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Research Article
Comparative Studies on the Lipid Profile of Shea (*Vitellaria paradoxa* C.F. Gaertn.) Fruit Kernel and Pulp

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Abstract

Background and Objective: The importance of shea tree (*Vitellaria paradoxa*) has led to the continuous investigation of the shea products (kernels and butter). This study examines comparatively the levels of fatty acids, phospholipids and sterols in the samples of *Vitellaria paradoxa* fruit kernel and pulp. **Materials and Methods:** The fatty acid, phospholipid and sterol compositions were determined from the kernel and pulp samples prepared from the fresh fruits of *Vitellaria paradoxa* using gas chromatography method. **Results:** The quality parameters of fatty acids investigated in the kernel and pulp samples were: SFA (16.14 and 16.33%); MUFA (57.37 and 52.27%); PUFA (26.49 and 31.40%); MUFA/SFA (3.56 and 3.20); PUFA/SFA (1.64 and 1.92); EFA (26.43 and 31.20%); oleic/linoleic (2.27 and 1.67), respectively. The total phospholipids present in the kernel and pulp were 102.33 and 58.11 mg/100 g, respectively. Phosphatidylethanolamine content was the highest in both samples. The concentrations of phytosterols were of low values except in sitosterol (273.59 and 38.49 mg/100 g) and stigmasterol (90.26 and 12.16 mg/100 g) for the kernel and pulp samples, respectively. **Conclusion:** The result revealed that pulp sample was richer in EFA and PUFA/SFA ratio compared to that of kernel while the values recorded for phosphatidylserine for the two samples were too low and do not meet USFDA standards.

Key words: Fatty acids, phospholipids, phytosterols, *Vitellaria paradoxa* fruit

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Shea (*Vitellaria paradoxa* C.F. Gaertn) is of the family Sapotaceae and divided into two sub-species: *Niltica* and *Paradoxa*[1]. The shea tree is indigenous to the savanna belt in sub-saharan Africa, extending across 19 countries from Mali in the west to Ethiopia and Uganda in the east which is from 16°W to 34°E longitude and 1°N to 15°N latitude[2-3]. The difference between the two sub-species occurs primarily in the consistency of the butter content found within the nut[4]. Shea is considered a sacred tree by many communities and ethnic groups and plays important roles in religious and cultural ceremonies where it is also believed to have some spiritual protective powers. It has been claimed to possess potentials to improve nutrition, boost food supply in the annual hungry season, foster rural development and support sustainable land care[5].

Many works have been reported on the studied samples such as prospects for the development of the shea nut industry in Ghana[6], triterpene alcohol and fatty acid composition of shea nuts from 7 African countries[7], variation of the physical properties of sea nut (*Vitellaria paradoxa* Gaertn) kernels during convective drying[8], evaluation of some physicochemical properties of shea-butter (*Butyrospermum paradoxum*) related to its value for food and industrial utiltzation[9], regional variation in shea butter lipid and triterpene composition in 4 African countries[10], fatty acids and triglycerides of Cameroon shea butter[11], evaluation of the quality of west African shea butter (*Vitellaria paradoxa*)[12], nutritional values and indigenous preferences for shea fruits (*Vitellaria paradoxa* C.V. Gaertn. F) in African agroforestry parklands[13], development of mechanical expression rig for dry extraction of shea better from shea kernel[14], the non-glyceride saponifiables of shea butter[15], nutritional composition of shea (*Vitellaria paradoxa*) fruit pulp across its major distribution zones in Nigeria[16], nutrient and anti-nutrient composition of shea (*Vitellaria paradoxa*) kernel and pulp in the north east, Nigeria[17] and compositional and toxicological studies on shea butter[18]. Thus, most information available in the published articles has not reported comparable composition of phytosterols and phospholipids of shea fruit kernel and pulp.

Therefore, this study is aimed at assessing the fatty acids, phospholipids and phytosterols composition of shea (*Vitellaria paradoxa* C.F. Gaertn.) fruit kernel and pulp in north-east, Nigeria as this will encourage their maximum and wider utilization as part of everyday human diet.

MATERIALS AND METHODS

Collection and treatment of samples: Some fresh shea fruits were collected from a farmer at Wukari local government area, north-east Nigeria sometime in July, 2017. The fruits were de-pulped and the fruit pulp, comprising the epicarp and mesocarp were oven-dried at 45°C to a constant weight for 6 days. The de-pulped seeds of the shea fruit were cracked to remove the kernel which was also oven-dried at the same temperature of 45°C for 4 days. The two different dried samples were finely milled for laboratory analyses.

