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Evaluation of the nutritional potentials of *Lageneria breviflora* seeds

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Abstract

The proximate, phytochemicals, volatile organic compounds and mineral compositions of *Lageneria breviflora* seeds were evaluated in this study using standard methods. The results showed appreciable amount of nutrient. Proximate composition showed that the seed contains 54.87% carbohydrate, 15.37% fat and 3.65% crude protein. Anthocyanin (77.55µg/g) had the highest concentration among the phytochemicals present. Majority of the phytochemicals that were predominantly present were of flavonoid class. Oxalate, tannin, and alkaloid were found in appreciable quantities. The following volatile organic compouds were detected acetone, acetic acid, butanal, formaldehyde, methane, benzene, xylene, toluene, and monoterpenes. The analysis of the mineral showed appreciable amount of minerals with magnesium having the highest amount(15.263ppm). *Lageneria breviflora* seed is a potential source of important nutrients and pharmacological compounds.

Keywords; *Lageneria breviflora* seed; Proximate composition; Phytochemicals; Volatile compounds

1. Introduction

Plant seeds are a good source of food for animals as well as humans, majority of food consumed by human beings are seeds. The drastic increase in the world's population in the last two centuries has driven up the need for food and increased food demand. Most third world countries depend for basic diet of carbohydrates, fats and proteins on a very limited number of crop species that are conventional (Mayes *et al.,* 2012; Ezim *et al*., 2020) while leaving others neglected and abandoned. Some of these neglected plant species (NPs) are generally rich in nutrients and health-promoting compounds with preventive effects against malnutrition and some chronic diseases, they have a huge potential in improving nutrition and health of indigenous communities (Ezim and Ezeanyika, 2021). Underutilized plants species are limited by loss of local knowledge, lack of research, lack of communication and limited market opportunities (Okigbo and Ugwu, 2021). Hence, creating awareness, market opportunities, research and new knowledge will enhance their utilization. Foods of animal origin are often unaffordable to low-income house-hold, various neglected crops can offer an alternative source of micronutrients, vitamins, as well as health-promoting secondary plant metabolites and improving agricultural diversity. Plants contain many bioactive compounds that produce definite physiological and biochemical actions in human body (Ujowundu *et al.*, 2010).

Lageneria breviflora is one of the neglected plant species, it belongs to the family cucurbitacae. It is commonly called wild colocynth in English language, while in Nigerian native Yoruba language, it is called 'tagiri', in Igbo, it is called 'anyummo'. It is a perennial tendril climber. The fruit of Lagenaria breviflora is widely used in folklore medicine in West Africa as a herbal remedy for the treatment of human measles, digestive disorders, and as wound antiseptics (e.g. umbilical incision wound), while livestock farmers use it for Newcastle disease and coccidiosis treatment in various animal species, especially poultry (Adedapo*et al.*, 2013). There is no record of the use of the seed in human nutrition, thus this work explored the nutritional potentials of this seeds.

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2. Material and methods

2.1. Sampe Preparation

Fresh *Lageneria breviflora* fruits were collected for a local farm at Igwuruta in Ikwerre Local Government Area, Rivers State, Nigeria. The fruits were carefully peeled off and the seeds were carefully removed. Drying of seeds (to a constant weight) was done in a laboratory oven at 50˚C. The dried seeds were ground into fine powder using a laboratory mill

2.2. Proximate Analysis

Moisture, lipid, ash and crude fibre contents were determined following standard methods of the Association of Official Analytical Chemists (AOAC, 1990) The organic nitrogen content was quantified using the micro Kjeldahl method, and an estimate of the crude protein content was done by multiplying the organic nitrogen content by a factor of 6.25 (Sosulski and Imafidon, 1990). Total carbohydrate content was calculated by difference.

2.3. Determination of Minerals

Calcium, potassium, manganese, selenium, copper, magnesium, iron, zinc and sodium were determined using FS 240 Varian atomic absorption spectrophotometry method while chloride and phosphorous were determined by colorimetric methods.

2.4. Determination of phytochemicals and volatile compounds

The phytochemicals and volatile compounds were determined using Gas Chromatography with Flame-Ionization Detection (GC-FID)

One gram of the sample was weighed and transferred in a test tube and 15ml ethanol and 10ml of 50% m/v potassium hydroxide was added. The test tube was allowed to react in a water bath at 60^{o}C for 60mins. After the reaction time, the reaction product contained in the test tube was transferred to a separating funnel. The tube was washed successfully with 20ml of ethanol, 10ml of cold water, 10ml of hot water and 3ml of hexane, which was all transferred to the funnel. These extracts were combined and washed three times with 10ml of 10%v/v ethanol aqueous solution. The solution as dried with anhydrous sodium sulfate and the solvent was evaporated. The sample was solubilized in 1000ul of pyridine of which 200ul was transferred to a vial for analysis.

