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NUTRITIONAL AND FUNCTIONAL PROPERTIES OF FLOUR FROM THE PALM (*ELAEIS GUINEENSIS*) WEEVIL *RHYNCHOPHORUS PHOENICIS* LARVAE CONSUMED AS PROTEIN SOURCE IN SOUTH CÔTE D'IVOIRE

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In Côte d'Ivoire, the oil palm (*Elaeis guineensis*) weevil *Rhynchophorus phoenicis* is among the most renowned larvae, widely appreciated and consumed for its delicious flavour. The present work is conducted to highlight the chemical composition and functional properties of flour from these larvae collected in the south of the country. The larvae were collected from dead trunks of oil palms at Anyama (5°29'40" N and 4°03'06" W) in Côte d'Ivoire. The fresh larvae were oven dried and ground to obtain crude flour. Chemical composition and functional properties were investigated using standard methods. The results revealed that dry *R. phoenicis* larva is an excellent source of nutrients mainly consisting of proteins (31.93 %), fats (25.21 %) and minerals. It provides 158.62 % and 129.08 % of the Recommended Dietary Allowances of iron and magnesium respectively, after 100 g dry larva consumption. These findings make it useful in human diet to prevent malnutrition caused by proteins and some minerals deficiencies. Dry *R. phoenicis* larva flour showed high water and oil absorption capacity (281.73 and 139 % respectively). Moreover, this flour exhibited good dispersibility (71 %), wettability (5 min) and foam stability (50 %) making it suitable for nutritional and industrial applications.

Keywords: chemical composition, functional properties, flour, weevil *Rhynchophorus phoenicis* larvae, *Elaeis guineensis*

Introduction

Entomophagy or eating insects has a long history as part of human diets and a large number of insect species are consumed in many parts of the world (FAO, 2004). Within the context of sustainable diet, entomophagy has a significant role to play in assuring food security and improving the livelihood of many peoples in the world. In America, Asia and Africa, many people eat insect as regular part of their diets in a similar way as eating meat or fish (Raubenheimer and Rothman, 2012;

Van Huis *et al*., 2013). In many parts of Africa, eating insects was practiced as a traditional heritage (Christensen *et al*., 2006).

As a food source, insects are highly nutritious. Many insect species contain as much as or more protein than meat or fish. Some insects, especially in the larval stage (caterpillars), are also rich in fat and most insects contain significant percentages of amino acids and essential vitamins and minerals (Durst and Shono, 2010).

Caterpillars are among the world's most diverse groups of edible insects. The consumption of caterpillars is especially pervasive in sub-Saharan Africa, where 30 percent of all edible insect species are caterpillars (Van Huis, 2003). In Côte d'Ivoire, some caterpillars like *Imbrasia oyemensis* (Akpossan *et al*., 2015a; Akpossan *et al*., 2015b) and *Oryctes owariensis* (Assiélou *et al*., 2015a; Assiélou *et al*., 2015b) were previously described for their nutritive value and the functional properties of their flours. Authors have shown that these larvae are excellent sources of nutrients mainly consisting of proteins, fats and minerals. They contain high percentage of unsaturated fatty acids. Their flours also exhibit good functional characteristics for use in many food industries. However, to our knowledge, there are no reports concerning the biochemical properties of the palm weevil *Rhynchophorus phoenicis* which is among the most renowned larvae, widely appreciated and consumed, for its delicious flavour, in the country. Indeed, the larvae attack palm species especially oil palm *Elaeis guineensis*. Thus, adult females lay a few hundred eggs on the new leaves of the plant or directly in the palm trunk. The weevil larvae burrow into the palm heart, causing its death. Then, palm weevil larvae are typically collected, washed and fried for consumption (Fasoranti and Ajiboye, 1993). So, the present work is conducted to highlight the chemical composition and functional properties of flour from *R. phoenicis* larvae collected in the south of Côte d'Ivoire, with the aim to reveal their suitability for nutritional and industrial applications. It is noteworthy that the behaviour of protein in food systems during processing, manufacturing, storage and preparation could be affected by intrinsic physicochemical characteristics also called functional properties (Onimawo and Akubor, 2005).

Materials and methods

Larvae Collection and Flour Preparation

Weevil *R. phoenicis* larvae were collected from dead trunks of oil palms at Anyama (5°29'40" N and 4°03'06" W) in Côte d'Ivoire. After collection, the larvae were kept alive in the palm juice and then transported to the laboratory for flour preparation. Fresh larvae (1 kg) were cleaned with distilled water, then drained and oven dried at 65°C for 72 h. Dried larvae were ground using a blender to obtain crude flour.

