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Analysis of Pesticide Residues in Some Market Garden Crops in Santa, North West Cameroon

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Abstract

The increased use of pesticides in crop production, especially in Santa, has left consumers with great fear and uncertainty regarding the fate of residues after consumption in the human body. This study was aimed at performing a multi-residue analysis of crop samples to detect and quantify pesticide residues present, and thus determine their level of contamination. Twelve (12) crop samples (4 samples each of *Solanum tuberosum* (Irish potato), *Solanum lycopersicum* (tomato), *Capsicum annum* (sweet pepper) and *Apium graveolens* (celery)), were randomly collected from four villages in Santa. The samples were analyzed using the QuEChERS Buffered AOAC 2007.01 method. The results showed the presence of pesticide residues in 58.3% of the samples. Cypermethrin and chlorpyrifos insecticides were detected, though cypermethrin was found in traces. After quantification, the concentration of chlorpyrifos was determined to be far above the CODEX Maximum Residue Limits (MRLs) in 28.6% of the samples. Samples with this high concentration of chlorpyrifos were: tomato from Pinyin and Baligham (2.0 and 1.8 µg/g respectively), whereas the Codex MRLs for tomato is 1.0 µg/g. This study raised concerns on the contamination of some garden crops by pesticide residues and calls for the assessment of the health situation of consumers and the analysis of pesticide residues in drinking water resources in this area.

Keywords

Pesticide Residue, Market Garden Crops, Contamination, Santa

1. Introduction

Market gardening is the commercial production of vegetables, fruits, flowers and other plants, on a scale larger than a home garden, yet small enough that many of the principles of gardening are applicable. Market garden crops can therefore be defined as crops cultivated mostly on a small scale for commercial purposes [1]. The need to produce greater quantity and quality of such market garden crops by pest control has led to an intensive use of pesticides by farmers. Pesticides are substances that are used in agriculture for the protection of crops against insects, fungi, weeds and other pests [2]. They constitute one of the major inputs in the agricultural production of crops [3]. Pesticides are used worldwide to protect crops before and after harvest in farms, homes and soil treatment [4]. Despite the importance of pesticides in crop production, they can be very dangerous to human beings if poorly managed. Uncontrolled application of pesticides leads to high concentration of residues in food crops at harvest. These residues can cause acute and/or chronic health effects depending on the type, quantity and ways in which a person is exposed [5]. According to Garcia *et al.* [6], pesticides must be used according to Good Agricultural Practice (GAP). The application of GAP is not always the case in Africa because of lack of proper spray equipment, pesticide risk assessment tools and the low level of expertise on pesticide risk assessment and management. According to the United Nations (UN) food experts [7], about 200,000 people die each year from toxic exposure to pesticides across the world. Pesticide analysis in food crops is therefore a call for concern [3] [8].

About 80% of the population of Santa currently depends on agriculture for living. Generally, the variation of soil types, together with the climate peculiarities, favours the cultivation of a large variety of crops, ranging from tubers and cereals like cassava and maize to garden crops like cabbage and carrot. It is worth noting that Santa Sub-division is the single highest producer of carrots in the North West region of Cameroon. The soil is gradually losing its fertility due to over-exploitation and the use of poor techniques of cultivation. Common crops grown include cabbage (*Brassica oleracea*), carrots (*Daucus carota*), maize (*Zea mays*), beans (*Phaseolus vulgaris*), tomato (*Solanum lycopersicum)*, sweet pepper (*Capsicum annum*), Irish potato (S*olanum tuberosum*), celery (*Apium graveolens*) and a host of others (Santa Council Development Plan, 2011). Among these crops, tomato, sweet pepper, Irish potato and celery were selected for this study. Tomato is a fruit from the nightshade family, native to South America. Even though botanically it is considered as a fruit, it is generally prepared as a vegetable. It contains lycopene, an antioxidant which has been linked to many health benefits such as reduced risk of heart diseases and cancer. Tomato is also a great source of vitamins. Sweet pepper is a cultivar group of the species *capsicum annum*. The cultivar of the plant produces fruits of different colours such as: red, orange, yellow, green, white and purple. They are botanically classified as fruits but in the culinary context they are considered as vegetables. Irish potato is the world's fourth largest food crop, following, rice, wheat and maize. Potatoes are edible tubers which are available all over the world, and all year long. They are relatively cheap to grow and rich in nutrients [9]. Celery is a vegetable which belongs to the apiaceae family. It has a low calorie crunchy stalk and is an integral part of many cuisines. In addition to having a low calorie level, celery is beneficial to the digestive system and the cardiovascular system. According to the United States Department of Agriculture (USDA) National nutrients Database, celery contains minerals like calcium, sodium, copper, magnesium, iron and potassium. It equally contains fatty acids and vitamins. The seeds of the plant are also used in medicine to help relieve pain [10].

