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Nutritive Value and Anti-Nutritional Factors in *Hibiscus sabdariffa*

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Abstract: The seeds of *Hibiscus sabdariffa* were analysed for nutritional and anti-nutritional factors. The concentration (mg g^{-1}) of minerals indicates potassium to be the highest 1505.0. Magnesium and calcium were 322.2 and 218.2 (mg g^{-1}) respectively. The concentration (mg g^{-1}) of sodium, aluminium, manganese and chlorine were 12.5, 46.6, 7.6 and 24.5 respectively. Protein, lipid, soluble carbohydrate and fibre contents were 19.84, 28.10 33.0 and 6.33% respectively. The result of anti-nutritional factors indicates that the concentration (mg 100 g^{-1}) of phytate, hydrogen cyanide and trypsin inhibitory activity were 5.90, 0.29 and 9.23, respectively. Tannins and saponin contents were 0.16 and 2.20% respectively. The result of the analysis showed that the seeds of *Hibiscus sabdariffa* could serve as a good food supplement for man and livestock if further processing methods are employ to eliminate the little toxicant inherent in the seeds.

Key words: Nutrition, anti-nutrition, seeds *Hibiscus sabdariffa*

INTRODUCTION

Hibiscus sabdariffa L. is a short-day annual plant that belongs to the family *Malvacease*. It is believed to originate from India, now cultivated throughout the tropics[1]. It grows to a height of about 3.5 m tall and variously coloured dark green to red leaves alternate, glabrous, long-petiolate, palmately divided into 3-7 lobes, with serrate margins. The flowers are large, short peduncled, red to yellow with dark centre. Caspules are 5 cm long, 5.3 cm wide; it has a deep penetrating taproot[2].

The leaves and calyx are used as vegetable in many tropical countries. In the west indices and elsewhere in the tropics, the fleshly calyces are used for making Roselle wine, jelly, syrup, gelatin, refreshing beverages, pudding and cakes; while the dried Roselle are used for tea, jelly, marmalade, ices, ice-cream, sherbets, butter, pies, snacks, tarts and other desserts[3]. The oil is edible and is used as a substitute for the crude castor oil. The residual meal is used for food, either in soup or made up with beans and other articles into cakes[4]. In Nigeria, districts where the locust bean is scarce, the seeds of *H. sabdariffa* were used as a substitute[5].

Medicinally, the leaves of *H. sabdariffa* are emollient and are much used in Guinea as a diuretic, refrigerant and sedative. The fruits are anti-scorbutic; the succulent calyx, boiled in water is used as a drink in bilious attacks[6]. It is on record that the Philippines used the bitter root as an apentive and cough remedy[7]. Reported that simulated ingestion of the plant extract decrease the rate of absorption of alcohol, lessening the intensity of alcohol effects in chickens.

Despite the beneficial uses of this crop and its cosmopolitan nature, there is dearth of information about the crop. The present study was conducted to generate information about the chemical composition and the nutritional and anti-nutritional qualities of the seeds of these plants obtained in Gwer local government area of Benue State, Nigeria.

MATERIALS AND METHODS

Hibiscus sabdariffa seeds were collected from Mbalom, Yonov district of Gwer LGA of Bneue State in January, 2005. The seeds and leaves were identified at the Herbarium of the Department of Biological Sciences, Ahmadu Bello University Zaria. The seeds were dried at 45°C in an-air circulated oven for 72 h, ground with porcelain mortar and pestle to fine particles and stored in screw-capped plastic containers.

Determination of minerals content: The ground sample was pulverized and approximately 150 mg sub-sample was weighed and wrapped in polyethylene films. For the elements leading to short-lived activation products, the sample was packed and sealed in 7 cm^3 rabbit capsules. Irradiation with thermal neutrons was carried out at an outer irradiator channels (i.e., B_4) of the Nigeria Research Reactor-1 (NIRR-1) operating at a thermal neutron flux setting of $2x10^{//}$ n/cm^2s, which corresponds to a neutron flux of $1x10^{//}$ n/cm^2s in the outer channels. After the irradiation, measurement of induced radionuclide was performed by a PC-based gamma ray spectrometry set-up. This consists of a HPGC detector coupled to a computer

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based Multi-Channel Analyzer (MCA) via electronic modules. The relative efficiency of detector is 10% and an energy resolution of 1.95KeV at gamma-ray energy of 1332 KeV belonging to ^{60}Co. Through appropriate choice of cooling time, detector's dead time was controlled to be less than 10%. Identification of gamma-ray of product radionulides through their energies and quantitative analysis of their concentrations was achieved using the gamma-ray spectrum analysis software, WINSPAN- 2004. The certified reference material IAEA- soil-7 was used as the standard, while two certified reference materials, GSD-II and GSR-5 were used as analytical quality control materials to validate the procedure for all the elements.

