Chemistry Research Journal, 2020, 5(6):189-194

Available online www.chemrj.org

Research Article

ISSN: 2455-8990
CODEN(USA): CRJHA5

Variations on the Physiochemical, Microbiological and Selected Heavy Metals of Different Palm Oil Samples Sourced from Galadima, Tarauni, Sabon-Gari, Yan-Kura of Kano State and Samples from Kogi and Edo States of Nigeria

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Abstract This study involves the physiochemical, microbiological and selected heavy metals variations of palm oil samples sourced from *galadima, tarauni, yan-kura* markets of kano State and samples from Kogi and Edo States of Nigeria. The samples were evaluated using standard documented procedures. The result showed; samples from Edo had: oil acid value 0.73±0.13, iodine value 29.25±0.09, peroxides value 2.68±0.09, saponification value 194.31±2.21, moisture content 1.13±0.56, ester value 195.04±2.08. The samples from Kogi had; acid value 0.88±0.11, iodine value 11.54±0.24, peroxides value 5.79±0.62, saponification value 187.49±0.56, moisture content 1.60±0.10, ester value 186.61±0.45. The *sabon-gari* samples had; oil acid value 0.86±0.17, iodine value 30.37±0.34, peroxides value 3.25±0.95, saponification value 188.66±1.88, moisture content 1.19±0.59, ester value187.8±1.71. *Galadima* samples had: acid value 0.89±0.14, iodine value 29.31±0.47, peroxides value 6.8±0.16, saponification value 182.62±3.65, moisture content 1.82±0.67, ester value 181.73±3.5. *taruani* samples had; acid value 0.56±0.13, iodine value 29.63±0.81, peroxides value 5.79±0.62, saponification value 194.07±0.47, moisture content1.43±0.71, ester value 193.51±0.44. Yan-kura samples had: acid value 0.98±0.09, iodine value 30.78±0.26, peroxides value 6.2±0.14, saponification value 184.045.00, and moisture content1 21±0.60, ester value 183.06±4.91. The microbiological analysis of the samples revealed *sabon-gari* samples had1.0 x 10^{-5}, galadima 7.6 x 10^{-3}, Tarauni 7.2 x 10^{-4}, Kogi 1.2 x 10^{-5}, Yan-kura 1.1 x10^{-5} and Edo 6.8 x 10^{-3} cfu/ml respectively. The heavy metals analysed were cadmium (Cd), arsenic (As), mercury (Hg) and lead (Pb). The concentrations of Cd ranged between 0.001 *(ppm)* Tarauni to 0.019 *(ppm)* Galadima, As *(ppm)* ranged between 0.01 Edo and Kogi to 0.047. Hg *(ppm)* 0.01 Edo to 0.19 galadima and Pb *(ppm)* 0.05 Kogi to 0.18 galadima. From the results obtained, it shows that Yan-kura has the most polluted oil due to the exposure to the environment. The samples collected are safe for consumption as at time of analysis, however continuous monitoring is required.

Keywords palm oil, physiochemical, microbiological, heavy metals

Introduction

Palm oil is derived from the *mesocarp* of the fruit of the oil palm. It has light yellow to red color [1]. Red palm oil get its name from its characteristic dark red color, which come from carotenes, such as alpha-carotene, beta-carotene

and lycopene which are responsible for high vitamin A content [1]. The oil palm (*Elaeis guinensis*) is West Africa's most important oil producing plant. The fruit produces two distant type of oil. The orange to red crude palm oil which is extracted from the mesocarp and brown light yellow crude palm kernel oil extracted from the seed (kernel). The former consists of mainly palmitic and oleic acids and the latter, mainly lauric acid. Both oils are important in the world trade. Crude palm oil (CPO) is the richest natural source of carotenoilds and cotrienols. While it's semi-solid consistency at a tropical room temperature is mainly due to the presence of triacylgycerols of palmitic and oleic acid [2]. Human use oil palm as far as 5,000 years back, in the late 1800s archaeologists discovered a substance that they conducted was originally palm oil in a tomb at Abydos dating back to 3,000 BC. It is believed that traders brought oil palm to Egypt.

