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# Chemical Composition of Water Lily (*Nymphaea lotus*) Bulbs

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# Abstract

Water lily (Nymphaea lotus) bulbs from Mankhanwa River in Ibi - Nigeria were quantified for their biochemical composition. Proximate composition, anti-nutritional factors, minerals, vitamins and phytochemical contents in the plant bulb were assayed using Association of Official Analytical Chemists (AOAC) standard methods. Values were obtained in duplicate and results tabulated using mean  $\pm$  standard deviation. The result of the proximate analysis shows the following composition: crude fat (5.07  $\pm$ 0.01%), crude fiber (13.30  $\pm$  0.02), crude protein (21.66  $\pm$  0.01), ash (8.34  $\pm$  0.01), moisture (9.72  $\pm$  0.00) and carbohydrate (41.92  $\pm$  0.06%). The present research also revealed that Nymphaea lotus bulb contains some vital phytochemicals: phytate (3.68  $\pm$ 0.01 mg/100g), tannin (13.63  $\pm$  0.01), saponins (4.89  $\pm$  0.01) and lycopene (1.96  $\pm$  0.01) and very rich in minerals: phosphorus (635.39  $\pm$  0.01 mg/100g), potassium (742.89  $\pm$ 0.04), sodium (431.53  $\pm$  0.01), zinc (8.16  $\pm$  0.01) and magnesium (87.46  $\pm$  0.01). Other minerals obtained in this research include: cadmium, lead and copper in trace quantity. Vitamin A  $(51.36 \pm 0.02 \text{ mg}/100\text{g})$  and C  $(24.65 \pm 0.02)$  were found in moderate quantities. Other vitamins obtained in trace amount include: B1, B2 and E. Anti-nutrients were found to also exist in trace quantities. Information from this biochemical quantification suggests that the bulb from this important aquatic plant can be incorporated as a good source of nutrients in food industries for flour production, pharmaceutical industries for drug production as well as in the cosmetic industries.

# **1. Introduction**

An observable feature in most of the lakes and ponds in Taraba State, Nigeria is the richness of their aquatic flora. Water lilies among the aquatic flora are the most abundant in all the water bodies in the state. Their presence to an observer is that of an aesthetic and serene aquatic environment. The white water lily type is predominant and grows freely in most environments in Taraba State more specifically in Ibi (where samples for this research were obtained) and Gindin-Doruwa areas of the state. The plant has wide-round leaves that float flat on the water surface. They are greenish in colour thus able to carry out photosynthesis. These white water lilies produce bulbs that develop from the matured flowers. The bulbs look like the onion bulbs though greenish in colour and contain numerous little seeds inside them unlike the onion bulbs. People within Ibi area eat the bulb and describe it to be tasteless and slimy like okra. The bulbs easily deteriorate when harvested from its environment and exposed to different environmental condition without following necessary preservation precaution. This may be due to high

microbial activities occurring either on the bulb or inside the bulb from the aquatic environment. According to Obot and Ayeni [1], *Nymphaea lotus* is one of the foremost aquatic macrophytes that have been identified in Nigerian freshwater bodies. Despite this fact, Mohammed and Awodoyin [2] stated that there is dearth of information on water lily even though it is abundant in fresh water bodies in Nigeria. In advocating for utilization of *Nymphaea lotus* as supplement to fish/livestock feeds, Mohammed et al. [3] examined water lily from Tatabu flood plain of North Central Nigeria and discovered that they indeed posses some nutritional values. This current research specifically examined the nutritional, anti-nutritional and phytochemical contents in *Nymphaea lotus* bulbs from Mankhanwa River in Ibi Local Government Area of Taraba State.

## 2. Materials and Methods

#### 2.1. Sample Collection

Fresh bulbs of *Nymphaea lotus* were collected from Mankhanwa River in Ibi LGA., Taraba State, Nigeria on April 13<sup>th</sup>, 2016 at exactly 9:00 am and the second sample was also collected on 2<sup>nd</sup> May, 2016. The bulbs were carefully rinsed with clean water; air dried for two (2) weeks in the laboratory and the biochemical compositions determined using AOAC (2000) standard methodologies.

