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Full Length Research Paper

Physicochemical properties and fatty acid composition of freshly prepared palm oil

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Elaeis guineensis **commonly known as the African palm oil, a monocotyledon widely used due to its nutritional and economic importance. The effect of deterioration of the harvested palm fruit on the physicochemical properties and fatty acid composition of palm oil was investigated. The palm fruit was processed after each period of deterioration (fresh, 7, 14 and 21 days after harvest) for oil production. AOAC Standard methods were used for the analysis. The result shows that the acid value (4.0±0.08, 4.94±0.33, 6.08±1.17 and 6.68±0.44) mg KOH/g, peroxide value (12.25±0.21, 14.53±0.78, 13.90±0.14, and 14.90±0.24) mg KOH/g, flash point (304.5±0.71, 300.5±2.12, 301.5±0.71 and 308.5±0.17°C) and boiling point (342±4.24, 339±1.41, 350±1.41 and 348±2.82°C) for fresh, 7, 14 and 21 days, respectively were above National Agency for Food and Drug Administration Control (NAFDAC) permissible limits. The free fatty acid value, saponification value, iodine value and melting point were below NAFDAC permissible limits. The fatty acid components showed that C_{12} for day 7 was significantly high when compared with other groups. Also linolenic acid (18:3) composition was significantly different at day 21 when compared with the values for other days. The result reveals that some physiochemical properties of the oil (acid value, free fatty acid, refractive index, specific gravity, cloud point and boiling points) increased in days 14 and 21 oil. Thus, the quality of oil processed within seven days of harvest of the palm fruits was better.**

Key words: *Elaeis guineensis,* physicochemical, fatty acid composition, deterioration.

INTRODUCTION

Elaeis guineensis (oil palm) is a monocotyledon belonging to the genus *Elaeis.* The palm fruit has an outer edible layer known as the mesocarp, an inedible layer of shell and an edible kernel inside the shell (Naiyana, 2003; Edo et al., 2022). It is mostly found in Southeast Asia, especially Malaysia and Indonesia which constitutes about 85% of the total world production (Sime et al., 2014). It is native to many West African countries where local population has used its oil for culinary purposes (Naiyana, 2003) and for production of soaps, detergents, pharmaceutical products and cosmetics (Yeong et al., 2012). It is medicinally used in approximately 35 out of

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Africa's 48 countries and it is used in the treatment of infection and digestive system disorder (Gruca et al., 2014). Palm oil thrives well in many African countries including Nigeria, Ghana, Benin, Liberia, Angola and Congo. Rafiu et al. (2022) noted that crude oil contains the highest concentration of agriculturally derived carotenoids when compared with other vegetable oils. It is known to be among the richest plant source of vitamin A. Sumathi et al. (2008) researched on the utilization of oil palm as a source of renewable energy, revealed that huge quantities of *E. guineensis by*-products are developed to produce value added products such as methane gas, bio-plastics, organic acids, activated carbon and bio-compost. They added that the biodiesel obtained from *E. guineensis* is degradable, non-toxic and has significantly fewer emissions than petroleum-based diesel (petrol diesel) when burned. Palm oil is now the most widely used oil in the world.

Physicochemical properties are referred to as the physical and chemical properties of a substance and these include refractive index, acid value, saponification, specific gravity, free fatty acid, viscosity, peroxide value, iodine value, cloud point, flash point, melting point and boiling point. They are qualitative properties of palm oil and do not indicate the position of bonds or the amount of olefinic carbon, but rather it provides an overall status of unsaturation level of oil. These parameters are very useful in evaluating the quality of the palm oil (Tulan et al., 2009). Peroxide value is used as a measure of the extent to which rancidity (incomplete oxidation or hydrolysis of fat) has occurred during storage. It can be used as an indication of the quality and stability of fats and oil (Tulan et al., 2009; Mac-Arthur et al., 2021). It increases with storage time and temperature.

Due to the ever-increasing utilization and demand for palm oil for domestic and industrial purposes like production of paint, soap, pharmaceuticals for local and for export, coupled with a decline in the production of commonly used source of oil in Nigeria, palm oils are produced on large scale for immediate and future use (Edo et al., 2022). Since palm oil processing is a major source of income and employment to a wide area of population in Africa especially in Nigeria, it has been observed that the red colour of the harvested palm fruit begin to change to dark brown as the fruit deteriorated (Mac-Arthur et al., 2021). Also edible vegetable oils are prone to quality deterioration through oxidation and microbial degradation resulting in nutritional loss and off-flavors (Mac-Arthur et al., 2021). The quality of the oil depends on the deterioration degree of harvested fruit. The degree of deterioration contributes to the formation of oxidation products that are reactive and toxic, which ultimately pose health risks inflammation (Tulan et al., 2009; Ruswanto et al., 2020), thus the need to investigate the effect of deterioration of the harvested palm fruit on the physicochemical properties and fatty acid composition of freshly prepared palm oil.

MATERIALS AND METHODS

Sample collection

The palm fruit sample was collected from Elele Community in Ikwerre Local Government Area of Rivers State, Nigeria. The coordinates of Elele community is 5.6663°N and 6.4948°E. The plant was identified by Dr. Green of Plant Science and Biotechnology Department of Rivers State University, Port Harcourt.

