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(RESEARCH ARTICLE)

Monitoring and assessment of pesticide residue in cucumber from four selected markets in Port Harcourt

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Abstract

Investigation of pesticide residues of cucumber was carried out in Choba, Borokiri, Mile 1 and Mile 2 markets, Port Harcourt. The pesticide residues of fruits obtained in open markets were monitored and compared with established safety limits. The pretreated samples were Soxhlet extracted and analysed for groups of pesticide most especially OCPs content using a Gas Chromatograph coupled with an Electron Capture Detector (GC-ECD). A total of 12 OCPs and other residues were detected in most of the samples with the percentage occurrence ranging from 35% - 100%, and the levels found exceeding their Maximum Residue Limits (MRLs). The MDL of the samples was 0.0408 mg/kg α Chlordane and 0.0723 mg/kg endosulfan sulfate; while the limit of quantitation (LOQ) of these pesticides were 0.0047mg/kg and 0.3952 mg/kg respectively. Among the pesticides determined 58.7% insecticides (14), 22.7% fungicides (5) and 17.9% Herbicide (4) were detected in the tomato and cucumber samples. The mean concentration of Metoxychlor was excessively higher in samples obtained across the four markets. Also, the concentration of O'ρ Pρ, DDT (0.18±0.20) was higher than its metabolite, (0.13±0.03) in Oil Mill and Mile 3 markets (0.14±0.02) which might portend danger for our health.

Keywords: MDL; LOQ; Pesticides; Residues; Markets; Fruits

1. Introduction

Cucumber, (*Cucumis sativus* L.) that belongs to the family *Cucurbitaceae* and important vegetable crops widely cultivated throughout the world in a range of agroecological environments (Lower and Edwards, 1986) .It is the fourth most important vegetable crops after tomato, cabbage and onion (Tatlioglu, 1997), and comprises of 90 genera and 750 species. It is one of the oldest cultivated vegetable crops and is cultivated in nearly all countries of temperature zones. It is a thermophilic and frost-susceptible plant species, growing best at temperatures above 20 °C (Thoa, 1998). It is a creeping plant which is widely cultivated for its edible fruit. The nutritional value of the cucumber is low, but its delicate flavour makes it popular for salads and relishes. The cucumber can be grown in frames or on trellises in greenhouses in cool climates and is cultivated as a field crop and in home gardens in warmer areas. Cucumber have some health benefits such as Detoxification of the body, it keeps the brain and kidneys healthy, Relieves constipation, lowers the risk of cancer, keeps the body cool and hydrated, freshens the breath and its a natural remedy for intestinal worm. A good diet rich in vegetables has been shown to be an important factor in reducing the risk of diseases.

Pesticides are toxic substances that are designed to control pests ranging from insects, fungus, bacteria and weeds which at the same time are known to be toxic to other living species including humans (Yanez *et al.,* 2002; Toxipedia, 2015). Adeniyi (2016) defined pesticides as chemicals known for repelling, controlling or killing pests, which could be an

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herbicide used to control weeds; fungicides against fungi; insecticides in combating insects or rodenticides against rodents.

Pesticides due to mankind food shortage has been used to reduce Post- harvest losses in food production, Nevertheless, the use of pesticides has left the challenge of food safety and health hazard (Erhunmwunse, 2012; Carvalho, 2016). The continual usage of pesticides can lead to their accumulation on soils which can lead to pesticides overload on our soils and consequently their uptake by the plant under favorable conditions. Bertolote*et al.* (2008) reported that an estimated three million farmers in developing countries are known to have experienced acute poisoning from pesticides and 18,000 of these aforementioned farmers have been reported to die from this. Nigeria is not excluded from this as Consumer Safety Bulletin [CBS] (2008); Shaibu (2008) reported incidences of food poisoning and deaths in Cross River, Taraba and Gombe States in 2007 and 2008. Pesticide residues on vegetables constitute a possible risk to consumers and have been a human health concern. When a chemical is used as recommended on the label of the product, any residues that occur should not exceed the maximum residue levels (MRLs). Residues detected in excess of the MRL rarely constitute a toxicological concern. A good knowledge of the pesticide concentration is necessary to properly assess human exposure. Health risk assessment of pesticide residues in contaminated vegetables is performed in developed countries (Akoto et al. 2013; EFSA 2013); however, these residues are minimally explored in developing countries (Vieira et al. 2014)

Verguchte*t al*. (2006); Agency for Toxic Substances and Disease Regulatory (ATSDR) (2002); Wallige*t al.* (2002) posited in their studies that exposure to pesticides may increase risk of Parkinson's disease, cancer, sterility in males and neurological disorders. Verguchte*t al*. (2006); posited exposure to pesticides could result to Parkinson's disease. In this aforementioned authors research study, specific pesticides such as organochlorines, organophosphates or carbamates insecticides or herbicides have all been found out to be guilty.

The broad objective of the study is to assess pesticide residue in Tomato and cucumber from four selected markets in Port Harcourt

While the specifics were to:

- To identify pesticide residue in cucumber fruit
- To analyze and compare the residual levels with established safety limits of four different markets in Port-Harcourt.
- To evaluate the dietary exposure of pesticide residues in cucumber

2. Material and methods

2.1 Sample Collection and Preparation

A total of 12 samples of Cucumber consisting of three each from four different markets in Port Harcourt were randomly collected from Choba(4.7988° N, 7.0009° E) Mile 2(4.8585° N, 7.0648° E), Borokiri(4.8664° N, 6.9991° E) and Mile 1(4.8042° N, 6.9924° E) Markets in June, 2021. Cucumber sample collections was made at these wholesale markets that are popularly patronized. The samples (300g each) were packed in black polyethylene plastic bags, labelled and transported to the laboratory. Each of the samples was cut into pieces and blended with a blending machine and then poured in a specimen bottle, with each bottle containing each of the samples. Specimen bottles were labelled and stored in a refrigerator at 4°C prior to further analysis. Samples collected were based on Codex Alimentarius Commission (CAC, 1999).

