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PHYTOCHEMICAL, PROXIMATE, AND METAL CONTENT ANALYSIS OF *CITRULLUS LANATUS* (WATERMELON) SEED.

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ABSTRACT
The present study examined the phytochemical, proximate and metal content of *Citrullus lanatus* (watermelon) seed. The dried seeds were pulverized into fine powder and a portion of it was extracted with methanol, and distilled water, and freeze-dried using a lyophilizer. The results of phytochemical screening showed that the seeds contained terpenoids, glycosides, steroids, alkaloids, flavonoids, coumarins and quinones in high amount, while phlobatannins and anthraquinones were not detected. Proximate analysis indicated low moisture content (10.40 ± 0.10 %), ash (6.60 ± 0.20 %), crude fat (14.60 ± 0.25 %), fibre (42.80 ± 1.80), high protein (7.70 ± 0.16%), and nitrogen-free extracts (NFE) (18.30 ± 1.35%), while metal analysis showed the presence of sodium (30.60 ± 0.25 mg/kg), calcium (0.97 ± 0.04 mg/kg), zinc (0.25 ± 0.00 mg/kg), magnesium (5.98 ± 0.05 mg/kg), and potassium (20.12 ± 0.07 mg/kg). Potassium and sodium were present in the highest quantity, while zinc occurred in the least amount. Lead and cadmium were not detected. The seeds of *C. lanatus* is a rich source (reservoir) of phytochemicals, macro- and micro nutrients.

Key words: Phytochemicals, metals, carbohydrates, protein, fibre

INTRODUCTION

Diets rich in phytochemicals such as carotenoids and phenolic compounds have been associated with a reduced risk of certain types of cancer, cataracts, inflammation, cardiovascular, muscular degeneration and neurodegenerative diseases (Bueno, *et al.*, 2012; Sergent, *et al*., 2010; Snyder, *et al*., 2011; Tanaka *et al.*, 2012). Tropical fruits consumption is increasing on domestic and international scale due to growing recognition of their nutritional and therapeutic values (Alves *et al*., 2008). Phytochemicals, the biologically active and naturally-occurring compounds in plants provide health benefits for humans. They contribute to plants colour and flavor, and are found in different parts of the plants such as root, stem, leaves, fruits and seeds. They have biological properties such as antioxidant activity, antimicrobial effect, modulation of detoxifying enzymes, stimulation of the immune system, and general modulation of hormonal activity. As a result of these properties, researchers are interested in their health benefits. The identification and quantification of phytochemicals in pulps and by-products of tropical fruits are of utmost importance in order to substantiate their potential health benefits in human nutrition.

Fruits occupy a part of daily diet of the rich and rarely the poor and there are many parts of a fruit that are not considered edible, and so are usually thrown away. One such fruit is *Citrullus lanatus*, which is taken by all but the seeds are thrown away and generally not included in regular diet. The juice or pulp of *Citrullus lanatus* is considered as the edible portion but rind and seeds are discarded as major solid wastes (Bawa and Bains, 1977). The fruit has numerous small black seeds embedded in the middle of the flesh, while the embryo completely fills the seed. The seeds have sweet and nutritious kernels. *Citrullus lanatus* is of the *cucurbitaceae* family and several studies have shown that seeds of the *cucurbitaceae* are potential sources of nutrients such as protein, minerals and lipids as well as ingredients for native medicine. The plant is related to the cantaloupe, squash and pumpkin and other plants that grow on vines (on the ground). It is high in fibre, citrulline and arginine (Collins *et al*., 2007; Oyeleke *et al*., 2012). *Citrullus lanatus* seeds are highly nutritional; they are rich sources of protein, B-group of vitamins, minerals (such as magnesium, potassium, phosphorous, sodium, iron, zinc, manganese and copper), fat, as well as phytochemicals (Braide, *et al.*, 2012). The seeds are known to have economic benefits especially in countries where cultivation is on the increase. They are used to prepare snacks, milled into flour and used for sauces. Oil from the seeds are used in cooking and incorporated into the production of cosmetics (Jensen *et al*., 2011). The aim of this study was to determine the phytochemicals and minerals present in the seed of *Citrullus lanatus*.

MATERIALS AND METHODS

Sample collection and preparation

Citrullus lanatus fruits were purchased from a major market in Benin City, Edo State, Nigeria. Only healthy looking fruits were collected. The seeds obtained from their pods were shade dried, until a constant weight was obtained. The dried seeds were pulverized into

fine powder using a mechanical blender. The powder was weighed and kept away from light before extraction.

Preparation of extract

Exactly, 2 kg of the powdered seeds was extracted with 5 litres of methanol (95 %) and distilled water for 96 h with constant stirring at intervals. The extracts (aqueous and methanol) were filtered using muslin cloth and concentrated using a rotary evaporator, freeze-dried and kept refrigerated.

Proximate composition

The powdered seeds were used for the determination of the moisture, crude protein, crude fat, crude fibre, carbohydrate, ash and mineral contents using standard methods (A.O.A.C, 2005).