Sample preparation

The harvested palm fruit was divided into four and labeled (1 [fresh], 7, 14 and 21) based on the number of days the fruits were allowed to stay before processing to obtain the oil. At the end of each period of deterioration, the fruits were boiled and pound traditionally. The pound fruit mash was then transferred to an open bowl and washed carefully with warm water and squeezed manually to obtain a red viscous fluid which was heated (temperature) for 30 min for traces of water to evaporate and finally sieved using metal basket to obtain a clear red palm oil. The processed palm oil was stored in an airtight plastic container at 25°C.

Method of data analysis

Determination of quality indices

The acid values, free fatty acid, iodine value, peroxide value, saponification value, refractive index, specific gravity, viscosity and free fatty acid profile were determined using the AOAC (2012) as summarized subsequently.

Determination of physical properties

Melting, cloud, flash, and boiling points were determined according to the method described by Pike (2003).

Determination of acid value

Twenty-five milliliters of diethyl ether was mixed with 25ml alcohol and 1 ml phenolphthalein (1%) and neutralized with 0.1 M NaOH. 10 g of oil was dissolved in the solvent and titrated with aqueous 0.1 M NaOH, shaken until pink color which persisted was obtained.

Acid Value = Titre volume (ml) × 5.61 / Of sample used

Determination of iodine value

Palm oil was poured in a beaker and weighed; the weight was calculated by dividing 20 by the highest expected iodine value. 10 ml of carbon tetrachloride was added to the oil and dissolved, 20 ml of Wiji's solution was further added and kept in the dark for 30 min. A 15 ml solution of potassium iodine was then added with water (100 ml) and mixed. The solution is then titrated (liberating the iodine) with 0.1 M sodium thiosulphate using starch as an indicator. A blank sample was also prepared with 10 ml of carbon tetrachloride, the sample result (V_1) and that of blank (V_2) was recorded.

Iodine Value = (v2 - v1) × 1.269 / Weight of the sample used If (V_2-V_1) is greater than, $V_1/_2$ will be repeated using a smaller amount of sample.

Determination of peroxide value (PV)

One gram of oil was weighed into a clean dry boiling tube, 1 g of powdered potassium iodide and 20 ml of solvent mixture (2 volume of glacial acetic acid + 1 volume of chloroform) was added. The resulting solution was then placed in a water bath for 30 s and allowed to boil vigorously; when this is done it was poured quickly into a flask containing 20 ml of potassium iodide solution (5%). The sample solution was then titrated with 0.002 N sodium thiosulphate solution using starch as an indicator.

$$PV = 2 \times V$$

where V= volume of sodium thiosulphate used, 2 = (N × 1000) / W, N=normality of sodium thiosulphate used, and W = weight of sample used.

Determination of saponification value

The sample (2 g) was weighed into a conical flask and 25 ml of alcoholic potassium hydroxide solution was added, the mixture was then heated in boiling water for 1 h.

Potassium hydroxide (KOH) was then titrated with 0.5 M HCl using phenolphthalein as an indicator. A blank sample was also prepared and back titrated accordingly. The sample and blank titers (V_1 and V_2) were recorded.

$$\text{Saponification value} = (v2 - v1) \times 28.05 / \text{Weight of sample}$$

Determination of specific gravity

Fifty milliliters pycnometer bottle (manufacturer, model) was washed thoroughly with detergent, water, petroleum ether and dried and weighed, the bottle was then filled with water and weighed. After that it was filled with the oil sample and weighed.

$$\text{Specific gravity} = \text{Weight of Xml} / \text{Weight of Xml water}$$

Determination of refractive index

Abbe refractometer was reset with a light compensator; the oil sample was then seared on the lower prism of the Abbe refractometer. Light was passed by means of bangle mirror which reflected in the form of a dark background. The telescope tubes were adjusted until the black shadow appeared central in the crosswire indicator; it is then read.

Determination of viscosity

The viscometer (manufacturer, model) was first calibrated using water as the sample at 26 to 28°C. For each sample 200 ml of the oil was measured into a 250 ml beaker and the rotating spindle of the viscometer was immersed in it. The viscosities of the samples were then read from the monitor of the viscometer.

Fatty acid profile

Five hundred milligrams of the sample was weighed in a 25 ml centrifuge; 2 ml of water was added and mixed to dissolve. It was then kept at room temperature for 15 min. 5 ml of internal standard (C11:0 FAME + C13:0 TAG) was added at 2 mg/ml in methyl tert-butyl ether and 5 ml of 5% (w/v) methanoic sodium methoxide solution was added also. The tube was then closed and vortexed for 10 s.

After 10 s (time starts when sodium methoxide is added), 2 ml of hexane was added and at 210 s 10 ml of neutralization solution (10% disodium hydrogen citrate / 15% sodium chloride in water). It was shaken gently using vortex mixer and centrifuged at 1,750 rpm for 5 min. 200 µl of supernatant was transferred into 10 ml flask and diluted to mark with hexane.

Smoke, flash and fire point

Ten milliliters volume of oil was poured into an evaporating dish, a thermometer was suspended at the center of the dish without it touching the bottom of the dish, and the temperature was gradually raised using a hot plate.

The temperature at which the sample gives off a thin blush smoke continuously is noted as the smoke point; similarly the temperature at which the oil started flashing without supporting combustion is noted as the flash point, the temperature at which it starts to support combustion is known as the fire point.

Fatty acids composition

This was determined with Gas Chromatography Flame Ionization Detector (GC-FID) (manufacturer, model) in accordance with AOAC method (2012). The conditions were: Column: SP™ -2560, 100 m × 0.25 mm I.D, 0.20 µm; oven: 60°C (1 min), 15°C/min to 165°C (1 min), 2°C/min to 225°C (20 min); inj. Temp.: 250°C; carrier gas: helium, 0.8 mL/min; detector: FID, °C; injection: 1 µL, 10:1 split; liner: 4 mm I.D., split/splitless type, wool packed single tapers FocusLiner™ design; sample: *cis/trans* FAME Column Performance Mix.

