



Evaluation of Secondary Metabolites and Nutritional Composition of N-Hexane Extract of Onion (*Allium Cepa*) Peel

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ABSTRACT

Allium cepa (onion) belongs to the family Liliaceae and found mainly in temperate region. Due to its significant medicinal properties, nutritional worth, and energy value, *Allium cepa* (onion) imparts numerous health benefits to users. This study was conducted to investigate the phytochemical, mineral and vitamin composition of *Allium cepa* (onion) peels. The phytochemical screening revealed that the peels contained moderate amount of alkaloids (29.4%), and flavonoid (16.1%). Phenol was present in minute quantity of 0.64% and saponin content was appreciable (8.5%). However, tannin was absent. The phytochemical screening also revealed the presence of ANTI nutrients such as oxalate and phytate in minute quantities of 2.64% and 0.17% respectively. However, hydrogen cyanide was absent. The minerals screened for were Ca, Mg, Fe and P, and the result showed that the plant was rich in Ca (12.65%), followed by Mg (1.24%), while the other two were more or less absent. The vitamin analysis revealed the plant has extremely low vitamin contents of A and C. The result of the analysis, having shown onion is a good carrier of Phytochemicals such as alkaloid, flavonoids, saponins, and minerals such as Ca, and Mg, concludes that onion peel can contribute appreciably to human health as an antimicrobial as well as an alternative source of nutritional supplements in as much as it suggests further investigation for other nutritional components of the plant part.

Keywords: *Allium cepa*; Phytochemistry; Mineral; Vitamins; n-Hexane

INTRODUCTION

Many higher plants, otherwise called herbs, are major sources of natural products used as pharmaceuticals, agrochemicals, flavouring agents, fragrances, and ingredients in food additives [1]. *Allium cepa* is a twin herb to *Allium sativum* (garlic), and is a dominating member of kitchen cabinet used as a wet condiment having ability to get rid of variety of disease conditions of the body and give miraculous relieve [2]. *Allium cepa* (onion) belongs to the family Liliaceae and found mainly in temperate region [3]. Onion is a crop of great economic importance in all over the world [4]. It is widely cultivated, second only to tomato, and is a vegetable bulb crop known to most cultures and consumed worldwide [5].

The major nutrient attributed to onion is vitamin C, although other vitamins are contained in it [6]. Due to its significant medicinal properties, nutritional worth, and energy value, *Allium cepa* (onion) imparts numerous health benefits to users [7]. Onion has been reported to possess anticarcinogenic properties [8,9], especially in oesophageal, stomach, lungs, prostate and developing brain carcinomas [10]. Its aqueous extract was also reported to have antileishmanial activity against *Leishmanial promastigotes* [11]. Further other activities of *Allium cepa* includes antifungal, antibacterial, promoting cardiovascular health, reducing high blood pressure and insulin resistance, etc. [8,9].

The bioactivity of plants is a function of phytochemicals [12], and these phytochemicals (from the Greek word photo, meaning plant) are bioactive chemical compounds, naturally occurring in plants and provide health benefits for humans beyond those attributed to micro and macro nutrients [13]. Phytochemicals are not essential nutrients and are not required by the human body for sustaining life. However, they have important properties to prevent or fight some common diseases [14]. Some of them are alkaloids, which have been reported to possess anti-microbial activities [15], anti-malarial activity, anti-hypertensive activity, to treat rheumatism [16], tannins, possessing anti-microbial activity [17], anti-diarrhoeal and anti-inflammatory activities, etc. [18], flavonoids, reported to possess antioxidant properties [16], be anti-allergens, exhibit anti-inflammatory potential [15,16], and as well, possess anti-microbial potential [19-21], saponins, reported to be an anti-bacterial, anti-fungal, anti-viral, insecticidal, and anti-inflammatory agent, etc. [16], terpenoids, reported to be anti-bacterial, anti-fungal, anti-viral, anti-protozoan, anti-allergen [22], antioxidant, anti-inflammatory, anti-diabetic agent [14], cardiac glycosides which are used traditionally to treat cardiac arrhythmias and congestive heart failure [14].

Vitamins are organic chemical compounds found in tiny amounts in natural food stuffs. They are needed and/or required by an organism as a vital nutrient to sustain life because they play an important role in normal metabolism process, growth, and vitality. They are usually received either from diet (which is considered their major source), or from supplements [23]. Depending on chemical nature, vitamins are classified as either fat-soluble (vitamins A, D, E, K), or water-soluble (the vitamin Bs and C) [24].