The work is aimed at ascertaining the physiochemical, microbiological and selected heavy metals of different palm oil sample consumed from different markets of *Galadima, Tarauni market, Sabon-gari, Yan-kura* of Kano state and sample from Kogi and Edo states.

Materials

The following materials were used for the study; wijis solution, thiosulphate solution, glacial acetic acid (BDH Chemicals England), Concentration nitric (Merck Chemicals), Sulphuric acid (BDH Chemicals), hydrogen peroxide (BDH Chemicals England), Hot air oven (Gellenkamp, UK), desiccators, Atomic absorption spectrometer (Shimadzu, Model-AA7000 with air acetylene flame at 324.8 nm), Incubators, Muffle furnace (Gellenkamp, UK), colony counters (Hanna, USA).

Sampling

The palm oil samples that were used for the study were collected in triplicate from oil market in size locations namely: *Galadima, Tarauni, Sabon-gari, Yan-kura,* in Kano state, Kogi and Edo state Nigeria.

Experimentation

Determination of acid value

The oil sample (1.0g) was boiled with 50.0 cm^3 ethanol, and then was allowed to cool and two drops of phenolphthalein indicator was added. The resulting solution was titrated against 0.1 mol/dm^3 NaOH until a pink colour was obtained [3].

The acid value was calculated using equation;

$$Acid\ value\ content = VxMx5.61/weight\ of\ sample$$

V is the titre value and W is the weight of oil, M is the molar mass NaOH.

Determination of iodine value

The method described by Marshall *et al* [4] was adopted. About (0.5 g) will be placed in 250.0 cm^3 conical flask and 10.0 cm^3 of anhydrous chloroform was added. This was followed by 30.0 cm^3 of solution and the flask was stopped and allowed to stand in the draw for 30 minutes and which potassium iodine (10.0 cm^3 of 15% v/v) was added to the content of the flask so as to wash down any iodine that might present on the stoppard. The resulting solution was titrated with sodium thiosulphate solution (0.14 M) until the light yellow colour form disappears. The determination for the blank will be conducted in the same manner but without the oil. The iodine value was calculated as:

$$Iodine\ Value\ content = \frac{(B-S)xMx12.69}{W}$$

B and S was titre value of blank and sample respectively, M is molarity of sodium thiosulphate, 12.69 is the conversion factor from meq sodium thiosulphate to gram molecular weight of iodine and W is the weight of oil [3].

Determination for peroxides value

One gram of oil sample was weighed into a clean dry boiling tube. While still liquid, one gram of powdered potassium iodide and 20 cm^3 of solvent mixture (2.0 volume glacial acetic acid and 2.0 volume chloroform.) were added. The tube was placed in boiling water so that the liquid boils within 30 sec. and allows boiling vigorously for not more than 30 seconds.

The content was placed quickly into a flask containing 20.0 cm^3 of potassium iodide solution (5.0 %) the tube was filled twice with 25.0 cm^3 water and was titrated with 0.002 mol sodium thiosulphate using starch indicator.

A blank will be performed at the same time. The peroxide value was calculated using equation [4].

$$Peroxide\ Value\ \frac{meq}{Kg} = \frac{(S-B)xMx1000}{weight\ of\ sample}$$

Where; B and S titre values of blank and sample respectively, M is the Molarity of $Na_2S_2O_3$.

Determination of saponification value

Two grams of the oil sample was weighed into clean dried conical flask and 25.0 cm^3 of alcohol potassium hydroxide will be added. The flask was heated for an hour with frequent shaking. 1.0 cm^3 phenolphthalein indicator was added and the hot excess alkali titrated with 0.5 mol/dm^3 hydrochloric acid (HCl) until it reaches the end point where it turns colourless.

