

Comparative Amino Acid Compositions of *Uvaria chamae* Stem Bark and Poly Herbal Mixture

P. M. Aja^{1,*}, E. U. Ekpono¹, N. A. Obasi³, D. C. Obasi² and J. Nwaeke²

¹Department of Biochemistry, Faculty of Science, Ebonyi State University, Abakaliki, Nigeria e-mail: ovumte@yahoo.com; Patrick.aja@ebsu-edu.net

²Evangel University, Akaeze, Abakaliki, Ebonyi State, Nigeria

³Department of Medical Biochemistry, Faculty of Basic Medical Sciences, Alex Ekwueme Federal University, Ndufu-Alike, Ikwo, Nigeria

* Corresponding author

Abstract

The study evaluated comparative amino acids compositions of Uvaria chamae stem bark and Ruzu bitters. The amino acids compositions were determined using amino acid analyzer. The result of amino acid composition showed that eighteen amino acids were detected in both samples. Ten of the detected amino acids were essential amino acids and eight were non-essential. Glumatic acid had the highest concentration of 10.90 and 5.15 g/100g protein in Uvaria chamae stem bark and Ruzu bitters followed by aspartic acid with values of 8.40 and 3.44 g/100 g protein in Uvaria chamae stem bark and Ruzu bitters respectively. Leucine was the next amino acid in Uvaria chamae stem bark and Ruzu bitters followed by arginine. Uvaria chamae stem bark had the highest level of total amino acids of 72.66 g/100 g protein and Ruzu bitters had 32.17 g/100 g protein. For the EAA, it was 34.41 g/100 g for Uvaria chamae >17.44 g/100 g for Ruzu bitter. The highest essential amino acid (EAA) was leucine (6.13 and 3.56 g/100 g) in Uvaria chamae stem bark and Ruzu bitters. The total sulphur amino acid was generally low at 1.01-1.78 g/100 g but the % Cysteine in total sulphur amino acid (TSAA) was slightly high at 47.05% for Ruzu bitters but lower in Uvaria chamae stem bark (27.44%). The percentage coefficient variance (CV %) of the amino acid values were generally high with

Received: April 23, 2019; Accepted: May 20, 2019

Keywords and phrases: amino acid, Uvaria chamae, Ruzu bitters, comparative study.

Copyright © 2019 P. M. Aja et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

the exception of isoleucine, phenylalanine, lysine, methionine, leucine, cysteine and alanine with respective CV % values of 27.32, 31.97, 33.68, 37.50, 37.53, 38.81 and 39.05 while rest of CV % values ranged from 50.69-94.53 showing the gap of the amino acid values in the two samples to each other. The results of this study indicate that *Uvaria chamae* stem bark is richer in essential amino acid while % Cys/TSAA value is higher in Ruzu bitters.

Introduction

Medicinal plants, since times immemorial have been used in virtually all cultures as a source of medicine. Their enormous usefulness in the primary health care system cannot be over emphasized (Newman et al. [36]). Traditional medicine has a long history and has been defined as the sum total of the knowledge, skills and practices based on the theories, beliefs and experiences indigenous to different cultures, whether explicable or not, used in the maintenance of health, as well as in the prevention, diagnosis, improvement or treatment of physical and mental illnesses (WHO [24]). The World Health Organization notes however that inappropriate use of traditional medicines or practices can have negative or dangerous effects and that further research is needed to ascertain the efficacy and safety of several of the practices (WHO [24]). The widespread use of medicinal plant preparations obtained from commonly used traditional herbs and medicinal plants have been traced to the occurrence of natural products with medicinal properties (Hoareau and DaSilva [26]). There has been an increasing reliance on the use of medicinal plants in Western Societies, which has been traced to the extraction and development of several drugs from these plants as well as from traditionally used as herbal remedies (UNESCO [25]). Evidence of the therapeutic effects of medicinal plants is seen in their continuous use. It is estimated that about 25% of all modern drug prescription are directly or indirectly derived from plants (UNESCO [25]). Such drugs include: quinine, reserpine, ephedrine, ipecac and morphine that have been in widespread use for a long time, and more recently adopted compounds such as the anti-malarial artemisinin.

Medicinal plants or healing herbs are used in treating and preventing specific ailments and diseases and as such are considered to play a beneficial role in health care (Srivastava et al. [27]). The World Health Organization estimated that 80% of the populations of developing countries rely on traditional medicines, mostly plant drugs, for their primary health care needs (Srivastava et al. [27]). Medicinal plants represent a

