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## The protein quality of raw leaf, seed and root of *Moringa oleifera* grown in Rivers State, Nigeria

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### ABSTRACT

This study was carried out to ascertain the quality of the root, seed and leaf proteins of *Moringa oleifera* grown in Rivers state Nigeria. The amino acid compositions were analyzed with the HP 6890 gas chromatography. A total of 17 amino acids were recorded with Glutamate having the highest concentration in g/100g protein (13.53, 14.76, 15.14) and Methionine having the least concentration (0.76, 0.97, 0.95) in the root, seed and leaf of *M.oleifera* respectively. On comparing the essential amino acids obtained in the root, leaf and seed of *M. oleifera* to the World Health Organization (WHO) reference pattern for essential amino acids, it was observed of the Limiting amino acids were isoleucine (43.94%), Lysine (62.71%), Isoleucine (55.59%) in the root, seed and leaf of *Moringa oleifera*. These results show that leaf and seed *M. oleifera* are good sources of amino acids for both man and livestock. Also, the exceptionally high level of arginine in the *M. oleifera* seeds makes it an excellent source of fortification of cereal weaning food.

**Keywords:** *Moringa oleifera*, amino acid, reference pattern, protein quality, fortification.

### INTRODUCTION

The knowledge that proteins differ in quality dates as back as far back as the 19th century. In 1915, the great American nutritionist Mendel divided protein into two classes; First class protein which when fed to rats allowed growth and second class protein which when fed to the rats allowed growth failure [1]. It has been the general assumption that proteins of animal origin such as meat, fish are of good quality while those of plant origin are of low quality. Undeniably, this assumption holds true in most cases, however current research on plant proteins [2,3,4,5] shows that some of them can compete favorably with proteins from animal sources.

Protein is essential for growth and development of living organism and it constitute 80-90% of all organic substances in animal body [6]. Protein quality is currently measured by the type of amino acids, eight of which are essential because they are not manufactured by the animal. These include Lysine, Leucine, Isoleucine, Methionine, Phenylalanine, Threonine, Tryptophan and Valine [7]. Dietary protein with all essential amino acids in the proportion required by the body is said to be of high quality. 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 said to be the limiting amino acid [8].

*Moringa oleifera* is the most widely cultivated species of genus *Moringa* in the family Moringaceae [9]. It is an exceptionally nutritious vegetable tree with a variety of potential uses [9,10,11]. *M. oleifera* is actively cultivated by

the World Vegetable Centre in Taiwan, a centre for vegetable research with a mission to reduce poverty and malnutrition in developing countries through improved production and consumption of vegetables. As a traditional food in Africa, it has the potential to improve nutrition, boost food security, foster rural development and support sustainable land care.[12]. *M. oleifera* is especially promising as a proteinous food source in the tropics because the tree is in full leaves in dry season when other foods are typically scarce [13]. *M. oleifera* leaves are a significant source of B vitamins, vitamin C, provitamin A, vitamin K, Manganese and other essential nutrient [14].

Protein energy malnutrition a form of malnutrition prevalent in developing countries affects children the most because they have less protein intake [15]. Moringa trees have helped to combat malnutrition especially among infants and nursing mothers. Three non-governmental organizations in particular-Tree for life, Church World Service and Educational Concerns for Hunger organization have advocated Moringa as natural nutrition for the tropics [16].

This paper reports on the amino acid composition and essential amino acid scores of the seed, leaf and root of *Moringa oleifera*. It also examines their protein quality on comparing them with the W.H.O reference pattern [17,18,19].

## MATERIALS AND METHODS

### 2.1 Collection and Preparation of Test Samples:

The roots, leaves and seeds of *Moringa oleifera* were collected from Mich farm at Eleme community, River state Nigeria and duly authenticated at the herbarium in the Faculty of Biological sciences University of Port Harcourt, Nigeria. The *M.oleifera* seeds, leaves and roots were cleaned and then dried at 60C in an air circulated oven, grounded with mortar and pestle to fine particles and stored in screw capped containers. Chemical analyses were carried out in the ground samples.

### 2.2 Reagents

All reagents used in this study were of analytical grades with high purity.

### 2.3 Determination of Amino Acid Composition in the *M.oleifera* root, seed and leaf.

#### 2.3.1 Processing

5.0g of the each sample was extracted with petroleum ether (40-60 °C) using soxhlet extractor for six hours [20]. 30mg of defatted samples was weighed into an extraction thimble of the Soxhlet extraction apparatus and hydrolyzed with deionized water [21]. The amino acid content of each sample was recovered with 30ml of Methylene chloride using the Soxhlet extraction apparatus [21]. The extract was then concentrated to 1ml using a rotary evaporator.

