***Original Research Article***

# Quality and Risk Assessment for Public Health of Polycyclic Aromatic Hydrocarbons in Crude Palm Oil Locally Produced from South-South zone, Nigeria

**Quality of the English language should be improved. In most of the parts, punctuations and grammatical errors are there. These should be addressed.**




## Abstract

**Background**: Oil palm, *Elaeis guineensis Jacq* is a perennial crop mainly cultivated for its vegetable oil. The fatty acid composition of CPO full form is mostly composed of palmitic the fatty acid composition of CPO is mostly composed of palmitic, stearic, oleic, and linoleicacids, stearic oleic, and linoleic acids. **Objectives**: This study evaluated the quality of locally produced crude palm oil (CPO) from six states of South-South zone, Nigeria. **Methods:** The parameters of CPOs were characterized by specific gravity (SG), acid value (AV), percentage of free fatty acid (%FFA), and saponification value (SV) and peroxide value (PV). Polycyclic aromatic hydrocarbons (PAHs) in CPO were quantified using standard procedures, the related risk due to daily consumption of 25 g was characterized by lifetime cancer risk (LCR) and margin of exposure (MOE). **Results:** The study observed that the mean values of the parameters were 0.931 SG; 5.01 mgKOH $g^{-1}$ AV; 2.28% FFA; 177.27 mgKOH $g^{-1}$ SV and 29.65 meq$O_2$ $kg^{-1}$ PV. Apart from %FFA, the others did not meet the requirements recommended by the Codex Alimentarius Commission of the joint WHO/FAO. Diagnostic ratios of PAHs suggested a mixture of petrogenic and pyrogenic origins. The MOEs showed low health concern while 16PAH-LCR values which ranged from 2.48E-04 - 4.48E-04 were unacceptable. The screening value 0.003 was significantly ($p > 0.05$) lower than the computed total BaP equivalent (BaPeq) values indicating potential health concerns. It is recommended that CPO processing and storage methods should be properly monitored to prevent quality reduction and contamination, which might have adverse effect on consumers.


**Keywords**: Crude Palm Oil, Physicochemical Properties, Polycyclic Aromatic Hydrocarbons, Margin of Exposure, Lifetime Cancer Risk

## 1. INTRODUCTION

Oil palm, *Elaeis guineensis Jacq*. thrives in the humid, tropical region where it originated and is perennial crop mainly cultivated for its vegetable oil [1]. There are two important species of palm trees (Genus: *Elaeis*), *E. guineensis* and *E. oleifera*. *E. guineensis*, mainly found in West Africa, is commercially cultivated and a high-yielding specie than the *E. oleifera* mainly in

Americas [2, 3]. The two types of oil that can be obtained from oil palms are crude palm oil (CPO), which is obtained from the fruit's fleshy mesocarp, and palm kernel oil (PKO), which is obtained from the fruit's endosperm or kernel [2]. Ripe fruits have 70–75% oil in the mesocarp, which makes up the fruit's overall weight. Glycerides make up the majority of the CPO, with trace amounts of non-glyceride components [4]. Free fatty acids (FFAs), moisture, contaminants, trace metals, and minor components are among the non-glyceride components; on the other hand, the fatty acid composition of CPO is mostly composed of palmitic (44%), stearic (4%) oleic (40.6%), and linoleic (9.8%) acids [5]. For the culinary and chemical industries, it is therefore a great source of fatty acids, minerals, and vitamins [6]. Numerous foods, as well as home, industrial, and health items, contain both vegetable oils derived from oil palm. Over half of all vegetable fats and oils consumed are made from palm oil, a traditional food item that is widely consumed worldwide [7]. It is one of the main types of fats and oils produced in Nigeria, a staple in the majority of regional meals eaten by Nigerians, and a valuable component of the diets of many people [8, 9]. Reddish and rich in nutrients, CPO has been shown to treat a number of illnesses, including cancer and heart conditions [10, 11]. It has been reported that Malaysia, Indonesia and Nigeria are the top three countries in the world for CPO production, having produced more than 85% of the global supply [12].

Because of its lipophilic nature, CPO, like other vegetable oils and foods, can be readily contaminated by organic compounds like polycyclic aromatic hydrocarbons (PAHs). Because of its well-known carcinogenic, teratogenic, and mutagenic properties [13, 14], the presence of PAHs in vegetable oils and foods is a serious health concern. This issue has been thoroughly researched [14, 15, 16]. Vegetable oils contamination by PAHs may be due to environmental pollution, unhealthy farming methods, or oil processing [17].

Generally, PAHs are released into the environment from natural and man-made sources which could either be of pyrogenic, petrogenic, biogenic or diagenetic origin [14, 18]. Petroleum seeps and unintentional spills, and terrestrial runoff are the main ways that petroleum products—both refined and crude—emit petrogenic PAHs. High temperature burning and/or pyrolysis of organic matter, biomass and fossil fuels including coal are characteristics of PAH of pyrogenic (or pyrolytic) origin [14, 18]. Biogenic PAHs are released from biological generation and degradation processes while PAHs of diagenetic origin are from chemical or biological transformation of natural organic matter.

PAHs have been demonstrated to have negative effects on humans, including the formation of DNA adducts by their metabolites, which can cause structural changes and subsequent mutations [19]. According to reports, exposure to PAHs can lead to adverse effects on the respiratory and cardiovascular systems, increase the risk of aberrant neurobehavioral development in children, and induce asthma in children [20. 21]. Limits for PAHs in different foods have been established by numerous regulatory organizations worldwide due to the negative effects of exposure to PAHs. The scientific opinion on PAHs in food published by the European Commission's EFSA concluded that benzo[a]pyrene was not a suitable indicator for the presence of PAHs in food. Instead, the total amount of four PAHs—benzo[a]anthracene (BaA), chrysene (Chr), benzo(a)pyrene (BaP), and benzo(b)fluoranthene (BbF)—is a more appropriate indicator for the

presence of PAHs in food as they are classified as mutagenic, genotoxic and carcinogenic [22]. The German Society for Fat Science recommended 0.025 mg $kg^{-1}$ for total PAHs [23, 24] and the European Commission has recommended maximum permissible levels (EU limits) of PAHs in vegetable oils and fats of 0.002 mg $kg^{-1}$ for BaP and 0.01 mg $kg^{-1}$ for PAH4 (sum of BaA, Chr, BaP, and BbF) [25]. Because of these negative consequences and the growing amount of PAHs in the environment, monitoring PAHs is a key priority in all programs that assess ecological and public health risks.

In order to assess and evaluate the quality of edible palm oil, the following physicochemical characteristics are helpful: specific gravity, acid value, acid value, percentage of free fatty acid (%FFA), saponification value, ester value, and peroxide value [26, 27]. The goals of this study were to: (i) assess the quality of CPO samples collected from South-South zone, Nigeria *Elaeis guineensis* growing areas and compare them to national and international standards; (ii) measure the levels of PAHs in the samples from South-South Nigeria following traditional processing methods; and (iii) estimate the risk to consumers associated with dietary exposure to PAHs on consumption of the products under examination using the margin of exposure (MOE) approach and lifetime cancer risk (LCR).

## 2. MATERIALS AND METHODS

### 2.1 Area of Study

There are six geopolitical zones in Nigeria of which South-South is one of them and comprises of six states, namely: Akwa, Ibom, Bayelsa, Cross, River, Delta, Edo, and Rivers. The map of study area showing sampling sites, is shown in Figure 1[28].