Santa sub division is an area where huge amounts of market garden crops are cultivated with an intensive use of pesticides. The misuse of pesticides in this area as indicated by Sonchieu *et al.* [11] has left consumers with fear and uncertainty regarding the fate of residues in the human body after the consumption of crops. Some outdated and banned pesticides in developed countries are still being used in developing countries [12]. Also, there is insufficient legislation on pesticides in Cameroon, coupled with the lack of proper spray equipment [13]. From every indication, most of the market garden crops consumed in Cameroon are contaminated with pesticides. It was, thus, necessary to determine the amount of pesticide residues in market garden crops and assess their level of toxicity in order to save the population from pesticide poisoning and death. In line with the above problem, this research was aimed to analyze pesticide residues in samples of *Solanum lycopersicum*, *Capsicum annum*, S*olanum tuberosum* and *Apium graveolens* from Pinyin, Njong, Mbei and Baligham villages of Santa in order to assess the level of crop contamination.

2. Materials and Methods

2.1. Description of the Study Site

According to the Santa Council Development Plan (CDP, 2011), Santa Sub-Division is one of the seven administrative units of Mezam Division in the North West Region of Cameroon. It lies between longitude 9˚58' and 10˚18' East of the Greenwich Meridian and between latitude 5˚42' and 5˚53' North of the Equator. Santa which is the Sub-divisional headquarter is situated some 20 km to the south of Bamenda, the North West Regional capital and about 60 km North of Bafoussam, the Regional capital of the West Region. It is located to the Southern part of the Region and bounded by Balikumbat and Galim to the East; Batibo and Wabane Sub-Divisions to the South; Bali to the West; Bamenda I to the North; Bamenda II to the North West and Bababjou and Mbouda to the South. The total surface area is estimated at 533 sq km. It is made up of 10 villages which are: Akum, Alateneng, Awing, Baba II, Baligham, Mbei, Mbu, Njong, Pinyin and Santa. The map of Santa is shown in **Figure 1**.

2.2. Sample Collection and Preservation

Samples were collected from Pinyin, Baligham, Mbei and Njong villages in Santa. About 1 kg each of sweet pepper, tomato, Irish potato and celery were randomly collected. A total of 12 samples were collected and each sample was

