the use of lindane for more than 20 years [8]. Lindane is a white, powdery solid substance, stable at ambient temperatures. The Stockholm Convention on Persistent Organic Pollutants classified (α-HCH) and (β-HCH) as Persistent organic pollutants (POPs), since 2009 due to the chemical's ability to remain for a long time in the environment, bioaccumulative, biomagnifying, and long-range transport capacity. Solid phase extraction (SPE) is one of the most commonly used sorbent techniques in analyzing pesticide residues. It has to do with the omission of extracts containing target analytes through a column filled with the desirable sorbent (which was previously conditioned by an appropriate solvent or solvent mixture), or passing the required solvent through the SPE column to which a suitable amount of sample was previously added) [9]. The coextractants from the SPE column can be eluted successfully if selective solvent are used, and then the target analytes or the elution of analytes can be direct, where undesirable coextractants derived from the sample matrix remain in the SPE column. SPE has many attractive features when compared with the traditional methods of extraction. It is easy to operate, less costly; it is being automated and uses small amounts of solvent [9]. Apart from this, SPE is the multifunctional techniques, since the purification and the concentration occur in the same step. However, some limitations are associated with, SPE technique. This has to do with lower yields (recovery), i.e. slightly lower sensitivity, in situations where there is "clogging" of the SPE column (blocking of the sorption centers by solid and oily components originating from the sample) [9]. The matrix nature has much influence on the solid phase micro-extraction (SPME) efficiency. Since the analytes distribution coefficients are partially determined by analytes-matrix interaction, appropriate matrix modification can increase the analytes partition coefficients. For instance, the presence of chloride and sulfate ion increases the ionic strength of the solution, which makes a large number of compounds less soluble [10]. The amount of analytes that would be adsorbed on the fiber will depend on the thickness and polarity of the active fiber layer, sampling mode (direct sampling - microextraction (DM-SPME) from solution, and headspace sampling–microextraction (HS-SPME) from gas phase, the nature of the sample and the analyte (analyte polarity, its molecular weight, pH value, nature of matrix), the mode and speed of the sample mixing, the solid phase micro-extraction (SPME) duration, the temperature at which it is performed, [11]. The most commonly used SPE sorbents in pesticide residues determination are: reverse- phase octadecyl (C_{18}), normal-phase aminopropyl ($-NH_2$) and primary-secondary amine (PSA), anion-exchanger three-methyl ammonium (SAX) and adsorbents such as graphitized carbon black (GCB). Normal-phase sorbents such as florisil ($MgSiO_3$), aluminum oxide (Al_2O_3) and silica (SiO_2) are usually applied in combination with the previously mentioned sorbents. The SPE cartridge should be chosen depending on the physicochemical properties of pesticides that are searched for in a particular sample, and the nature of the sample matrix (12). Researches indicate that the GCB sorbent is suitable for extraction of compounds of different polarity, and that it causes retention of pigments (carotenoids and chlorophylls) and sterols, and now are widely used for purification of plant extracts [12; 13]. On the other side, PSA sorbent proved to be effective in removing polar compounds and fatty acids, while the SAX is suitable for the removal of fatty and other organic acids and sugars [14]. Activated charcoal proved to be a good solution for removal of pigments from tomato extracts [15], and cabbage and carrots [16], while the florisil gave good results in pyrethroids determination in lettuce, cabbage, cauliflower, carrot, green pepper and green beans [17]. The most of the SPE methods are based on a combination of two or three cartridges. As GCB is suitable for removal of lipids, waxes and other nonvolatile, non-polar coextractants with high molecular weight, it is usually used in combination with other sorbent. (Solid-phase extraction) SPE technique was used by Pang *et al*. [18] for determination of 446 pesticides in some vegetable crops in Ghana and found out that the $C_{18}/GCB/-NH_2$ combination was the best. Except for slightly lower recoveries obtained for onion and leeks, the method was shown as good choice for all other samples) [9]. Pesticide residues have been determined in several food crops in Nigeria. This study aimed at adding another step in pesticide residues determination in commonly cultivated food crops within the university community as to ascertain their consumption safety and potential efficacy in agricultural markets as income drive of the university and state so as to advise the targeted consumers appropriately.

2. Study Area

The study was carried out at University of Agriculture commercial rice and vegetable farm sites designated as study site ‘A’ and ‘B’ respectively which are about 500 m apart. ‘A’ covers an area of 210 m (700 ft) long and 180 m (600 ft) width, while site ‘B’ covers an area of 77 m (258 ft) in length and 76 m (265 ft) width. The two sites A and B lie within latitude 7^0 47’ 0’’N, longitude

8^0 44’ 40’’E and latitude 7^0 39’ 10’’N, longitude 8^0 49’ 46’’E, respectively. These areas are found in the flood plain of river Benue which are usually surrounded by a large mass of sand stone and alluvium. The study areas were also found to be covered by a large deposits sandy soil, silt and laterites. The main crops commonly cultivated in sample site B include *Telfaria occidentalis* (Fluted Pumpkin) locally called *Ugwu*, *Solanum lycopersicum* (tomatoes), *Amaranthus hybridus* (African spinach) commonly called ‘green’, *Solanium marcrocarpon* (Garden egg), *Daucus carota* (Carrot plant), *capsicum* (red bell pepper) [fruits and leaves] locally called *Tar tase* soya beans etc.