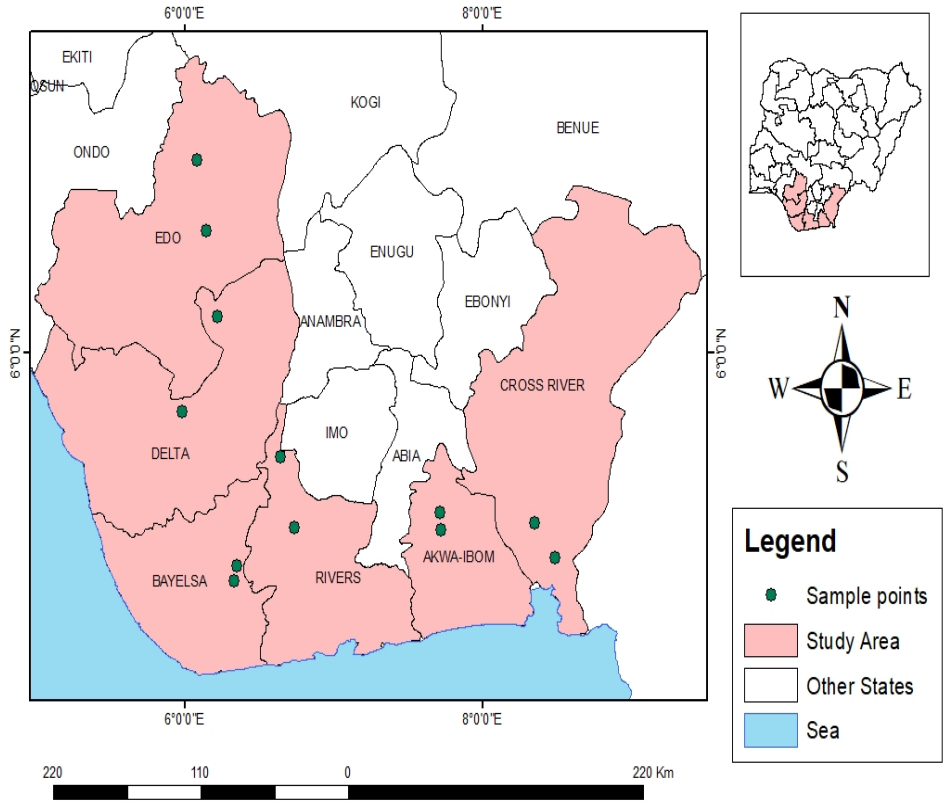


Figure 1: Map of study area [28]

## 2.2 Sample collection

The palm oil samples were collected in clean sterilized and oven dried amber glass bottles from local producers directly from their oil mills during extraction. Total of thirty-six samples were collected, six samples from each state (code named AKS, BYS, CRS, DES, EDS, and RVS), from the six states of South-South Nigeria (Table1). The oil samples were packed in plastic bags and stored in darkness under room temperature in the laboratory.

**Table 1: Coordinates of states in Study area**

| State | Town (Local Government Area) | Latitude | Longitude |
|---|---|---|---|
| Akwa Ibom (AKS) | Afaha Obong (Abak) | 5.0420 | 7.7227 |
| | Ukana Ikot (Obot Akara) | 5.1327 | 7.7166 |
| Bayelsa (BYS) | Elebele (Ogbia) | 4.7740 | 6.3268 |
| | Aghoro (Ekeremor) | 4.8559 | 6.3421 |
| Cross-Rivers (CRS) | Adiabo Efut (Odukpani) | 5.185 | 8.1330 |
| | Ikot Edem Odo (Akpabuyo) | 4.9008 | 8.4811 |
| Delta (DES) | Owa Agbor (Ika North East) | 6.2086 | 6.2230 |
| | Kokori (Ethiope East) | 5.6879 | 5.9841 |
| Edo (EDS) | Iruekpen (Esan West) | 6.6546 | 5.1396 |

| | | | |
|---|---|---|---|
| | Uzalla (Owan East) | 6.6546 | 5.1396 |
| Rivers (RVS) | Omuku (Ogba/Egbema/Ndoni) | 5.4512 | 6.6409 |
| | Elele Alimini (Emuoha) | 5.0599 | 6.7300 |

### 2.3 Chemicals and reagents

All chemicals and reagents used in this study were of analytical grade. The surrogate standard, mixture containing naphthalene-d8, acenaphthene-d10, phenanthrene-d10, chrysene-d12, and perylene-d12 (Sigma-Aldrich), was added to the samples before extraction and used as internal standards for quantification. Working solutions of 16PAHs were prepared in acetonitrile (100 ng/mL). All working standard solutions were freshly prepared weekly and stored at -4 °C in the dark in order to maintain the integrity of the PAH compounds. Distilled water was used throughout the work for preparation and dilution of standard solutions

### 2.4 Sample extraction and clean-up

The extraction procedure as described [29] was adopted in this study. An aliquot (1 g) of the sample was dissolved in 12 ml of n-hexane-DMF 1:2 (v/v), agitated for 20 s, ultrasonicated for 10 min, and followed by centrifugation for 5 min at 4000 rpm. The lower DMF phase was then transferred into a 100 ml separatory funnel. The extraction process was carried out with a fresh portion of 8 ml of N,N-DMF. The N,N-DMF extracts were combined and washed with 50 ml of a 4% saline solution, and 8 ml of n-hexane was used to back-extract the washed extract three times, each time with a fresh portion of n-hexane. The combined hexane extracts were washed twice with 50 mL of 4% saline solution, and then heated to 70 ºC. A gentle stream of nitrogen gas was used to evaporate the n-hexane extract to approximately 1 ml. A column packed with 2 g of aluminium oxide (5% deactivated, upper part) and 2 g of silica gel (5% deactivated, lower part) was used for the clean-up of the concentrated extract. The PAHs were eluted with 5 ml of n-hexane/dichloromethane (1:2 v/v) and the eluted fraction was subsequently evaporated to 1 mL with a gentle flow of pure nitrogen. The samples were stored at -4 ºC until the analyses were completed.

### 2.5 GC–MS analysis of PAHs

The analyses of the extracted samples were performed using a GC apparatus (Agilent Technology 7890A, USA) with a mass spectrometer (Agilent Technology 5975C, USA) coupled with a DB-5 ms Column (30 m length × 0.25 mm inner diameter × 0.25 μm film thickness, Agilent Technology, USA). The oven initially prepared at 80 °C for 1 min was heated to 245 °C at a rate of 4 °C $min^{-1}$. And then, the temperature of oven was ramped up to 270 °C at a rate of 30 °C $min^{-1}$ and finally held for 10 min. The carrier gas (helium) flow was set at 1.5 ml $min^{-1}$ and the injector temperature and injection volume were set at 320 °C and 1 μl, respectively. The analyte was injected to GC–MS with a splitless mode. Temperature of MS source was 250 °C and spectrometry of MS was obtained by using selective ion monitoring (SIM) modes with the electron ionization (EI) at 70 eV [30].

### 2.6 Analytical determinations

The physicochemical parameters examined and quantified include specific gravity, acid value and free fatty acids, saponification value, and peroxide value. The specific gravity was determined using a pycrometer gravimetric method [31]. Acid value and free fatty acids were determined by adopting an existing method [32]. Saponification values were determined by the titrimetric method [33], while the peroxide value was determined based on the iodometric method [34]. Ester value is determined as the difference between saponification and acid value.

### 2.7 Digestion Methods for 16 Polycyclic aromatic hydrocarbons and Sample Analysis

A validated HPLC-FLD method was used for the determination of PAHs in vegetable oils [21]. The CPO samples (2 g) were put into 15 mL polypropylene centrifuge tubes, and then 10 mL of ethylene acetate:cyclohexane (1:1, *v/v*) was added. The samples were vortexed for 1 min and sonicated for 15 min. After centrifugation (4500 r/min for 3 min), the supernatant was collected and purified. The upper phase was placed into a 5 mL vial and the sample was injected in the gel permeation chromatography (GPC) system with a purification column (200 mm × 25 mm, styrene divinylbenzene copolymer gel, Bio-Beads S-X3). Ethylene acetate: cyclohexane (1:1, *v/v*) was used as the mobile phase at a column flow rate of 5 mL/min. The eluate containing the target PAH was collected from 19 to 50 min, evaporated, and concentrated into a small volume using a vacuum rotary evaporator. After that, it was transferred and dried to near dryness using a pressure blowing concentrator and re-dissolved in 0.5 mL of acetonitrile. Finally, the extract was transferred into an auto-sampler vial for determination by HPLC-FLD.

### 2.8 Laboratory Quality Control (QC)

All glassware and apparatus to be used through the entire analysis were first washed with tap water and detergent in order to keep the analyte from contamination. Then, they were soaked with deionized water and followed by rinsing with 0.5 N $HNO_3$, and rinsed again with distilled water and a corresponding analyte solution to be analysed based on its necessity.

