

Unlocking the Nutritional Potential of Watermelon Seed Derivatives: A Comparative Analysis of Cake, Oil, and Powder

¹Muhammad Aminu Abubakar, ²Bello Mukhtar, ²A. Hamza, ¹Imoisi Chinyere, ¹Bright Ayenwenre Omoike and ³Usman Sani Bello

¹Department of Industrial Chemistry, Mewar International University, KM 21, Abuja-Keffi, Expressway, Masaka, Nasarawa State, Nigeria

²Department of Chemical Engineering, Ahmadu Bello University Zaria, P.M.B. 1094, Zaria, Kaduna, Nigeria

³Department of Petroleum and Gas Processing Engineering, Kaduna Polytechnic, P.M.B. 2021, Kaduna, Nigeria

ABSTRACT

Background and Objective: Watermelon seed cake, a by-product of watermelon seed oil extraction, is a potentially valuable source of nutrients. However, its chemical composition and proximate analysis are not well documented. This study aims to investigate the proximate composition and chemical characteristics of watermelon seed cake, oil, and powder, to explore their potential applications in food, feed, and industrial sectors. **Materials and Methods:** Watermelon seed cake, oil, and powder were obtained from a local oil mil. Proximate analysis was performed using standard methods, and the chemical composition was analyzed. The watermelon seed passed several processes which including cleaning, drying, grinding, and particle sizing, to obtain the powder. The ash content was determined by incineration, fat content was assessed by the use of solvent extraction through the Soxhlet apparatus, protein content was quantified using the micro-Kjeldahl Method, and crude was measured by sequential digestion and ashing. Carbohydrate was calculated by difference. The oil was extracted using the Soxhlet technique, which is the common method for oil extraction. **Results:** Proximate analysis revealed that watermelon seed cake contained. 11.18% moisture content, 3.57% ash content, 18.9% crude fibre, 13.72% crude fat, 10.47% crude protein, 42.16% carbohydrate, and 89.35% dry matter. The oil contained specific gravity of 0.92 g/cm³, acid value of 3.5 mg KOH/g, peroxide value of 2.9 mg KOH/g, iodine value of 75.0 mg KOH/g, saponification value of 149.0 mg KOH/g, viscosity (at 26.7°C) of 50.00 cP, refractive index of 1.45 while the powder contained moisture content 10.65%, ash content 5.1%, crude fibre 7.9%, crude fat 2.77%, crude protein 5.95%, carbohydrate 67.65% and dry matter 89.35% **Conclusion:** This study provides valuable information on the proximate analysis of watermelon seed cake, oil and powder. The results highlight the potential of watermelon seed cake, oil, and powder as a nutritious ingredient for food and feed applications, while underscoring the importance of sustainable utilization of these underutilized resources.

KEYWORDS

Proximate analysis, nutritional comparison, watermelon seed, cake, powder

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INTRODUCTION

Watermelon (*Citrullus lanatus*) is one of the most widely consumed fruits globally, with a rich history dating back over 4,000 years to ancient Africa. It was cultivated in Egypt around 20000 BCE and later introduced to Asia by Persian traders. Watermelon was brought to Europe by the Moors in the 10th century and later introduced to the Americas by European colonizers. Beyond its refreshing and nutritious fruit, watermelon processing generates a significant by-product: watermelon seeds. These seeds are a treasure trove of nutrients, including protein, oil, carbohydrates, crude fiber, fat, dry matter, and essential minerals^{1,2}.

Despite their potential, watermelon seeds are often discarded or underutilized, representing a significant loss of valuable resources. However, with growing concerns about food security, sustainable resource management, and the need for novel functional ingredients, there is a renewed interest in harnessing the nutritional and industrial potential of watermelon seeds^{2,3}.

Watermelon seeds can be processed into various forms, including cake, oil, and powder, each with unique properties and potential applications in food, feed, and industrial sectors. The cake, for instance, can serve as a valuable source of protein and fiber in animal feed or as a nutritious ingredient in human food products. The oil, rich in unsaturated fatty acids, has potential applications in the cosmetics, pharmaceutical, and food industries. The powder, with its high protein and fiber content, can be used as a functional ingredient in food products, such as baked goods, snacks, and beverages^{2,3}.

Despite the vast potential of watermelon seed derivatives, there is a significant knowledge gap regarding their nutritional and chemical composition. This lack of information hinders the development of value-added products and limits the exploitation of these underutilized resources. Therefore, this study aims to conduct a comprehensive proximate analysis and nutritional comparison of watermelon seed cake, oil, and powder^{4,5}.