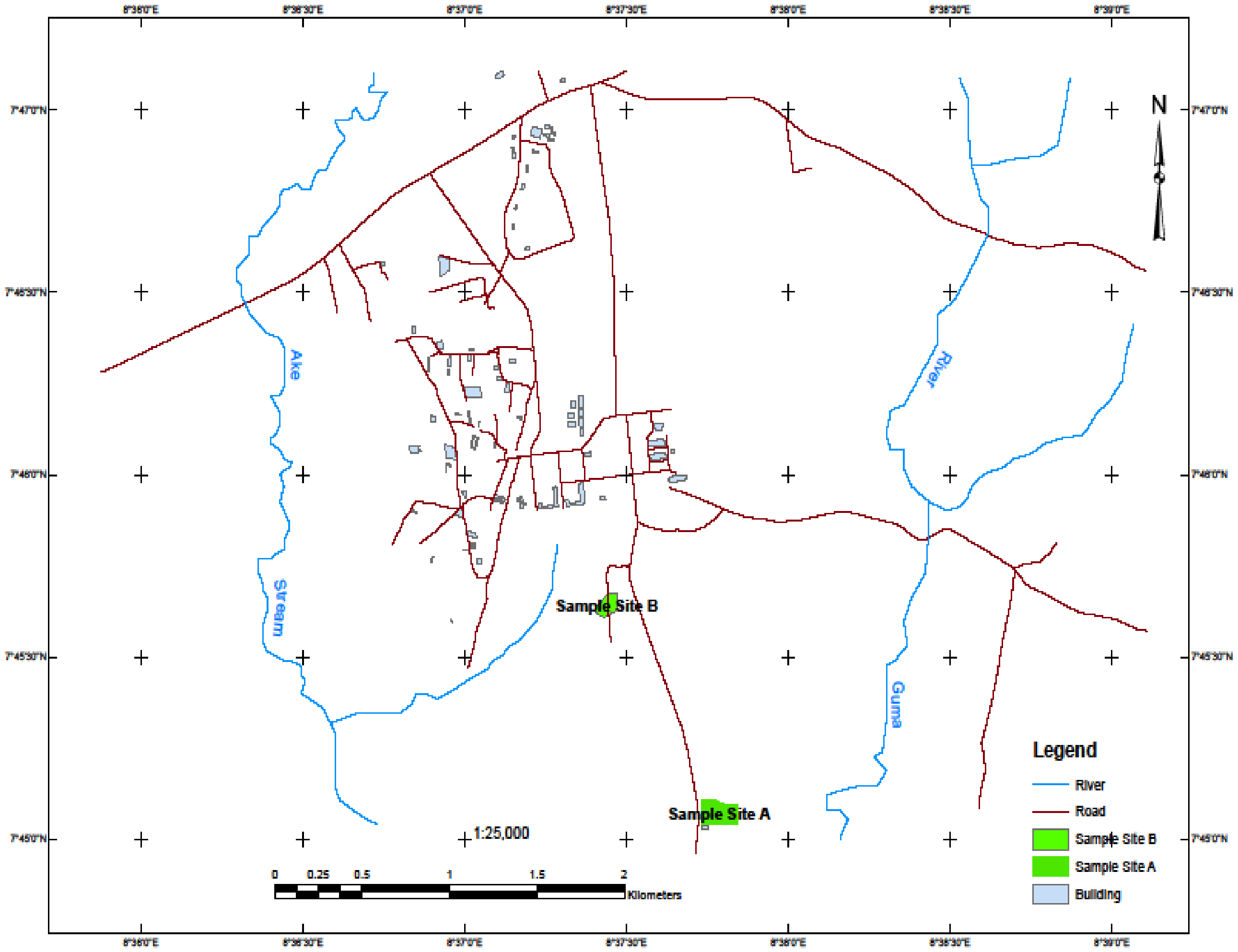


Figure 1: Map of South Core, University of Agriculture, Makurdi showing the Sampling Sites

3. Materials and Methods

Analytical grade acetone, sodium sulphate, ethyl acetate, n-hexane, sodium bicarbonate, silica gel and Flrorisil were supplied by Central Drug House (CDH), India. Pesticide quantification was achieved by using a GC-MS (Shimadzu QP-2010).

Laboratory Materials

All reagents were of analytical grade except otherwise stated

Sample Collection

(a) Soil Samples

Multiple soil samples were collected in July, 2015, within a depth of 0-15 cm layer using soil auger of 20.7 cm or 8.2’’ claw length. Each soil sample was a composite of fifteen subsamples of soil collected using random sampling method within a grid from a particular sample point and was mixed thoroughly together to obtain a homogenized representative sample. A grid was also established by locating the approximate centre of the field (rice farm) and dividing the farm in to three rows and columns, respectively, having a total of nine (9) paces apart according to a method described by Dem *et al.* [19] was established. Materials like roots, stones and pebbles were removed and bulked. The bulk sample was reduced to about 1 kg by dividing the thoroughly mixed sample into four equal parts. The two opposite quarters were discarded while the remaining two quarters were remixed and the process was repeated until the desired sample size was obtained. Nine soil samples were collected from sample site A’ i.e. rice farm while one sample was taken from vegetable plot, sample site ‘B’. Two control samples were obtained about 16 cm (54 ft) each away ahead and before the sampling station, respectively from sample site ‘A’ where no farming activity was expected to have taken place. Samples obtained were wrapped in aluminum foil, well labelled and transported to the laboratory in a bucket and stored at ambient temperature for further treatment.

(b) Vegetable Samples

Two replicate vegetable samples were obtained from tomato leaves and fruits *(S. lycopersicum)*, pepper leaves and fruits [red bell pepper fruit locally called ‘*tartase*’ (*Capsicum*), Ugwu leaves (*T. occidentalis*), garden egg leaves and fruits (*S. marcrocarpon*), carrot leaves (*D. carota*) and African spinach called ‘Green’ (*A. hybridus*) randomly from each of the selected points as distributed in the farm. A total of fourteen samples of food crops were obtained, wrapped in aluminium foil, clearly labeled and placed in polythene bags and then transported in a cooler to the laboratory.

3.1 Extraction of Pesticides

3.1.1 Soil

Extraction of soil samples was carried out by the method described by Ize-Iyamu *et al*. [20]. Exactly 10 g of each sample and 20 g of anhydrous sodium sulphate (Na_2SO_4) was ground into dry powder. The ground powder sample was extracted with 150 mL of a mixture of acetone and n-hexane (2:1). After extraction, the extract was transferred into a round bottomed flask connected to a pre-weighed receiver through a Liebig condenser and concentrated to about 20 mL on a water bath maintained between 50 and 55^0 C. The remaining solvent in the concentrated extract was evaporated using a rotary evaporator. The almost–dry extracts were cleaned up in a micro-column as described in America Society for Testing and Materials Standards (ASTM) [21]. Two (2) grams of activated silica gel was packed into a chromatographic micro-column of 10 mm internal diameter and approximately 10 cm long. The silica gel was conditioned with 10 mL n-hexane while the sample extracts were dissolved in 5 mL n-hexane before they were loaded on to the separate micro-column. Elution of each of the sample was carried out with 50 mL of ethyl acetate: n-hexane mixture (9:1). The eluents were then concentrated on a rotary evaporator at about 45^0 C and under a gentle stream of nitrogen gas. The almost-dry concentrates were then dissolved in 2 mL acetone and transferred into vials for injection into the Gas Chromatograph.

