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Fatty Acid and Amino Acid Profile of Emperor Moth Caterpillar (*Cirina forda*) in Paikoro Local Government Area of Niger State, Nigeria

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Abstract The proximate, amino acid, fatty acid and mineral compositions of emperor moth caterpillar (*Cirina forda*) were investigated. A high protein and lipid contents of 31.40 ± 0.3 and 16.12 ± 1.1 (% dry weight) respectively were recorded. The insect was found to contain varying proportions of essential and non-essential amino acids and also saturated and unsaturated fatty acids. An ash content of 7.62 ± 0.11 (% dry weight) was obtained containing high proportions of calcium, sodium, potassium, magnesium and phosphorus (32.24, 45.50, 65.04, 67.31, and 111.00 mg/100g) respectively. Moderate amounts of iron, manganese and zinc were also detected. A K/Na ratio of 1.53 ± 0.04 obtained suggests that this insect could be a potential source of diet for the management of hypertension. The high protein and carbohydrate contents also suggest that the emperor moth caterpillar could serve as an alternative source of protein and other nutrient supplements in human and animal diet.

Keywords Emperor Moth Caterpillar, Fatty Acid, Amino Acid, Proximate Composition, Mineral Composition

1. Introduction

Insects are widely spread all over the world; many of them are well known and appreciated for their characteristics [1]. Entomophagy (the habit of eating insects as food) is a well established custom in many parts of the world [2], and many edible insects have become a food resource that is being tapped by various communities. In Europe, Asia, Australia and Africa, people feed on different stages of various insects. Early records by Ene [3], showed that the ancient Greek-Parthians and Nasamines ate locust and grasshoppers as food. The French feed on the abdomen of the beetle *Rhizotrogus assimilis*. In Nigeria several species of insects are prominent items of commerce in the village markets and many families make fairly good living from selling insects [4].This has contributed significantly to the reduction of protein deficiencies [5]. The caterpillar of the pallid emperor moth *(Cirina forda)* is of the order, *Lepidoptera* and family, *Saturniidae*. It is an insect pest of *Butyrospermun paradoxum*, the shearbutter tree and is widely accepted as a food source and is also an important item of commerce in many Nigerian states such as Oyo, Kwara, Kogi, Niger, Kaduna and Benue [5]. The larvae of this insect are processed into the dried form and consumed as a delicacy served as snacks or as an essential ingredient in vegetable soups along with carbohydrate food in Southern Nigeria and many homes in Africa [6, 7, 8]. The nutritional qualities of edible insects have been studied by various authors including Mbata and Chidumayo [9], and Ogbadu [10]. They have been found to contain protein and abundant fat calories. Ande [11] recorded high protein content and 21.45% ether proportion in *C. forda* but the quality was not reported. He concluded that they may be a good nutritional source to man. In Niger State, only a few of these edible insects have been analyzed for their nutritional content. The quality of the nutrients in most of the popularly consumed insects is fragmentary and inadequate. Lack of such information is standing in the way of the utilization of these relatively rich protein and lipid sources for the dietary requirements of man.

This study therefore attempts to evaluate the proximate composition, mineral content, the fatty acid and amino acid profiles of emperor moth caterpillar from Paikoro Local Government, Niger State, and to compare these values with reports on other plant and animal protein and lipid sources. This will provide a baseline data and a reference point for the study of nutritional potentialities of other edible insects which will encourage greater exploitation to meet other malnutrition needs in the rural communities.

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2. Materials and Methods

2.1. Sample Collection, Preparation and Analysis

Fresh samples of the emperor moth caterpillar (*Cirina forda*) were handpicked from the crowns of Shear butter trees in ten different farmlands between the months of May to August, 2010 in Tutungo Village, Paikoro Local Government, Niger State. The samples were washed and sun-dried for 72 hours, milled into powder ready for analysis.