### 2.9 Sources of PAHs in crude palm oils

Source identification (or apportionment) of PAHs in the environment has been well studied over the last four decades and various techniques have been proposed as ideal tools. Molecular diagnostic ratios (MDRs), relationships between the concentrations of PAH congeners, are the most common methodology, often preferred due to its simplicity [14, 18]. The MDR values are used to distinguish between pyrogenic and petrogenic sources of PAHs, or PAHs derived from fuel or coal/grass/wood combustion, PAHs that come from traffic, fresh PAHs emissions, etc. In this study, of the 16PAHs assayed in the CPO samples, only nine (Nap, Acy, Ace, Flu, Phe, Ant, Flt, Pyr, and Chr) were detected and among the combustible PAHs (Flt, Pyr, BaA, Chr, BkF, BbF, IdP), Flt, Pyr, and Chr only were detected, (∑CombPAHs (= Flt + Pyr + Chr) [35, 36]. Regarding their aromatic ring numbers, the 9 PAHs of this study can be classified into two classes: low molecular weight (LMW-PAHs), composed of 2-ring and 3-ring PAHs (Nap, Acy, Ace, Flu, Phe and Ant), and high molecular weight (HMW-PAHs) composed of 4-ring PAHs (Flt, Pyr, and Chr) [37].

In this study, the following five MDRs were applied: Phe/Ant, Ant/(Ant+Phe), Flt/(Flt+Pyr), low molecular weight PAHs (LMW-PAHs)/high molecular weight PAHs (HMW-PAHs) and combustible PAHs (∑CombPAHs)/total PAHs (∑PAHs) [36, 38]. The LMW-PAHs detected are composed of 2-ring and 3-ring PAHs (Nap, Acy, Ace, Flu, Phe, and Ant) and the HMW-PAHs are composed of 4-ring, 5-ring and 6-ring PAHs (Flt, Pyr, and Chr). The results are interpreted as follows: Ant/Phe ratio > 0.1 pyrogenic; < 0.1 petrogenic, Ant/(Ant+Phe) ratio >0.1 pyrogenic - biomass combustion and <0.1: petrogenic; Flt/(Flt+Pyr) <0.4: petrogenic, 0.4 - 0.5: pyrogenic - petroleum combustion (e.g. combustion engines, and furances), and >0.5: pyrogenic - biomass combustion (e.g. grass, wood, or coal combustion), LPAHs/HPAHs (<1: pyrogenic and >1: petrogenic) and ∑CombPAH/∑PAH < 0.3: petrogenic, 0.3-0-7: mixed sources, > 0.7: pyrogenic [14, 38, 39].

### 2.10 Exposure Assessment

The BaP was used as the representative PAH for the toxicity equivalent factors (TEFs) to express the relative carcinogenic risk of PAHs. The BaP equivalent concentration (BEC) was computed for individual PAH congeners by multiplying its concentration with its corresponding toxic equivalent factor (TEF) and the sum of the BECs is the total BaP equivalent (BaPeq). The BaPeq was estimated for ∑PAHs and ∑PAH4 (BaA, Chr, BaP, and BbF). The BEC and BaPeq for the PAHs in the CPO samples were calculated according to Eqns. (1) and (2) [40]:

$$BEC_i = C_i \times TEF_i \quad (1)$$

$$BaPeq = \sum BEC_i \quad (2)$$

where $C_i$ is the concentration of $i^{th}$ PAH congener detected in the sample (mg $kg^{-1}$), $TEF_i$ is corresponding TEF of congener i and $BEC_i$ is the BaP equivalent concentration of $i^{th}$ congener in CPO. The TEF values used in this study were associated with the cancer evidence from individual PAHs. The TEFs of the nine detected PAHs are 0.001 for Nap, Ace, Flu, Phe, Flt, Pyr, and 0.01 for Ant and Chr [16]. The sum of all detected 9PAHs ($BaPeq_{PAHs}$) was used to assess the lifetime cancer risk while $BaPeq_{PAH4}$ was used to evaluate MOE.

The chronic daily intake (CDI) of PAH exposure levels for BaP, PAH4, and ΣPAHs were calculated using the BaPeq according to Eqn. (3) [16]:

$$CDI\ (mg\ kg^{-1}\ bw\ d^{-1}) = \sum \frac{BaPeq \times IR \times EF \times ED}{BW \times AT} \quad (3)$$

where: BaPeq = Concentration of total PAH or PAH4 (mg BaPeq $kg^{-1}$) for each CPO sample; IR (kg $day^{-1}$) is the average daily intake of CPO, 25 g based on 9.125 kg per capita consumption rate [41]; EF is the exposure frequency in days (350 days $year^{-1}$); ED is the exposure duration (year), exposure estimations were performed in two age groups: children (1-15 years old, ED = 14) and adults (>16 years old, ED = 39.7); BW is the average body weight, for Nigerians: 21.1 kg for children and 63 kg for adults [42]; and AT is the average exposure time (equal to lifespan for carcinogens); an average life expectancy of 54.7 years for a Nigerian [43]. Four PAHs (PAH4) are defined as the sum of BaP, Chr, BaA, and BbF.

### 2.11 Risk Characterization

The risk characterization combines data from hazard and exposure assessments. The parameters that were utilized to quantitatively evaluate the risk associated with consuming CPO samples contaminated with PAHs were margin of exposure (MOE), lifetime cancer risk (LCR), and screening value (ScV).

### 2.12 Margin of Exposure

The MOE is a useful and acceptable tool for health risk characterization for a given population exposed to genotoxic and carcinogenic substances such as BaP and PAH4 (sum of BaA, Chr, BaP and BbF) and is recommended by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) and the European Food Safety Agency (EFSA) [44, 45, 46]. It is the ratio between the benchmark dose lower confidence limit ($BMDL_{10}$) determined in experimental animals. The MOE is computed by Eqn. (4) [22]:

$$MOE = \frac{BMDL_{10}}{CDI} \quad (4)$$

where MOE = Margin of Exposure, $BMDL_{10}$ = Benchmark dose lower confidence limit (mg kg-bw$^{-1}$ day$^{-1}$) and CDI = chronic daily intakes (mg kg-bw$^{-1}$ day$^{-1}$) due to BaPeq $_{PAH4}$.

The $BMDL_{10}$ value was set by the dose-response analysis for tumour type. $BMDL_{10}$ for BaP and the sum of 4 PAHs ranged from 0.07 to 0.20 mg kg-bw$^{-1}$ day$^{-1}$ and from 0.34 to 0.93 mg kg-bw$^{-1}$ day$^{-1}$ based on total tumour-bearing animals, respectively [22]. The EFSA CONTAM Panel in 2008 chose the lowest values of 0.07 and 0.34 mg kg-bw$^{-1}$ day$^{-1}$ for BaP and PAH4, respectively. In this study, BaP was not detected in any of the CPO samples, so the CDI used in the calculation of MOE is the one computed taking into account BaPeq due to PAH4 only. The MOE values $<10^4$ are interpreted as possible concern to human health, $10^4$-$10^5$ as low concern to human health, $>10^5$ as negligible concern as long as actions are taken to minimize further exposure, while $>10^6$ values are translated as negligible concern [47].

### 2.13 Cancer Risk

This is the lifetime likelihood that an individual will get cancer due to exposure to a certain pollutant or combination of contaminants in the environment. According to USEPA [48], cancer risk (CR) is computed by Eqn. (5):

$$CR = CDI_{PAHs} \times CSF \quad (5)$$

where $CDI_{PAHs}$ is the total CDI due to BaPeq $_{PAHs}$ of 9 PAHs computed by Eq. (3) and CSF is the cancer slope factor of BaP (geometric mean, 7.3 mg$^{-1}$ kg-bw day [48].

If CR < 1 x $10^{-6}$ (<1 chance in a million population of the risk of developing cancer), then the level of risk is considered acceptable and without negative consequences; values in the interval 1 x $10^{-6}$ – 1 x $10^{-4}$ (1-100 chances in a million population of the risk of developing cancer) denote the potential risk but acceptable, and values greater than 1 x $10^{-4}$ (>1 chance in 10,000 population of the risk of developing cancer) correspond to serious risk and unacceptable [14]

### 2.14 Screening value

In order to determine the carcinogenic effects that consuming the sampled CPO would pose to people, screening value (ScV) was analysed. It is described as the edible oil's chemical threshold concentration at which there may be a public health risk. The ScV for carcinogenic effect was computed using Eqn. (6) [16]:

$$\text{ScV (mg kg}^{-1}) = \frac{RL}{CSF} * \frac{BW}{IR} \quad (6)$$

where RL = maximum acceptable risk level (dimensionless) and was set to 1 x $10^{-1}$ [49], CSF = oral slope factor ($mg^{-1}$ kg day), BW = body weight (kg), IR = intake rate (kg $day^{-1}$).

### 2.5 Statistical analysis

Each parameter was determined in triplicate for each sample, and the average values for each state were expressed as mean ± SD. Kruskall-Wallis ANOVAS were used for multiple comparisons. The Beauvais-Pearson test was used to assess the degree of linear dependence amongst the various parameters assessed.