Proximate Composition

The moisture content was determined by drying in an oven at 105 °C for 24 h to constant weight (AOAC, 2012). The crude protein content was calculated from nitrogen contents (N×6.25) obtained using the Kjeldahl method by AOAC **(**2012**)**.

The crude fat content was determined by continuous extraction in a Soxhlet apparatus for 8 h using hexane as solvent (AOAC, 2012). The total ash content was determined by incinerating in a furnace at 550 °C (AOAC, 2012). The crude fibre content was determined by taking about 3.0 g sample as portion of carbohydrate that resisted sulfuric acid (1.25%) and NaOH (1.25%) digestion followed by sieving (75 µm), washing, drying and ignition to subtract ash from fiber (AOAC, 2012). The amount of total soluble sugars was estimated by Phenol sulphuric acid reagent method (Dubois *et al.*, 1951). The amount of reducing sugars was determined by using 3,5-dinitrosalicylic acid (DNSA) colorimetric procedure developed by Bernfeld (1955). The carbohydrate content was determined by difference. Energetic value was calculated using Atwater factors of 4 × % protein, 4 × % carbohydrate and 9 × % fat and then taking the sum. Minerals including calcium, magnesium, iron, zinc, copper, manganese and potassium were determined using an Atomic Absorption Spectrophotometer, AAS (Model 372, Perkin-Elmer, Beaconsfield, UK) by wet digestion while phosphorous level was determined using the phosphovanado molybdenate method (AOAC, 2012).

Functional Properties

Water Absorption Capacity (WAC) and Water Solubility Index (WSI)

The water absorption capacity and solubility index were evaluated according to Phillips *et al*. (1998) and Anderson *et al*., (1969) methods, respectively. Two grams (M_0) of flour were weighed into a centrifuge tube and 50 mL of distilled water were added. The content of the centrifuge tube was shaken for 30 min in a KS 10 agitator. The mixture was kept in a water-bath (37ºC) for 30 min and centrifuged (Ditton LAB centrifuge, UK) at 5000 rpm for 15 min. The resulting sediment (M_2) was weighed and then dried at 105ºC to constant weight (M_1). The WAC was then calculated as follows:

$$\text{WSI (\%)} = \frac{M_0 - M_1}{M_0} x\, 100 \quad (1)$$

$$\text{WAC (\%)} = \frac{M_2 - M_1}{M_2} x\, 100 \quad (2)$$

Oil Absorption Capacity (OAC)

For the oil absorption capacity, Beuchat's method (1997) was used. One gram of flour sample was mixed with 10 mL of oil for 30 min in a mixer (Vari-whirl-mixing control, set at fast speed). Afterwards, the sample was allowed to rest at room temperature for 30 min. It was then centrifuged at 5000 rpm for 30 min (Ditton LAB centrifuge, UK) and the volume of the supernatant was measured in a 10 mL graduated cylinder. The density of the oil was determined, too. The volume of oil absorbed was multiplied by the density of the oil to determine the weight of the oil so absorbed.

$$\text{OAC (\%)} = \frac{(V_1 - V_2)\, x\, P}{W} x\, 100 \quad (3)$$

where V_1: initial volume of the oil used; V_2: remained oil volume (not absorbed); P: density of the oil used; W: weight of the sample.

Foaming Capacity and Foam Stability

The foaming capacity (FC) and stability (FS) of flour were studied according to Coffman and Garcia's method (1977). Three g of flour were transferred into clean, dry and graduated (50 mL) cylinders. The flour sample was gently leveled and the volumes noted. Distilled water (30 mL) was added to the sample; the cylinder was swirled with a vortex (Genius 3 France) for 5 min. Initial foam volume was registered and then the cylinder was allowed to rest for 120 min while the change in foam volume was recorded every 15 min.

$$\text{FC } (\%) = \frac{V_t - V_0}{V_0} \; x \; 100 \tag{4}$$

$$\text{FS } (\%) = \frac{FC}{FC_0} \; x \; 100 \tag{5}$$

Where V_0 is the original volume of the sample (mL), V_t is the total volume after different times (mL) and FC_0 is the initial foaming capacity (FC).

Bulk Density Measurement

The volume and bulk density were determined according to Okezie and Bello's modified method (1988), by pouring 2 g of flour into a 10 mL measuring cylinder, and then holding the cylinder on a vortex vibrator for 1 min to obtain a constant volume of the sample. The volume of the sample was recorded against the scale on the cylinder. The bulk density value was calculated as the ratio of mass of the powder and the volume occupied in the cylinder.