3.1.2 Vegetable

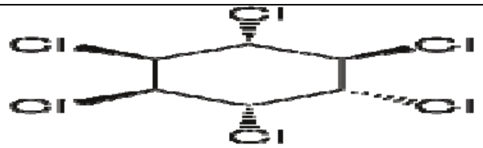
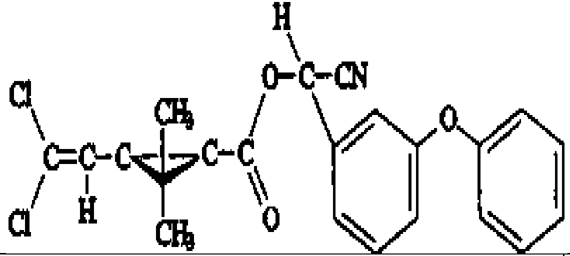
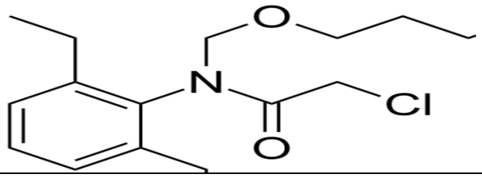
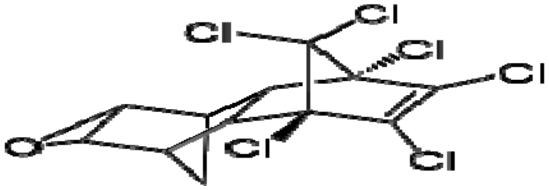
A method described by Ize-Iyamu [20] was adopted and modified. Each sample was chopped with a sharp knife on a chopping board. This was blended in a salton elite glass blender to obtain a homogenous representative sample. The knife, chopping board and blender were washed with distilled water to avoid cross contamination. Twenty (20) grams of sodium sulphate (Na_2SO_4) and 5 g of sodium hydrogen carbonate (IV) ($NaHCO_3$) were weighed and added to a separatory funnel. Samples to be blended (10 g) were added followed by 40 mL of ethyl acetate ($CH_3COOCH_2CH_3$). The mixture was shaken for ten minutes and allowed to settle. The supernatant (extract) was decanted into a round bottomed flask. This was evaporated in a BUCHI Rotavapor R-200 rotary evaporator [20].

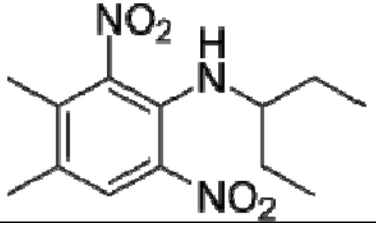
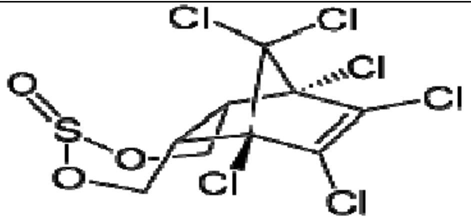
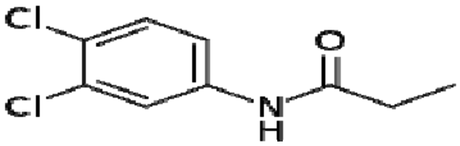
3.1.3 Cleanup Process

Ize-Iyamu *et al*. [20] described a method of clean up which was adopted. In order to remove any interfering substances co-extracted with the pesticide residues, the extract will be cleaned up. Activated florisil (1.5 g) was packed into a column that was plugged with glass wool. The column was further packed with 0.5 and 1.0 g of activated charcoal and sodium sulphate (Na_2SO_4), respectively. 10 mL of ethyl acetate was used to condition the column prior to clean up. Extract was transferred unto a florisil column using a Pasteur pipette and then waited until it was eluted. The sample tube (round bottomed flask) was rinsed with 2 mL ethyl acetate and was repeated twice to get all the extract from the tube. 9 mL of ethyl acetate was put on the column after the last rinse. The solvent (extract) was concentrated (evaporated) to dryness in a Rotavapor R- 200 rotary evaporator. The dried residue for each sample was dissolved in 1.5 mL of isooctane and picked into GC vials for analysis.

4. 0 Results and Discussion

Table 1: Selected Properties of Pesticide Residues Determined

Pesticide	Structure	Properties
[22 (α-HCH): α-1,2,3,4,5,6-hexachlorocyclohexane		Formula: $C_6H_6Cl_6$, molar mass: 290.83 g/mol, melting point:156-161^0, assay: 99 %, solubility: 1000 µg/mL in methanol
[22] Cypermethrin: (R,S)-alpha-Cyano-3-phenoxybenzyl-2,2-dimethyl(1R, 1S)-cis,trans-3-(2,2-dichrovinyl) cyclopropane-carboxylate)		Formula: $C_{22}H_{19}Cl_2NO_3$, Molar mass: 416.30 g/mol, **Purity:** commercial prepara tion (94.2%). **Melting point:** 80.5°C, **Vapour pressure:** 1.9x10^{-7} pascals at 20°C, **Solubility (g/l at 20°C): Density:** 1.23 kg/l at 20°C, Octanol-water partition coefficient **(P):** 2.0 x 10^6
[22]Butachlor :*N*-(Butoxymethyl)-2-chloro-*N*-(2,6-diethylphenyl)acetamide		Formula: $C_{17}H_{26}ClNO_2$, molar mass:311.85 g mol $^{-1}$, appearance: Light yellow oil, density: 1.0695 g/cm^3, solubility in H_2O: 20 mg/L (20 °C), LD_{50} (median dose) 1740 mg/kg.
[23] Dieldrin: 1,2,3,4,10 ,10-hexachloro-6,7-epoxy-1,4,4α,5,6,7,8,8α-octahydro-1,4-endo,exo-5,8-dimethanonaphthal ene		Formula: $C_{12}H_8Cl_6O$, molar mass: 380.91 g/mol, appearan ce: colourless to light tan crystals, density: 1.75g/cm^3, melting point: 176 to 177 °C, boiling point: 385 °C, solubili y in H_2O: 0.02 %

Pendimethalin (3,4-Dimethyl-2,6-dinitro-*N*-pentan-3-yl-aniline)		Formula:$C_{13}H_{19}N_3$O4, molar mass: 281.31 g·mol^{-1}, density: 1.17g/cm^3, melting point: 47 to 58 °C (117 to 136 °F; 320 to 331 K): boiling point: 330 °C (626 °F; 603 K): solubility in H_2O: 0.275 ppm
[23] Aldrin (1,2,3,4,10,10-Hexachloro-1,4,4a,5,8, 8a-hexahyd ro-1,4:5,8-dimethanonap hthalene)		Formula: $C_{12}H_8Cl_6$, molar mass: 364.90 g·mol^{-1}, density: 1.60 g/mL, appearance: colourless, melting point: 104 °C (219 °F; 377 K), solubility in H_2O: :slightly soluble (0.003, vapour pressure: 7.5 × 10^{-5} mmHg @ 20 °C
Endosulfan: (6,7,8,9,10,10-Hexachl oro-1,5,5a,6,9,9a-hexah ydro-6,9-methano-2,4,3-benzodioxath iepine-3-oxide		Formula: $C_9H_6Cl_6O_3S$, molar mass: 406.90 g·mol^{-1}, appearan ce: Brown crystals, odour: slight sulfur dioxide odour, density: 1.745 g/cm^3,melting point: 70 to 100 °C (158 to 212 °F; 343 to 373 K), Boiling point: decomposes, solubility in H_2O: 0.33 mg/L, vapour pressure: 0.00001 mmHg (25 °C)
Propanil: *N*-(3,4-Dichlorophenyl)propanamide		Formula: $C_9H_9Cl_2NO$, molar mass: 218.08 g/mol, appearance: White crystalline solid (pure), Brown powder (impure), melting point: 91 to 93 °C (196 to 199 °F; 364 to 366 K), solubility in H_2O: 225 ppm

