Comparative Evaluation of Proximate Compositions, Functional and Physicochemical Properties of Raw Melon Seeds of Five Members of Cucurbitaceae Family

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Citation

Abstract
Comparative study of the natural fermentation of five melon seed varieties (Citrullus vulgaris (CV), Citrullus lanatus (CL), Colocynthis citrullus (CC), Cucurbita pepo (CP), and Cucumeropsis edulis (CE)) is presented. Each melon seed variety was procured, dried, dehulled and ground into flour for the determination of functional properties and proximate composition of raw seeds. Free Fatty Acid (FFA), Acid Value (AV), Total Titrable Acidity (TTA), and proximate composition were determined. Data obtained were analysed statistically at p≤0.05. The results of functional properties showed variations in behaviour. While CV was significantly different (p≤0.05) in bulk density (1.70g/cm^3), oil absorption capacity (1.55g/ml) and swelling index (1.36g/cm), CL was significant in foam capacity (5.36%) and CE in emulsion capacity (50.17%).

1. Introduction

Melon seeds such as Pumpkin (Ugboguru) Cucurbitapepo L (CP), Watermelon Citrulluslanatus (Thunb.) Mansf (CL), Regular melon (Egusi) Citrullus vulgaris (Schrad) (CV), Egusi melon (Ahuru) –Cucumeropsis edulis (Hook. f.) Cogn (CE) Egusi kirikiri-Colocynthis citrullus (L) O. Ktze (CC) are cucurbit crops that belong to the Cucurbitaceae family with fibrous and shallow root system. They are tendril climbers or crawling annual crops, mostly grown as subsidiary crops interplanted with early maize and yam in some savannah belt of Nigeria, West African. Melons are major food crops with several varieties which serve as a major food source [1]. Cucurbit sp. are among the economically most important vegetable crops worldwide and are grown in both temperate and tropical regions. Egusi melon is a creeping annual and inter-cropping plant used in traditional farming practices. It thrives well on rich light soil in the hot climate regions of Africa [2].

Melon is an important component of most Nigerian diets. A valuable vegetable oil is extracted from the seeds, while the ground seed is used to prepare various delicacies including cake and soup. Melon is consumed in many parts of Nigeria, yet despite its
nutritional and commercial value; its production remains low, even with its good market price, melon is still produced by farmers on a small scale. Consequently, despite the vast nutritional, economical, usability, consumption and medicinal significance of other melon seeds, apart from Citrullus vulgaris, little detail on their proximate, functional and physicochemical properties are available to international readership. The functional properties of proteins is a reflection of the complex interactions between the composition, structure, conformation, physicochemical properties of the protein per se, other food components and the nature of the environment in which these are associated or measured [3, 4, 5, 6]. Therefore, the objective of this study is to evaluate proximate compositions, functional and physicochemical properties of raw seeds of five melon varieties.

2. Materials and Method

2.1. Materials

Five varieties of melon seeds from Cucurbitaceae family were purchased from farmers in OkporoOrlu, Izombe, Mbieri and Ekeonunwa market Owerri in Imo State, and Ohafia in Abia State. The melons included:

a) Pumpkin (Ugboguru) -Cucurbitapepo (CP)
b) Watermelon-Citrullus lanatus (Thunb.) Mansf (CL)
c) Regular melon (Egusi)-Citrullus vulgaris (Schrad) (CV)
d) Egusi melon (Ahuru)Cucumeropsis edulis (Hook.f.) Cogn (CE)
e) EgusiKiriri-Colocynthis citrullus (L) O. Ktze (CC)

Raw Melon Seed Preparation for Analysis

The seeds of each melon fruit were removed from the pulp matrix, sundried and shelled manually. The endocarps (fleshy pulp) were sundried for 2 days before grinding with a blender to obtain seed meal that passed through a 30 mesh sieve. Each seed meal was packed in a separate plastic container, carefully labelled and stored in the refrigerator until it was required for use [7].

Functional Properties of each Raw Melon Seed Variety

Functional properties of the raw melon seed varieties were determined as described by [8, 9].

Bulk Density

A 10 ml-graduated cylinder was gently filled to mark with melon seed meal. The filled cylinder was gently tapped on a laboratory bench about 10 times until there was no further diminution of the sample level after filling to the 10ml mark. The procedure was adopted for each of the melon seed meal sample, and the bulk density was calculated using Equation 1 below:

\[
\text{Bulk density (g/ml) } = \frac{\text{Mass (weight) of sample}}{\text{Volume of material after tapping}} \quad \text{(Eqn. 1)}
\]

2.2. Wettability

A small quantity, (1.0g) of melon seed meal was poured into a 25ml graduated cylinder. A finger was placed at the open end of the cylinder inverted and clamped at a height of 10cm from the surface of a 600ml beaker containing 500ml of distilled water. The finger was removed to allow the sample to be poured. The time required for all the melon meal to become wetted and penetrate the surface of the distilled water was recorded as the wettability for that melon variety seed meal.