#### 2.3.2 Gas Chromatography Analysis

First, the instrument was calibrated by injecting 1ml of the standard mixture of amino acid solution into the column inlet of the HP 6890 gas chromatograph. The pulsed flame photometric detector (PFPD) in the chromatograph monitors the outlet stream. Thus the retention time of each component (amino acids) is determined. The results from the gas chromatograph are channeled to a data station where an instrument specific software program interprets and translates instrument control and data acquired from the gas chromatography process. This action ascertains the efficiency of the standard mixture of amino acids and calibrates the instrument. The entire procedure was then repeated for the analyses of 1ml of each of the test samples. The period of analysis lasted for about 60mins.

#### 2.3.3 Gas Chromatography Conditions

The gas chromatograph was HP 6890 model. The carrier gas was Hydrogen while the injection temperature was split injection. The split ratio was 20:1. The inlet temperature and column type were 250°C and HP5 respectively. The column dimensions were 30mmx0.25x 0.25 micro meters. The oven program initial temperature was 60°C. The first ramping was 8°C per minute maintained for 2 minutes while the second ramping was 12°C per minutes maintained for 2 minutes. The detector was pulsed flame photometric detector. Detector temperature, Hydrogen pressure and compress air were 320°C, 20psi and 35psi respectively.

## RESULTS

The amino acid profile in g/100g protein of the raw root, leaf and seed of *Moringa oleifera* are shown in **Table 1**. **Table 2**, shows the concentration of the Total, Essential, Non-essential, Acidic, Neutral, Sulfur, Basic, Aromatic amino acids (in g/100g protein) and Predicted Protein Efficiency ratio [22] of the *M. oleifera* root leaf and seed. The essential amino scores of the raw root, leaf and seed sample of *Moringa oleifera* based on the WHO reference pattern [17,18,19] are shown in **Table 3**.

**Table 1: The amino acids profile of the root seed and leaf of *Moringa oleifera* (in g/100g protein)**

Amino acids	Root	Seed	Leaf
Glycine	4.60	5.00	5.15
Alanine	3.36	3.23	3.43
Serine	3.61	4.25	4.20
Valine	3.03	3.09	3.36
Threonine	3.94	3.22	4.38
Isoleucine	1.84	4.35	2.33
Aspartate	6.01	6.14	6.86
Lysine	3.62	3.24	3.60
Glutamate	13.53	14.76	15.14
Methionine	0.76	0.97	0.95
Phenylalanine	3.98	4.53	4.26
Histidine	1.91	2.01	1.90
Arginine	1.74	8.06	1.88
Leucine	5.02	5.27	5.22
Tyrosine	2.43	2.33	2.20
Cysteine	2.42	2.02	2.05

**Table 2: The Concentration of Essential, Non-essential, Acidic, Neutral, Basic and Aromatic amino acids (g/100g protein) in raw root, leaf and seed of *M. oleifera***

Amino acid	Root	Leaf	Seed
Total Amino acid (TAA)	64.53	69.59	74.86
Total Non-Essential Amino acid (TNEAA)	38.70	41.72	40.12
% TNEAA	60.00	60.00	53.60
Total Essential Amino acid (TEAA)	25.83	27.87	34.73
% TEAA	40.00	40.00	46.40
Total Neutral Amino acid (TNAA)	17.85	19.49	20.93
% TNAA	27.70	28.00	28.00
Total Acidic Amino acid (TAAA)	19.54	22.00	20.91
% TAAA	30.30	31.60	27.90
Total Aromatic Amino acid (TArAA)	6.42	6.46	6.86
% TArAA	9.90	9.30	9.20
Total Sulfur Amino acid (TSAA)	3.18	3.00	2.99
% TSAA	4.90	4.30	4.00
Total Basic Amino acid (TBAA)	7.26	7.36	13.31
% TBAA	11.30	10.60	17.80
Predicted Protein Efficiency Ratio (P-PER)	1.56	1.68	1.67

**Table 3: Essential Amino Scores\* of the raw root, leaf and seed of *M. oleifera* based on the W.H.O Reference pattern**

Amino acid	RP**	Root (%)	Leaf (%)	Seed (%)
Lysine	5.17	69.93	69.66	62.71***
Histidine	1.77	107.78	106.76	113.53
Threonine	3.47	113.66	126.20	92.68
Valine	4.81	62.92	69.81	64.14
Methionine	1.53	49.48	62.09	63.72
Isoleucine	4.19	43.94***	55.59***	103.78
Leucine	7.03	71.40	74.30	74.99
Phenylalanine	3.01	131.21	141.41	150.51

\* Reference pattern in g/100g protein.

