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Proximate, Physicochemical, Elemental, and Amino Acid Profile of *Fragaria x ananassa* (Strawberry) Grown on the Plateau

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Abstract

This study evaluated the proximate, phytochemical, elemental, and amino acid composition of *Fragaria* × *ananassa* fruit. Proximate analysis revealed high carbohydrate (33.43%) and ash (28.48%) contents, alongside moderate crude fiber (21.26%) and low protein (3.24%) and fat (1.50%) levels. Qualitative and quantitative phytochemical screenings indicated high concentrations of phenols, saponins, and sugars, with ethanol extracts yielding greater amounts than aqueous extracts. Elemental analysis showed magnesium (18.99 mg/kg) and calcium (14.01 mg/kg) as the most abundant essential metals. However, chromium (0.31 mg/kg) and platinum (0.41 mg/kg) levels slightly exceeded WHO/FAO permissible limits, suggesting potential health concerns. Amino acid profiling identified aspartic acid (0.1493 mg/ml) and glutamic acid (0.0952 mg/ml) as predominant.

Keywords: Strawberry, Fragaria x ananassa, amino acid profile, proximate, phytochemical, elemental analyses.

INTRODUCTION

Strawberries *Fragaria x* ananassa are globally cherished fruits recognized for their unique flavor, attractive red color, and rich nutritional profile [1]. Traditionally, various parts of the strawberry plant—including the fruit, leaves, and roots—have been used in ethnomedicine for treating ailments such as gastrointestinal disorders, liver complaints, skin conditions, and as diuretics or mild laxatives [2,3]. The fruit, in particular, is widely used in herbal teas to relieve diarrhea, support kidney function, and reduce inflammation [4].

Strawberries are notable sources of vitamin C, which contributes significantly to diabetes management [5], enhanced immune function [6], and may retard leukemia development [7]. Among red berries, strawberries particularly stand out due to their qualitative and quantitative characteristics [8]. Widely consumed [9], their nutrient content varies with cultivation systems [10], maturation stages [11], climate conditions [12], and post-harvest practices [13-14].

The primary attributes influencing strawberry acceptability include their red color, sweet taste, and juicy texture [15], with color mainly resulting from anthocyanin accumulation during maturation [11]. Nutritionally, strawberries surpass fruits like citrus, guava, and apple in protein, mineral, and vitamin content [8, 16, 17]. In Nigeria, particularly in Plateau State, strawberries hold economic importance. They are highly valued not only for sensory qualities but also for their phytochemical content [18], offering antioxidant, anticarcinogenic [19], antimutagenic [20], anti-inflammatory [21], and antihypertensive benefits [22].

Despite their cultivation, gaps persist in understanding the nutritional properties of local Nigerian strawberries, with challenges linked to pest management, post-harvest losses, storage methods, and nutritional profiling. Strawberries are particularly perishable, with shelf-life of 1-2 days at room temperature, necessitating prompt consumption or processing [23]. Effective temperature management remains vital to extending post-harvest life. Genetically, strawberries are a hybrid species. The cultivated garden strawberry, *Fragaria* × *ananassa*, popularly known for low-growing habits and aggregate fruits with embedded seeds, is an octoploid hybrid that originated approximately 250 years ago through a natural cross between the North American

species *F. virginiana* and the South American species *F.* chiloensis [8].

Strawberries are processed into various products like jams, yogurts, and cosmetics, leveraging their abundant nutrients, including vitamins (C, E, K, B-complex) and minerals like iron, calcium, and potassium [24-27]. Additionally, their bioactive compounds—phenolic acids, flavonoids, tannins, alkaloids, and terpenoids—possess antioxidant, anti-inflammatory, antimicrobial, and anticancer properties [28-32]. Understanding these attributes is essential for maximizing strawberries' health and economic potential.

Recent studies emphasize strawberries' enhanced bioactive content under organic farming conditions [33], improved antioxidant stability with cold storage innovations [34], and promising applications in functional foods and pharmaceuticals [35]. Advances in metabolomics profiling reveal key compounds responsible for strawberries' therapeutic properties [36], while novel breeding techniques target enhancing their shelf-life and nutritional quality [37]. These developments underline the growing significance of strawberries in health promotion and economic sustainability.

MATERIALS AND METHODS

Collection of Plant Material and Extraction

Fresh strawberry fruits (*Fragaria* × *ananassa*) were collected from Chaha, Vom, located in Jos South Local Government Area (LGA) of Plateau State, Nigeria. The fruits were thoroughly washed with clean water, air-dried at room temperature, and subsequently ground into a fine powder using a mortar and pestle. A total of 307 g of the powdered fruit was weighed and stored in a sterile, dry bag for subsequent analyses.