2.3. Emulsion Capacity

Two grams (2.0g) of melon seed meal sample were blended with 20ml of distilled water at room temperature for 30 seconds in a warring blender at 1600 rpm. After complete dispersion, 20ml of refined vegetable oil was added and blending continued for another 30 seconds. The mixture was then centrifuged at 1,600 rpm for 5min. The volume of oil separated from the sample after centrifugation was read directly from the tube.

2.4. Oil and Water Absorption Capacity

One gram (1.0g) of melon seed meal was weighed into a centrifuge tube. The sample was mixed thoroughly with 10ml distilled water (for water absorption) or oil (for oil absorption) for 30secs. The sample was allowed to stand for 30min at room temperature and then centrifuged at 5000 x g for 30min. The volume of free water or oil (the supernatant) was read directly from the graduated centrifuge tube. The result is expressed as gram of water or oil retained respectively.

\[
\text{Volume of oil or water used} = x \text{ ml}
\]

\[
\text{Volume of supernatant} = y \text{ ml}
\]

\[
\text{Volume of oil or water absorbed} = x - y \text{ ml} \quad \text{(Eqn. 2)}
\]

2.5. Gelling Point

This was determined according to the method of Narayana and Rao (1982). Five grams (5.0g) of each melon seed meal sample was poured into a beaker (250ml) respectively. Each meal sample was dispersed to make 50ml suspension using distilled water. A thermometer was clamped on a retort stand with magnetic stirrer and heating system. The heating and stirring continued until the suspension began to gel, the corresponding temperature was recorded. The temperature at boiling point was also recorded.

2.6. Foam Capacity

Two grams (2.0g) of melon seed meal were dissolved with 100ml of distilled water in a warring Blender at 1600 rpm for 5min. The suspension was poured into a 250ml measuring cylinder and the volume after 30sec was recorded.

Foam capacity is expressed as percent increase in volume and is calculated (Eqn. 3) thus:

\[
\text{Foam capacity} = \frac{\text{Vol after whipping} - \text{Vol before whipping}}{\text{Vol before whipping}} \times 100 \quad \text{(Eqn. 3)}
\]
2.7. Swelling

One gram (1.0g) of each melon seed meal was weighed into a dry clean test tube respectively. The meal was gently leveled and the volume noted. Ten milliliters (10ml) of distilled water was added in each case. The content of each tube was swirled and then allowed to stand for 60minutes, while the change in volume was recorded every 15 minutes. The swelling power index of each melon seed flour sample was calculated as multiple of the original volume (Olawuni et al, 2013)

\[
\text{Swelling index} = \frac{H_2}{H_1}
\]

(Eqn. 4)

Where \(H_1\) = Initial height
\(H_2\) = Final height

2.8. Proximate Composition of Raw and Boiled Melon Seed Varieties

The standard methods of [9] No. 945.38; 942.05; 955.04C; 942.05 and 920.39A were used for the analysis of proximate composition of the melon seed varieties respectively.

2.9. Determination of Moisture Content

Two grams (2.0g) of the melon seed meal for each variety were weighed into dry and weighed crucibles respectively. The crucibles with contents were transferred into moisture extraction oven at 105°C for 2h. The crucibles with contents were cooled in a desiccator and weighed. The process was repeated until a constant weight was obtained. The loss in weight obtained for each sample (using Eqn. 5) represented the moisture content for that melon seed variety.

\[
\% \text{moisture content} = \frac{\text{Initial wt of sample} - \text{wt of oven dried sample}}{\text{Initial wt of sample}} \times 100
\]

(Eqn. 5)

2.10. Determination of Ash Content

Two grams (2g) of the melon seed meal of each melon seed variety were weighed into a dry and weighed porcelain crucible. These were then charred over a Bunsen flame before igniting in the muffle furnace at 550°C for 30 minutes. The ash contents were cooled in a desiccator and weighed. The percentage loss in weight during combustion was calculated (with eqn. 6) as the ash content.

\[
\% \text{ash content} = \frac{\text{Weight of Ash}}{\text{Weight of sample}} \times 100
\]

(Eqn. 6)