#### 2.2. Proximate Composition of the Sample

#### 2.2.1. Moisture Content

Moisture contents of the various oilseeds were determined using oven (GenlabMiNO/30 UK). Differences in weight are calculated as:

% Moisture =  $\frac{(Wt \text{ of } dish+Sample \text{ before } Drying) - (Wt \text{ of } Dish+Sample \text{ after } Drying) \times 100}{Wt \text{ of } Sample \text{ Taken}}$ 

#### 2.2.2. Protein Analysis

Protein levels in sample using the Kjeldahl Nitrogen determination of protein as reported by AOAC [4], with results adjusted through calculation as:

$$\% N = \frac{0.00014 \times Titre \ value \times 50 \times 100}{Wt.of \ sample \ taken}$$
(2)

% Protein = N 
$$\times$$
 6.25 (3)

#### 2.2.3. Amino Acid

Using the AOAC [4] methodology for Amino acid analysis, Buck Scientific (USA) (BLC10/11- model) High Performance Liquid Chromatography (HPLC) equipped with UV 338nm detector, column with C18, 2.5 x 200mm,  $5\mu$ m column and a mobile phase of 1:2:2 (100mM sodium sulphate, pH 7.2; acetonitrile; methanol (v/v/v) at a flow rate of 0.45 ml/minute and an operating temperature of 40°C. The various amino acids were determined.

### 2.2.4. Determination of Anti- nutrition and Phytochemicals

Antinutritional and Phytochemical contents in *Nymphaea lotus* bulb was carried out using Buck Scientific (USA) (BLC10/11 – model) High Performance Liquid Chromatography (HPLC) system fitted with a fluorescence detector (excitation at 295nm and emission at 325nm) and an analytical silica column (25cm x 4.6mm ID, stainless steel,  $5\mu$ m). The mobile phase used was hexane: tetrahydrofuran: iso-propanol (1000:60:4 v/v/v) at a flow rate of 1ml/min. The methodology is according to AOAC [4]. The concentration of the anti-nutrient was calculated using the following formula:

Where:

[Conc. Of anti-nutrient] = concentration of Anti-nutrient in ppm

(1)

[STD] = concentration of standard A sample = peak area of sample A STD = peak area of standard V HEX = Volume of hexane

Wt. Sample = weight of sample [4].

## 2.2.5. Ash Content

The dry ashing method of AOAC [4] was adapted. Calculation for ash content was as shown below:

% Ash = 
$$\frac{Wt.of Dish + Ash - Wt.of Dish x 100}{Wt.of Sample Used}$$

# 2.2.6. Determination of Fat in Nymphaea lotus Bulb

Determination of fat in *Nymphaea lotus* bulb was according to AOAC [4] methodologies for analysis of food stuff.

The percentage weight of fat contents was calculated viz:

% Fat Content = 
$$\frac{\text{Weight of Extract}}{\text{Weight of Sample}} X 100$$
 (5)

## 2.2.7. Determination of Fibre Nymphaea Lotus Bulb

Association of Official Analytical Chemists (AOAC) [4] standard methods for determination of food stuff was used to ascertain the fibre content in *Nymphaea lotus* bulb sample. The loss in weight on ignition was multiplied, by 100. The result gave the percentage of fibre in the sample.

### 2.2.8. Digestion Procedures for Mineral Analysis (Dry Ash Procedure)

Mineral analysis of sample (dry water lily bulb) was carried out using AAS with specific lamps (for all mineral elements and heavy metals) and flame photometer (for Na and K) using air acetylene flame integrated mode and quantity concentration of unknown from the calibration curve of standards [4].

## 2.2.9. Determination of Water and Fat Soluble VITAMINS by Isocratic HPLC

Buck Scientific (USA) (BLC10/11- model) HPLC equipped with UV 325nm and UV 254nm detectors was used for fat and water soluble vitamins determination of sample respectively. AC18,  $4.6 \times 150$ mm,  $5\mu$ m column and a mobile phase of 95.5 (methanol: water) was used at a flow rate of 1.00ml/minute and an ambient operating temperature.

