CHARACTERIZATION OF FATTY ACIDS OF SESAME, ROSELLE AND SMOOTH LOOFAH SEEDS FLOUR

Habibat Omolara Adubiaro[1*], Abdul Ademola Olaleye[2] and Michael Ogunmola Oguntokun[3]

[1]Department of Chemistry, Faculty of Science, Federal University Oye-Ekiti, Ekiti State, Nigeria

[2]Department of Chemistry, Faculty of Science, Federal University Dutse, Jigawa State, Nigeria

Department of Animal Production and Health (Nutrition Laboratory), The Federal University of Technology, Akure, Ondo State, Nigeria

ABSTRACT

The fatty acids composition of three seeds flour: sesame, roselle and smooth loofah were determined using standard reagents and analytical techniques. The most abundant fatty acids (%) were linoleic acid (25.0-45.8), oleic acid (22.2-38.8), palmitic acid (8.77-25.5) and stearic acid (3.94-5.48). The following fatty acids recorded 0.00% concentration: lauric, myristic, margaric, lignoceric, palmitoleic, erucic and arachidonic acids; margaric acid (roselle); lauric and margaric acids (smooth loofah). Total saturated fatty acid (SFA) ranged from 15.1-41.4%; total monounsaturated fatty acid (MUFA) fell within 25.9-38.8%) and total polyunsaturated fatty acid (PUFA) range was 32.7-46.0%. 2n-6/3n-3 ratios in roselle (8.47) and smooth loofah (11.1) were in the neighborhood of the recommended 5-10; the value in sesame (186) was far above. Statistical analysis results, using Chi-square with critical value set at $\alpha_{= 0.05}$ showed that significant difference was observed only in palmitic, lignoceric and linoleic acids. However, significant difference existed in energy contributions by all the fatty acid groups: SFA, MUFA and PUFA among the three samples. The samples could therefore be considered as potential sources of palmitic, oleic and linoleic acids; they contain high concentrations of unsaturated fatty acids.

Keywords: Fatty acids, sesame, roselle, loofah

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INTRODUCTION

Sesame (*Sesamum indicum*) belongs to the family Pedaliaceae and genus sesamum. Sesame seeds vary in size, colour, depending on the variety; the seeds from northeast region of India are black; those from eastern region are brown to black and from south are predominantly red while those from other regions are white (Bisht *et al*., 1998). It is considered to be the oldest oilseed crop known to man, domesticated well over 5000 years ago. It is widely distributed in the tropical parts of Africa and Asia (Saydut *et.,* 2008); Ethiopia was said to be its original home (Bisht *et al*., 1998). Banerjee & Kole (2009) reported that the major sesame producers are India, Myanmar, China and Sudan producing about sixty-eight percent of the total world consumption. The seeds are used for the production of oil, paste, salads and in various food formulations. The traditional Indian medical practice of *Ayurveda* uses sesame oil to pacify stress-related symptoms (Cain & Shelton, 2001). Sesame seed oil (or til oil) is rich and of high nutritive quality and stability (Nayar & Mehra, 2002). In the eastern parts of the world, sesame has long been considered as a 'health food' that provides high energy and prevents ageing (Yoshida *et al*., 2007). It is said to be plant breeder's dream crop because of its great genetic diversity (Banerjee & Kole, 2009).

Roselle (*Hibiscus sabdariffa*) belongs to the plant family *Malvaceae* and a species of *Hibiscus* native from Asia (India to Malaysia) (Mahadevan *et al*., 2009). It is common in warm countries such Malaysia, Nigeria India and Saudi Arabia; in Nigeria, it is popularly called *Zobo*. The plant is primarily cultivated in most places for the production of bast fibre, used as a substitute in binlap production (Rost & Weier, 1979). Roselle is an important and popular medicinal plant; roselle seeds are used as laxative, tonic and in treating debility (Perry, 1980). The leaves are emollient and are used in Guinea as a diuretic, refrigerant and sedative (Anhwange *et al*., 2006). In nutrition, the seeds are subjected to solid-state fermentation process to produce a meat substitute condiment called 'furudu' in Sudan (Yaquob *et al*., 2004).

Smooth loofah (*Luffa cylindrical*) is a member of Cucurbitaceae family; it looks like cucumber. The leaves are oval, simple, five to seven lobed- dentate, dark green and frequently hairy. Loofah (in English) refers to the fruits of the two species *Luffa aegyptiaca* and *Luffa acutangula*. *Luffa aegyptiaca* grows well in a well-drained soil that is rich in organic waste. The fruits are smooth and cylindrical shaped with white flesh. The fruits of the two species are eaten at young stage as vegetables. The fully developed fruits are used as scrubbing sponge in bathrooms and kitchens.The loofahs are also used for filters and for stuffing pillows, saddles and slippers. United States being the major importer of loofahs from Asia, there is an increasing interest in domestic production (Dupriez & Leener, 1989). Tanobe *et al*. (2005) reported that the fibre of Brazilian sponge-gourds was chemically characterized in order to improve the fibre properties so as to enhance its large scale commercial utilization. There is little information on the nutritional profile, especially fatty acids of these samples: sesame, roselle and smooth loofah thereby making them suffer neglect in many parts of the world. The present study therefore aims at investigating their fatty acids composition. It is hoped to update the composition table of food ingredients.