2.2 Sample Collections

- Samples collected was based on Codex Alimentarius Commission (CAC, 1999).
- Sources of selected vegetable (Borokiri, Mile 1, Choba and Mile 2 Markets).
- 1kg of each sample of wet fruits was randomly collected into polythene bags and replicated eight times

Research work and Laboratory analyses were carried out in the Crop Protection Laboratory, Uniport and Austino Research Laboratory, Port Harcourt, Rivers State

2.2.1 Reagents, Equipment's and materials

The reagents used were Air-zone grade, Nitrogen gas-UPH grade, n-hexane and methylene chloride for pesticide residue analysis and anhydrous sodium sulphate, standard of the pesticide (internal standard) (Merck, Germany). These

reagents were procured through various sales representatives of the producing companies resident in Nigeria. Equipment and materials such as Autos ampler vials, 150µL vial inserts, and crimp seals, Vail crimper and decrimper, 10 micro-literautosampler, 2.5mL airtight syringe or 3mL disposable hypodermic syringe, 30-m x 0.25-mm or 0.32-mm ID fused-silica capillary column chemically bonded with SE-54 (DB-5 or equivalent), 1µm film thickness and Soxhlet extractor.

2.3 Sample Preparation

A representative portion of the cucumbers and tomatoes was blended. A 20-g aliquot of sample is homogenized, and a 10-g aliquot is spiked with the labeled compounds. The sample is mixed with anhydrous sodium sulfate, allowed to dry for 30 minutes minimum, and extracted for 18 - 24 hours using methylene chloride in a Soxhlet extractor. The extract is evaporated to dryness, and the lipid content is determined. Thus, a method or analysis blank is a laboratory prepared sample, which is known to contain no analyte.A method blank shall be run for each analyte once every 20 samples or part thereof. Though Analytes of interest should not be detected in the method blank. A duplicate sample is a complete duplicate analysis of a the sample, including extraction. The laboratory prepares duplicate samples by splitting a sample at the time of sub-sampling and prior to extraction (note: this cannot be done with samples containing phase-separated hydrocarbons PSH or sheen). The variation between the original and the duplicate analysis provides an estimate of the precision of the method. A duplicate sample shall be run for each analyte once every 20 samples or part thereof and the precision shall be expressed as a relative percentage difference (RPD %). The RPD % is between 70-130%. If outside this range, laboratory must flag this result in the final report and comment on likely reasons for this.

2.4 Extraction Procedure

Extracts for pesticide analysis may be subjected to a sequential methylene chloride-n-hexane (1:1) clean up specifically for these analytes, 40µL of the sample was injected into a gas chromatograph equipped with either a narrow- or wide-bore fused-silica capillary column and either an electron capture detector (GC/ECD) or an electrolytic conductivity detector (GC/ELCD).

2.5 Clean-up procedures

A column of about 15cm (Length) x 1cm (Internal diameter) was packed first with glass wool and then with about 7.5g activated silica gel prepared in a slurry form in DCM. About 5g of anhydrous sodium sulphate was placed at the top of the column to absorb any water in the sample or the solvent; pre-elution was done with 15mL of DCM, without exposing the sodium sulphate layer to air, so as to prevent the drying up and cracking of the packed silica gel absorbent. The reduced extract was run through column and allowed to sink below the sodium sulphate layer. Elution was done with 3 x 10mL portions of DCM. The eluate was collected and the accompanying solvent was evaporated to dryness under stream analytical grade nitrogen (99.99%). The dried eluate was reconstituted with 1mL 2, 2, 4-trimethylpentane (Isooctane) in readiness for instrumental analysis.

2.6 Qualitative identification and quantitative evaluation

GC–MS analysis was carried out using an Agilent 6890 gas chromatograph with a 5973 MS detector equipped 30-m x 0.25-mm or 0.32-mm ID fused-silica capillary column chemically bonded with SE-54 (DB-5 or equivalent), 1-µm film thickness. (Agilent). The following temperature ramp was used: injector at 250 °C, oven initially at 200 °C, held for 1 min and heated to 230 °C (1.5 °C min-1, then held for 10 min). The characterization and identification of pesticides, from the sample was completed in the SCAN mode with the m/z range varied from 35 to 450. The flow rate of the nitrogen as carrier gas was 1mL min-1; manual injection; the injection volume was 1µL. The composition of the pesticides from the sample was determined using an Agilent 6820 gas chromatograph equipped with 30-m x 0.25-mm or 0.32-mm ID fused-silica capillary column chemically bonded with SE-54 (DB-5 or equivalent), 1-µm film thickness.Injection port. The initial oven temperature was 200 °C, which was held for 1 min, subsequently increased to 230 °C at 1.5 °C min-1 and then held for 1 min. The injector was set at 250 °C, and the detector at 280 °C. Nitrogen was used as the carrier gas at a flow rate of 1 mL min-1. The split ratio was 50:1, and the sample size was 1 µL.

2.7 Quality Control Measures

Quality control measures were adopted to ensure that the analytical procedures used compiled strictly with appropriate standards and specifications and also to further ensure that the results obtained were reliable, valid and of high quality included the determinations of percentage recovery and detection limit.

2.8 A laboratory control sample

(LCS) is a reference sample either prepared by the laboratory or purchased from an external source, which contains a known amount of analyte. A laboratory control sample shall be run for each analyte once every 20 samples or part thereof and be reported in % recovery. The percentage recovery is between 70-130%. If outside this range, laboratory must flag this result in the final report and comment on likely reasons for this.