2.11. Determination of Crude Protein Content

Total nitrogen and protein content of each melon seed meal were determined by Kjeldahl methods as described by James (1996) and Onwuka (2005). Approximately 0.2g of the meal was weighed into a Kjeldahl digestion flask, into which 1g of copper sulphate, 1 tablet of Kjeldahl, 25ml of concentrated sulphuric acid and a few glass beads were added. The mixture was digested under a fume cupboard until a clear solution was obtained. All the digests were carefully transferred into a 100ml volumetric flask and made up to mark using distilled water. A 50ml portion of the digest was mixed with equal volume of 40% NaOH solution in a micro Kjeldahl (Markham distillation apparatus) unit and distilled. The distillate was collected into 10ml of 4% boric acid solution containing 3drops of mixed indicator (bromocresol green-methyl red). A total of 50ml distillate was collected and titrated against 0.02N H\(_2\)SO\(_4\) solution to a color change from initial bluish-green color to pink (end point). Percentage total nitrogen and percentage crude proteins were calculated with Equations 7 and 8 respectively thus:

\[
\% \text{Nitrogen} = \frac{T \times N \times V{f} \times 14}{W} \times 100
\]

(Eqn. 7)

\[
\% \text{Crude Protein} = \% \text{Nitrogen} \times C
\]

(Eqn. 8)

Where: \(N\) = Normality of the acid
\(V_f\) = Total volume of digest
\(V_t\) = Volume of digest distilled
\(W\) = Weight of sample digested
\(T\) = Volume of acid used to titrate the sample
\(C\) = Correction factor (6.25)

2.12. Determination of Crude Fibre Content

Two grams (2.0g) of defatted sample (defat with petroleum ether if more than 10% fat) were weighed into a 250ml beaker containing 200ml of 0.125m tetraoxosulphate IV acid (H\(_2\)SO\(_4\)). The mixture was heated in a steam bath at 70 – 90°C for 2 hours, allowed to cool and then filtered using a muslin cloth over a Buckner funnel. The residue was washed three times with hot water to remove acid and then poured into a beaker containing 200ml of potassium hydroxide (KOH). The residue was heated again as before, filtered and the residue washed three times with hot water, then with alcohol and water. The final residue was put in a pre-heated crucible and dried at 120°C to a constant weight. It was then incinerated in the muffle furnace at 550°C for 30min (sample becomes ash white), cooled in a desiccator and weighed. The percentage crude fibre is calculated thus:

\[
\% \text{Crude fibre} = \frac{\text{Wt of oven dried} - \text{wt of ash}}{\text{Initial wt of sample}} \times 100
\]

(Eqn. 9)

2.13. Determination of Crude Fat Content

Five grams (5.0g) of melon seed meal were weighed into a filter paper, wrapped carefully and put in the holder of the Soxhlet extraction apparatus. A clean dried and weighed Soxhlet extraction flask was half filled with diethyl ether and the whole apparatus was assembled together. The flask was placed on the heating mantle and heated at 35°C and the fat was extracted for 3 hours. At the end of the three hours, the sample holder was disconnected and the sample removed. The equipment was reassembled with only the extraction flask and
its oil content. The flask was heated at 34°C and the solvent evaporated leaving the oil in the flask. The oil was then dried in a moisture extraction oven in order to remove the solvent residues in the oil. The dried oil was cooled in a desiccator and reweighed. The drying, cooling and reweighing of sample were repeated until a constant weight was obtained. The fat content determined was calculated thus:

\[
\% \text{Crude fat} = \frac{\text{Wt of flask} - \text{wet wt of empty flask}}{\text{Wt of sample}} \times 100 \text{(Eqn. 10)}
\]

2.14. Carbohydrate Content Determination

The carbohydrate content of the melon seed meals was obtained by difference as below:

\[
\% \text{CHO} = 100 - (\% \text{MC} + \% \text{fat} + \% \text{protein} + \% \text{fibre} + \% \text{crude ash}) \quad \text{(Eqn. 11)}
\]

Where: CHO = Carbohydrate
MC = Moisture Content

2.15. Determination of Physicochemical Properties

2.15.1. Free Fatty Acid and Acid Value

Free fatty acid and acid value of each fermenting mash was taken at 24h intervals for 96h using the method of Onwuka (2005). Two grams (2g) of fermenting paste was dissolved in a mixed neutralized solvent (25ml diethyl ether + 25ml alcohol + 1ml phenolphthalein (1%)) and carefully neutralized with 0.1N sodium hydroxide (NaOH) and titrated with aqueous 0.1M NaOH, constantly shook until a pink colour persisted for 15 sec. The titre value was recorded and calculated thus:

\[
\text{Acid Valu} = \frac{Titrable \text{acidity}}{\text{Wt of sample (g)}} \times 100 \quad \text{(Eqn. 12)}
\]

\[
\text{FFA (as oleic acid)} = \frac{X \times 0.0282g}{W} \quad \text{(Eqn. 13)}
\]

Where \( X = \text{Titre Value} \)

