**Level of Organochlorine Pesticide Residues in Selected Soft Drinks Commonly Sold in Benin City Markets**

**Abstract;** This research studied the levels and risk assessment of pesticide residues in selected soft drinks such as sprite and coca-cola commonly sold in Benin City markets. A total of twenty-five organochlorine pesticide (OCP) residues in these samples were analyzed using with gas chromatography- electron captured detector (ECD). The estimated chronic daily intake (ECDI) of aldrin in soft drinks (sprite) was determined based on European Union (EU) and United States Agency of Toxic Substances and Disease Registry (ATSDR) standards, using two population age groups (child and adult). The results revealed the levels of organochlorine pesticide residues among the soft drinks. Aldrin, had a value of 0.100mg/kg in soft drink (sprite), while coca-cola were below detection limit (0.00±0.00). The study, thus suggest that OCPs concentration in sprite is relatively safe for human consumption. The study therefore calls for continuous monitoring of agricultural farmlands because continuous exposure to pesticide contaminated soft drinks sold in Benin City markets (sprite) could affect the health of consumers.

**Keywords**; organochlorine pesticides, sprite, coca-cola, chromatogram

INTRODUCTION

Pesticides are chemical substances generally used in agriculture to increase the yield, improve the quality, and extend the storage life of agricultural produce (10). They help society with immense benefits, such as increasing agricultural output and in the control of diseases.

There has been an alarming rate about ingestion of pesticide residues by adults and children who consume soft drinks. Consumption of soft drinks has increased over time because of its demand. Xenobiotics are a major cause for concern all over the world, given their persistence to their metabolite residues after degradation by artificial or natural means and negative effects on humans (12).

UNDER PEER REVIEW

In 2001, about 74 percent of soft drinks sold in the United States of America (USA) were reported to contain at least one pesticide residue greater than the acceptable level (13). This increase in level of pesticide residues has risen in developing countries and the fastest growing markets in Africa, Asia, South and Central America, Eastern Mediterranean (13).

The toxic effects of these pesticides has caused them to be banned in developed and many growing nations. Moreover, some growing countries still use them (14).

In Nigeria, there have been reports of some levels of pesticide residues in soft drinks (8). The results have often revealed contamination by organochlorine pesticides in soft drinks sold in Benin City markets (14). The organochlorine pesticides (OCPs) include aldrin, endrin, dieldrin, chlordane, heptachlor, DDT, toxaphene, endosulfan 1, and hexachlorobenzene (HCB) (17). These dreaded organochlorine pesticides get into the raw materials and water for processing soft drinks via ; (lack of awareness, inappropriate use of pesticides, waste from industrial chemical production, pesticides runoff from agricultural areas, sewage and refuse dump) . Because of their potency, efficiency and low cost compared with alternative pesticides, organochlorine pesticides (OCPs) are still being used by some farmers (5). The use of OCPs such as DDT has been outlawed since 1990 in Nigeria.

These compounds are characterized by high persistent, low polarity, low aqueous solubility and high lipid solubility (lipophilicity). They are ecotoxic, non-biodegrable and able to bioaccumulate and biomagnify in humans via consumption of contaminated soft drinks which may pose health risk to human (8) . To estimate the risk of detected pesticides to humans (young children and adults) through drinking/ingestion of contaminated sprite, the hazard quotient (HQ) method was employed (17).

Hazard quotient (HQ) = ECDI/RfD

Where ECDI is the estimated chronic daily intake

This was obtained by using the Formula ECDI (ingestion) =Cs IR(sprite) x EF xED/BW xAT where Cs is the concentration of sprite samples, IR (sprite) is the ingestion rate of sprite, EF is the exposure frequency, ED is the exposure duration, BW is the body weight, and AT is the average life Span (8).

The aim of this study were to assess the level of organochlorine pesticide residues in soft drinks (sprite and coca-cola) sold in markets in Benin City.

UNDER PEER REVIEW

## MATERIALS AND METHOD

### Study area

Benin City have alot of super markets located in different localities in the city where soft drinks (sprite and coca-cola) are sold for final consumption. The markets include Ekiosa, New Benin, Oba market, Ikpoba hill market and Uselu market. The soft drinks (sprite and coca-cola) are usually transported from within the state or other neigboring Western, Eastern, Southern or Northern part of the country.

