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Amino acid composition of the seeds of *Moringa oleifera* (Lam), *Detarium microcarpum* (Guill & Sperr) *and Bauhinia monandra (*Linn.*)*

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ABSTRACT

The seeds of *Moringa oleifera* (Lam), *Detarium microcarpum* (Guill & Sperr) and *Bauhinia monandra* (Linn) were analysed for amino acids contents. The concentrations (g/100g protein) of the essential amino acids lysine, cystine, valine, methionine, isoleucine, leucine, phenylalanine and threomine were M. oleifera 3.21, 2.09, 3.05, 1.09, 4.01, 5.74, 4.24 and 3.03, *D. microcarpum* 2.14, 1.07, 2.13, 0.74, 2.35, 2.35, 2.5 and 2017, *B. monandra* 2.86, 1.11, 3.54, 1.54, 2.31, 2.13, 3.77 and 2.70 respectively. Other non –essential amino acids were in various quantities in the seeds. The statistical analysis of variance (ANOVA) showed that there was no significant difference in the amount of amino acids in the seeds of these plants. Therefore these plants can serve as good sources of amino acids for both man and livestock.

INTRODUCTION

Recently, the analysis of nutritional value of wild plant materials attracted attention due to the fact that they contain significant amount of essential nutrients that can be used for both human consumption and in the formulation of animal feeds. The proximate composition of seed protein of some wild plants of Nigerian origin showed that, they could be adequately used in the formulation of animal feeds provided the level of their toxic substances are known [1]. For instance, *Amaranthus viridis* had been reported to be an excellent source of protein. Its amino acid composition compared favourably to that of the World Health Organisation (WHO) protein standard [2]. The protein content of the seeds of Mexican wild lupins *Lentils (Splendens, rotundiflorus, elegans, Simulans, exaltatus*) and mango were found to be very high and could be used for animal feeds if alkaloids inherent in them were eliminated [3,4,5].

Protein is essential for growth and development of living organisms and it constitute 80 – 90% of all organic substances in animal body. Protein quality is measured by the type of amino acids present. There are twenty different types of amino acids, eight of which are essential because they are not manufactured by the animal's body. This include Lysine, Leucine, Isoleucine, Methionine, Phenylalanine, Threonine, Tryptohan and Valine [7]. The amino acid histidine is essential for the growth and development of children but it is only synthesized by adults. Other non-essential amino acids that are required to maintain health, can be synthesized by the body if supplied with necessary nitrogen. These non-essential amino acids include alanine, arginine, aspartic acid, cystine, glutamic acid, glycine, hydroxyproline, proline, serine and tyrosine [8]. Dietary protein with all the essential amino acids in the proportion required by the body is said to be a high quality protein. If the protein is low in one or more of the essential amino acids, the protein is of low quality. The amino acid that is in short supply is called limiting amino acid [9]. Generally, many plant proteins are low in one of the essential amino acids. A combination of plant proteins, such as grains, with pulse or seeds leads to a high quality protein which is just as good as protein from animal foods.

This paper reports on the amino acid composition in the seeds of the wild *Moringa oleifera* Lam (family: moringacea), *Detarium microcarpum* Guill & Sperr (family: caesalpinioideae) and *Bauhinia monandra* Linn. (family: caesalpiniaceae) in Zaria, Nigeria.

EXPERIMENTAL

Sample collection and preparation

The seeds were collected from villages around Samaru-Zaria, Kaduna state, Nigeria. The samples were deposited at the Herbarium in the Department of Biological Sciences, Ahmadu Bello University, Zaria for identification and filing. The seeds were oven dried at 60^{o}C in an air circulated oven, ground with porcelain

mortar and pestle to fine particles and stored in screw capped plastic containers. Chemical analyses were carried out on the ground samples.

Moisture Content

A clean crucible was dried to a constant weight in air- circulated oven at 105^0C. A known weight of the seed sample was placed in the crucible and dried in the oven at 105^0C to constant weight for 2 hours. The crucible and its content was cooled in a desiccator and weighed. The moisture content was calculated and expressed in percentage [10]

Ash and Organic Matter Content

A crucible was pre-heated in a muffle furnace at 500ºC cooled in desiccator and weighed. 2g of the sample was transferred into the crucible and weighed. The crucible and its content was kept in the muffle furnace at 525ºC until white ash was obtained after twelve hours. The organic matter content was calculated by subtracting the percent ash content from 100 [10].

Protein Content

2g ground sample was weight into a 300cm^3 Kjedahl digestion flask and few antidumping granules (1.0g of K_2S0_4 and anhydrous $CuS0_4$) were added to the flask. 25cm^3 of concentrated sulphuric acid (98% w/w) was added. The flask was fixed on the Kjedahl digestion jack and heated slowly at first until frothing subsides and then vigorously with occasional rotation (to ensure even digestion and avoid overheating of the content) until a clear solution was obtained. The solution was diluted to 250cm^3 and poured in to the Kjedahl distillation flask. Few drops of phenolpthalein and 70cm^3 of 40% (w/v) sodium hydroxide solution were added. The ammonia liberated was trapped in 4% (w/v) Boric acid solution (100cm^3) contained in a 500cm^3 conical flask to which two drops of methyl red indicator was added. Distillation was stopped after colour change. The content of the conical flask was titrated with 0.1 mol dm^{-3} hydrochloric acid. The percentage protein was calculated [11].

