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RESEARCH ARTICLE

PHYSICO-CHEMICAL PROPERTIES AND METAL IONS CONTENT OF OIL EXTRACTED FROM CICKET (*Brachytrupes membranaceus)* IN BOSSO LOCAL GOVERNMENT AREA OF NIGER STATE, NIGERIA

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ABSTRACT

The proximate, mineral composition, physico-chemical properties and fatty acid compositions of oil extracted from cricket (*Brachtrupes membranaceus*) were investigated. The result of the proximate analysis showed that the insect have ash, moisture, carbohydrate, protein and lipid contents of 6.40 ± 0.11 %, 4.50 ± 0.51%, 25.13±0.13%, 16.80 ± 0.22% and 18.52 ± 0.12 % dry weight respectively. High proportions of sodium, potassium calcium, iron and phosphorus (127.50, 72.50, 9.21, 27.10 and 126.90 g/100g were detected respectively. Manganese, zinc and copper were also detected with K/Na ratio of 0.57 ± 0.02. The oil extracts exhibited good physico-chemical properties with varying proportions of both saturated and unsaturated fatty acids which have potentials to be developed for food or pharmaceutical uses. The acid value and free fatty acids were low confirming the stability and edibility of the oil. With the high carbohydrate, crude fiber and mineral contents, cricket could serve as an alternative source of protein and other nutrient supplements in human and animal diet.

Key words: Proximate analysis, physico-chemical properties, fatty acids, mineral composition, metal ions, cricket.

INTRODUCTION

The part played by insects in the nutrition, art, customs and beliefs of indigenous communities cannot be underestimated (Dunkel, 1998). Insects have played important roles in the history of human nutrition in Africa, Asia and Latin America (Bodenheimer, 1951). Various communities eat insects for food, while others relish them as delicacies. Insects commonly consumed are mostly those which can be collected in large numbers and in Nigeria these include locust in the gregarious phase, winged termites, caterpillars, grasshoppers and the large African cricket "*Brachytrupes" (*Ene, 1963). Holt (1969) advocated the eating of a select list of vegetable eating insects, which he described as "clean, palatable, wholesome and nutritious". DeFoliart, (1992) concludes that insects are really the forgotten food crop particularly because of the contemporary reliance on junk foods. He proposes adding nutritional value to staple diets, and maximizing ecological benefits with edible insects. Crickets (*Brachytrupes membranaceus)* commonly called "Gyare" in Hausa, "Ere" in Yoruba and "Ententen" in Igbo, belong to the family– *Gryllidae* and are somewhat similar to grasshoppers because they are nocturnal and they have similar body structure including jumping hind legs. There are about 900 species of crickets, but the common black cricket (*Gryllus assimilis)* is brown to black in appearance with front wing varying in length, covering half to entire abdomen. They have flattened bodies and long antennae (Gorochov and Mostovski, 2008). Some species may not have wings but the wings are generally held flat over the body. Crickets, like all other insects are omnivores and scavengers, feeding on organic materials as well as decaying plant materials. Information on the quality of the nutrients in most of the popularly consumed insects in Niger State is 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, physico-chemical properties and the fatty acid profiles of oil extracted from cricket in Bosso Local Government Area of Niger State. This will provide baseline data and a reference point for the study of nutritional potentialities of edible insects which will encourage their greater exploitation to meet other nutritional needs in rural communities.

MATERIALS AND METHODS

Sample Collection and Preparation

Fresh samples of adult crickets were collected from several farmlands in Bosso Local Government Area of Niger State, between the months of July and September 2010. The samples were identified by entomologists at the Federal University of Technology, Minna. After removal of wings and intestines the samples were sun-dried for 72 hours to constant weight. The dried samples were made into powder with laboratory pestle and mortar. The powder was sieved through a 2 mm mesh sieve and stored in plastic bottles for analysis. Triplicate determinations were made in each case.

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Proximate analysis

The moisture, ash and crude fiber contents were determined by the methods of Official Analytical Chemist (AOAC, 2000). Total nitrogen (% N) was determined by the micro- Kjeldahl method (AOAC, 2000) 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 *et al.*, (1957) method. The solvent was evaporated on a rotary evaporator and the oil obtained was dried in an oven at 60^0C to remove all traces of solvent. Percentage Nitrogen free extract (carbohydrate) was obtained by difference. All determinations were in triplicate.