MATERIALS AND METHODS

Sample Preparation

Sesamum indicum, *Hibiscus sabdariffa* and *Luffa cylindrical* seeds were purchased from various local markets in Ado-Ekiti, Ekiti State of Nigeria and were later identified in Plant Science Department, Ekiti State University, Ado-Ekiti. The samples were screened to eliminate the defective ones; they were sun-dried properly, decorticated, dry-milled, sieved and packed separately in polythene bags and stored prior to use. All the analyses were carried in duplicate.

Determination of Ether Extract

An aliquot (0.25g) of each sample was weighed in an extraction thimble and 200ml of petroleum ether (40-60°C boiling range) was added. The thimble containing the sample was extracted for five hours with the aid of Soxhlet extractor. The extracted fat was removed from the heating mantle when it was almost free of petroleum ether; oven dried at 105°C for one hour, cooled in a desiccators and the weight of dried oil was determined.

The Fatty Acid Methyl Ester Analysis

50mg of the extracted fat content of the sample was saponified for five minutes at 95°C with 3.4ml of 0.5MKOH in dry methanol. The mixture was neutralized by using 0.7 MHCl. 3ml of 14% boron trifluoride in methanol was added. The mixture was heated for five minutes at 90°C for complete methylation. The fatty acid methyl esters were thrice extracted from the mixture with redistilled n-hexane. The content was concentrated to 1ml for gas chromatography analysis and 1µL was injected into the injection port of gas chromatograph (GC).

GC Conditions for the Analysis of Fatty Acid Methyl Esters. GC: HP6890 powered with HP ChemStation rev. A 09.01[1206] software; Injection Type: Split Injection Split Ratio: 20:1; Carrier Gas: Nitrogen, Inlet Temperature: 250°C, Column Type: HP INNOWAX,Column Dimensions: 30m x 0.25mm x 0.25µm

Oven Programme. Initial temperature at 60°C. First ramping at 10°C/min for 20min, maintained for 4min. Second ramping at 15°C/min for 4min, maintained for 10min; Detector: FID (Flame ionization detector) Detector Temperature: 320°C; Hydrogen Pressure: 22 psi, Compressed Air: 35 psi

Statistical Analysis

Mean, standard deviation (SD) and coefficient of variation percent (CV%) were determined (Oloyo, 2011). Chi-square

(χ^2) values were also calculated by setting them at critical value of $a_{= 0.05}$. This is to see if significant differences existed in the fatty acid values among the samples.

RESULTS AND DISCUSSION

The percent levels of various fatty acids in sesame (*sesamum indicum*), roselle (*Hibiscus sabdariffa*) and smooth loofah (*Luffa cylindrical*) are presented in Table 1. Osborne & Voogt (1978) reported that major portion of man's energy supplies was provided by fat, providing more than twice as much energy as proteins or carbohydrates. The following fatty acids recorded 0.00% (of total fatty acid): lauric acid (sesame and smooth loofah), margaric acid (sesame, roselle and smooth loofah), myristic acid, lignoceric acid, palmitoleic acid, erucic acid and arachidonic acid (sesame). Low levels were recorded for the following fatty acids (% total fatty acid): lauric acid in roselle (0.116); myristic acid in roselle and smooth loofah (0.190 and 0.729 respectively); arachidic acid in sesame (0.688); behenic acid in all the samples (0.207-0.421); palmitoleic acid in roselle and smooth loofah (0.574 and 0.598 respectively); erucic acid in smooth loofah (0.691); a-linolenic acid sesame (0.246). Among the saturated fatty acids (SFA), palmitic acid (16:0) had the highest concentrations with values ranging from 8.77-25.5%. Several literature evidences indicated that palmitic acid is the most abundant SFA in nature. High levels of palmitic acid in this study compared well with the following reports: groundnut seeds (10.1-13.0%) (Adeyeye & Agesin, 2012); melon seeds flour (9.54-10.8%) (Adeyeye & Olaleye, 2015); millet, sorghum, rice and maize (10.9-21.0%) (Adeyeye & Ajewole, 1992); *Clarias gariepinus*, *Chana striatus* and *Tilapia zillii* (22.3-23.5%) (Olaleye *et al*., 2014); skin and muscle of *Oreochromis niloticus* (27.8-28.3%) (Adeyeye, 2011). The fat around the heart is highly saturated due to high concentration of palmitic and stearic acids (Daley *et al*., 2010). Though, both palmitic and stearic acids elevate serum cholesterol (Hegsted *et al*., 1993) but stearic acid may not be hypercholesterolemic as other SFAs due to its conversion to oleic acid (monounsaturated fatty acid) (Bonanome & Grundy, 1988). Some beneficial effects had been reported for saturated fatty acids in nutrition. Watkins *et al*. (1996) showed that for calcium to be incorporated into the skeletal structure, at least 50% of the dietary fats should be saturated. Nanji *et al*. (1995) indicated that SFAs protect the liver from negative effects of alcohol and other toxins and also enhance the immune system (Kabara, 1978). Among the monounsaturated fatty acids (MUFAs), oleic acid (18:1) had the highest concentrations in all samples (22.2-38.8%). Kris-Etherton (1999) showed that diets high in MUFA improves glycemic control in individuals with non-insulin dependent diabetic mellitus who maintain normal body weight and individual with elevated insulin also may benefit from a high monounsaturated fatty acid diets. The levels of C18:1 in this study were higher than those reported for three varieties of melon seeds flour (8.44-11.5%) (Adeyeye & Olaleye, 2015); edible larva of *Cirina forda*