Amino Acids Content of Samples

3.0g of ground sample was extracted with petroleum ether (40 – 60ºC) using soxhlet extractor for six hours [11]. 30mg of the defatted samples was weighed into a glass ampoule and 7.0cm^3 of 6.0mol/dm^3 hydrochloric acid was added. Oxygen was expelled by passing nitrogen into the ampoule (to avoid possible oxidation of some amino acids during hydrolysis). The ampoule was sealed with Bunsen flame and put in an oven preset at 105ºC for 22 hours, after which it was allowed to cool, broken at the tip and the content filtered. The filtrate was evaporated to dryness at 40ºC under vacuum in a rotary evaporator. The residue was dissolved with 5.0cm^3 of acetate buffer (pH 2.0), stored in plastic bottle and kept in deep freezer for 24 hours. Five to ten microlitres of the hydrolysate was loaded on the Technicon Sequential Multi-Sample (TSM) amino acid analyzer (DNA 0209) made by Technicon (Ireland) Ltd. This was dispensed into the cartridge of the analyzer and the analysis lasted for 76 minutes [12].

RESULTS AND DISCUSSION

The moisture content of the seeds are generally low (Table 1) this is an indication that they can be stored for a long time without the development of moulds. *Detarium microcarpum* moisture content was 4.68%, Akpata and Miachi [13] reported a lower value of 3.5%, the differences could be due to environmental factors and time of the experiment. The moisture content of *M. oleifera* observed was 3.76%, Campden and Chlorlery- wood [14] reported a higher value of 4.5%. *Bauhinia monandra* moisture content (7.55%) was high, but this value fall below the 15% moisture

Table 1: Moisture, ash, organic matter and protein contents of *Moringa oleifera,Detarium microcarpum, Bauhinia monandra.*

	Moringa oleifera	Detarium microcarpum	Bauhinia monandra
Moisture content (%)	3.76 ± 1.73	4.69 ± 0.1	7.55 ± 0.3
Ash content (%)	3.60 ± 0.05	2.77 ± 0.16	4.44 ± 0.07
Organic matter content (%)	96.4 ± 0.05	97.23 ± 0.16	95.56 ± 0.07
Protein Content(%)	40.31 ± 1.63	35.96 ± 1.63	33.09 ± 2.30

n = 3

Table 2: Amino acid content g/100g protein of *Moringa oleifera, Detarium microcarpum, Bauhinia monandra*

Amino acids	*Moringa oleifera*	*Detarium microcarpum*	*Bauhinia monandra*
Lysine	3.21	2.14	2.86
Phenylalamine	4.24	2.58	3.77
Leucine	5.74	2.35	2.13
Isoleucine	4.01	2.35	2.31
Methionine	1.00	0.74	1.54
Valine	3.05	2.13	3.54
Cystine	2.09	1.07	1.11
Threonine	3.03	2.17	2.70
Glutamic acid	14.43	9.78	11.75
Arginine	8.00	5.66	6.74
Aspartic acid	6.88	4.79	6.02
Serine	4.22	3.12	4.58
Glycine	4.96	2.41	3.09
Alanine	3.22	2.10	2.99
Histidine	2.20	1.13	2.36
Proline	2.09	2.09	2.37
Tyrosine	2.37	1.06	3.18

content required as safe storage limit for plant food materials [2].

Ash content is the measure of the total mineral content of a material. From the result (Table 1), the ash content of *B. monandra* was 4.44%. This value is higher than that of *D. microcarpum* (2.77%) and *M. oleifera* (3.60%). *Detarium microcarpum* ash content (2.77%) is lower than the value reported by Akpata [13].

The organic matter content of a plant material is the measure of the total lipids, proteins and carbohydrates. *Detarium microcarpum* has the highest value (97.23%) of organic material than the other two seeds. According to Dalziel [15], the seeds of this plant are highly nutritious and are used as food stuff for cattle by the Nupe people of Niger state. The organic matter of *M. oleifera* (96.4%), agrees with the report of Ram [16] who described the seeds as highly nutritious. *Bauhinia monandra* had 95.5% organic matter, this value is lower than that of the other two plants. Generally, the seeds of the three plants can be described as having high nutritional value.

Moringa oleifera had the highest protein content of 40.31% (Table 1), however Makkar [17] reported a lower value of 38.4%. The result compared with other proteins of plant seeds like soya beans (*Glycine max*) 51.4%, groundnut (*Arachis hypogaea*) 51.3% and cottonseed (*Gossypium* spp) 51.5%[18,21]. *Detarium microcarpum* protein observed is 35.96%, Akpata [13] reported a high value of 37.1%. The difference could be in environmental factors. The results proved that these plants are potential sources of proteins.

