

Assessment of the Protein Quality of Some Edible Insects And Mollusks as Potential Food Sources

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Abstract

The amino acid profile of *Rhynchophorus phoenicis*, *Zonocerus variegatus*, *Helix pomatia* and *Littorina littoria* were determined using a Technicon sequential multi sample amino acid analyzer (TSM). The protein qualities of the samples were also estimated. The result of the study showed that *R. phoenicis* and *Z. variegatus* contain predominantly glutamic acid (13.71 and 12.96 g/100 g protein) and aspartic acid (10.22 and 10.53 g/100 g protein) respectively. The predominant amino acids in *H. pomatia* were arginine (12.25 g/100g) and lysine (12.03 g/100 g), while in *L. littoria*, it was glutamic acid (9.54 g/100 g protein). The total essential amino acid content of *L. littoria* was low (29.29 g/100 g) in comparison with 66.81, 54.70, and 50.38 g/100 g observed in *H. pomatia*, *R. phoenicis* and *Z. variegatus* respectively. The predicted protein efficiency ratio (P-PER) of the samples ranged from 2.12 in *L. littoria* to 3.58 in *H. pomatia*. The limiting amino acid for *R. phoenicis* and *Z. variegatus* were isoleucine and valine respectively, while for *H. pomatia* and *L. littoria*, it was methionine or cysteine and threonine respectively. The results revealed that the species of insects and mollusks studied contain nutritionally useful quantities of essential amino acids. Thus, they are very good sources of protein comparable to conventionally used protein foods.

Key words: *Rhynchophorus phoenicis*, *Zonocerus variegatus*, *Helix pomatia*, *Littorina littoria*, Amino acid.

1.0 Introduction

1.1 Protein Quality of Foods

Protein is one of the major nutrients required for growth and normal function of the body of a living organism. The quality of protein in a food source is determined by its amino acid make up and the bioavailability of these amino acids. Thus, the quality score of a protein is calculated by comparing the amino acids in the protein to human needs. The prime consideration in the evaluation of protein quality of different protein food sources is their essential amino acid composition and the efficiency with which the protein in the food can support growth also known as the protein efficiency ratio (Jessica *et al.*, 2011). The high cost of protein rich foods have led to the inability of people especially the poor to afford them. In many parts of the world, the poor has trouble obtaining protein rich food. Their menus often consisted of the local staple made up mainly of high carbohydrate diet and low quality protein foods. This is a fact of general interest known to have contributed to protein deficiency and malnutrition commonly seen in children in the developing world today (Lyons, 2000). Kwashiorkor is the disease that results when calories intake is adequate but protein is scanty. It is one of the consequences of protein malnutrition with symptoms such as fatty liver, edema, loss of skin color and deterioration of the digestive system.

Marasmus may also set in when protein malnutrition is coupled with lack of calorie intake. In this situation, muscles are wasted as they are degraded for protein synthesis or supply (Igwe *et al.*, 2011).

All over the world, acculturation has affected many human activities including types of food consumed. Some foods which include local delicacies like insects and some species of mollusk that maybe rich with high biologic value and protein efficiency ratio have long been neglected because of ignorance and the people associating their consumption with poverty. From pre-historical times to the era of cave man, human beings utilized insects and mollusks as ready sources of protein. This may be because they are cheap, cost effective and are affordable to the common man. Edible insects and mollusks have been reported to be important sources of high protein to rural dwellers and many city dwellers in Nigeria (Faboranti & Ajiboye, 2000). In Europe, Asia, Australia, America and Africa, people feed on different stages of various insects. Early record showed that the ancient Greek Pathians and Nasamines ate locust and grasshoppers as food; the North Americans, and Indians fed on rocky mountain locust, ants, grasshoppers, crickets and mollusks (Fish & Fish, 1989). The French also feed on snails and periwinkle which serve as treats for them. Because of the reported high protein content

of different species of insects and mollusks, their cost effectiveness and affordability, the present study was designed to assess the protein quality of African palm weevil larva, variegated grasshopper, edible land snail and periwinkle.