The physico-chemical properties of selected pesticides and their chemical structures are presented in **Table 1.**

4.1 Quality Assurance Parameters/ Validation of Analytical Process

Vegetable Crops

To ensure good data quality, the efficiency of the analytical method (the extraction and clean-up methods) was determined by recoveries of an internal standard. One blended vegetable sample was spiked with a 50 µL of 100 ng/mL internal standard (isodrin) and extracted under the same conditions as the analytes. To check for cross contamination and interferences, a blank sample was analysed.All glassware used for the analysis were washed detergent, then hot water, rinsed with tap water and then with distilled water. They were further rinsed with acetone then kept till the next day at 40^0 C in an oven.

Analysis: The residues were analysed by Varian gas chromatography CP-3800 equipped with ^{63}Ni electron capture detector that allowed the detection of contaminants even at trace level concentrations (in the lower micrograms per gram range). The GC conditions and the detector response were adjusted so as to match the relative retention time and response. The conditions used for the analysis are summarised in **Table 2**.

Table 2: Analytical conditions for pesticide analysis in edible crops using GC-MS

Analysis Conditions	Description
Column	Capillary column coated with ZB-5 (30 m*0.25 mm, 0.25 Nm film thickness)
Carrier gas and make-up gas	Nitrogen
Flow rate	1.0 and 29 mL/min. respectively
Injector temperature operating at spitless mood	225 ^{0}C
Oven temperature	225 ^{0}C
Detector temperature	300 ^{0}C
Column oven temp.	60 ^{0}C for 2 min. & at 180 ^{0}C/min. Up to 300 ^{0}C
GC injection volume	1.0 nL

4.2 Soil Analytical parameters

The internal standard technique was employed to analyse the extracted samples. The organochlorine standard containing a mixture of eight (8) organochlorine compounds of high purity (alpha HCH, butachlor, aldrin, cypermethrin, dieldrin, pendimethalin, endosulphan, and propanil) were prepared at concentrations ranging from 0.10 to 2.00 ppm, with anthracene added as internal. The modern Shimadzu GC-MS QP-2010 was employed in analysing the standard. Both the control and main soil samples extract from clean-up were then analysed under the same conditions as the standard. They were analysed using the Selective Ion Mode (SIM) with m/z values ranging from 65 to 264. The efficiency of the method was validated using recovery studies. The reference materials were fortified with four organochlorine compounds at two concentration levels at 0.1 and 1.0 mg/kg without pesticide residues. This was carried out in triplicates and the same method of extraction and clean-up was followed. The observed retension times (RT) for each organochlorine compound in the standard under the conditions used as well as the percentage recoveries of selected pesticides are given in **Tables 3** and **4**, respectively. Satisfactory results were obtained for all the pesticides at the two fortification levels. The recovery of the organochlorine pesticides was in the range of 80 – 97 % and 83 – 91 % for 0.1 and 1.0 mk/kg, respectively. These results showed that the method has suitable range with good reproducibility.

Table 3: Retention time of organochlorine standards

Standard	Alpha-HCH	Butachlor	Aldrin	Cypermethrin	Dieldrin	Pendimethalin	Endosulfan	Propanil
Retention time (min)	8.31	8.89	9.58	10.00	10.64	11.43	12.10	12.61

Table 4: Percentage recovery of selected organochlorine pesticides

Pesticide	Retention Time (min.)	Fortification (mg/kg)	% Recovery
Butachlor	8.89	0.1	97
		1.0	90
Cypermethrin	10.00	0.1	93
		1.0	91
Endosulfan	12.10	0.1	80
		1.0	83
Propanil	12.61	0.1	85
		1.0	87

Table 5: Physico-chemical properties of soil from Sampling Sites

			Particle size distribution					
S/No	**Sample ID**	**pH†**	**% sand**	**% clay**	**% silt**	**Cond. µScm^{-1}**	**CEC (Cmolk^{-}**	**Bulk Density (g.cm^{-3})**
1	A	7.16	65.4	20.6	14.0	872	7.79	1.40
2	B	7.30	75.4	16.6	8.00	116	8.12	1.36

pH†: pH of samples in water, Cond.: Conductivity

Table 5 shows some major physico-chemical parameters of soil samples taken from both sites. Particles size distribution was determined using a method described by Gee and Or [24]. Bulk density determination was also carried out according to standard procedure described by Blake and Hartge [25]. The pH in both sites was determined to be comparatively close and remained slightly alkaline (7.16 and 7.30). A neutral to moderately alkaline (6.5 to 8.0) pH value was also reported by [26]. The physical and chemical characteristics of the soil system includes moisture content, organic matter, clay contents, and pH, which have influence on the sorption/desorption and degradation of pesticides and their access to groundwater and surface waters [27]. The bulk density (gcm^{-3}) have close values of 1.40, 1.36, for sites 'A'and 'B' respectively. Nwite [28] reported values of 1.2 ± 1.00 for forest land and 1.30±0.15 for continuous cropping system within a soil profile depth of 0-15 cm in a similar research carried out in Abakaliki, Nigeria. Cation exchange capacity, CEC (Cmolk^{-1}) have very close range of values and 7.79, 8.12 for sample sites A and B, respectively. AariffKhan and Kamalakar [26] also reported low to medium in CEC (11.5 – 26.5 C mol (p+) kg-^{1}) which was only slightly higher. Electrical conductivity ranged between 116 – 872 µScm^{-1} for sites A and B. Soil sample in site A has high sandy content (65.4 %) with relatively higher percentage of clay and silt (20.6 and 14.0 %) as compared to site B. Perhaps, this necessitates its use for growing of paddy rice, a rice variety commonly known as ''CP Farrow 44'' as reported by the farmer officially in charge of the university rice farm in the site. Soil sample in site B has higher percentage of sandy soil (75.4 %) than A with relatively lower clay content (16.6 %) followed by silt (8.00 %). Nwite [28] also reported values of 38 ± 0.7, 14 ± 0.7 and 48 ± 0.7 for sand, clay and silt for continuous cropping system in Ghana within same soil depth. Variations in soils properties could be attributed to the nature of parent materials [29]. The physical properties of Soil influence the occurrence and growth the movement through soil of water and its dissolved nutrients and chemical pollutions [20, 31]. The fate of pesticides in soil and water environments is influenced by the physico-chemical properties of the pesticide, the properties of the soil and water systems (presence of clay materials, organic matter, pH), climate, biology, and other factors [32].