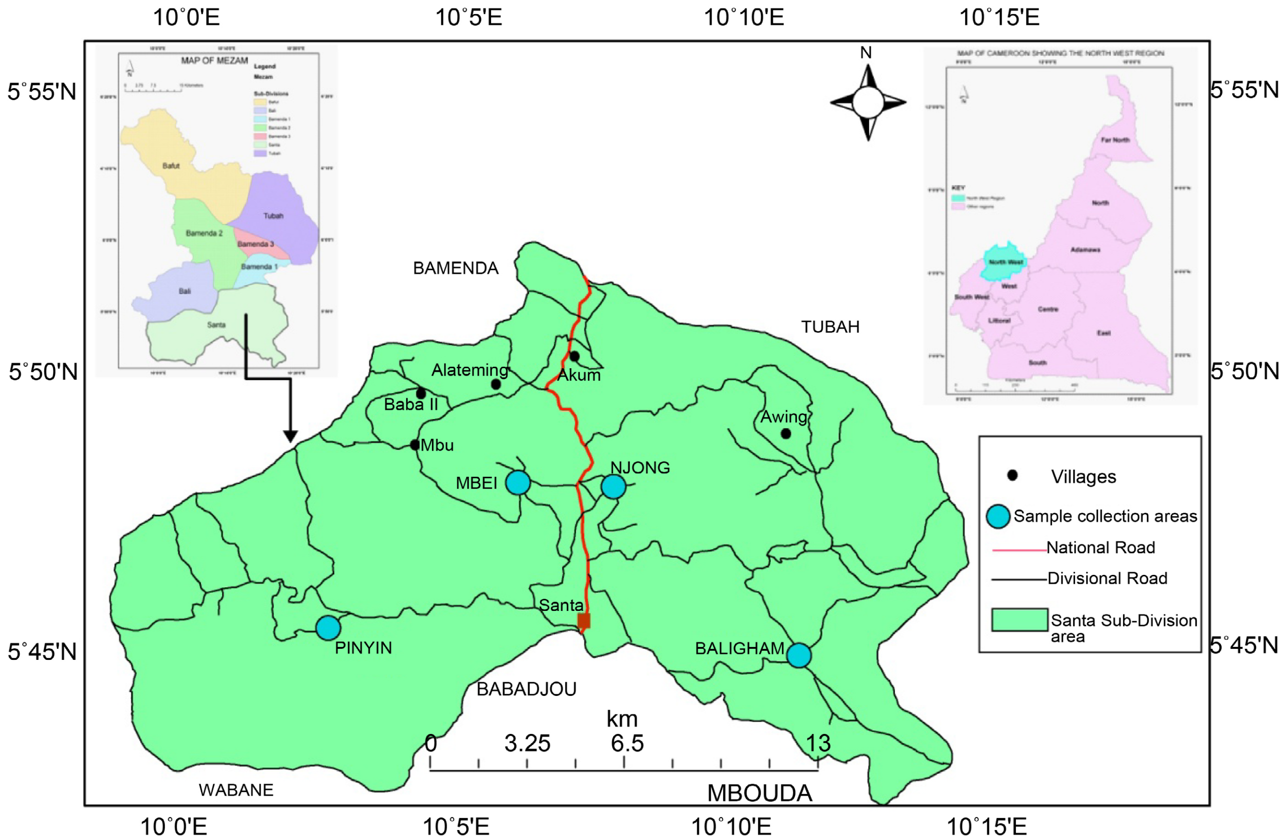

Figure 1. Map of Santa area showing sample collection sites.

weighed using a portable weighing balance. The weighed samples were parceled in aluminum foil, labeled and placed in Ziploc polythene bags before transportation in a cooler with ice blocks to the laboratory. On arrival at the laboratory, the samples were checked and confirmed to be in a good state and were placed in a refrigerator at −20˚C. Each cleaned sample was blended to obtain a homogenous mixture.

2.3. Analytical Techniques

The technique used in sample analysis was the Multi-residue QuEChERS AOAC official 2007.01 method. This method has the ability of detecting several pesticides at the same time. The analysis was done at the DRCQ laboratory of MINADER in Yaoundé, Cameroon. Extraction was done using acetonitrile solvent. Clean up was achieved via the addition of appropriate amounts of primary secondary amine (PSA), anhydrous magnesium sulphate and carbon-18, followed by centrifugation. Each cleaned sample was then analyzed using a Gas Chromatograph with a mass spectrometric detector. After the analysis, the chromatogram of retention time against peak areas was obtained for all the samples. The pesticides were identified by matching the retention time on the chromatograms to those in the database of the instrument. Examples of chromatograms are shown in Figure 2 & Figure 3 below.