Extraction and Concentration

For the extraction, 150 g of the finely powdered strawberry (*Fragaria* × *ananassa*) sample was subjected to Soxhlet extraction using 500 mL of a 70:30 ethanol-to-water (v/v) solvent mixture for 24 hours. The extraction was carried out at an ideal temperature range of 60–70 °C, consistent with the recommendations of the Association of Official Analytical Chemists (AOAC, 2019), to prevent the degradation of thermolabile compounds. This hydroethanolic solvent system was chosen for its ability to extract both moderately polar and highly polar compounds, including phenols, flavonoids, tannins, saponins,

terpenes, anthraquinones, alkaloids, coumarins, and reducing sugars [38].

Ethanol enhances the solubility of moderately polar phytochemicals such as flavonoids and terpenes, while water facilitates the extraction of polar compounds like anthocyanins, phenolic acids, sugars, and some amino acids. This dual-polarity ensures efficient recovery of antioxidant-rich bioactive compounds while preserving their structural integrity [39].

Additionally, the extraction aimed to isolate amino acids such as tryptophan, threonine, isoleucine, leucine, lysine, methionine, glutamic acid, phenylalanine, tyrosine, valine, arginine, histidine, alanine, aspartic acid, cysteine, glycine, proline, and serine, which are biologically essential and often co-extracted with polyphenols in aqueous-alcoholic solvents [40]. These amino acids contribute to the nutritional and therapeutic relevance of strawberries.

The resulting extracts were concentrated using a hot water bath at temperatures below 50 °C to retain bioactivity and stored at 4 °C for subsequent qualitative and quantitative analyses.

Proximate Analysis

All the proximate analysis of moisture content, ash content, crude fat, crude fibre, crude protein, and carbohydrate of *Fragaria* × *ananassa* were determined using standard analytical method according to (AOAC 2005).

Determination of Moisture Content

An aluminum moisture pan was washed, dried in an oven, cooled in a desiccator, and weighed. Approximately 100 g of the sample was placed in the pan and weighed. The pan with the sample was then oven-dried at a temperature range of 80–100 °C, checked at 3-hour intervals, until a constant weight was achieved. After cooling, the final weight was recorded, and the moisture content (%) was calculated. Drying within this temperature range, rather than at a fixed 100 °C, was necessary due to the highly perishable nature of strawberries, which typically have a shelf life of only 1–2 days at room temperature. Using a moderate drying range allows for gradual moisture removal while minimizing structural or chemical alterations.

After cooling, the final weight was recorded. Moisture content (%) was calculated as:

$$\frac{(W_2) - (W_3)}{(W_2)} \quad X \quad 100$$

Where,

Weight of pan + wet sample (W_2)

Weight of pan and dry sample (W_3)

Weight of empty pan (W_1)

Determination of Ash Content

A clean crucible was dried in a muffle furnace, cooled in a desiccator, and weighed. About 10 g of the *Fragaria* × *ananassa* sample was added and reweighed. The crucible and sample were gradually heated from 200 °C to 450 °C in a muffle furnace for 4–5 hours to obtain ash. This temperature range was used to prevent charring and spattering due to the fruit's high sugar and moisture content. After cooling in a desiccator, the final weight was recorded.

Ash content (%) was calculated as:

$$\frac{(W_3) - (W_1)}{(W_2) - (W_1)} \text{ X } 100$$

Weight of crucible + sample (W_2)

Weight of crucible + ash (W_3)

Weight of empty crucible (W_1)

Determination of Fibre Content

10 g of *the* sample was weighed into a 50 mL conical flask, and 100 mL of 1.25% sulfuric acid (H_2SO_4) was added. The mixture was gently boiled for 30 minutes under reflux and filtered through a poplin cloth using a Buchner funnel. The residue was washed with hot distilled water and then treated with 100 mL of 1.25% sodium hydroxide (NaOH), 2–3 mL of distilled water, and 3–5 drops of vegetable oil as an anti-foaming agent. It was again gently boiled for 30 minutes, filtered, and thoroughly rinsed with hot distilled water, 10–15 mL of 95% ethanol, and 10–15 mL of ether. The residue was oven-dried at 105 °C for 2 hours, cooled in a desiccator, and weighed. It was then ashed in a muffle furnace at 300 °C for 30 minutes, cooled, and reweighed. Crude fibre (%) was calculated as. Crude fibre (%) was calculated as:

$$\frac{(W_2) - (W_3)}{(W_2)} \text{ X } 100$$

Where,

Weight of residue + crucible (W_2)