Minerals are solid inorganic substances that form crystals, and are classified depending on how much of them are needed. Many minerals are critical for enzymes function, others are used to maintain fluid balance, build bone tissues, synthesize hormones, transmit nerve impulses, and contract and relax muscles, and protect against harmful free radicals. Minerals possess definite chemical composition and structure, nearly five percent of human body is composed of inorganic materials (minerals) [25]. Even though minerals yield no energy, they have important roles to play in many activities in the body, and every form of living matter requires these inorganic elements or minerals

for their normal life processes. However, some of these elements cause a health hazard when foods containing them are ingested [25].

Minerals are classified as trace [25,26], which are only required in few milligrams or less (e.g., molybdenum, selenium, zinc, iron, iodine), and macro minerals; which are required in hundreds of milligram (e.g., calcium, magnesium, potassium, sodium, and phosphorus) [26].

MATERIALS AND METHODS

Collection of Plant Material

Fresh onion (*Allium cepa*) peels were gathered from Naze market in Owerri, Imo State and was identified by a botanist from the Department of Environmental Microbiology Federal Polytechnic Nekede, Owerri, Imo State.

Preparation of Plant Material

The onion peels were washed with clean water and air dried at room temperature for 2-3 weeks, after which, they were grinded into powder using a pulverizing machine, and stored in an air-tight container till extraction.

Extraction of the Plant Material

The extraction method was done by maceration. 150 g of the powdered plant material was soaked in 600 ml of N-hexane. The mixture was kept for 3 days in tightly sealed vessels at room temperature and agitated several times daily. It was then filtered through muslin cloth. Further extraction of the residue was repeated 3-5 times until the extraction liquid was clear and colourless, indicating that no more extraction from the plant materials was possible. Then the extracted liquid was subjected to water bath evaporation to remove the solvent.

Phytochemical Analysis

Qualitative analysis

Alkaloid determination: 0.5 g of the extract was mixed in 8 ml of 1% HCL, this was warmed and filtered. 2 ml of the filtrate was treated with Mayer's reagent. The presence of alkaloid showed turbidity [27].

Phenol determination: 10 mg of the extract was treated with 2 drops of ferric chloride solution. Formation of bluish black color indicated the presence of phenol [27].

Flavonoid determination: 0.5 g of the extract was dissolved in 5 ml ethanol, and 1 ml concentrated sulphuric acid was added, followed by 0.5 g of Mg. A pink or red coloration that disappear on standing for 3 minutes indicates the presence of flavonoids [27].

Tannin determination: 0.5 g of the extract was dissolved in 2 ml of distilled water, 2 drops of diluted ferric chloride solution was added. A dark green or blue green coloration would indicate the presence of tannins [27].

Saponin determination: 0.5 g of the extract was added to 5 ml of distilled water in a test tube. The solution was shaken vigorously and observed for a stable persistent froth for 20 minutes [27].

Oxalate determination: The titration method as described by Agbaire [28] was followed. 1 g of sample was weighed into 100 ml conical flask. 75 ml 3 M H₂SO₄ was added and stirred for 1 hr with a magnetic stirrer. This was filtered using a Whatman No 1. filter paper. 25 ml of the filtrate was then taken and titrated while hot against 0.05 M KMnO₄ solution. The presence of oxalate would be indicated by a faint pink color which would persist for at least 30 sec.

Phytate determination: 2 g of the sample was soaked in 100 ml of 20% HCl for 3 hours, and then filtered. 50 ml of the filtrate was measured into 250 ml beaker and 100 ml distilled water was added, followed by 10 ml of 0.3% ammonium thiocyanate solution as an indicator. The solution was titrated with standard iron (III) chloride solution which contained 0.00195 g iron per 1 ml, and an end point of 5 minutes persistent brownish-yellow coloration would mean the presence of phytate [29].

Hydrogencyanide determination: The alkaline titration method of Agbaire [28] was used for the determination. 100 ml of sample was steam-distilled into a solution of NaOH. The distillate was treated with dilute KI solution. This was then titrated against 0.02 M AgNO₃ solution. A change from clear to a faint but permanent turbid solution as an endpoint would mean the presence of hydrogen cyanide.

Quantitative analysis

Alkaloid quantification: 1 g of sample was weighed in a 250 ml beaker and 150 ml 10% acetic acid prepared in ethanol was added to it. The solution mixture was covered and allowed to stand for a time interval of 4 hrs. After the required time interval, the mixture was filtered and the resultant filtrate was concentrated on water-bath to reduce its volume up to a quarter of its original volume. Conc. NH₄OH was added to the extract sample drop wise until complete precipitation. The solution was allowed to settle, precipitate collected, washed with dilute NH₄OH and filtered. The residue obtained was completely dried and weighed to calculate the percentage of alkaloids in the test sample [30].