1.2 *Rhynchophorus phoenicis* (African palm weevil larva)

The African palm weevil larva (Figure 1A) is an edible worm that is traditionally called “Erurungwo” or “Nza” in eastern part of Nigeria. It belongs to the family of curculionidae, order coleoptera and is scientifically known as “*Rhynchophorus phoenicis*”. It is a species of snout beetle also known as sago palm weevil. The adults are large reddish brown beetles about 3 cm long, with a characteristic long curved rostrum and strong wings. The neonate larva is yellow-white in colour, segmented, legless and have a chitinous head capsule that is darker brown than the rest of the body. They have powerful horizontal conical jaws with which they burrow from the axils of the leaves to the crown where they feed voraciously. *R. phoenicis* is found in wide geographical areas spanning many different climates such as Africa, south Asia and southern American (Kalshoven & Laem, 2001). The weevil is attracted to dying or damaged parts of palms, cut or split palm trunks and can also attack undamaged palms as well as dying sugar cane. It is an important pest of palm especially the oil palm, *Elaeis guineensis*, and date palm (Gomez and Ferry, 1998). Its presence in the trunk of a tree is detectable by the noise made by the larva when feeding.

1.3 *Zonocerus variegatus* (variegated grasshopper)

Z. variegatus (variegated grasshopper; Figure 1B) are insects that belong to the family Pyrgomorphidae. They are found in sub-saharan African and generally in temperate regions where there is enough sunshine and rainfall. The variegated grasshopper has a yellow green body, 1.4-2.2 inches in length with yellow, orange, white and black markings. The wings are usually very short, covering only half of the abdomen but long winged individuals are also known. The larva is black with bright yellow sparkles. *Z. variegatus* feeds on a variety of plants especially relatives of peas, cassava leaves, garden egg, banana leaves, okro, etc. The larva feeds in group forming clusters of tens or hundreds of individual on a single plant. The fully winged adults are recalcitrant to fly, apparently relying on their bright warning colors to discourage most predators (Arneht, 2001; Gade,

2002).

1.4 *Helix pomatia* (Land Snail)

Snail is a common name which is applied to most members of the mollusk class, gastropod that have coiled shells in their adult stage. Gastropod, the largest and most diverse class of the phylum mollusk has about 75,100 known living species. The members of gastropod that lack shell are known as slugs (Ponder & Limdberg, 1997). Snails that respire (land snail) through lungs belong to the group *pulmonata* while those using gills (aquatic snail) form a paraphyletic group. As with all mollusks, snails are characterized by having a true coelom; a body divided into three parts of head, visceral mass and muscular foot; and organ system for circulation, respiration, digestion, excretion, nerve conduction and reproduction (Towel, 1989). Like other gastropod, their body plan involves torsion or trusting during larval development where by the visceral mass twists 180 degrees in relation to the head bearing the mantle cavity to the anterior of the animal. Thus the gills and renal openings are near the front of the animal. Land snails range greatly in size and are all hermaphrodites (Benami & Heller, 2005). Most snails are of herbivorous nature, though a few land species and many marine species may be oviparous or carnivorous. They break up their food using the radial, achitinous surface containing microscopic hooks called cuticle. With this the snail scrapes at food which is then transferred to the digestive tract. The land snail has a shell that is creamy to light brown often with indistinct brown colour bands. When retracted into their shell, some snails protect themselves with an anatomical structure called operculum which has a pleasant scent when burnt (Kearney *et al.*, 1998).

Helix pomatia (Figure 1C) is the most commonly seen and edible land snail found in Nigeria. It lives in the forest and open habitats, garden, vine yards, especially along rivers confined to calcareous substract. Representatives live in the garden, wood land, desert and on mountain, small ditches, banks of rivers and lakes, estuaries, mid floods, the rocky depths of the oceans and other ecological niches (Olgunoglu & Kennedy, 2005; Olgunoglu & Olgunoglu, 2009).