Statistical analysis

Data obtained were subjected to statistical analyses, mean, standard deviation and one-way analysis of variance to determine the significant difference among the days and compared at 95% confidence limit using SPSS program version 17.

RESULTS AND DISCUSSION

Table 1 shows the effect of days (day 1 [fresh], 7, 14 and 21) of palm fruit deterioration on the physicochemical properties of freshly prepared palm oil. The result shows that the acid value, peroxide value, flash point and boiling point of the sample were above National Agency for Food and Drug Administration Control (NAFDAC) permissible limit while the free fatty acid, saponification value, iodine value and melting point were below NAFDAC permissible limit. The values for days 14 and 21 were significantly ($p<0.05$) higher when compared with the value for day 1. Acid value is a measure of the free fatty in palm oil and it is used to estimate the amount of palm oil that is lost during refining process (Enyoh et al., 2017). From the table, it was observed that the acid value of the oil sample increased as the number of days increased and

Table 1. The effect of palm fruit deterioration on the physicochemical properties of freshly prepared palm oil.

Unit	Parameter	Day 1	Day 7	Day 14	Day 21	NAFDAC (2019)
mgKOH/g	Acid value	4.0 ± 0.08^{df}	4.94 ± 0.33^{ce}	$6.08\pm1.17^{*ce}$	$6.68\pm0.44^{*ce}$	0.6
mgKOH/g	Free Fatty acid	1.96 ± 0.07^{bcf}	2.45 ± 0.11^{acf}	$2.81\pm0.26^{*ace}$	$3.09\pm0.14^{*bce}$	3.5
mgKOH/g	Saponification value	189.05 ± 1.63^{adf}	178.29 ± 5.67^{adf}	$136.52\pm4.07^{*bcf}$	$157.49\pm4.24^{*bde}$	190-209
mEq/kg	Peroxide value	12.25 ± 0.21^{bdf}	$14.53\pm0.78^{*ace}$	$13.90\pm0.14^{*ace}$	$14.90\pm0.24^{*ace}$	10
Wijs	Iodine value	50.13 ± 2.21^{bdc}	$30.19\pm1.71^{*acf}$	$35.90\pm1.83^{*acf}$	46.10 ± 2.54^{bde}	50 -55
ºBx	Refractive index	1.31±0.14	1.39±0.03	1.46±0.07	1.58±0.19	1.449-1.456
$Kgm^{-1}s^{-1}$	Viscosity	160.0 ± 4.24^{bdf}	$133.0\pm7.07^{*ace}$	$136.5\pm6.36^{*ace}$	$126.50\pm4.94^{*ace}$	N/A
Kgm^{-3}	Specific gravity	0.90±0.04	0.91±0.06	0.93±0.07	0.97±0.01	0.90-0.92
ºC	Cloud point	10.39 ± 0.01^{bdf}	$9.74\pm0.08^{*adf}$	$10.89\pm0.07^{*bce}$	11.09 ± 0.158^{bce}	10ºC
ºC	Flash point	304.5 ± 0.71^{bdf}	$300.5\pm2.12^{*adf}$	301.5 ± 0.71^{acf}	$308.5\pm0.17^{*bde}$	250ºC
ºC	Melting point	4.94 ± 0.08^{adf}	4.99 ± 0.01^{adf}	$4.69\pm0.01^{*bcf}$	$4.19\pm0.01^{*bde}$	37-40ºC
ºC	Boiling point	342 ± 4.24^{de}	339 ± 1.41^{adf}	$350\pm1.41^{*bce}$	348 ± 282^{be}	300ºC

Values are expressed as mean ± standard deviation. Values in each row with superscript *, a,b; c,d; e,f; are significantly different at P <0.05. Values with superscript * differ significantly when comparing day 1 with other days, values with superscript a,b differ significantly when comparing day7 with other days, values with superscript c,d differ significantly when comparing day14 with other days and values with superscript e,f differ significantly when comparing day 21 with other days.
Source: Authors

the values obtained were above NAFDAC permissible limits. High acid value could imply poor refining of the oil, thus lowering the palm oil quality. Also, the values reported in this study were higher than that reported by Akinola et al. (2010) and Rafiu et al. (2022).

Free fatty acid is one of the most important quality parameters in edible palm oil. The amount of free fatty acids in palm oil is an indicator of the quality of the palm oil and high level of free fatty acids is an indication of lipid oxidation (Akinola et al., 2010). The free fatty acid value is half the acid value and for this study, it ranged from 1.96±0.07 to 3.09±0.14 mg KOH/g. and it increased as the days increase. The value for day 21 was significantly higher as compared to the values for days 1 and 7. However, the values obtained were below NAFDAC limit, which indicates that the level of oxidation of the palm fruits at the time of processing was low (Akinola et al., 2010). The increase in the free fatty acid (FFA) values as the day increases are in tandem with the findings of Taage et al. (2012) who reported that FFA values of processed palm oil could be influenced by the degree of deterioration.

Saponification value indicates the molecular weight of triglycerides of oils (Abulude et al., 2015). The saponification values obtained showed that values for days 1 and 7 were significantly ($p<0.05$) higher when compared with the values for days 14 and 21, and all the values were lower than NAFDAC permissible limit (190-209) mg KOH/g. Agbaire et al. (2012) reported in their study that the higher the saponification value, the better the crude palm oils suited for soap production, thus processing the oil within seven days of harvest will yield good oil suitable for soap production.