Determination of nutritional content: The samples were analysed for proximate composition (moisture, ash, crude protein, lipids, soluble carbohydrate and crude fibre). The moisture content of the seeds was determined by oven drying to a constant mass at 105°C. The ash content was determined by heating the sample in a muffle furnace at 550°C. The lipid was extracted with petroleum ether 40-60°C using a soxhlet for four h. The micro-Kjedahl procedure was adopted for the determination of protein, while the Anthrone procedure was used for the determination of soluble carbohydrate[8,9].

Determination of hydrogen cyanide: The alkaline titration procedure was adopted[9]. 10 g of ground sample was soaked in a mixture of 200 cm^3 of distilled water and 10 cm^3 of orthophosphoric acid. The mixture was left overnight to release all bounded hydrocyanic acid. The mixture was distilled until 150 cm^3 of the distillate was collected. 20 cm^3 of distillate was taken into a conical flask containing 40 cm^3 of distilled water. 8 cm^3 of 6 $moldm^{-3}$ aqueous ammonia and 2 cm^3 of 5% potassium iodide solution were added. The mixture was titrated with 0.02 $moldm^{-3}$ silver nitrate to faint but permanent turbidity.

Determination of phytate: Ground sample (4 g) was soaked into 100 cm^3 of 20% hydrochloric acid for five h and filtered. 25 cm^3 of the filtrate was placed in a conical flask and 5 cm^3 of 0.3% Ammonium thiocyanate solution was added. The mixture was titrate with standard Iron (III) chloride solution until a brownish-yellow colour persisted for 5 min[10].

Determination of tannins: Tannins were determined by the vanillin-HCl procedure, which was based on an acid-catalysed addition of vanillin to flavonols. These reactions are determined colorimetrically at 500 nm[11].

Determination of saponins: The ground sample (10 g) was mixed with 100 cm^3 of 20% aqueous ethanol in a beaker and agitated with a magnetic stirrer for 12 h at 55°C. The solution was filtered using whatman No.1 filter paper, the residue was re-extracted with 200 cm^3 of 20% aqueous ethanol. The extracts were mixed and reduced to about 40 cm^3 under vacuum. The extract and 20 cm^3 diethyl ether were poured into a 250cm^3 separating funnel and shaken vigorously. The aqueous layer was discarded. The process of purification continued until a colourless aqueous extract was obtained. The pH of the remaining aqueous solution was adjusted to 4.5 by adding 4 g of NaCl and the solution then shaken successively with 60 and 30 cm^3 portions of n-butamol. The butamol extract was washed twice with 10cm^3 of 50% aqueous sodium chloride. Then evaporated to dryness in a fume cupboard, to give the saponin, which was weighed and expressed as percentage[12].

Determination of trypsin inhibitor activity: Ground samples (0.25 each) were extracted in 12.5 cm^3 of 0.01M NaOH at pH 9.4 to 9.6 using an ultra-Turrax at 20,000 rpm for five minutes (2×2.5 min) with intermittent cooling by keeping the tubes containing the samples in an ice bath. The contents were centrifuged at 3500 g for 15 min and the supernatants were collected; this supernatant was centrifuged a second time at 9500 g, following which the supernatants were collected by slowly pipetting between the residue at the bottom and the fatty layer on top. These solutions were used for the assay after appropriate dilution (with distilled water), based on pre-assay trial result[13].

RESULTS AND DISCUSSION

From Table 1, the mineral content of *H. sabdariffa* indicates the concentration of potassium to be highest 1505 mg g^{-1}. Magnesium contents was 322.2 mg g^{-1}. The concentration of calcium was 218.2 mg g^{-1}, sodium 12.5 mg g^{-1}, while the concentration of manganese, Aluminium and chlorine were 7.6 mg g^{-1}, 46.0 mg g^{-1} and 24.5 mg g^{-1}, respectively. The values indicate that *H. sabdariffa* seeds are good sources of potassium, calcium and magnesium. Potassium, plays a critical role in the transmission of nerve impulses, muscle contraction and maintenance of normal blood pressure, calcium helps in bones repairing. Lack of calcium in the body results in the breakdown of bones. Manganese is necessary for normal body metabolism and important enzyme reactions; maintenance of normal nerve, brain and thyroid functions. Its deficiency is uncommon but can affect brain, glucose tolerance, normal reproduction, skeletal and cartilages formation.

