

# Vitamin E Content of Traditionally Processed Products of Two Commonly Consumed Oilseeds - Groundnut (*Arachis Hypogea*) and Melon Seed (*Citullus Vulgaris*) in Nigeria

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## Abstract

The levels of vitamin E after processing of melon (*Citrullus vulgaris*) seed and groundnut (*Arachis hypogea*) into some of their commonly consumed forms in Nigeria, was investigated.

Raw melon seeds were processed into roasted melon, fermented melon (*ogiri*), defatted fried cake (*robo*) and melon seed oil, using available traditional methods. Raw groundnut was also processed using traditional methods, into; roasted groundnut, fried groundnut cake (*kulikuli*) and groundnut oil. The raw and processed products were analyzed for vitamin E using a RP-HPLC (Reverse Phase-High Performance Liquid Chromatography) system after the samples had been saponified and the vitamin extracted from them.

Results showed that vitamin E levels in the processed melon seed products were less than that in the raw melon, with the exception of the melon oil which had the highest amount of vitamin E in this group (16.1 mg/100g, 3.9 mg/100 g, 12.0 mg/100g, 6.1 mg/100g and 20.1 mg/100g for raw melon, roasted melon, fermented melon seeds (*ogiri*), defatted fried cake (*robo*), and melon oil respectively). There was a statistically significant difference in the level of vitamin E in the raw melon compared with the levels in the processed products ( $p < 0.05$ ) except the melon oil which was not. In the groundnut products, the vitamin E levels were higher in the processed products than in the raw groundnut: 8.9 mg/100g, 16.7 mg/100g, 13.1 mg/100g and 48.1mg in the raw groundnut, roasted groundnut, defatted fried cake (*kulikuli*), and groundnut oil, respectively. The difference in vitamin E levels in the raw and processed groundnut products was found to be statistically significant ( $p < 0.05$ ). Interestingly, groundnut and melon seed oils were found to have the highest vitamin E content.

This study showed that groundnut and melon seeds are good sources of vitamin E, most especially the oils of these seeds.

**Keywords:** Vitamin E (tocopherol); Oilseeds; Groundnut; Melon seed; Processing

## Introduction

Vitamin E is a term used for a group of related compounds, which have been isolated from plant materials especially plant oils [1]. It is an important micronutrient in human nutrition. This is because it functions as an antioxidant together with other antioxidants like vitamin A and C, copper, selenium and zinc [2]. These antioxidants help to fight free radicals in the body. Vitamin E is also involved in the metabolism of all cells and protects vitamin A and essential fatty acids from oxidation in the body cells and prevents breakdown of body tissues. Foods rich in vitamin E have been reported to protect the body from non-communicable diseases such as cancer, coronary heart disease among others [3].

Plant sources especially vegetable oils and products made from them are the richest sources of vitamin E [1]. In Nigeria, groundnut (*Arachis hypogea*) and melon seeds (*Citrullus vulgaris*) are among the most popularly consumed plant foods that are oilseeds [4,5]. They are processed and prepared in different ways by virtually all-ethnic groups in Nigeria and they form part of many of the Nigerian traditional food products, diets and snacks.

Groundnuts are a high value crop that can be marketed with little processing, but are extremely versatile and can be used in a wide range of products. It is the third most abundantly cultivated oilseed in the world and over the years it has played an important role in the economy of Nigeria [6]. They are also the third most important source of vegetable protein [7]. In Nigeria, they are primarily consumed as roasted or

boiled whole nuts or processed to extract the oil [8]. In the eastern part of Nigeria, groundnuts are made into a local peanut butter called '*osé orji*' and eaten with garden eggs. In northern Nigeria, groundnut is popularly made into a snack called *kulikuli*. This is a defatted fried cake obtained after oil has been extracted from the groundnut.