2.2. Proximate Analysis

The moisture, ash and crude fiber contents were determined by the methods of Association of Official Analytical Chemists (AOAC) [12]. Total nitrogen (%N) was determined by the micro- Kjeldahl method [12], and crude protein was obtained by using a Nitrogen Protein conversion factor of 6.25. The oil (lipid %) content was obtained by extracting 5 g of the powdered sample with chloroform: methanol (2: 1) mixture as described by the modified Folch method [13]. The solvent was evaporated on a rotary evaporator and the oil obtained was dried in an oven at 60 ^{o}C to remove all traces of solvent. Percentage Nitrogen Free Extract (Carbohydrate) was obtained by difference applying the formula:

100 % - (% protein + % lipid + % ash + % fibre)

All determinations were in triplicate.

2.3. Mineral, Fatty acid and Amino Acid Analyses

Sodium and Potassium were determined using Gallenkamp flame analyzer, while Calcium, Magnesium, Iron, Manganese, Zinc and Copper were determined using Buch 211 Atomic Absorption Spectrophotometer. Phosphorus content was determined using the phosphovanado molybdate colorimetric method and the results read on JENWAY 6100 spectrophotometer. The oil in the sample was extracted using chloroform: methanol (2:1) mixture, as described by modified method by Folch [13]. The extracted oil was hydrolysed and the fatty acids converted to their fatty acid methyl derivatives and their concentrations determined using GC/MS. The defatted samples were hydrolysed and after derivatization, the amino acid profile was obtained using WATERS 1525 binary HPLC coupled with multiwavelength fluorescence detector. Duplicate determinations were carried out in each case.

Table 1. Proximate composition (%) of Emperor moth caterpillar

Parameter	*Composition
Ash	7.62 ± 0.11
Moisture	5.40 ± 0.01
Crude protein	31.40 ± 0.30
Crude fat	16.12 ± 1.10
Crude fiber	10.80 ± 0.10
Carbohydrate	28.66 ± 0.01
Energy (kJ/ 100g)	1617.46

*means ± SD of three (3) determinations

3. Results and Discussion

The result of the proximate chemical composition of emperor moth caterpillar is presented in Table 1.

The moisture content of *C. forda* is 5.40 ± 0.01%. Subhachae, *et al.* [14] reported higher moisture content of 8.6 ± 0.1 and 12.8 ± 0.1% for two varieties of edible ants. Omotoso [8], recorded a slightly higher value of 10.85 ± 0.38 % moisture for emperor moth caterpillar. Low moisture content indicates a good shelf life characteristic; the low moisture content of the insect shows that it can be stored for a long period without deterioration. The ash content of 7.62 ± 0.11% was obtained in moth caterpillar. Akinnawo and Ketiku [15] and Omotoso [8] reported ash content of 7.12 ± 0.32 % for larva of *Cirina forda* which is close to the value of 7.62 ± 0.11% of the larva of moth caterpillar obtained in this work. The quantity of ash in a sample supposedly indicates the amount of minerals. The relatively high ash content in this work shows that the insect could be a good source of minerals. The crude protein content obtained is 31.40 ± 0.3 %. Ekpo, *et al.* [16] obtained value of 33.41 ± 0.20 for *Macrotermis bellicosus.* Subhachae *et al.* [14] documented a value of 31.5 ± 0.5% as protein content of sun-dried edible black ants cultivated in China. Akinnawo and Ketiku [15] reported a protein content of 33.12 ± 0.08 % dry matter for *Cirina forda*. Crude protein contents as high as 30 % have been reported for termites [17]. These values are similar to the ones obtained for moth caterpillar in this work. The reported protein values of beef meat (22.6%), chicken (18.83%) as well as fish which range from 15.55% in cat fish to 23.33% in tuna fish by Onimawo and Egbekun [18] are in agreement with the protein values in this work. The crude protein content from the larva of moth caterpillar falls within the protein range of 15 – 60 % previously reported for various forms of *Lepidopterous* edible insects [19]. Since the insect have high yield of crude protein, they could serve as good protein source contributing significantly to the recommended human daily protein requirement of 23 – 56% stipulated by National Research Council (NRC) [20], for man and animals, particularly in developing countries where the cost of conventional protein sources are expensive. The crude fibre content (10.80 ± 0.18%) was found in moth caterpillar This value is close to crude fibre content of 9.40 ± 0.16% reported by Akinnawo and Ketiku [15] for moth caterpillar. Mbah and Elekima [21] documented higher fibre content of 13.12 ± 0.15 and 14.13 ± 0.20% in *Periplaneta americana* and *Componotus sp.* High crude fibre in food is known to promote digestibility and enhance health benefits such as reduction of the risk of gastrointestinal cancers. The high value of crude fibre from moth caterpillar indicates that, they could be used to complement animal roughages in addition to other uses. The crude fibre content in this insect could be due to chitin found normally in insects which can reduce serum cholesterol and serve as a haemostatic agent for tissue repairs [22]. Carbohydrate content of 28.66 ± 1.03 % was found in moth caterpillar. Mbah and Elekima [21] obtained value of 20.01 ± 0.10% for *Zonocerus variegatus*, result which is similar to that obtained in this work. Subhachai, *et al.* [14] found carbohydrate content of 12.4 ± 1.1% in sun-dried edible ants. Ife and Emeruwa [17], reported carbohydrate content of 15.05% in the larva of *Oryctes monoceros.* These differences are not unusual due to differences in species and environment. The high carbohydrate content found in this insect could serve as an energy source to man or animals when compared with other carbohydrate sources.