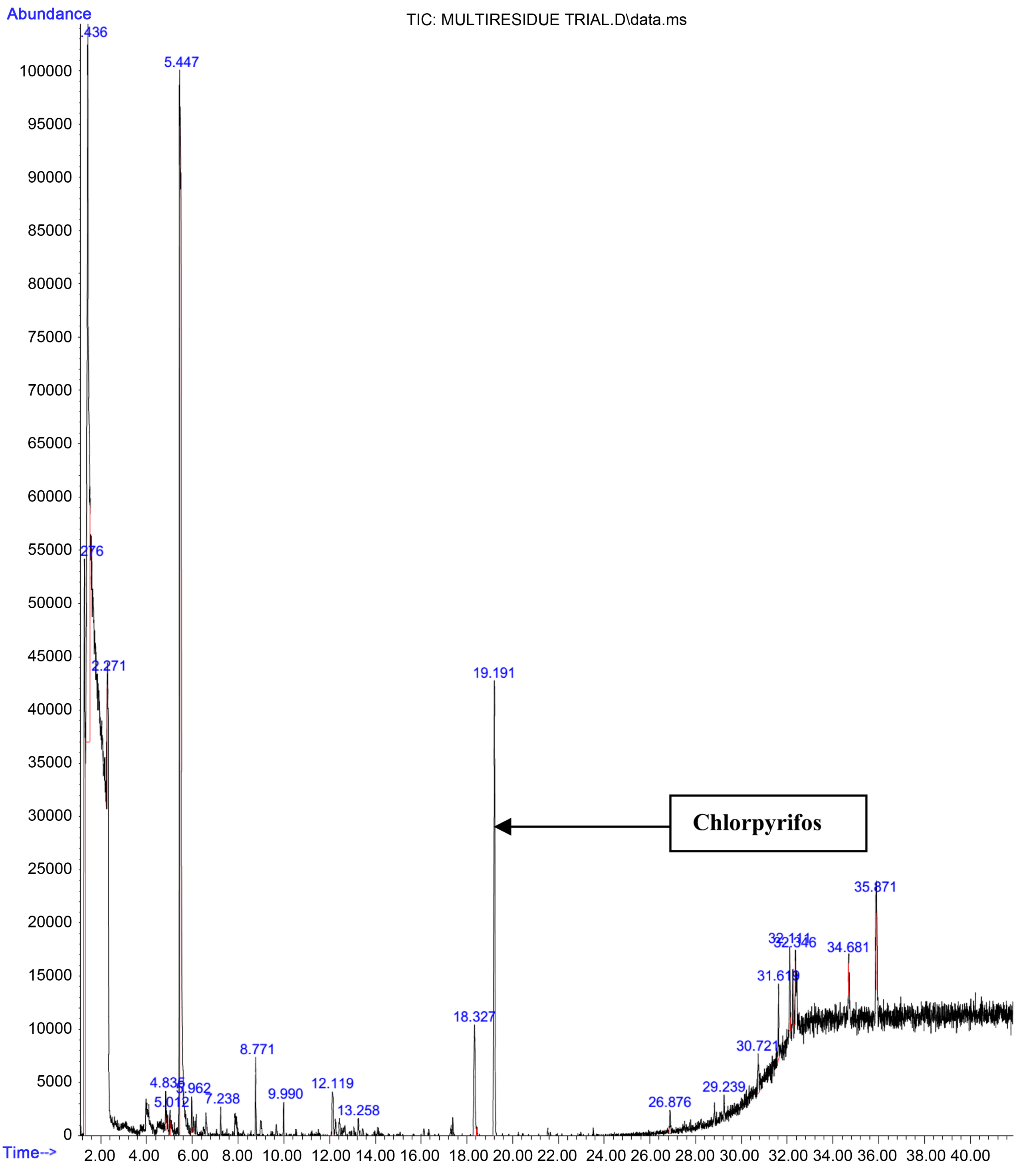

Figure 2. Chromatogram of the tomato sample from Baligham.

To quantify the pesticide residues detected, a standard solution of the pesticide was prepared, and used to constitute five calibration standards of which 1.5 μl of each was analyzed. The peak areas obtained from each chromatogram are shown in **Table 1**.