From Table 2, the observed moisture content of *H. sabdariffa* was 5.98%. Moisture is the water content of

Table 1: Mineral composition of *Hibiscus sabdariffa* seeds

Elements	Concentration mg g^{-1}
Ca	218.2±3.5
Mg	322.2±2.4
K	1505.0±7.2
Na	12.5±1.3
Al	46.0±1.0
Mn	7.6±10
Cl	24.5±30

Table 2: Proximate composition of *Hibiscuss sabdariffa* seeds

Elements	Concentration mg g^{-1}
Ash (%)	5.55±0.11
Moisture (%)	5.98±0.11
Protein (%)	19.84±0.21
Lipids (%)	28.10±0.44
Soluble Carbohydrate (%)	33.0±0.14
Fibre (%)	6.33±0.07

(Values are mean±SE)

Table 3: Anti-nutritional content of *Hibiscus sabdariffa* seeds

Elements	Concentration mg g^{-1}
Phytate mg 100 g^{-1}	5.90±0.23
HCN mg 100 g^{-1}	0.29±020
Tannins (%)	0.16±0.6
Saponin (%)	2.2±0.3
Trypsin inhibitory mg 100 g^{-1}	9.23±0.4

(Values are mean±SE)

feed. A knowledge of the moisture content is needed for the optimum processing of food, for example milling of cereals, mixing of dough to optimum consistency and for producing bread with the best grain, texture and freshness retention[14] . The value observed in this plant agrees with those of *Hibiscus esculentus, Milletha thonningii, Albiza zygia, Afzellia bella* which were given as 5, 4.71 7.80 and 6.81%, respectively[15].

The ash content of *H. sabdariffa* was 5.55%, the ash content of plant material is a measure of its inorganic content. The value compared favorable with other legumes that serve as good source of certain minerals. The protein content of *H. sabdariffa* was 19.8%. The value falls within protein representation in most legumes, which is about 17-30%[16]. The value also indicates that the seeds of this plant are good source of protein. The crude lipid content of the seeds was observed to be 28.10%. This value is higher compared to those reported by Duke[3] for the same plant. The difference could be due to environmental factors and type of solvent employed for the extraction. Soluble carbohydrates content was 33,0% while fibre content was 6.33%. Nour[17] observed that apart from their nutritional and metabolic functions, carbohydrates are important natural sweeteners and raw materials for various fermentation products. The value of carbohydrate observe is relatively lower compare to those of *Diospyrous mespiliformis, Daneia ogea* and *A. bella* which were given as 77, 74 and 54%, respectively[18]. The crude fibre content of the seeds of this plant falls within the values reported for most legumes[19]. The presence of high crude fibre in food material decreases dry matter digestibility in animals. The high crude fibre content therefore shows good nutritive value of a feed material[13].

The phytate content of *H. sabdariffa* seed Table 3 was observed to be 5.9 mg 100 g^{-1}. Miller[3] reported that phytate content of legumes in many cases vary depending upon the variety and/or cultivar, climate conditions, location, type of soil and year during which they are grown. High level of phytic acid are of nutritional significance as phytic acid might decrease biovailability of minerals. The insoluble complexes, so-formed, resist breakdown in the digestive tract, resulting in a reduced availability of these minerals for the non-ruminants. The phytic acid could, however be substantially eliminated by processing methods such as soaking and cooking[3]. The Hydrogen cyanide content was 0.29 mg 100 g^{-1}. Anhwange[20] reported that high hydrogen cyanide content in diet causes neurological respiratory, carbiovascular and thyroid debilities. The low hydrogen cyanide content of the seeds is an indication that it is a good source of diet.

Tannin contents of *H. Sabdariffa* seed was 0.16%. Tannins in legumes are mostly found in the seed coat thus its removal may be expected to reduce the content. The nutritional effect of tannins is mainly related to their interaction with protein[13,21]. Tannin content of the seeds was found to be in trace amount. However, soaking and cooking processes may eliminate the tannins content significantly. Saponin content of *H. sabdariffa* was low, 2.20%. saponins are glycoside containing a polycyclic aglycone moiety of either C_{27} steroid or C_{30} triterpenoid attached to a carbohydrate. They are found naturally in plants especially desert plants[22]. They have a characteristic bitter taste; foaming properties and can cause injuries to the digestive mucosa and haemocytic changes in blood[21].The trypsin content of *H. sabdariffa* was 9.23 mg 100 g^{-1}. This value is relatively lower than 16.6 mg reported for raw soyabean meal[13].

Acute shortage of conventional foodstuffs for feeding of livestock in developing countries has forced planners and nutritionalist to look for unconventional food resources wherein there is no competition with humans. These unconventional food resources are rich in anti-nutritional factors and sometimes have low biological value. *Hibiscus sabdariffa* seeds were found to be very good sources of protein, fats and carbohydrate. The low values of anti-nutritional factors also make the seeds a good source of nutrients. Therefore efforts should be make to use the seeds of *H. sabdariffa* as foodstuff efficiently by removing the anti-nutritional factors and increasing their biological values by chemical or biotechnological means.

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