2.9 Percent recovery (%R) determination

Recovery data was obtained at the three speaking levels of pesticides in the matrix each day using blank cucumber and tomato samples in accordance with European Commission (EC) guidelines (EC 2009, 2013). Blank samples (2.0g) after homogenization were spiked by addition of appropriate volumes of pesticide standard mixture in hexane/acetone (9:1 v/v) solution and were left for 1h (equilibration times) and then prepared according to the procedures described in the sample preparation section. Method accuracy and precision were evaluated by performing recovery studies. The precision was expressed as the relative standard deviation (RSD). Accuracy can be measured by analyzing the samples with known concentration and comparing the measured values with the true values.

2.10 A Matrix spike (MS) and a Matrix spike duplicate

(MSD) is a laboratory prepared sample, which contains a known amount of analyte, which in most cases is comparable to a relevant regulatory level, and is analysed in the laboratory. The results for the matrix spike and the matrix spike duplicate are reported in percentage recovery of the spiked amount. The matrix spike and matrix spike duplicate shall be conducted on an actual soil or water sample from a batch of samples. The matrix spike and matrix spike duplicate shall be run for each analyte once every 20 samples or part thereof. The MS and MSD % recovery value is between 70-130%. The Project Manager must consider the need for specific types of matrix spikes e.g. trip matrix spikes, for particular projects and these requirements, which may differ from location to location, should be defined and agreed prior to commencement of a project.

2.11 Method Detection Limit (MDL)

The lowest level at which an analyte can be detected, but not necessarily quantified. Usually this is determined by statistical analysis of the calculated concentration of multiple injections (commonly seven) at a concentration estimated to be below the practical reporting limit. A signal to baseline noise level of 2 or 3 is used as the MDL of the analyte. MDL studies must be performed annually or every time major changes affect the method, instrument or as part of newanalysttraining.

2.12 Determination of Limit of Detection (LOD)

The limit of quantification (LOQ) was defined as the lowest concentration of the analyte that could be quantified with acceptable precision and accuracy. The limit of detection (LOD) was defined as the lowest concentration of the analyte in a sample which could be detected but not necessarily quantified. The LOQ and LOD were evaluated as the signal-to-noise ratio (S/N) of 10:1 and 3:1 for the pesticide, respectively.

The limits of detection (LOD) were evaluated by the determination of concentrations that gave signals equal to the blank based on the empirical and more specific definition described by Miller and Miller (2000) using the relationship.

$$yC = yB + 3SB$$

Where, yC = analyte signal equivalent to detection limit; yB = blank signal; and SB = standard deviations of the blank. From the value of yC, the analyte concentration corresponding to the detection limit was evaluated for GC-ECD determination of pesticides.

2.13 Limit of Quantitation

Statistically determined concentration level that produces quantitative measurements in the target matrix with acceptable precision. A signal to baseline noise level of 10 to 20 is used as the limit of quantitation of the analyte.

2.14 Statistical Analysis

All data will be subjected to Analysis of variance (ANOVA). Significant means will be separated using New Duncan multiple test (Using SPSS, Version 2.0).

3. Results

Table 1 Percentage recovery (%R) values in cucumber

The percentage recovery (%R) values of some of the analytes in the cucumber samples in the range of 82% β BHC and 100% O'ρ DDE including Methoxychlor are shown in Table 1. The detection limit of method used, (MDL) which is the lowest level at which the analyte was detected though not necessary quantified of the samples ranged 0.0010-0,0408 in α Chlordane to 0.0408-0.1035 endosulfan sulfate; while the limit of quantitation (LOQ) is 0.0039-0.0055 in α Chlordane and 0.2539-0.5364 had by endosulfan sulfate. (Table 1&2).

Table 1 Percentage recovery, Method of detection limit and limit of quantitation for insecticides

Pesticide	%Recovery RSD	MDL (mg/kg)	LOQ (mg/kg)
Organochlorine			
O'ρ DDT	85	0.0852	0.3209
Pρ DDT	90	0.0025	0.0062
O'ρ DDE	100	0.0066	0.0218
Methoxychlor	100	0.0021	0.0055
Perthane	86	0.0045	0.0079
Octachlorostyrene	98	0.0297	0.1039
α BHC	87	0.0066	0.0167
β BHC	82	0.0031	0.0073
ϒ BHC	95	0.0038	0.0084
α Endosulfan	86	0.0049	0.0119
Endosulfan sulfate	83	0.0721	0.3952
α Chlordane	93	0.00175	0.0047
Organophosphorus			
Diazinon	83	0.0166	0.0045
Malathion	93	0.0042	0.0451

Table 3 Mean concentration of pesticide residue in tomato and cucumber samples across the four markets (Fruit garden, Rumuokoro, Mile 3 and Oil mill Markets)

The mean concentration of different group of pesticides viz; Organochlorine (OC), Organophophorus (OP), Fungicide (F) and Herbicide (H) pesticide residues in cucumber samples obtained from four markets (Choba, Borokiri, Mile 1 and Mile 2 markets) in Port Harcourt are presented in Tables 2 &3. Among the pesticides detected, 58.7% insecticides (12 Organochlorine& 2 Organophosphorus insecticides), 22.7% fungicides (5) and 17.9% Herbicide (4) were detected in the tomato and cucumber samples. Table 3 showed the frequency occurrence of pesticides residue recorded were 45% forα BHC and 85% for αEndosulfanin tomato samples obtained from the fruit garden markets. The observed levels of α Chlordane were 0.02±0.01 and 0.31±0.01 for O'ρ DDE which is relatively higher than Standard Maximum Residue Limit. The percentage occurrence for Organophosphorus compound, Diazinon and Malathion were the same, 45% while their concentration range between 0.10±0.05 to 0.22±0.02. However, the reverse was the case in Borokiri market where these residues recorded different percentage occurrence but similar mean concentration .Similarly,the concentration of residues in tomato samples from the Borokiri markets was least in Methoxychlor (0.02±0.01) while higher concentration was recorded in Endosulfan sulfate (0.20±0.90) among the insecticides detected. The frequency occurrence of these pesticides residue ranged from 35%O'ρ DDT% to 100% isomer (αβ) BHC in cucumber samples obtained in Borokiri.