Weight of sample from the muffle furnace (W_3)

Weight of sample (W_1)

Determination of Crude Fat Content

A weighed thimble was filled with about 10 g of the sample and weighed again. A 500 mL round-bottom flask was also weighed. The Soxhlet extractor, fitted with a reflux

condenser, was used with petroleum ether as solvent. The system was heated to allow extraction for 5–6 hours. Afterward, the solvent was evaporated, and the residue was oven-dried at 100 °C, cooled in a desiccator, and weighed. The % extracted lipid is given by the formula. Crude fat (%) was calculated as:

Weight of the residue (W_4)

Weight of round bottom flask (W_3)

Weight of the sample (W_2)

Weight of the thimble (W_1)

Determination of Protein Content

The crude protein content was determined by the Kjeldahl method involving digestion, distillation, and titration:

Digestion: 0.5 g of the sample was mixed with 2 mL HCl, 3 mL HNO_3, 45 mL distilled water, and 1 g mercury oxide (catalyst). The mixture was digested on a hot plate under a fume cupboard at 250 °C for 30–40 minutes.

Distillation: The digest was distilled using a Kjeldahl apparatus and diluted to 100 mL with distilled water.

Titration: The distillate was titrated against 0.02 N HCl.

The percentage nitrogen and protein were calculated as:

Nitrogen free extract

$$\frac{[(V_1 - V_2)(14.01 \times N)]}{W \times 1000} \times 100$$

%NFE =

Where:

N = Normality of acid, W = Weight of the sample, V_1 = Titre value, V_2 = Blank value, %protein = % Nitrogen free extract x conversion factor.

Conversion factor, 6.25 was used

Determination of Carbohydrate Content

The carbohydrate can be represented by the stoichiometric formula (C_nH_2O) where n is the number of carbon in the molecules. Therefore, the ratio of carbon to hydrogen to oxygen is 1:2:1 in the carbohydrate molecules. The total carbohydrate was determined by difference in method in which the sum of the % of moisture, ash, crude fibre, crude fat and protein content were subtracted from the sample. That is, (100 - % moisture content, ash content, crude fiber content, crude fat content and protein content).% Carbohydrate = 100 − (% Moisture + % Ash + % Crude Fibre + % Fat + % Protein)

Qualitative Phytochemical Analysis

The extract was screened for the presence of major phytochemicals following the AOAC Official Methods of Analysis (2019 edition) [41].

The tests conducted include phenols, flavonoids, tannins, saponins, terpenes, anthraquinones, alkaloids, coumarins, and reducing sugars.

Each test was performed in triplicates and interpreted based on specific observable colour changes or precipitate formation.

Test for Phenols: To 1 mL of the extract, 2 mL of distilled water and a few drops of 10% ferric chloride were added. The formation of a bluish-green or dark blue colour indicated the presence of phenolic compounds.

Test for Flavonoids (Sodium Hydroxide Test): 1 mL of 10% sodium hydroxide solution was added to 1 mL of the extract. The appearance of a bright yellow colouration, which turned colourless upon the addition of dilute hydrochloric acid, confirmed the presence of flavonoids.

Test for Tannins: 1 mL of 5% ferric chloride solution was added to 1 mL of the extract. A blue-black or greenish-black precipitate indicated the presence of tannins.

Test for Saponins (Froth Test): 2 mL of the extract was mixed with 2 mL of distilled water and shaken vigorously in a test tube. Formation of a stable, persistent froth layer of at least 1 cm height after 10 minutes indicated the presence of saponins.

Test for Terpenes (Salkowski Test): To 2 mL of the extract, 2 mL of chloroform was added, followed by 2 mL of concentrated sulfuric acid (carefully poured down the side). The formation of a reddish-brown interface confirmed the presence of terpenes.

Test for Anthraquinones (Borntrager's Test): 1 mL of the extract was boiled with 0.02 mL of 5% dilute sulfuric acid, cooled, and filtered. The filtrate was shaken with 5 mL of chloroform. The chloroform layer was separated and shaken with 1 mL of 10% ammonia solution. A pink to red colouration in the ammoniacal layer indicated the presence of free anthraquinones.

Test for Alkaloids (Mayer's Test): 2 mL of the extract was acidified with 1 mL of 5% dilute hydrochloric acid, followed by the addition of a few drops of Mayer's reagent. The formation of a creamy white or greenish precipitate confirmed the presence of alkaloids.

Test for Coumarins: 2 mL of the extract was mixed with 2 mL of 2% sodium hydroxide solution and heated in a boiling water bath for 3 minutes. Four drops of concentrated hydrochloric acid were then added. The

appearance of a cloudy white precipitate or fluorescence under UV light indicated the presence of coumarins.