1.5 *Littorina littoria* (periwinkle)

The edible periwinkle (Figure 1D), *Littorina littoria* is a prosobranch gastropod of the family Littorinidae. It is one of the most common and largest shore gastropods of the Irish coast. Common periwinkles are native to the north eastern Atlantic Ocean, along the coasts of northern Spain, Scotland,

Ireland, Scandinavia and Russia. They can attain a length of approximately 35 cm. The head and tentacles of the animal are uniformly covered with dense transverse black lines. Sexes are separate and easily distinguished by the presence of penis on the right hand side of the male and a whitish ovipositor in the equivalent area of the female. But under certain conditions, the female may show abnormal development of a penis known as pseudohermaphroditism (Casey *et al.*, 1996). Mature shell height range from approximately 10.6-52.8 mm and is broadly ovate, thick, sharply pointed except when eroded. The shell contains 6-7 whorls with some fine threads and wrinkles. The shell lacks an umbilicus. The inside of the shell has a chocolate brown color (Reid, 1996).

1.6 Objectives of the Study

There is decreasing interest in the consumption of insects and mollusks because they have been labeled as food for poor people. This unacceptable belief is complicated by the decrease in availability and ease in accessibility of high protein containing foods to both rural and urban low and middle income families. Thus exposing such people to protein-deficiency associated diseases. The present study is therefore an attempt to assess the protein quality of *R.phoenicis*, *Z.variegatus*, *H. pomatia*, and *L. littoria*. It is hoped that this will contribute information to food composition table and revitalize interest of people towards consumption of these delicacies.



A



B



C



D

Figure 1: A. *Rhynchophorus phoenicis* larva; B. *Zonocerus variegatus* (variegated grasshopper); C. *Helix pomatia* (snail); D. *Littorina littoria* (periwinkle).

2.0 Materials And Method

2.1 Sample collection

Healthy larva of *Rhynchophorus phoenicis* were collected from a wine tapping terminal in Oba town, beside Oba River, Oba, Anambra State. Variegated grasshoppers were collected from a natural farm field in Eziobodo, Owerri, while the snails and periwinkles were purchased from the EkeukwuMarket, Owerri, Imo State. Twenty samples of each animal were collected. The species were specifically identified at the School of Agriculture and Agricultural Technology, Federal University of Technology, Owerri.

2.2 Sample processing

The palm weevil larva, de-winged grasshoppers and de-shelled snails and periwinkles were washed thoroughly with distilled water to remove sand particles and drained. They were each dried at 40°C in an oven to a constant weight and ground into coarse powder using a ceramic mortar. The samples were stored in labeled containers and their percentage moisture content determined as described by AOAC (1990).

2.3 Determination of Protein and Amino Acid Compositions

The nitrogen content of the ground samples was determined by the standard micro-Kjeldahl method, as described by Pearson (1976), and the percentage nitrogen obtained was converted to crude protein by multiplying with a factor of 6.25.

The total amino acid (AA) compositions of the ground samples were quantified using the ion-exchange chromatography-based Technicon Sequential Multi-sample (TSM)AA analyser (Technicon Instruments Corporation, New York) method described by Spackman *et al.*, (1958). Exactly 200 mg of each ground sample was first defatted thrice with equal volumes of chloroform/methanol mixture (1:1). Then, 30 mg of each defatted sample was put into a glass ampoule, 7 mL of 6 M HCl was added and oxygen expelled by passing nitrogen into the ampoule. The sealed ampoule was put in the oven, preset at 105 ± 5 °C for 22 h, allowed to cool, broken open at the tip and the content filtered to remove humus. The filtrate was then evaporated to dryness at 40°C under vacuum in a rotary evaporator. The residue was dissolved in 5 mL acetate buffer (pH 2.0) and 10 µL of each sample was dispensed into the cartridge of the TSM AA analyser and analysed. The amounts of the various AAs were calculated using the chromatogram generated as described by Spackman *et al.*, (1958) and expressed as g/100 g protein.