Table 2 Percent recovery, Method of detection limit and limit of quantitation for Fungicide and Herbicide

Pesticide	%Recovery RSD	MDL (mg/kg)	LOQ (mg/kg)
Fungicide			
Captan	89	0.0029	0.0055
Chlorotalonil	98	0.0037	0.0069
Hexachlorobenzene	96	0.0044	0.0083
Quintozene	89	0.0021	0.0054
Tecnazene	80	0.0054	0.0093
Herbicide			
Atrazine	95	0.0107	0.0353
Hexazinone	90	0.0126	0.0395
Simazine	98	0.0056	0.0161
Cyanazine	84	0.0072	0.0182

Table 3 Mean concentration of pesticide residues in cucumber samples from open market

Pesticide	Choba		Borokiri			
	Mean +SD	% Occurrence	Mean +SD	% Occurrence	MRL	%above MRL
Organochlorine						
O'ρ DDT	0.19±0.59	62	0.08±0.65	35	0.05	100
Pρ DDT	0.19±0.59	50	0.08±0.65	70	0.05	100
O'ρ DDE	0.31±0.01	75	0.17±0.01	85	0.05	100
Methoxychlor	0.14±0.40	65	0.02±0.01	95	0.05	100
Perthane	0.16±0.02	65	0.19±0.01	55	0.02	100
Octachlorostyrene	0.14±0.04	55	0.14±0.02	55	0.02	100
α BHC	0.07±0.08	45	0.14±0.12	100	0.01	100
β BHC	0.07±0.08	60	0.14±0.12	100	0.01	100
ϒ BHC	0.07±0.08	62	0.14±0.12	65	0.01	100
α Endosulfan	0.11±0.10	85	0.11±0.10	65	0.002	100
Endosulfan sulfate	0.17±0.02	55	0.20±0.90	64	0.002	100
α Chlordane	0.02±0.01	75	0.13±0.02	65	0.01	100
Organophosphorus						
Diazinon	0.22±0.02	45	0.15±0.05	80	0.10	100
Malathion	0.10±0.05	45	0.15±0.02	55	0.10	100

n = 10 for each market; SD = Standard Deviation; ND = Not Detected; MRL = Maximum Residue Limit

Table 4 Mean Concentration of Pesticide Residue in Cucumber from Fruit garden and Borokiri Market

Table 4 showed the mean concentration of 0.13±0.01 by Octachlorostyrene and three isomersBHC (α,βϒ) in cucumber obtained in Fruit garden market compared with little or no residue recorded by these organochlorine compounds in Borokiri market. The frequency occurrence of pesticides residue recorded was relatively low ranging from 40% to 70% in cucumber samples compared with other markets. The observed levels of Endosulfan sulfate was relatively high, 0.35±0.05 and 0.26±0.01 in both markets compared to other markets. This closely followed by metabolite of DDT, O'ρ DDE which is relatively higher than other pesticide residue in both markets. The percentage occurrence for Endosulfan sulfate (85%) is exceedingly high compared to pesticide residue in the two markets. The percentage occurrence of Malathion in cucumber sample obtained in Choba was higher, while the least was recorded by Diazinon. Though Diazinon recorded high mean concentration of pesticide residue in cucumber obtained from Borokiri. The ban Organochlorine insecticides shifted the attention on the Organophosphorus compound.

Table 4 Mean concentration of pesticide residues in cucumber samples from open market

Pesticide	Choba		Borokiri			
	Mean +SD	% Occurrence	Mean +SD	% Occurrence	MRL	%above MRL
Organochlorine						
O'ρ DDT	0.08±0.05	58	0.04±0.04	58	0.05	100
Pρ DDT	0.08±0.05	35	0.04±0.04	50	0.05	100
O'ρ DDE	0.15±0.02	25	0.01±0.00	45	0.05	100
Methoxychlor	0.10±0.01	42	0.20±0.01	55	0.05	100
Perthane	0.06±0.01	68	0.05±0.00	55	0.02	100
Octachlorostyrene	0.13±0.01	70	0.17±0.01	65	0.02	100
α BHC	0.13±0.08	50	0.01±0.00	65	0.01	100
β BHC	0.13±0.08	40	0.01±0.00	62	0.01	100
ϒ BHC	0.13±0.08	70	0.01±0.00	60	0.01	100
αEndosulfan	0.17±0.01	40	0.18±0.01	55	0.002	100
Endosulfan sulfate	0.35±0.05	50	0.26±0.01	85	0.002	100
α Chlordane	0.13±0.02	85	0.19±0.02	70	0.01	100
Organophosphorus						
Diazinon	0.04±0.01	35	0.16±0.02	45	0.10	100
Malathion	0.02±0.01	100	0.02±0.01	75	0.10	100

n = 10 for each market; SD = Standard Deviation; ND = Not Detected; MRL = Maximum Residue Limit

Table 5 Mean Concentration of Pesticide Residue in Cucumber From Choba and Borokiri Market

Table 5 showed high mean concentration of Captan (0.28±0.02) in cucumber obtained from Fruit garden, while Tecnazen recorded equally high mean concentration 0.22±0.03 in Fruit garden and 0.37±0.01 in Rumuokoro at relatively low percentage occurrence across the markets under study. Quintozene followed closely in mean concentration by the proportion of pesticide residue obtained in cucumber from Borokiri market at relatively high percentage occurrence of 75%. Cyanazine recorded higher mean concentration (0.23±0.01) (0.16±0.01) in the two markets compared with other pesticide residues at relatively high percentage of occurrence (65%). This shows increase in the use of herbicide as a means of controlling weeds. However, all the pesticide residues detected samples were above their respective MRLs (Table 5).