## 3. RESULTS AND DISCUSSION

### 3.1 Physicochemical Properties

The selected physicochemical properties of CPO samples such as specific gravity, acid value, free fatty acids, saponification value, and peroxide value of CPO samples analysed are presented in Table 2, reported in terms of mean plus standard deviation (n=3).

The mean SG value determined in this study (0.931) is higher than the permissible limits of Codex and NAFDAC/SON. Higher value of SG indicates the solid content (impurities) in the CPO samples [31].

Acid value (AV) is an important indicator of CPO quality. Av is the number of milligrams of KOH required to neutralize the free fatty acids present in one gram of CPO sample. Low AVs of oil indicate the resistance to rancidity and reveal the resistance of oil to gum formation and corrosion and CPO can be stored for a longer time. Free fatty acid (%FFA) content is a conventional expression of the percentage of mass fraction of total oil. It is expressed as the percentage by weight (wt%) of palmitic acid in oil. It is, therefore, one of the most important oil quality parameters and a good indicator of impairment of oil quality or the level of deterioration caused by hydrolytic rancidity or enzymes and edibility of the oil [50]. The fatty acids obtained ranged from 1.10 - 3.54 (wt% palmitic acid), were below the 5% standard requirement [51, 52, 53], and and therefore, are edible. These values are indicative of the freshness of the palm oil samples. The results were comparable with the previous work of Madubuike [27]. High FFAs content in CPO is an unfavourable condition as it results in rancidity which adversely affects the quality and may pose health risks including cancer and inflammation because of the formation of toxic and reactive oxidation products [54]. High FFAs need to be reduced to below 5% palmitic acid for edible oils usage.

The saponification value (SV) is employed as a quality control index. Sv SV represents the amount of potassium hydroxide (KOH) (in mg) required to saponify one gram of oil under the

conditions specified. It is an indication of the average molecular weight (or chain length) of triglycerides (fatty acid) of oil. In other words, SVs provide useful information as to the quantity, type of glycerides, and the average molecular weight of all fatty acids which are present in a given oil sample [55]. High saponification value indicates high proportion of low fatty acids since SV is inversely proportional to the average molecular weight or chain length of triglycerides [56]. The result (Table 2) shows that the mean SVs of oil from the producing states (expect CRS) are below the minimum of 195 mg KOH $g^{-1}$ oil of NAFDAC [51] and SON [52] and 190 mg KOH $g^{-1}$ oil of Codex Alimentarius [53] thus did not meet the standard. The mean SV (207.32 mg KOH $g^{-1}$) of CPO samples from CRS was significantly ($p<0.05$) higher than that of CPOs from all the other producing states in the following trend: DES > BYS > AKS > EDS > RVS (Table 2). The low saponification values recorded in comparison with the NAFDAC [51] and SON [52] and Codex Alimentarius [53] might be due to high level of impurities as indicated [57]. The combined mean SV of samples CPO (177.28 mg KOH $g^{-1}$) was found to be lower than a previous study [31]. Their low SVs, except samples from CRS (207.32 mg KOH $g^{-1}$), are indication that the CPOs do not have potential to be used as raw materials for soap and other cosmetic industry.

Ester value (EV) is the number of milligrams of potassium hydroxide (KOH) required to hydrolyze the esters present in one gram of oil sample. It is the difference between saponification value and acid value. Higher EV indicates the presence of high amount of esters and low molecular weight fatty acid content.

Peroxide value (PV) is a measure of oxygen chemically bound to an oil or fat as peroxides, particularly hydroperoxides, and expressed as milliequivalent of peroxide per kilogram ($meqO_2$ $Kg^{-1}$). The PV of oil is a useful index to assess the extent to which rancidity is produced due to autoxidation reactions occurred during storage of the oil, i.e., deterioration or spoilage (undesirable odours and flavours). It is used as an indication of the quality, stability, and age of edible fat and oil [50, 58].

High peroxide values could result from a high degree of unsaturation and found to increase with the storage time, temperature, light, and contact with atmospheric oxygen [54, 59]. The lower peroxide numbers, the better and/or fresher the oil. In general, the peroxide value increased with storage time. The PV values for all CPO samples ranged from 9.74 - 17.41 meq/kg (Table 2). The mean values of PV of CPO samples from EDS and RVS were within the range of Codex Alimentarius and NAFDAC/SON, the rest were higher and showed relatively susceptibility to oxidative rancidity.

The peroxide value of CPOs from this work was higher than that of Ndife et al. [60] and Raji et al. [61]. High peroxide values result in more oxidative products and free radicals which could have health implications which include cancer, heart attack, ageing, oxidative stress, and even death [25, 62].

### 3.2 Polycyclic Aromatic Hydrocarbon Concentration

Thirty-six samples of locally produced crude palm oil were collected from six states of the South-South zone, Nigeria, quantified for exposure assessment of the 16 USEPA priority PAHs.

Table 3 and Figure 2 provide a statistical summary of the mean concentrations, total PAHs of detected PAHs, and distribution pattern of the PAH congeners including PAH4 in the CPO samples. Among the 16 priority PAHs analysed, only nine PAHs (Nap, Acy, Ace, Flu, Phe, Ant, Flt, Pyr and Chr) were detected in most of the sample (Table 3). The remaining PAHs (BaA, BbF, BkF, BaP, DAh, BgP, IdP) - all 5-ring and 6-ring PAHs- were not detected in all the CPO samples. Among the nine detected PAH congeners, not more than eight congeners were detected in any one sample; CPO samples from AKS contain the highest number (8) while those from DES contain the least (4), in decreasing order of AKS (8) >CRS (7) > BYS (6) = EDS = RVS > DES (4).

It was observed that the mean concentrations of PAH congeners and the mean of total concentrations of PAHs (∑PAH) in this study varied significantly depending on sampling state. The mean concentrations of the PAH congeners ranged from 1.6 mg $kg^{-1}$ (Phe) to 6.90 mg $kg^{-1}$ (Acy) while mean ∑PAH ranged from 12.9 to 38.93 mg $kg^{-1}$ with a mean value of 27.52 mg $kg^{-1}$.

Table 2: Physicochemical properties of crude palm oil from South-South zone, Nigeria and Regulatory limits

| Sample States | | SG | SV | AV | FFA | EV | PV |
|---|---|---|---|---|---|---|---|
| AKS | Mean±SD | 0.939±.004 | 167.69±.52 | 5.72±0.49 | 2.87±0.25 | 161.97±.99 | 16.32±1.25 |
| | Range | 0.926-0.933 | 167.32-168.05 | 5.37-6.06 | 2.70-3.05 | 161.27-162.67 | 15.44-17.2 |
| BYS | Mean±SD | 0.936±.004 | 186.77±12.75 | 3.77±1.22 | 1.89±0.62 | 182.98±11.52 | 16.42±.84 |
| | Range | 0.932-0.94 | 177.75-195.78 | 2.91-4.64 | 1.46-2.33 | 174.84-191.13 | 15.82-16.42 |
| CRS | Mean±SD | 0.932±.002 | 207.32±.92.89 | 5.69±2.93 | 2.87±1.48 | 191.62±75.82 | 17.11±1.97 |
| | Range | 0.930-0.933 | 273-141.64 | 3.62-7.77 | 1.82-3.908 | 138.01-245.23 | 16.04-18.48 |
| DES | Mean±SD | 0.932±0.002 | 189.09±7.98 | 5.06±3.14 | 2.55±1.58 | 210.7±48.83 | 17.41±1.94 |
| | Range | 0.93-0.933 | 183.45-194.09 | 2.84-7.28 | 1.43-3.66 | 176.17-245.23 | 16.64-18.78 |
| EDS | Mean±SD | 0.922±0.002 | 164.55±28.47 | 4.54±2.99 | 2.28±1.51 | 160.01±31.47 | 16.26±1.97 |
| | Range | 0.920-0.923 | 144.42-184.68 | 2.42-6.66 | 1.22-3.35 | 137.76-182.26 | 14.29-18.23 |
| RVS | Mean±SD | 0.935±0.005 | 148.19±4.96 | 5.25±0.51 | 2.64±0.26 | 142.94±4.455 | 12.09±3.33 |
| | Range | 0.93-0.94 | 144.69-151.7 | 4.89-5.61 | 2.46-2.82 | 139.79-146.09 | 9.74-14.45 |
| Codex Alimentarius (2019) | | 0.896-0.910 | 189-199 | 10% | 5% | 189.4-208.4 | $\leq$ 15 |
| NAFDAC (2019) | | 0.898-0.907 | 195-205 | 10% | 5% | - | - |
| SON (2000) | | 0.897-0.907 | 195-205 | 3.5 | 3–5% | - | 10 |