Ether extract: A quantity of 5 g each of oven dried sample of shea (*Vitellaria paradoxa*) fruit kernel and pulp was extracted for 5 h in Soxhlet apparatus with 250 mL of petroleum ether (40-60°C boiling range) of analytical grade (British Drug Houses, London). The extraction flask was removed from the heating mantle when it was almost free of petroleum ether, oven dried at 105°C for 1 h, cooled in a desiccator and used for further analyses[19].

Preparation of fatty acid methyl esters and analysis: The oil extracted from each sample was converted to the methyl ester using the method described by Adeyeye and Adesina[20]. A 50 mg aliquot of the dried oil was saponified for 5 min at 95°C with 3.4 mL of 0.5 M KOH in dry methanol. The mixture was neutralized by 0.7 M HCl and 3 mL of 14% boron trifluoride in methanol was added. The mixture was heated for 5 min at 90°C to achieve complete methylation. The fatty acid methyl esters were analyzed using a HP gas chromatograph [HP gas chromatograph powered with HP ChemStation rev a09.01 (1206) software (GMI, Inc., Minnesota, USA)] fitted with a flame FID and a computing integrator while nitrogen was used as the carrier gas. The injection port and the detector were maintained at 310 and 350°C, respectively while the column initial temperature was 250°C rising at 5°C min^{-1} to a final temperature of 310°C. A polar (HP INNO Wax) capillary column (30, 0.53 mm and 0.25 μm) was used to separate the esters. The peaks were identified by comparison with standard fatty acid methyl esters obtained from Sigma Chemical Co., (St. Louis MO, USA). However, the quantitative evaluation was carried out on the base of gas chromatography peak areas of the different methyl esters. The heptadecanoic ester was used to calculate the response factor for fatty acids which was found to be 0.96. Three determinations were made for each sample.

Phospholipids analysis: The phospholipids content of shea (*Vitellaria paradoxa*) fruit kernel and pulp oils was determined by gas chromatography (GC) as it was earlier described by Adeyeye and Adesina[20]. About 0.01 g of the extracted fats was added to the test tube. To ensure complete dryness of the oil for phospholipids analysis, the solvent was completely removed by passing the stream of the nitrogen gas on the oil. About 0.04 mL of chloroform was added to the content of the tube and it was followed by the addition of 0.10 mL of chromogenic solution. The content of the tube was heated at a temperature of 100°C in a water bath for about 1 min. The content was allowed to cool, 5 mL of the hexane was added and the tube with its content shook gently several times. The solvent and the aqueous layers were recovered and allowed to be separated. The hexane layer was recovered and allowed to be concentrated to 1.0 mL for gas chromatography using flame photometric detector. The conditions for phospholipid analysis include H.P 5890 powered with HP ChemStation REV. A 09.01 (1206) and split injection ratio of 20:1; nitrogen as carrier gas; inlet temperature, 250°C; column type, HP5; column dimension: 30, 0.25 mm and 0.25 µm; oven program: Initial temperature at 50°C; first ramping at 10°C min^{-1} for 20 min, maintained for 4 min while second ramping at 15°C min^{-1} for 4 min, maintained for 5 min. Detector: PFPD Detector temperature was 300°C; hydrogen pressure, 20 psi; compressor air: 35 psi.

Sterol analysis: The sterol analysis was done as described by AOAC[19]. The aliquots of the extracted fat were added to the screw-capped test tubes. The samples were saponified at 90°C for 30 min using 3 mL of 10% KOH in ethanol to which 0.20 mL of benzene had been added to ensure miscibility. Deionized water (3 mL) was added and 2 mL of hexane was added in extracting the non-saponifiable materials. Three different extractions, each with 2 mL of hexane were carried out for 1 h, 30 and 30 min, respectively. The hexane was concentrated to 1 mL in the vial for gas chromatography analysis and 1 µL was injected into the injection pot of GC. The GC conditions of analyses were similar to the GC conditions for methyl esters analyses.

Statistical evaluation: The descriptive statistical analysis done was the determination of mean, standard deviation and coefficient of variation percent.

RESULTS

The percentage composition of the various fatty acids in the samples of *Vitellaria paradoxa* is presented in Table 1.

Table 2 displays the summary of the quality parameters of fatty acids of both kernel and pulp samples of *Vitellaria paradoxa*.