Table 6 Mean Concentration of Pesticide residue in Cucumber from Mile 1 and Mile 2 Markets

Table 5 Mean concentration of pesticide residues in cucumber samples from open market

Pesticide	Choba		Borokiri			
	Mean +SD	% Occurrence	Mean +SD	% Occurrence	MRL	%above MRL
Fungicide						
Captan	0.28±0.02	55	0.13±0.01	60	0.05	100
Chlorotalonil	0.15±0.15	35	0.03±0.01	45	0.05	100
Hexachlorobenzene	0.19±0.02	52	0.11±0.01	65	0.02	100
Quintozene	0.12±0.02	65	0.21±0.01	75	0.02	100
Tecnazene	0.22±0.03	48	0.37±0.01	45	0.02	100
Herbicide						
Atrazine	0.13±0.01	58	0.08±0.02	60	0.10	100
Hexazinone	0.06±0.01	60	0.15±0.01	55	0.10	100
Simazine	0.03±0.01	70	0.12±0.02	90	0.02	100
Cyanazine	0.16±0.01	65	0.23±0.01	65	0.02	100

n = 10 for each market; SD = Standard Deviation; ND = Not Detected; MRL = Maximum Residue Limit

Table 6 Mean concentration of pesticide residues in cucumber samples from open market

	Mile 1		Mile 2			
Pesticide	**Mean +SD**	**% Occurrence**	**Mean +SD**	**% Occurrence**	**MRL**	**%above MRL**
Organochlorine						
O'ρ DDT	0.07±0.04	35	0.10±0.06	35	0.05	100
Pρ DDT	0.07±0.04	60	0.10±0.06	69	0.05	100
O'ρ DDE	0.36±0.01	42	0.03±0.01	40	0.05	100
Methoxychlor	0.03±0.01	72	0.35±0.00	90	0.05	100
Perthane	0.13±0.01	65	0.02±0.00	55	0.02	100
Octachlorostyrene	ND	40	0.17±0.01	55	0.02	100
α BHC	0.14±0.07	72	0.08±0.06	50	0.01	100
β BHC	0.14±0.07	70	ND	80	0.01	100
ϒ BHC	0.14±0.07	75	0.08±0.06	100	0.01	100
αEndosulfan	0.52±0.00	35	0.11±0.01	65	0.002	100
Endosulfan sulfate	0.17±0.02	42	0.18±0.99	64	0.002	100
α Chlordane	0.10±0.01	80	0.08±0.01	58	0.01	100
Organophosphorus						
Diazinon	0.25±0.01	60	0.18±0.01	80	0.10	100
Malathion	0.24±0.02	50	0.09±0.01	38	0.10	100

n = 10 for each market; SD = Standard Deviation; ND = Not Detected; MRL = Maximum Residue Limit

Table 6 residue in cucumber got from Mile 1 market. Also themetabolite of DDT recorded high mean concentration (0.36±0.01) of pesticide residue compared to other compound in cucumber obtained in Mile 1. Whereas this pesticide residue is exceedingly low (0.03±0.01) in Mile 2 market. Likewise Methoxychlor recorded high mean concentration (0.36±0.01) in cucumber in Mile 2 market. The isomers of DDT were however lower as pesticide residue detected in all the samples obtained from the four markets. Among the isomer of BHC, β BHC was not detected from cucumber samples in Mile 2 market. Also, Octachlorostyrene was not detected in cucumber obtained in Mile 1 market. However, in Mile 1 ,α,β,Y, BHC recorded similar concentration (0.14±0.02) at relatively same percentage of occurrence. Lindane one of the banned insecticide is applied as a pest control. Organophophorus compound like Diazinon had high frequent Occurrence of 80% and mean concentration (0.18±0.01) compared to Malathion that recorded the least (0.09±0.01) in Oil (Table 6).

Table 7 Mean Concentration of Pesticide Residue in Cucumber from Mile 1 and Mile 2 Markets.

Table 7 showed that fungicide, Quintozene recorded higher mean concentration (0.42±0.19) (0.31±0.01) pesticide residue in (Mile 1 & Mile 2), followed closely by Chlorotalonil (0.39±0.01) andCaptan (0.31±0.01) in both markets and with percentage occurrence between 55 and 80%%.The fungicides residues recorded the lowest mean concentration for Hexachlorobenzene0.01±0.01 in Mile 1, whereas Captan had 0.08±0.01 in Mile 2 market. Thus Simazine and Cyanazine had pesticide residue mean concentration of 0.27±0.02 compared to other herbicide detected in cucumber samples from Mile 1 whereas Hexazinone recorded higher mean concentration (0.13±0.01), while Atrazine had the least (0.04±0.02) in cucumber obtained from Mile 2 market (Table 7).