Phenol quantification: First of all, fat-free sample was prepared by defatting 1 g sample with 100 ml di-ethyl ether employing soxhlet apparatus for a time interval of 2 hrs. Phenol quantification was done by utilizing spectrophotometric method. The fat-free sample was boiled for 15 min with ether (50 ml) for complete extraction of phenolic components. 5 ml extract was pipetted into 50 ml-flasks, and 10 ml distilled water was added. After this, 2 ml NH₄OH solution was added followed by addition of 5 ml conc. amyl alcohol. After making samples up to the mark, they were left to react for 30 min to develop color which was measured at 505 nm against standard curve [30].

Flavonoid quantification: 1 g plant sample was repeatedly extracted with 100 ml 80% aqueous methanol kept at room temperature and finally filtered through Whatman # 42 filter paper (125 mm). The filtrate was transferred into crucible and left for complete evaporation over water bath. After drying, the sample was weighed until constant weight obtained [30].

Tannin quantification: 500 mg sample was weighed in 50 ml plastic bottle. 50 ml distilled water was added and left for shaking on mechanical shaker for a time period of 1 hrs. This was filtered and made up to the mark in 50 ml volumetric flask. 5 ml of filtrate was pipetted out in a test tube and mixed thoroughly with 2 ml FeCl₃ (0.1 M) in HCl (0.1 N) and potassium ferrocyanide (0.008 M). The absorbance at 120 nm was measured spectrophotometrically against the standard curve [30].

Saponin quantification: Determination Procedure of Ali *et al.* [30] was used for saponin quantification. 1 g plant sample was dispersed in 150 ml of 20% aqueous ethanol. The suspension was heated for 4 hrs on water-bath at 55°C with continuous stirring. The mixture after filtration was re-extracted with another 150 ml 20% aqueous ethanol. The combined extracts were reduced over water-bath to 40 ml by heating at 90°C. The concentrate was transferred to separating funnel after addition and vigorous shaking with 20 ml diethyl ether. The aqueous layer obtained was

collected and ether layer discarded. This purification step was repeated. 50 ml n-butanol was added and combined n-butanol extracts were washed twice with 10 ml aqueous NaCl (5%). The remaining solution was transferred to water-bath and heated till complete evaporation. After evaporation, samples were dried in oven to achieve a constant weight. Saponin content was calculated as percentage yield of the sample.

Oxalate determination: The titration method as described by Agbaire [28] was followed. 1 g of sample was weighed into 100 ml conical flask. 75 ml 3 M H₂SO₄ was added and stirred for 1 hr with a magnetic stirrer. This was filtered using a Whatman No 1. filter paper. 25 ml of the filtrate was then taken and titrated while hot against 0.05 M KMnO₄ solution until a faint pink color persisted for at least 30 sec. The oxalate content was then calculated by taking 1 ml of 0.05 m KMnO₄ as equivalent to 2.2 mg oxalate.

Phytate content determination: This was determined by the method of Agbaire [28] 100 ml of the sample was extracted with 3% trichloroacetic acid. The extract was treated with FeCl₃ solution and the iron content of the precipitate was determined using Atomic Absorption spectrophotometer. A 4:6 Fe/P atomic ratio was used to calculate the phytic acid content.

Hydrogen cyanide determination: The alkaline titration method of Agbaire [28] was used for the determination. 100 ml of sample was steam-distilled into a solution of NaOH. The distillate was treated with dilute KI solution. This was then titrated against 0.02 M AgNO₃ solution. The endpoint was obtained when there was a change from clear to a faint but permanent turbid solution. The hydrogen cyanide content was determined by taking 1 ml of 0.02 m AgNO₃ as equivalent to 1.08 mg HCN.

Mineral analysis

Test for calcium: The test was done according to Ibitoye [29]. 0.4 g of the sample was weighed into a beaker, and 10 ml conc. HCl was added. The mixture digested, and then small quantity of water was added. The resulting solution was filtered into a 50 ml volumetric flask and was made up to the mark with water, after which, 5 ml was pipetted into a conical flask and 50 ml H₂O was added, followed by 5 ml of 20% KOH, 5 drops of 2% KCN, 5 drops of 5% hydroxylamine hydrochloride, and a pinch of calcium indicator, and the mixture was titrated with 0.01 M EDTA until it gave a deep blue color.

Test for magnesium: The analysis was done as described by Achikanu et al., [31]. 5 g of the sample were digested with 10 ml of 5 N conc. hydrochloride. The mixture was placed on a water bath and evaporated to almost dryness, after which, it was cooled and filtered into 100 ml standard flask and diluted to volume with distilled water. 5 ml of the sample was pipette into a test tube, and then 1 ml of 0.67 N H₂SO₄ was added. 1 ml of 0.05% titan yellow was added also. 1 ml of 0.01% gum acacia was added and 2 ml of 10% NaOH was also added and the solution was mixed and absorbance was taken at 520 nm against the blank.