Peroxide value (PV) is a useful indicator of the early stages of rancidity during storage of palm oil. It can be used to check the presence of unsaturation and to determine autoxidation (oxidative rancidity) as peroxides are intermediates of the autoxidation reaction (Onyeka et al., 2005). The degree of deterioration of the fruit before processing the oil affects its taste, odor and colour due to oxidative rancidity. The result showed that the palm oil from the palm fruit processed immediately after harvest (day 1) had a significant low PV when compared with other days (7, 14 and 21). The values obtained were all above the NAFDAC permissible limit (10) mEq/kg. High peroxide value is a definite indication of high level of unsaturation and susceptibility to oxidative rancidity (Enyoh et al., 2017).

The Iodine value is a measure of the degree of unsaturation (C=C) in relation to the amount of fat or oil (Hasan et al., 2016). The result showed that iodine value of the oil processed immediately (day 1) was significantly high when compared with other days (7 and 14). Iodine value of day 1 oil (50.13±2.21) is within the NAFDAC recommended range of 50 to 55, while the values for other days were below the range. The value for day 21 is similar to that reported by Akinola et al. (2010) and Rafiu et al. (2022). These low values suggest that the oil has low level of unsaturation and might not be susceptible to oxidation (Hasan et al., 2016).

Refractive Index (RI) measures how much light bends when travelling through oil and it also assess the purity of the palm oil. RI is mostly used as a quality control measure (Sarkar et al., 2015). It helps to indicate the possible chances of spoilage and higher RI indicates rancidity in palm oil (Sarkar et al., 2015). The values obtained showed no significant difference for all the days and the values for days 1, 7 and 14 were within the limit.

Table 2. The effect of storage duration on the fatty acid component of freshly prepared palm oil.

Component	Day 1	Day 7	Day 14	Day 21
C12	4.77 ± 0.06^{bcf}	$8.43 \pm 0.61*^{adf}$	4.82 ± 0.01^{bcf}	$3.66 \pm 0.14*^{bde}$
C14	0.22 ± 0.04^{bd}	$6.62 \pm 0.47*^{ad}$	$1.16 \pm 0.07*^{bc}$	--
C16	25.48 ± 0.65^{bdf}	$41.02 \pm 2.23*^{adf}$	$66.81 \pm 1.48*^{bcf}$	$34.74 \pm 1.29*^{bde}$
C18	14.40 ± 0.72^{bf}	$3.50 \pm 0.21*^{a}$	--	$3.49 \pm 0.09*^{e}$
C18:1	4.26 ± 0.06^{bdf}	$7.55 \pm 0.38*^{af}$	$8.19 \pm 0.23*^{acf}$	$27.10 \pm 1.27*^{bde}$
C18:2	9.22 ± 0.32^{bde}	$5.06 \pm 0.08*^{adf}$	$3.60 \pm 0.06*^{bcf}$	8.99 ± 0.49^{bde}
C18:3	11.74 ± 0.36^{bdf}	$9.40 \pm 0.07*^{acf}$	$9.63 \pm 0.17*^{acf}$	$4.32 \pm 0.03*^{bde}$
C20:2	5.22 ± 0.16^{bf}	$0.18*^{af}$	--	$3.38 \pm 0.13*^{be}$
C20:3	$1.90 \pm 0.0.9^{df}$	--	$7.79 \pm 0.14*^{cf}$	$3.58 \pm 0.04*^{de}$
C20:4	5.89 ± 0.15^{df}	--	7.80 ± 0.52	$1.14 \pm 0.08*^{de}$
C20:5	7.75 ± 0.19^{bdf}	$6.64 \pm 0.05*^{adf}$	7.92 ± 0.11^{bcf}	$2.12 \pm 0.04*^{bde}$
C22	4.94 ± 0.08^{bdf}	$3.16 \pm 0.06*^{adf}$	$3.82 \pm 0.06*^{bcf}$	$1.99 \pm 0.01*^{bde}$
C24	$4..45 \pm 0.15^{bf}$	$0.01 \pm 0.00*^{af}$	--	$0.01*^{bc}$

Values are expressed as mean ± standard deviation. Values in each row with superscript *, a,b; c,d; e,f; are significantly different at P <0.05. Values with superscript * differ significantly when comparing day1 with other days, values with superscript a,b differ significantly when comparing day 7 with other days, values with superscript c,d differ significantly when comparing day 14 with other days and values with superscript e,f differ significantly when comparing day 21 with other days.
Source: Authors

The significant decrease observed during the early stages of processing (days 1 and 7) could be as a result of the degree free fatty acids and oxidation (Akinola et al., 2010).

Viscosity is a measure of internal friction of the oil molecules. Relative high viscosity of palm oil may be due to intermolecular attraction between the long chain structures of the triacylglycerol molecules. The result revealed that the viscosity of the oil sample immediately after harvest was significantly higher as compared to the values obtained after 7, 14 and 21 days of deterioration.

Specific gravity also known as Relative Density is one of the most basic bio-diesel feed stock properties because of its correlation with heating value (Kenechi et al., 2017). The result of this findings showed that the specific gravity of the first two days of processing (days 1and 7) were within the NAFDAC acceptable limit of 0.90 - 0.92 kg/m^3 while the later (14 and 21 days) values were higher than the standard. High specific gravity of oil is an indication of the level of spoilage and adulteration of the palm oil (Kenechi et al., 2017).