\*\* Amino acid scores = [g/100g of amino acid in sample ÷ (g/100g of amino acid in reference protein)] x 100.

\*\*\* Limiting amino acid.

## DISCUSSION

The amino acid profile in Table 1 showed that a total of 17 amino acids were found present in the seed, leaf and root of *M.oleifera* grown in southern Nigeria. This trend is similar to that reported by Anhwange et al[23] for *M. oleifera* grown in northern Nigeria. The amino acid Glutamate appeared to have the highest concentration in g/100g protein (14.76, 13.52 and 15.14) while Methionine had the least concentration (0.97, 0.76 and 0.95) in the seed, root and leaf of *M.oleifera* respectively. It is interesting to note that the amino acid Arginine was exceptionally high in the *M.oleifera* seed (8.06g/100g protein). This value is approximately equal to the value reported by Anhwange et al[23]. Since Arginine is essential for children and a reasonable level was present in the *M.oleifera* seed. The *M.oleifera* seed will thus serve as a good source of fortification of cereal weaning foods.

Among the essential amino acids, Leucine was the highest (5.02, 5.27, 5.22 g/100g protein) in the root, seed and leaf of *M.oleifera*. Leucine is responsible for regulating 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 [24]

Results in Table 2 showed that the total amino acid concentration in the seed (74.86g/100g protein) was higher when compared to those found in the leaf (69.59g/100g protein) and root (64.53g/100g protein) respectively. This indicates that *M. oleifera* seed is a better source of protein when compared to its leaf and root.

It was observed in the present study that the contents of total essential amino acid (TEAA) were (25.83, 27.87 and 34.73g/100g protein) in the root, leaf and seed respectively were approximately half of the value for egg reference protein (56.60 g/100g protein) reported by Paul et al [25] and their values compare favourably to some literature values (g/100g protein): 33.60 in *Anarcadium occidentale* [26]; 31.20 in *Parkia biglobosa* [2]; 37.60 - 51.80 in six different varieties of dehulled *Sphingostylis sternocarpa* flour [3]; 22.1 in endosperm of ripe coconut [27]; 45.2 in pigeon seed [28]; 53.4 in melon seed, 38.3 in pumpkin seed and 53.6 in gourd seed respectively [29]; 44.4 in Soyabean [30].

The Total Sulfur amino acid content (3.18,3.00 and 2.99 g/100g protein) in the root, leaf and seed of *M. oleifera* respectively were generally lower than the 5.8g/100g crude protein recommended for infants [18]. However, the Total Aromatic amino acid content (6.42,6.46 and 6.86 g/100g protein) in the root, leaf and seed respectively suggests them as an ideal infant protein as it is approximately equal to the recommended level 6.8g/100g protein [18]. The aromatic amino acids are precursors of epinephrine and thyroxine which play key hormonal roles in organisms [31].

It can also be observed that the percentage ratios of TEAA to TAA in the *M. oleifera* root (40.00), leaf (40.00) and seed (46.40) were higher than the 39% considered ideal protein food for infants, 26% required for children and 11% required for adults [18]. **Table 2** results further revealed that the Predicted Protein Efficiency Ratio of *M.oleifera* seed (1.67), leaf (1.68) and root (1.56) respectively were higher than the 1.21 in Cowpea, 1.62 in Millet ogi and 0.27 in Sorghum ogi [32] but lower than the 4.06 in modified corn ogi [32].

The Essential amino acid scores of the *M. oleifera* root, leaf and seed shown in **Table 3** revealed that these 3 morphological parts of *M.oleifera* had appreciable amino acid scores. Phenylalanine had the highest score in the *M. oleifera* seed, leaf and root while the limiting amino acids were Isoleucine (43.94%), Lysine (62.71%) and Isoleucine (55.59%) in the root, seed and leaf respectively. This implies that *M. oleifera* root, seed and leaf can serve as good sources of amino acid for man and livestock.

## CONCLUSION

In summary, this study established the fact that the *M. oleifera* root, seed and leaf protein are proteins of good quality and as such are suitable for animal feeds and human diet provided the toxicants present in them are removed. A further comprehensive study of the effect of processing on the protein quality is recommended as in most cases they are cooked before consumption.

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