consistent part of the natural biodiversity endowment of many Countries in Africa (Okigbo et al. [28]). Also, modern pharmacopoeia still contains at least 25% drugs derived from plants and many others which are synthetic analogues built on prototype compounds isolated from plants. Medicinal uses of plants range from administration of the roots, barks, stems, leaves, and seeds to the use of extract and decoction from plants (Ogbulie et al. [29]). The medicinal properties or values may be present in one of all the plants parts like roots, stem, back, leaves, flower, fruit or seeds. In fact, with all the progress in synthetic chemistry and biotechnology, plants are still indispensable source of drugs and natural products on the basis of their therapeutics (Ogbulie et al. [29]). Among all, plants like Kalancho epinnata, Kajanuskajan, Pterocarpus sanalinoides, Moringa lucida, Alstonia boonei, Azadirachta indica, Khaya grandifoliola and others have been scholarly proved effective in the treatment of malaria and other microbial infection (Turner [40]). In Nigeria, the local people are known for using natural herbs and herbal formulae for addressing various kinds of blood deficiencies. In south-eastern Nigeria, the roots of Uvaria chamae among others, are considered excellent natural herbal blood boosters, used especially for debilitating conditions, acute blood loss and blood deficiency diseases (Obadoni and Ochuko [30]).

Uvaria chamae belongs to the family of Annonaceaea. It is a small tree that grows to about 4.5m high. It is commonly found in the Savanna and rain forest region of Nigeria and other African countries. It is called "Mmimi ohia" in Igbo, "Kas kaifi" in Hausa and "Akisan" in Yoruba (Ogueke et al. [37]). It is a plant with both medicinal and nutritional values. It used as sedative, analgesic and cardio-protective and for the treatment of gonorrhea, catarrhal inflammations, amenorrhea and prevention of miscarriage among several other uses (Okwu and Josiah [32]). The fruits are yellow when ripe and have a sweet pulp which is widely eaten. The fruit carpels are in finger-like clusters. All parts of the plant are fragrant. The root barks, stem barks and leaves have a wide spread medicinal use. In Nigeria a decoction of the stem is used on the treatment of diarrhea (Igoli et al. [33]). Personal interaction with traditional medicinal practitioners indicated that they use the various parts of the plant in the treatment of cough, various stomach problems and urinary tract infections. They also apply the sap from the root, stem and leaf to wounds and sores for quick and proper healing. The use of this plant and its extracts in treatment of infections is very popular amongst the traditional medical practitioners of South Eastern Nigeria. Uvaria chamae commonly called clustered pear or bush banana (Nne-nwe) is a small tree whose parts (the leaves) are used as concoction

for treatment of malaria. A decoction of its roots, mixed with the roots of *Anthocleista djalonesis*, *Salacia nitida* and *Cnestis ferruginea* is used in the treatment of gonorrhea. *Uvaria chamae* is known to have cytotoxic activity (Philipov [34]).

Poly herbal mixture is a blend of several herbs. It is a herbal medicine consisting of three key ingredients namely *Uvaria chamae*, *Colocynthis citrullius* and *Curculigo pilosa*. It is use to manage and control various health related problems such as diabetes, high blood pressure, typhoid and malaria, fibroid, arthritis, gonorrhea, staphylococcus and infertility.

The amino acids found in nature occur either in free form or as linear chains in peptides and proteins. Analysis of amino acids plays a significant role in the study of the composition of proteins, foodstuffs and other materials of biological origin. In multicellular organisms, most of the proteins are based on L-amino acids that have a great influence in both human and animal nutrition, health maintenance, and possess potent therapeutic applications (Ambrogelly et al. [23]). Plants subjected to different environmental and physiological stresses can accumulate amino acids in their system that play pivotal role in combating stress. The amino acids produced in plant systems act as osmolyte, regulate ion transport, modulate stomata opening, activate phyto-hormones and growth substances, generate chelating effect on micronutrients and play a vital role in the detoxification of heavy metals (Zhao et al. [22]). They are also responsible for the synthesis and functional properties of specific enzymes, gene expression, and redox-homeostasis. Most importantly, in higher plants the amino acids are directly related to plant stress physiology and have diverse preventive and recovery effects.

Aim and objectives

The aim of this study was to determine and compare the amino acids profile of *Uvariaa chamae* stem bark and Ruzu bitters (a poly herbal mixture).

Materials and Methods

Materials

The equipment, chemicals and reagents used in this analysis were of analytical grade and quality.

Biological Material

The stem bark of Uvaria chamae and Polyherbal mixture were used for this study.

Collection of plant materials

Fresh stem barks of *Uvaria chamae* used in the study were collected from Umuka-Okposi Ohaozara L.G.A, Ebonyi state. This indigenous local plant was identified and authenticated by a taxonomist Professor S. C. Onyekwelu of the Department of Biological Sciences, Ebonyi State University, Abakaliki, Ebonyi State and the poly herbal mixture used in the study was purchased from Ruzu bitters head office in Abakiliki.

Methods

Preparation of plant materials

The fresh stem bark of the *Uvaria chamae* were cut into pieces and air-dried in the laboratory at room temperature for one week. The dried sample was pounded to certain texture (using pestle and mortar). The sample was further milled into fine powder using a mechanical grinder, sieved and stored in an air tight plastic container for analysis.