The peak areas obtained from the different concentrations were used to plot a

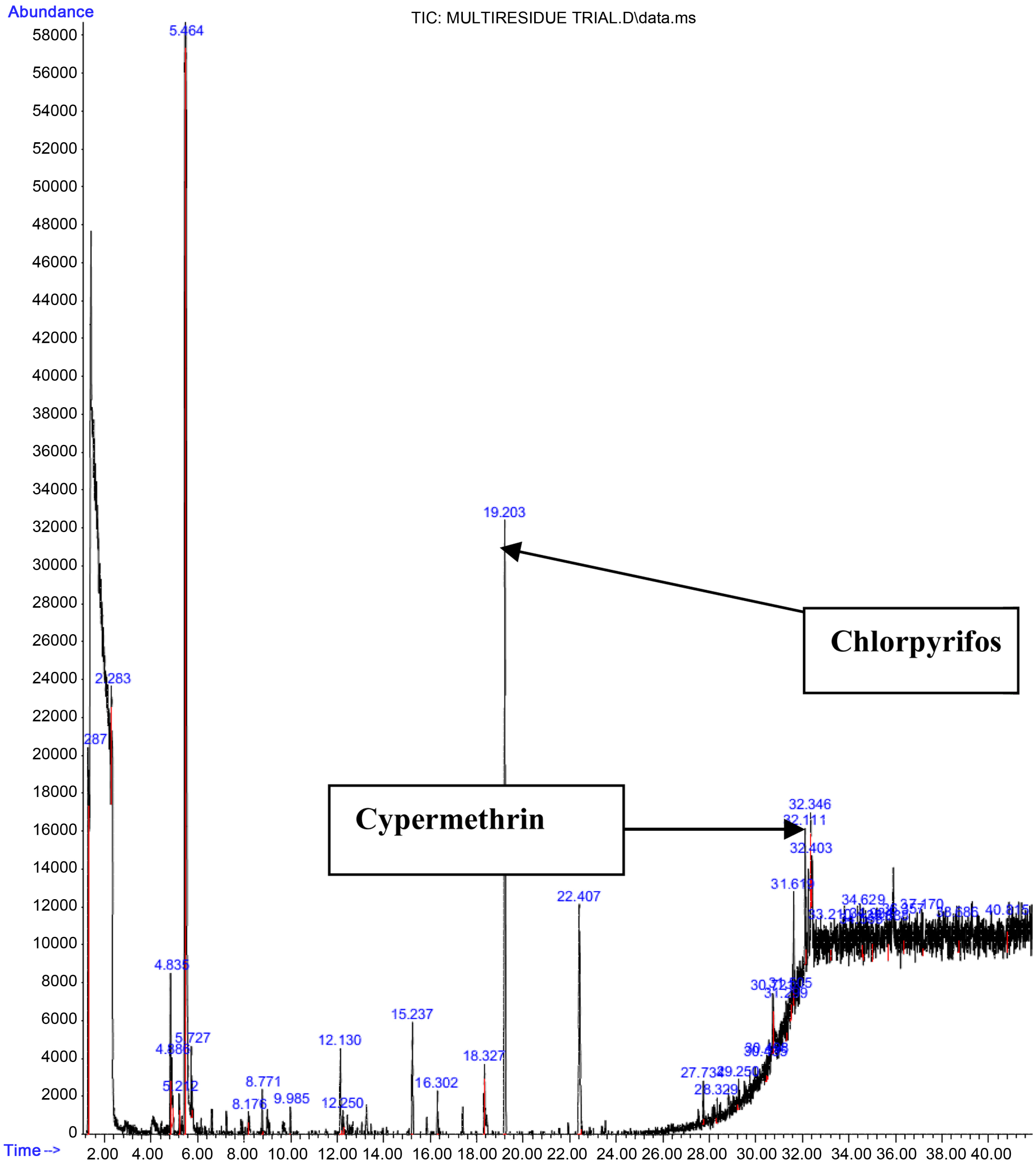

Figure 3. Chromatogram of sweet pepper sample from Mbei.

calibration curve as shown in Figure 4.