### 2.2.10. Statistical Analysis

Statistical Analysis was carried out with the use of standard Students T-distribution test using Statistical Package for Social Sciences (SPSS) version 21 and values presented as mean  $\pm$  standard deviation.

## 3. Result

The results of the analysis involved proximate composition, mineral analysis, vitamin contents, antinutritional components as well as phytochemical composition of *Nymphaea lotus*. The results obtained are shown below:

Table 1. Proximate composition of water lily (Nymphaea lotus), bulb.

PARAMETERS	0/0
Crude Protein	$21.66 \pm 0.014$
Crude Fat	$5.07 \pm 0.014$
Crude Fiber	$13.30 \pm 0.021$
Ash	$8.34 \pm 0.007$
Moisture	$9.72 \pm 0.000$
Carbohydrate	$41.92 \pm 0.056$

All values are expressed as mean  $\pm$  standard deviation of duplicate results.

Table 2. Vitamins composition of water lily bulb.

PARAMETER S	mg/100g	
Vitamin A	$51.360 \pm 0.020$	
Vitamin B <sub>1</sub>	$0.085 \pm 0.001$	
Vitamin B <sub>2</sub>	$0.065 \pm 0.007$	
Vitamin C	$24.650 \pm 0.021$	
Vitamin E	$1.340 \pm 0.014$	

All values are expressed as Mean  $\pm$  Standard deviation of duplicate results.

Table 3. Mineral content of the water lily bulb.

PARAMETERS	mg/100g
Potassium	$742.89 \pm 0.040$
Phosphorus	$635.39 \pm 0.020$
Sodium	$431.53 \pm 0.010$
Zinc	$8.16 \pm 0.014$
Cadmium	$1.37 \pm 0.021$
Lead	$0.012 \pm 0.000$
Iron	$41.38 \pm 0.021$
Copper	$12.36 \pm 0.022$
Magnesium	$87.46 \pm 0.007$

All values are expressed as Mean  $\pm$  Standard deviation of duplicate results.

Table 4. Anti-Nutrients content in water lily bulb.

PARAMETERS	mg/100g
Phytates	$3.68 \pm 0.014$
Nitrates	$0.47 \pm 0.021$
Oxalate	$8.76 \pm 0.014$
Tannins	$13.63 \pm 0.010$
Saponins	$4.89\pm0.014$

All values are expressed as mean  $\pm$  standard deviation of duplicate results.

Table 5. Phytochemical content in water lily bulb.

PARAMETERS	mg/100g
Cyanides	$0.84 \pm 0.01$
Caffeine	$4.68 \pm 0.01$
β-Carotene	$5.62 \pm 0.01$
Lycopene	$1.96 \pm 0.01$
Hesperidins	$6.56 \pm 0.01$
Rutin	$0.87 \pm 0.02$
Diadzein	$3.62 \pm 0.01$
Genistein	$7.54 \pm 0.02$

All values are expressed as mean  $\pm$  standard deviation of duplicate results.

Table 6. Table of Nitrogen Free Extract and Gross Energy.

% NFE	Energy Kcalorie/100g
$55.22 \pm 0.035$	353.13 ± 0.042

## 4. Discussion

#### 4.1. Proximate Composition

The proximate composition of the water lily bulb is presented in table 1. The crude protein value (21.66 ± 0.014%) in Nymphaea lotus bulb obtained in this research was higher in comparison to the one gotten from the root sample  $(1.02 \pm 0.190\%)$ , leave sample  $(5.82 \pm 0.220\%)$ , and seed sample  $(1.04 \pm 0.170\%)$  [5]. This may be due to microhabitat variation in the study area or due to differences in the part of the plant used which showed that the bulb contain much of the crude protein than the other parts of the plant. Crude protein is an important feed ingredient normally used as a major ingredient in fish feed formulation [3], especially those obtained from Nymphaea lotus bulb. This shows that the bulb can be used in animal/fish feed formulation, infant and adult nutrient additive. The crude fat obtained from the bulb  $(5.07 \pm 0.014 \%)$  in this research was not in the same range with those obtained in the leave  $(2.18 \pm 0.290\%)$  and root sample  $(2.00 \pm 0.500\%)$  [5]. But the value in the leaves  $(4.83 \pm 0.210\%)$  obtained by Muhammed *et al.* [3], agree with the findings from this research which is important in correlating to vitamin absorption. The carbohydrate content in the bulb of Nymphaea lotus (41.92  $\pm$  0.056%) examined here was higher among the rest of the proximate components