Determination of amino acid profile

The Amino Acid profile of the known sample was determined using the method described by Benitez [21]. The known sample was dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Applied Bio-systems PTH Amino Acid Analyzer.

Defatting sample

The sample was defatted using chloroform and methanol mixture of ratio 2:1. About 300mg of the sample was put in extraction thimble and extracted for 15 hours in soxhlet extraction apparatus (AOAC [20]).

Nitrogen determination

A small amount (0.115 mg) of ground sample was weighed, wrapped in whatman filter paper (No.1) and put in the Kjeldhal digestion flask. Concentrated sulphuric acid (10 ml) was added. Catalyst mixture (0.5 g) containing sodium sulphate (Na₂SO₄), copper sulphate (CuSO₄) and selenium oxide (SeO₂) in the ratio of 10:5:1 was added into the flask to facilitate digestion. Four pieces of anti-bumping granules were added. The

flask was then put in Kjeldhal digestion apparatus for 3 hours until the liquid turned light green. The digested sample was cooled and diluted with distilled water to 100ml in standard volumetric flask. Aliquot (10 m1) of the diluted solution with 10ml of 45% sodium hydroxide was put into the Markham distillation apparatus and distilled into 10ml of 2% boric acid containing 4 drops of bromocresol green/methyl red indicator until about 70 ml of distillate was collected.

The distillate was then titrated with standardize 0.01 N hydrochloric acid to grey coloured end point.

Percentage Nitrogen =
$$\frac{(a-b) \times 0.01 \times 14 \times V}{W \times C} \times 100$$

where:

a = Titre value of the digested sample

b = Titre value of blank sample

V =Volume after dilution (100ml)

W = Weight of dried sample (mg)

C = Aliquot of the sample used (10ml)

14 = Nitrogen constant in mg.

Hydrolysis of the sample

Thirty grams of the defatted sample was weighed into glass ampoule. Seven ml of 6N of Hydrochloric acid (HCl) was added and oxygen was expelled by passing nitrogen into the ampoule (this is to avoid possible oxidation of some amino acids during hydrolysis e.g. methionine and cystine). The glass ampoule was then sealed with Bunsen burner flame and put in an oven preset at $105^{\circ}C\pm5^{\circ}C$ for 22 hours. The ampoule was allowed to cool before broken open at the tip and the content was filtered to remove the humins. It should be noted that tryptophan is destroyed by 6N hydrochloric acid during hydrolysis.

The filtrate was then evaporated to dryness using rotary evaporator. The residue was dissolved with 5ml of acetate buffer (pH 2.0) and stored in plastic specimen bottles, which were kept in the freezer.

Loading of the hydrolysate into analyzer

The amount loaded was 60 micro litres. This was dispensed into the cartridge of the analyzer. The analyzer is designed to separate and analyze free acidic, neutral and basic amino acids of the hydrolysate.

Method of Calculating Amino Acid Values: An integrator attached to the Analyzer calculates the peak area proportional to the concentration of each of the amino acids.

Determination of tryptophan

The tryptophan in the known sample was hydrolyzed with 4.2 N Sodium hydroxide as described by (Spies [35]).

Defatting Sample

Thirty grams of the dried sample was weighed into extraction thimble and the fat was extracted with chloroform and methanol (2:1 mixture) using soxhlet extraction apparatus as described by AOAC [20] the extraction lasted for 15hrs.

Nitrogen determination

A small amount (200 mg) of ground sample was weighed, wrapped in whatman filter paper (No.1) and put in the Kjeldhal digestion flask. Concentrated suphuric acid (10ml) was added. Catalyst mixture (0.5) containing sodium sulphate (Na₂SO₄), cooper sulphate (CuSO₄) and selenium oxide (SeO₂) in the ration of 10:5:1 was added into the flask to facilitate digestion. Four pieces of anti-bumping granules were added. The flask was then put in Kjeldhal digestion apparatus for 3 hours until the liquid turned light green. The digested sample was cooled and diluted with distilled water to 100ml in standard volumetric flask. Aliquot (10 ml) of the diluted solution with 10ml of 45% sodium hydroxide was put into the Markham distillation apparatus and distilled into 10ml of 2% boric acid containing 4 drops of bromocresol green/methyl red indicator until about 70ml of distillate was collected. The distillate was then titrated with standardize 0.01N hydrochloric acid to grey coloured end point, the percentage nitrogen in the original sample was calculated using the formula:

Percentage Nitrogen =
$$\frac{(a-b) \times 0.01 \times 14 \times V}{W \times C} \times 100$$

where:

- a = Titre value of the digested sample
- b = Titre value of blank sample
- V = Volume after dilution (100ml)
- W = Weight of dried sample (mg)
- C = Aliquot of the sample used (10ml)
- 14 = Nitrogen constant in mg.