2.4 Estimation of protein quality parameters

The total amino acid profiles of the samples were estimated by calculating the Total Essential AA (TEAA), excluding Trp content; by summing the Arg, His, Ile, Leu, Lys, Met, Phe, Thr and Val contents; Total Non-Essential AA (TNEAA) by summing the Ala, Asp, Cys, Glu, Gly, Pro, Ser and Tyr contents; Total Neutral AA (TNA) by summing all the AAs excluding the TAAA, TBAA and Trp; Total Acidity AA (TAAA) by summing the Asp and Glu contents; Total Basic AA (TBAA) by summing the Arg, Lys and His contents; Total Sulphur-containing AA (TSAA) by summing up the Met and Cys contents; Total Branched-Chain AA (TBCAA) by summing the Leu, Ile and Val contents; Total Aromatic AA (TArAA), excluding Trp content, by summing the Phe and Tyr contents; percentage occurrences of these AA groupings were also calculated (Igwe *et al.*, 2012; Ibegbulem *et al.*, 2013). The Predicted Protein Efficiency Ratio (P-PER) was also determined using the equation of Alsmeyer *et al.*, (1974) i.e. P-PER = - 0.468 + 0.454 (Leu) - 0.105 (Tyr).

2.5 Estimation of total essential amino acid scores

The TEAA scores of the processed samples were calculated as the ratio of the actual amount (mg) of each EAA per g of the protein to the required amount (mg) of that EAA per g of a reference protein as described by FAO/WHO (1973) and Wardlaw and Kessel (2002) using the FAO/WHO/UNU (1981) provisional scoring pattern.

2.6 Statistical Analysis

All parameters determined were expressed in dry weight basis. The data generated were expressed as mean ± standard deviations and as simple percentages. The data were analysed using coefficient of variations, one-way analysis of variance and Tukey posthoc tests with the aid of a computer-based statistical package, Graph Pad Prism 5.3 (Graph Pad Software Inc. USA). Inferences were taken at confidence level of 95% (Sanders, 1990).

3.0 RESULTS AND DISCUSSION

3.1 Nitrogen and Protein Content of Samples

Table 1 presents the percentage nitrogen and total protein contents (g/100 g dry sample) of the insects; *R. phoenicis* and *Z. variegatus* as well as the mollusks; *H. pomatia* and *L. littoria* studied. It shows that *H. pomatia* (9.50 ± 0.18% and 59.37 ± 0.13 g/100 g) has appreciably more nitrogen and protein

TABLE 1: Percentage nitrogen and total protein (g/100 g dry sample) contents of the *R. phoenicis*, *Z. variegatus*, *H. pomatia* and *L. littoria*.

Parameter	Insects		Mollusks	
	<i>R. phoenicis</i>	<i>Z. variegatus</i>	<i>H. pomatia</i>	<i>L. littoria</i>
Nitrogen (%)	7.55 ± 0.04 ^a	8.49 ± 0.06 ^b	9.50 ± 0.03 ^c	8.81 ± 0.05 ^d
Protein (g/100g)	47.19 ± 0.15 ^a	53.06 ± 0.21 ^b	59.37 ± 0.13 ^c	55.06 ± 0.17 ^b

Values are mean ± standard deviations of triplicate determinations. Values bearing different alphabet letters per row are statistically significant (p<0.05).