Melon seeds (*Citrullus vulgaris*) are very popular as a condiment in Nigerian local soup. The melon plant is grown in all regions of Nigeria. The seeds are consumed within the region in which they are produced. Melon seeds can be obtained in the local markets under the names of '*egusi*' (Yoruba), '*ogili*' (Ibo), '*ogi*' (Benin), and '*iguana agushi*' (Hausa) and '*dènde*' (Fulani) [9]. The residual protein is fried into a protein rich cake known as *igbalo* [10] or *robo* which is consumed as snack. The melon seed is also used in the production of a popular fermented condiment called *egusi* (Yoruba), the production of which is largely on a traditional small-scale, house-hold basis under highly variable conditions [11,12]. This condiment is also known to contribute to the

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caloric and protein intake and is generously added to soups as low-cost meat substitute by low-income families in parts of Nigeria [11].

Several studies have reported on the nutritive value of groundnut and melon seeds and their products in Nigeria [11-15] but little attention has been given to their vitamin E content. Therefore determining the level of vitamin E in groundnut and melon seeds and their products are important because of the high consumption level in Nigeria, as well as interest in dietary standards, nutrient requirements, and antioxidant potential of foods with vitamin E and the reported protective effect of this vitamin. This is in the light of the current burden of non-communicable diseases which is on the increase in Nigeria and also for those involved in giving dietary advice.

Thus the aim of this study was to investigate the levels of vitamin E in raw and traditionally processed groundnut and melon seed products. They include roasted groundnut, *kulikuli* (fried groundnut cake) groundnut oil, roasted melon seed, *robo* (defatted fried cake) and melon seed oil.

## Materials and Methods

### Sample Preparation

Groundnut (*Arachis hypogea*) and melon seeds (*Citrullus vulgaris*) were purchased at a local market in Ibadan (Bodija market), Oyo state in Nigeria. The processing/ preparation of the groundnut and melon seeds into their different products; roasted groundnut, groundnut oil and fried groundnut cake (*kulikuli*) and roasted melon, fermented melon (*ogiri*), melon oil (*ororo egusi*) and fried melon cake (*robo*), respectively were carried out according to the available folk methods (tradition and local methods). All the products were prepared from the same batch of raw samples purchased i.e., of both groundnut and melon seeds.

### Production of roasted groundnut

Shelled raw groundnut (household measure; 8 milk tins) was put in a tray and sorted to remove stones, dirt and grit. It was put into a bowl containing tap water, for three minutes. The groundnut was stirred with the hands while still in water and afterward drained using a sieve. The groundnut was then salted to taste. The groundnut was left that way in the sieve till the following morning.

Groundnut was roasted locally using clean hot sand, in an open aluminium pot and local fire made with firewood. The groundnut was poured into the sand and was constantly stirred using a wooden spoon to ensure uniform roasting and that it does not get burnt. The roasting lasted for about 30 minutes. The groundnut was separated from the sand using a raffia basket and then left to cool by spreading it out on a flat surface. It was afterward dehulled manually by rubbing gently with the hands and then winnowed. The clean roasted groundnuts was bottled and stored in a cool dry and dark cupboard.

### Production of groundnut oil and fried groundnut cake (*kulikuli*)

Shelled groundnut (household measure; 10 milk tins (one 'congo')) was first sorted to remove stones and dirt. It was then roasted in a dry open aluminium pot using charcoal fire, for about 30 minutes. It stirred with a spoon at intervals to prevent it from burning and also to ensure uniform roasting. Groundnut was butter color after roasting. It was allowed to cool, and then skinned and winnowed. It was taken to a local mill and ground to paste without adding of water.

A little quantity of water (about ¼ of a liter) was warmed in a round bottom aluminium pot and the groundnut paste was added to it and stirred using a wooden stick for 5 minutes on low heat (heat of charcoal fire was reduced by sprinkling some water on it). The pot was brought down from the fire and the stirring continued. At intervals, warm water was again sprinkled on the paste while stirring continued until the oil began to flow out gradually. As oil flowed out, the cake or residue became harder. The oil was collected into a plate and the semi hard groundnut cake was moulded into desired shapes and fried in the extracted oil. The whole process was completed in about 3 ½ hours. The oil and *kulikuli* were kept in dry bottles.