### Sample Collection

Random sampling of soft drinks ; sprite and coca-cola were purchased from the various markets in Benin city. However, samples were provided to the analytical laboratory for GC analysis (2)

### Organochlorine pesticide residue extraction

The persistent organochlorine pesticide residues from soft drinks (sprite and cocacola) samples were analyzed chromatographically with gas chromatography equipped with electron capture detector (GC/ECD) as described by (17).

Thirty grams (30 g) aliquot of well mixed samples was placed separately into a solvent rinsed beaker. 50:50 mixed with acetone were prepared. One milliliter (1 ml) of decachlorobiphenyl was added and mixed thoroughly using glass stir rod. 1.5 g of anhydrous sodium sulfate ($Na_2SO4$) were weighed and added to the sample and mixed thoroughly to form a free flowing powder. 50ml of the solvent were mixed and added to the samples. Samples were placed in the Sonicator and Sonicated for about 10 – 15 minutes at about $60^0C$. The extract was decanted into a round bottom flask. The procedure was repeated once more an additional 50 ml of solvent were mixed, Sonicated and allowed to settle in the beaker before it was decanted into the round bottom flask. The samples were extracted with 2ml concentrated rotary evaporator. 5 ml of hexane was added to the extract. It was then allowed to evaporate to reduce the volume to 2 ml. The final hexane volume was 2 ml, giving final sample weight of 15g/ml.

UNDER PEER REVIEW

### Sample cleanup

A syringe was used to transfer 2 ml of hexane into a 10 ml vial extract in a fume cupboard. 5ml of the 1:1 Sulfuric acid were carefully added, It was ensured there was no exothermic reaction. The cap of the flask was capped tightly and vortexed for 1-min. The phases were allowed to separate for at least 3mins, the upper hexane layer was observed and ensured that it was not highly colored. After the cleanup procedure, each fraction was evaporated with rotary evaporator and concentrated in a nitrogen stream to 1 ml. The fractions were analyzed for pesticide residues. The organochlorine residues concentrations were determined using a Hewlett-Packard ($H_P$) 5890 series II equipped with electron capture detector with auto-sampler. The chromatographic separation was achieved by using an HP-1 of 30 x 0.25 mm internal diameter (ID). The chromatographic temperature program was kept at 100 $^{O}$C for 1 min; increased to 200$^{O}$C for 2 min. The injection volume was 1ml. The detector temperature was maintained at 300$^{O}$C.

UNDER PEER REVIEW

### Calculation for sample analysis

The concentration of each analyte range in a sample was calculated directly from the Instrument using the Data Analysis Software. The final sample weight and the dilution factor can be use in the batch file and the final results would be generated by the software. OR

The use of the Response Factor Method could be employed (18).

$$Cf = \frac{Area\ (p) \times Rf \times Vf \times DF \times 1000}{Wi}$$

Where:

Cf = Final Sample concentration (mg/l)

Area (p) = Measured area of peak (peaks)

Wi = Initial weight extracted (g dry weight)

Vf = Final extract volume (ml).

Df = Dilution factor of sample or extract if diluted.

RF= Response factor from the calibration standard calculation

$$RF = \frac{\text{Concentration (P)}}{\text{Area (P)}}$$

Concentration (p) = Concentration of peak or Total concentration of range

Area = Area of peak or total across range.

UNDER PEER REVIEW

**Estimated Standards for Acceptable and Chronic Daily Intake of Soft Drinks used in this study**

| | Unit | Child | Adults |
|---|---|---|---|
| Ingestion rate (IR) | L/day | 1 | 1.41 |
| Exposure frequency (EF) | Day/year | 350 | 350 |
| Exposure duration (ED) | Year | 6 | 30 |
| Body weight (BW) | kg | 10 | 70 |
| Average life span (AT) | Day | 2190 | 8760 |