TABLE 2: Amino acid compositions (g/100 g protein) of edible insects and edible mollusks

Amino acid	Insects		Mollusks	
	<i>R. phoenicis</i>	<i>Z. variegatus</i>	<i>H. pomatia</i>	<i>L. littoria</i>
Lysine ^a	7.41	6.34	12.03	6.12
Histidine ^a	5.26	4.51	3.23	1.57
Arginine ^a	8.17	8.00	12.25	3.91
Aspartic acid	10.22	10.53	3.86	5.92
Threonine ^a	4.99	4.83	6.88	1.78
Serine	4.30	5.02	1.30	1.24
Glutamic acid	13.71	12.96	9.69	9.54
Proline	3.40	3.40	0.65	0.85
Glycine	4.23	4.04	0.34	0.39
Alanine	4.56	3.94	3.17	4.02
Cystenine	1.32	1.19	0.53	0.53
Valine ^a	5.29	4.36	7.03	3.43
Methionine ^a	2.76	2.55	2.08	1.15
Isoleucine ^a	3.83	4.02	8.73	3.08
Leucine ^a	9.72	8.84	9.06	5.88
Tyrosine	3.54	3.83	0.64	0.81
Phenylalanine ^a	7.27	6.93	6.43	2.37

^a = Essential amino acid

TABLE 3: Classification of amino acid compositions (g/100 g protein) of *R. phoenicis* larva, *Z. variegatus*, *H. pomatia* and *L. littoria*

Groups	<i>R.phoenicis</i>	<i>Z.variegatus</i>	<i>H.pomatia</i>	<i>L.littoria</i>
Total Amino Acid (TAA)	98.98	95.29	86.99	52.59
Total Non-Essential Amino Acid (TNEAA)	45.35	44.91	20.18	23.30
% TNEAA	45.82	47.13	23.20	44.31
Total Essential Amino Acid (TEAA)With Histidine	54.70	50.38	66.81	29.29
TEAA-Without Histidine	49.44	45.87	64.49	27.72
%TEAA With Histidine	55.26	52.87	76.80	55.70
% TEAA Without Histidine	49.95	48.14	74.13	52.71
Essential Aliphatic Amino Acid	23.83	22.05	31.7	14.17
%EAAA	23.83	23.14	36.44	26.94
Essential Aromatic Amino Acid	7.27	6.93	6.43	2.37
%EA _R AA	7.34	7.27	7.39	4.51
Total Neutral Amino Acid (TNAA)	55.28	52.95	46.84	25.53
% TNAA	55.28	55.57	53.85	48.55
Total Acidic Amino Acid (TAAA)	23.93	23.49	13.55	15.48
% TAAA	23.93	24.65	15.58	29.40
Total Basic Amino Acid (TBAA)	20.84	18.85	26.60	11.6
% TBAA	20.84	19.78	30.58	22.06
Total Sulphur Amino Acid TSAA	4.08	3.74	2.61	1.68
%TSAA	4.12	3.92	3.00	3.19
%Cysteine in TSAA	32.35	31.82	20.31	31.55
Total Hydroxyl Amino Acid (THAA)	9.36	9.85	8.18	3.02
%THAA	9.46	10.34	9.40	5.74
Predicated Protein Efficiency Ratio (P-PER)	3.57	3.14	3.58	2.12

TABLE 4: Amino acid Scores (AAS) of *R. phoenicis*, *Z. variegatus*, *H. pomatia* and *L. littoria*

Amino Acid	PAAESP (g/100 g protein)	Amino Acid Scores (AAS)			
		<i>R. phoenicis</i>	<i>Z. variegatus</i>	<i>H. pomatia</i>	<i>L. littoria</i>
Ile	4.0	0.96	1.01	2.18	0.77
Leu	7.0	1.39	1.26	1.29	0.84
Lys	5.5	1.35	1.15	2.19	1.11
Met+Cys	3.5	1.16	1.07	0.75	0.48
Phe+Tyr	6.0	1.80	1.80	1.18	0.53
Thr	4.0	1.25	1.21	1.72	0.45
Val	5.0	1.06	0.87	1.41	0.69
Total	35.0	8.97	8.37	10.72	4.87

PAAESP=Provisional Amino Acid (Egg) Scoring Pattern.