### Production of roasted melon seeds

Shelled melon seeds were sorted and cleaned to remove dirt and bad seeds. A clean pot was put on a hot plate to dry and the seeds were poured into the pot. The melon was stirred in the pot constantly until all the seeds were moderately brown and evenly roasted. This took about 5 minutes. It was bottled and kept in a cool dry and dark cupboard until time for analysis.

### Production of fermented melon seeds (*ogiri*)

This *ogiri* was prepared using the method of the indigenous producers of *Ivwe-Uromi* in Edo State. Shelled and already sorted melon seeds (household measure; 2 milk tins) were spread on a tray and sun dried for a day. The seeds were reduced to a coarse mill by pounding in a wooden mortar. Two liters of water was put into an open pot and brought to boil using local fire (firewood). The pounded seeds were put into the pot of boiling water and left to boil leaving the pot uncovered, while stirring with a spoon at intervals. It was left to boil for several hours until water was completely dried such that when cooked paste was squeezed with the hand, the only liquid that came out was the oil.

After the melon had cooled, it was transferred into a stainless steel plate, covered lightly to allow for minimum air and then kept in a warm corner of the kitchen for four days to ferment. On the fourth day, the fermented melon was further reduced to a smoother fine paste using the local miller. It was moulded into a big bolus and kept on a tray in a slanting position till the following day, so that the oil could flow out and be collected.

On the fifth day, the fermented melon was cut into bits and wrapped in clean steamed banana leaves. The oil collected from it was sprinkled over the wrapped leaves, which were then further wrapped in a bigger package together such that it would not receive direct heat and steamed over dry heat for another day, before being ready for use. It was stored in an ordinary freezer until time for analysis.

### Production of melon seed oil (*ororo egusi*) and fried melon cake (*robo*)

Already sorted and cleaned melon seeds were roasted in an open dry pot using an electric cooker. They were turned constantly until they became very brown (10-15 minutes). The roasted seeds were left to cool and then taken to the mill. It was milled with a wet miller into paste without adding water. The resulting paste was kneaded with the hands in a bowl continuously, while adding little quantities of water at intervals. The kneading was done for about one hour after which the oil began to flow out and the cake or concentrate became harder. The oil was collected into a pot and the cake was rolled into small round balls and fried in the extracted oil. After the oil and fried cake had cooled, they were kept in covered bottles.

All prepared products apart from the fermented melon seeds, were

stored in a cool dry and dark cupboard. This was one of the precautions taken to prevent loss of the vitamin E. From literature, vitamin E is said to be destroyed by light and one of the ways to protect samples for the analysis of this vitamin, is to keep them away from light.

## Analytical Procedure

### Sample Preparation

A dry mill blender in the laboratory was used to reduce all dry samples to smooth flour. They were all kept in separate cellophane bags, labeled and put into an airtight container and kept away from light. The groundnut oil was bottled and kept in a cupboard away from light.

### Vitamin E Analysis

**Saponification and extraction:** Saponification involves the heating of the sample in a strong alkaline medium. This step is needed to remove the excess fatty acids before HPLC analysis (<http://www.cyberlipid.org/vitet/vite002>) i.e., it is used to split associated triglycerides and thus prepare a tocopherol concentrate (Ames 1972; 226).

**Apparatus:** Test tubes, analytical balance, measuring cylinder, 2 ml and 5 ml pipettes, heating mantle, mercury thermometer, oven (Gallenhamp Model 1H-100).

**Reagents:** KOH (60g of KOH dissolved in 100 ml d/w), absolute ethanol, ethanolic pyrogallol (6g in 100 ml absolute ethanol), normal/physiological saline (9g NaCl in 100 ml d/w), hexane, ethyl acetate, isopropanol.