Table 7 Mean concentration of pesticide residues in cucumber samples from open market

Pesticide	Mile 1		Mile 2			
	Mean +SD	**% Occurrence**	**Mean +SD**	**% Occurrence**	**MRL**	**%above MRL**
Fungicide						
Captan	0.31±0.01	55	0.08±0.01	68	0.05	100
Chlorotalonil	0.39±0.01	38	0.21±0.02	75	0.05	100
Hexachlorobenzene	0.21±0.02	55	0.01±0.01	80	0.02	100
Quintozene	0.42±0.19	48	0.31±0.01	65	0.02	100
Tecnazene	0.13±0.01	38	0.10±0.02	50	0.02	100
Herbicide						
Atrazine	0.11±0.01	60	0.04±0.01	85	0.10	100
Hexazinone	0.19±0.01	30	0.13±0.01	58	0.10	100
Simazine	0.14±0.02	35	0.05±0.00	85	0.02	100
Cyanazine	0.27±0.02	55	0.02±0.00	60	0.02	100

n = 10 for each market; SD = Standard Deviation; ND = Not Detected; MRL = Maximum Residue Limit

4. Discussion

This study established the presence of different group of pesticides residues in tomato and cucumber samples from the selected markets in Port Harcourt, South South, Nigeria. The level of precision was shown by the percent recovery and limit of detection. The MDL of the samples was 0.0408 mg/kg α Chlordane and 0.0723 mg/kg endosulfan sulfate; while the limit of quantitation (LOQ) of these pesticides were 0.0047mg/kg and 0.3952 mg/kg respectively. Among the pesticides determined 58.7% insecticides, 22.7% fungicides and 17.9% Herbicide were detected in the tomato and cucumber samples. Pesticide include Chlordane, Hexachorbe, Perthane, Atrazine, Hexazinone, and Malathion, DDEetc, This quite agrees with the study of Islam *et al.* (2015) who reported that pesticide residues in cucumber samples from local markets detected were Endosulfan, Mancozeb, Perthane and atrazine (about 50 ppm) in one out of three samples. Ali and (2020) and Kaya and Tuna (2019) also investigated pesticide residues in cucumbers and detected thiamethoxam residues as 0.025 mg/kg. Similarly, Adeniyi (2016) in his findings detected the likes of several pesticide residues of Endosulfan 2, Glyphosate, Methoxychior, Atrazine and Malathionas pesticide residues found in cucumber. The mean

concentration of Metoxychlor was excessively higher in samples obtained across the four markets. Also, the concentration of O'ρ Pρ, DDT was higher than its metabolite in Mile 2 and Mile 1 markets which might portend danger for our health. This implicated regular use of common all purpose insecticides that still available in the markets despite outlawed by the government. This group of pesticide residues is frequently detected in food items in Southwestern, Nigeria (Ogah et al., 2011; 2012; Sosan et al., 2015; Sosan and Oyekunle, 2017). The preponderance and high concentrations of Endosulfan sulfate and isomer in all the markets monitored underscored to the fact that these compounds are still much in use despite its ban by the government. Also, Reksa-Naik and Prasad (2006) stated that endolsulfan residues in wheat were reported in all market samples in India. In Cameroon, Sonchieu et al., (2010) reported Lindane and Endosulfan in maize samples with mean concentrations higher than the EU-MRL for the insecticides. The detection of high Methoxychlor, DDT and O'ρ Pρ metabolite in Mile 2 and Mile 1 markets might be as a result of the past usage which resulted in bioaccumulation or probably arising from the cultivation of the crop on contaminated soils where application of insecticides were intense. In Mile 1 and MilE 2 markets, isomer of BHC and Octachlorostyrene pesticides residue were not detected. This has proven that less quantity of organochlorine compounds are used in these markets compared with Choba and Borokiri. This is in contrary to the report by Bakore et al., (2004) that all of the wheat samples from India were contaminated with various organochlorine pesticide residues of DDTs and its metabolites and its isomers. The organophosphorus group (Diazinon and Malathion) pesticides analysed recorded the highest occurrence when compared with organochlorine compound in Mile 2 and Mile 1 markets. This could be attributed to the ban on Lindane marketed as Gammalin 20EC which made it difficult to get in the markets.

Based on acceptable dietary intake, the pesticides residue detected in cucumber has a value less than 100% of the ADI estimates given by WHO (1 – 20Mg/kg) (WHO, 1997). Dietary estimates recorded for most of the pesticide residues are above the EU-MRL (0.05mg/kg) (European Commission 2005). The diversity of classes of pesticide residues such as organophosphorus (Malathion, and Diazinon) detected in this study show that the proposed method to determine residues of pesticides in fruits is rapid, simple, sensitive and uses smaller amount of organic solvents, reducing the risk for workers and the environment.

5. Conclusion

This study assessed the levels of pesticide residues tomato fruit from four selected markets in Port Harcourt. The results indicated that 100% of the samples were contaminated with pesticide residues, however tomato samples from open markets in Port Harcourt were contaminated with pesticide residues above the standard Maximum Residue Limits (MRLs). Therefore contamination may pose a danger to human since fruit contamination is the main route of exposure to all group of pesticides.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest.

References

[1] Agency for Toxic Substances and Disease Regulatory (ATSDR) (2002). Toxicological profile for Methoxychlor. US Department of Health and Human Services; 1-235.

[2] Akhtar Naveed, Arshad Mehmood, Barkat Ali Khan, Tariq Mahmood, Haji Muhammad Shoaib Khan and Tariq Saeed. 2011. Exploring cucumber extract for skin rejuvenation. African Journal of Biotechnology, Vol. 10(7): 1206-1216.

[3] Aktar, M.W., Sengupta, D and Chowdhury, A. (2009). Impact of pesticides use in agriculture: their benefits and hazards. Journal of InterdisciplinaryToxicology; 2(1):1–12.

[4] Aprea, C., Colosio, C., Mammone, T., Minoia, C and Maroni, M. (2002). Biological monitoring of pesticide exposure: a review of analytical methods Journal of Chromatography B, 769 (2):191-219.

[5] Bempah, C.K., Buah-Kwofie, A., Denutsui, D., Asomaning, J. and Tutu, A. O. (2011). Monitoring of pesticide residues in fruits and vegetables and related health risk assessment in Kumasi Metropolis Ghana. Research Journal ofEnvironmental and Earth Sciences 3:761–771.