SG = Specific gravity, SV= Saponification Value (mg KOH/g), AV = Acid value mg KOH/g, FFA = Free fatty acid (wt% palmitic acid), EV= Ester Value (mg KOH/g), PV= Peroxide value ($meqO_2/Kg$)

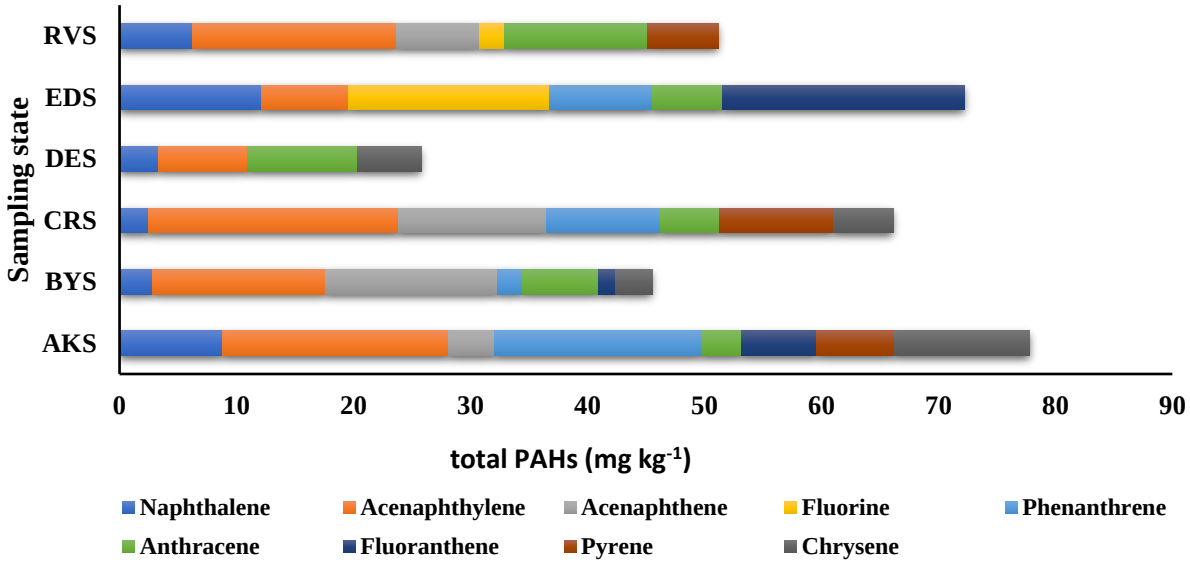


Figure 2: The total PAHs and distribution pattern in the CPO samples from producing states of South-South zone, Nigeria

The mean ∑PAHs values were 516-1557 times greater than the limit of 0.025 mg $kg^{-1}$ prescribed by the German Society for Fat Science for total PAHs [23, 24]. The levels of PAHs in the CPO samples were higher than levels reported by Eze et al. [63]. Such a high PAH content suggests that producers using local methods will make greater efforts to improve processing practices to reduce the possibility of PAH contamination in palm oil.

The 3-ring PAHs contributions of the ∑PAHs investigated in this study were in the following decreasing trend: 86% (in BYS) > 79% (in RVS) > 71.53% (in CRS) > 66% (in DES) > 57% (in AKS) > 55% (in EDS). The 4-ring PAHs contributions of the ∑PAHs were in the following increasing order: 7% (in BYS) < 8% (in RVS) < 21% (in DES) < 24% (in CRS) < 28% (in EDS) < 32% (in AKS). The 3-ring PAHs were the most abundant PAHs in the samples and its mean concentration varied from 8.49 to 21.17 mg $kg^{-1}$, which constitutes approximately 55% (EDS) - 86% (in BYS) of the ∑PAHs. The 2-ring PAH (i.e. Nap) was maximum in EDS (17% of ∑PAHs). The compositional patterns of the PAHs in the oil samples followed the decreasing order: 3-ring > 4-ring > 2-ring.

The samples have 33.33% of PAH4 and this constituted 7.7% of sample ∑PAHs. The mean PAH4 (BaA, Chr, BaP, and BbF) concentration values, mainly Chr, of the samples were many orders of magnitude (155-579 times) higher than the EU maximum permissible limit of 0.01 mg $kg^{-1}$ [25].

The LMW-PAHs (2- and 3-ring) accounted for about 93% of ∑PAHs in BYS, ~ 92% in RVS, ~ 79% in DES, ~ 76% in CRS, 71% in EDS and 68% in AKS while the HMW-PAHs (4-ring) ~ 32% in AKS, ~ 29% in EDS, ~ 24% in CRS, ~ 21% in DES, ~ 8% in RVS. The percentage of the LMW-PAHs ranged from 68- 93% with a mean of ~ 80%. Polycyclic aromatic hydrocarbons of petrogenic origin tend to be of lower molecular weight (2-3 rings) [64].

### 3.3 Sources of PAHs in the crude palm oil

An analysis of the identification of possible sources of PAHs in food products is essential for risk evaluation and developing strategies for minimizing the concentrations of these compounds in food materials. In this study, the computed values for different PAH molecular diagnostic ratios to detect the potential sources of PAHs in these CPOs are shown in Table 4. The Ant/Phe values in these CPOs ranged from 0.193 - 2.991, suggesting that the PAHs are of pyrogenic sources may be the most impactful in the CPO samples examined. The values of the Ant/(Ant + Phe) obtained ranged from 0.161 - 1.00, indicating PAHs of biomass combustion origin. The Flt/(Flt + Pyr) values in these oil samples ranged from 0.493 -1.00, suggesting a possible mixture of petrogenic and biomass combustion origins. The LMWPAHs/HMWPAHs values in the samples of CPO ranged from 0.739 – 3.547 suggesting that contributors to the PAH load were from petrogenic sources. The ratio of ∑CombPAHs/∑PAHs varied from 0.074 to 0.317 which are indicative of pyrogenic origin. Thus, the PAHs in the CPO samples from the study area can be attributed to a combination of mainly pyrogenic (fuel and biomass combustion) origin of PAHs with a small association from petrogenic origin. These PAH sources resulting from anthropogenic activities of petrogenic and pyrogenic origin include illegal artisanal petroleum refining/bunkering activities, gas flaring, indiscriminate burning of petroleum products, and unwholesome farming practices, accidental and intentional (vandalisation) spills of petroleum, improper disposal of used engine oils, and discharges from generator houses.

### 3.4 Exposure assessment

As already mentioned, it is customary to ascertain the total carcinogenicity of all PAHs found in palm oil in terms of the BaPeq and to assess exposure in the form of chronic daily intake (CDI). These parameters were computed in two ways: using strictly PAH4 contributors ($BaPeq_{PAH4}$ and $CDI_{PAH4}$), as well as using the detected 9PAH contributors ($BaPeq_{PAHs}$ and $CDI_{PAHs}$) and presented in Table 5.

### 3.5 Risk characterization

There is no benchmark dose of lower confidence limit ($BMDL_{10}$) health-based guidance available in the literature that encompasses all 16 PAHs. Therefore, from the dietary exposures ($CDI_{PAH4}$ and $CDI_{PAHs}$) the risks due to dietary exposure to PAH4 and PAH16 were characterized by calculating the MOE and LCR, respectively. The results obtained are presented in Table 5.

In this study, the MOEs attributed to oil ingestion varied between $1.06 \times 10^4$ - $3.97 \times 10^4$ in adults and $1.01 \times 10^4$ - $3.77 \times 10^4$ in children. The MOEs for PAH4 are greater than $1 \times 10^4$, indicating low health concern. The CR values obtained from the consumption of the CPO samples ranged between $2.48 \times 10^{-4}$ - $4.43 \times 10^{-4}$ and $2.36 \times 10^{-4}$ - $4.21 \times 10^{-4}$ for children and adults, respectively (Table 5). These CR values were above the unacceptable value of $1 \times 10^{-4}$ (one case in 10,000 population with the risk of developing cancer or cancer-related illnesses) [14].