**Procedure:** 2g of each sample were weighed into Pyrex test tubes in triplicates and they were arranged in a test tube rack. The samples were saponified by addition of equal volumes of KOH (60%, w/v) and 2 ml absolute ethanol and 5 ml of ethanolic pyrogallol (6%, w/v) as antioxidant. The mixture was digested for 45 minutes at 70°C. They were cooled after digestion, in a beaker of cold water (18°C) for 20 minutes. 15 ml of physiological saline (9%, w/v) was then added to free saponified extracts from alkali.

Tocopherols were extracted from the mixture with 15 ml of hexane/ethyl acetate (9/1, v/v). The test tubes were corked tightly and the contents were mixed, by shaking vigorously for about 2-5 minutes and left to stand for 1 hr 30 minutes during which the test tubes were shaken again at intervals, to ensure maximum extraction of vitamin E.

Two distinct layers were formed after the addition of the extracting solvents. 6 ml of the clear upper layer formed in each of the test tubes was collected using pipettes, into clean 10 ml test tubes and the lower layer was discarded. The contents of the test tubes were evaporated to dryness in an oven. The dry residues were reconstituted in a known volume of hexane/isopropanol (99/1, v/v). The test tubes were corked tightly, wrapped in black cellophane bags and kept in a refrigerator before the HPLC analysis. All experiments were performed under low light because it has been reported in literature that tocopherols are unstable in presence of light.

### High Performance Liquid Chromatography Analysis

**Apparatus:** CECIL HPLC (series 1000): UV detector (high performance variable wavelength monitor) absorbance range- 0.02, wavelength 280 nm, flow rate 2 ml/minute, pressure 5.3 mpa (milipascal), mobile phase: methanol (CHROMASOLV for HPLC) and water (98/2); recorder (ABB GOERZ recorder, model: SE 120), chart speed 30 cm/hour, chart voltage 1000 mV; microlitre syringe, stationary phase:RP18 column 10 µm, 25 cm (length), ¼ OD (outside diameter).

**Standard:** one ampoule (5ml) dl- $\alpha$ - tocopherol acetate (density 0.95g/ml) (produced by BDH chemicals)

**Procedure:** The eluant (mobile phase) was degassed by boiling in a water bath (with shaker) at 50°C for a minimum of four hours. A stock standard solution of 1000 ppm (parts per million) of dl- $\alpha$ -tocopherol acetate was obtained by dissolving 0.1 ml of standard in 100 ml n- hexane/isopropanol (99/1). The working standard solutions were obtained by diluting 0.5 ml, 1.0 ml, 1.5 ml, 2.0 ml and 2.5 ml of stock standard solution with n-hexane/isopropanol to 10 ml respectively (which represented concentrations of 50, 100, 150, 200 and 250 ppm respectively). They were each injected directly unto the column with a microlitre syringe and the injection volume was 20 µl. It was monitored by their absorbance at 280 nm with a UV detector. All determinations were made in duplicates. The peak heights were measured and the mean peak heights were the various concentrations of the standard prepared. This was used to draw a calibration graph.

20 µl of each of the samples which were already reconstituted in a known volume of n-hexane/ isopropanol were injected individually unto the liquid chromatography. Each sample was injected twice. The results obtained were reproducible and retention time for all samples was approximately four minutes.

**Fat content was determined according to Official methods of analyses of association of analytical chemist [16]:** The fat content was determined using the soxhlet extraction method.

**Statistical Analysis:** Results are presented as mean and standard deviation from analytical duplicate. One-way analysis of variance was carried out to determine if there were significant differences between the vitamin E levels in the raw and different processed products, using SPSS version 17. The first one consists of mean comparison of 2 data $\times$ 4 types of processed products from groundnuts (raw, roasted, defatted fried cake and oil) for vitamin E levels. The second was mean comparison of 2 data $\times$ 5 types of processed products from melon seeds (raw, roasted, fermented, defatted fried cake) for vitamin E levels. The third was a comparison of all the samples of both groundnut and melon seeds together. A  $p$ -value $<$ 0.05 was taken as significant.