UNDER PEER REVIEW

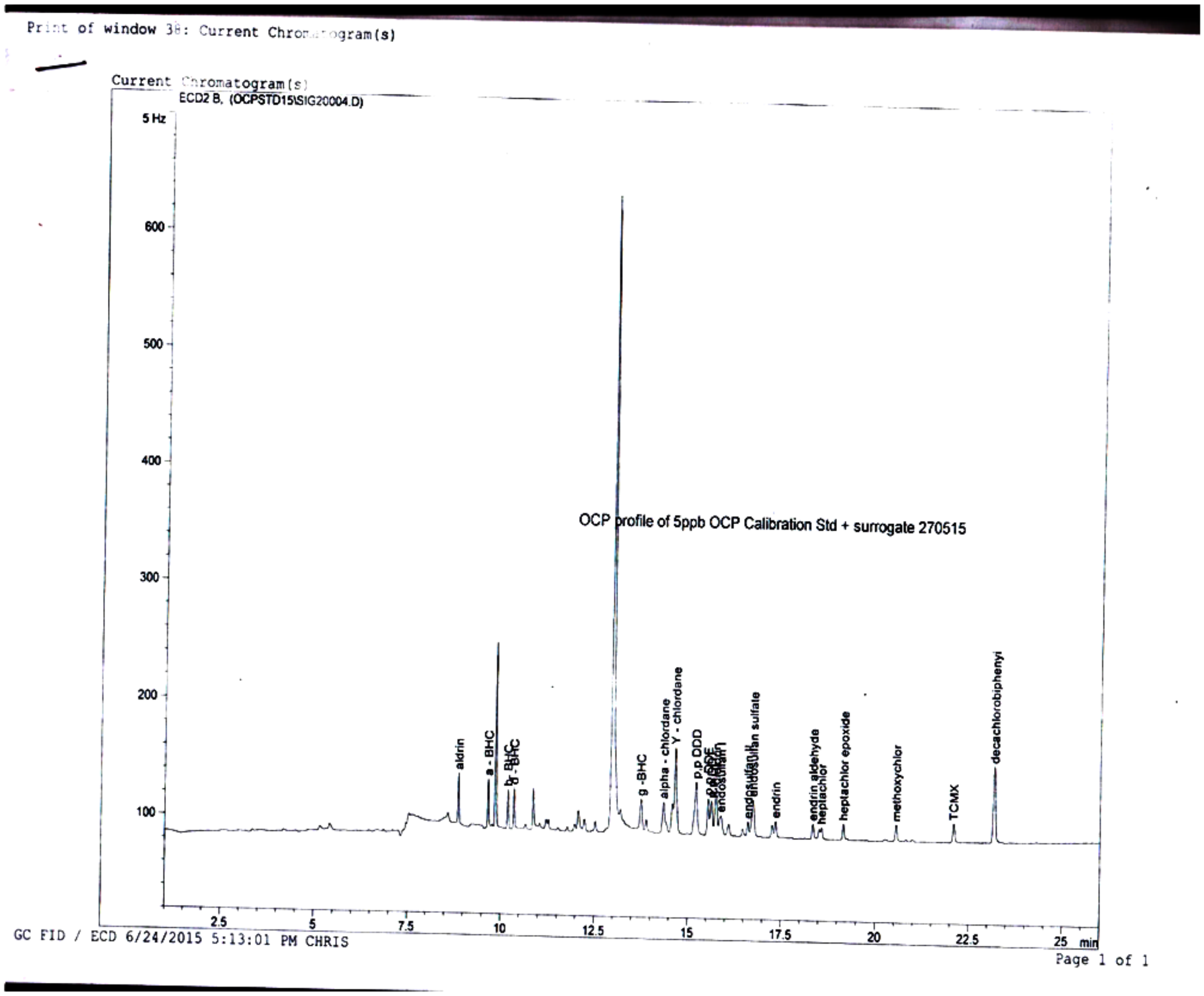


Figure 1; Standard of organochlorine pesticide used in this study

## Results and Discussion

The concentration of pesticide residues in soft drinks (sprite and coca-cola) sold in Benin city, Edo state has been investigated. Figure 1, 2 and 3 shows the representative chromatogram standard and samples. No interference peaks were obtained for the blank sample chromatogram at the same retention time as the targets compounds. The mean recovery values for the spiked samples are shown in Table 1. The procedure employed in this study is reproducible, efficient and reliable for the analysis of OCPs as stipulated by EU guidelines for evaluating accuracy and precison method (7).

The chromatogram result in this study is in consonance with that of (1) who assessed soft drinks in Addo Ekitti, Ogun and Benin. Reports from this investigation reveal that organochlorine pesticide residues ranged from below detection limit(BL)( 0.00±0.00) to 0.0001mg/kg in (Table1).