Dispersibility Measurement

The dispersibility of flour was measured according to the method developed by Mora-Escobedo *et al*. (1991). One gram of flour was dispersed in distilled water in a 100 mL stoppered measuring cylinder. Then distilled water was added to reach a volume of 30 mL, the mixture was vigorously stirred and allowed to settle for 3 h, the volume of settled particles was subtracted from 30 and multiplied by 100 and reported as percentage dispersibility.

Wettability Determination

The method described by Onwuka (2005) was adopted. One gram of flour sample was measured into a 10 cm^3 measuring cylinder. The cylinder was inverted at 10 cm above the water contained in a 600 mL beaker. The finger was used to close the cylinder preventing the flour sample from falling. By removing the finger and giving the cylinder a gentle tap, the flour sample was discharged into the water surface. The time taken by the sample to get completely wet was recorded as the time of wettability.

Statistical Analysis

All experiments in this study are reported as means of three replicate analyses. One-way analysis of variance (ANOVA) was carried out to compare the mean values. Differences in the mean values were determined using Duncan's multiple range tests (SAS, 1990).

Results and discussion

Proximate composition

The proximate composition of the flour obtained from palm weevil *R. phoenicis* larvae is summarized in table 1. The moisture content of 5.92 % obtained in this study is not very different from the one reported by Assiélou *et al.* (2015a) for flour from the larvae of *Oryctes owariensis* but it is lower compared to the moisture content found by Okaraonye and Ikewuchi (2009) for *Oryctes rhinoceros* larvae flour (16.73 %). This should be advantageous since the lower the moisture content of a product to be stored the better the shelf stability of such products (Sanni *et al.*, 2006). Consequently, low moisture ensures higher shelf stability of dried product. The moisture content of a food is indicative of the dry matter in that food, however low residual moisture content in confectionaries is advantageous in that microbial proliferation is reduced and storage life may be prolonged if stored inside appropriate packaging materials under good environment conditions.

As regards the ash content (3.87 %), it is higher than those of several edible insects such as *Macrotermes bellicosius* (2.90 %), *Macrotermes natalensis* (1.90 %), *Cirina forda* (1.50 %), *Oryctes boas* (1.50 %) and *Zonocerus variegatus* (1.20 %) commonly eaten in South-western Nigeria (Banjo *et al.*, 2006). This suggests at first that this edible insect would be a good source of minerals as the ash content indicates a rough estimation of the mineral content of the product.

The carbohydrate content of the studied larvae (23.04 %) is higher than those of most edible insects as reported by Alamu *et al.* (2013) and Assiélou *et al.* (2015a). Long *et al.* (2007) have revealed that insects have considerable amounts of polysaccharides that can enhance the immunity function of the human body. As regards the total and reducing sugars amount (2.15 and 0.18 % respectively), it seems that these carbohydrates are mainly composed of polysaccharides including fibres which are very beneficial to health.

Proteins constitute the major biochemical component of the studied larvae with a registered value of 37.93 %. Protein content is higher than those reported for edible insects in south-western Nigeria (from 6% for *Brachytrypes* spp. to 30% for *Analeptes trifasciata*) (Banjo *et al.*, 2006). The high protein content is an indication that the palm weevil *R. phoenicis* larvae can be of value in human and animal rations, thus replacing more costly sources of animal protein that are usually absent in the diet of rural dwellers in developing countries. Furthermore, the crude fat content of 25.21 % could be an advantage since some studies have shown that fats are vital in the structural and biological functioning of cells, helping in the transport of nutritionally essential fat-soluble vitamins (Omotoso, 2006) and they are also essential in diets as they increase the palatability of foods by absorbing and retaining their flavours (Aiyesanmi and Oguntokun, 1996). This could explain easily why the larva is very appreciated and renowned for its delicious flavour. The fat content obtained in this study is higher compared to those of some larvae consumed in Côte d'Ivoire such as *Imbrasia oyemensis* (23.96 %) (Akpossan *et al*., 2015a) and *Oryctes owariensis* larvae (18.88 %) (Assiélou *et al*., 2015a). Therefore, due to its fat content, *R. phoenicis* larvae should be a good energy source since according to Aiyesanmi and Oguntokun (1996), lipids are the main energy source which can reduce the consumption of protein and help detoxification. The energy levels provided by the studied larva (higher than 400 kcal/l00 g DW) give about 25 % of the daily energy intakes recommended for a 70 kg person. It is well known that the energy content is mainly affected by the proportion of fat in the sample. The present results point out that edible insects can be used as a source energy diet (Ramos-Elorduy *et al*., 1997). The energy value found in this work (470.77 kcal/100 g) was in the interval of values reported by Ramos-Elorduy *et al.* (1997) for 78 species of edible insects in Mexico, with calorie content ranging from 293 to 762 kcal/100 g.