Mineral and fatty 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.

Determination of physico-chemical properties of the oil

The crude oil sample was characterized for specific gravity, refractive index, saponification, iodine and peroxide values using AOAC (2000) Official methods of analysis. The Acid value was also determined and the percentage free fatty acid (as oleic) was calculated.

Preparation of Fatty Acid Methyl Ester (FAME) derivatives

The extracted oil was hydrolyzed and the fatty acids converted to their acid methyl derivatives and their concentrations determined using GC/MS. The oil was first methylated by dissolving 0.2 g of the oil in a quickfit conical flask with 6 cm^3 of methanolic NaOH (2 g NaOH in 100 cm^3 methanol) and refluxed for 10 minutes. To this sample, 10 cm^3 of a mixture of 30 cm^3 HCl and 20 cm^3 methanol was added and refluxed again for 10 minutes. To the mixture, 10cm^3 of n-hexane was added and refluxed again for 2 minutes and then cooled. Finally, 10 cm^3 distilled water was added and the lower aqueous layer separated from the methylated oil. CCl_4 was added to remove excess water. The methylated oil was dissolved in pure hexane and introduced into the injector of a GC/MS- gas chromatographic system at an injection temperature of 250^OC using Helium as the carrier gas at a pressure of 100.2 kPa. A linear velocity of 46.3 cm/sec was maintained with splitless injector interfaced to a 5973 mass selective detector. The fatty acids were eluted as peaks whose retention times were measured by the mass spectrometer detector and compared with those of known standards of the Wiley library (Mclaffety and Stauffer, 1988). Individual fatty acids were identified with those of standards. Duplicate determinations were carried out in each case.

RESULTS AND DISCUSSION

Result of proximate composition of cricket is shown in Table 1. The study reveals that the percentage moisture content is 4.50 ± 0.5 %. Subhachae, *et al*. (2010) reported higher moisture content of 8.6 ± 0.1 and 12.8 ± 0.1% for two varieties of edible ants. Low moisture content indicates a good shelf life characteristic. The low moisture content of cricket in this study shows that the insect can be stored for a long period before deterioration. The quantity of ash in a sample supposedly indicates the amount of minerals. The average ash content of 6.40 ± 0.1 % shows that this insect could be a good source of minerals. Ekpo, *et al.* (2009) obtained ash content of 7.38 ± 0.11 and 5.79 ± 0.13 % for *Imbrasia bellina* and *Ranchophorus phoenicis* respectively, results similar to that obtained in this study.

The crude protein content was found to be 16.80 ± 0.2%. 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 are in agreement with the protein value in this work. Since the insect has 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) 1980, for man and animals, particularly in developing countries where the cost of conventional protein sources are expensive. The crude fibre content (28.65 ± 0.4 %) found in cricket in this work is similar to that recorded by Subhachae, *et al.* (2010) which was 26.4 ± 1.4% for edible black ants. High crude fibre in food is known to promote digestibility and enhance health benefits such as reduction of the risk of gastrointestinal cancers (Salvin, *et al.* 1997). The high value of crude fibre in cricket indicates that, they could be used to complement animal roughages in addition to other uses. The crude fibre in the insects could be due to chitin found normally in insects which can reduce serum cholesterol and serve as a haemostatic agent for tissue repairs (Goodman, 1989). Carbohydrate content of 25.13 ± 0.13 % was found in cricket. Mbah and Elekima (2007) obtained values of 18.73 ± 0.03 and 20.01 ± 0.10% for *Macrotermis nigeriensis* and *Zonocerus variegatus* respectively, results which are lower to those obtained in this work.

These differences could be 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 cricket is 1398.05 ± 2.83 Kj/100g. Teffo, *et al.* (2007) obtained energy value of 2600 kJ/100g for the larva of the edible Stink-bug. The high value in this work suggests that the insect could be a good energy source. The crude lipid content of the insect is 18.52 ± 0.2 %. Dung Wang, *et al. (*2004) reported lipid content of 10.3% (dry weight) for cricket which is lower than the value of 18.52 ± 0. 2 % obtained for cricket in this work. Adedire and Aiyesanmi, (1999) found values of 17.65 ± 3.24% for adult grasshopper and 22.93 ± 3.37% crude fat for immature grasshopper. Fat is important in diets as it is a source of fat soluble vitamins and promotes their absorption. Fat acts as temperature sensors and serve as substrates for eicosanoids. They also add taste, colour and lubricity to diets. The high lipid content from the insect studied in this work makes it a good source for the supply of cheap and readily available animal fat.