The metabolisable energy calculated for this insect is 1617 kJ/100 g. Teffo, *et al.* [23] obtained energy value of 2600 kJ/100 g for the larva of the edible Stink-bug which is higher than that in this study. The high value suggests that it could be a good energy source. The result of the mineral profile of pallid moth caterpillar is shown in Table 2.

Table 2. Mineral composition of Emperor moth caterpillar (*C. forda*)

Mineral	*Concentration
Sodium	45.50 ± 0.25
Potassium	65.04 ± 2.02
Calcium	32.24 ± 0.06
Magnesium	67.31 ± 0.21
Iron	12.85 ± 0.10
Manganese	7.53 ± 1.15
Zinc	3.71 ± 0.21
Phosphorus	111.0 ± 0.40
K/Na ratio	1.53 ± 0.04
Ca/P ratio	0.29 ± 0.01

*means (mg/ 100g dry weight) ± SD of three determinations

The ash content of a sample is a reflection of the minerals it contains. It is therefore not surprising that the emperor moth caterpillar is rich in minerals as earlier shown by the high ash content in Table 1.

Sodium concentration of 42.50 ± 0.25 mg/100g (dry weight) was found in moth caterpillar. The values of 45.26± 0.01 for Sodium by Omotoso [8] for *Cirina forda* are quite similar with values of 42.50 ± 0.25 mg/100g obtained in this work. The RDA for sodium is 1500 mg for an adult. The amount of sodium from this insect is encouraging because sodium assists in maintaining the proper acid-balance and in controlling osmotic pressure that develops between the blood and cells due to ionic concentration differences. Potassium is an essential mineral nutrient and plays an important role in the synthesis of amino acids and proteins. Moth caterpillar has potassium content of 65.04 ± 2.02 mg/100g. The value of 64.02 ± 0.02 mg/100g for Potassium obtained by Omotoso [8] for *Cirina forda* are quite similar with values of 65.04 ± 2.02 mg/100g for moth caterpillar obtained in this work. Potassium intake has been found to lower blood pressure by antagonizing the biological effect of sodium. A high intake of potassium has been reported to protect against increasing blood pressure and other cardiovascular risks [24]. A K/Na ratio of less than 1.0 is recommended. The K/Na ratio found in this insect therefore suggests that it could be a potential component of diets for the management of hypertension. Calcium concentration of 32.24 ± 0.06 mg/100g was obtained from moth caterpillar. Banjo, *et al*. [25] in their own work found calcium content of 42.16 mg/100g for grasshopper, result similar to 32.24 ± 0.06 mg/100g for moth caterpillar in this work. Calcium is important in the diets of children and adults for effective bone and teeth development. It is needed for the formation of muscles, heart and digestive system development. The amount of calcium in the insect suggests that its consumption can increase the calcium in the body and contribute tremendously to the blood clotting process. Phosphorus like calcium is also involved in calcification of bones and teeth. It plays a vital part in the oxidation of nutrients in the form of phosphate groups in ATP. The value of phosphorus in the insect though lower when compared to the values obtained for beef (156.0 mg/100g), liver (313 mg/100g) and eggs (218 mg/100g), suggest that the insect is a good source of phosphorus. Banjo, *et al.* [25], reported Phosphorus value of 110.8 mg/100g for *Cirina forda,* result similar to those obtained in this work.