Table 1. Proximate composition (g/ 100 g DM) of the oil palm weevil *R. phoenicis* larvae flour

Parameters	**Values (%)**
Moisture	05.92 ± 0.30
Dry matter	94.08 ± 0.30
Ash	03.87 ± 0.21
Protein	37.93 ± 0.61
Fats	25.21 ± 0.00
Carbohydrate	23.04 ± 0.11
Crude fibre	04.03 ± 0.02
Total sugars	02.15 ± 0.01
Reducing sugars	00.18 ± 0.00
Energy (kcal / 100 g DM)	470.77 ± 0.07

Values given are the averages of at least three experiments ±SE; DM: Dry matter.

Mineral element composition

Table 2 presents the mineral composition of *R. phoenicis* larvae flour. Results revealed that potassium (1192.00 mg/ 100 g), magnesium (516.33 mg/ 100 g), calcium (225.66 mg/ 100 g), phosphorus (93.40 mg/ 100 g) and iron (12.69 mg/ 100 g) are the most predominant minerals. Thus, the consumption of 100 g dry larva would provide 158.62 % and 129.08 % of the Recommended Dietary Allowances of iron and magnesium respectively. It is noteworthy that iron deficiency is a major problem in human diets in the developing countries, especially for pregnant women and children (0 to 5 year) in Africa. Indeed, in many countries of this continent, one in two pregnant women and about 40 % of preschool children are believed to be anaemic (Van Huis *et al*., 2013). Health consequences include poor pregnancy outcomes, impaired physical and cognitive development, increased risk of morbidity in children and reduced work productivity in adults. Anaemia is a preventable deficiency but contributes to 20 percent of all maternal deaths (FAO, 2001). So, the inclusion of these larvae in the daily diet could improve iron status and help to prevent anaemia in this part of the world.

Concerning magnesium and zinc, they prevent cardiomyopathy, muscle degeneration, growth retardation, impaired spermatogenesis, immune-logic dysfunction and bleeding disorder (Chaturvedi *et al.*, 2004). Indeed, Zinc deficiency is a core public health problem, especially for child and maternal health. Zinc deficiencies can lead to growth retardation, delayed sexual and bone maturation, skin lesions, diarrhea, alopecia, impaired appetite and increased susceptibility to infections mediated via defects in the immune system (FAO, 2001).

Table 2. Mineral element composition of the oil palm weevil *R. phoenicis* larvae

Minerals	Contents (mg/ 100 g)	Intake recommendation for 25 year old males (mg per day)*	Intake for 100 g dry larva (%)
Phosphorus	93.40 ± 0.07	700	13.34
Potassium	1192.00 ± 11.53	4700	25.36
Calcium	225.66 ± 1.15	1000	22.56
Magnesium	516.33 ± 0.57	400	129.08
Iron	12.69 ± 0.07	8	158.62
Manganese	0.53 ± 0.00	2.3	23.04
Copper	0.15 ± 0.00	0.9	16.66
Zinc	2.68 ± 0.02	11	24.36
Lead	0	-	0
Cadmium	0	-	0

Values given are the averages of at least three experiments ±SE

*Dietary reference intakes (DRIs): recommended dietary allowances and adequate intakes, minerals, Food and Nutrition Board, Institute of Medicine, National Academies.

Functional properties

Table 3 depicts the functional properties of *R. phoenicis* larvae flour. This flour shows a dispersibility index of 71 % which is similar to the values reported for sorghum-wheat composite flour as reported by Adebowale *et al.* (2012). This dispersibility index is relatively high indicating that *R. phoenicis* larvae flour has the ability to scatter or disperse over a wide surface area and, therefore, it would be an ideal raw material in various food products, since the higher the dispersibility, the better the flour reconstitutes in water to give a fine and consistent paste (Kulkarni *et al.*, 1991). The bulk density obtained in this study (0.31 g/mL) was significantly lower than the one found for *Imbrasia oyemensis* flour (1.04 g/mL) (Akpossan *et al.*, 2015a). This suggests that *R. phoenicis* flour is less heavy; hence it would occupy more space and would require relatively more packaging materials. However, according to Adebowale *et al.* (2008), bulk density is generally affected by particles size and the density of flour blend. Bulk density is an indication of the porosity of a product which influences package design and could be used in determining the type of packaging material required, material handling and application in wet processing in the food industry (Kinsella, 1987).