Table 6 shows the mean concentration (mg/kg) of pesticides residues present in Edible crops. In *D. carota* (carrot), Cypermethrin has the highest residue concentration of 1.62 ±0.01 mg/kg, while pendimethalin has a close concentration of (0.58 ± 0) mg/kg. Alpha-HCH has the least concentration of residues in this sample (0.001 ± 0); they are in order of; Cypermethrin > pendimethalin > propanil > butachlor > Alpha-HCH. However, aldrin, dieldrin and endosulfan residues were not detected. The distribution of the entire pesticide residues concentration in this crop ranged between 1.62 - 0.001 mg/kg. **Table 6** also shows the mean concentration (mg/kg) of pesticide residues in *C. anuum* (fruits and leaves). The pesticide residue concentration (mg/kg) ranged between 1.63 ± 0.66- 0.002 ± 0.001. Cypermethrin indicated the highest mean concentration (1.63 ± 0.66) while alpha-HCH gave the lowest residual concentration (0.002 ± 0.001) mg/kg. Aldrin, dieldrin and endosulfan were not detected as well. Others with residual concentration (mg/kg) decreasing progressively are pendimethalin, propanil and butachlor (0.61 ± 0.24, 0.07 ± 0.03 and 0.03 ±0.01), respectively. Cypermethrin exceeded the maximum residue limits of 0.1 mg/kg in fruits or berries [33. Cypermethrin is not soluble in water and has a strong tendency to adsorb to soil particles [34]. There are four major routes through which pesticides reach water body, it may percolate, or leach, through the soil, it may be carried to the water as runoff, or it may be spilled. Factors that affect a pesticide's ability to contaminate water include its water solubility, the distance from an application site to a body of water, weather, soil type, presence of a growing crop, and the method used to apply the chemical [35, 36, 37]. *Telfaria occidentalis* was analysed for pesticide residual content. *T. occidentalis* was analysed for pesticide residual content. The mean concentration (mg/kg) of each pesticide residue in the sample is also presented in **Table 6**. The pesticides ranged from concentrations (mg/kg) of 1.64 ± 0.6 - 0.001 ± 0.001, which represents cypermethrin and alpha-HCH, respectively. The concentrations (mg/kg) of other pesticide residues in order of magnitude are 0.55 ± 0.2, 0.06 ± 0.02, and 0.02 ±0.01 for pendimethalin, propanil and butachlor, respectively. Rao *et al*. [38] also observed that very low levels of butachlor residues were detected in rice grain below the maximum residue limit of 0.5 mg/kg. Maximum residue value of 0.0095 mg/kg was observed at 2 kg/ha of herbicide (with poultry manure) during wet season. However, in both seasons and in all the treatments, the detected residues were below maximum residue limits of 0.05 µg/g [39]. Aldrin, dieldrin and endosulfan were not detected in any of the *T. occidentalis* (ugwu leaves) and fruits designated as UL. **Table 6** shows the mean concentration (mg/kg) of pesticide residues in *S. lycopersicum* (garden egg). Pesticides ranged from a concentration (mg/kg) of 0.84 ± 0.7- 0.002 ± 0.001. The highest concentration being cypermethrin and the lowest being alpha-HCH. Other concentration (mg/kg) in order of magnitude are 0.29 ± 0.2, 0.07 ± 0.02, and 0.03 ± 0.01 for pendimethalin, propanil and alpha- HCH, respectively. Aldrin, dieldrin and endosulfan were not detected in any of the tomato leaves and fruits designated as TL and TF. The residual concentration (mg/kg) of pesticides in *A. hybridus* (African spinach) is shown in **Table 6**. The concentration of the pesticide residues ranged from 1.61± 0.5 - 0.001 ± 001, which represents cypermethrin and alpha-HCH pesticides, respectively. The concentration of other pesticides in decreasing order are 0.58±0.2, 0.06±0.02 and 0.02 ± 0.01 for pendimethalin, propanil and butachlor, respectively. Aldrin, dieldrin and endosulfan were not detected in the vegetable sample, meaning that it was absent in the cultivated soil and consequently the crop. Pesticide residues analysis was also carried out to find out the residual concentration in *S. macrocarpon* (garden egg fruits and leaves). The mean concentration of each incipient is presented in **Table 6** as well. The concentration (mg/kg) decreases from cypermethrin (1.62 ± 0. 01**),** pendimethalin (0.57 ± 0.01)**,** propanil (0.06 ± 0.01), butachlor (0.02 ±0.01) and alpha-HCH (0.001 ± 0), being the least**.** Aldrin, dieldrin and endosulfan were also not detected in this sample. A two-tailed Pearson correlation analysis was carried out (at p = 0.01 and 0.05) to show relationships among individual pesticides in various vegetable crops. The results were significant between butachlor and alpha-HCH, pendimethalin and cypermethrin (p = 0.01) with a very high positive values (r = 1.000 and 0.985), respectively while it was significant between prapanil and alpha-HCH, propanil and butachlor (p = 0.05) with high positive correlation value (r = 0.857), respectively. The high positive values obtained suggest strong interaction between these pesticides in the crops, indicating that the presence of one quantitatively induces the availability of its pairs. Others pesticides showed high negative correltion values indicating absence of meaningful interaction among them. The presence of these pesticides may not be wholly attributed to only direct application within the farm site. Aldrin, dieldrin, endrin, heptachlor, hexachlorobenzene are Persistent organic pollutants (POPs) which resist degradation and thus remain in the environment for years. Some POPs have the ability to volatilize and travel great distances through the atmosphere to become deposited in remote regions. Such chemicals may have the ability to bioaccumulate and biomagnify and can bioconcentrate up to 70,000 times their original concentrations [40]. These can affect non-target organisms in the environment and increase risk to humans by disruption in the endocrine, reproductive, and immune systems. Other plant (branching habit, plant Surface, plant species and varieties, Plant Maturity or Age of the Plant) as well as environmental (temperature and Humidity, rain fall, wind, light) factors also influenced pesticides distribution in crops [40].