By elucidating the chemical and nutritional properties of these derivatives, this research seeks to provide valuable insights into their potential uses and applications, ultimately contributing to the sustainable utilization of watermelon seeds and the development of novel functional ingredients.

MATERIALS AND METHODS

Study area and sites: This study was conducted in Zaria, Kaduna State, Nigeria, within the span of seven months, specifically from May, 2024 to December, 2024. It is located at 11.12'N Latitude and 7.73'E Longitude and it is situated at an elevation of 640 m above sea level. The population of Zaria is 766,000, making it one of the most populous cities in Kaduna.

Sample collection and analysis

Preparation of watermelon seed powder

Extraction of oil from water melon seed: The extraction of oil from water melon seed was carried out using the method described by Abubakar *et al.*^{6,7} 2024 with slight modifications. In this method, 30 g of 425 microns particle size of the water melon seed sample was weighed into a thimble covered with a cotton plug and was inserted into the Soxhlet extractor. The 375 mL of solvent-hexane was poured into the round-bottom flask and connected to the extractor from above. A rubber hose attached to the inlet and outlet holes of the condenser was connected to a water tap, where water flowed in through the inlet hole and flowed out through the outlet. The round-bottom flask containing solvent (hexane) was placed on a heating mantle. The heating mantle was set at 80 and 100°C, which supplied heat to the bottom of the flask placed on the heating mantle. Solvent in the flask gained heat and evaporated through the Soxhlet tube and heated the bottom of the cooled condenser, where the vapors were condensed and the condensate fell back into the body of the thimble to capture the oil in the feed and flow back into the flask. The heating continued for some time, and a mixture of the oil extract and solvent was observed in the flask. The solvent is removed through evaporation^{3,4}.

Preparation of water melon seed cake: The cake was obtained as a by-product of oil extraction from water melon seeds, which will further be processed to give the seed powder after undergoing drying, which was milled into fine powder.

The watermelon seeds were processed using various unit operations.

Particle separation: This was done manually to remove dirt and any unwanted particles from the seed.

Drying: The seeds were either dried naturally (sun-dried) or artificially using the dryer to remove moisture from the seed, in this project, artificial drying was employed.

Size reduction: This was done using a blender or a grinding machine to blend or to grind the sample or the seed to reduce its size to increase the surface area so that sieving will be easier and perfectly done.

Sieving: This was done to separate fine particles from the coarse ones using a sieve.

Extraction of oil: The Soxhlet extraction method was employed, which is the most common extraction technique for oil extraction, in which 30 g of 425 microns particle size of the water melon seed sample was weighed into a thimble covered with a cotton plug and was inserted into the Soxhlet extractor. The 375 mL of solvent-hexane was poured into the round-bottom flask and connected to the extractor from above. A rubber hose attached to the inlet and outlet holes of the condenser was connected to a water tap, where water flowed in through the inlet hole and flowed out through the outlet. The round-bottom flask containing solvent (hexane) was placed on a heating mantle. The heating mantle was set at 80 and 100°C, which supplied heat to the bottom of the flask placed on the heating mantle. Solvent in the flask gained heat and evaporated through the Soxhlet tube and heated the bottom of the cooled condenser, where the vapors were condensed and the condensate fell back into the body of the thimble to capture the oil in the feed and flow back into the flask. The heating continued for some time, and a mixture of the oil extract and solvent was observed in the flask. The solvent is removed through evaporation^{3,5}.

Determination of the yield of watermelon seed oil extracted: The 30 g (W_1) of the sample was placed in the thimble, and about 137 mL of normal hexane was poured into the round flask. The apparatus was heated at 70°C and allowed for 24, 4, 6 and 8 hrs of continuous extraction using the Soxhlet apparatus. The experiment was repeated for different particle sizes, temperatures, and solvents.

Determination of protein: The dried powdered samples were extracted by stirring with 50 mL of 50% methanol (1:5 w/v) at 25°C for 24 hrs and centrifuged at 7,000 rpm for 10 min. 0.2 mL of extract was pipette out and the volume was made to 1.0 mL with distilled water. The 5.0 mL of alkaline copper reagent was added to all the tubes and allowed to stand for 10 min. Then, 0.5 mL of Folin's Ciocalteu reagent was added and incubated in the dark for 30 min. The intensity of the colour developed was read at 660 nm⁶.