Table 1: Fatty acid composition of shea (*Vitellaria paradoxia*) kernel fruit and pulp

Fatty acid (%)	Kernel	Pulp	Mean	SD	CV (%)	K-P	Difference (%)
Palmitic acid (C16:0)	12.07	9.78	10.93	1.62	14.82	+2.29	18.97
Margaric acid (C17:0)	0.030	0.095	0.063	0.046	73.54	-0.07	216.67
Stearic acid (C18:0)	3.07	4.19	3.63	0.79	21.82	-1.12	36.48
Arachidic acid (C20:0)	0.427	0.464	0.446	0.026	5.87	-0.04	8.67
Behenic acid (C22:0)	0.265	0.883	0.574	0.437	76.13	-0.618	233.20
Lignoceric acid (C24:0)	0.274	0.918	0.596	0.455	76.40	-0.644	235.04
Palmitoleic acid (C16:1)	0.152	0.506	0.329	0.250	76.08	-0.354	232.89
Oleic acid (C18:1)	57.03	51.12	54.08	4.18	7.73	+5.91	10.36
Erucic acid (22:1)	0.192	0.643	0.418	0.318	76.38	-0.451	234.90
Linoleic acid (18:2)	25.11	30.56	27.84	3.85	13.85	-5.45	21.70
α-linolenic acid (18:3)	1.32	0.641	0.981	0.480	48.97	+0.679	51.44
Arachidonic acid (20:4)	0.059	0.196	0.128	0.097	75.98	-0.137	232.20
Total	100	100					

SD: Standard deviation, CV: Coefficient of variation, K-P: Kernel-Pulp

Table 2: Quality parameters of shea (*Vitellaria paradoxia*) fruit kernel and pulp

Parameters	Kernel	Pulp	Mean	SD	CV (%)	K-P	Difference (%)
Total SFA	16.14	16.33	16.23	0.097	0.60	-0.194	1.20
Total MUFA	57.37	52.27	55.03	2.501	4.65	+5.105	8.90
Total PUFA	26.49	31.40	28.94	2.454	8.48	-4.908	18.53
DUFA	25.11	30.56	27.84	2.73	9.81	-5.45	21.70
Total UFA	83.86	83.67	83.76	0.097	0.12	+0.197	0.23
MUFA/SFA	3.56	3.20	3.38	0.18	5.33	+0.36	10.11
PUFA/SFA	1.64	1.92	1.78	0.14	7.87	-0.28	17.07
Total EFA	26.43	31.20	28.82	2.39	8.28	-4.77	18.05
O/L	2.27	1.67	1.97	0.30	15.23	+0.6	26.43

SD: Standard deviation, CV: Coefficient of variation, K-P: Kernel-Pulp, SFA: Saturated fatty acid, MUFA: Monounsaturated fatty acid, O/L: Oleic/linoleic ratio

Table 3: Phospholipids composition of shea (*Vitellaria paradoxia*) fruit kernel and pulp

Phospholipid (mg/100 g)	Kernel	Pulp	Mean	SD	CV (%)	K-P	Difference (%)
Phosphatidylethanolamine (PE)	60.78	33.49	47.14	19.30	40.94	+27.29	44.90
Phosphatidylcholine (PC)	32.84	20.77	26.81	8.53	31.84	+12.07	36.75
Phosphatidylserine (PS)	3.89	1.81	2.85	1.47	51.61	+2.08	53.47
Lysophosphatidylcholine (LC)	0.226	0.0446	0.14	0.13	94.73	-0.2205	97.70
Sphingomyelin (SM)	0.211	0.099	0.16	0.08	50.99	+0.11168	53.00
Phosphatidylinositol (PI)	4.37	1.87	3.12	1.77	56.66	+2.50	57.20
Phosphatidic acid (PA)	0.017	0.03	0.02	0.096	40.88	-0.014	81.33
Total	102.33	58.11	80.24	31.38	367.65	43.82	424.35

SD: Standard deviation, CV: Coefficient of variation, K-P: Kernel-Pulp

Table 4: Phytosterol composition of shea (*Vitellaria paradoxia*) fruit kernel and pulp

Phytosterol (mg/100 g)	Kernel	Pulp	Mean	SD	CV (%)	K-P	Difference (%)
Cholesterol	0.0725	0.0723	0.0724	0.0014	0.20	+0.0002	0.28
Cholestanol	0.0587	0.0587	0.0587	0	0.00	0	0.00
Ergosterol	0.0178	0.0178	0.0178	0	0.00	0	0.00
Campesterol	26.63	6.50	16.57	14.23	85.93	+20.13	75.59
Stigmasterol	90.26	12.16	51.21	55.23	107.84	+78.1	86.53
5-Avenasterol	6.86	1.40	4.13	3.86	93.48	+5.46	79.59
Sitosterol	273.59	38.49	156.04	166.24	106.54	+235.1	85.93
Total	397.49	58.70	228.10	239.56	393.99	338.79	327.92

The levels of various phospholipids in the two samples are shown in Table 3.

