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Amino Acid and Heavy Metal Composition of *Afzelia africana* Leaves

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Abstract Malnutrition is one of the major problems in Nigeria and other developing countries. The country is blessed with rich soil and favorable climate, but lack of irrigation facilities has resulted to seasonality; and ignorance of nutritional value of lesser-known vegetables have contributed to scarcity of vegetables for the increasing population. This study investigated the amino acid and heavy metal composition of *Afzelia africana* leaves. Sixteen amino acids were found in varying proportions in the protein of the vegetable. All essential amino acids, except methionine and tryptophan were present in good quantities. Phenylalanine had the highest concentration of 2.27g/100g protein while isoleucine had the lowest concentration of 1.71g/ 100g protein. The heavy metal analysis was done by atomic absorption spectrometry. Results obtained showed the presence of the essential metals: iron (8.49ppm) and nickel (0.03ppm); and the non-essential metals: lead (0.75ppm) and mercury (0.64ppm). Results revealed that the protein contained in the leaves of *Afzelia africana* is of high quality; however the presence of the toxic metals is an issue of great concern.

Keywords: *Afzelia Africana, amino acids, vegetable, heavy metals, FAO/ WHO limit*

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1. Introduction

Malnutrition is a state in which one's physical function is impaired such that one can no longer maintain adequate bodily performance processes such as growth, pregnancy, lactation, physical work, resistance and recovery from disease [1]. It is estimated that approximately 1.6 billion people are malnourished [2,3]. According to the Vanguard Newspaper, the fifth report on World Nutrition Situation states that stunting affects 147 million pre-school children in developing countries and Nigeria constitutes about 10 million of such children [4]. The consequences of malnutrition can be dire. Effects include maternal mortality and premature birth caused by iron deficiency; night blindness and impaired immune system caused by insufficient vitamin A. In 2001, 54% of deaths in children in developing countries were associated with child malnutrition [5]. Vegetables supply nutrients that combat malnutrition. It has been reported that under-consumption of fruits and vegetables is among the top 10 risk factors leading to micronutrient malnutrition and is associated with the prevalence of chronic non-communicable diseases [6]. In Nigeria, most vegetables consumed by people are seasonal. They thrive mostly during the wet season and are scarce during the harmattan/dry season. Though these vegetables can be grown all year round, lack of irrigation facilities has made it virtually impossible to produce enough vegetables for the increasing population. The lesser-known vegetables therefore supplement the more-known vegetables. *Afzelia africana* leaves belong to such lesser-known vegetables. *Afzelia africana*, also known as African mahogany or African oak is a large deciduous tree. It is a leguminous tree which belongs to the family of Fabaceae, sub-family Caesalpiniaceae and found in humid and dry forests. The genus name *Afzelia* was named in honour of Adam Afzelius, a Swedish botanist who made the first collection during his visit to Sierra Leone in the eighteenth century [7]. It is widely distributed in many African countries including Benin, Burundi, Cameroon, Central African Republic, Cote d'Ivoire, Gambia, Ghana, Guinea-Bissau, Kenya, Liberia, Mali, Nigeria, Senegal, Sierra-Leone, Sudan, Tanzania and Uganda. In Nigeria, it is called *akpalata* in Igbo, *apa* in Yoruba, *yiase* in Tiv, *ukpo* in Idoma and *kawa* in Hausa.

Afzelia africana is a very attractive medium sized tree. The stem has relatively thick, unequal buttresses with a light concave profile, generally 1 to 1.5m tall and 1 to 2m wide. The reddish-grey, scaly bark, about 2cm thick, exfoliates in rounded patches and serves as protection against frequent bush fires of the dry season. The leaves are bright green and turn yellowish during harmattan season in Nigeria. The fruits are flat, hard, dark brown or black pods. Each pod contains several oblong black bean-shaped seeds with bright orange aril. *Afzelia africana*

survives very harsh weather conditions and effects of some global climate change [8]. Several studies on the plant showed that it contains vital nutrients such as protein, fat, micro and macro minerals. Antinutrients such as haemagglutinnin, oxalate, phytic acid, saponins, tannins and trypsin inhibitor have been found in the tree foliage [9]. It has been shown that every part of the plant is of great importance [10]. Wood from *Afzelia africana* is used in carpentry; canoe and house building; furniture making; flooring and heavy construction; wood carvings and other traditional uses [11]. The seeds contain about 31% fat and may be a source of seed oil for domestic and industrial uses. Seeds are also milled into flour which serves as soup thickener or substitute for wheat flour in biscuits and doughnuts.