Table 6: Mean Concentration of Pesticides residues present in Edible Crops

Pesticide	**Mean Concentration (mg/kg) in Edible Crops**					
	Daucus carota	*Capsicum anuum*	*Telfaria Occidentalis*	*Solanum lycopersicum*	*Amaranthus Hybridus*	*Solanum macrocarpon*
Alpha-HCH	0.001 ± 0	0.002 ± 0.001	0.001±0.001	0.002 ± 0.001	0.001±001	0.001±0
Butachlor	0.02 ± 0.01	0.03 ± 0.01	0.02 ±0.01	0.03 ± 0.01	0.02±0.01	0.02 ±0.01
Aldrin	ND	ND	ND	ND	ND	ND

Cypermethrin	1.62 ± 0.01	1.63 ± 0.70	1.64 ± 0.60	0.84 ± 0.70	1.61±0.50	1.62 ± 0. 01
Dieldrin	ND	ND	ND	ND	ND	ND
pendimethalin	0.58 ± 0	0.61 ± 0.20	0.55 ± 0.20	0.29 ± 0.20	0.58±0.20	0.57 ± 0.01
Endosulfan	ND	ND	ND	ND	ND	ND
Propanil	0.05 ± 0	0.07 ± 0.03	0.06 ± 0.02	0.07 ± 0.02	0.06±0.02	0.06 ± 0.01

ND: Not detected

The total mean concentration (mg/kg) of pesticide residues in soil for sites 'A' and 'B' are presented in **Table 7.** Organochlorine pesticide residues were detected in almost all the soil sample except Alpha-HCH, cypermethrin and endosulfan which were not detected in all soil sample from site A but were detected in samples from site B. Endosulfan was only determined in very small quantity in soil from the vegetable farm but was not detected in any of the vegetable crops. This indicates that the pesticide was probably applied on soil from site B or washes off from nearby vicinity where micro-farming and students' domestic activities from residence take place but was not used on the commercial rice farm (site A). Endosulfan was present in similar research conducted in Ghana, where human health risk assessment was performed on the results obtained. The analysis indicated cancer risk for adults and children as a result of the presence of endosulfan and chlopyrifos. Endosulfan is not registered in Ghana as a pesticide for use on vegetables, therefore the detection of endosulfan in several samples indicates misuse of agrochemicals among Ghanaian farmers [41]. Conversely, aldrin and dieldrin were not detected in soil from sample site B, but were present in all soil samples from site A (rice farm), meaning that they were preferably applied only to the rice during cultivation but not applied to the vegetable crops. However, hazards associated with aldrin are that they are potential occupational carcinogen [42. The predominant pesticides analysed were propanil, butachlor and pendimethalin. Pendimethalin gave the highest mean concentration (mg/kg) of 13.8 ± 3.8, followed by butachlor (11.0 ± 3.3) and propanil (4.76 ± 0.94). Both butanol and propanil were above the residue limits. Cypermethrin, which was absent in soil samples from site 'A' was sufficiently present and ranged the highest in soil samples from site 'B'. Two control soil samples (CA1 and CA 2) were taken from extremes sides of site A, where farming activities were expected to be low or has not taken place over a long period of time. No significant pesticides concentration was obtained as they were below the instrument detection limit except butachlor, propanil and pendimethalin having the concentration of 0.01, 0.01 and 0.02, respectively in CA1 only. The farmer in charge of the university rice farm reported that 2,4, D-amine combined with Nominee gold (a post emergent, broad spectrum systemic herbicide for all types of rice cultivation i.e. direct sown rice, rice nursery and transplanted rice) were regularly used in the farm as selective post emergence weed control pesticides against rice within cropping seasons. Two-tailed Pearson's correlation analysis was carried out to infer possible interactions among pesticides in different soil sites (p = 0.01, 0.05), respectively. The concentration of butachlor and alpha-HCH was significant but high negative value (-0.999 at p = 0.01) as presented in **Table 8**. This indicates that as the concentration of one increases, the other reduces, a negative interaction. This could possibly suggests a side reaction or chelating property that limits availability of the other within the substrate or the vicinity. The same effect was observed between aldrin and alpha-HCH, pendimetalin and alpha-HCH, endosulfan and butachlor, endosulfan and aldrin, propanil and alpha-HCH, propanil and endosulfan. A significant positive correlation values were observed (r = 1, 0.999, 1) between cypermethrin and alpha-HCH, pendimethalin and butachlor as well as endosulfan and cypermethrin), respectively. This shows active interaction within the soil. How long the pesticide remains in the soil depends on how strongly it is bound by soil components and how readily it is degraded [27]. It also depends on the environmental conditions at the time of application, *e.g.*, soil water content, temperature and Humidity, rainfall, wind, light.

Table 7: Total Concentration (mg/kg) of Pesticide Residues in Soil for both Sites

	Sampling Sites									
Pesticide	A1	A2	A3	A4	A5	A6	A7	A8	A9	B1	Mean ± SD
Alpha-HCH	ND	ND	ND	ND	ND	ND	ND	ND	ND	0.51	0.51 ± 0
Butachlor	12.0	12.0	12.0	11.9	12.3	12.1	12.1	12.2	12.2	1.73	11.0 ± 3.3
Aldrin	0.09	0.10	0.12	0.09	0.05	0.11	0.10	0.06	0.08	ND	0.09 ± 0.02
Cypermethrin	ND	ND	ND	ND	ND	ND	ND	ND	ND	4.45	4.45 ± 0
Dieldrin	0.12	0.13	0.13	0.14	0.11	0.09	0.12	0.1	0.13	ND	0.12 ± 0.02
pendimethalin	14.6	15.2	15.0	14.7	15.1	15.1	15.1	15.0	14.8	3.05	13.8 ± 3.8
Endosulfan	ND	ND	ND	ND	ND	ND	ND	ND	ND	0.01	0.01 ± 0
Propanil	4.83	5.23	5.13	4.57	5.09	5.16	4.94	5.33	5.15	2.15	4.76 ± 0.94

ND: Not detected

Table 8: Pearson correlation analysis of pesticides in soil sampling sites

	Alpha-HCH	**Butachlor**	**Aldrin**	**Cypermethrin**	**Dieldrin**	**Pendimethalin**	**Endosulfan**	**Propanil**
Alpha-HCH	1							
Butachlor	-0.999 **	1						
Aldrin	-0.797**	0.781**	1					
Cypermethrin	1.000**	-0.999**	-0.797**	1				
Dieldrin	-0.124	0.103	0.110	-0.124	1			
Pendimethalin	-0.999**	0.999**	0.798**	-0.999**	0.100	1		
Endosulfan	1.000**	-0.999**	-0.797**	1.000**	-0.124	-0.999**	1	
Propanil	-0.973**	0.977**	0.756*	-0.973**	-0.058	0.979**	-0.973**	1