## Results

Table 1 shows the fat content of the raw and processed products of groundnut and melon seeds. The fat content of ranged from 19.0  $\pm$  0.15% in defatted fried groundnut cake (*kulikuli*) to 54.4  $\pm$  0.4% in raw groundnut. While in melon seed fat content ranged from 18.6  $\pm$  0.05% in defatted fried melon seed cake (*robo*) to 56.4  $\pm$  0.36% in roasted melon seeds.

The vitamin E content of raw groundnut and melon seeds and their products are presented in Table 2. The table indicates that the oils had the highest level of vitamin E: 48.1  $\pm$  0.88 mg/100g for groundnut oil and 20.1  $\pm$  20.1.30 mg/100g for melon seed oil. It was observed that the vitamin E levels in groundnut were higher in the processed product than in the raw groundnut. The raw groundnut had the lowest vitamin

	Groundnut ( <i>Arachis hypogea</i> )	Melon seeds ( <i>Citullus vulgaris</i> )
	Fat (%)	Fat (%)
Raw	54.4 $\pm$ 0.4	54.7 $\pm$ 0.10
Roasted	50.6 $\pm$ 0.3	56.4 $\pm$ 0.36
Fermented ( <i>Ogiri</i> )	-	28.9 $\pm$ 0.62
Defatted fried cake	19.0 $\pm$ 0.15	18.6 $\pm$ 0.05

**Table 1:** Fat content of raw and processed groundnut and melon seeds.



	Groundnut ( <i>Arachis hypogea</i> )	Melon seeds ( <i>Citrullus vulgaris</i> )
	Vitamin E (mg/100g)	Vitamin E (mg/100g)
Raw	8.9 ± 0.66	16.1 ± 0.88
Roasted	16.7 ± 0.23	3.9 ± 0.00
Fermented ( <i>Ogiri</i> )	-	12.0 ± 0.55
Defatted fried cake	13.1 ± 0.46	6.1 ± 0.62
Oil	48.1 ± 0.88	20.1 ± 2.30

**Table 2:** Vitamin E content of raw and processed groundnut and melon seed products.

E levels (8.9 ± 0.66 mg/100g). For melon seeds the vitamin E levels were lower in the processed products than in the raw melon seeds, apart from melon seed oil which was higher in vitamin E than the raw melon seed.

When subjected to statistical analysis, the difference between the levels of vitamin E in raw groundnut, roasted groundnut, groundnut oil, were statistically significant ( $p < 0.05$ ). The difference in vitamin E levels between roasted groundnut, and defatted fried groundnut cake, were not statistically significant ( $p > 0.05$ ). In the melon seed group, the vitamin E levels between raw melon seed, roasted and defatted melon seed cake were statistically significant ( $p < 0.05$ ). However there was not statistical difference between raw melon seeds and melon seed oil ( $p > 0.05$ ). The most statistical difference in vitamin E levels was between groundnut oil and all the other samples ( $p < 0.05$ ).

## Discussion

The purpose of this work was to determine the levels of vitamin E in the raw and traditionally processed forms of two commonly consumed oil seeds in Nigeria: groundnut (*Arachis hypogea*), and melon (*Citrullus vulgaris*) seeds. The health benefits associated with oilseeds are thought to reflect their nutritional profile including their nutrient density, fatty acid profile and presence of bioactive compounds that offer protection, among which is vitamin E [17,18]. There have been reports of the beneficial effects of groundnut consumption on risk of coronary heart disease [17,19]. They are a source of energy of high density, protein, carbohydrate, fibre, phosphorus, iron, magnesium, niacin and a source of vitamin E, an antioxidant [20]. Vitamin E is active compound that participate in mopping up free radicals and also take part in oxidative stress [21].