2.15.2. Titrable Acidity

Total titrable acidity of the fermenting melon seeds as described by [10]. was adopted. A small quantity (2.5g) of fermenting melon seed samples were macerated with 50ml of distilled water. Three (3) drops of phenolphthalein indicator were added. This suspension was titrated with 0.1N NaOH to a pink end point which persisted for 15sec. Titrable acidity was expressed in percentage lactic acid as:

\[
\% \text{Titrable acidity} = \frac{N \times V \times \text{Eq wt}}{W \times 1000} \times 100 \quad \text{(Eqn. 14)}
\]

Where \( N = \text{normality of NaOH (in Eq/ml)} \)
\( V = \text{volume of titrant (ml)} \)
\( \text{Eq wt} = \text{equivalent weight of predominant acid (mg/m.Eq)} \)
\( W = \text{weight of sample (g)} \)
\( \alpha = \text{factor relating mg to gram (mg/g)} \)

2.16. Statistical Analysis

The data obtained from this study were statistically analyzed for Analysis of Variance (ANOVA) using SAS 9.2 or Windows 2007 and Windows XP. The means were separated using Fisher’s Least Significant difference (LSD) at \( P<0.05 \) confidence level.

3. Results and Discussions

3.1. Functional Properties of Raw Melon Seed Flours

The results of functional properties of the raw melon seed varieties’ flours as shown in Table 1 include:

3.2. Bulk Density

Bulk density ranged from the lowest 0.65±0.03g/ml for Citrullus lanatus (CL) to the highest 1.74±0.04g/ml for Citrullus vulgaris (CV). Similar values were obtained by Olawumi et al (2013) in their work on the effect of pH and temperature on functional and physicochemical properties of Asparagus bean (Vignaesquis pedalis) flour. There were significant differences (p<0.05) in bulk density values of samples, with CV being significantly different from CC (Colocynthis citrullus) and CP (Cucurbita pepo), which were in turn significantly different from CE (Cucumeropsis edulis).
and CL (Citrullus lanatus). This showed that CV was denser than all the other melon seed varieties, followed by CP and CC while CL was the least when ground into flour for soup or stew. High bulk density is desirable since it helps to reduce paste thickness and ease the dispersibility of food powders [11], [12, 13]. This could be the reason why CV is regularly and comfortably being used in soup making.

According to [14], bulk density of foods increases with increase in starch content. [15] however said that bulk density is generally affected by the particle size and the density of the flour, and it is very important in determining the packaging requirement, material handling and application in wet processing in the food industry. However, [16] holds the view that reduction in bulk density has nutritional and economic significance as more of the products can be eaten, resulting in high nutrient intake. Additionally, [17] reported that low level of bulk density could be an advantage in the formulation of baby weaning foods. [18] also asserted that low bulk density is an advantage in the preparation of complementary foods, because high bulk density limits the caloric and nutrient intake per feed of a child, which can result in growth faltering. This therefore placed CL and CE at advantage over CV, CP and CC for use in food formulation.

### 3.3. Foam Capacity (FC)

The foam capacity of CL (5.36±0.02%) was found to be significantly different (p<0.05) from that of CV (3.72±0.29%) and other melon seeds as shown in Table 1. These values were lower than those obtained for Asparagus bean isolate (3.94%), full fat (8.45%), defatted (12.46%) and concentrate (5.33%) (Olawumi et al, 2013). From this result, CE, CP and CC do not have the ability to retain stable foam when whipped, therefore they may not do well as an aerating or foaming agent in food formulation [19]like ice cream. However, they could be used to enhance the nutritional value of food [20] While CL and CV may have the ability to incorporate air by themselves or in a mixture with other ingredients and to hold aerated structure so that it can be set by heat or other means. Foamability is related to the rate of decrease in the surface tension of the air-water interface caused by absorption of protein molecules. [21, 22] Good foamability can be linked with flexible protein molecules that can reduce surface tension. Highly ordered globular proteins which are relatively difficult to surface denature, give low foamability. Therefore, it does appear that CE may be rich in globular protein.

### 3.4. Emulsion Capacity (EC)

The emulsion capacities of the flours of the melon seed varieties ranged from 46.85% (CL) to 50.17% (CE) as shown in Table 1. There were significant differences (p<0.05) in emulsion capacities of melon seed flours, with CE being significantly different from CV and CC. These were in turn different from CP and then CL. [23] reported that emulsion capacity and stability are higher in protein with globular nature. Proteins aid in formation and stabilization of emulsion [24]; hence the relatively high emulsion capacity obtained for CE could be due to the nature and type of the protein. However, [25] obtained emulsion capacities of flour and protein concentrate of 450 and 690ml oil/g dry sample respectively in their work. They attributed their observation to the presence of carbohydrates and fibres in the flour. Therefore, the relatively low emulsion capacity obtained for CL could be traceable to its high carbohydrate content (17.59%) and fibre (5.73%), compared to the high emulsion capacity of CE which had (6.71%) carbohydrate and (5.32%) fibre.