Tender leaves and shoots of *Afzelia africana* are consumed as delicacies in yam dish [8]. The leaves are used as forage during dry season to feed ruminants [12]. The leaves are eaten cooked as vegetable and young leaves are mixed with ground cereals before cooking [13]. The nitrogen-rich leaves also serve as soil improver especially when mulching and littered. A leaf decoction mixed with *Syzygium guineensis* leaves and xylopia fruit forms a drink used to treat oedema [11]. The flowers are used as condiment in sauces. In traditional medicine, an infusion of the bark is used against paralysis; a decoction against constipation. The crushed bark mixed with honey is used in veterinary medicine while the ash can be prepared with Shea butter as soap and used against lumbago. The root of the plant can be pulverised and employed in the treatment of various ailments and disease conditions such as hernias, gonorrhoea, and stomach ache. *Afzelia africana* is one of the non conventional vegetables obtained from forests, and supplements the conventional ones obtained from farms and home gardens. Despite the usefulness of this plant, it is on the verge of extinction, owing to population explosion, urbanization and the resultant degradation of natural forests; and the preference of farmers to produce the conventional vegetables.

Though many studies have been carried out on the plant, none has investigated the individual amino acids present in the nitrogen-rich leaves which are consumed by man and animals. This study was therefore carried out to investigate the amino acid and heavy metal composition of the leaves.

2. Materials and Methods

2.1. Sample Preparation

Fresh leaves of *Afzelia africana* were obtained from Umuahia, Abia state of South-eastern Nigeria. They were air-dried, pulverized and stored in air-tight containers for analysis.

2.2. Amino acid Analysis

The amino acid profile was determined using the method described by [14]. The sample was dried to constant weight, defatted, hydrolyzed, evaporated and loaded into the techno sequential multi-sample amino analyzer. All reagents used were of analytical grade

2.3. Heavy Metal Composition

Wet digestion method was done using the acid mixture of nitric acid, perchloric acid and sulphuric acid. The digested sample was filtered and diluted. The resulting extract was then analyzed with the FS 240 Varian Atomic Absorption Spectrophotometer using methods described by [15].

3. Results

Table 1. Amino acid composition of leaves of *Afzelia africana*(g/ 100g protein)

Amino acid	Afzelia africana	FAO Ref protein
Aspartic acid	1.14	-
Threonine	1.69	2.80
Serine	1.19	-
Glutamic acid	1.09	-
Proline	0.60	-
Glycine	1.03	-
Alanine	1.01	-
Valine	1.95	4.20
Cysteine	-	-
Methionine	-	2.20
Isoleucine	1.71	4.20
Leucine	2.67	4.20
Tyrosine	1.18	-
Phenylalanine	2.27	2.80
Lysine	2.48	4.20
Histidine	2.19	-
Arginine	1.28	-
Ornithine	4.89	-

Table 2.Percentage comparison of the essential amino acid composition of the vegetable with the FAO standard (sample/standard x 100) %

Amino acids	Afzelia Africana
Threonine	60
Valine	46
Isoleucine	41
Leucine	64
Phenylalanine	81
Lysine	59

Table 3. Heavy metal composition of *Afzelia africana* leaves

Heavy metals	Concentration(ppm)
Chromium	0.00
Lead	0.75
Mercury	0.64
Iron	8.49
Nickel	0.03
Cadmium	0.00
Arsenic	0.00

4. Discussion

Amino acids are chemical units from which proteins are formed as well as the end products of protein digestion. They are useful to human beings and animals. Sixteen amino acids were found in varying proportions in the vegetable. Seven essential amino acids were present. These are leucine, lysine, isoleucine, histidine, phenylalanine,