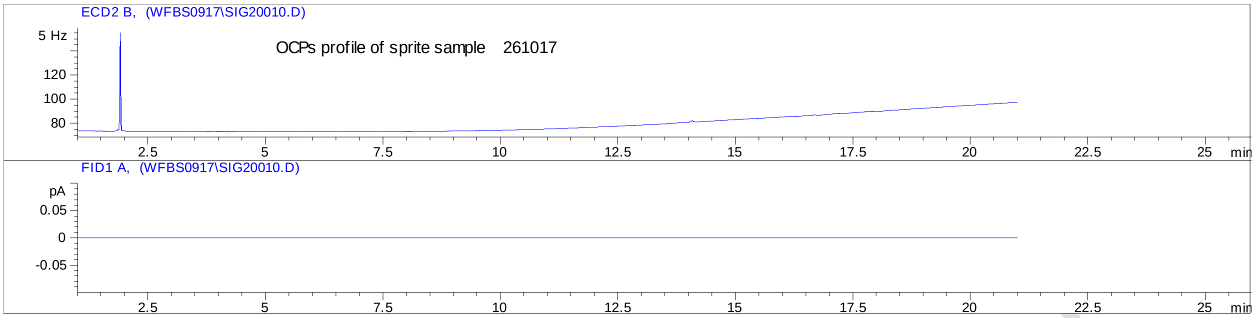


**Figure 2; Chromatogram of sprite samples analysed**

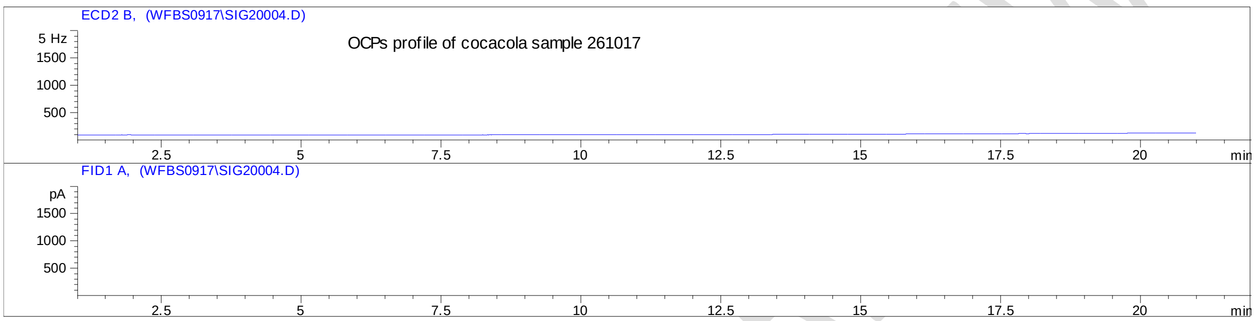


**Figure 3; Chromatogram of coca-cola samples analysed**

Twenty five (25) pesticides were assessed in these consumable products (Table.1) with aldrin having the highest mean concentration (0.100mg/kg) in sprite sample.

**Table 1; Mean concentration of pesticide residues in soft drinks**

| COMPONENT | Sample 1 sprite | Sample 2 coca-cola |
|---|---|---|
| **ALDRIN** | 0.100±0.00 | 0.00±0.00 |
| **a – BHC** | 0.00±0.00 | 0.00±0.00 |
| **b – BHC** | 0.00±0.00 | 0.00±0.00 |
| **d – BHC** | 0.00±0.00 | 0.00±0.00 |
| **Gamma - BHC (LINDANE)** | 0.00±0.00 | 0.00±0.00 |
| **ALPHA – CHLORDANE** | 0.00±0.00 | 0.00±0.00 |
| **GAMMA – CHLORDANE** | 0.00±0.00 | 0.00±0.00 |
| **ATRAZINE** | 0.00±0.00 | 0.00±0.00 |
| **p,p DDD** | 0.00±0.00 | 0.00±0.00 |
| **p,p DDE** | 0.00±0.00 | 0.00±0.00 |
| **p,p DDT 4, 4 DDT** | 0.00±0.00 | 0.00±0.00 |
| **DIEDRIN** | 0.00±0.00 | 0.00±0.00 |
| **ENDOSULFAN 1** | 0.00±0.00 | 0.00±0.00 |