The calibration curve gave the linear equation:

$$y = 36408x - 26580 \quad (1)$$

where y = Area of chlorpyrifos as given by each chromatogram

x = Sample concentration; $x = (y + 26580)/36408$

Table 1. Peak areas and concentrations.

Concentration (mg/kg)	Peak Area
0.5	14,899
1	38,626
2	87,762
4	111,369
8	160,569

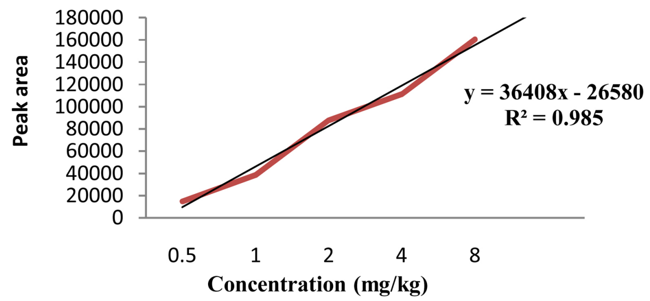

Figure 4. Calibration curve of chlorpyrifos.

To calculate the concentration of the pesticide residue in the crop sample, the following formula was used.

$$\text{Conc. of pesticide residue}(\mu g/g) = \frac{\text{Conc. of sample}\left(\frac{\mu g}{ml}\right)\text{from calibration curve} \times \text{Vol. of extract}(ml)}{\text{Sample weight}(g)} \quad (2)$$

3. Results and Discussion

Results of the analysis are presented in the tables that follow. Table 2 shows the summary of qualitative results obtained from the analysis. The frequency of pesticide residue detection with respect to the type of crop is shown in Table 3.

Table 2 shows that the two main pesticides detected in the garden crops were chlorpyrifos ($C_{22}H_{19}Cl_2NO_3$) and cypermethrin ($C_9H_{11}Cl_3NO_3PS$). Chlorpyrifos was detected in celery from Mbei, Irish potato from Njong, tomato from Baligham and Pinyin, Sweet pepper from Baligham, Mbei and Pinyin. Cypermethrin, which occurred in traces, was detected in tomato from Pinyin and sweet pepper from Mbei and Pinyin. Table 3 shows that 58.3% of the crops had pesticide residues. 33.3% of the crops contained chlorpyrifos only while 25% contained both chlorpyrifos and cypermethrin. 66.7%, 66.7%, 33.3% and 0.0% of Irish potato, celery, tomatoes and sweet pepper respectively were pesticide free. The lack of residues in some of the crops may be explained by the fact that pesticides were not used on the crops at all; contact pesticide residues present could have been flushed out by the effective washing that was done before the analysis; the crops were harvested after the pre-harvest period or the pesticide residues present could not be detected by the instrument. On the other hand, the presence of

Table 2. Summary of qualitative results of pesticides detected.

Village	Sample name	Pesticides detected	Type of pesticide	Peak area	Retention time (min)
Njong	Potato	Chlorpyrifos	Insecticide	113,625	19.191
Mbei	Potato	-		-	
Pinyin	Potato	-		-	
Njong	Celery	-		-	
Mbei	Celery	Chlorpyrifos	Insecticide	26,597	19.202
Pinyin	Celery	-		-	
Baligham	Tomato	Chlorpyrifos	Insecticide	140,152	19.191
Mbei	Tomato	-			
Pinyin	Tomato	Chlorpyrifos	Insecticide	156,749	19.202
		Cypermethrin trace (Quality <50%)	Insecticide	-	32.106
Baligham	Sweet pepper	Chlorpyrifos	Insecticide	56,933	19.203
Mbei	Sweet pepper	Chlorpyrifos	Insecticide	142,174	19.208
		Cypermethrin trace (Quality <50%)	Insecticide	-	32.111
Pinyin	Sweet pepper	Chlorpyrifos	Insecticide	104,194	19.203
		Cypermethrin trace (Quality <50%)	Insecticide	-	32.111

Table 3. Frequency and percentage of contamination of samples by pesticides.