### 3.5. Oil and Water Absorption Capacities

There were significant differences (p<0.05) in oil absorption capacities of the flours of melon seed varieties with CC, CL and CV being significantly different (p<0.05) from CE and CP (Table 1). The values ranged from the lowest (CP=1.33g/ml) to the highest (CC=1.57g/ml). These values corresponded with the value (1.43g/ml) obtained for asparagus bean flour [20], and higher than that obtained for Trichosanthes cucumerina (snake ground) seed flour 54% (0.54g/ml) [25], pumpkin seed flour 87% (0.87g/ml), soy flour 84.4% (0.84g/ml) [26]. The result showed that melon seeds may have higher flavor retention than other legumes of lower oil absorption capacity. This high OAC of melon seeds could be due to the high hydrophobic protein of the seeds.

Invariably high oil absorption capacities show that the melon seeds increase mouth feel when used in food preparations such as meat analogues [20]. OAC is the ability of the product to entrap oil. It is an important functional property for such applications as meat replacers and extenders mainly because it acts as flavour retainer and increases the mouth feel of foods [21] hence the significant difference in retention capacity of CC and CV.

The water absorption capacity of the melon seeds flours (Table 1) ranged from the lowest 1.79g/ml (CC) to the highest 3.40g/ml (CE and CL). The result showed a significant difference (p<0.05) in the water absorption capacities of CE and CL from those of CV, CP and CC. The values were higher than those obtained for other legumes example soy flour (130% i.e. 1.30g/ml) and Pigeon pea flour (138% i.e. 1.38g/ml) [25], and that obtained by [20] for asparagus bean flour (1.330g/ml). According to [20], low values of water absorption capacity of the different flour samples suggest that asparagus beans flour is less hydrophobic than other legume flours. The melon seed flours absorbed more water, which could imply that they have both hydrophobic and hydrophilic moieties [27] in their molecules and hence are soluble in relatively polar solvents. It has been reported that water binding by starch is a function of several parameters including size, shape, conformational characteristics, hydrophilic and hydrophobic balance in the molecule. Other parameters are lipids and carbohydrates associated with proteins, thermodynamic properties of the system, physicochemical environment (pH, vapour pressure, temperature, and so on) and the solubility of starch molecules. 

Nature of starch has also been found to have effect on water absorption capacity [3]. High water absorption capacity is also attributed to loose structure of starch polymers while low values indicate compactness of the structures [3, 5]. CE and CL being the highest, shows that they can also be used as thickeners for soup and stew.

3.6. Wettability of the Melon Seed Flours

Wettability is the time required for the sample to become completely wet [8]. There were significant differences in the wettability (Table 1) of the flours of melon seed varieties: CE (6.67 sec/g) was significantly different (p< 0.05) from CC and CP, while CV (1.03 sec/g) was the least. CE took longer time to become completely wet which implies that it has less water affinity than the other melon seeds. Conversely CV has the greatest affinity to water, hence a small wettability. However, [9] described wettability as the ability of powdery particles to adsorb water on their surfaces, thus initiating reconstitutions, and that it largely depends on the particle sizes. This could mean that CV and CL will reconstitute easily in food systems upon hydration.

3.7. Swelling Indices of the Flours of Melon Seeds

The swelling indices of the flours of melon seed varieties (Table 1) ranged from the lowest (CL) 1.12 g/ml to the highest (CV) 1.36 g/ml. Statistical analysis showed significant differences (P<0.05) in swelling index among the melon seeds: CV (1.36 g/ml) was significantly different from CP (1.34 g/ml) and CC (1.30 g/ml) which were in turn different from CE (1.21 g/ml) and CL (1.12 g/ml). The differences in swelling indices could be attributed to the differences in the inter-molecular starch bound in each of the raw melon seeds which allowed it to absorb water and swell. [28] The morphological change of the granules during swelling depends on the origin of the starch, [29] and starch swelling behaviour not only depends on the starch origin but also on the amylose content, [30] the extent of chemical cross-binding within the granules and non-carbohydrate substances such as lipids or phosphates [31]. High amylose content as well as the presence of higher numbers of stronger intermolecular bonds may also reduce swelling [32], hence the low swelling index of CL.