UNDER PEER REVIEW

| | | |
|---|---|---|
| **ENDOSULFAN 11** | 0.00±0.00 | 0.00±0.00 |
| **ENDOSULFAN SULFATE** | 0.00±0.00 | 0.00±0.00 |
| **ENDRIN** | 0.00±0.00 | 0.00±0.00 |
| **ENDRIN ALDEHYDE** | 0.00±0.00 | 0.00±0.00 |
| **HEPTACLOR** | 0.00±0.00 | 0.00±0.00 |
| **HEPTACHOR EPOXIDE** | 0.00±0.00 | 0.00±0.00 |
| **METHOXYCHLOR** | 0.00±0.00 | 0.00±0.00 |
| **DIAZINON** | 0.00±0.00 | 0.00±0.00 |
| **PHOSPHORO METHYL GLYCINE** | 0.00±0.00 | 0.00±0.00 |
| **TCMX** | 0.00±0.00 | 0.00±0.00 |
| **CARBAMATE** | 0.00±0.00 | 0.00±0.00 |
| **DECACHLOROBIPHENYL** | 0.00±0.00 | 0.00±0.00 |
| **TOTAL OCP (mg/Kg)** | **BDL** | **BDL** |

KEY :OCP; Organochlorine pesticide residue, BDL;Below detection limit

The relative presence of these organochlorine pesticide residues in soft drink (sprite) showed that farmers still use them for the control of pest. This is because of their potency, efficiency and low cost compared with alternative pesticides (banned or not) (15).

Several activities (volatilization, photolysis, penetration through the plant surface, inadequate training of personnel, inappropriate use of pesticides) could be attributed to the levels of chlorinated hydrocarbon compounds in soft drink sold in Benin City markets (8). The findings in this study is in consonance with the findings of (12) observed that organochlorine pesticide residue in sprite ranged from 0.100 to 0.75mg/kg in Benin City, Addo Ekitti, Ogun, Lagos and Ghana metropolis.

Previous studies have demonstrated that organochlorine pesticide like gamma lindane is toxic and can affect non target organisms other than the organisms of interest, thereby causing great menace to ecosystem and to consumers (4).

The use of organochlorine pesticides for the control of pest by farmers is worldwide . These compounds are characterized by high persistent, low polarity, low aqueous solubility and high lipid solubility (lipophilicity). They are ecotoxic, non-biodegrable and able to bioaccumulate and biomagnify in living organisms (16). The major problems are their toxic effects such as interfering with the reproductive

UNDER PEER REVIEW

systems and foetal development as well as their ability to cause cancer, cardiovascular disease, asthma and other health related diseases .

**Table 2: Estimated chronic daily intake (CDI) and hazard quotient (HQ) for OCPs in sprite**

| Pesticide | Concentration | child | | Adult | | RQ |
|---|---|---|---|---|---|---|
| Aldrin | 0.0001 | ECDI | HQ | ECDI | HQ | |
| | | 1.8E-04 | 8.3E-06 | 2.4E-06 | 5.8E-04 | |
| Hazard index | | 8.3E-06 | | 5.8E-04 | | 1.89E-08 |

However, the estimated chronic daily intake (ECDI) of (aldrin) in soft drinks (sprite) (non dietary), risk quotient (RQ), hazard quotient (HQ) and the hazard index (HI) in (Table 2) were calculated using two population groups with varying body weights (35 and 70kg) (Ezemonye *et al*., 2008). The study revealed that the risk quotient (RQ) were (1.89E-08), estimated chronic daily intake (ECDI) for both groups were (1.8E-04) and (2.4E-06) (Table 2). The hazard quotient (HQ) were (8.3E-06) and (5.8E-04). Hazard index (HI) (8.3E-06) and (5.8E-04) were also estimated for both population groups. The result in this study is in consonance with the result of (6) which was (BL -0.256mg/kg) in Ghana and Addo Ekitti.

Organochlorine pesticide residues at any given concentration are highly toxic, bioaccumulate and not readily biodegraded (12). Though the estimated concentrations in this study were minute, ecological risk assessment showed that there is a potential of toxic effects to soft drink (sprite) upon exposure to organochlorine pesticides. Risk projections for humans from non-dietary intake also revealed that there is potential for cancer effects. Projections showed that both children and

UNDER PEER REVIEW

adult were at high health risk. The result in this study is in line with the United States Agency of Toxic Substances and Disease Registry (3) and European union (7) standard for estimation of OCPs (0.0100 to 1mg/kg) in sprite that could be considered as unsafe to human.

**Conclusion**

The study, thus suggest that OCPs concentration in sprite is relatively safe to human. The study therefore calls for continuous sensitization to farmers from the use of these dreaded chemicals, that could contaminate the raw materials for processing and periodic investigation of soft drinks (sprite ) sold in Benin city markets.

UNDER PEER REVIEW

**COMPETING INTERESTS DISCLAIMER:**

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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UNDER PEER REVIEW

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UNDER PEER REVIEW