Determination of crude lipid content: The lipid content was determined. A clean, dried 500 mL round bottom flask, containing a few anti-bumping granules, was weighed (W_1) and 300 mL of Petroleum ether (40-60°C) for extraction was poured into the flask fitted with a Soxhlet extraction unit. The extractor thimble containing twenty grams of the sample was fixed into the Soxhlet extraction unit. The round-bottom flask and a condenser were connected to the Soxhlet extractor, and cold water circulation was put on. The heating mantle was switched on, and the heating rate was adjusted until the solvent was refluxing at a steady rate. Extraction was carried out for six hours. The solvent was recovered, and the oil was dried in the oven at 70°C for 1 hr. The round-bottom flask containing the oil was cooled in the desiccator and then weighed W_2 .

The lipid content was calculated thus⁷:

$$\text{Crude lipid content (\%)} = \frac{W_2 - W_1}{\text{Weight of sample}} \times 100$$

Determination of crude protein

Protein digestion: Exactly 1.5 g of the defatted sample in an ashless filter paper was dropped into a 300 mL Kjeldahl flask. Twenty-five milliliters of H₂SO₄ and 3 g of digesting mixed catalyst (weighed separately into an ashless filter paper) were dropped into the Kjeldahl flask. The flask was then transferred to the Kjeldahl digestion apparatus. The sample was digested until a clear green colour was obtained. The digest was cooled and diluted to 100 mL with distilled water⁸.

Distillation of the digest: The 20 mL of the diluted digest was measured into a 500 mL Kjeldahl flask containing anti-bumping chips, and 40 mL of 40% NaOH was slowly added by the side of the flask. A 250 mL conical flask containing a mixture of 50 mL of 2% Boric acid and 4 drops of mixed indicator was used to trap the ammonia liberated. The conical flask and the Kjeldahl flask were then placed on the Kjeldahl distillation apparatus, with the tubes inserted into the conical flask and the Kjeldahl flask. The flask was heated to distill out NH₃ evolved. The distillate was collected into the boric acid solution. From the point when the boric acid turned green, 10 min were allowed for complete distillation of the ammonia present in the digest. The distillate was titrated with 0.1 M HCl.

Calculation:

$$N (\%) = \frac{14 \times M \times V_t \times T_v}{\text{Weight of sample (mg)} \times V_a} \times 100$$

$$\text{Weight of sample (mg)} \times V_a$$

$$\text{Crude protein (\%)} = \text{Nitrogen (\%)} (N_2) \times 6.25$$

Where:

M = Actual molarity of acid

T_v = Titre volume of HCl used

V_t = Total volume of diluted digest

V_a = Aliquot volume distilled

Tests for carbohydrates

Fehling's test: The 5 mL of Fehling's solution was added to 0.5 mg of extract and boiled in a water bath. The formation of yellow or red precipitate indicates the presence of reducing power.

Benedict's test: The 5 mL of Benedict's solution was added to 0.5 mg of extract and boiled in a water bath. The appearance of red or yellow, or green precipitate indicates the presence of reducing sugars^{5,6,9}:

$$100 - \frac{(\text{weight in grams [protein+fat+moisture+ash] in 100 g of food})}{([\text{protein+fat+moisture+ash}] \text{ in 100 g of food})} \times 100 = \text{Carbohydrate}$$

$$\text{Carbohydrate} = 100 - \frac{([\text{protein+fat+moisture+ash+ fibre}] \text{ in 100 g of food})}{([\text{protein+fat+moisture+ash+ fibre}] \text{ in 100 g of food})} \times 100$$

Determination of crude fibre content: The 2 g of sample were weighed out into a round-bottom flask. The 100 mL of 0.25 M sulphuric acid solution was added, and the mixture was boiled under reflux for 30 min. The hot solution was quickly filtered under suction. The insoluble matter was washed several times with hot water until it was acid-free. It was quantitatively transferred into the flask, and 100 mL of

hot 0.31 m sodium hydroxide solution was added, and the mixture was boiled again under reflux for 30 min and quickly filtered under suction. The insoluble residue was washed with boiling water until it was base-free. It was dried to constant weight in the oven at 100°C, cooled in a desiccator, and weighed (C1) was then incinerated in a muffle furnace at 550°C for 2 hrs, cooled in the desiccator, and reweighed (C2)^{5,10}.

Calculation:

$$\text{Loss of weight on incineration} = C1 - C2 \times 100 \text{ Weight of original sample}$$

Determination of Ash content: The porcelain crucible was dried in an oven at 100°C for 10 min, cooled in a desiccator, and weighed (W1). The 2 g of the sample were placed into the previously weighed porcelain crucible and weighed (W2). The sample was first ignited and transferred into a furnace, which was then set at 550°C. The sample was left in the furnace for eight hours to ensure proper ashing. The crucible containing the ash was then removed, cooled in the desiccator, and weighed W3.