[6] Bertolote, J. M., Fleischmann, A., Eddleston, M. and Gunnell, D. (2008). Deaths from Pesticide Poisoning: Are we lacking a global response. Europe PMC Funders Group Author Manuscript Br J Psychiatry; 189: 201–203.

[7] Borş, A. M., Meghea, I. and Borş, A.G. (2012). Agricultural and Biological Sciences: "Pesticides- Recent Trends in Pesticide Residue Assay". (Ed) Soundararajan. R.P. 3.0 license. Romania.

[8] Carvalho, F.P. (2017). Pesticides, Environment, and Food Safety. Food and Energy Security; 6(2):48-60.

[9] Codex Alimentarius Commission (1999) Recommended Methods of Sampling for the Determination of Pesticide Residues for Compliance with MRLs, CAC/GL 33-1999.

[10] Cohen, L.A. (2002) Nutrition and prostate cancer: a review. Ann. N.Y. Acad. Sci. 963: 148–155. In: Kristie Canene-Adams, Jessica K. Campbell, Susan Zaripheh, Elizabeth H. Jeffery, and John W. Erdman, Jr. 2005. The Tomato as a Functional Food. Journal of Nutrition, 135: 1226-1230.

[11] Cserháti, T and Szőgyi, M. (2012). Chromatographic determination of pesticides in foods and food products. European Chemical Bulletin; 1(3-4),58-68.

[12] Erhunmwunse, N.O., Dirisu, A. and Olomukoro, J.O. (2012). Implications of Pesticide Usage in Nigeria. Tropical Freshwater Biology; 21 (1): 15- 25.

[13] Etonihu, A. C, Aminu, B. A, Ambo, A. I and Etonihu, K. I. (2011). Iodine content and Pesticide residues of Some Nigerian grains. Continental Journalof Agricultural Science; 5 (1): 26-32.

[14] FAO (2002). Submission and evaluation of pesticide residues data for the sestimation of maximum residue levels in food and feed. Rome: Food and Agriculture Organisation.

[15] Food Standards Agency (2010). What are Pesticide residues? www.food.gov.uk/.../pesticidesmainqa/pesticidesqaq03. Retrieved 21/7/2011.

[16] Garcia, F, P., Cortés Ascencio, S.Y., Oyarzun, J.C.G., Hernandez, A.C and Alavarado, P.V. (2012). Pesticides: classification, uses and toxicity. Measures of exposure and genotoxic risks. Journal of Research in EnvironmentalScience and Toxicology; 1(11): 279-293.

[17] Garcia, F, P., Cortés Ascencio, S.Y., Oyarzun, J.C.G., Hernandez, A.C and Alavarado, P.V. (2012). Pesticides: classification, uses and toxicity. Measures of exposure and genotoxic risks. Journal of Research in EnvironmentalScience and Toxicology; 1(11): 279-293.

[18] Global Environment Monitoring System – Food Contamination Monitoring and Assessment Programme (GEMS/Food) in collaboration with Codex Committee on Pesticide Residues (1997). Guidelines for predicting dietary intake of pesticide residues (revised). Programme of Food Safety and Food Aid World Health Organization, 1-41.

[19] Ijyothi, S. and Devia, S. (2016). Review on-Pesticide Residues of Fruits and Vegetables. International Journal of Research in Applied, Natural and SocialSciences; 4 (11): 71-78.

[20] James, O.A (2016). "Detection and Alleviation of Pesticide Residue in Food and Water" (2016). LSU Doctoral Dissertations. 2808. https://digitalcommons.lsu.edu/gradschool_dissertations/2808.

[21] Joint FAO/WHO Codex Alimentarius Commission.; Joint FAO/WHO Food Standards Programme.;Food and Agriculture Organization of the UnitedNations.; World Health Organization. (2000) Pesticide residues in food: methods of analysis and sampling. Codex Alimentarius, Vol. 2a. Rome; Great Britain.

[22] Jørs, E. (2016). Acute pesticide poisoning among Bolivian small-holder farmers - frequency,risk factors and prevention. Unit for Health Promotion, Department of Public HealthUniversity of Southern Denmark Publications of the Unit for Health Promotion Research, Series A; No 10. 2016

[23] Kapoor, U, Srivastava, M. K, Srivastava, A. K, Patel, D. K, Garg, V and Srivastava, L. P. (2013). Analysis of imidacloprid residues in fruits, vegetables, cereals, fruit juices, and baby foods, and daily intake estimation in and around Lucknow, India. Environmental Toxicology Chemistry,32 (3):723-727.

[24] Katsikantami, I., Colosioc, C., Alegakisb, A, Tzatzarakis, M N, Vakonaki, E, Rizosa, A, K, Sarigiannis, D. A, and Tsatsakis, A. M. (2019). Estimation of daily intake and risk assessment of organophosphorus pesticides based on biomonitoring data. The internal exposure approach. Food and Chemical Toxicology, 123: 57-71.

[25] Kegley, S. E., Hill, B. R., Orme S. and Choi A. H. (2016). PAN Pesticide Database, Pesticide Action Network, North America.http:www.pesticideinfo.org; 1-5.

[26] Lehotay, S. J. (2007). Determination of pesticide residues in foods by acetonitrile extraction and partitioning with magnesium sulfate: collaborative study. Journal of AOAC International, 90 (2):485-520.

[27] Mahugija, M.A.J., Khamis, F.A. and Lugwisha, E.H.J (2017).Assessment of pesticide residues in tomatoes and watermelons (fruits) from marketS in Dar ESSalaam, TanzaniaJ. Appl. Sci. Environ. Manage. May 2017Vol. 21 (3) 497-501.