The phytosterol composition in *Vitellaria paradoxa* fruit kernel and pulp samples is presented in Table 4.

DISCUSSION

The most concentrated SFA in both samples was palmitic acid (C16:0) with values of 12.07% (kernel) and 9.78% (pulp) followed by stearic acid (C18:0) with values of 3.07% (kernel) and 4.19% (pulp). The values recorded for both palmitic and stearic acids in the present study are in close agreement with the results of palmitic and stearic acids of seed samples from literature[21-23]. Margaric acid, arachidic acid, behenic acid and lignoceric acid are all saturated fatty acids which were present in small quantities with none of them recording up to 1.0% in both samples. The most pre-dominant MUFA was oleic acid (C18:1) with values of 57.03 and 51.12% for kernel and pulp samples, respectively. This is in agreement with the report of Aremu *et al.*[24] and Grosso *et al.*[25], that linoleic and oleic acids are major fatty acids in many plant seeds such as peanut, soybean, broad bean, lentil, cashew nut and garden pea. Oleic acid has been regarded as MUFA and has been shown to decrease HDL-cholesterol concentrations which affect positively cardiovascular disease risk[26]. The values of oleic acid recorded for both samples in the present study are comparable with the value reported for *Artocarpus altilis* (56.78%)[23]; however higher than the values reported for African locust bean and mesquite bean (32.24 and 30.96%, respectively)[27]. Oleic acid is the biosynthetic precursor of a family of fatty acids with the (n-9) terminal structure and with chain length of 20-24 or more carbon atom[21]. The linoleic acid (C18:2) values in both samples (25.11 and 30.56%) are comparable with *Luffa cylindrica*[28] and *Brachystegia eurycoma*[29] as reported by some researchers. The value of (C18:2) in the pulp (30.56%) is greater than that of kernel (25.11%) by 21.70% while the value of alpha linolenic acid (C18:3) in kernel (1.32%) is higher than that of pulp (0.641%) by 51.44% (Table 1). Linoleic acid plays a significant role in the skin. In dry skin, it strengthens the lipid barrier of epidemic, protects against transepidermal loss of water and normalizes the skin metabolism. Linoleic acid is natural component of sebum[30]. The LA and ALA have critical roles in the membrane structure and as precursors of eicosanoids, which are potent and highly reactive compounds. Since they compete for the same enzymes and have different biological roles, the balance between the LA and ALA fatty acids in the diet can be of considerable importance[31]. The CV (%) is highly varied and ranged from 5.87 in (C20:0) to 76.40 in (C24:0) (Table 1).

The SFA levels were 16.14 and 16.33% with a percentage difference of 1.20% in favour of pulp sample. The MUFA contents were 57.37% (kernel) and 52.27% (pulp) with a difference of 8.90% in favour of kernel. The total SFA contents in this report are lower than TSFA values of 24.80 and 20.50% reported for raw and boiled tigernut samples[32], 54.51% reported for dehulled African yam bean[33], 40.20 and 43.00% reported for African locust bean and mesquite bean, respectively[27], 34.68% reported for bambara groundnut[34]. However, the reported values of 12.3% for groundnut[35], 15.2%

for soybean[36] and 9.0-12.9% for processed pinto bean[37] are lower than the values reported in this work. The values revealed for the total UFA were 83.86 and 83.67% for kernel and pulp samples, respectively. They are higher than that of tigernut sample with 75.20% (in raw) and 79.50% (in boiled)[32], *Parkia biglobosa* with a reported value[38] of 33.69%, *Vigna subterranea* with a reported value[34] of 65.32% and mesquite bean with a reported value[27] of 56.90%. TUFA in this study is of great concern because report has shown that fats and oils with high unsaturation are particularly susceptible to oxidation and intakes of food containing oxidized lipid increase the concentration of secondary proxidation products of liver[39]. The high amount of TUFA makes kernel and pulp of *Vitellaria paradoxa* as special fruit for nutritional applications. The ratios of MUFA/SFA and PUFA/SFA were 3.56 and 3.20; 1.64 and 1.92, respectively. These ratios are important in the determination of detrimental effects of dietary fats. The higher the PUFA/SFA ratio, the more nutritionally useful is the oil[33]. It has been reported that the severity of the disease condition such as atherosclerosis is closely associated with the proportion of total energy supplied by PUFA and SFA[40]. The ratio of oleic/linoleic (O/L) has been associated with high stability and potentiality of the oil for deep frying fat[27]. The O/L levels were 2.27 in kernel and 1.67 in pulp sample. These values are lower than that of *Anarcadium occidentale* (12.28%)[24], but compared with that of raw tigernut (2.11)[32] and *Artocarpus altilis* (2.21)[23]; hence, kernel and pulp oils of *Vitellaria paradoxa* may be stable compared with peanut oil and may be useful as frying oil. Linoleic acid contents (25.11 and 30.56%) constituted the DUFA while total NEFA gave 73.57 and 68.8% for the kernel and pulp samples, respectively.