contents than *L. littoria* (8.81 ± 0.14 % and 55.06 ± 0.17 g/100 g) and *Z. variegatus* (8.49 ± 0.16 % and 53.06 ± 0.18 g/100 g), with *R. phoenicis* having the least contents at 7.55 ± 0.14 % and 47.19 ± 0.15 g/100 g respectively. Although, both animal groups showed appreciable protein contents, the mollusks have more crude protein content than the insects. Previous studies have also shown appreciable but varied amounts of protein content in these animals; *H. pomatia* (12.87 ± 0.13 %), *L. littoria* (22.47 ± 1.12 %), *Z. variegatus* (26.8 %) and *R. phoenicis* (31.4 %) (Ozogul *et al.*, 2005; Bayo *et al.*, 2006). Results of the present study show that the protein contents of *H. pomatia*, *L. littoria*, *R. phoenicis* and *Z. variegatus* are comparable to the conventionally used protein supplements such as soybean meal (48 % crude protein) and fish meal (50-55 % crude protein) (Anand *et al.*, 2008) by having a high protein content range of 47.19 – 59.37 g/100 g. Interestingly, the insects studied, *R. phoenicis* and *Z. variegatus* have approximately similar or even more protein content when compared with the other edible insects such as *Tenebriomolitor* (60.2 %) (Ramous-Elorduy *et al.*, 2002) and *Macrotermes nigeriensis* (20.94 %) (Igwe *et al.*, 2011).

3.2 Amino Acid Composition of Samples

The concentration of amino acids in the insects (*R. phoenicis* and *Z. variegatus*) shows that glutamic and aspartic acids were highest in content at values of 13.71 and 10.22 g/100 g protein in *R. phoenicis* and 12.96 and 10.53 g/100 g protein in *Z. variegatus* respectively (Table 2). Cysteine had the lowest value in both insects. The essential amino acids with high values included leucine, phenylalanine and arginine in these insects. Table 2 also presents the amino acid concentrations of the mollusks; *H. pomatia* and *L. littoria*. Lysine, leucine and arginine were found to be the highest essential amino acids (EAA) in the land snail, followed by glutamic acid, leucine, isoleucine, valine, threonine and phenylalanine. This is in agreement with the report of Olgunoglu and Olgunoglu, (2009) which showed that lysine, leucine and arginine constituted the highest essential amino acids (EAA) in garden and land snails. For *Littorina littoria* glutamic acid recorded the highest value, with lysine, leucine and arginine being the highest essential amino acids (EAA) with values of 6.12, 5.88 and 3.91 g/100 g protein respectively. On the other hand, cysteine, glycine, tyrosine and proline recorded lowest values in both mollusk species.

Cereal proteins that are key staples in diets around

the world are often low in lysine and, in some cases, lack the amino acids tryptophan (e.g. maize) and threonine, which are important essential amino acids. In some insect species, these amino acids are very well represented. The results of the present study shows that the following essential amino acids lysine, histidine, arginine, threonine, valine, leucine and phenylalanine are also very well represented in the insects and mollusks studied. The distribution of amino acid contents shows that there were more essential amino acids in the *H. pomatia* and the insects than in *L. littoria*.

3.3 Classifications of the Amino Acid Composition of Samples

The classification of amino acid composition in Table 3 revealed that the TEAA, TNEAA, TAAA and TBAA in *R. phoenicis* and *Z. variegatus* gave values of 54.70, 43.35, 23.93 and 20.84 g/100 g and 50.38, 44.91, 23.49 and 18.85 g/100 g respectively, while those of *H. pomatia* and *L. littoria* gave values of 23.20, 20.18, 13.55 and 26.60 g/100 g and 44.31, 23.30, 15.48 and 11.6 g/100 g respectively.