Test for Reducing Sugars (Benedict's test): 1 mL of the extract was treated with 2 mL of Benedict's reagent and heated in a boiling water bath for 5 minutes. A brick-red, yellow, or green precipitate indicated the presence of reducing sugars.

Quantitative Analysis Method

The extract (10 g) was weighed and dissolved in 1.0 ml of distilled water. The solution was transferred to centrifuge for 10 minutes. 1.0 ml of the upper layer was measured and transferred to the spectrophotometer. To read the absorbance at 436 nm, distilled water was used as blank. Calculating at absorbance 436 nm, =

$$\frac{A \times V \times DF}{\varepsilon \times L \times W}$$

Where, A = Absorbance, mV = Volume of extract, DF = Dilution factor, ε = Molar absorptivity, L = Path lens, W = Sample weight.

Elemental Analysis

Fifty gram of dried sample was placed in a crucible and heated in a muffle furnace, gradually raising the temperature from 100 ℃ to 250 ℃ for 1–2 hours. After cooling in a desiccator, 20 g of the ash was digested by adding 20 mL HNO_3, 10 mL HCl, and 10 mL distilled water. The mixture was heated on a hot plate under a fume hood for 30–40 minutes, cooled, and diluted with 50 mL distilled water. It was then filtered twice using Whatman No. 1 filter paper. The filtrate was analyzed using Atomic Absorption Spectrophotometry (A.A.S) with element-specific hollow cathode lamps and adjusted wavelengths to detect essential and heavy metals.

Amino Acid Content

A 100 g sample of the extract was placed in a round-bottomed flask, and 6 mL of sulfuric acid was added. The flask was connected to a reflux apparatus and gently heated for 24 hours to hydrolyze peptide bonds. After cooling in a desiccator, the aqueous layer containing amino acids was separated and drained into a clean container. The amino acids were then analyzed using an amino acid analyzer.

RESULTS AND DISCUSSION

The proximate composition of *Fragaria* × *ananassa* fruit powder (Table 1) reveals a high carbohydrate ($33.43 \pm 1.63\%$) and crude fibre content ($21.26 \pm 0.15\%$), suggesting its significant contribution to energy provision and digestive health. This finding agrees with Zhang *et al.* [42], who reported that fruits like strawberries offer

substantial carbohydrates and fibre beneficial for maintaining gut health. The low fat content ($1.50 \pm 0.17\%$) and moderate protein level ($3.24 \pm 0.14\%$) further support its value for low-fat, balanced diets, aligning with observations by de Souza *et al.* [43] regarding the nutritional properties of fruits.

Phytochemical screening (Table 2) indicated the presence of phenols, flavonoids, saponins, and tannins in both ethanolic and aqueous extract, with ethanolic extraction yielding higher quantities (Table 3). The superior extraction efficiency of ethanol corresponds to the findings of Bhattacharjee *et al.* [44], who emphasized the effectiveness of organic solvents in retrieving bioactive compounds. Notably, high concentrations of phenols (6.01 ± 0.01 mg) and saponins (5.70 ± 0.02 mg) were recorded in the ethanolic extract, which implies a strong antioxidant potential, consistent with Shahidi and Ambigaipalan [45] and Khan *et al.* [46], who highlighted the health benefits of phenolic and saponin- rich foods. Flavonoids and tannins, also detected at appreciable levels, are well known for their anti-inflammatory, cardioprotective, and antimicrobial activities [47, 48].

The elemental composition analysis (Table 4) showed magnesium (18.99 ± 0.02 mg/kg) and calcium (14.01 ± 0.01 mg/kg) as the dominant minerals. Magnesium's abundance is critical for enzymatic reactions, muscle function, and metabolic regulation, in agreement with Rosanoff *et al.* [49]. Calcium is similarly vital for bone health and metabolic functions [50]. Minor elements such as iron (0.55 ± 0.01 mg/kg) and sodium (1.50 ± 0.00 mg/kg) were also detected but in lower concentrations, indicating that while strawberries contribute to micronutrient intake, they may need to be complemented with other dietary sources to meet daily requirements.

Heavy metal analysis (Table 5) revealed very low concentrations of cadmium(0.02 ± 0.01 mg/kg) and lead (0.01 ± 0.00 mg/kg), both within safe limits established by WHO [51]. However, chromium (0.31 ± 0.01 mg/kg) slightly exceeded the recommended threshold (0.25 mg/kg), raising concerns about environmental contamination during cultivation. This result highlights the ongoing need for environmental monitoring

in agricultural practices, as stressed by Gupta and Gupta [52].