The amino acid content of the seeds were generally high (Table 2). Among the essential amino acids, leucine was highest in *M. oleifera* (5.74g/100g protein) than in *D. microcarpum* (2.34 g/100g protein) and *B. monandra* (2.13 g/100g protein). Leucine is responsible for regulating the blood sugar concentrations, growth and repairs of muscles/tissues, hormone production, wound healing and energy production. Its deficiency causes dizziness, headaches, fatigue, depression, confusion, irritability and hypoglycemia in infants [20]. Phenylalanine is also highest in *M. oleifera* (4.24 g/100g protein) than in *D. microcarpum* (2.58 g/100g) protein while in *B. monandra* it is 3.77 g/100g protein. It is another essential amino acid that is used by the brain to produce norepinephrine (a chemical that transmits signals between the nerve cells and the brain). It keeps the body alert and reduces hunger pains. It is an antidepressant and helps in improving memory and its deficiency could result in slow growth, liver damage, and skin lesions [21]. Isoleucine content was high in *M. Oleifera (*4.01 g/100g protein), 2.35 g/100g protein in *D. microcarpum* and 2.31 g/100g protein in *B. monandra*. This amino acid helps in development and repair of muscles, development of hemoglobin and acts as energy regulator. Its deficiency results in ailments similar to

leucine deficiency. Lysine concentration was highest in *M. Olifera* (3.21 g/100g protein), while *B Monandra* was 2.86g/100g protein and 2.14g/100g protein in *D. microcarpum*. Lysine insures the adequate absorption of calcium, help the formation of collagen, in addition it aids the production of antibodies, hormones and enzymes. Lysine deficiency may result in tiredness, inability to concentrate, irritability, bloodshot eyes, retarded growth, hair loss, anemia and reproductive problems [20].

Threonine concentrations in these seeds were 3.03g/100g protein in *M. oleifera*, 2.17 g/100g protein in *D. microcarpum* and 2.70 g/100g protein in *B. monandra*. These values are considered to be high when compared to the World Health Organisation protein standard [2]. Threonine is necessary in the body because, it produces antibodies, prevent fat buildup in liver and assist metabolism and assimilation [20]. It is an important constituent of collagen, elastin and enamel protein. Its deficiency has been associated with skin disorders and weakness [21]. *Bauhinia monandra* had the highest content of valine (3.54, g/100g protein) while *M. oleifera* had 3.05 g/100g protein and *D. microcarpum* had 2.13 g/100g protein. Valine promotes mental vigor, muscle coordination and calm emotions [22]. Methionine is a sulphur containing amino acid, its concentration was highest in *B. monandra* (1.54 g/100g protein), while *M. oleifera* and *D. microcarpum has 1.0* g/100g protein and 0.74 g/100g protein respectively. It functions as a supplier of sulphur, which prevents disorders of hair, skin and nails. It prevent arterial fat buildup, regulate ammonia formation and creates ammonia free urine which reduces bladder irritations; its deficiency results in similar symptom like phenylalanine [21]. From the result obtained, histidine had the highest concentration occurring in *B. monandra* (2.36. g/100g protein), *M. oleifera* (2.20, g/100g protein), while *D. microcarpum* content was 1.13. g/100g protein. Histidine is essential especially in children, it is used for growth, tissue repairs and histamine development [22]. Cystine concentration in the seeds was 2.09 g/100g protein in *M olefera*, 1.07 g/100g protein in *D. microcarpum* and 1.11 g/100g protein in *B. monandra*. This shows that cystine values are relatively low in *D. microcarpum* and *B. monandra* but high in *M. oleifera*. It is a sulphur containing amino acid which acts as an antioxidant and protects the body from radiation and pollution. It also aids protein synthesis and prevents cellular changes. In addition, it deactivate free radicals and neutralize toxins.

Among the non-essential amino acids, *Moringa oleifera* had 14.48, 8.0, 6.88 and 4.22 (g/100g protein) of glutamic acids, arginine, aspartic acid and serine respectively. On the other hand, *B. monandra* had 11.75, 6.74, 6.02 and 4.58 (g/100g protein) as concentrations for glutamic, arginine, aspirtic acid and serine respectively. While glutamic acid in *D. microcarpum* was 9.78g/100g protein, arginine (5.66g/100g protein) aspartic acid (4.79g/100g protein) and serine (3.12 g/100g protein).

The result indicates that *M. oleifera* is a good source of essential amino acids. This agrees with Ram [16] who reported that, *M. oleifera* seeds contained all the essential amino acid in appreciable quantities. Similarly, the amino acid composition of *B. monandra* seeds confirmed that, the plant is a good source of amino acids [19]. Statistical analysis of variance (ANOVA) showed that, there was no significant difference between the mean values of the total amino acids composition in the three plants at 95% confident limit. This implies that, all these plants can equally serve as good sources of amino acids for man and livestock.

CONCLUSION

In conclusion, these wild plants contained amino acids in appropriate quantities that can serve as supplementary potential sources of essential amino acids to man and livestock, provided toxicants present in them are eliminated.

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