Cloud point is the temperature at which one component of a mixture of liquid begins to solidify on cooling, resulting to invisible cloudiness. The cloud point is closely related to degree of unsaturation as increase in the degree of unsaturation of a sample results to a decrease in the cloud point of the given sample (Khalid et al., 2011). The degree of the cloud point of the sample for all the days except day 7 was above NAFDAC limit of 10°C

The Flash Point is the temperature at which the decomposition products produced from frying oils can be ignited. The flash points of the sample in all the days were greater than the NAFDAC range (250°C) for flash point of palm oil (Khalid et al., 2011).

Table 2 shows the effect of deterioration period on the fatty acid component of freshly prepared palm oil. Fatty acid composition is peculiar to each oil sample because it affects the physicochemical properties of the oil sample (Aremu et al., 2015). Lauric acid also known as Dodecanoic is a medium chain saturated fatty acid with 12-carbon atom chain. It can be metabolized into ketone bodies, which are important energy source for extra-hepatic organs in the body, such as the brain, heart and muscle (Sodamade et al., 2013). The result showed that oil for day 7 had significant ($p<0.05$) increase in the composition of lauric and Myristricacid when compared with oils for other days. Myristic acid is a long chain saturated fatty acid with 14 carbons and it accumulates fat in the body. Moderate consumption of myristic acid improves long-chain omega-3 fatty acids level in plasma phospholipid, which could exert improvement of cardiovascular health parameters in human (Sodamade et al., 2013).

Palmitic acid (C16:0) is a saturated fatty acid and it is a principal constituent of refined palm oil. It is the most common saturated fatty acids in the human body and it represents 20 to 30% of total fatty acids in membrane phospholipids and adipose triacylglycerol. It helps in formation of cell membranes, lung secretion and signaling molecules, while also storing and utilizing energy within cells and modifying proteins (Arzoo and Bakeet, 2014). The result revealed that the composition of palmitic acid for day 14 oil was significantly higher when compared with other days.

Stearic acid (C18) also called Octadecanoic is a saturated straight chain fatty acid consisting of 18 carbon atoms without double bonds. Stearic acid has been shown to have a neutral effect on blood total and low-density lipoprotein (LDL) cholesterol levels (Arzoo and Bakeet, 2014). The result revealed that the value for day one oil was significantly ($p<0.05$) higher as compared to the values for other days. Oleic acid (C18:1) is a mono-unsaturated omega-9 fatty acids (Sodamade et al., 2013). Excess of oleic acid blocks the body from using linoleic acid. From the Table 2, the value for day 21 oil was significantly different when compared with other days.

Linoleic acid also known as omega-6 is a poly-unsaturated omega-6 fatty acid. It possesses two more double bonds and lacks several hydrogen atom that are found in saturated fatty acids and it is an important constituent of neuronal membrane phospholipids (Arzoo and Bakeet, 2014). Linolenic acid is an omega-3 polyunsaturated acids which helps to lower blood pressure and also reduce serum triglyceride (Srilakshmi, 2008). The result showed that the values of C18:2 (linoleic acid) and C18:3 (Linolenic acid) for day 1 oil was significantly different than the oil of other days, the values showed that C18:3 decreased as the fruit deteriorate.

Conclusion

The result reveals that the acid value, peroxide value, flash point and boiling point of the sample were above NAFDAC permissible limit while the free fatty acid, saponification value, iodine value and melting point were below NAFDAC permissible limit. The result also shows that most physiochemical properties (such as acid value, free fatty acid, refractive index, specific gravity, cloud point and boiling points) increased in days 14 and 21 oil, thus it could be concluded that allowing the palm fruit to deteriorate after harvest beyond seven days could reduce the quality of oil.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Full Length Research Paper

Biochemical exploration of pancreatic function in market gardeners exposed to pesticides in Bobo-Dioulasso, Burkina Faso

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Market gardeners use pesticides that could contain disruptors affecting pancreatic function. The objective was to explore exocrine and endocrine pancreatic functions in market gardeners exposed to pesticides. This prospective study was conducted on a cohort of 50 market garden producer participants. Sociodemographic and clinical variables, as well as information on pesticides used, were collected alongside blood samples. Quantitative determination of total insulinemia in serum was conducted using a chemiluminescence microparticle immunoassay. Lipasemia, pancreatic amylasemia, triglyceridemia, and glycemia were measured through colorimetric or enzymatic methods. Reference intervals for glycemia (4.11-6.50 mmol/L), lipasemia (13-60 U/L), pancreatic amylasemia (13-53 U/L), triglyceridemia (<1.70 mmol/L), and insulinemia (2.60-24.90 µU/L) were considered. Fisher's exact test was employed to examine the significance of associations, with a p-value set at 0.05. The market garden producers included in the study were males with a median age of 38 years. They were most frequently exposed to active substances such as glyphosate (89.47%), paraquat chloride (36.84%), and 2,4-D dimethylamine salt (15.79%). Hyperinsulinemia (21.05%), hypoglycemia (44.00%), and hypertriglyceridemia (10.53%) were noted. An increase in catalytic activity of pancreatic enzymes was observed, including hyperlipasemia (2.63%) and hyperamylasemia (13.16%). Through insulinemia, a disorder in the endocrine secretion of the pancreas was noted, which may not be solely attributed to the pesticides used. Further comparative studies are needed.

Key words: Pancreas, market gardeners, pesticides.