(13.9%) (Akinnawo & Ketiku, 2000); three fresh water fish samples (11.1-13.7%) (Olaleye *et al*., 2013); *Vigna subterranean* (6.69-9.16%) (Adeyeye *et al*., 2015). However, the other two MUFAs recorded low concentrations: palmitoleic acid (0.00-0.598%); erucic acid (0.00-3.10%). The most concentrated polyunsaturated fatty acid (PUFA) was linoleic acid (18:2, n-6) in each of the samples with range of values from 25.0-45.8%. These values were higher than the following literature reports: larvae of three insect samples (14.0-22.8%) (Olaleye *et al*., 2017); *Vigna subterranean* seed parts (13.9-20.9%); (Adeyeye *et al*., 2015); *Cirina forda* larva (8.1%) (Akinnawo & Ketiku, 2000). The concentration of α-linolenic acid (18:3, n-3) ranged from 0.246-2.95%. These values were comparable with 1.17-1.30% reported for fresh water fish samples (Olaleye *et al*., 2014) and 1.64-1.81% for melon seeds flour (Adeyeye & Olaleye, 2015); higher than 0.010-0.041% for insects larvae (Olaleye *et al*., 2017). α-linolenic acid was reported to show significant anti-coronary heart disease effects in humans, both in clinical and epidemiological studies (Hayes, 2002). Generally, polyunsaturated fatty acids have been shown to lower moderately serum cholesterol and low density lipoprotein (LDL) cholesterol levels (WHO/FAO, 1994) and their deficiencies could result in kidney and liver disorders.

Table 2 presents the summary of fatty acids into saturated, monounsaturated and polyunsaturated fatty acids (i.e SFA, MUFA and PUFA respectively). The calculated MUFA/SFA, PUFA/SFA and 2n-6/3n-3 ratios are also shown in Table 2. Total SFA in the samples were in the range 15.1-41.4% and the highest concentration was recorded for *Hibiscus sabdariffa* (41.4%). Literature values for SFA were (%): three varieties of melon seeds flour (14.9-22.2) (Adeyeye & Olaleye, 2015); Thai edible insects (26.4-35.4) (Li-Feng *et al*., 2006); African yam bean seeds flour (50.6-57.1) (Adeyeye *et al*., 1999). The levels of MUFA were *Sesamum indicum* (38.8%), *Hibiscus sabdariffa* (25.9%) and *Luffa cylindrical* (28.8%). MUFA/SFA in this study ranged from 0.626-2.57; only *Sesamum indicum* had a value (2.57) greater than 1.00% (i.e. MUFA>SFA); others fell below 1.00. The relative proportion of SFA to PUFA in the samples ranged from 0.790-3.05. Only *Hibiscus sabdariffa* had the least value of less than 1.00 (0.790); others had higher PUFA than SFA. PUFA/SFA ratio is important in determining the detrimental effects of dietary fats; the higher the PUFA/SFA value, the more nutritionally useful is the dietary oil (Honatra, 1974). The present levels would prevent atherosclerosis tendencies (Honatra, 1974). The ratios of 2n-6/3n-3 in this study were in the range 8.47-186. Only *Sesamum indicum* had value (186) far above the recommended 5-10 (WHO/FAO, 1994). Too much omega-6 (2n-6) fatty acid in the diet creates an imbalance that can interfere with production of important prostaglandins (Kinsella, 1988).