Amino acid profiling (Table 6) showed that aspartic acid (0.1493 ± 0.0002 mg/ml) and glutamic acid (0.0952 ± 0.0003 mg/ml) were predominant, with essential amino acids like leucine, lysine, and threonine also present. These amino acids play pivotal roles in protein synthesis, neurotransmitter functions, and overall metabolic health [53]. The presence of essential and non-essential amino acids enhances the nutritional significance of *F.* × *ananassa*, corroborating the functional food potential described by Kaur and Kapoor [54].

Table 1. Result for Proximate Composition of *Fragaria x ananassa* fruit

Parameter	**Percentage value (%)**
Moisture content	12.09±0.60
Ash content	28.48±0.54
Crude fibre content	21.26±0.15
Crude fat	1.50±0.17
Crude protein	3.24±0.14
Carbohydrate	33.43±1.63

Value reported as Mean ± SD for triplicate analysis n = 3

Table 2. Phytochemical Screening Result for Fruit of *Fragaria x ananassa*

Constituent	Extract with ethanol	Extract with distilled water
Phenols	+++	+++
lavonoids	++	++
Tannins	++	+
Saponins	+++	++
Terpenes	++	+
Anthraquinones	+	-
Alkaloids	++	+
Coumarins	++	++
Sugar	+++	+++

Keys: *+++ = High concentration, ++ = Moderate concentration + = Low concentration,* **- =** *Not detected*

Table 3. Quantitative Phytochemical Screening Test Result for fruit of *Fragaria x ananassa*

Constituent	Extract with ethanol (mg)	Extract with distilled water (mg)
Phenols	6.01±0.01	4.50±0.01
Flavonoids	3.20±0.01	2.50±0.00
Tannins	3.50±0.02	1.90±0.01
Saponins	5.70±0.02	2.80±0.01
Terpenes	3.00±0.01	1.60±0.01
Anthraquinone	1.40±0.02	0.05±0.02
Alkaloid	3.90±0.02	2.00±0.01
Coumarins	3.00±0.00	2.53±0.01
Sugar	5.03±0.03	4.98±0.03

Value reported as Mean ± SD for triplicate analysis n = 3

Table 4. Result for Essential metal concentration of *Fragaria x ananassa*

Elements	Concentration (mg/kg)
Fe	0.55±0.01
Na	1.50±0.00
Ca	14.01±0.01
Mg	18.99±0.02

Value reported as Mean ± SD for triplicate analysis n = 3

Table 5. Result for Heavy Metal Concentration of *Fragaria x ananassa*

Elements	Concentration (mg/kg)	WHO/FAO permissible limit (mg/kg) (WHO 2016)
Cd	0.02±0.01	0.20
Pb	0.01±0.00	0.30
Cr	0.31±0.01	0.25
Pt	0.41±0.01	3.00

Value reported as Mean ± SD for triplicate analysis n = 3

Table 6. Result for Amino acid constituent of *Fragaria x ananassa*

Constituent	Concentration (mg/ml)
Tryptophan	0.0084±0.0001
Threonine	0.0196±0.0001
Isoleucine	0.0135±0.0002
Leucine	0.0326±0.0002
Lysine	0.0242±0.0003
Methionine	0.0025±0.0003
Glutamic acid	0.0952±0.0003
Phenylalanine	0.0184±0.0003
Tyrosine	0.0227±0.0002
Valine	0.0184±0.0003
Arginine	0.0255±0.0002
Histidine	0.0122±0.0001
Alanine	0.0319±0.0001
Aspartic acid	0.1493±0.0002
Cysteine	0.0065±0.0002
Glycine	0.0245±0.0002
Proline	0.0193±0.0002
Serine	0.0233±0.0002

Value reported as Mean ± SD for triplicate analysis n = 3

CONCLUSION

The study confirms that strawberries from Plateau State are nutritionally rich, characterized by significant levels of carbohydrates, fibres, essential minerals, and bioactive compounds. Overall, the findings substantiate the nutritional and therapeutic value of *Fragaria* × *ananassa*, positioning it as a promising candidate for functional food development. However, the slight chromium contamination and the high ash content observed necessitate improved environmental monitoring and management to prevent potential contamination from soil or agricultural inputs. Ensuring food safety, while promoting local cultivation, consumption, and processing of strawberries, could offer substantial health and economic benefits.

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If not applicable, please expunge this section.

CONFLICT OF INTEREST

Authors declare no conflict of interest.

ETHICAL STATEMENT

This work required no ethical statement.

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