A blank titration was carried at the same time. The saponification value was calculated using equation;

$$SV\ content = \frac{(S-B)xMx56.1}{weight\ ofsample}$$

Where S = Sample titre value, B = Blank titre value, M = Molarity of HCl (0.5 M) and 56.1 = Molecular weight of potassium hydroxide [3].

Unsaponifiable Matter

After the titration of the saponification value, the resultant solution was made alkaline again with 1.0 cm^3 of aqueous 3.0 M potassium hydroxide solution. And was then transfer to a separator and washed in with water.

The solution was extracted while still just warm three times with 50.0 cm^3 quantities of diethyl ether and also poured into separator containing 20.0 cm^3 water. After the third extract was added, the combined ether extracts was shaked with the first 20ml of wash water and vigorously with another 20.0 cm^3. The third was washed twice with 20.0 cm^3 of aqueous 0.5 M potassium hydroxide solution and at least twice with 20.0 cm^3 quantity of water until the wash water is no longer alkali to phenolphthalein. The ether extract was poured until a weighed flask, the solvent was evaporated off and the residue dried at about 550.0 ºC, cooled and weighed [3].

Refractive Index

The refractive index was determined using Abbe's refractor meter. The Abbe's refractor meter was reset with a light compensator (20.0 ºC) the oil sample was smeared on the lower prism of the instrument and closed. Light was passed by means of angled mirror, the reflected light appear in form of a dark background. The fine adjustment of telescope tube was used until the black shadow appears at central in the cross wire indicator. The refractive index was read off and recorded [3].

Determination of Rancidity

The rancidity of the oil sample was determined qualitatively using Kries Test. About 5.0 cm^3 of the oil samples was placed in 100.0 cm^3 test tube and then mixed vigorously with 5.0 cm^3 of universal indicator and 5.0 cm^3 of concentrated HCl for about 20.0 seconds. The presence of pink colour indicates incipient rancidity [3].

Determination of pH value

The pH of the oil sample was determined using a pH meter, about 30.0 cm^3 of the oil sample was poured into a beaker, then the pH meter electrode was immersed into the beaker containing the oil sample and the pH values was recorded [5].

Determination of Moisture Content

Gravimeter method was used for moisture determination. About 5.0 g of sample was weighed into evaporation dish. It was placed in an oven and maintained at 105.0 ºC for 3 hours interval. After drying to a constant weight, the sample was cooled in a desecrator and re-weighed using analytical balance [6].

$$Percentage\ Moisture\ content = \frac{(b-c)x100}{b-a}$$

Where b = weight of crucible and sample

c = weight of crucible and dried oil

a = weight of crucible only.

Determination of Ester Value

The ester was determined by subtracting acid value from saponification value [6].

Microbiological Analysis

Media Preparation

28.0 grams of nutrient agar was suspended into 1000.0 ml of distilled water. It was boiled to dissolve the medium completely. It was then dispensed as desired and sterilized by autoclaving at 15.0 pounce pressure (121 ºC) for 15 minutes and was allowed to cool.

Serial Dilution

10.0 ml of the oil sample was dissolved with ethanol. It was then transferred into 90.0 ml of distilled water (stock solution). 9.0 cm^3 of distilled water was added into six test tubes, each was labeled 10^{-1}, 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5}, 10^{-6}. 1.0 cm^3 of the oil sample from the stock solution was added into the first test tube (10^{-1}) and was shaked vigorously then 1.0 cm^3 of the first sample (10^{-1}) added into the second sample (10^{-2}).This same step was repeated down to the sixth test tube [2].

Pour Plate Technique

Into a clean six petri-dish, 1.0 cm^3 of the sample in the test tube (10^{-4}, 10^{-5}, 10^{-6}). Each petri-dish labeled with the name of sample was then added into the petri-dish that is 1.0 cm^3 each of 10^{-4} was poured into the petri-dish and was labeled A and B the steps was followed on 10^{-5}, 10^{-6}. All the six dishes were filled with already prepared agar to the top. It was then allowed to solidify before turning the petri-dish upside down and was kept in the incubator for 48 hours before taking the reading [2].