Omotoso [8] in a separate study recorded value of 62.31 ± 0.01 mg/100g magnesium in *Cirina forda* larva. This value is in agreement with that obtained for emperor moth caterpillar (67.31 ± 0.21 mg/100g) in this work. Differences in mineral contents may be due to variations in the dietary habits of the insects or as a result of different ecotypes. Differences may also be due to the age of the insects. Calcium and Magnesium play significant roles in photosynthesis, carbohydrate metabolism and act as binding agents of cell walls. Calcium assists in teeth development while Magnesium plays a role in regulating the acid-alkaline balance in the body [8]. It also helps to maintain normal muscle and nerve function, supports a healthy immune blood and regulates blood sugar levels. The Iron content of 54.70 ± 1.0 mg/100g obtained by Subhachai, *et al.* [14] from sun-dried ants is higher than the value of 12.85 ± 0.10 mg/100g obtained in this work. Lower values of Iron content for moth caterpillar have been reported by Banjo, *et al.* [25]. Iron can serve as an antioxidant and can prevent cardiomyopathy and growth retardation. The iron content of 12.85, mg/100g in moth caterpillar, compares favorably with corresponding values in conventional animal products like liver (11.4 mg/100g). The recommended dietary allowance of iron is 2-5 mg/day [20]. The high content of iron in the insect is encouraging because iron deficiency is a problem in the diets of pregnant women in the developing world [26]. Iron facilitates the oxidation of carbohydrates, proteins and fats. Iron is one of the mineral elements which may be lacking in an average diet and so there is the need to be conscious of taking diets rich in iron especially the vulnerable group. The Manganese level in moth caterpillar is 7.53 ± 1.15mg/100g. Akinnawo and Ketiku [15], reported value of 7.0 mg/100g of manganese in *Cirina forda* which is in agreement with the findings of this study. Akinnawo and Ketiku, (2000) reported values of 8.6 mg/100g zinc in *Cirina forda* which is higher than zinc content (3.71 mg/100g) for moth caterpillar found in this work. Zinc can prevent cardiomyopathy and growth retardation. Its deficiency can lead to loss of appetite and impaired immune function. Reports have shown that Zinc deficiency causes hair loss, diarrhea, delayed sexual maturation, impotence and eye and skin lesions [27].

The amino acid profile of emperor moth caterpillar is shown in Table 3. The nutritional value of any food depends largely on the quality of the protein it contains, which in turn is determined to a large extent, by the amino acid composition. From the results, the insect is rich in essential amino acids such as leucine, isoleucine, phenylalanine, methionine, valine, threonine, and lysine. The presence of these amino acids is important since there is an increasing cost of conventional protein sources in the third world. In a recent study, Ife and Emeruwa [17], found the concentration of Valine in *Oryctes monoceros* as 2.64 g/100g. This result is comparable with the value of 3.246 ± 0.008 g/100g found in this study. The FAO/WHO/UNO [28] recommended daily intake of valine (an aliphatic amino acid) for adults is 26mg/kg body weight.

Table 3. Amino acid profile of Emperor moth caterpillar (*C. forda*)

Amino acid	Composition (g/100g dry weight)
Valine	3.246 ± 0.008
Tryptophan	3.873 ± 0.175
Threonine	2.989 ± 0.038
Histidine	1.709 ± 0.001
Methionine	0.965 ± 0.171
Lysine	2.949 ± 0.643
Leucine	3.344 ± 0.554
Isoleucine	2.459 ± 0.322
Phenylalanine	2.302 ± 0.971
Aspartic acid	6.551 ± 0.062
Serine	2.888 ± 0.039
Glutamic acid	7.379 ± 0.066
Glycine	2.943 ± 0.081
Alanine	3.040 ± 0.014
Arginine	2.834 ± 0.081
Proline	3.072 ± 0.057
Total essential amino acid(TEAA)	23.836
Total non-essential amino acid (TNEAA	29.16
Total amino acid analysed (TAA)	53.00
% TEAA	44.97
%TNEAA	55.02
TEAA/TNEAA	0.82