The percentage ash content was calculated as^{5,9,10}:

$$\text{Ash conten (\%)} = \frac{W3 - W1}{W2 - W1} \times 100$$

Determination of moisture content: A clean crucible was dried to constant weight in an air oven at 105°C, cooled in a desiccator, and weighed (W1). The 2 g of sample were accurately weighed into the previously labeled crucible and reweighed (W2). The crucible was dried in an oven to a constant weight (W3).

The percentage moisture content was calculated thus^{5,10}:

$$\text{Moisture content (\%)} = \frac{W2 - W3}{W2 - W1} \times 100$$

Determination of specific gravity: The specific gravity is determined by dividing the density of a material by the density of water at 4 degrees celsius. For the calculation, the density of the material and that of water must be expressed in the same unit^{3,4}:

$$\text{Specific gravity} = \frac{\text{Density of material}}{\text{Density of water}}$$

Determination of acid value: A known mass of oil sample was weighed and dissolved in a measured volume of neutralized ethanol, and was then titrated with KOH standard solution by adding few drops of phenolphthalein as an indicator until a faint pink color persisted. The acid value was calculated using the formula³⁻⁵:

$$\text{Acid value} = \frac{\text{Volume of KOH used (mL)} \times \text{Normality of KOH}}{\text{Weight of oil sample (g)}} \times 56.1$$

Determination of peroxide value: A known amount of oil sample was weighed and dissolved in a solvent mixture, and an iodide solution was added and mixed well, and the mixture was allowed to stand for a set time in the dark. Distilled water was added, and the mixture was titrated with thiosulfate solution until the blue color of the starch disappeared. Blank titration was done using only the solvent mixture. To calculate the peroxide value, the formula below was used^{3,4,10}:

$$\text{Peroxide value} = [(\text{Volume of sodium thiosulfate used for sample} - \text{Volume of sodium thiosulfate used for blank}) \times \text{Normality of sodium thiosulfate}] \times \left(\frac{1000}{\text{Weight of oil sample}} \right)$$

Determination of iodine value: A known amount of oil sample was reacted with a known amount of iodine, and how much of the iodine was left was measured, and this gave the iodine value^{2,3}.

Determination of saponification value: A known amount of oil sample was weighed, and a known volume of standardized KOH solution was added. The mixture was heated under reflux for a set time to ensure complete saponification.

After which was then cooled to room temperature, followed by the addition of a few drops of phenolphthalein indicator. Titration was conducted with a standard HCL solution until the pink color just disappeared. Blank titration was also conducted with the same amount of KOH solution without the sample to account for any impurity.

Saponification value was calculated using the formula:

$$SV = (56.1 \times (A - B)) \times \frac{F}{W}$$

Where:

- SV = Saponification value
- 56.1 = Molar mass of potassium hydroxide
- A = Volume of KOH used for blank titration
- B = Volume of KOH used for the sample titration
- F = Is the factor of the KOH solution
- W = Weight of the sample^{3,4}

Determination of viscosity: A known quantity of oil was poured into the viscometer's sample holder, and it was ensured to fill the recommended level, and the viscometer was set to the desired temperature of 27°C.

The viscometer (rheometer) was started and was allowed to rotate at a constant speed, and shear stress was applied to the sample. The viscometer measured the resulting shear stress, which allowed the dynamic viscosity of the oil to be determined. The operation was repeated severally to ensure accuracy and reliability of the result^{5,10}.

Determination of refractive index: A small drop of oil was placed on the prism of an Abbe refractometer, which is the standard tool for measuring the refractive index of liquids like oil. The temperature of 27°C was maintained for accurate readings as refractive index changes with temperature, the refractometer directly displays the refractive index value^{3-5,11}.

RESULTS AND DISCUSSION

Table 1-3 explain the results of the proximate analysis conducted on the watermelon seed oil, cake, and powder. This was purposely carried out to find the chemical compositions present in the cake, oil, and powder, and their significance and importance were explained in the discussion section.

Findings from Table 1 and 2 showed that the moisture content, ash content, crude fibre, crude fat, crude protein, carbohydrate, and dry matter are: 10.65, 5.1, 7.9, 2.72, 5.95, 67.68 and 89.35%, in watermelon seed (powder) and that of watermelon cake is 11.18, 3.57, 18.9 and 13.72, 10.47, 42.16 and 89.35%, in Table 1 and 2, respectively for the seed cake and powder.

Table 3 also showed that the watermelon oil has a specific gravity of 0.92 g/cm³, acid value 3.5 mg KOH/g, peroxide value 2.9 mg KOH/g, iodine value 75.0 mg KOH/g, saponification value 180.0 mg KOH/g, viscosity 50.00 cP, and refractive index 1.39 in Table 3, respectively.