3.8. Gelling Point (Temperature)

Gelation is an aggregation of denatured molecules and protein concentration, especially globulin fraction. Interactions between proteins, carbohydrates and lipids have been reported to be responsible for the gelation capacity of legumes and oil seed proteins [23, 33] had the opinion that higher carbohydrate content gives better gelating property in food products. Statistically, a significant difference (p < 0.05) was found between the flours of melon seeds of CV, CP, CC, CL, and CE in their gelling point (Table 1). Work done by Yusuf et al (2007) showed that snare gourd seed flour (also in Cucurbitacae family) required a lower concentration for gel formation than most oil seeds and legume flours. This could be useful applications in food systems such as sausages emulsion, and sauces that require thickening and gelling [20]. obtained a gelling point of 96°C (for full fat Asparagus flour) and 94°C (for defatted flour) and concluded that the property would make the flours suitable in food systems where thickening and gelling properties are required. This could be the reason why melon seeds generally are used as soup thickeners particularly CV. However, [23] associated the variation in gelling properties to different constituents-proteins, lipids and carbohydrate that make up the legume. The result showed that these other melon seeds (CP, CC, CL and CE) could also be used in food systems that require thickening just as CV. Hence they could be used in soup making.

4. Physicochemical Properties of Melon Seed Varieties

The results of the determination of the physicochemical (FFA, AV and TTA) properties of raw, boiled, primary and secondary fermented melon seeds are shown in Table 2. The free fatty acid (FFA) of melon seeds increased as fermentation progressed in all the samples, ranging from the lowest values 2.47 mg/g – 21.27 mg/g (CC), 6.28 mg/g - 24.83 mg/g (CE); 6.07-25.93 mg/g (CL); 8.56-26.36 mg/g (CP) to 3.13-22.71 mg/g (CV) raw seeds to fermented products. Generally secondary fermented melon substrates had higher FFA than primarily fermented melon seeds. The highest value (26.36 mg/g) was obtained in CP during 144h into secondary fermentation while the least value (21.27 mg/g) was recorded for CC. Such trend has been observed by [34] during fermentation of melon seeds and by [35] for fermentation of fluted pumpkin. The increase in free fatty acid is indicative of the production of lipases by the fermenting bacterium. This is compatible with what is known about fermentation by Bacillus species. [36, 37]. Such observations have been made by previous workers [38]. Generally, secondary fermentation had higher FFA than primary fermented melon seeds. Though [39] obtained a contrary result in their work, with unfermented melon seeds having a higher fatty acid than the fermented; C. lanatus (52.10 mg/g- 38.40mg/100g) and C. vulgaris (53.50-43.20mg/100g).

This is exceptional because [40] had reported of low levels of lipase in melon. Low lipase activity in some fermented foods has been considered desirable because of problems of objectionable taste and development of rancidity [40, 41].
However, there are reports of beneficial effects of lipase in the development of characteristic flavours and aroma [42]. Significant lipolysis of legumes yields predominantly oleic, linoleic and linolenic acids [43, 44], and free fatty acids particularly oleic and linoleic (omega 3 and 6) are desirable since rancidity can be checked via alkalinities.

Acid value is the number of milligramme of potassium hydroxide required to neutralize the free acid in one gramme of the oil or fat sample [8, 45]. The acid value measures the extent to which the glycrides in the oil have been decomposed by lipase action. The decomposition is accelerated by heat and light. As rancidity is usually accompanied by free fatty acid formation, the determination is often used as a general indication of the condition and edibility of oils [8]. Acid value of the melon seeds increased as fermentation progressed, with CL and CP being significantly different (P<0.05) from CE which in turn differs from CV and then CC. This could be the explanation of the significant difference in flavour obtained in CC. However, this result shows that fermented melon seed condiments are likely to develop off-flavour if kept for a longer time especially under heat or light. [46] reported that lipid oxidation and resultant flavour impairment had seriously limited the storage potential of most fat – containing foods, hence the observation made: as the samples were left near a hearth, the oil content seemed to increase and the drop off became very visible.

Total titrable acidity (TTA) of the melon seed substrates increased significantly with fermentation. The values obtained (lowest-highest) were: for raw seeds (0.56-1.87%), boiled seeds (0.85-2.40%), primary fermented substrates (2.06-3.14%) and secondary fermentation (2.99-5.69%). The result showed a steady and significant increase in total titrable acidity with fermentation. Similar results were obtained by [39] for C. lanatus (0.11-2.40%), C. vulgaris (0.10-1.60%) and C. manni (0.20-2.00%) for unfermented and fermented melon substrates respectively [47], also reported a simultaneous increase in titrable acidity (2.83-20.2 and 19.6mg/g) for melon seed mash (C. vulgaris) fermented by mixed cultures and pure cultures of Bacillus spand Proteus sp. Similar trends have been observed during fermentation of some other protein rich plant material[48, 49].The increase in titrable acidity could be attributed to the production of various organic acids from utilization of the little amount of carbohydrate present [47] by few of the genera (Corynebacterium sp and Enterococcus sp) classified as lactic acid bacteria [50], and accumulation of other metabolites [39]. They may not be proteolytic, but may have contributed to the final flavour of the product through the production of organic acids and other products of carbohydrate metabolism.