Table 3: Mean concentration (mg $kg^{-1}$) (n=3) of priority PAHs of CPO samples

| | Rings | AKS | | BYS | | CRS | | DES | | EDS | | RVS | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Range | Mean±SD | Range | Mean±SD | Range | Mean±SD | Range | Mean±SD | Range | Mean±SD | Range | Mean±SD |
| Nap | 2 | ND-8.79 | 4.40±6.22 | ND-2.83 | 1.42±2.00 | ND-2.46 | 1.23±1.74 | ND-3.34 | 1.67±2.36 | ND-12.09 | 6.05±8.55 | 2.84-3.38 | 3.11±0.38 |
| Acy | 3 | ND-19.36 | 9.68±13.69 | ND-14.81 | 7.41±10.47 | 1.26-20.13 | 8.04±9.58 | ND-7.61 | 3.81±5.38 | ND-7.5 | 3.755±5.31 | 7.72-9.75 | 8.73±1.43 |
| Ace | 3 | ND-3.92 | 1.96±2.77 | 5.65-9.01 | 7.33±2.38 | 3.53-9.1 | 6.32±3.94 | ND | ND | ND | ND | ND-7.11 | 3.56±5.03 |
| Flu | 3 | ND | ND | ND | ND | ND | ND | ND | ND | ND-17.18 | 8.59±12.15 | ND-2.09 | 1.05±1.48 |
| Phe | 3 | ND-17.66 | 8.83±12.49 | ND-2.15 | 1.08±1.52 | ND-9.79 | 4.89±6.92 | ND | ND | ND-8.76 | 4.38±6.194 | ND | ND |
| Ant | 3 | ND-3.4 | 1.7±2.40 | ND-6.46 | 3.23±4.57 | ND-5.01 | 2.51±3.54 | 3.48-5.88 | 4.68±1.70 | ND-6.05 | 3.03±4.28 | 3.61-8.66 | 6.18±3.64 |
| Flt | 4 | ND-6.47 | 3.24±4.58 | ND-1.51 | 0.08±1.07 | ND | ND | ND | ND | 3.91-16.74 | 10.33±9.07 | ND | ND |
| Pyr | 4 | ND-6.65 | 3.33±4.70 | ND | ND | 3.91-5.81 | 4.86±1.34 | ND | ND | ND | ND | 2.85-3.17 | 2.01±0.23 |
| Chr | 4 | ND-11.58 | 5.79±8.19 | ND-3.1 | 1.55±2.19 | ND-5.15 | 2.57±3.64 | ND-5.47 | 2.74±3.87 | ND | ND | ND | ND |
| ∑PAHs | | ND-66.15 | - | ND-45.52 | - | ND-66.15 | - | ND-25.78 | - | ND-72.23 | - | ND-51.18 | - |
| ∑PAH4 | | ND-11.58 | - | ND-3.1 | - | ND-5.15 | - | ND-5.47 | - | - | - | - | - |

ND – Not detected

The results of the cancer risk evaluation indicated that there are 248 - 443 chances in a million population of developing cancer. Therefore, adverse health effects due to PAHs may arise from the lifetime consumption of CPO samples from South-South Nigeria. The CPO samples from BYS and AKS had the highest and lowest MOEs, respectively, associated with their consumption. Conversely, the highest and lowest associated CR on consumption of CPO samples were from AKS and BYS. This is in line with the observation of Yousefi et al. [65].

Screening value (ScV) was assessed in this study to determine the health effects on consumers of PAH-contaminated CPO. The computed $BaPeq_{PAHs}$ values ranged from 0.117 to 0.209 mg $kg^{-1}$ and significantly ($p < 0.05$) higher than the ScV 0.003 mg $kg^{-1}$ for CPO (Table 4). This may suggest a potential adverse health effects for consumers of CPO locally produced in South-South zone, Nigeria.

Table 4: Molecular diagnostic ratios for PAHs in study area

| Sampled State | Ant/Phe | Ant/(Ant+Phe) | Flt/(Flt+Pyr) | LMW/HMW | Comb PAH/PAH |
|---|---|---|---|---|---|
| AKS | 0.193 | 0.161 | 0.493 | 1.769 | 0.317 |
| BYS | 2.991 | 0.749 | 1.000 | 3.547 | 0.074 |
| CRS | 0.513 | 0.339 | - | 2.060 | 0.244 |
| DES | - | 1.000 | - | 0.739 | 0.212 |
| EDS | 0.692 | 0.409 | 1.000 | 1.705 | 0.286 |
| RVS | - | 1.000 | - | 2.009 | 0.082 |

Table 5: Total BaP equivalent (BaPeq), chronic daily intake (CDI), margin of exposure (MOE), and lifetime cancer risk (LCR) values for CPO samples

| Sampled state | BaPeq $_{PAH4}$ (mg kg$^{-1}$) | BaPeq $_{PAHs}$ (mg kg$^{-1}$) | CDI$_{PAH4}$ (mg kg$^{-1}$ day$^{-1}$) | CDI$_{PAHs}$ (mg kg$^{-1}$ day$^{-1}$) | MOE$_{PAH4}$ | LCR$_{PAHs}$ |
|---|---|---|---|---|---|---|
| **Child of average body weight of 21.1 kg** | | | | | | |
| AKS | 0.116 | 0.209 | 3.37E-05 | 6.07E-05 | 1.01E+04 | 4.43E-04 |
| BYS | 0.031 | 0.117 | 9.01E-06 | 3.40E-05 | 3.77E+04 | 2.48E-04 |
| CRS | 0.052 | 0.145 | 1.50E-05 | 4.22E-05 | 2.27E+04 | 3.08E-04 |
| DES | 0.055 | 0.159 | 1.59E-05 | 4.63E-05 | 2.14E+04 | 3.38E-04 |
| EDS | - | 0.144 | - | 4.18E-05 | - | 3.05E-04 |
| RVS | - | 0.157 | - | 4.55E-05 | - | 3.32E-04 |
| ScV | | 3.45E-03 | | | | |
| **Adult of average body weight of 63 kg** | | | | | | |
| AKS | 0.116 | 0.209 | 3.20E-05 | 5.76E-05 | 1.06E+04 | 4.21E-04 |
| BYS | 0.031 | 0.117 | 8.56E-06 | 3.23E-05 | 3.97E+04 | 2.36E-04 |
| CRS | 0.052 | 0.145 | 1.42E-05 | 4.00E-05 | 2.39E+04 | 2.92E-04 |
| DES | 0.055 | 0.159 | 1.51E-05 | 4.40E-05 | 2.25E+04 | 3.21E-04 |
| EDS | - | 0.144 | - | 3.97E-05 | - | 2.90E-04 |
| RVS | - | 0.157 | - | 4.32E-05 | - | 3.16E-04 |
| ScV | | 3.45E-03 | | | | |

ScV - Screening, BaPeq $_{PAH4}$ – Total BaP equivalent of PAH4, BaPqe $_{PAHs}$ - BaP equivalent of detected 9 PAHs, CDI$_{PAH4}$ - chronic daily intake of PAH4, CDI$_{PAHs}$ - chronic daily intake of detected 9PAHs

## 4. CONCLUSION

Health risks due to consumption of PAH-contaminated vegetable oils are a major concern requiring regular monitoring to detect changes as recommended by regulatory agencies. This study investigated the quality of edible palm oil from the six states in the South-South zone of Nigeria by assessing important parameters such as specific gravity, free fatty acid content, acid value, saponification value, ester value, and peroxide value. Additionally, PAHs in the oil

samples were quantified and related risks due to consumption were characterized by margin of exposure, lifetime cancer risk, and screening value. The results indicated that the CPO samples were of low quality as the parameters were below the requirements of NAFDAC/SON and Codex Alimentarius. The CPO samples are adjudged to be highly contaminated with PAHs as 100% of CPO samples examined exceeded the EU and the German Society for Fat Science permissible limits. Molecular diagnostic ratio for PAH source evaluation suggested a mixture of petrogenic and pyrogenic origins. The margins of exposure for both children and adults were at the borderline of possible health concern. The lifetime cancer risk values for PAHs for children and adults were unacceptable, above the regulatory limit of USEPA while the $BaPeq_{PAHs}$ (carcinogenic toxic quotients) for both children and adults were higher than screening value. The study therefore, concluded that consumers of CPO locally produced in South-South zone, Nigeria, could have serious health risk concerns and regular monitoring is required to detect specifications.

