Assessment of Nutritional Composition of Atama (*Heinsia crinita*) and its Fungal Pathogens

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Abstract

The research on the assessment of nutritional composition of fresh Atama (Heinsia crinta) leaves and it's associated pathogens was carried out. Atama leaves were purchased from Mile 3 Market, Port Harcourt, Nigeria and analysed in the Laboratory of the Department Plant Science and Biotechnology, Rivers State University, washed and sent to the Department of Food Science and Technology for assessment of the Nutrient quality.The results of the Proximate analysis of fresh Atama leaf revealed that the leaf is rich in 26.4±0.1 of moisture, 4.3±0.1 of ash, 15.2±0.1 of lipid, 15.4±0.1 of fibre, 18.4±0.2 for protein and carbohydrate for 20.07±0.2. Mineral assessment revealed the presence of calcium, iron, potassium, sodium, magnesium, phosphorus. However, calcium was (300mg/300mg), iron (50mg/50mg), magnesium (156mg/156mg), phosphorus (149mg/149mg), potassium, sodium (52mg/53mg). Determination of vitamin revealed only the presence of vitamin A,C, and Thaimime. Phytochemical screening showed the availability of carotenoid, polyphenol, flavonoid, lignan, while anti nutrient, showed glycoside, oxalate but no values for saponin. Three fungal organisms (saccharomyces cerevisiae, candida sp and Mucor spp) were isolated to be responsible for the spoilage of Heinsia crinita. However saccharomyces cerevisiae sp had an incidence of 20% and candida spp with an incidence of 20% and Mucor spp had an incidence of 60% respectively. Consumption of Atama leaf is advised as they supply adequate energy and minerals for metabolic processes needed in maintenance of life, although it still prone to fungi spoilage.

Keywords: Atama (Heinsia crinita), nutritional quality and fungal pathogens.

INTRODUCTION

Heinsia crinita (commomly known as bush apple) is one of the green leafy vegetables plentifully found in Akwa Ibom State, Niger Delta region of Southern Nigeria. According to Etuk, *et* al., (1998), it grows undomesticated in the forest or selectively cultivated near the home and is called Atama leaf. It is a perennial rain forest shrub with woody stem and branches. It is classified as white or dark based on the taste and leaf colour by indigenes of Akwa Ibom State in Nigeria. The two varieties resemble each other morphologically. However, the dark variety is green and bitter than the white variety. Like other leafy vegetables, the harvested leaf has short shelf life. Consequently, most people trading on the leaf usually sundry it and sold as direct product. The leaves of both varieties (fresh or dried) are used to prepare traditional soup called "atama" soup which is cherished by most consumers for its beautiful aroma and lasting sweet taste in the mouth long after consumption. (Etukudo *et al.,* 2003). Atama is an underutilized crop and according to Jain *et al.,* (2013), an underutilised crop are domesticated plant species that have been used for centuries or more for their food, fibre fodder, oil or medical properties, but have been reduced in important over time. Underutilised crops are often considered minor crops because they are less important than staple crops and agricultural commodities in terms of global production and market value. Some of these crop species may be distributed globally, but are restricted to a more local production and consumption system.

Underutilised crops were once grown into discuss for a variety of agronomic, genetic, economic and cultural reasons. Farmers and consumers are using these crops less because they are in some way not competitive with other species in the same agricultural environment. Traditional crops typically do not meet modern standards for uniformity and other characteristics as they have been neglected by breeders from the private and public sectors (stamp *et al*, 2012).

Over the years there has been an increasing need to identify and isolate the fungi associated with the spoilage of Atama leaves. Thus spoilage refers to any change in the condition of food, in which the food becomes less palatable or even toxic, these changes may be accompanied by alterations in taste, smell, appearance or texture. A great variety of nutrient are found in Atama leaves and they play a role in food culture of Nigerians and African as well as their sister green leafy vegetable. (Mensah, 2008).

MATERIALS AND METHODS

Sample collection and preparation

Healthy atama leaves were purchased from mile 3 market in Port Harcourt. The leaves were divided into two; a part was washed and transported to the Department of Food Science and Biotechnology RSU for nutrient and anti-nutrient analysis. The other part was kept until visible signs of spoilage were noticed for mycological studies.

Nutrient analysis

The proximate composition was determined according to the method of AOAC (1990). Mineral contents were determined according to the methods of AOAC (2010).Analysis of Vitamin C was determined based on the method described by the Association of Analytical Chemist (AOAC, 1990).Analysis of vitamin B1 was determined according to the method of Okorie, (2010).

Determination of Anti-nutrient

Saponin content of the samples was determined by the method described by Harborne (1973), oxalate was determined according to the method of Onwuka (2005), tannin by Pearson, (1976), flavonoid and polyphenols was determined AOAC, (1990).