INTRODUCTION

In Burkina Faso, as in other West African countries, cities are undergoing rapid population growth accompanied by accelerated urbanization (Yemadji et al., 2017). This trend has driven the expansion of urban and peri-urban agriculture due to increased food needs, rising living standards, and the necessity to diversify sources of income (Kêdowidé et al., 2010). Vegetable production remains the predominant agricultural activity in the larger cities of Burkina Faso (Robert et al., 2018). Nevertheless, crop losses (approximately 30%) resulting from damage caused by pests, nematodes, and weeds constitute a significant agricultural challenge for producers (Grunder et al., 2018).

In fact, a considerable proportion (about 59 to 100%) of market garden producers in major production centers such as Bobo-Dioulasso, Ouagadougou, and Ouahigouya resort to periodic pesticide application without adhering to the recommended dosage (Naré et al., 2015; Son et al., 2017). Additionally, a substantial number of synthetic pesticides employed by market gardeners lack registration by the Sahelian Pesticides Committee (CSP) and are consequently unauthorized for use in Burkina Faso (Toé et al., 2013). This widespread improper utilization of highly toxic chemical pesticides has adverse implications for human health (Son et al., 2018).

Pesticide exposure presents a significant public health concern, with over 200,000 cases of accidental poisoning reported annually (Liang et al., 2018). Furthermore, the contribution of pesticide exposure to pancreatic dysfunction remains unexplored in this region. The objective of this study is to assess the pancreatic function of market gardeners exposed to pesticides.

MATERIALS AND METHODS

Site and type of study

This is a prospective study that was conducted from July 10 to September 15, 2019. The study took place in the region of "Hauts-Bassins" precisely in periurban district 3 (Dogona) and 5 (Kuinima) (Figure 1).

Population and sampling

The study focused on a cohort of 50 market garden producers. The inclusion criteria were: being male or female of any age; owning a farm in the study area; using pesticides on the farm during the study period; and agreeing to participate in the study. This study was authorized by the Ethics Committee for Health Research of Burkina Faso, deliberation n ° 2018-7-083. Written informed consent was obtained from all market garden producers included in this study.

They were alerted about the high morbidity rate observed in persons using pesticides, including hormonal disorders. Their participation was completely voluntary. Biological samples were well labeled and all data were processed in anonymity.

Collection and processing of blood samples

A venepuncture was carried out using a needle with a vacutainer support. Three (03) ml of blood was taken under strict aseptic conditions. The blood was then packaged in a dry tube and transported directly to the laboratory in coolers containing the tube racks. At the laboratory, the blood samples were centrifuged, aliquoted and stored at a temperature of -20°C prior to analysis.

Analysis of biochemical parameters

The analysis of blood samples was done in the biochemistry department in "Centre Hospitalier Universitaire Sourô SANOU de Bobo-Dioulasso". Quantitative determination of total insulinemia in serum was carried out by a chemiluminescence microparticle immunoassay (CMIA) and lipasemia, pancreatic amylasemia, triglyceridemia, glycemia were measured either by the colorimetric and enzymatic methods on HITACHI Cobas® 6000.

Data analysis

Data were analyzed with XLSTAT 2016.02.27444 (Lumivero, Colorado, USA). Statistical parameters such as medians, minimum, maximum and frequencies were expressed for data. We have considered reference intervals for glycemia (4.11-6.50 mmol/L), lipasemia (13-60 U/L), pancreatic amylasemia (13-53 U/L), triglyceridemia (<1.70 mmol/L) and insulinemia (2.60-24.90 µU/L). Fisher's exact test was used to assess the significance of associations with a p value < 0.05.

RESULTS

Socio-demographic and clinical characteristics

Market garden producers included were male with a median age of 38 years (minimum-maximum: 22-69 years). Market gardeners with primary education had a proportion 30.77%, secondary education 13.46% and illiterate 55.77%. Clinical signs frequently reported were colds (29.31%), coughs (25.86%), headaches (22.41%), and dizziness (12.07%). No clinical signs were reported for 39.47%.

Pesticide used

Seventeen pesticides were cited and used by the market gardeners. From these pesticides, 64.70% were registered by CSP. According to World Health Organization (WHO), 5.88% and 47.05% were in toxicity class U, II and III respectively. Glyphader 75 (58.00%), Gramopat super (32.00%) and Kalache (22.00%) were the most used pesticides. Their active substance were glyphosate (89.47%), paraquat chloride (36.84%) and 2,4-D dimethylamine salt (15.79%), endosulfan (5.26%), thirame (5.26%), acetamipride (2.63%) and