Determination of heavy metals

10.0 cm^3 of the oil sample was weighed into a beaker, 20. 0 cm^3 of concentrated nitric acid was added and the solution transferred into the khjedal tube, the tube was inserted into the khjedal digester at 550.0 ºC for three hours in the fume cupboard after when the sample has been digested it was allowed to cool and 20.0 cm^3 of 0.1 N nitric acid was added and transferred into a sample bottle for analysis.

Table 1: The chemical properties on palm oil sample

Properties	**Edo** ***(X±SD)***	**Tarauni** ***(X±SD)***	**Yan-kura** ***(X±SD)***	**Galadima** ***(X±SD)***	**Kogi** ***(X±SD)***	**Sabon-gari** ***(X±SD)***
Acid value (mgKOH/g)	0.73±0.13	0.56±0.13	0.89±0.09	0.89±14	0.88±0.11	0.86±0.17
Iodine value I_2/g	29.25±0.09	29.63±0.81	30.78±0.26	29.31±0.47	11.54±0.24	30.37±0.34
Peroxide value (mg KOH/g)	2.68±0.09	5.79±0.62	6.2±0.14	6.8±0.16	2.97±0.22	3.25±0.95

Saponification (mgKOH/g)	194.31±2.21	194.07±0.47	184.04±5.00	182.62±3.65	187.49±0.56	188.66±1.88
Unsaponification	Detected	Detected	Detected	Detected	Detected	Detected
Rancidity	ND	ND	ND	ND	ND	ND
Ester value	195.04±2.08	193.51±0.44	183.06±4.91	191.73±0.35	191.73±0.35	187.8±1.77
Moisture content (%)	1.13±0.56	1.43±0.71	1.21±0.62	1.82±0.67	1.62±0.10	1.19±0.59

Table 2: The total bacterial load on palm oil

Sample	Dilution factor	No of colonies (Cfu/ml)	No of organism (Cfu/g)
Edo	10^{-6}	68	6.8×10^{-3}
Kogi	10^{-6}	124	1.2×10^{-5}
Yankura	10^{-6}	112	1.1×10^{-5}
Sabon gari	10^{-6}	108	1.0×10^{-5}
Tarauni	10^{-6}	72	7.2×10^{-4}
Galadima	10^{-6}	76	7.6×10^{-3}

Table 3: The heavy metal analysis on palm oil

Sample	Cd (ppm) (X±SD)	As (ppm) (X±SD)	Hg (ppm) (X±SD)	Pd (ppm) (X±SD)
Edo	0.007±0.01	0.009±0.01	0.01±0.03	0.04±0.02
Kogi	0.012±0.02	0.009±0.01	0.01±0.03	0.05±0.01
Yan-kura	0.011±0.01	0.008±0.01	0.13±0.02	0.93±0.03
Sabon-gari	0.010±0.01	0.014±0.02	0.04±0.02	0.22±0.02
Tarauni	0.001±0.02	0.021±0.03	0.10±0.013	0.37±0.01
Galadima	0.019±0.01	0.047±0.01	0.19±0.01	0.18±0.02