The analysis of phytochemical was performed on a BUCK M910 Gas chromatography equipped with a flame ionization detector. A RESTEK 15-meter MXT-1 column (15m x 250um x 0.15um) was used. The injector temperature was 280^{0}C with split less injection of 2ul of sample and a linear velocity of 30cms^{-1}, Helium 5.0pa.s was the carrier gas with a flow rate of40 mlmin^{-1}. The oven operated initially at 200^{0}C, it was heated to 330^{0}C at a rate of 3^{0}C min^{-1} and was kept at this temperature for 5min. The detector operated at a temperature of 320^{0}C.

Phytochemicals were determined by the ratio between the area and mass of internal standard and the area of the identified phytochemicals. The concentration of the different phytochemicals is expressed in ug/g.

3. Results

Table 1 Proximate composition of *Lagenaria breviflora* seeds

Parameters	**(%)**
Carbohydrate	54.88 ± 0.06
Crude protein	13.65 ± 0.03
Fat	15.37 ± 0.03
Crude firbre	2.14 ± 0.04
Ash	5.26 ± 0.02
Moisture	8.70 ± 0.03

Table 2 Phytochemical composition of *Lagenaria breviflora* seeds

Parameters	**CLASS**	**(ug/g)**
Proanthocyanin	Flavonoid	52.0518
NarinSgin	Flavonoid	27.3762
Quinine	Alkaloid	16.9488
Flavan-3-ol	Flavonoid	29.3094
Anthocyanin	Flavonoid	77.5544
Sapogenin	Steroids or triterpens	38.9238
Spartein	Alkaloid	18.7590
Phenol	Flavonoid	38.5210
Flavonones	Flavonoid	29.8090
Steroids		21.0716
Epicatechin	Flavonoid	5.4888
Kaempferol ppm	Flavonoid	4.4411
Phytate		11.368
Flavones	Flavonoid	4.9866
Naringenin	Flavonoid	25.4369
Resveratol	Flavonoid	37.3469
Tannin		23.0274
Ribalinidine	Alkaloids	31.3808
Catechin	Flavonoid	52.1338
Oxalate		20.2039
Total		**566.1290**

Table 3 Volatile organic acids in *Lagenaria breviflora* seeds

Parameters	**(ug/l)**
Acetone	0.1123
Acetic acid	0.0096
Butanal	0.0238
Carbon disulphide	0.1607
Formaldehyde	0.0461
Benzoylecgonine	0.0264
Methane	0.0471
Benzene	0.0399
Monoterpenes	0.0110
Ethylene glycol	0.0675
Xylene	0.0234
Toluene	0.0006

Table 4 Minerals composition of *Lagenaria breviflora* seeds

Parameters	Ppm
Iron	0.276
Zinc	0.499
Magnesium	15.263
Potassium	8.893
Sodium	4.893
Calcium	8.343

Table 5 Minerals composition of *Lagenaria breviflora* seeds

Parameters	mg/l
Phosphorus	7.444 ± 0.03
Chloride	34.87 ± 0.02

4. Discussion

The proximate composition of *Lagenaria breviflora* seed revealed high carbohydrate (54.88%) content compared to some other conventional fruits in the Cucurbitaceae family. The value is higher than *Cucumis metuliferus* (Jelly melon) seeds which contains 50.24% carbohydrate (Achikanu*et al.,* 2020) and *Cucumis sativus* (Cucumber) seeds which contains 50.1% (Niyi*et al.,* 2019) thus wild colocynth seeds can be considered as a potential energy source. The seeds contains appreciable amount of protein (13.65%) thus can be vital in the repairing of worn out body tissues and also contribute significantly to growth (Murray *et al.,* 2009). The protein content of the seeds (13.65%) was found to be lower than those of *Amaranthus viridus (34%)* (Pandey*et al*, 2006), 31% for *Talinum triangulare* (Akindahunsi and Salawu, 2005). The proximate analysis of the *Lagenariabreviflora* seeds showed the high content of dietary fat (15.73%.). Dietary fats increases palatability by flavor absorption and retention although consumption of dietary fats in large quantity can lead to cardiovascular disorders (Anita *et al,* 2006). Ash is an index of mineral content of a material and the ash content of the *Lagenaria breviflora* seed was relatively high when compared to conventional seeds in the same family.

Minerals in adequate amount ensure the normal physiological functions and are also required for some enzymatic processes in the body (Chatterjea and Shinde, 2012). Potassium is very important in maintaining body fluid volume and osmotic equilibrium, regulation of muscles and nerve irritability, control of glucose absorption and enhancement of normal retention of protein during growth (NRC, 1989), the result showed high amount of potassium in the seeds (8.89ppm. Chloride values were high in the seeds (38.87 ml). Chlorides (chlorine compounds) play an essential role in the electrical neutrality and pressure of extracellular fluids and in the acid-base balance of the body (Rolfes*et al.,* 2012*)*.