The total titrable acidity of CL was significantly different (P<0.05) from that of CP; which was also significantly different from CE, CC and CV in secondary fermentation. The differences could be due to the intrinsic properties of the seed to support bacteria growth [40].

### Table 3. Proximate Composition of Raw, Boiled, Primary and Secondary Fermented Melon Seed Varieties.

<table>
<thead>
<tr>
<th>TRT</th>
<th>SC</th>
<th>Proximate composition parameter</th>
<th>MC</th>
<th>F</th>
<th>CrP</th>
<th>A</th>
<th>CF</th>
<th>CHO</th>
<th>DM</th>
</tr>
</thead>
<tbody>
<tr>
<td>CC</td>
<td>5.60±0.05</td>
<td>22.88±0.01</td>
<td>3.50±0.05</td>
<td>7.64±0.54</td>
<td>10.54±0.05</td>
<td>94.03±0.55</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CE</td>
<td>8.93±0.02</td>
<td>28.73±0.06</td>
<td>4.57±0.03</td>
<td>5.35±0.06</td>
<td>6.72±0.01</td>
<td>91.16±0.19</td>
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<tr>
<td>CL</td>
<td>5.29±0.06</td>
<td>27.15±0.23</td>
<td>4.20±0.17</td>
<td>5.73±0.01</td>
<td>17.52±0.12</td>
<td>94.70±0.01</td>
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<tr>
<td>RAW</td>
<td>CP</td>
<td>6.26±0.06</td>
<td>30.47±0.02</td>
<td>5.15±0.22</td>
<td>7.58±0.12</td>
<td>9.12±0.17</td>
<td>93.63±0.23</td>
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<tr>
<td></td>
<td>CV</td>
<td>7.19±0.01</td>
<td>26.61±0.17</td>
<td>3.30±0.12</td>
<td>6.42±0.02</td>
<td>5.37±0.04</td>
<td>92.76±0.12</td>
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<td>Mean</td>
<td>6.66±1.36</td>
<td>27.17±2.62</td>
<td>4.14±0.72</td>
<td>6.56±0.97</td>
<td>9.85±4.39</td>
<td>93.26±1.30</td>
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<tr>
<td>LSD</td>
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<td>0.239</td>
<td>0.252</td>
<td>0.177</td>
<td>0.175</td>
<td>0.519</td>
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</table>

### 5. Proximate Compositions of Melon Seeds

Table 3 shows the results of the determination of the proximate compositions of raw or boiled melon seeds and seeds used in primary and secondary fermentations.

Crude Protein: C. pepo (CP) was significantly different (p<0.05) in protein content with a value of 30.47% in the raw sample analysis (Table 3) followed by CE (28.73%), CL (27.15%), CV (26.61%) and CC (22.88%). These results corresponded closely to values obtained by [51, 52, 53, 54] Higher values have been reported by [54] for Cucumeropsis manni (34.86%) and Cucumeropsis edulis (35.31%) and [55] for C. lanatus (34.07%) and C. manni (31.53%). However, the result in Table 3 is still higher than the result of [55] for unfermented C. pepo (20.2%). But the values obtained for C. pepo corresponded with literature (30%), C. lanatus (30 – 40%) as given by [57] and C. edulis was 36% (Schippers 2000).

Fat: The Fat content of the raw melons (being oil seeds) were CC (49.71%), CE (45.72%), CL (40.43%), CP (41.50%) and CV (51.20%). These results agreed closely with report of [39, 51, 52, 55, 53 54]. According to [57], melon seed kernels contain about 45% oil, which is sold as a commodity itself, while [58] reported a fat content of 20-45% (Citrullus lanatus), and 40-50% oil (Cucurbita pepo). This showed that the data obtained in this research fell within the limits. There were significant differences (p<0.05) in the fat contents of the melon seed varieties. CV was significantly different (p<0.05) from CC which in turn was significantly different from CE, CP and then CL (the least). These melon seeds are all from Cucurbitaceae family (Schippers, 2000) but from different genera and species which could be the reason for the variation.

Moisture content of the melon seeds ranged from 5.29-
partly with report of [56, 54]. The ash content of all the products. Moisture contents of the raw seeds were lower than products respectively. They also observed a decrease in ash content from 8.93% (Raw) to 21.32-25.12% (secondary fermented) melon seeds. A slight decrease in ash content was observed in all the melon seeds (from raw seeds to fermented substrates) except in CL. [39] reported an increase in ash content of C. lanatus (2.83±0.3 - 2.97±0.2) and C. vulgaris (2.68±0.2-4.80±0.3) for unfermented and fermented products respectively. They also observed a decrease in ash content of Cucumeropisismanni from 3.05±0.5 (unfermented) to 2.91±0.7% (fermented). However, these authors did not specify whether their observations were made from raw or boiled melons for the unfermented, and whether the fermented was from primary or secondary fermentations.