[28] Martinez-Dominguez, G., Plaza-Bolaños, P., Romero-González, R and Frenich, A.G. (2014). Multiresidue method for the fast determination of pesticides in nutraceutical products (Camellia sinensis) by GC coupled to triple quadrupole MS. Journal of Sep Science, 37(6):665-674.

[29] Muir, D and Sverko, E. (2006). Analytical methods for PCBs and organochlorine pesticides in environmental monitoring and surveillance: a critical appraisal. Annals of Bioanalytical Chemistry, 386:769–789.

[30] Niimura, N. (2011). Effectiveness of Selected Reaction Monitoring for rapid assay of Cypermethrin Residue in PerillaLeaves. Advances in MaterialsPhysics and Chemistry; 1: 1-5.

[31] Oerke, E. C. (2006). Crop losses to pests. J Agr Sci.;144:31-43.

[32] Oerlemans Kirsten, Diane M. Barrett, Carme Bosch Suades, Ruud Verkerk, Matthijs Dekker. (2006). Thermal degradation of glucosinolates in red cabbage. Food Chemistry 95, 19-29.

[33] OGBOLE, A.C (2019). Determination of selected pesticide residues in some foodstuffs sold in four markets in Makurdi metropolis Benue state Nigeria using gas chromatography-mass spectrometry in a thesis submitted to postgraduate school Benue state university Makurdi Nigeria.

[34] Oluwarantimi, I. M and Faruruwa, F. (2016). Determination of Pesticide Residues in Fruits and Vegetables in Kaduna Metropolis, Nigeria. International Journal of Environmental Science and Toxicology Research, 4 (10): 185-189.

[35] Pang, G. F., Fan, C. L, Liu, Y. M., Cao, Y. Z., Zhang, J. J., Li, X. M., Li, Z. Y., Wu, Y. P. and Guo, T. T. (2006). Determination of residues of 446 pesticides in fruits and vegetables by three-cartridge solid-phase extraction-gas chromatography-mass spectrometry and liquid chromatography-tandem mass spectrometry. Journal of AOAC International. 89 (3):740-71.

[36] Pretty J, and Bharucha Z. P. (2015). Integrated Pest Management for Sustainable Intensification of Agriculture in Asia and Africa. Insects.;6(1):152-82.

[37] Rahman, SIandChima, C. D. (2018). Determinants of Pesticide Use in Food Crop Production in South eastern Nigeria. Agriculture. Molecular DiversityPreservation International (MDPI); 8 (35):1-14.

[38] Schreinemachers P, and Tipraqsa P.(2012). Agricultural pesticides and land use intensification in high, middle and low income countries. Food Policy.;37(6):616-26.

[39] Shaibu, I. (2008). 30 Agrochemical products banned in Nigeria after deaths. Vanguard 14th May, 2008. Retrieved from http://www.organicconsumers.org/news/30/agrochemical-products-bannednigeria- after-deaths on 17/09/2017.Consumer Safety Bulletin (2008): NAFDAC regulated products. AbujaNigeria: National Agency for Food and Drug Administration and Control; 9p.

[40] Tomlin, C. (1994). The Pesticide Manual. 10th Ed. Crop Protection Publications; UK; 1-10.

[41] Toxipedia. Toxipedia, Pesticides 2015 [cited 2015 15/10]. Available from: http://www.toxipedia.org/display/toxipedia/Pesticides.

[42] Vergucht, S, de Voghel, S, Misson, C, Vrancken, C, Callebaut, K, Steurbaut, W, Pussemier, L, Marot, J, Maraite, H and Vanhaecke, P. (2006). Health and environmental effects of pesticides and type 18 biocides. Report from the contract AP/02/05A between the Belgian Science Policy and Department of Crop Protection Chemistry, Ghent University; Veterinary and Agrochemical Research Centre (VAR), and Environmental Consultancy and Assistance; 1- 455.

[43] Verkerk Ruud and Matthus Dekker. (2004). Glucosinates and Myrosinase Activity in Red Cabbage (Brassica oleraceaL. Var. Capitata f. rubraDC.). after Various Microwave Treatments. Journal of Agricultural Food Chemistry, 52,7318-7323.

[44] Wallig, M.A, Haschek, W.M, Rousseaux, C. G and Bolon, B. (2018). Male Reproductive System. Fundamentals of Toxicologic Pathology; 3rd Edition, Elsevier, 553-597.

[45] Wilkinson, S. and Chodak, G. W. (2003) Critical review of complementary therapies for prostate cancer. J. Clin. Oncol. 21: 2199–2110. In: Kristie Canene-Adams, Jessica K. Campbell, Susan Zaripheh, Elizabeth H. Jeffery, and John W. Erdman, Jr. 2005. The Tomato as a Functional Food. Journal of Nutrition, 135: 1226-1230.

[46] World Health Organisation (2004). Classification of Pesticides; Geneva. Cox, C. (1995). Glyphosate part 2: Human Exposure and Ecological Effects. Journal of pesticides Reform; 15 (4): 14-27.

[47] Yanez L., Ortiz, D., CalderoN, J., Batres, L., Carrizales, L. and Mejia, J, (2002). Overview of human health and chemical mixtures: problems facing developing countries. Environ Health Perspect.;110Suppl 6:901-9.

[48] You, J, Weston, D. P and Lydy, M. J. (2004). A Sonication Extraction Method for the Analysis of Pyrethroid, Organophosphate, and Organochlorine Pesticides from Sediment by Gas Chromatography with Electron-Capture Detection. A r c h i v e s o f Environmental Contamination and Toxicology, 47, 141–147.

[49] Zacharia, J. T (2011). Identity, Physical and Chemical Properties of Pesticides, Pesticides in the Modern World - Trends in Pesticides Analysis. (Ed.), ISBN: 978-953-307-437-5, InTech.