Mineral composition

The result of the mineral profile of cricket is shown in Table 2. The result shows high values of Na^+, K^+, Fe^{2+}, and phosphorus. The value of sodium is higher than other minerals. The RDA

Table 1: Proximate Composition (% dry wt) of Cricket

Parameter	Moisture	Ash	Crude Protein	Crude Lipid	Crude Fibre	Carbohydrate (by difference)	Energy (kJ/100g)
Mean Value	4.50+ 0.51	6.40+0.11	16.80+0.22	18.52+0.12	28.65+0.42	25.13+0.13	1398.05+2.83

Values are means ± SD of three determinations.

Table 2: Mineral Composition (mg/100 g dry weight) of Cricket

Mineral	Amount (mg/100g)
Na	127.50 ± 0.50
K	72.50 ± 0.50
Ca	9.21 ± 0.01
Mg	0.13 ± 0.01
Fe	27.10 ± 1.03
Mn	2.40 ± 0.01
Zn	1.90 ± 0.10
Cu	2.05 ± 0.10
Pb	0.00 ± 0.00
P	126.90 ± 0.10
K/Na	0.57 ± 0.02
Ca/P	0.07 ± 0.01

*****Values are means ± SD of three determinations

for sodium is 1500 mg for an adult. The amount of sodium from this insect is encouraging because it 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. The amount of potassium (72.50 ± 0.5% obtained in this work is higher than the value of 64.02 ± 0.02 mg/100g Potassium recorded by Omotoso, (2006) for *Cirina forda*. 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 (Langford, 1983). A K/Na ratio of less than 1.0 is recommended. The K/Na ratio (0.57 ± 0.02) found in this insect therefore suggests that it could be a potential component of diets for the management of hypertension. Calcium concentration of 9.21± 0.01 mg/100g was found in cricket**.** Banjo, *et al.* (2006) reported higher value of 21.0mg/100g of Calcium in *Macrotermis bellicosus*. In a recent work by Subhachai, *et al.* (2010), sun-dried ants from Zhejiang were found to contain 49.1 ± 0.7 mg/100g Ca. Such variations could be due to specie differences and mode of feeding among various insects.

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. The recommended dietary allowance of calcium is 120 mg/day for children and 800mg for adults. The calcium content in the insect though low suggests that their consumption can increase the calcium in the body and contribute tremendously to the blood clotting process. The phosphorus concentration in the insect is 126.9 mg/100g. Banjo, *et al.* (2006), reported Phosphorus values of 138.0, 131.2 and 110.8 mg/100g for *Macrotermis bellicosus,* Grasshopper and *Cirina forda respectively* results very similar to that obtained in this work. 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 values of phosphorus in cricket 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. Magnesium concentration was very low in the insect. Banjo, *et al.* (2006) reported similar low values of 0.15 mg/100g Magnesium in *Macrotermis bellicosus*. Differences in mineral contents may be due to variations in the dietary habits of the insects as a result of different ecotypes and 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. It also helps to maintain normal muscle and nerve function, and regulates blood sugar levels. The recommended dietary allowance of iron is 2 - 5 mg/day (NRC, 1980). The iron content of 27.10 mg/100g in cricket found in this work compares favourably with corresponding values in conventional animal products like liver (11.4 mg/100g), beef (1.9 mg/100g) and eggs (2.1 mg/100g) documented by Fox and Cameron, 1980). 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 (Orr, 1986). Iron can serve as an antioxidant and can prevent cardiomyopathy and growth retardation. Iron facilitates the oxidation of carbohydrates, proteins and fats.