The concentration of **tryptophan (Try)** is 3.873 ± 0.175 g/100g. This high value of Tryptophan could be due to the method of hydrolysis and derivatization used for the analysis of tryptophan. Tryptophan content of 2.10 g/100g has been reported by Ife and Emeruwa [17]. The concentration of **threonine (Thr)**, a hydroxyl containing amino acid, in the insect is 2.989 ± 0.038 g/100g.

Aremu and Olaofe [29] and Ghaly and Alkoaik [30] quoted threonine concentrations of 3.9, and 4.3% in *Tilapia quineensis,* and American bollworm, values slightly higher than those obtained in this work. Ife and Emeruwa [17] have reported Threonine value of 2.90g/100g for *Oryctes monoceros*, which is very close to the threonine value of 2.989g/100g obtained for emperor moth caterpillar in this work. The RDA of threonine is 15 mg/kg body weight or 8.8 mg/g protein.

The concentration of **lysine (Lys)** is 2.949 ± 0.643 g/100g. Ife and Emeruwa [17] quoted a value of 2.83 g/100g as the lysine content in *Oryctes Monoceros* larva. The value of lysine quoted by Teflo, *et al.* [23] for edible stink-bug, *E. delegorguei* (2.4 g/100g), is in agreement with values of moth caterpillar in this study. The recommended dietary requirement of lysine is 30 mg/kg body weight [28].

The concentration of **leucine (Leu)** is 3.344g/100g. The leucine content of (3.12 g/100g) reported by Oyarzum, *et al.* [31] for *Macrotermis falciger* is similar to values in this work. The value of 6.5 and 6.30 g/100g leucine was reported by Aremu and Olaofe [29] and Ife and Emeruwa [17], for tilapia fish and *Oryctes monoceros* are quite higher than the value obtained in this study. This may not be unconnected with environmental and specie differences. The recommended daily amount of leucine (Leu) in adult humans is 39 mg/kg body weight [28].

The concentrations of **isoleucine (Ile)** obtained from this study 2.459 ± 0.322 g/100g. Ife and Emeruwa [17] also reported value of isoleucine as 3.04 g/100g in the larva of *Oryctes monoceros.* In an earlier work, Oyarzum, *et al.* [31] reported lower values of 1.69 ± 0.05% as the isoleucine content in *Macrotermis falciger.* Aremu and Olaofe [29] and Ghaly and Alkoaik [30] reported isoleucine content of 4.2 % and 4.4% in *T. quineensis* and American bollworm respectively, values which are higher than those obtained in this work. The recommended daily amount of isoleucine prescribed by WHO [28], is 20 mg/kg body weight for adults.

The concentrations of **phenylalanine (Phe)** is 2.302± 0.971 g/100g which is very similar to phenylalanine content of 2.3g/100g documented by Teffo, *et al.* [23] in *Delegorguei spinola*.. Ghaly and Alkoaik [30] also reported phenylalanine value of 2.0 g/100g in maize- stalk borer, which is similar to the value in this study. Ife and Emeruwa [17] quoted higher value of 4.65 g/100g as the phenylalanine content of *Oryctes monoceros* larva. The RDA of phenylalanine has been given as 25 mg/kg or 1750 mg/70 kg body weight [29].

All the insects exhibited reasonable concentrations of essential amino acids, and they can therefore be exploited as human food. Cereal based diets common in developing countries could receive a boost with the inclusion of these insects in the diet.