Elemental analysis

This was carried out on the pulverized seeds to determine the iron, cadmium, lead, and zinc contents using atomic absorption spectrometer (AAS) and the contents of magnesium, calcium, sodium, and potassium using flame photometer based on the method of A.O.A.C, (1984).

Phytochemical analysis

The methanol and aqueous extracts were screened for their phytochemical contents according to standard methods (Trease and Evans, 2002; Tiwari *et al*., 2011; Boakye *et al*., 2015).

STATISTICAL ANALYSIS

One-way analysis of variance (ANOVA) was used to analyze the results using Graph Pad Prism Demo (6.07). Data are presented as mean ± SEM.

RESULTS

Phytochemical screening of the methanol and aqueous extracts

Terpenoids, glycosides, steroids, alkaloids, flavonoids, coumarins and quinones were detected in high amount, while phlobatannins and anthraquinones were not detected.

Table 1: Qualitative phytochemical analysis

Parameters	**Extracts**	
	Methanol	**Aqueous**
Steroids	+ +	+++
Terpenoids	+	+++
Cardiac glycosides	+ +	+++
Saponins	ND	++
Alkaloids	+	+
Flavonoids	+	+
Tannins	+	ND
Phlobatannins	ND	ND
Coumarins	+ +	++
Quinone	+	++
Anthraquinones	ND	ND
Fixed oils	++	ND
Carbohydrates	+	++

+ = present, + + = relatively abundant, +++ = relatively more abundant, ND = not detected

Proximate analysis

The percentage compositions of moisture, ash and crude protein were significantly lower ($p < 0.05$) than those of fat, crude fibre and NFE.

Table 2: Proximate analysis of the pulverized seeds of *Citrullus lanatus*

Parameter	%
Moisture content (Dried sample)	10.40 ± 0.00
Ash content	6.60 ± 0.20
Fat content	14.40 ± 0.25
Crude fibre	42.80 ± 1.80
Crude protein	7.70 ± 0.16
Nitrogen free extract	18.30 ± 1.39

Data are presented as mean ± SEM (n = 3)

Elemental analysis

The concentrations of potassium and sodium were significantly higher ($p < 0.05$) than those of magnesium, zinc and calcium, while lead and cadmium were not detected.

Table.3: Elemental analysis of *Citrullus lanatus* seed

Metals	**Concentration (mg/kg)**
Zinc (Zn)	0.25 ± 0.00
Lead (Pb)	0.00 ± 0.00
Magnesium (Mg)	5.98 ± 0.05
Potassium (K)	20.12 ± 0.07
Sodium (Na)	30.60 ± 0.25
Calcium (Ca)	0.97 ± 0.04
Cadmium (Cd)	0.00 ± 0.00

Data are presented as mean ± SEM (n = 3)

DISCUSSION

The present study examined the phytochemical, proximate and metal content of *Citrullus lanatus* (watermelon) seed. In the present study, terpenoids, glycosides, steroids, alkaloids, flavonoids, coumarins and quinones were detected in high amount, while phlobatannins and anthraquinones were not detected. This is in agreement with the report of Ali *et al*., (2012), who showed that alkaloids and terpenes are widely distributed in the genus "*citrullus"*. Phytochemicals have biological properties such as antioxidant activity, antimicrobial effect, modulation of detoxifying enzymes, stimulation of the immune system and general modulation of hormonal activities (Narasinga, 2003). The low moisture and ash contents showed that the powdered sample may contained more of the organic components and so less liable to spoilage by microbial contamination if properly stored (African Pharmacopoeia, 1986). Moisture content is a major quality factor in the preservation of some food products and it affects food stability (Nielsen, 2010).The relatively low moisture content is an indication that flours made from these seeds may have high shelf-life especially when properly packaged. Proteins are essential component of the diet needed for survival of animals and humans, which function basically in nutrition by supplying adequate amounts of required amino acids (Pugalenthi *et al.*, 2004). The high level of crude fibre in the seeds is probably the reason why many individuals tend to spit them out due to the discomfort of chewing. However, this component provides the bulk necessary for the proper peristaltic action in the intestinal tract thereby aiding digestion, contrary to public opinion of the seeds being a source of constipation and indigestion. The Recommended Daily Allowance (RDA) of protein, fibre, fat and carbohydrate for men are 56 g/day, 38 g/day, 20 - 35 % calories, and 130 g/day, respectively. The present study showed the presence of a number of metals in *citrullus lanatus* seeds. The chief metal components were magnesium, potassium, sodium, calcium and zinc, as previously reported by Stefania *et al.*, (2009). Nutritionists have considered the role of metals such as zinc, copper and iron in human fitness (Udayakumar and Begum, 2004), and recommended that they should be considered for preparation of herbal drugs (Sadia *et al*., 2011).

CONCLUSION

The seeds of *C. lanatus* is a rich source (reservoir) of phytochemicals, macro- and micro nutrients.

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