Concerning wettability, result reveals that *R. phoenicis* larvae flour wets very quickly (5.00 min) compared to full-fat and defatted flours from *Imbrasia oyemensis* larvae with wettability values of 50.33 and 9.67 min, respectively, as reported by Akpossan *et al*. (2015a). Moreover, these authors have suggested that defatting significantly influences the wettability. Thus, when flour wets quickly, it might be due to the absence of fat since in this work, the flour was defatted before the study of its functional properties.

Generally, the ability of flours to absorb water is attributed to their protein or carbohydrate content. The observed water absorption capacity of the studied flour could therefore be attributed to its proteins and carbohydrates content as they both are the major biochemical constituents found. According to Aremu *et al*. (2009), the difference in protein structure and the presence of different hydrophilic carbohydrates might be responsible for variation in the water absorption capacity of the flours. Indeed, flours with high water absorption capacity have more hydrophilic constituents such as polysaccharides. The water absorption capacity of dry *R. phoenicis* larvae flour (281.73 %) was found to be higher compared to those of *O. owariensis* larvae flour (Assiélou *et al*., 2015a), soy bean flour (130%) (Padilla *et al*., 1996) or African yam bean flour (118-179%) (Oshodi *et al*., 1997). This high value of water absorption capacity of the flour is an indication that it would be useful as a functional ingredient used in the formulation of some foods such as sausage, dough, processed cheese, soups and baked products confirmed by Olaofe *et al*. (1998). The oil absorption capacity of 139.00 % obtained in this study was found to be higher than the values reported for full fat and defatted flours from *Imbrasia oyemensis* larvae (Akpossan *et al*., 2015a). Oil absorption capacity is important since oil acts as flavour retainer and increases the palatability of foods (Aremu *et al*., 2009). Adebowale and Lawal (2004) have reported that variations in the presence of non-polar side chains, which might bind the hydrocarbon side chains of oil among the flours, explain differences in the oil binding capacity of the flours. This result suggests that *R. phoenicis* larvae flour is a high flavour retainer and may therefore find useful application in food systems such as ground meat formulations.

The results (Table 3) show that dry *R. phoenicis* larvae flour is not a good foaming agent, with a foaming capacity of only 6.06 %. This value is lower than the value of flour from *O. owariensis* larvae reported by Assiélou *et al.* (2015a), although, the foam stability (50 %) obtained in this study (after 120 min) was higher than the one found by the same authors. This result suggests that the defatted *R. phoenicis* larvae flour may not be suitable in food system such as cake and ice cream which requires a high percentage of foam. The basic requirements of proteins as good foaming agents are the ability to adsorb rapidly at air water interface during bubbling and to undergo rapid conformational change and rearrangement at the interface. Low foamability can be related to highly ordered globular proteins, which resists surface denaturation. For the stability of the foam, proteins must form a cohesive viscoelastic film via intermolecular interactions (Fennema, 1996).

Table 3. Functional properties of defatted flour from the oil palm weevil *R. phoenicis* larvae

Parameters	Values
Dispersibility (%)	71.00 ± 0.30
Hydrated density (g/mL)	1.25 ± 0.02
Bulk density (g/mL)	0.31 ± 0.02
Porosity (%)	31.25 ± 0.90
Wettability (min)	5.00 ± 2.04
Foaming capacity (%)	6.06 ± 0.00
Foam stability (%)	50.00 ± 0.00
Water absorption capacity (%)	281.73 ± 5.74
Water solubility index	42.50 ± 2.00
Oil absorption capacity (%)	139.00 ± 3.05

Values given are the averages of at least three experiments ±SE (n = 3)

Conclusions

To sum up this report, it appears that dried *R. phoenicis* larva is an excellent source of nutrients mainly consisting of proteins (31.93 %), fats (25.21 %) and minerals. This edible insect was found to be rich in minerals, especially iron and magnesium. This finding makes it useful in human diet to prevent undernourishment caused by proteins and some minerals deficiencies. As regards functional properties, flour from dried *R. phoenicis* larva showed high water and oil absorption capacity indicating its usefulness in many food formulations such as sausages and bakery products. Moreover, this flour exhibited good dispersibility, wettability and foam stability making it suitable for nutritional and industrial applications.

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