It is an antioxidant (Buss, *et al*., 2003). The iron in meat does not only enhance the absorption of iron from other sources such as cereals, but increases the level of iron absorption in the blood and prevent anemia which is widely spread in developing countries (Bender, 1992). The levels of Manganese and Zinc are low in the insect. In a recent report, Ife and Emeruwa, (2011) reported concentrations of 1.21 and 7.00 mg/100g for manganese and zinc respectively in the larva of *Oryctes monoceros.* 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 diarrhea, delayed sexual maturation, impotence and eye and skin lesions (Ryan-Harshman and Aldoori, 2005). The daily requirement of Copper is 1–3 mg (Fox and Cameron, 1980). Subhachai, *et al*. (2010), however reported values of 2.4 ± 0.5 mg/100g mean concentration of copper in sun-dried edible ants. This value is close to that obtained for cricket (2.05± 0.10 mg/100g) in this work. Copper forms part of several enzyme systems including cytochrome oxidase and tyrosinase.

It is associated with iron and catalyses oxidation-reduction mechanisms concerned with tissue respiration (Fox and Cameron, 1980). Lead was not detected in cricket. Subhachai, *et al*. (2010), documented value of 0.4 ± 0.1 mg/100g mean concentrations of Pb from sun-dried edible ants. Lead is very toxic. It is a potent neurotoxin and has a cumulative effect on vital organs (Licata, *et al*., 2004). The absence of lead in this insect should not be overlooked because its presence could be due to fodder contamination, climatic factors such as winds and the use of pesticide compounds. Therefore it is necessary to monitor the level of lead over time for better monitoring of its presence in these insects since human activities get sophisticated day by day thereby polluting the terrestrial environment.

Physico-chemical properties

The results of some determined physico-chemical properties of the crude oil extracted are shown in Table 3. The properties of the oil revealed that, the oil is brownish in colour and solid at room temperature with solidification temperature in the range of 10 to 12^0C which is in agreement with values of 12- 14^0C obtained by Ekpo, *et al*. (2009) for oils of *Macrotermis bellicosus* and *Rhynchophorus phonicis* larval oils. The Specific gravity and refractive index of the oil are 0.9300 ± 0.01 and 1.301 ± 0.01 respectively. The refractive index is a reflection of the optical clarity of the crude oil sample relative to water. The value obtained in this work falls within the range common for vegetable oils. The acid value of 3.14 ± 0.06 mg KOH/g was obtained. Acid values are used to measure the extent to which triglycerides in the oil have been decomposed by lipase action and other physical factors such as light and heat into free fatty acids (FFAs) in a given amount of oil.

Acid values and free fatty acids (as % Oleic acid) are important indexes of physico-chemical properties of oil used to indicate the quality, age, edibility and suitability of oil for a particular use. Ekpo, *et al*. (2009) quoted acid value of 3.60 ± 0.06 and 3.5 ± 0.06 for *Macrotermis bellicosus* and *Ranchophorus phoenicis* larvae which are close to acid value of 3.14 ± 0.06 mg KOH/g in this work.. The low level acid value and free fatty acid and low peroxide values show that the oils from these insects have high stability to oxidation, an indication of its lower susceptibility to rancidity and lipase action. The low acid values also are a testimony to the freshness of the crude oil and suggest that these fatty acids were not degraded from processing stage. These parameters are a measure of the level of spoilage of oil, hence these low values is a reflection of the freshness and edibility of the oil (Onyeike and Oguike, 2003). The saponification value from Table 3 is 190.3 ± 0.30 mg KOH/g. Ekpo, *et al*. (2009) recorded saponification values of 193.4 ± 0.3 and 198.9 ± 0.25 mg KOH/g for *Macrotermis bellicosus* and *Rhynchophorus phoenicis* respectively.

Table 3: Physico-chemical Properties of Oil Extracted from Cricket

Parameters	Values
Appearance	Brown
Specific gravity	0.9300 ± 0.01
Refractive index	1.301 ± 0.001
Solidification value(^{0}C)	10-12
Acid value(mgKOH/g)	3.14 ± 0.06
Free Fatty Acid(% as Oleic)	1.57 ± 0.01
Wij's Iodine value(g I_2/100g)	115.0 ± 1.00
Saponification value(mgKOH/g)	190.3 ± 0.30
Peroxide value (mEq O_2/Kg)	11.6 ± 0.20