Discussions

The physicochemical properties of palm oil samples as shown in table one revealed that the acid value ranged from (Edo 0.73±0.13), 0.88 ±0.11 (kogi) 0.86 ± 0.17 (Sabon gari) 0.98 ± 0.09 (Yankura) 0.89±0.14 (Galadima) 0.56±0.13 (Taruani). The result shows that Sabon gari has the highest acid value. Acid value is taken as an important indicator of oxidation of oil. The acid value is the number of mg (milligram) of potassium hydroxide required to neutralize the free acid of the substance. The Iodine Value is to determine the amount of unsaturation contained in fatty acid, it ranged from 29.63±0.81 (Taruani) 29.25±0.09 (Edo) 29.31±0.47 (Galadima) 11.54 0.24 (kogi) 30.78±0.26 (Yan kura) 30.37±0.34 (Sabon gari). The result shows that Yankura has the highest iodine value. The peroxides value in oil samples indicares the state of oxidation of a substance under observation. If the oxidation proceed for long it makes the oil rancid and gives an unpleasant smell to the substance. The peroxides value of the samples: ranged from 2.68±0.09 (Edo) 2.97± 0.22 (Kogi) 5.79±0.62 (Taruani) 6.2±0.14 (Yankura) 3.25±0.95 (Sabon gari) 6.8 ±0.16 (Galadima). The result shows that Galadima samples have the highest peroxides value. The Saponification value is a measure of the total free and combined acid especially in fat expressed as the number of milligram of potassium hydroxide required for the complete saponification of one gram of substance. The saponification value of the samples ranged from 194.07 ± 0.47 (Taruani) 184.04±5.00 (Yan kura) 188.66 ±1.88 (Sabon-gari) 194.31±2.21 (Edo) 187.49±0.56 (kogi) 182.62±3.65 (Galadima). The result shows that Edo has the highest saponification value. Moisture content of oil samples an important parameter in assessing the quality of oil sample. The moisture content of any food is an index of it water activity. High moisture content is an indication of ease spoilage. The moisture content of the samples ranged from 1.13±0.56 (Edo) 1.43±0.71 (Taruani) 1.21±0.60 (Yankura) 1.19±0.59 (Sabon gari) 1.82±0.67 (Galadima) 1.60±0.10. The moisture content obtained is higher than standard of 0.29 % [7]. The high moisture content obtained will discourage the storage stability of palm oils. Ester value of oil sample is an amount of mg of KOH required to saponificate esters contained in one gram of oil. Ester value: ranged between

(Taruani 193.5±0.44), (Yankura 183.06±4.91) 187.8±1.71 (Sabon-gari) 195.04±2.08 (Edo) 186.61±0.45 (kogi) 181.73±3.5 (Galadima). The higher the ester value the more intact the ester bond between the glycerol molecule and fatty acid. Therefore the oil samples analysed are of high quality and can be able to be stored for a long time [8].

The microbiological analysis of the samples revealed *sabon-gari* samples had 1.0 x 10^{-5}, galadima 7.6 x 10^{-3}, Tarauni 7.2 x 10^{-4}, Kogi 1.2 x 10^{-5}, Yan-kura 1.1 x10^{-5} and Edo 6.8 x 10^{-3} cfu/ml respectively. The results revealed the samples collected are contaminated with micro organisms.

The result of heavy metals analysis using Atomic Absorption Spectrophotometer is shown in table 3. The results generally show the presence of cadmium (Cd), arsenic (As), lead (Pb), mercury (Hg) in palm oil samples. The concentrations of Cd ranged between 0.001 *(ppm)* Tarauni to 0.019 *(ppm)* Galadima, As *(ppm)* ranged between 0.01 Edo and Kogi to 0.047. Hg *(ppm)* 0.01 Edo to 0.19 galadima and Pb *(ppm)* 0.05 Kogi to 0.18 galadima. From the results obtained, it shows that Yan-kura has the most polluted oil due to the exposure to the environment.

Conclusion

Adulteration is very common among palm oil sellers especially in this part of the world, understanding the physical and chemical content of palm oil is therefore important. The results of the study indicate the suitability of the palm oil samples for both domestic and industrial uses. Current study highlight the need of implementation of food safety law at all level of palm oil processing specially within supply chain to ensure safe product to the consumers. Some micro organisms found in palm oil often lead to deterioration in their chemical quality. Hence the presence or absence of micro-organisms can be considered as quality determinant of palm oil. The microbial quality of palm oil is essential because of the adverse role played by most lipophillic micro-organisms in human and animal health.

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