Mycological studies

Preparation of mycological medium

Sterilization of conical flask, slides, Petri dishes and all the equipment needed for the experiment was carried out in the laboratory. The glass wares were sterilized in the oven at 120^oC for an hour after washing with soap, while other equipment were surface sterilized with 70% ethanol to reduce microbial contamination (Chuku, 2009). Inoculating loops and scalpels were sterilized by dipping for 20 seconds in 70% ethanol and heated to red hot. The mycological medium used was Sabouraud Dextrose Agar prepared in a conical flask using the standard method. The mouth of the flask was plugged with non-absorbent cotton wool and wrapped with aluminium foil. The conical flask containing the mycological medium was autoclaved at 121^{o} C and pressure of 1.1kg cm-3 for 15 minutes. The molten agar was allowed to cool to about 40 ^{o} C and dispensed into Petri dishes at 15mls per plate and allowed to further cool and solidify.

Isolation of fungi from Atama

The direct plating method of Mehrotra and Aggarwal, (2003) was adopted where 0.5cm portion of diseased atama leaves were cut and dipped in 70% alcohol for 30 seconds and rainsed in distilled water before directly inoculated onto Sabouraud Dextrose Agar in Petri dishes containing ampicillin to hinder the growth of bacteria and this was done in triplicate. The inoculated plates were incubated for 5 days at ambient temperature of 25^{o} C ± 3^{o} C (Chuku, 2009). The entire set up was observed for 7 days to ensure full grown organisms. Pure cultures of isolates were obtained after a series of isolations.

Identification of fungal organisms from Atama

Microscopic examination of fungal isolates was carried out by the needle mount method (Cheesebrough, 2000). The fungal spores were properly teased apart to ensure proper visibility. The well spread spores were stained with cotton blue in lacto phenol and examined microscopically using both the low and high power objective. The fungi were identified based on their spore and colonial morphology, mycelia structure and other associated structures using the keys of Barnett and Hunter, (1998).

Determination of percentage incidence

The percentage incidence of fungal occurrence was determined by the formula stated below (Chuku *et al.*, 2019):

$$\frac{X}{Y} \times \frac{100}{1} = \text{\% incidence}$$

Where:

X= total number of each organism in a variety

Y= total number of all identified organism in a variety.

Statistical Analysis of Data

Data obtained from this study were expressed as mean ± SEM. Analysis of group data will be done by one-way analysis of the variance (ANOVA) followed by Duncan test for the establishment of significances. Statistical significance was set at (P<0.05).

RESULTS AND DISCUSSION

Table 1: Proximate Composition of Atama Leaf

Parameters	Composition (%)
Moisture	26.4 ± 0.1
Ash	4.6 ± 0.1
Lipid	15.2 ± 0.1
Fibre	15.4 ± 0.1
Carbohydrates	20.07 ± 0.2
Protein	18.4 ± 0.2

Table 2: Mineral Content of Atama Leaf

Parameters	Composition (mg/100g)
Calcium	301.67 ± 2.89
Iron	53.34 ± 3.05
Magnesium	157.34 ± 2.3
Phosphorus	48.67 ± 0.58
Potassium	309.00 ± 6.08
Sodium	53.00 ± 1.00

Table 3: Vitamins content of Atama Leaf

Parameters	Composition (mg/100g)
Vitamin C	119.0 ±1.00
Vitamin A	0.24 ±0.58
Thiamin	0.54 ±0.00

Table 4: Antinutrient and Phy-tochemical composition of Atama Leaf

Parameters	Composition (%)
Carotenoids	24.60±0.10
Polyphenols	6.27 ±0.10
Flavonoids	2.17±0.01
Lignant	1.9 ±0.02

Table 5 Fungal isolates from Atama leaves and their percentage incidences.