** Correlation is significant at the 0.01 level (2-tailed)
* Correlation is significant at the 0.05 level (2-tailed)

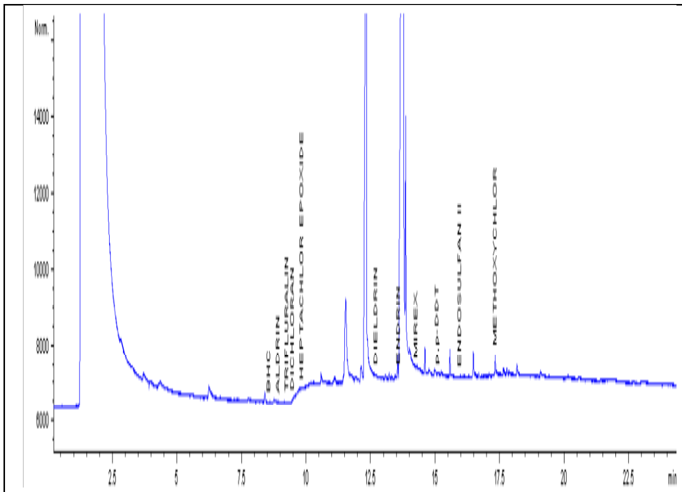


Figure 2: GC-MS Chromatogram of Soil Sample in Site A1

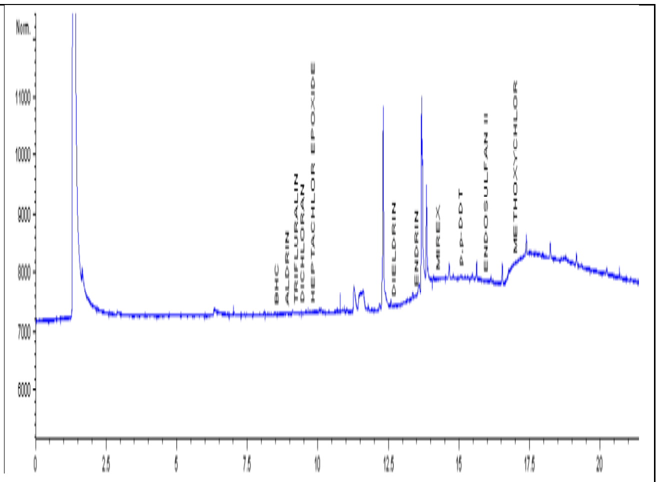


Figure 3: GC-MS Chromatogram of Soil Sample B 2

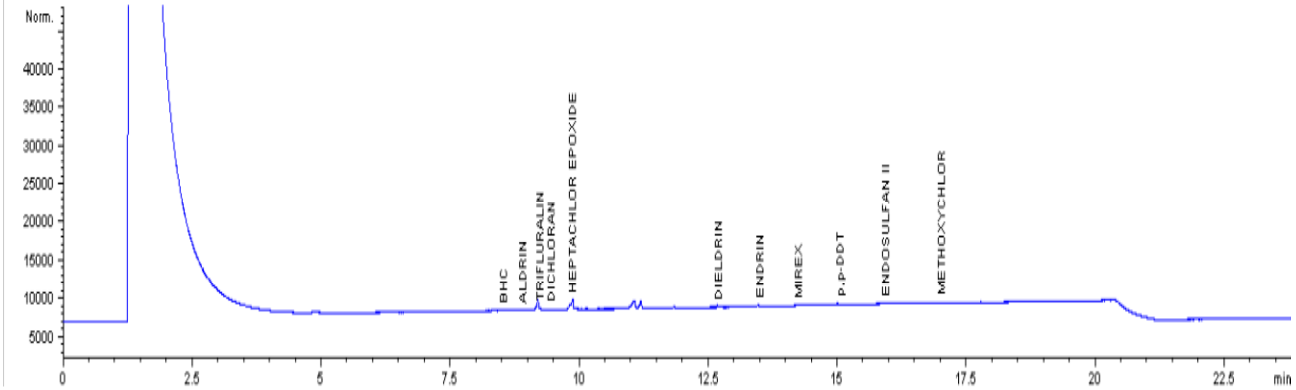


Figure 4: GC-MS Chromatogram of Control Soil Sample (A1)

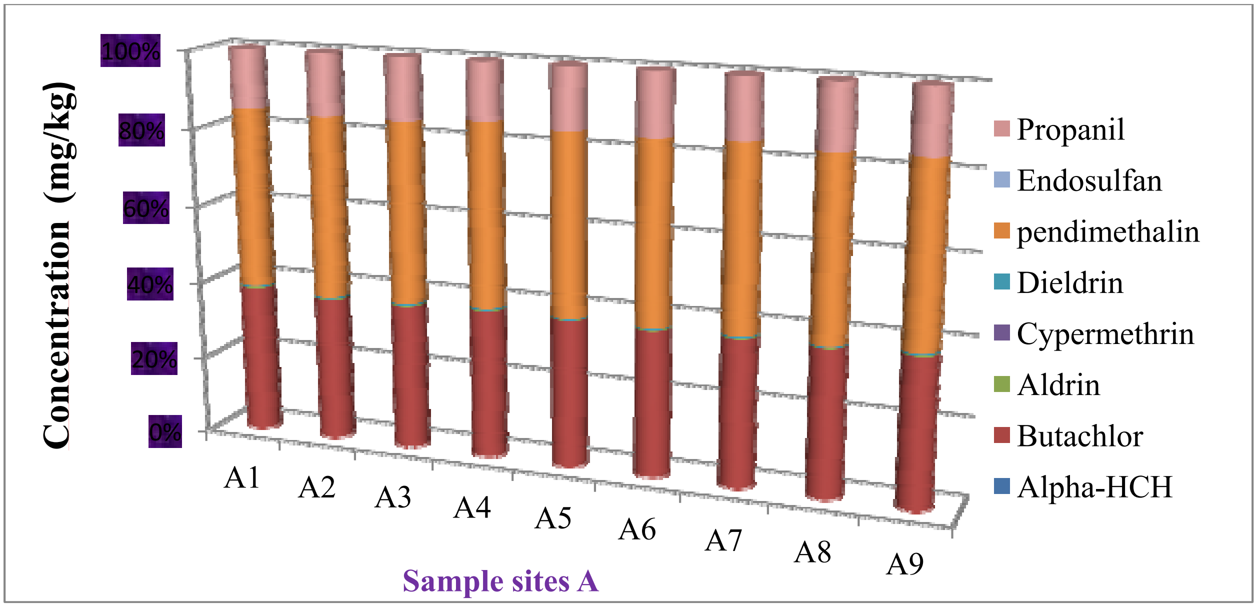


Figure 5: Concentration (mg/kg) of pesticide residues in each soil sample in Site A

Figure 2 shows the Chromatogram of Soil Sample in Site A. The chromatogram of soil sample taken from Sample B is shown in **Figure 3. Figure 4** shows the chromatogram of control soil Sample (A1) also taken from the sampling site A.