## Data Availability

All relevant data are within the paper

## REFERENCES


1. Arunachalam V. Oil Palm, In: Editor(s): Arunachalam V. Genomics of Cultivated Palms, Elsevier, 2012. p 29-48.
2. Goh KJ, Wong CK, Ng PHC. Oil Palm. In: Thomas B, Murray BG, Murphy DJ, editor(s). *Encyclopedia of Applied Plant Sciences* (2nd edition): Academic Press, 2017. p 382-390.
3. Nizam AFA, Mahmud MS. Food quality assurance of crude palm oil: a review on toxic ester feedstock. *Oilseeds & fats Crops and Lipids*, 2021 Mar;28(23):1-14.
4. Bahadi MA, Japir A, Salih N, Salimon J. Free fatty acids separation from Malaysian high free fatty acid crude palm oil using molecular distillation. *Malaysian J. Anal. Sci.* 2016 May; 20(5):1042-1051.
5. Tarmizi AHA, Siew WL, Kuntom A. Production of palm oil reference materials for the determination of solid fat content. *J. Food Qual.* 2008 Dec;31(6):673-685.
6. Ogunsola AOR, Oluwusi GO. Methods of palm oil processing in Ogun state, Nigeria: a resource use efficiency assessment. *Am. Int. J. Contemp. Res.* 2014 Aug;4:173-179.
7. Gonzalez-Diaz A, Pataquiva-Mateus A, García-Núñez JA. Recovery of palm phytonutrients as a potential market for the by-products generated by palm oil mills and refineries–A review. *Food Biosci.* 2021 June;41:100916.
8. Imoisi OB, Ilori GE, Agho I, Ekhator JO. Palm oil, its nutritional and health implications (Review). *J. Appl. Sci. Environ. Manage.* 2015 Mar;19(1):127-133.
9. Olafisoye OB, Fatoki OS, Oguntibeju OO, Osibote OA. Accumulation and risk assessment of metals in palm oil cultivated on contaminated oil palm plantation soils. *Toxicol. Rep.* 2020 Jan;7:324-334.

10. Odia OJ, Ofori S, Maduka O. Palm oil and the heart: A review. *World J. Cardiol.* 2015 Mar;7(3):144-149.
11. Lv C, Wang Y, Zhou C, Ma W, Yang Y, Xiao R, Yu H. Effects of dietary palm olein on the cardiovascular risk factors in healthy young adults. *J. Food Nutr. Res.* 2018 July;62.
12. Absalome MA, Massara CC, Alexandre AA, Koffi G, Chantal GGA, Ferdinand D, Rhedoor AJ, Coulibaly I, George TG, Brigitte T, Marion M, Jean-Paul C. Biochemical properties, nutritional values, health benefits and sustainability of palm oil. *Biochimie*, 2020 Nov;178:81-95.
13. Moslen M, Miebaka CA, Boisa N. Bioaccumulation of Polycyclic Aromatic Hydrocarbon (PAH) in a bivalve (*Arca senilis*- blood cockles) and health risk assessment. *Toxicol. Rep.* 2019 Sept;6:990-997.
14. Onyegeme-Okerenta BM, West OL. Toxicological Evaluation of Contamination by Potentially Toxic Elements (PTEs) and Related Risks in the Surface Waters of Three Tidal Streams of the Niger Delta, Nigeria. *Asian J. Environ. Ecol.* 2023 May;20(4):67-83.
15. Sanchez-Arevalo CM, Olmo-Garcia L, Fernandez-Sanchez JF, Carrasco-Pancorbo A. Polycyclic aromatic hydrocarbons in edible oils: An overview on sample preparation, determination strategies, and relative abundance of prevalent compounds. *Compr. Rev. Food Sci. Food Saf.* 2020 Oct;19:3528-3573.
16. Onyegeme-Okerenta BM, West OL, Chuku LC. Concentration, Dietary Exposure and Human Health Risk Assessment of Total Petroleum and Polycyclic Aromatic Hydrocarbons in Seafood from Coastal Communities in Rivers State, Nigeria. *Sci. Afr.* 2022 July;16:e01186.
17. Mancini A, Imperlini E, Nigro E, Montagnese C, Daniele A, Orrù S, Buono P. Biological and Nutritional Properties of Palm Oil and Palmitic Acid: Effects on Health. *Molecules* 2015 Sept;20(9):17339-61.
18. Davis E, Walker TR, Adams M, Willis R, Norris GA, Henry RC. Source apportionment of polycyclic aromatic hydrocarbons (PAHs) in small craft harbor (SCH) surficial sediments in Nova Scotia, Canada. *Sci. Total Environ.* 2019 Nov;691:528-537.
19. Malarkey DE, Hoenerhoff MJ, Maronpot RR. Carcinogenesis. In *Fundamentals of Toxicologic Pathology*, 3rd ed.; Wallig MA, Haschek WM, Rousseaux CG, Bolon B. Eds.; Academic Press, 2018. p 83–104.
20. Famiyeh L, Chen K, Xu J, Sun Y, Guo Q, Wang C, Lv J, Tang YT, Yu H, Snape C, He J. A review on analysis methods, source identification, and cancer risk evaluation of atmospheric polycyclic aromatic hydrocarbons. *Sci. Total Environ.* 2021 Oct;789:147741.
21. Liu Q, Wu P, Zhou P, Luo P. Levels and Health Risk Assessment of Polycyclic Aromatic Hydrocarbons in Vegetable Oils and Frying Oils by Using the Margin of Exposure (MOE) and the Incremental Lifetime Cancer Risk (ILCR) Approach in China. *Foods*, 2023 Aug; 12(4):811.
22. EFSA. European Food Safety Authority. Polycyclic Aromatic Hydrocarbons in food: Scientific opinion of the panel on contaminants in the food chain. *The EFSA Journal*, 2008 Aug;724:1–114.

23. Teixeira VH, Casal S, Oliveira MBPP. PAHs Content in Sunflower, Soybean and Virgin Olive Oils: Evaluation in Commercial Samples and During Refining Process. *Food Chem.* 2007 Jan; 104(1):106–112.
24. Junmin J, Miaomiao J, Yaxin Z, Jie H, Shangde S. Polycyclic Aromatic Hydrocarbons Contamination in Edible Oils: A Review. *Food Rev. Int.* 2023 Oct;39:9:6977-7003
25. EC. Commission Regulation (EU) 2023/915 of 25 April 2023 on maximum levels for certain contaminants in food and repealing Regulation (EC) No 1881/2006. Official Journal of the European Union L, 2023 April; 119/103:1–55.
26. Tagoe SMA, Dickinson MJ, Apetorgbor MM. Factors influencing quality of palm oil produced at the cottage industry level in Ghana. *Int. Food Res. J.* 2012 Jan; 19(1):271 – 278.
27. Madubuike PC, Ezigbo IJ, Ekpe IO, Onukwube SI. Evaluation of the Quality and Level of Adulteration of Palm Oil Obtained from Different Locations in Enugu Metropolis, Nigeria. *Int.J. Multidiscip. Sci. Engineer.* 2015 June;6(6):23-26.
28. Isiodu VC, Onyegeme-Okerenta, BM, Essien EB. Dietary exposure and risk characterization of selected toxic metals in crude palm oil (*Elaeis guineensis Jacq*) from six states in Niger Delta, Nigeria. *Magna Scientia Adv. Res. Rev.* 2024 Jan;10(01):025–037.
29. Wu S, Yu W. Liquid–liquid Extraction of Polycyclic Aromatic Hydrocarbons in Four Different Edible Oils from China. *Food Chem.* 2012 Sept; 134(1):597–601.
30. Oyibo JN, Wegwu MO, Uwakwe AA, Osuoha JO. Analysis of total petroleum hydrocarbons, polycyclic aromatic hydrocarbons and risk assessment of heavy metals in some selected finfishes at Forcados Terminal, Delta State, Nigeria. *Environ. Nanotechnol. Monit. Manag.* 2018 May;9:128-135.
31. Riris ID, Silalahi A. Assessment of the quality of crude palm oil from *Elaeis Guineensis* growing areas of the PT. Langkat Nusantara Kepong-PTPN II North Sumatera. *Agrium: Jurnal Ilmu Pertanian*, 2017 June; 20(3):185-189.
32. Chong CL. Measurement and maintenance of palm oil quality. In Palm oil. AOCS Press 2012. p. 431-470.
33. Nwakodo CS, Chukwu M, Iwuagwu MO, Odom TC. Effect of Processing Methods and Storage Time on Chemical Properties of Palm Oil. *Res. J. Food Sci. Nutr*. 2019 April; 4(2): 37-47.
34. ISO. (International Organization Standardization) ISO 3960:2017 Animal and vegetable fats and oils – Determination of peroxide value – Iodometric (visual) endpoint determination. 2017 Feb; BS EN ISO 3960:2017.
35. Zhang D, Wang JJ, Ni H G, Zeng H. Spatial-temporal and multi-media variations of polycyclic aromatic hydrocarbons in a highly urbanized river from South China. *Sci. Total Environ.* 2017 Mar.;581-582:621–628.
36. Khan R, Islam MS, Tareq ARM, Naher K, Islam ARMT, Habib MA, Siddique MAB, Islam MA, Das S, Rashid MB, Ullah AKMA, Miah MMH, Masrura SU, Bodrud-Doza M, Sarker MR, Badruzzaman ABM. Distribution, sources and ecological risk of trace

elements and polycyclic aromatic hydrocarbons in sediments from a polluted urban river in central Bangladesh. *Environ. Nanotechnol. Monit. Manag.* 2020 Dec.;14:100318.