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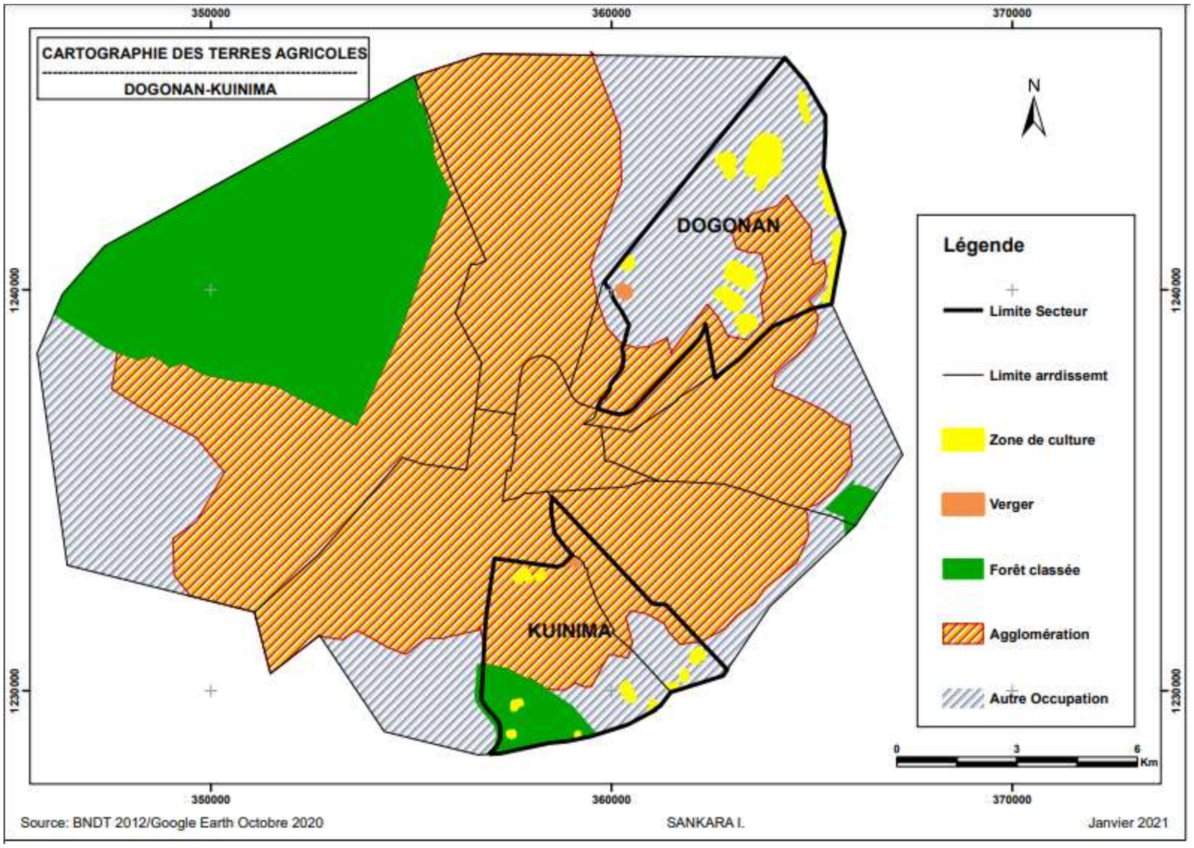

Figure 1. Agricultural land mapping.

lamda-cyalothrine (2.63%).

Market gardeners and pesticide used

Market gardeners who had received at least one training on good pesticide use represented 4.00% ; and those who had received no training at all were 96.00%. Among our market gardeners, 52.00% had less than ten years pesticide use experience; 36.00% had [10-20[years' experience and 12.00% had [20-30[years' experience. They were exposed to a single active substance (47.37% and 5.26% for glyphosate and paraquat chloride) or a combination of 2 (34.21%) or 3 (13.16%) active substances (Table 1).

Biochemical parameters analysis

We noted hypoinsulinemia (2.63%) and hyperinsulinemia (21.05%), but no significant association with glyphosate (p= 0.604), paraquat chloride (p=0.388) exposure. A significant association was observed with 2,4-D dimethylamin salt exposure and hyperinsulinemia (p=0.039). Hypoglycaemia (44.74%) and hyperglycaemia (2.63%) were observed in market gardeners but were not significantly associated with glyphosate (p= 0.650), paraquat chloride (p=0.579) or 2,4-D dimethylamin salt exposure (Table 1). No significant association was observed for 10.52% of market gardeners with both hyperinsulinemia and hypoglycemia (p= 0.793).

An increase in the catalytic activity of pancreatic enzymes was observed: hyperlipasemia (2.63%), hyperamylasemia (13.16%). Hypertriglyceridemia was 10.53% and was not significantly associated with any active substance exposure (p=0.372) or lipasemia level (p=0.826) (Table 2).

DISCUSSION

Biological studies on pancreatic function in response to pesticide exposure are scarce in Burkina Faso. Consequently, we were unable to locate any data on the prevalence of toxicity resulting from pesticide exposure, which could have enabled us to make comparisons with our findings. Moreover, we encountered a lack of biological crop promoters in these peri-urban environments that could have served as a control group.

Market gardening appears to be a predominantly male

Table 1. Single or combined active substances in pesticides used by market gardeners.

Active substances	Frequency (%)
Single active substance	52.63
Glyphosate	47.37
Paraquat chloride	5.26
Two active substances	34.21
Glyphosate+Paraquat chloride	23.68
Glyphosate+2,4-D dimethylamine	7.89
Endosulfan+Thirame	2.63
Three active substances	13.16
Glyphosate+2,4-D dimethylamine+Paraquat chloride	5.26
Glyphosate+2,4-D dimethylamine+Endosulfan	2.63
Glyphosate+Endosulfan+Thirame	2.63
Paraquat chloride+Lamda-cyhalothrine+Acetamipride	2.63

Table 2. Frequency of biochemical parameters level and active substance exposure.