Isolates	Incidence %
Candida sp.	20
Saccharomyces cerevisiae	20
Mucor sp.	60

The proximate composition of Atama leaf recorded 26.4% for moisture, 4.2% for Ash, 15.1% for lipid, 15.4% for fibre, 20.3% for carbohydrate, and 18.6% for protein respectively. This result is higher than Asystasia gangetica reported by Chuku *et al.,* (2018). High moisture contents of Atama leaf help in maintaining the protoplasmic contents of the cells. (Etukudo, 2003), also supports greater activity of water soluble enzymes needed for metabolic activities of the leaf (Gbadamosi *et al.* 2011). However, high moisture content makes vegetables susceptible to spoilage, (Etukudo, 2008). Microorganisms that cause spoilage are known to thrive in foods containing high moisture content (Gbadamosi *et al.,* 2011). According to Gbadamosi (2011), Atama has a great amount of mineral elements present in it, hence it is very nutritious. But Ukam, (2008) reported that it could be the reverse of it, that Atama contains toxic metals which also contribute to the high ash percentage in it. Protein has been reported to be an important part of diets that is used in body building and repair of tissues. (Onward *et al.,* 2009). Fibres are non-hydrolysable polysaccharides which can be in soluble or insoluble state and increase faecal bulk. They form complex with protein, sugars and cholesterol. When taken in excess, they help to reduce the risk of colon cancer and scrub out the intestines leaving a much healthier digestive system. Dietary fibre helps in maintaining bowel movement and can prevent diverticulitis aiding the absorption of trace elements in the guts. Ash content in Atama is directly proportional to the mineral elements presents in it. This implies that the higher the ash content, the more nutritional the plant would be. The human system needs constant supply of Adenosine triphosphate (ATP) to function properly by striking a balance between external and internal factors. This energy form of ATP could be gotten from carbohydrate. The deficiency of carbohydrate in our diet has resulted in poor mental function, fatigue, endurance and lack of stamina. The fats and oil from vegetables have been shown to possess hypolipidemic activity and therefore could ameliorate the occurrence of diseases associated with hyperlipidemia like coronary artery disease, myocardial infarction cerebrovascular accident and hypertension (Onward *et al.,* 2009). Fibre is known to cleanse the digestive tract, remove potential carcinogens from the body, as well as keep blood sugar levels under control. (Ukam, 2008) carbohydrate provides the body with fuel and energy that is required for daily activities and exercise.

The mineral composition of the present study has shown that Atama possessed several mineral components. The results obtained indicated that calcium (C_a), iron (F_e), magnesium (Mg) phosphorous (p), potassium (k) and sodium (Na) recorded 300mg, 50mg, 156mg, 49mg, 52mg and 3mg respectively. The mineral content result is higher than *A. gangetica*, reported by Chuku *et al*., (2018). Magnesium (mg) is important in decreasing blood pressure by dilating arteries and preventing abnormal heart right. (Etukudo, 2003), it is important as functioning to protect the soft tissues from hardening and protects the linings of the arteries from stress due to fluctuation in blood pressure. (Ezekiel *et al.*, 2016 and AOAC, 1990).

The vitamin content of the current study has shown that Atama leaf have vitamin A, C, Thiamin(mg). Vitamin A, C and thiamin recorded 0.24 $\pm$0.58, 119.0 $\pm$1.00, and 0.54$\pm$0.00 respectively. The vitamin content of this study iss higher than *Pleurotus Ostreatus* reported by Agbagwa *et al.*, (2020) and lower than citrus peels reported by Chuku *et al.,* (2020). The presence of vitamin helps in the amelioration and prevention of diseases in the body which help to maintain a healthy life style. (Ekpa, 1991 and Okafor, 1983).

The anti-nutrient result revealed the presence of carotenoid, polyphenols, flavonoid and lignant. This result is higher than green pea reported by Chuku *et al*., (2019).

Oxalate and tanins is a known component of herbs used in traditional medicine used for the treatment of common headaches, fevers, diarrhoea, anaemia, high blood pressure and family infertility. It is used for the treatment of cardiac infections along with other oilment such as cough, and chest pain (Mensah, 2008).

The result of fungi isolates revealed a total of three fungal organisms (*Saccharomyces cerevisiae*, *Candida spp* and *Mucor spp*) to be associated with the spoilage of *Heinsia crinita*. *S.cerevisiae* and *candida spp* had 20% incidence while *Mucor* spp recorded 60% incidence respectively (Chuku, *et al.,* 2019). Different researchers have identified these organisms to be responsible for the spoilage of Atama (*Heinsia crinita).* Chuku *et al.*, (2019) also identified *Mucor spp* as mycoflora of green Pea (*Pisum sativum*), Wogu and Ighile, (2014) identified *Mucor spp* and *Saccharomyces cerevisiae* to be spoilage organisms of avocado fruit. Jidda and Musa, (2016) isolated *Candida tropicalis* and *Saccharomyces cerevisiae* as spoilage Fungi of garden egg fruit. Wekhe *et al.*, (2020) identified *Mucor spp* to be associated with the spoilage of different varieties of Bread fruit. The low percentage incidence of *Saccharomyces cerevisiae* is attributed to their bitter nature and it disagrees with the research of Chuku and Agbagwa 2020 on post harvest fungal pathogens of Beet root (*Beta vulgaris*), and the research of Strausbaugh, (2008) on the ability of yeast to cause Beet root rot.

In general, Atama is rich in nutrients and anti-nutrients and its compound can be used in the development of novel drugs for future use but however, they are prone to fungal contaminations. Therefore, proper care should be taken during harvest and storage to reduce contamination.

CONCLUSION

The research carried out on the leaves of *Heinsia crinita* has proved to be beneficial. The phytochemicals present has revealed that the leaves posses potent antioxidant properties. Also the nutritional content of the leaves support their use as food. The presence of bioactive compounds and pharmacological activities proved the potency of plant in the development of novel drugs for future use.

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