Test for Iron (Fe): The analysis was done as described by Butnariu et al., [32]. 10 g of the sample was weighed into a pre-weighed crucible and ashing was done in a muffle furnace at 400 degrees centigrade. The crucible was cooled in a desiccator at room temperature and the ash was moistened with concentrated sulphuric acid and heated on a heating mantle until no sulphuric acid fumes was given off. The ash in the crucible was then heated to a constant weight in a muffle furnace at 600 degrees centigrade, and then 0.5 g of the ash was dissolved in 50 ml of 5% HCl to

obtain a solution for analysis using the AAS. Standard solution was prepared and calibration curve was drawn using AAS.

Test for phosphorus: Phosphorus content was determined as described by Achi *et al.*, (2017). [33] Ash solution of the plant sample was prepared by weighing 5gms into a crucible and ashing it at 550°C in a muffle furnace for 5 hours, and the residue was dissolved in 100 ml of deionized water. Suitable salt of phosphorus was used to make the standard, and the lamp was fixed. The standard solution was injected to calibrate the AAS using acetylene gas. An aliquot of the ash solution was injected and the concentration was obtained from the AAS.

Vitamin analysis

Test for vitamin A: 1 g of the sample was weighed and dissolved in 20 ml N-hexane in a test tube for 10 minutes. 3 ml of the upper extract was transferred into a dry test tube and was evaporated to dryness. After which, 0.2 ml acetic anhydride chloroform reagent was added, followed by 2 ml of 50% Trichloroacetic Acid (TCA) in chloroform and the absorbance was read at 620 nm [31].

Test for vitamin C: The analysis was carried out as described by Achikanu *et al.*, [31]. 5 g of the sample was weighed and dissolved in 10 ml of 0.4% italic acid in a test tube for 10 minutes, followed by 5 minutes centrifuging and then filtration. 1 ml of the filtrate was transferred into a dry test tube. 9 ml of 2,6-dichlorophenol indophenol was added and the absorbance was taken at 520 nm (Tables 1-4).

RESULTS

All the results are shown in Tables 1-4 below.

Table 1. Qualitative phytochemical result of onion (*Allium cepa*) peels

Parameters	Plant Extract
Alkaloid	++
Phenol	+
Flavonoid	++
Tannin	-
Saponin	+
Oxalate	+
Phytate	+
Hydrogen cyanide	-

Table 2. Quantitative phytochemical result of onion (*Allium cepa*) peels

Parameters	Composition
Alkaloid	29.40%
Phenol	0.64%
Flavonoid	16.10%
Tannin	0.1%
Saponin	8.5%
Oxalate	2.64%
Phytate	0.17%
Hydrogen cyanide	700 mg/kg (0.07%)

Table 3. Mineral composition of onion (*Allium cepa*) peels

Parameters	Composition (%)
Calcium (Ca)	12.65
Magnesium (Mg)	1.24
Iron (Fe)	0.06
Phosphorus (P)	0.00%

Table 4. Vitamin composition of Onion (*Allium cepa*) peels

Parameters	Composition (mg/100 g)
A	0.4359
C	1.106

DISCUSSION

The result of the analysis revealed that the plant has rich alkaloid content (29.4%), followed by flavonoid (16.10%), saponin (8.5%), oxalate (2.64%), phenol (0.64-1%), then phytate (0.17%). Alkaloid-containing plants can be used as neuropharmaceuticals, anticancer agents, anti-microbials, as well as sedatives and insecticides [34]. The flavonoid content in the plant means the plant part can be employed as a cyto toxicant, an anti-microbial agent, anti-inflammatory agent, anti-tumour agent and as a powerful antioxidant [35,36]. The saponin in content of onion peel presents it as an antimicrobial, insecticidal agent. It further means that the plant can be employed as an antioxidant and hypoglycemia agent [37]. Phenol in plants means they have anti-bacterial and anti-fungal potential [38]. However, the phytate and oxalate content of onion peel stands as a threat to its use, as phytate are capable of chelating bivalent minerals like Ca, Fe, Zn, and Cu, as well as starch, protein and enzymes which determines the bioavailability of these components [39]. Oxalate when taken in excess can cause diarrhoea, gastroenteritis and renal damage [40].

The mineral composition of onion peel showed it is rich in Ca (12.65%), followed by Mg (1.24%). Calcium helps in regulating hormonal secretion and nerve impulse, as well as maintains skeleton [41]. Magnesium serves as an anti-inflammatory agent [42] and helps lower blood pressure in adults [43].

CONCLUSION

The result of the analysis, having shown onion is a good carrier of Phytochemicals such as alkaloid, flavonoids, saponins, and minerals such as Ca, and Mg, concludes that onion peel can contribute appreciably to human health as an antimicrobial as well as an alternative source of nutritional supplements.

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