threonine, and valine as shown in Table 1. Results showed appreciable quantity of leucine (2.67g/100g) protein which is about 64% when compared with the FAO reference protein (Table 2). This value is acceptable since leucine helps to lower blood sugar and also helps to slow the deterioration of muscle tissue by increasing the combining of muscle proteins [16]. Isoleucine and valine had low values of (1.71 and 1.95) g/100g protein respectively; which give 41% and 46% when compared with the FAO standard. Isoleucine is needed for the healing and repair of muscle tissue, skin cells and bones. This essential amino acid is particularly useful to athletes. It also regulates blood sugar. Valine is needed for muscle metabolism and coordination; it supplies energy to the muscle tissue to promote muscle growth and tissue repair. During intense physical activity, it supplies the muscles with extra glucose for energy production, thereby preventing breakdown of muscle fibres. High levels of valine could lead to hallucinations. Appreciable amounts of lysine and threonine were obtained (2.48 and 1.69) g/100g protein; and when compared to the FAO standard had 59% and 60% respectively. Lysine helps in the absorption and conservation of calcium; formation of collagen, and conversion of fatty acids to energy. It is needed for synthesis of enzymes and hormones. Phenylalanine had a high value of 2.27g/100g protein and 81% when compared with the FAO standard. It converts to the amino acid tyrosine, needed for production of brain chemicals, thyroid hormones and protein. It is beneficial for healthy nervous system; boosts memory and learning. It elevates mood and alertness and may be useful against depression. The amino acids, tryptophan, cysteine and methionine were absent or too low to be detected. It has been reported that cysteine and methionine are commonly deficient in green leafy vegetables [17].This means that sources of these amino acids should be consumed together with the vegetable. The presence of arginine and histidine in the vegetable indicates that it could be recommended for children since they need arginine and histidine in their foods [18]. These results show that the protein in *Afzelia africana* leaves is of high grade.

The presence of heavy metals in the vegetable could be as a result of contamination during growth, processing or handling. Heavy metals are harmful because they are not biodegradable; have long biological half-lives and accumulate in different parts of the body. Vegetables which are contaminated can therefore be toxic and produce undesirable side effects [19]. Prolonged consumption of unsafe concentrations of heavy metals may lead to accumulation of heavy metals in the kidney and liver of humans, thus, causing disruption of numerous biochemical processes, and leading to cardiovascular, nervous, kidney and bone diseases [20]. Table 3 shows the presence of four heavy metals: iron, lead, mercury, and nickel and the absence of chromium, cadmium and arsenic. Iron, an essential metal had the highest concentration of 8.49ppm. The permissible limit set by FAO/WHO in edible plants was 20ppm [21]. However, the WHO limit has not yet been established for iron for medicinal plants [22]. Iron is an essential element for man and animals. It is an integral part of many proteins and enzymes that maintain good health. It facilitates the oxidation of carbohydrates, protein and fat to control body weight, which is very important factor in diabetes [23]. Low iron content causes gastrointestinal infection, nose bleeding and myocardial infection [23]. Excess amounts result in toxicity and even death [24]. Result obtained showed the concentration of lead to be 0.75ppm. It has been reported that the permissible limit set by FAO/WHO in edible plants was 0.43ppm [21, 25]. However the WHO, China, Malaysia and Thailand set 10ppm as the limit for medicinal herbs [22]. Lead is a non-essential metal. The target organs for lead poisoning are the bones, brain, blood, kidneys, and the thyroid glands. Reports have shown that lead exposure causes severe anaemia, permanent brain damage, neurological disorders, reproductive problems, diminished intelligence and many other diseases [26]. The concentration of mercury is 0.64ppm as shown in Table 3. This value is above the permissible limit set by FAO/WHO in edible plants and medicinal herbs which were 0.02ppm and 0.1ppm respectively. Recently,there is growing interest in mercury and its effects on humans and wild life, due to the discovery of mercury as the causative agent in the Minamata disease in Japan[27].High levels of mercury can permanently damage the brain, kidneys, and developing foetus. Exposure to methyl mercury is more injurious to children than adults because it interferes with their normal development, as more of it passes into their brains. The concentration of nickel obtained was 0.03ppm as shown in Table 3. This value is within the permissible limit set by FAO/WHO in edible plants (1.63ppm). However, for medicinal plants, the WHO limit has not yet been established for nickel. Nickel, though an essential metal has been identified as a suspected carcinogen and adversely affects lungs and naval cavities [22]. Nickel plays an important role in the production of insulin and so is needed in minute quantity. Its deficiency results in the disorder of liver [22].

5. Conclusion

This study has shown that *Afzelia africana* leaves contain appreciable amounts of amino acids which indicate that the protein is of good quality. The amino acids play multiple roles in health promotion and protection. However, the presence of the heavy metals, iron, lead, mercury and nickel is an issue of great concern since these metals can be toxic even at very low concentrations. Although the leaves of *Afzelia africana* are consumed in many regions of Africa especially during the dry season when the conventional vegetables are scarce, caution should be applied to avoid the accumulation of toxic metals in the body which have adverse effects.

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