Hydrolysis of the sample

Thirty grams of the defatted sample was weighed into glass ampoule. Ten ml of 4.2 M NaOH was added and oxygen was expelled by passing nitrogen into the ampoule. The glass ampoule was then sealed with Bunsen burner flame and put in an oven preset at $105^{\circ}C \pm 5^{\circ}C$ for four hours. The ampoule was allowed to cool before broken open at the tip and the content was filtered to remove the humins. The filtrate was neutralized to pH 7.00 and evaporated to dryness at 40°C under vacuum in a rotary evaporator. The residue was dissolved with 5ml of borate buffer (pH 9.0) and store in plastic specimen bottles, which were kept in the freezer.

Loading of the hydrolysate into TSM analyzer

The amount loaded was 5 micro litres. This was dispended into the cartridge of the analyzer. The period of an analysis lasted for 76 minutes.

Method of calculating amino acid values

An integrator attached to the analyzer calculates the peak area proportional to the concentration of each of the amino acids.

Method of calculating amino acid values from the chromatogram peaks: The net height of each peak produced by the chart recorder of TSM (each representing an amino acid) was measured. The half-height of the peak on the half-height was accurately measured and recorded. Approximate area of each peak was then obtained by multiplying the height with the width of half-height.

Calculation of other protein quality parameters

Determination of the ratio of total essential amino acids (TEAA) to the total amino

acids (TAA), i.e. (TEAA/TAA), total sulphur amino acids (TSAA), percentage cystine in TSAA (% Cys/TSAA), total aromatic amino acids (TArAA), total neutral amino acids (TNAA), total acidic amino acids (TAAA) and total basic amino acids (TBAA) were estimated from the results obtained for amino acids profiles.

Results

Table 1 shows the amino acid (AA) composition for each sample. Glumatic acid had the highest concentration of 10.90 and 5.15 g/100 g protein in Uvaria chamae stem bark and poly herbal mixture respectively and it is an acidic amino acid. On the other hand, whilst aspartic acid was the second highest concentrated amino acid in Uvaria chamae stem bark and poly herbal mixture with the values of 8.40 and 3.44 g/100 g protein respectively, leucine was the third highest amino acid in Uvaria chamae stem bark and poly herbal mixture followed by arginine. Aspartic acid is a non-essential and acidic amino acid; leucine is an essential amino acid and is a neutral amino acid. The highest essential amino acid (EAA) was leucine (6.13 and 3.56 g/100 g) in Uvaria chamae stem bark and poly herbal mixture. The CV % of the amino acid values were generally high with the exception of isoleucine, phenylalanine, lysine, methionine, leucine, cysteine and alanine with respective CV % values of 27.32, 31.97, 33.68, 37.50, 37.53, 38.81 and 39.05 whilst rest of CV % values ranged from 50.69-94.53 showing the gap of the amino acid values in the two samples to each other. Table 2 and 3 show the concentrations of total AA (TAA), total essential AA (TEAA), total acidic AA (TAAA), total neutral AA (TNAA), total sulphur AA (TSAA), total aromatic AA (TArAA) and their percentage levels.

Amino Acids	Uvaria chamae	Polyherbal mixture	Mean	S.D	%CV
*Leucine	6.13	3.56	4.85	1.82	37.53
*Lysine	3.53	2.17	2.85	0.96	33.68
*Isoleucine	3.01	2.23	2.62	0.55	21.37
*Phenylalanine	4.79	3.02	3.91	1.25	31.97
*Trytophan	1.05	0.26	0.66	0.56	84.85

Table 1. Amino Acid Composition of *Uvaria chamae* stem bark of dry weight and poly herbal mixture in g/100 g protein.

*Valine	4.50	1.81	3.16	1.90	60.13
*Methionine	0.91	0.53	0.72	0.27	37.50
				••=•	
Proline	3.05	0.61	1.83	1.73	94.53
*Arginine	5.16	2.06	3.61	2.19	60.67
Tyrosine	2.92	1.03	1.98	1.34	67.68
*Histidine	2.11	0.80	1.46	0.93	63.70
Cystine	0.85	0.48	0.67	0.26	38.81
Alanine	4.02	2.28	3.15	1.23	39.05
Glutamic acid	10.90	5.15	8.03	4.07	50.69
Glycine	4.11	0.90	2.51	2.27	90.44
*Threonine	3.22	1.00	2.11	1.57	74.41
Serine	4.00	0.84	2.42	2.24	92.56
Aspartic acid	8.40	3.44	5.92	3.51	59.29

*= Essential Amino acid

Table 2. Concentrations of essential, non-essential, acidic, neutral, and sulphur, aromatic amino acid in g/100 g crude protein of *Uvaria chamae* stem bark in dry weight and poly herbal mixture.