Biochemical parameters level	Glyphosate		p-value	Paraquat chloride		p-value	2,4-D dimethylamin		p-value
	No	Yes		No	Yes		No	Yes	
Glycemia (mmol/L)			0.650			0.579			0.118
High	0.00	2.63		2.63	0.00		2.63	0.00	
Low	2.63	42.11		31.58	13.16		31.58	13.16	
Normal	7.89	44.74		28.95	23.68		50.00	2.63	
Insulinemia (µU/ml)			0.604			0.388			0.039
High	0.00	21.05		15.79	5.26		15.79	5.26	
Low	0.00	2.63		0.00	2.63		0.00	2.63	
Normal	10.53	65.79		47.37	28.95		68.42	7.89	
Triglyceridemia (mmol/L)			0.372			1.00			1.00
High	2.63	7.89		7.89	2.63		10.53	0.00	
Normal	7.89	81.58		55.26	34.21		73.68	15.79	
Amylasemia (U/L)			0.446			0.337			0.570
High	2.63	10.53		5.26	7.89		13.16	0.00	
Normal	7.89	78.95		57.89	28.95		71.05	15.79	
Lipasemia (U/L)			1.00			0.043			1.00
High	0.00	2.63		0.00	2.63		2.63	0.00	
Normal	10.53	81.58		63.16	28.95		76.32	15.79	
low	0.00	5.26		0.00	5.26		5.26	0.00	

activity. These results align with those of Sawadogo (2016), who reported a 100.00% male participation rate. In contrast, the Western Regional Directorate of the National Center for Scientific and Technological Research (DRO/CNRST, 2016) found male representation at 94.10% and females at 5.86%. This variation could potentially be attributed to differences in sample size.

The age range of participants spanned from 22 to 69 years. These findings indicate that pesticide application is an activity involving individuals across different age categories. The educational attainment of surveyed market gardeners was generally low, with 55.77% being illiterate. A substantial number had not received any training in proper pesticide use techniques. The combination of limited education and lack of training constrains the safe application of pesticides. Furthermore, market gardeners with limited educational backgrounds might face challenges in comprehending label information, thereby increasing their exposure risk.

Similar observations were made by Toé (2010), who highlighted that the educational levels of producers do not facilitate the establishment of systems aimed at reducing health and environmental risks. Nonetheless, it is possible to educate gardeners in reading and writing in local languages such as Dioula, as suggested by Toé (2010). This could substantially enhance pesticide safety. The survey findings indicated that 52.00% of farmers had fewer than ten years of experience in pesticide use, while 12.00% possessed extensive experience exceeding twenty years. Long-standing experience in pesticide use could be a significant factor contributing to effective pesticide utilization.

However, extended experience could also serve as a toxicological risk factor. In the field, we have observed that some market gardeners who have been using pesticides for an extended period do not set the best example. This exposure to pesticide hazards might be further compounded by the utilization of unregistered pesticides as determined by the CSP. According to the WHO, 5.88% of the pesticides fell under class U, implying that they did not pose an acute hazard under proper usage conditions. Classes II and III accounted for 47.05% each, with Class II classified as moderately hazardous and Class III as slightly hazardous. Various pesticides contained one or more active substances, such as glyphosate and paraquat chloride, both of which have been shown to impact the pancreas. This aligns with the observations made by the Pesticide Action Network Updates Service (PANUPS, 2001) in 2001, which identified endosulfan as a hazardous substance restricted by the Sahelian Pesticide Committee (CSP), potentially causing disruptions in the human body's hormonal system and leading to increased birth defects, sexual abnormalities, and reproductive disabilities.

According to vegetable growers, unregistered pesticides, especially those originating from countries like Ghana and Nigeria, are more affordable than products offered by domestic distribution companies. Similar findings were reported by Ibrango (2014), who noted that these growers favored cheaper pesticides from Ghana. In terms of the assay of biological variables, our study found that 21.05% of pesticide users exhibited hyperinsulinemia, with 10.52% of them also experiencing hypoglycemia. This could potentially be attributed to a malfunction in the pancreatic endocrine function. Similar findings were observed by Jamshidi et al. (2009) and Bousquet (2016), who found increased insulin levels and pancreatic cancer in rats exposed to diazinon and glyphosate, respectively. Patients demonstrating both hyperinsulinemia and hypoglycemia could be explained by the natural role of insulin, an antihyperglycemic hormone, wherein elevated insulin levels lead to lower blood sugar.

The observed hypoglycemia in 44.74% of patients could be attributed to the 12-hour fasting period prior to collection, as well as the inactivity of pancreatic β cells, which stimulate antihypoglycemic hormone secretion. Hyperglycemia among our patients might be linked to a dysfunction of pancreatic α cells. Hyperlipasemia was detected in 2.63% of patients, and hyperamylasemia in 13.16% of patients. The escalation in catalytic activity of pancreatic enzymes could indicate acute pancreatitis, prompting a significant release of these enzymes. Bismuth et al. (1982) made similar observations during acute paraquat intoxication, reporting hyperlipasemia and hyperamylasemia in 26.00% of cases. Hypolipasemia in 5.26% of patients could point toward chronic pancreatitis, wherein lipase secretion diminishes over time. These biological disorders, whether acute or chronic in nature, could be attributed not only to the duration of exposure but also to the quantity used. Hypertriglyceridemia was noted in 8.00% of patients, which might be due to a deficiency in lipase secretion affecting the conversion of triglycerides into fatty acids.

Conclusion

This survey is characterized by poor control of pesticide use techniques and the choice of unregistered pesticides with lesser-known toxicity. Biochemical parameters showed biological disorders in lipasemia, amylasemia (pancreatic enzymes), insulinemia (pancreatic endocrine hormone), and hormonal regulation disorders of triglyceridemia and glycemia. However, the results obtained cannot be considered representative, as market gardeners have cited and used several types of pesticides, making it difficult to assign responsibility to each active substance.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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