The result showed that kernel sample was of higher phospholipid content than the pulp sample. The phosphatidylethanolamine (60.78 mg/100 g) and phosphatidylcholine (32.84 mg/100 g) showed greater concentrations in the kernel sample with difference (%) of 44.90 and 36.75 compared to the values recorded for pulp sample, respectively. Phosphatidylserine and phosphatidylinositol values (3.89 and 4.37 mg/100 g) in kernel sample are also higher compared with that of pulp sample (1.81 and 1.87 mg/100 g) with difference (%) of 53.47 and 57.20, respectively. The minor phospholipids in both samples were lysophosphatidylcholine, sphingomyelin and phosphatidic acid and none of them has concentration up to 1.0 mg/100 g. The values of phosphatidylethanolamine recorded in this work for the two samples are in agreement with the report of Wirtz[41], that phosphatidylethanolamine is usually the most abundant phospholipid in animals and plants, often amounting to almost 50% of the total and as such they are building block of membrane bilayer. The PE is found in all living cells; although in human physiology, it is found particularly in nervous tissue such as the white matter of brain, nerves, neural tissue and in spinal cord[42]. The low values recorded for phosphatidylserine and phosphatidic acid for both samples are of great concern. The US Food and Drug Administration (USFDA) have stated that consumption of PS may reduce the rate of dementia and cognitive dysfunction in the elderly people; in young people, it reduces mental stress and increases mental accuracy and stress resistance[43]. PS supplementation promotes a desirable hormonal balance for athletes and might reduce the physiological deteriorations that accompanies over training and/or overstretching[42]. Phosphatidic mediates cellular functions through different modes of action such as membrane tethering, modulation of enzymatic activities and structural effects on cell membranes. Processes in which phosphatidic plays a role include; signaling pathways in cell growth, proliferation, reproduction and responses hormones in biotic and abiotic stress[42]. However, consumption of this fruit (kernel and pulp) may not participate well in these functions. The coefficient of variation (%) varied from 31.84 in PC to 94.73 in LC (Table 3).

Phytosterols are natural components of plant origin forming cell membrane and occur in small quantities in many fruits, vegetables, nuts, seeds, cereals, legumes, vegetable oils and other plants. They are abundantly present in the fat soluble fractions of all the plants and food containing plant based raw materials including principally oils, cereals, pulse and dried fruits[44]. Systematic reviews studying the efficacy of phytosterols have shown that phytosterols enriched foods can significantly lower LDL cholesterol[45]. Plant sterols have also been described as anti-inflammatory and anti-cancer compounds[46]. The total phytosterol levels in the present study were 397.49 mg/100 g (kernel) and 58.70 mg/100 g (pulp) with the higher value being in kernel sample. The result showed that all the phytosterol components had higher concentration in kernel sample compared to pulp with difference (%) ranged from 0.28 in cholesterol to 86.53 in stigmasterol. This suggested that fruit kernel of *Vitellaria paradoxa* can be regarded as a better source of sterols when compared to the pulp sample. The CV (%) values ranged from 0.20 in cholesterol to 107.84 in stigmasterol.

CONCLUSION

The research has focused on the lipid composition of *Vitellaria paradoxa* fruit kernel and pulp. The results showed

that the total UFA was higher than the total SFA in kernel and pulp samples; thereby making the fruit fats good for human health. The quality parameters such as MUFA/SFA, PUFA/SFA and O/L were all observed to be good. The values recorded for PS (3.89 and 1.81 mg/100 g) for both samples were too low and do not meet the USFDA standards. However, the result of phytosterol composition showed that both samples may be a good source of phytosterol. The lipid analysis of *Vitellaria paradoxa* fruits collected from north-east Nigeria will add to the available food composition table.

SIGNIFICANCE STATEMENT

This study discovered that kernel and pulp samples have high values of UFA that make *Vitallaria paradoxa* as special fruit for nutritional applications. The contents of phosphatidylethanolamine and phosphatidylcholine that were revealed in this study will be useful to the researchers for their main applications in drug delivery systems. Likewise the quantitative analysis of the sterol composition will provide baseline information for future research on *Vitellaria paradoxa* fruits and other similar plants.

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