Pesticides detected	Number of samples in which pesticide residues were detected	Percentage of contamination	Total contamination
Chlorpyrifos	4	33.3%	58.3%
Cypermethrin	-	00.0%	
Cypermethrin/ Chlorpyrifos	3	25.0 %	

chlorpyrifos residues in some samples indicated that it was used in excess. This is because chlorpyrifos is a contact pesticide which is not normally absorbed into the plant tissues when used according to good agricultural practice. It has been reported that farmers repeatedly spray different pesticides on crops to protect their crops from various pests without any awareness about their hazardous effect on health [4]. The presence of both chlorpyrifos and cypermethrin residues in sweet pepper samples from Mbei and Pinyin showed that farmers in these villages use more pesticides than those in Baligham. This implies that sweet pepper was vulnerable to insect attack in the studied area.

After substituting the various concentrations from the calibration curve (**Table 1**), into Equation (2), the concentration of chlorpyrifos was obtained for all the samples and the values are presented in **Table 4** against the maximum residue limits of chlorpyrifos given by Codex.

Table 4 shows that the concentration of chlorpyrifos was above the Codex MRLs in 28.6% of the crops. Chlorpyrifos concentration in tomato from Pinyin and Baligham was 2.0 and 1.8 μg/g respectively, which was above the Codex Maximum Residue Limit of 1.0 μg/g for tomato. These high concentrations of chlorpyrifos may be due to the fact that farmers in these villages use excessive doses of chlorpyrifos on tomato; the crops were sprayed and harvested when the pre-harvest period was not over; or spraying frequencies were not respected.

Chlorpyrifos (O, O-diethyl O-(3, 5,6-trichloro-2-pyridyl) phosphorothioate) is an organophosphate pesticide that is used to kill a number of pests [14] [15]. Its structure is presented in **Figure 5(a)**.

Table 4. Comparison of concentration of chlorpyrifos with Codex MRLs.

Sample	Village	Concentration of chlorpyrifos (μg/g)	MRL by CODEX Alimentarius commission(μg/g)	Remarks
Irish potatoes	Njong	1.5	2.0	Below MRL, safe for consumption
Celery	Mbei	0.6	1.0	Below MRLs, safe for consumption
Tomato	Pinyin	2.0	1.0	Doubles MRLs, not safe for human consumption
	Baligham	1.8	1.0	Far above MRLs, not safe for human consumption
Sweet pepper	Baligham	0.9	2.0	Below MRLs, safe for consumption
	Mbei	1.9	2.0	Very closed to MRLs
	Pinyin	1.4	2.0	Below MRLs, safe for consumption

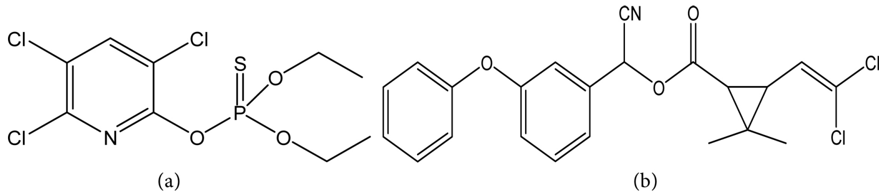

Figure 5. Structures of Chlorpyrifos (a) and Cypermethrin (b).