Values are means ± SD of three determinations

This is in agreement with the values of 190.3 ± 0.30 mg KOH/g in this study. Saponification value is an index of average molecular mass of fatty acids in an oil sample and it measures the free as well as chain-bounded fatty acids. The high saponification values of the insect oils suggest that the mean molecular weight of fatty acids is also high or that the number of ester bonds is high. This might imply that the fat molecules are intact (Denniston, *et al*. 2004). This is in line with the lower susceptibility to lipase action and other oxidative means (Atasie, *et al*. 2009). Therefore, the high saponification value observed in the oil may suggest its use in the saponification industry. The iodine value of 115 ± 1.00 g I_2/100g was obtained in this work. Ekpo, *et al*. (2009) have earlier shown that *Macrotermis bellicosus* and *Rhynchophorus phoenicis* have iodine values of 108 ± 0.15 and 123.6 ± 0.24 g I_2/100g respectively. Lepidopterous larvae and *Phytophagous chrysomefids* were found to have iodine values of 112 to 159 and 106.6 to 118 g I_2/100g respectively (Ekpo and Onigbinde, 2007). Iodine value of oil is a measure of the degree of unsaturation of the oil. The iodine index found in this work is comparable to those reported for most insect lipids suggesting a reasonable amount of unsaturated fatty acids. The Peroxide value of 11.6 ± 0.2 m EqO_2/kg was obtained in cricket oil. Peroxide value is used as a measure of the extent to which rancidity reactions have occurred. The relatively low peroxide value observed in this work indicates that the oil may have resistance to lipolytic hydrolysis and oxidative deterioration when compared with other oils.

The fatty acid profile of the oil from cricket in Table 4 showed that the oil contain myristic (1.72%), stearic (3.06%), docosanedioc acid (5.73%) and eicosanoic acid (2.02%). Palmitoleic, docosanedioic, hexadecanoic acids and cyclopentadecanone are the most dorminant monounsaturated fatty acids (MUFA), while Myristic (C14:0), Palmitic (C16:0), Stearic (C18:0) and Arachidic (Eicosanoic, C20:0) acids are the dorminant saturated fatty acids (SFA). The presence of both saturated and unsaturated fatty acids in given oil is advantageous since both seem to compliment the function of one another. The high TUFA/TSFA ratio (1.73) in cricket oil is very similar to the value of 1.70 observed by Akinnawo and Ketiku, (2000) for *Cirina forda* suggesting that cricket oil could be useful in dietetic management of coronary diseases.

Table 4: Fatty acid Composition of Oil Extracted from Cricket

Peak no.	R. Time	Fatty acids	Trivial name	% Composition
1	26.372	methyl tetradecanoate	Myristic acid	1.72
2	27.202	Tetradecanoic acid	-	1.53
3	27.824	9-Hexadecenoic acid	Palmitoleic acid	1.76
4	27.972	Pentadecanoic acid	-	2.74
5	28.282	Octadecanoic acid	Stearic acid	3.06
6	28.642	Hexadecanoic acid	-	1.80
7	29.181	Docosanedioc acid	-	5.73
8	29.299	Hexadecanioc acid	-	2.07
9	29.465	Cyclopentadecanone	-	4.46
10	29.573	Octadecanoic acid	-	2.56
11	30.476	Eicosanoic acid	Arachidic acid	2.02
12	31.454	14-Methyl-8-hexadecyn-1-ol	-	2.32
13	31.591	Hexadecanoic acid	-	2.23

Conclusion

The present study have shown that the insect cricket, have the potential to provide substantial amounts of proteins, carbohydrates as well as crude fibre. In addition, they contain high amounts of lipids made up of both saturated and unsaturated essential fatty acids. The oil extracts exhibit good physico-chemical properties and therefore has the potential to be developed either for food, pharmaceuticals and chemical industries. In the absence of other essential fatty acids (EFA) in the selected insects such as Linoleic and Linolenic (Omega - 6 and Omega-3), such fatty acids may be obtained from other sources to compliment our diets. The insect is a rich source of important minerals such as sodium, potassium, calcium, magnesium, zinc, iron and phosphorus. Thus, this insect have the potential of being used as a source of minerals and can be used as nutraceuticals or alternative nutritional food source in the diet of low income people whose diets are usually deficient in essential minerals. The K/Na ratio in the insect suggests that it could be a potential component of diets for the management of hypertension.

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