Amino Acids	Uvaria chamae	Polyherbal Mixture	Mean	S.D	CV%
TAA	72.66	32.17	52.42	28.63	54.62
TEAA	34.41	17.44	25.93	9.61	37.06
TNEAA	38.25	14.73	26.49	14.82	55.95
TNAA	22.68	11.31	16.11	8.04	49.92
TAAA	19.30	8.59	13.95	7.57	54.27
TBAA	10.80	5.03	7.92	4.08	51.52
TSAA	1.76	1.01	1.39	0.53	38.13
TArAA	8.76	4.31	6.54	3.15	48.17
TAA/TEAA	2.11	1.85	2.29	0.28	12.23

Total amino acid (TAA), Total Essential amino acid (TEAA), Total Non-essential amino acid (TNAAA), Total sulphur amino acids (TSAA), Total aromatic amino acids (TArAA), Total neutral amino acids (TNAA), Total acidic amino acids (TAAA) and Total basic amino acids (TBAA)

Amino Acids	Uvaria chamae	Polyherbal mixture	Mean	S.D	CV%
%TEAA	47.36	54.21	44.03	5.35	12.15
%TNEAA	59.74	52.19	55.97	5.34	9.54
%TNAA	31.12	35.16	33.14	2.86	8.63
%TAAA	26.56	26.70	26.63	0.10	0.38
%TBAA	14.86	15.64	15.25	0.55	3.61
%TSAA	2.42	3.14	2.78	0.51	18.35
%TArAA	12.06	13.40	12.73	0.95	7.46
% Cysteine in TSAA	27.44	47.05	37.25	13.87	37.24

Table 3. Percentage concentrations of essential, non-essential, acidic, neutral, sulphur, aromatic and cysteine in total sulphur amino acid in g/100 g crude protein of *Uvaria chamae* stem bark of dry weight and polyherbal mixture.

%= percentage

Total amino acid (TAA), Total Essential amino acid (TEAA), Total Non-essential amino acid (TNAAA), Total sulphur amino acids (TSAA), Total aromatic amino acids (TArAA), Total neutral amino acids (TNAA), Total acidic amino acids (TAAA) and Total basic amino acids (TBAA)

Discussion

The result of this study showed that glumatic acid had the highest concentration of 10.90 and 5.15 g/100 g protein in *Uvaria chamae* stem bark and poly herbal mixture respectively and it is acidic amino acid (Table 1). On the other hand, while aspartic acid was the second highest concentrated amino acid in *Uvaria chamae* stem bark and poly herbal mixture with the values of 8.40 and 3.44 g/100 g protein respectively, leucine was the next amino acid in *Uvaria chamae* stem bark and Ruzu bitters followed by arginine (Table 1). Aspartic acid is a non-essential and acidic amino acid; leucine is an essential amino acid and is a neutral amino acid. The highest essential amino acid (EAA) was leucine (6.13 and 3.56 g/100 g) in *Uvaria chamae* stem bark and Ruzu bitters (Table 1). This is in line with the report of Olorunfemi et al. [18] which reported that *Moringa oleifera* leaves had the highest level of total amino acids (76.4 g/100 g) and followed by the root (70.9 g/100 g) while stem had 65.4 g/100 g as the lowest.

The result also supported the report of Omoyeni et al. [38] which revealed a high

value of glutamic acid, aspartic acid and leucine and low level of cystine, histidine, methionine and serine in dried leaves of *Melanthera scandens*. Adeveye et al. [41] also reported that the glutamate value of 10.38 g/100 gcp in P. mildbraedii was the same as that reported in S. indicum and B. aegyptiaca and also with the leaves of F. asperifolia and F. sycomorus which followed the same trend in both fermented and unfermented Cocoa nibs. Another study on Amino acid compositions of Luffa cylindrica seed (Oyetayo and Ojo [19]) showed that Luffa cylindrica seed contains a high proportion of essential amino, glutamic acid was the most abundant non- essential acid found while arginine was the most concentrated essential amino acid. Aremu et al. [1] made a similar observation in L. cylindrica seed kernel. Osibona et al. [2] reported that the most abundant amino acids in *Clarias gariepinus and Tilapia zillii* the two fish species were glutamic acid, aspartic acid, leucine and lysine ranging from 9.49 % to 18.16 %. Oyetayo and Ojo [19] reported that the total amino acid concentration was 72.71 g/100g protein and the total essential amino acid concentration was 38.76 g/100g protein in Luffa cylindrical seed flour. Arginine (9.75 g/100g protein) was the most concentrated essential amino acid while the Lysine/Arginine ratio was 0.52 in Luffa cylindrical seed flour (Oyetayo and Ojo [19]). Igwenyi et al. [3] reported that the concentration of arginine in Irvigna gabonesis and Citrullus colocynthis were 5.01 and 9.32 g/100g protein respectively. The concentration of histidine was 2.40 and 4.77 g/100g for Irvigna gabonesis and Citrullus colocynthis respectively (Igwenyi et al. [3]). The seeds were also high in non-essential amino acids such as cysteine in Irvigna gabonesis while glutamic acid and aspartic acid were high in both seeds. The common culture of combining soups made with these seeds will effectively compensate for deficiencies in the balance of nutrients especially, the limiting essential amino acids in both seeds. The result of this study was not in correlation with the report of Aja et al. [4] which reported that ethanolfruit extract of *Phoenix dactylifera fruit* (Dates fruit) sold in Abakaliki, Ebonyi State, Nigeria revealed that cysteine was not detected in the sample and nineteen (19) other amino acids were detected with proline (0.89%) and tryptophan (0.78%) as the major amino acids. Trimethysone (<0.1 %) was also detected in the sample.