The mean concentration of pesticide residues in individual soil sample taken from site A is presented in **Figure 5**. The mean concentration (mg/kg) of pesticide residues in soil sample in site B is shown in **Figure 6**. Cypermethrin showed the greatest peak, indicating highest concentration followed by pendimethalin, propanil, butachlor and alpha-HCH. Aldrin and dieldrin were not detected in all the soil samples from this site as they were reportedly not applied. No good linear relationship was statistically observed between pesticide concentration and distribution as the square of correlation coefficient was (r = - 0.08), indicating a very low negative correlation. This is because the concentration of pesticides was only based on those ones selectively applied and consequently present in the soil for adsorption.

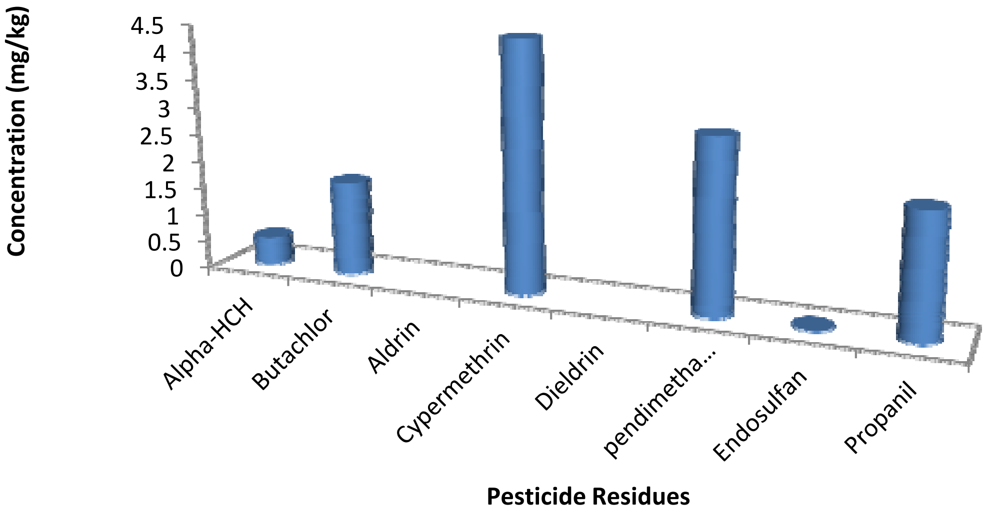


Figure 6: Mean Concentration (mg/kg) of pesticide residues in soil samples in Site B

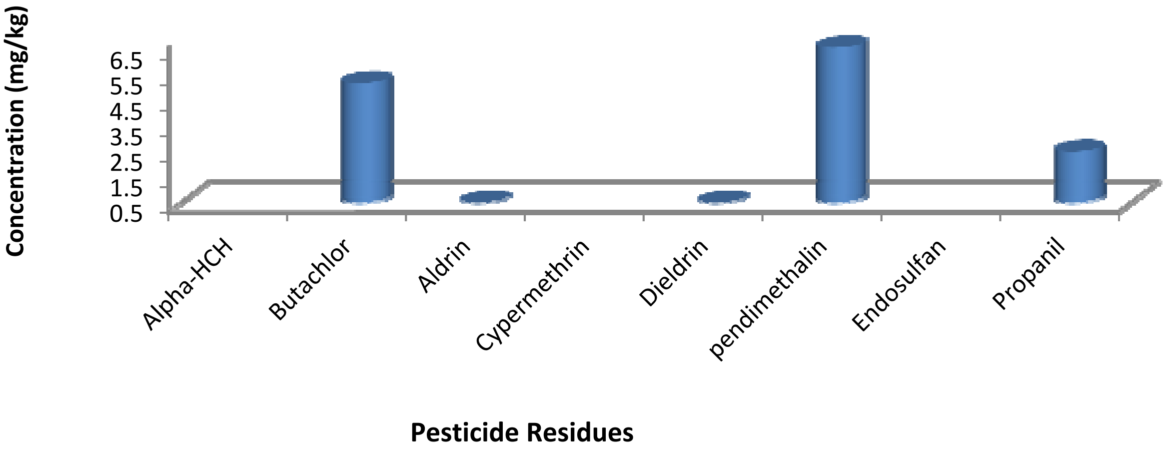


Figure 7: Mean Concentration (mg/kg) of pesticide residues in soil samples in Site A

Figure 7 shows the mean concentration (mg/kg) of all the pesticide residues present in soil samples in site A. Pendimetalin gave the highest concentration peak followed by butachlor, propanil, dieldrin and aldrin which are close in terms of the magnitude of their values. Alpha-HCH, cypermethrin and endosulphan were not detected. The consistent absence of these pesticides in sample site A indicated that they were not applied to the commercial rice farm within cropping seasons. Conversely, aldrin and dieldrin which were completely not detected in site B were present in small quantities in the rice farm (Site A).

5. Conclusion

Alpha-HCH, cypermethrin and endosulphan were not detected in all soil samples within the commercial rice farm (Site A) but were detected in the vegetable farm plot (Site B). Aldrin and dieldrin were not detected in site B were present in the rice farm in comparatively little amount. This generally indicates the selective use of these pesticides is based on the crops grown in each of these locations. However, run offs of effluents from the neighbouring farming environment cannot be over ruled. Therefore, the university farmers who are expected to be more enlightened should endeavour to sensitise the neighbouring communities as how to apply these chemicals to alleviate their residual toxicity to unsuspected consumers. The mean concentration (mg/kg) of all the pesticides of interest were within MRL, except cypermethrin in edible crops meaning that the concentrations of these pesticide residues are safe for now in many of the food crops, however, continuous research should be endorsed.

Acknowledgements

The authors wish to thank the following Postgraduate Diploma students of department of chemistry for participating in the sampling and sample preparation processes in the laboratory; Terver Tyongu Hir, Jerry Akowe, Alade Patience Otunsha and Mary Oyilema Onoja as well as the laboratory technologists, Mr. Julius for mutual cooperation during laboratory work.

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