Table 1: Proximate analysis of watermelon seed powder

Parameter	Values (%)
Moisture content	10.65
Ash content	5.1
Crude fibre	7.9
Crude fat	2.72
Crude protein	5.95
Carbohydrate	67.68
Dry matter	89.35

Table 2: Proximate analysis of watermelon seed cake

Parameter	Values (%)
Moisture content	11.18
Ash content	3.57
Crude fiber	18.9
Crude fat	13.72
Crude protein	10.47
Carbohydrate	42.16
Dry matter	89.35

Table 3: Proximate analysis of watermelon seed oil

Parameter	Values
Specific gravity	0.92 g/cm ³
Acid value	3.5 mg KOH/g
Peroxide value	2.9 mg KOH/g
Iodine value	75.0 mg KOH/g
Saponification value	180.0 mg KOH/g
Viscosity (at 26.7°C)	50.00 Cp
Refractive index (at 26.7°C)	1.45

This is the quantity of water contained in a material such as soil, rock, ceramics, wood, or even fruits. In the case of watermelon seed powder, it constitutes 10.6 and 11.8%. This shows that moisture is less present in powder than in the cake reason because in the cake it must have undergone extraction, and liquid or solvent must be added to aid the extraction, and as such increased the content of the moisture¹²⁻¹⁴.

Ash content in the powder is 5.1%, and that of the cake is 3.1%, which is significant and proves that the watermelon seed has medicinal value and health benefit. pectin can be extracted from the seed, which is a vital component in pharmaceutical applications^{6,10}.

Crude fibre constitutes 7.9 and 18.9% Of watermelon seed in powder and in cake form, which makes it good for people with issues of diabetes, constipation etc.⁹⁻¹⁴.

Crude fat constitutes 2.72 and 13.72% of watermelon seed in the powder and the cake, which makes it a good source of fat and aids metabolic and structural functions¹⁵⁻¹⁸.

Protein is one of the constituents of watermelon seed with 5.95 and 10.47% in the powder form and in the cake, which makes a good source of protein¹⁹⁻²⁷.

Carbohydrate constitutes 67.68 and 42.16% of the watermelon seed in the powder and the cakes, which makes and an excellent source of carbohydrate needed by all the tissues in our body for physical activities and brain function^{9,14,27}.

Dry matter constitutes 89.35% of the watermelon seed in the cake and in the powder, which makes it excellent for consumption by livestock to boost their nutrients²⁸.

Specific gravity of 0.92 g/cm³ indicates that the oil is less dense than water, non-drying characteristics, and possible high oleic acid content, and its suitability in applications like cosmetic, food, pharmaceutical, soap making industries^{2-13,15}.

Acid value of 3.5 mg KOH/g, high level of free fatty acids, and as such, it makes it suitable for applications like food, cosmetic industries

Peroxide value of 2.9 mg KOH/g indicates a low oxidation level, as such, good quality oil, low rancidity, and longer shelf life. Suitability for various applications includes food and beverage, cosmetic, and pharmaceutical applications^{2-13,15}.

Iodine value of 75.0 mg KOH/g indicates low unsaturated fatty acid content, hence greater stability, lower reactivity, and a different nutritional profile. Suitable for application in food and beverage, cosmetic application^{2-13,15}.

A saponification value of 180 mg KOH/g indicates the typical molecular weight of triglycerides which suggests average soap making, standard physical and chemical properties. It indicates its suitability in applications like soap making, cosmetics, and pharmaceuticals^{2-13,15}.

Refractive index of 1.45 suggests that it is within the typical range of for watermelon seed oil and indicates good quality, authenticity, good physical and chemical properties. Finds suitability in food, pharmaceutical, and cosmetic^{2-13,15}.

CONCLUSION

This comparative study investigated the proximate analysis and nutritional profiles of watermelon seed cake, oil, and powder. The results revealed distinct differences in the nutritional content of each product. The study highlights the potential of watermelon seed products as nutritious and valuable food ingredients. Watermelon seed cake and powder can serve as excellent sources of protein, fibre, ash content, dry matter, and minerals making them suitable for various food applications. Watermelon seed oil, rich in healthy fats, can be used as a cooking oil or cosmetic products.

SIGNIFICANCE STATEMENT

This study is significant because it provides critical insights into the proximate composition and chemical characteristics of watermelon seed cake, oil, and powder, which are essential for unlocking their potential as valuable ingredients in food, feed, and industrial applications. By exploring the nutritional and chemical properties of these underutilized resources, this research contributes to the development of sustainable and innovative solutions for food security, waste reduction, and value-added product development, ultimately benefiting the environment, industry, and human health.

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