Arginine (1.77-8.22 g/100g crude protein is essential for children and reasonable levels were present in the two samples: the lysine contents of the samples (2.17-3.53 g/100g cp) were about one half to the content of the reference egg protein (6.3 g/100g), and any of the samples will therefore serve as an average source for the amino acid. The study revealed that TEAA in *Uvaria chamae* stem bark was 34.41 g/100g protein which

is comparable to some literature values of nonconventional meat sources (g/100g protein): 35.1 for *Zonocerus variagatus* (Adeyeye [5]); 35.0 for *Macrotermes bellicossus* (Adeyeye [5]); 42.8 for *Limicolaria sp.*, 36.1 for *Archatina archatina*, 45.0 for *Archachatina marginata* (Adeyeye and Afolabi [6]); 38.6 for heart and 42.2 for liver of African giant pouch rat (*Cricetomys gambianus*) (Adeyeye and Aremu [7]) while that of poly herbal mixture was 17.44 g/100g protein. The contents of TSAA were generally lower than the 5.8 g/100 g cp recommended for infants (FAO/WHO [8]). The TArAA range suggested for ideal protein (6.8-11.8 g/100 g) FAO/WHO [8] has present value for *Uvaria chamae* slightly greater than the minimum and close to the maximum, i.e. 8.76 g/100 g cp. The TArAA are precursors of epinephrine and thyroxin (Robinson [9]).

The percentage ratios of TEAA to the TAA in the samples were 47.36 % (Uvaria chamae) and 54.211 % (poly herbal mixture) which were strongly comparable to that of egg (50 %) (FAO/WHO [16]), 43.6 % reported for pigeon pea flour (Oshodi et al. [10]), 43.8-44.4 % (beach pea protein isolate) (Chavan et al. [11]), 46.2 % (liver) and 46.3 % (heart) reported for African giant pouch rat (*Cricetomys gambianus*) (Adeyeye and Aremu [7]). The percentage ratios of TEAA to the TAA in the samples were well above the 39 % considered to be adequate for ideal protein food for infants, 26 % for children and 11 % for adults (FAO/WHO [8]). Most animal proteins are low in cystine (Cys) and hence in Cys in TSAA. For examples, (Cys/TSAA) % were 36.3 in *M. bellicosus* (Adeyeye [5]); 25.6 in *Z. Variegates* (Adeyeye [5]); 35.5 in *A. marginata*, 38.8 in *A. archatina* and 21.0 in *Limicolaria sp.*, respectively (Adeyeye and Afolabi [6]); 23.8-30.1 % in three fresh fish consumed in Nigeria (Adeyeye [12]) and 29.8 % in *Gymnarchus niloticus* (Trunk fish) (Adeyeye and Adamu [39]).

In contrast, many vegetable proteins contain substantially more Cys than Met, for examples, 62.9 % in coconut endosperm (Adeyeye [13] and Aremu et al. [1]); its range is 58.9-72.0 in guinea corn (Adeyeye et al. [14]); it is 50.5 % in cashew nut 43; it is 40.7 % in *Triticum durum* (Adeyeye [14]) and 44.4 % in *Parkia biglobosa* seeds (Adeyeye [15]). In the present study % (Cys/TSAA) values ranged from 27.44-47.05 which were much closer to the usual plant values. Thus, for animal protein, Cys is unlikely to contribute up to 50 % of the TSAA (FAO/WHO [16]). The % Cys/TSAA had been set at 50 % in rat, chick and pig diets (FAO/WHO [16]) but not in man. Cys can spare with Met in improving the protein quality and has positive effects on mineral absorption, particularly zinc (Mendoza [17]).

Conclusion

The study showed that *Uvaria chamae* stem bark had reasonable levels of total amino acid and essential amino acids greater than Ruzu bitters while % Cys/TSAA value is higher in poly herbal mixture than *Uvaria chamae*.