Table 3 also shows the concentrations of the non-essential amino acids found in emperor moth caterpillar. The concentration of **Aspartic acid (Asp)** is 6.551 ± 0.062 g/100g in emperor moth caterpillar. Aspartic acid rejuvenates cellular activity, cell formation and metabolism. The high value recorded in this insect is therefore a great advantage to consumers of these insects. Ife and Emeruwa [17] reported aspartic acid value of 7.18 g/100g similar to those of this work. The concentrations of **Glutamic (Glu) acid** is high (7.379 ± 0.066 g/100g) in emperor moth caterpillar. Aremu and Olaofe [29] recorded 13.8 g/100g glutamic acid content in *T.quineensis.* **Glycine (Gly)** concentration reported in this work compares with reports by Ife and Emeruwa [17]. Glycine is involved in the transmission of impulses in the nervous system. **Alanine (Ala)** content in emperor moth caterpillar is 3.040 ± 0.014 g/100g. Alanine strengthens the immune system and prevents build up of toxic substances in the body. The high alanine content observed in this insect could be an advantage. Ife and Emeruwa [17] reported similar alanine content of 2.34g/100g in *Oryctes monoceros* similar to that found in this study.

The fatty acid composition of oil obtained from moth caterpillar is shown in Table 4.

Table 4. Fatty acid composition of oil extracted from Emperor moth caterpillar (*C. forda)*

Fatty acid	Trivial name	Composition (%)
Undecane	-	3.10
Hexadecane	-	2.41
Pentadecane	-	2.06
Pentadecanoic acid	-	1.85
9 – Octadecenoic acid	Oleic acid	3.70
Hexadecanoic acid	Palmitic acid	7.35
Heptadecanoic acid	Margaric acid	3.67
9, 12 – Octadecadienoic acid	-	8.94
Octadecanoic acid	Stearic acid	9.65
Undecanoic acid	-	2.01
Eicosanoic acid	Arachidic acid	2.36
Docosanoic acid	Behenic acid	2.23
Tetracosanoic acid	Lignoceric acid	2.20
Squalene	-	3.59

Fourteen fatty acids were identified comprising 38.89% of total saturated fatty acids (TSFA), and 30.34% of total unsaturated fatty acids (TUFA). Saturated fatty acids identified include, Palmitic, C16:0 (7.35%), Stearic C18:0 (9.65%), Arachidic C20:0 (2.36%) and Docosanoic or Behenic acid C22:0 (2.23%). Polyunsaturated fatty acids (PUFA) account for 24.91%. The dominant unsaturated fatty acid being Oleic, C18:1 (3.70%). Hydrocarbons such as undecane (3.10%), hexadecane (2.41%) and pentadecane (2.06%) were also identified. Kayode, *et al.* [32], have observed that in insects, fatty acids may be complex having odd numbers of carbon atoms, branched chains or contain a variety of functional groups including hydroxyl or keto groups and even ring structures. The range of fatty acids observed in moth caterpillar oil is therefore not strange. The percentage of saturated fatty acid obtained in moth caterpillar is comparable to 35.5% and 29.6% reported for poultry and fish, but lower than 52.0% and 44.1% reported for beef and pork respectively [29]. Oleic acid, a monounsaturated fatty acid constitutes 68.14% of the total monounsaturates. Palmitic and Myristic acids have been demonstrated to raise low density lipoprotein (LDL) cholesterol and are therefore considered atherogenic [17].

The presence of these fatty acids in the oil of moth caterpillar seems to compliment the function of one another. The occurrence of Oleic acid may be an advantage as it could be readily converted to either α-linolenic or γ- linolenic or both which are essential fatty acids (EFAs). Being a good source of unsaturated fatty acid, moth caterpillar could be useful in the diet of humans [18]. The moderately high TUFA/TSFA ratio of moth caterpillar suggests that it could serve as an alternative dietary source for both humans and animals.

4. Conclusions

In this study fatty acids and amino acid profiles of emperor moth caterpillar (*Cirina forda*) were investigated using GC/MS and HPLC respectively. Proximate and mineral analyses were also carried out using methods of AOAC and flame analyzer and AAS respectively. The results obtained show that the insect is a good source of protein, essential amino acids, fatty acids and other nutrient supplements. Consumption of this unpopular insect is encouraged so as to reduce the pressure on other conventional protein sources among the poorer sector of the population and to alleviate the problem of nutrient deficiency.

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