Data obtained in this study indicate that groundnut and melon seeds and the products derived from them, all had considerable amounts of vitamin E. The vitamin content reported in this study was slightly different from the values reported in literature [22], which were 16.6 ± 5.17 mg/100g for raw groundnut, 11.3 ± 3.53 mg/100g for roasted groundnut and 25.7 ± 5.99 mg/100g for groundnut oil. As against 8.9 ± 0.66 mg/100g, 16.7 ± 0.23 mg/100g and 48.1 ± 0.88 mg/100g for raw groundnut, roasted groundnut and groundnut oil respectively. This difference could be due to variations in the processing methods used. Some authors reported a loss of vitamin E after raw groundnuts were roasted and milled during the manufacture of peanut butter [22]. However results obtained in this experiment were contrary in that there was no loss of vitamin E after roasting the raw groundnuts and defatted fried groundnut cakes (Table 2). However, in the case of melon seeds, the results showed that there was loss of vitamin E after raw melon seeds were processed into roasted, fermented and defatted fried melon seed cake forms, respectively (Table 2). Statistically, the difference in vitamin E levels was significant ( $p < 0.05$ ).

In agreement with previously published studies [5,20,23], our data show that groundnut and melon seeds have very high oil content

(Table 1), about 50% of the seed. In this study, both groundnuts and melon seeds oils had the highest vitamin E levels. This supports the fact that plant oils and products made from them are considered the richest sources of vitamin E [21]. In a study of ten grape seed varieties, the authors reported that grape seed oils were an excellent source of vitamin E [24]. In a review of the food potentials of unconventional oilseeds grown in Nigeria, the author reported that *Citrullus vulgaris* (melon seeds) had vitamin E values of approximately 19 mg/100g of its crude oil [25]. This value is in close agreement with the values observed in this study for melon seed oil (20.1 ± 2.30 mg/100g). The same author [25] also noted that melon oil was naturally rich in the kinds of nutrients which were found in oils considered to be good edible oils like groundnut oil and soyabean oil. Melon seed oil has been reported to have 71.9% unsaturated fatty acid of which 57.4% are polyunsaturated fatty acids and 14.5% monounsaturated and that it has a hypocholesterolemic effect [5]. Similarly, studies have revealed that frequent nut consumption including groundnuts have protective effects against coronary heart diseases [19,17,26]. All of these beneficial effects have been adduced to the bioactive compounds in these oilseeds, which include vitamin E.

This study presents us with more evidence of the nutritional value of our traditional oilseeds in Nigeria and their potential for increased dietary utilization to meet the dietary requirements for vitamin E in individuals. Since groundnut and melon seeds make a significant contribution to Nigerian diets, then their nutritional value in terms of vitamin E could offer protective effects against certain diseases like heart diseases. The use of groundnut and melon seeds especially their oils, which have been found to be high in vitamin E in this study should be explored in dietary formulations aimed at addressing cardiovascular diseases.

## Conclusion

This study has provided information on the vitamin E content of raw groundnut and some of its traditional products in Nigeria. Groundnut and melon seeds are oilseeds that will always be consumed in this part of the world and this work has demonstrated that in addition to their reported high protein and energy content, they are good sources of vitamin E (tocopherol) especially in the oils obtained from them. Most of the traditionally processed products contained sufficient vitamin E to meet RDA when consumed in adequate amount. Of particular note is the high oil content as well as the high content of vitamin E in the oil, which further proves that the vegetable oils are among the richest sources of vitamin E.

There is need for further studies on vitamin E content of Nigerian local foods especially the oilseeds and their antioxidant potential. Secondly, there is need to analyse the locally produced commercial groundnut oil sold in Nigeria for vitamin E. This is because the products are usually exposed to sunlight and other unfavourable conditions, when sold in the market. These practices might lead to loss of this important vitamin in the various food products.

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