Chlorpyrifos is a non-systemic insecticide that is effective on direct contact, ingestion and inhalation. Its insecticidal function is as a result of the inhibition of the enzyme acetyl cholinesterase, which regulates the neurotransmitter acetylcholine in the nervous system. Chlorpyrifos is one of the most widely used pests control products in the world and it is called a broad spectrum insecticide because it can kill a wide variety of insects [13]. Exposure to chlorpyrifos exceeding recommended levels has been linked to neurological effects, persistent developmental disorders and autoimmune disorders. Exposure to chlorpyrifos during pregnancy harms the mental development of children. Farmers have a high risk of exposure and when it is sprayed at home; residents equally have a high risk of exposure. The toxicity of chlorpyrifos is determined from its lethal dose, LD50, which is the amount of the chemical needed to kill 50% of a study population. The LD50 for chlorpyrifos is estimated to be 92 to 276 mg/kg of body weight [13]. As a result of its toxicity, the Environmental Protection Agency (EPA) banned chlorpyrifos for residential purposes in 2010. The agency banned its use on crops like tomato, and limited its use on other crops including apples, grapes and citrus. Chlorpyrifos moves to all parts of the body after exposure. It is not toxic per say, but when the body breaks it down, it forms a toxic form called chlorpyrifos Oxon [16]. This toxic form binds permanently to the enzyme which controls the messages that travel between nerve cells, that is, acetyl cholinesterase [16]. When chlorpyrifos Oxon binds to so many of the enzymes, it affects the normal functioning of the nerves and muscles. Chlorpyrifos thus causes acetyl cholinesterase inhibition in humans at high doses as it over stimulates the nervous -system causing nausea, dizziness, confusion and at very high exposure, it causes respiratory paralysis and death. Sonchieu *et al.* [11] studied pesticide application on some cultivated vegetables, and its health implications in Santa and reported similar results. They showed that spraying parameters like dose, spray frequency and pre-harvest periods are poorly practiced; and ailments such as abdominal pain, vomiting, skin irritations, cancer and reproductive problems are now common in Santa. 71% of the crops studied had chlorpyrifos concentration below the Maximum Residue Limits. These crops were: Irish potato from Njong, Celery from Mbei and sweet pepper from pinyin, Mbei and Baligham. These low residue concentrations do not have any effect on human health and the crops were, thus, safe for human consumption.

Cypermethrin is a synthetic pyrethroid pesticide used as a broad-spectrum insecticide in large scale agricultural applications and also in consumer products for domestic purposes. Its IUPAC name is [cyano-(3-phenoxyphenyl) methyl]3-(2, 2-dichloroethenyl)-2, 2-dimethylcyclopropane-1-carboxylate and its structure is shown in **Figure 5(b)**. Cypermethrin degrades easily on soil and plants but can be effective for weeks when applied to indoor inert surfaces. Cypermethrin is moderately toxic through skin contact or ingestion. Human beings are also exposed to cypermethrin toxicity by consuming contaminated crops and other products like fish in which it is highly toxic [17]. It causes skin and eye irritation. Symptoms of dermal exposure include; numbness, tingling, itching, burning sensation, loss of bladder control, seizures and possible death. In humans, cypermethrin is deactivated by enzymatic hydrolysis to several carboxylic acid metabolites which are eliminated in urine [18]. The two pesticides detected in this study (chlorpyrifos and cypermethrin) are a call for concern, as previous studies done on health risk among pesticide sellers in Bamenda (Cameroon) and peripheral areas, identified them to be prominently used on farm lands in this area [19].

4. Conclusions

This study was aimed at performing a multi-residue analysis of crop samples to detect and quantify pesticide residues present, and thus determine their level of contamination. The concentration of chlorpyrifos in Irish potato from Njong, celery from Pinyin and sweet pepper from Pinyin, Mbei and Baligham, was below the Codex maximum Residue limits (MRLs). These crops were, thus, safe for human consumption since they can not cause any negative health effects in humans. The concentration of chlorpyrifos in tomato from Pinyin and Baligham was 2.0 and 1.8 μg/g respectively against the Codex MRLs of 1.0 μg/g. Therefore, these concentrations were far above the Codex MRLs. These crops were therefore not fit for human consumption, because the high concentration of residues will cause severe negative health effects. The farmers in Santa in general, Pinyin and Baligham in particular, are called upon to use pesticides according to the labeled directions prescribed by the manufacturers in order to avoid crop contamination.

A broader research needs to be carried out in Santa to assess the level of pesticide residues. Furthermore, drinking water resources in this locality should be analyzed to determine the level of pesticides contamination.

Acknowledgements

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Data Availability

Relevant data are available for consultation if needed.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this article.

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