References

- M. O. Aremu, O. Olaofe, S. K. Basu, G. Abdulazeez and S. N. Acharya, Processed cranberry bean (*Phaseolus coccineus*) seed flours for African diet, *Canadian Journal of Plant Science* 90 (2010), 718-728. https://doi.org/10.4141/CJPS09149
- [2] A. O. Osibona, K. Kusemiju and G. R. Akande, Fatty acid composition and amino acid profile of two fresh water species, African catfish (*Clarias gariepinus*) and tilapia (*Tilapia zillii*), African Journal of Food, Agriculture, Nutrition and Development 9(1) (2009), 608-621. https://doi.org/10.4314/ajfand.v9i1.19216
- [3] I. O. Igwenyi, C. A. Eze, B. N. Azoro, C. E. Offor and C. P. Nwuke, Proximate, mineral and amino acid compositions of *Irvigna gabonesis* and *Citrullus colocynthis* used as soup thickener in South Easter Nigeria, *International Journal of Biotechnology and Biochemistry* 4(7) (2011), 493-499.
- [4] P. M. Aja, U. E. Uzuegbu, A. O. Opajobi, S. M. C. Udeh, E. U. Alum, M. C. Ominyi, C. Aloke and E. U. Ekpono, Amino acid profile, vitamin and reducing sugar compositions of ethanol fruit-extract of *Phoenix dactylifera* (date fruit) sold in Abakaliki, Ebonyi State, Nigeria, *International Journal of Biology, Pharmacy and Allied Sciences* 6(2) (2017), 349-362.
- [5] E. I. Adeyeye, Amino acid composition of variegated grasshopper (*Zonocerus variegates*), *Tropical Science* 45(4) (2005), 141-143. https://doi.org/10.1002/ts.9
- [6] E. I. Adeyeye and E. O. Afolabi, Amino acid composition of three different types of land snails consumed in Nigeria, *Food Chemistry* 85 (2004), 535-539. https://doi.org/10.1016/S0308-8146(03)00247-4
- [7] E. I. Adeyeye and M. O. Aremu, Amino acid composition of two fancy meats (liver and heart) of African gaint pouch rat (*Cricetomys gambianus*), *Oriental Journal of Chemistry* 27(4) (2011), 1409-1419.
- [8] FAO/WHO (1990). Protein quality evaluation. Report of Joint FAO/WHO Consultation Held in Bethesda, USA, 4-8 December, 1989. FAO, Rome, Italy.

- [9] D. E. Robinson, *Food Biochemistry and Nutrition Value*, London, UK: Longman Scientific and Technical, 1987.
- [10] A. A. Oshodi, O. Olaofe and G. M. Hall, Amino acid, fatty acid and mineral composition of pigeon pea (*Cajanus cajan*), *International Journal of Food Sciences and Nutritio* 43 (1993), 187-191. https://doi.org/10.3109/09637489309027541
- [11] U. D. Chavan, D. B. Mckenzie and F. Shaludi, Functional properties of protein isolates from beach pea (Lathynes maritimus L), *Food Chemistry* 74 (2001), 177-187. https://doi.org/10.1016/S0308-8146(01)00123-6
- [12] E. I. Adeyeye, Amino acid composition of three species of Nigerian fish: Clarias anguillaris, Oreochromis niloticus and Cynoglossus senegalensis, *Food Chemistry* 113(1) (2009), 43- 46. https://doi.org/10.1016/j.foodchem.2008.07.007
- [13] E. I. Adeyeye, The chemical composition of liquid and solid endosperm of ripe coconut, Oriental Journal of Chemistry 20(3) (2004), 471-476.
- [14] E. I. Adeyeye, Amino acids and sugar composition of Triticum durum whole meal flour, *Journal of Applied and Environmental Sciences* 3(2) (2007), 128-132.
- [15] E. I. Adeyeye, Amino acid composition of fermented African locust bean (Parkia biglobosa) seeds, *Journal of Applied and Environmental Sciences* 2(2) (2006), 154-158.
- [16] FAO/WHO, Protein quality evaluation, Report of Joint FAO/WHO Expert Consultation, FAO Food and Nutrition Paper 51, FAO, Rome, Italy, 1991.
- [17] C. Mendoza, Effect of genetically modified low phytic acid plants on mineral absorption, *International Journal of Food Sciences and Nutrition* 37 (2002), 759-767. https://doi.org/10.1046/j.1365-2621.2002.00624.x
- [18] O. Olorunfemi, E. I. Adeyeye and S. Ojugbo, Comparative study of proximate, amino acids and fatty acids of *Moringa oleifera* tree, *Elixir Applied Chemistry* 54 (2013), 12543-12554.
- [19] F. L. Oyetayo and B. A. Ojo, Food value and phytochemical composition of *Luffa cylindrica* seed flour, *American Journal of Biochemistry* 2(6) (2012), 98-103. https://doi.org/10.5923/j.ajb.20120206.02
- [20] AOAC (Association of Official Analytical Chemists), Official Method 18: (2006a), Fat content of raw and pasteurized whole milk, Official methods of analysis of the association of official analytical chemists (18th ed.) AOAC, Arlington, 2006.
- [21] L. V. Benitez, Amino acid and fatty acid profiles in aquaculture nutrition studies, In: S.S. De Silva (ed.), Fish Nutrition Research in Asia, Proceedings of the Third Asian Fish

Nutrition Network Meeting, Manila, Philippines: Asian Fisheries Society, 1989, pp. 23-35.