quantified. Considerably, high amount of carbohydrate in the bulb indicate that it can be used as a good source of energy as shown in table 6 where the energy level obtained from the plant bulb is  $353.13 \pm 0.042$  KCalorie/100g. Also the crude fiber value (13.30  $\pm$  0.021%) obtained in Nymphaea lotus bulb examined herein is quite appreciable and similar to that gotten by Muhammed et al. [3] in the rhizome  $(13.24 \pm$ 0.350%). Past studies have linked low fiber content in diets with health problems like heart disorder, bowel cancer and appendicitis [6]. Therefore, Nymphaea lotus bulb when consumed will prevent health problems linked with bowel cancer, heart disorder and appendicitis. The moisture content  $(9.72 \pm 0.000\%)$  in dry sample quantified was low which shows that the bulb can be processed and saved for animal farm use and human diet. Some level of moisture as obtained in this research is also adequate in order to retain the nutritional contents in the water lily bulb.

#### 4.2. Vitamin Composition

The vitamin content of the water lily bulb is shown in table 2. It is shown that the bulb contains a very good amount of Vitamin A  $(51.3 \pm 0.020 \text{mg}/100 \text{g})$ . Vitamin A protects the lining of respiratory digestion urinary tracts against infection and serves as a visual pigment of the vertebrate eye [7-8]. The bulb also contains a high content of vitamin C (24.65  $\pm$ 0.021 mg/100g). Ascorbic acid was earlier reported to enhance iron absorption [8], it also prevent debilitating diseases [9] as well as enhance quick wound healing and general wellbeing of humans. Vitamin C increases the health and resilience of tendons, ligaments and collagen. The water lily (Nymphaea lotus) bulb understudied also contains vitamin E but the value was not much,  $(1.34 \pm 0.014)$ mg/100g). Vitamin E intake was linked to a decreased incidence of prostate and breast cancer [10]. Vitamin  $B_1$  $(0.085 \pm 0.001 \text{ mg}/100 \text{ g})$  and Vitamin B<sub>2</sub>  $(0.065 \pm 0.007 \text{ g})$ mg/100g), were found to be low in the bulb. The composition of those vitamins present in the bulb like the vitamin C can be used as supplements in food product to increase their availability in the system.

#### 4.3. Mineral Composition

The mineral compositions of *Nymphaea lotus* bulb content is presented in table 3. Generally, the bulb of water lily contains important valuable mineral elements. The potassium content of the bulb (742.89  $\pm$  0.040mg/100g), was higher than all other mineral found within the bulb. Nervous system and muscles activities are attributable to the presence of potassium. Potassium helps to maintain the correct water balance in the cells of the nerves and muscles [11]. The water lily investigated also had appreciable high content of phosphorus (635.39  $\pm$  0.020mg/100g) which is considerably higher than the one obtained in the leave sample (0.31  $\pm$ 0.001ppm) and root sample (0.19  $\pm$  0.002ppm) [8]. This may be due to ecological variation or the part of plant being used as the bulb contains more of this mineral element. This mineral is integral in energy storage and helps maintain and repair cells and tissues [12]. Phosphorus is important for healthy bones and teeth; found in every cell; part of the system that maintains acid-base balance [13]. Therefore, phosphorus from water lily may be extracted and used for solving problems associated with bone defect, cell and tissue maintenance. The bulb also contains a high amount of sodium (431.53  $\pm$  0.010mg/100g) and magnesium (87.46  $\pm$ 0.010), but moderately rich in iron (41.38  $\pm$  0.020) and copper (12.36  $\pm$  0.020). Iron is an essential part of many enzymes and proteins. It helps red blood cells transport oxygen to all the parts of the human body. Iron also helps regulate cell growth and cell differentiation [12]. It helps keep muscles and nerves functioning normally and also helps to regulate heartbeat, supports the immune system and keeps the bones strong [12]. Iron is also part of a molecule (haemoglobin) found in red blood cell that carries oxygen in the body; needed for energy metabolism [13]. Copper is needed as a part of many enzymes; needed for iron metabolism [13].