Ash content: The ash content of the melon seeds (Table 3) corresponded closely with the report of [51, 55, 56] and partly with report of [56, 54]. The ash content of all the melon seeds ranged from 3.30-5.15% (raw), 3.67-5.36% (boiled), 3.04-4.76% (primary fermented) to 2.29-4.76% (secondary fermented). A slight decrease in ash content was observed in all the melon seeds (from raw seeds to fermented substrates) except in CL. [39] reported an increase in ash content of C. lanatus (2.83±0.3 - 2.97±0.2) and C. vulgaris (2.68±0.2-4.80±0.3) for unfermented and fermented products respectively. They also observed a decrease in ash content of Cucumeropisismanni from 3.05±0.5 (unfermented) to 2.91±0.7% (fermented). However, these authors did not specify whether their observations were made from raw or boiled melons for the unfermented, and whether the fermented was from primary or secondary fermentations.

Crude Fibre: Apart from CV which had an increase in crude fibre (6.42%raw-7.24%) during secondary fermentation, all the other melon seed varieties experienced a decrease in crude fibre content: CC (7.64-5.87%), CE (5.35-2.46%), CL (5.73-4.52%) and CP (7.58-6.43%) as shown in Table 3. However, the value in primary fermentation was lower (5.58%). [39] also reported higher crude fibre content in unfermented melon seeds: Cucumeropisismanni (3.30±0.1-1.99±0.1), C. lanatus (3.43±0.2-1.98±0.1) and C. vulgaris (3.85±0.2-3.75±0.2). The values obtained for raw C. lanatus corresponded closely with the report of Ogunlade et al (2011), but was a little higher than the values given by [51, 55, 53, 54] These melon seeds being agricultural produce, the variations resulting from intrinsic and extrinsic factors cannot be ruled out. However, CV was very significant in crude fibre content, followed by CP, CC and then CL, while CE had the least crude fibre content.

Carbohydrate: CL was highest in carbohydrate content (17.52%), followed by CC (10.54%), CP (9.12%) and CE (6.72%). CV (5.37%) had the least carbohydrate content (Table 3). The result obtained for CC corresponded closely to the report of [53] 10.88%, but a little higher than what Onawola et al (2011) obtained (7.86%). However, the reports of [54, 55] were higher than what were obtained in this work. Also the result obtained by [52, 53] for C. edulis and C. vulgaris respectively were higher than what were obtained. [40] had a higher report for C. vulgaris (7.6±0.02%) and lower result for C. lanatus (13.30±0.2%). The differences could be due to differences in species and soil variations and other intrinsic factors. CL was significantly different (p<0.05) from all the melon seeds in carbohydrate content for both raw and fermented products. Carbohydrate content of the melon seeds generally decreased with fermentation as follows: CC (10.54-3.32%), CE (6.72-4.70%), CL (17.52-6.14%), CP (9.12-2.26%) and CV (5.37-4.23%). Apart from CV, the reductions in carbohydrate of the other melon substrate were pronounced. Similar trend of result was obtained by Wakshama et al (2010) for C. pepo (2.3-33.6%) while [39] reported increase in carbohydrate content of CL (13.30-25.20%) and CV (7.60-23.60%) during fermentation. [57] also reported increased carbohydrates content for CL (16.31-23.43%).

Dry Matter: There were close ranges in the dry matter (Table 3) of raw melon seeds (91.16-94.74%); however, CL was highest (94.74%), followed by CC (94.03%), CP (93.63%), CV (92.76%) and then CE (91.16%). This also tallied with the trend in the carbohydrate content of the melon seeds, showing that there is a relationship between carbohydrate content and dry matter of the melon seeds. The higher the carbohydrate content, the higher the dry matter. However, dry matter decreased with boiling and fermentation. This may be due to leaching effect of cooking solvent (water) during boiling, and tissue breaks down due to heat. The change observed in that of fermentation may be due to utilization of metabolizable parts by the fermenting organisms. Also, the lower the moisture content the higher the dry matter and vice versa.

6. Conclusion

The findings of this research have shown that the other melon seeds which are underutilized can be good substitutes in food formulations, hence reducing the pressure on Citrullus vulgaris. Proximate composition of raw, boiled and fermented melon substrates showed that these melon seeds are nutritious especially in protein and carbohydrate contents and that boiling has no adverse effect on the nutrients.

References