The energy contributions by various fatty acid groups are shown in Table 3. SFA had the following contributions (kJ/100g). *Sesamum indicum* (559), *Hibiscus* *sabdariffa* (1532) and *Luffa cylindrical* (1132). The contributions of MUFA were: *Sesamum indicum* (1436), *Hibiscus sabdariffa* (958) and *Luffa cylindrical* (1066). For PUFA, it was 1702, 1210 and 1469 for *Sesamum indicum*, *Hibiscus sabdariffa* and *Luffa cylindrical* respectively. Lasserre *et al*. (1985) indicated that intake of polyunsaturated fatty acids by humans should not be more than 4% of the caloric total.

The statistical analysis of the samples using Chi-square (setting critical value at $a_{= 0.05}$) showed that in fatty acids composition (Table 1), significant difference existed among the samples only in palmitic, lignoceric and linoleic acids; in other fatty acids, no significant difference existed among the samples. In fatty acid groups (Table 2), significant difference was observed only in SFA and 2n-6/3n-3 while in energy contributions (Table 3), significant difference existed among the samples in all the parameters determined.

CONCLUSION

The results of this study showed that the samples contained unequal distribution of most of the parameters determined. The samples had palmitic and stearic acids as the predominant saturated fatty acids. Unsaturated fatty acids (UFAs) in the samples were significantly higher than saturated fatty acids with oleic and linoleic acids forming the bulk of the UFAs. With the exception of *Sesamum indicum*, 2n-6/3n-3 ratios in the samples were in the vicinity of 5-10 recommended ratios. The consumption of the samples should therefore be encouraged as they are capable of reducing the risk of heart related diseases due to high levels of UFAs.

Table 1 Fatty acid composition (%) of sesame, roselle and smooth loofah seeds flour

Fatty acid	SS	RS	SL	Mean	SD	CV%	χ^2	R
Lauric acid	0.00	0.116	0.00	0.039	0.067	172	0.230	NS
Myristic acid	0.00	0.190	0.729	0.306	0.378	124	0.935	NS
Palmitic acid	8.77	25.5	16.4	16.9	8.38	49.6	8.30	S
Stearic acid	5.45	3.94	5.48	4.96	0.881	17.8	0.313	NS
Margaric acid	0.00	0.00	0.00	0.00	0.00	0.00	0.00	NS
Arachidic acid	0.688	2.76	2.70	2.05	1.18	57.6	1.36	NS
Behenic acid	0.207	0.404	0.421	0.344	0.119	34.6	0.082	NS
Lignoceric acid	0.00	8.50	4.83	4.44	4.26	95.9	8.19	S
Palmitoleic acid	0.00	0.574	0.598	0.391	0.339	86.7	0.586	NS
Oleic acid	38.8	22.2	27.5	29.5	8.48	28.7	4.87	NS
Erucic acid	0.00	3.10	0.691	1.26	1.63	129	4.20	NS
Linoleic acid	45.8	25.0	32.3	34.4	10.6	30.8	6.47	S
α-linolenic acid	0.246	2.95	2.92	2.04	1.55	76.0	2.36	NS
Arachidonic acid	0.00	4.78	4.48	3.09	2.68	86.7	4.64	NS
Total	100	100	99.0	99.7	0.577	0.006	0.007	NS

SS = sesame; RS = roselle; SL = smooth loofah; SD = standard deviation; CV% = coefficient of variation percent; χ^2 = Chi- square; NS = not significant; S = significant; R = remark

Table 2 Summary of fatty acids into SFA, MUFA and PUFA (% total fatty acid)

Fatty acid	SS	RS	SL	Mean	SD	CV%	χ^2	R
SFA	15.1	41.4	30.6	29.0	13.2	45.5	12.1	S
MUFA	38.8	25.9	28.8	31.2	6.77	21.7	2.94	NS
PUFA	46.0	32.7	39.7	39.5	6.65	16.8	2.24	NS
MUFA/SFA	2.57	0.626	0.941	1.38	1.04	75.4	1.58	NS
PUFA/SFA	3.05	0.790	1.30	1.71	1.19	69.6	1.64	NS
2n-6/3n-3	186	8.47	11.1	68.5	102	149	302	S

Table 3 Energy contributions by the various fatty acid groups

Fatty acid		SS	RS	SL	Mean	SD	CV%	χ^2	R
SFA	kcal	134	366	271	257	117	45.5	106	S
	KJ	559	1532	1132	1074	489	45.5	445	S
MUFA	kcal	343	229	255	276	59.7	21.6	25.9	S
	KJ	1436	958	1066	1153	251	21.8	109	S
PUFA	kcal	407	289	351	349	59.0	16.9	20.0	S
	KJ	1702	1210	1469	1460	246	16.8	83.0	S

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