37. Barhoumi B, Beldean-Galea MS, Al-Rawabdeh AM, Roba C, Martonos IM, Bălc R, Kahlaoui M, Touil S, Tedetti M, Driss MR, Baciu C. Occurrence, distribution and ecological risk of trace metals and organic pollutants in surface sediments from a Southeastern European river (Someşu Mic River, Romania). *Sci. Total Environ.* 2019 April 10th;660:660-676.
38. Effiong IA, Bassey FI, Iwegbue CMA, Ekpa OD, Williams SA, Oguntunde FC, Osabor VN, Martincigh B. Polycyclic aromatic hydrocarbons in three commercially available fish species from the Bonny and Cross River estuaries in the Niger Delta, Nigeria. *Environ. Monit. Assess*. 2016 August;188(9):1-17.
39. Liu W, Ma L, Abuduwaili J, Li Y. Distribution, source analysis, and ecological risk assessment of polycyclic aromatic hydrocarbons in the typical topsoil of the Issyk-Kul Lake Basin. *Environ. Monit. Assess.* 2017 July; 189(8):398.
40. ATSDR. Agency for Toxic and Disease Registry. Guidance for Calculating Benzo(a)pyrene Equivalents for Cancer Evaluations of Polycyclic Aromatic Hydrocarbons. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Services. 2022. https://www.atsdr.cdc.gov/pha-guidance/resources/ATSDR-PAH-Guidance-508.pdf
41. WHO. Global Environmental Monitoring System – Food Contamination Monitoring and Assessment Programme (GEMS/Food). GEMS/Food Consumption Cluster Diets. 2006. <http://www.who.int/foodsafety/chem/gems/en/index.html> (accessed May 18th 2023).
42. Akinpelu AO, Oyewole OO, Adekanla BA. Body Size Perceptions and Weight Status of Adults in a Nigerian Rural Community. *Ann. Med. Health Sci. Res.* 2015 Sept; 5(5):358–364.
43. United Nations Development Programme (UNDP). The 2020 Human Development Report. [Assessed November 28, 2023]. Available from: https://www.undp.org/nigeria/publications/2020-human-development-report
44. JECFA. Joint FAO/WHO Expert Committee on food additives (FAO/WHO) Summary and conclusions; Sixty-fourth meeting, Rome, 2005 Feb;8-17.
45. EFSA. European Food Safety Authority. Opinion of the scientific Committee on a request from EFSA related to A harmonised approach for risk assessment of substances which are both genotoxic and carcinogenic. *The EFSA Journal*, 2005 Oct.; 282:1-31.
46. Okoye EA, Bocca B, Ruggieri F, Ezejiofor AN, Nwaogazie IL, Domingo JL, Rovira J, Frazzoli C, Orisakwe OE. Concentrations of polycyclic aromatic hydrocarbons in samples of soil, feed and food collected in the Niger Delta region, Nigeria: A probabilistic human health risk assessment. *Environ. Res.* 2021 July;202:111619.
47. EFSA. European Food Safety Authority. Scientific Opinion on risk-based control of biogenic amine formation in fermented foods. *The EFSA Journal,* 2011 Oct;9(10):2393.
48. USEPA. Conducting a human health risk assessments. EPA's Report on the Environment (ROE), U.S. Environmental Protection Agency, Washington, D.C, 2008 Final Report) EPA/600/R-07/045F (NTIS PB2008-112484), 2008.

49. USEPA. (US Environmental Protection Agency) 3rd ed. Office of Water; Washington DC: 2000. Guidance for Assessing Chemical Contaminant. Data for Use in Fish Advisories. Fish Sampling and Analysis.2000 Nov; (EPA823-R-95-007)
50. Mengistie M, Alemu A, Mekonnen A. Comparison of physicochemical properties of edible vegetable oils commercially available in Bahir Dar, Ethiopia. *Chem. Intl*. 2018 April; 4(2):130-135
51. NAFDAC. National Agency for Food and Drug Administration and Control. Fats-Oils-and-Foods-Containing-Fats-and-Oils-Regulations-2022. Federal Republic of Nigeria Official Gazette. 2023 May 16th ;110:B521-543.
52. SON. Standard Organisation of Nigeria. Standards for edible refined palm oil and its processed form. 2000;2-5
53. Hishamuddin E, Sulaiman N, Ahmad Bustamam FK, Yeoh CB. Recent Updates on the Codex Standard for Named Vegetable Oils (CXS 210-1999) in Relation to Palm Oil and Palm Kernel Oil. Palm Oil Developments, 2020 Dec;72:34-41.
54. Negash YA, Amare DE, Bitew BD, Dagne H. Assessment of quality of edible vegetable oils accessed in Gondar City, Northwest Ethiopia. *BMC Res. Notes.* 2019 Dec 4th; 12:793.
55. Mohammed IHA, Ali TB. Physicochemical characteristics of some imported edible vegetable oils in Iraq. *Res. J. Pharm. Biol. Chem. Sci*. 2015 Jan; 6(5):488-494.
56. Ivanova M, Hanganu A, Dumitriu R, Tociu M, Ivanov G, Stavarache C, Popescu L, Ghendov-Mosanu A, Sturza R, Deleanu C, Chira A. Saponification Value of Fats and Oils as Determined from 1H-NMR Data: The Case of Dairy Fats. Foods. 2022 May;11(10).
57. Odoom W, Edusei VO. Evaluation of Saponification value, Iodine value and Insoluble impurities in Coconut Oils from Jomoro District of the Western Region of Ghana. *Asian J Agric Food Sci*. 2015 Oct.;3(5).
58. Nangbes JG, Nvau JB, Buba WM, Zukdimma AN. Extraction and characterization of castor (Ricinus Communis) seed oil. *Int. J. Eng. Sci*.2013 July;.2(9):105-109.
59. Othman OC, Ngassapa FN. Physicochemical characteristics of some imported edible vegetable oils and fat marketed in Dar es salaam, Tanzania. *Tanzania J. Nat. Appl. Sci.* 2010 Jan.;1(2):138-147.
60. Ndife J, Ogbonna J, Okwunodulu IN, Ugwuona FU. Comparative quality evaluation of selected traditional processed palm oils (*Elaeis guineensis Jacq*) from South-east Nigeria. *Int. J. Agric. Environ. Bio-Res.* 2019 June;4(3):145-156.
61. Raji RO, Inengite AK, Godwin J, Ajibesin KK. Assessment of proximate and physicochemical properties of crude palm oil from south-west and South-South Nigeria. *Afr. J. Pure Appl. Chem.* 2022 July;16(2):40-56.
62. Adelagun ROA, Berezi EP, Fagbemi JO, Igbaro OJ, Aihkoje FE, Ngana O, Osondu G, Garba MS. Evaluation of level of rancidity of edible oil in some fried snacks food. *J. Chem. Soc. Nigeria*. 2023 feb. 28th;48(1):154-161.

63. Eze SO, Kalu NU, Eze CN. Polycyclic Aromatic Hydrocarbon (PAHs) contamination of palm oil samples from Ohafia Agricultural Zone of Abia State, Nigeria. *Eur J PA Sci*. 2015 Oct.; 3(3):1-14.
64. Mostert, M. M., Ayoko, G. A., Kokot, S. Application of chemometrics to analysis of soil pollutants. *Trends Anal. Chem.* 2010 May;*29*(5):430-445. https://doi.org/10.1016/j.trac.2010.02.009.
65. Yousefi M, Shemshadi G, Khorshidian N, Ghasemzadeh-Mohammadi V, Fakhri Y, Hosseini H, Khaneghah AM. Polycyclic aromatic hydrocarbons (PAHs) content of edible vegetable oils in Iran: A risk assessment study. *Food Chem Toxicol.* 2018 May;118:480–489.