#### 4.4. Anti-nutritional and Phytochemical Contents

Tables 4 and 5 represent selected anti-nutritional and phytochemical content of the water lily bulb. The antinutrients (Tannins = $13.63 \pm 0.010$ mg/100g; saponins = $4.89 \pm$ 0.014 mg/100g; oxalates =  $8.76 \pm 0.014 \text{mg}/100\text{g}$  and phytates=  $3.68 \pm 0.014$  mg/100g) obtained in the bulb were moderately low. Tannins had the highest value and foods rich in tannins are considered to be of low nutritional value [14]. Tannins are used chiefly in tanning leather, dyeing fabric, making ink, and in various medical applications [15]. Therefore the tannin content in the bulb can be extracted and used for making ink and for dyeing fabrics. The detergent properties of saponin have led to their use in shampoos, facial cleansers and cosmetic creams [16]. Therefore, saponins in the bulb can also be extracted by industries for producing shampoos, facial cleansers and cosmetic creams. The bulb also contains a significant amount of oxalate. One of the main applications of oxalic acid is in rust-removal, which arises because oxalate forms, water-soluble derivatives with the ferric ion [17]. Powdered oxalate is used as pesticide in bee keeping combating the bee mite. The anti-nutritional components which are obtained in low quantities in water lily bulbs in this research makes the bulb nutritional good, however, since the water lily bulbs are readily available, these anti-nutritional components may be exploited for their industrial usage as illustrated above.

There are also appreciable quantities of phytochemicals present in the water lily investigated. They contain good amount of beta-carotene  $(5.62 \pm 0.010 \text{ mg/100g})$ . Beta-carotene is an antioxidant; it protects the body from damaging molecules called free radicals. Free radicals damage cells through a process known as oxidation [18]. The beta-carotene component can be extracted and used to protect the body against free radical that may be threat to the cell. Phytic acid plays a role in pancreatic function and insulin secretion. And it may reduce the glycemic response from

meals, meaning one feel full for longer [19]. The value of phytate, saponins, oxalate, and tannin in the bulb was shown (table 5) to be higher than that obtained from the leaves [5]. This may be due to the part of the sample used or due to micro-habitat variation in the area. Genistein was also present in water lily bulb (7.54  $\pm$  0.020mg/100g). Genistein are known to suppress malignant cell migration, invasion metastasis in vitro and in vivo [20]. Therefore, it can be extracted and use for this purpose. This study also shows that there is low presence of lycopene  $(1.96 \pm 0.010 \text{ mg}/100 \text{ g})$ . Lycopene is also used for treating human papilloma virus (HPV) infection, which is a cause of uterine cancer. Some people also use lycopene for cataracts and asthma) prevention [21]. The bulbs also contain a moderate value of hesperidin (6.56  $\pm$  0.010mg/100g) which have been said to have growth inhibitory effect [20].

# 5. Conclusion

This research showed quantification of water lily bulbs with different biochemical composition such as carbohydrate, protein, lipid and fiber and also some essential element such as sodium, potassium, magnesium, phosphorus, and zinc in addition to anti-oxidant Vitamins (A, C and E). It also shows that there are anti-nutritional factors in the bulb which are low and may be applied to other uses in chemical and pharmaceutical industries. From the result it is observed that the bulb is a good source of nutrient for consumption, but due to the effect of some anti-nutritional composition it is best advisable that the bulb should be processed first by either fermentation, or any other method to reduce the anti-nutritional content like phytates before consumption. Therefore, people living around this area should embark on a method of processing the bulb both as food and other economic uses.

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