The total essential amino acid (TEAA) with histidine for *Z. variegatus* and *R. phoenicis* gave values of 50.38 and 54.70 g/100 g protein respectively which correspond to 53 % for *Z. variegatus* and 55 % for *R. phoenicis* larva. In the same vein, the total essential amino acid content (with histidine) for *H. pomatia* and *L. littoria* gave a value of 66.81 g/100 g protein for *H. pomatia* and 29.29 g/100 g protein for *L. littoria* corresponding to 76.80 % and 56.89 % respectively. This is comparable with the values obtained for *H. pomatia* by Kunle *et al.*, (2001) where TEAA with histidine was reported to be 67.1 g/100 g protein corresponding to 74.6% of the total amino acid content. Similarly, their total EAA contents were higher than the 1.7 g EAA content of egg white and 26.14 – 28.68 g/100 g protein of local wines made from *Elaeisguineensis* and *Raphiahookeris* reported by Connolly (2011) and Ibegbulem *et al.*, (2013) respectively. This suggests that their amino acids may adequately support protein synthesis in humans. For essential aliphatic amino acid (EAAA), *R. phoenicis* larva had a value of 23.83 g/100 g protein which is a little higher than that of *Z. variegatus*, where it recorded a value of 22.05 g/100 g protein. For *H. pomatia* and *L. littoria*, the EAAA values were 31.7 and 14.17 g/100 g protein respectively.

The acidic amino acid contents were 23.93 % for *R.*

phoenicis larva and 24.65 % for *Z. variegatus*. These were found to be greater than the contents of basic amino acids (TBAA) which gave values of 20.84 % for *R. phoenicis* larva and 19.78 % for *Z. variegatus*. This indicates that the amino acid content of proteins of *R. phoenicis* larva and *Z. variegatus* are more acidic in nature. On the other hand, the acidic amino acid content of *H. pomatia* was found to be lower than its total basic amino acid content. For *Littorina littoria*, the total acidic amino acid content was 29.39 % while the total basic amino acid value was 22.06 % which indicates that the amino acid contents of its protein are mainly acidic in nature.

The values of the total sulphur amino acid (TSAA) with cysteine showed *R. phoenicis* to be higher in value at 32.35 %, than *Z. variegatus* which was slightly lower at a value of 31.82 %. Similarly, the total sulphur amino acid (TSAA) content with cysteine showed *L. littoria* to be higher in value at 31.54 %, than *H. pomatia* which was lower (20.31 %). This is in agreement with reports of Jeffson *et al.*, (1999) which showed the TSAA for *L. littoria* to be 30.04 %, indicating that *L. littoria* might be a good source of TSAA.

The % Cysteine/TSAA of the studied animals ranged from 20.31 % in *H. pomatia* to 32.35 % in *R. phoenicis*, which are close to those observed earlier in *Macrotermesbellicosus* (36.3 %) (Adeyeye & Afolabi, 2004), *Archatinaarchatina* (38.8%) and *Archachatina marginata* (35.5 %) (Belavady *et al.*, 1963). However, our results of % Cys/TSAA failed to corroborate the assertion that Cysteine supplies up to one-third of the need for TSAA (Alsmeyer *et al.*, 1974). The % Cys contents of the samples were however lower than the 37.8 % reported for *Garcinia kola*, the 50.5% reported for *Anacardium occidentale* (Adeyeye *et al.*, 2007) and the 67.09 % reported for raw *Prosopisafriicana* by Igwe *et al.* (2012). Cysteine in animal proteins does not contribute more than 50 % of the TSAA. The % Cys in the studied samples were lower than the expected value of 50 % for animal sources. Cys is one of the AAs required for the synthesis of glutathione, a redox buffer. Nelson and Cox (2008) reported that glutathione maintains the sulphhydryl groups of proteins in the reduced state, the iron of haem in the ferrous (Fe²⁺) state and also acts as a reducing agent for glutaredoxin in deoxyribonucleotide synthesis. Under aerobic conditions the redox function of glutathione is also used to remove toxic peroxides formed in the normal course of growth and metabolism. Harris and Crabb (2006) reported that the synthesis of glutathione is limited by the