The result of the phytochemicals of *Lagenaria breviflora* seeds revealed the presence of active phytochemical constituents like the tannin, flavonoids, alkaloids and steroids in the seeds with the flavonoids being the most abundant. Flavonoids are used to combat bacteria, viruses, and it equally exhibits anti- neoplastic effect (Ali, 2009). Some health challenges such as heart disease, cancer, diabetes can be addressed with the use of flavonoids (Stauth, 2007). Flavonoids acts as signaling molecules, detoxifying agents, phytoalexins and helps in the stimulation of seed germination, temperature acclimatization, and provide drought resistance (*Roy et al.,* 2022). Flavonoids also reduce reactive oxygen species in plant tissue, which are generally generated due to infection or UV irradiation. Another important role they play is in the fragrance, color, and taste of fruits, flowers, or seeds. This fragrance and color attract pollinators that help pollination and dispersal of seeds (Griesbach,2010; Roy *et al*., 2022). Flavonoids have the property of UV absorption, which is why they are considered to play a role in the protection of plants from UV radiations . They exhibit anti-oxidant actions by the suppression of ROS formation either by inhibition of enzymes or by chelating trace elements involved in free radical generation, scavenging reactive oxygen species and up-regulation or protection of antioxidant defenses (Ferreyra et al., 2012 ; Mishra *et al., 2013*;Roy et al., 2022). Flavonoids give protection to plants from pathogens and herbivores. Flavonoids give color and fragrance to flowers and fruits in different plant species and attract pollinators.

These pollinators help to remove and grow seeds and plants. Anthocyanins which were found to have the highest concentration when compared with other flavonoids present in *Lagenaria breviflora* seeds are of huge potential. They can be used as natural colorant and also have added valued properties (Bridle and Timberlake, 1997). These properties include potential antioxidant capabilities, used as nutraceuticals, also as antimicrobial effect and chronic disease prevention. It has been traditionally used as a phytopharmaceutical, appetite stimulant, choleretic agent, and for treatment of many other diseases. Epidemiological studies show the relationships between anthocyanin-rich foods and CVDs, as well as the relationship between total anthocyanin intake and risk of developing these cardiovascular-related diseases. Anthocyanins also demonstrate *in vitro* anti-thrombotic effect (Rechner and Kroner, 2005). *Lagenaria breviflora* seeds also contained steroid containing compounds which are important in pharmacology due to their relationship with sex hormones (Okwu, 2001). The result also shows the presence of alkaloids. Alkaloids are nitrogen-containing compounds that occur naturally and are reported to possess antimalarial, anticancer, antiasthma (Kittakoop*et al,* 2014), analgesic, hypoglycemic and also antibacterial activities (Qiu*et al,* 2014). Alkaloid is used in the management of diabetes, bacterial and fungal infection (Awoyinka*et al,* 2007) also it stimulates anaesthetic action (Edeoga and Enata, 2001). The result also revealed the presence of tannin. Tannins are non-toxic but can become toxic to filamentous fungi such as yeast, and bacteria and organisms that take them may experience some physiological responses (Scalbert, 1991). The availability of tannin shows that *Lagenaria breviflora* seed can function as anti-fungal, anti-diarrheal, anti-oxidant, anti-haemorrhagic and anti-hemorrhoidal agents (Asquit and Butter, 1986). Tannins have been implicated in the speeding up blood clotting processes, reduction of blood pressure, modulation of immune-response and in reduction of plasma lipid (Chung *et al,* 1998). These phytochemicals were all found in the seed in appreciable amounts.

The results of volatile compounds, revealed the presence of several volatile compounds including acetone, acetic acid and formaldehyde. Although of low concentration, these volatile compounds plays signaling role in and between plants, (Baldwin and Schultz, 1983; Rhoades, 1983), attraction of pollinators, either through triggering innate behavioural responses or by providing a stimulus that can be associated with the presence of rewards (Schiestl and Johnson, 2013; Haverkamp *et al*., 2016). Monoterpenes was also found to be present in *Lagenariabrevifloras*eeds. This compound is also present in some other plants of the Cucurbitaceae family (Toulassi*et al,* 2015).They are potent in the prevention of carcinogenesis at both the initiation and progression stages (Gould, 1997).

Neglected and underutilized species like *Lagenaria breviflora* seeds offer the potential to diversify not only the human diet, but also increase the number of food in the food system , thereby improving food security. Introducing this species into the food system would increase the opportunity to produce nutrient-rich food and improve overall human nutrition and health. Also, the seeds are rich inphyto-constituents and could be sources of therapeutic compounds in prevention and management of various illnesses.

5. Conclusion

This study reveals that *Lagenaria breviflora* seed is a good source of carbohydrate, lipid , moisture and ash and hence can serve as alternative source of energy and other nutrients needed to fight malnutrition in developing countries . The presence of appreciable amount of protein shows that the seed is vital in the repairing of worn out body tissues and also contribute significantly to growth . The presence of biologically active compounds such as flavonoid, steroids , alkaloids are evidence that it could be used in the management of different ailments and can be used as a potential source of useful drug. It also demonstrated that *Lagenaria breviflora* seed extract contains volatile compounds including acetone, acetic acid andmonoterpenes which suggest that the seed is potent in the prevention of carcinogenesis at both the initiation and progression stages. Therefore *Lagenaria breviflora* seed represents potentially useful pulps for management of nutritionally related diseases.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence this research work.

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