- [22] H. Zhao et al., Regulation of zinc homeostasis in yeast by binding of the ZAP1 transcriptional activator to zinc-responsive promoter elements, *J. Biol. Chem.* 273(44) (1998), 28713-20. https://doi.org/10.1074/jbc.273.44.28713
- [23] A. Ambrogelly, S. Palioura and D. Söll, Natural expansion of the genetic code, *Nature Chemistry Biology* 3(1) (2007), 29-35. https://doi.org/10.1038/nchembio847
- [24] WHO, Health Statistics, Geneva, Switzerland: World Health Organization, 2012.
- [25] UNESCO, FIT/504-RAF-48 Terminal Report: Promotion of Ethnobotany and the Sustainable Use of Plant Resources in Africa, Paris, 1998, p. 60.
- [26] Lucy Hoareau and Edgar J. DaSilva, Medicinal plants: a re-emerging health aid, *EJB Electronic Journal of Biotechnology* 2(2) (1999), 23-24. https://doi.org/10.2225/vol2-issue2-fulltext-2
- [27] A. K. Srivastava, R. Srivastava and V. Dixit, Pharmacological studies on fruits of Melia azedarach Linn, *Journal of Research in Ayurveda and Siddha* 2 (1981), 260-300.
- [28] R. N. Okigbo, U. E. Eme and S. Ogbogu, Biodiversity and conservation of medicinal and aromatic plants in Africa, *Biotechnology and Molecular Biology Reviews* 3(6) (2008), 127-134.
- [29] J. N. Ogbulie, C. C. Ogueke and F. C. Nwanebu, African Journal of Biotechnology 6(13) (2007), 1549-1553.
- [30] B. O. Obadoni and P. O. Ochuko, Phytochemical studies and comparative efficacy of the crude extracts of some haemostatic plants in Edo and Delta States of Nigeria, *Global Journal of Pure and Applied Sciences* 8 (2001), 203-208. https://doi.org/10.4314/gjpas.v8i2.16033
- [31] J. N. Ogbulie C. C. Ogueke and F. C. Nwanebu, Antibacterial properties of Uvaria chamae, Congronema latifolium, Garcinia kola, Vemonia amygdalina and Aframomium melegueta, *African Journal of Biotechnology* 6(13) (2007), 1549-1553.
- [32] D. E. Okwu and C. Josiah, Evaluation of the chemical composition of two Nigerian medicinal plants, *African Journal of Biotechnology* 5(4) (2006), 357-361.
- [33] J. O. Igoli, O. G. Ogaji, T. A. Tor-Anyiin and N. P. Igoli, Traditional medicine practice amongst the Igede People of Nigeria, Part II, *Afr. J. Trad. CAM* 2(2) (2005), 134-152. https://doi.org/10.4314/ajtcam.v2i2.31112

- [34] S. Philipov, N. Ivanovska, R. Istatkova, M. Velikova and P. Tuleva, Phytochemical study and cytotoxic activity of alkaloids from Uvaria chamae P. Beauv, *Pharmazie* 55 (2000), 688-689.
- [35] Joseph R. Spies, Determination of tryptophan in proteins, Anal. Chemistry 39(12) (1967), 1412-1416. https://doi.org/10.1021/ac60256a004
- [36] D. J. Newman, G. M. Cragg and K. M. Snader, The influence of natural products upon drug discovery, *Natural Product Report* 17 (2000), 215-234. https://doi.org/10.1039/a902202c
- [37] C. C. Ogueke, J. N. Ogbulie and F. C. Nwanebu, Antibacterial properties of Uvaria chamae, Congronema latifolium, Garcinia kola, Vemonia amygdalina and Aframomium melegueta, African Journal of Biotechnology 6(13) (2007), 1549-1553.
- [38] O. A. Omoyeni, E. Aterigbade, R. O. Akinyeye and R. A. Olowu, Phytochemical screening, nutritional/anti-nutritional and amino acid compositions of Nigeria *Melanthera scandens, Sci. Revs. Chem. Commun.* 2(1) (2012), 20-30.
- [39] E. I. Adeyeye and A. S. Adamu, Chemical composition and food properties of *Gymnarchus niloticus* (Trunk fish), *Biosciences, Biotechnology Research Asia* 3(2) (2005), 265-272.
- [40] G. Turner, Cerebral malaria, *Brain Pathology* 7 (1997), 569-582. https://doi.org/10.1111/j.1750-3639.1997.tb01075.x
- [41] E. I. Adeyeye, R. O. Akinyeye, I. Ogunlade, O. Olaofe and J. O. Boluwade, Effect of farm and industrial processing on the amino acid profile of cocoa beans, *Food Chem.* 118 (2010), 357-363. https://doi.org/10.1016/j.foodchem.2009.04.127