availability of Cys. It therefore follows that consumption of these insects and mollusks may contribute to the Cys pool of tissues and organs. The predicted protein efficiency ratios (P-PER) for *R. phoenicis* and *Z. variegatus* were 3.57 and 3.14 respectively while in *H. pomatia* and *L. littoria*, the values were 3.58 and 2.12 respectively. Muller and Tobin (1980) reported that experimentally determined protein efficiency ratio (PER) usually ranged from 0.00 for a very poor protein to a maximum of just over 4. The results show that *H. pomatia* and the insects have higher and possibly more efficient or available protein than *L. littoria* and support the earlier observed lower content of essential and non-essential amino acids in *L. littoria* than in *H. pomatia* and the insects studied. However, P-PER results of the insects and mollusks compared favourably with those of processed common legumes (2.15 – 2.95), and male fresh water crab (2.70) (Igwe *et al.*, 2012).

The Leu/Ile ratios and % Leu-Ile values for *R. phoenicis* and *Z. variegatus* were 2.54 and 5.89, and 2.20 and 5.06 respectively while in *H. pomatia* and *L. littoria*, the values were 1.04 and 0.38, and 1.91 and 5.32 respectively. The results show that the samples contained more Leu than Ile. High Leu content in diet impairs tryptophan and niacin metabolism and might be a factor in pellagra development (FAO, 1995). However, it has been suggested that Leu/Ile balance is a more important factor than dietary excess of Leu alone in regulating the metabolism of Trp and niacin, and hence the disease process (Adeyeye, 2008). Furthermore, it has been reported that animals fed sorghum proteins containing less than 110 mg/gcp of Leu did not suffer from nicotinic acid deficiency (Igwe *et al.*, 2012). The present results show that *H. pomatia* and *L. littoria* have less ratio values of Leu/Ile than *R. phoenicis* and *Z. variegatus* indicating better protein quality. But it is worthy of note that these insects are only eaten as treats or supplements to normal animal diets and thus may not pose danger with respect to impairment of tryptophan and niacin metabolism. Meanwhile, the branched-chain amino acids (BCAAs) generally include leucine, valine, and isoleucine and are popular as dietary supplements in strength-training athletes. In particular, leucine has been implicated to improve athletic performance. However, a significant increase in blood ammonia

concentrations above normal values, plasma leucine concentrations, and urinary leucine excretion were observed with leucine intakes $>500 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$ calling for caution in the intake of high leucine containing diets (Elango *et al.*, 2012).

3.4 Essential Amino Acid Scores of Samples

Table 4 shows the essential amino acid scores of the edible insects and mollusks studied. It revealed that isoleucine is the limiting amino acid in *R. phoenicis* falling short of meeting the body requirement by only 4% if the insect is taken as the only source of diet protein. Similarly, valine and methionine plus cysteine are the first limiting amino acids in *Z. variegatus* and *H. pomatia* at deficiency percentages of 13 and 25 respectively. The first and second limiting amino acids in *L. littoria* are threonine and methionine plus cysteine. The EAAs in *L. littoria*, excluding lysine, fell short of meeting human body requirement by a range of 16 – 55 %. These observations further buttresses the earlier discussed lower quality of protein in *L. littoria* in comparison with *H. pomatia*.

4.0 Conclusion and Recommendation

In conclusion, the insects and mollusks studied are apparently good sources of protein supplements to meals and contain both essential and non-essential amino acids in quantities comparable with some animal proteins and other common legumes. Thus, they can serve well in complementing weaning food for children in the tropics where protein malnutrition is one of the major causes of child morbidity and mortality. Furthermore, the study has shown that these insects and mollusks, which are currently consumed largely by the rural populace is not inferior to conventional popular protein sources. Thus there is need to make these delicacies available in the urban and rural communities all year round. This will, not only help to counter the effect of protein deficiency diseases but would make these foods available to all and also create sources of income for rural dwellers that will be sourcing and marketing them. Government may assist them with finance, technological know-how and information dissemination via enlightenment programmes to ensure consumer awareness of the nutritional potentials these currently neglected protein foods.

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