



## **RHYNCHOPHORUS PALMARUM L. LARVA, AN EDIBLE INSECT IN CÔTE D'IVOIRE : NUTRITIONAL VALUE AND CHARACTERIZATION OF THE LIPID FRACTION**

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### **ABSTRACT**

There is a very serious worldwide nutritional problem due to foodstuff deficiency affecting low income groups living in rural areas and slum zones in urban cities, however there are many sources of underused staple not enough investigated on in spite of their enormous importance for a very large group of people.

The aim of this study is to assess nutritional quality of the palm tree grub called *Rhynchophorus palmarum* L. (larva) edible insects available and consume at Côte d'Ivoire, to inform the population about the benefits of health they provide and promote their intake on a daily diet basis. Proximate and chemical analysis was carried out on the larva, larval protein and oil of *Rhynchophorus palmarum* L. Chemical score of proteins was also evaluated. The following data were obtained: crude proteins 7.1%, lipids 21.8%, carbohydrates 9.0%, Ash 0.8%. Proteins were rich in the essential amino acids (with histidine, threonine, lysine, phenylalanine) with a high protein score (from 90% to 172%). The limiting amino acids were leucine, valine and methionine, with a score ranging from 53% to 74%. The oil had a high proportion of unsaturated fatty acids (52.4%), part of which were essential fatty acids, linoleic and linolenic acids. Results suggested that *Rhynchophorus palmarum* larva could be a basis of new food or feed products to improve their nutritional value and to highlight the health status of people of Côte d'Ivoire.

**Key words:** *Rhynchophorus palmarum*, Edible insects, Nutritional value, Larva oil, Protein.

### **INTRODUCTION**

The Food and Agriculture Organization (FAO) of the United Nations makes an effort

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to explore the opportunity insect rearing and gathering offer. In this way, with the support of the Government of the Netherlands, it was organized in 2012, an expert consultation on insects as food and feed at FAO's headquarters in Rome. Insects have played an important role in the history of human nutrition in Africa, Asia and Latin America. For example, in Southeast Asia weaver ant larvae and pupae are a very popular dish, In west Africa, Insects, mushrooms, snails and larvae such as the beetle *Rynchophorus phoenicis* F., *Macrotermes subhyalinus* and *Rynchophorus palmarum* L. were a cherished food among the many rural and urban communities<sup>1-4</sup>. There is a number of studies about the nutritional value of insects. Several authors showed that some of the insects which are pests have high nutritional qualities. Proximate composition of these insects have been studied from Central Africa, South Africa<sup>5</sup> and South America<sup>6</sup>. Insects provide high quality of proteins and supplements (minerals and vitamins)<sup>7</sup>. Recently, FAO Experts consultation<sup>8</sup> recognized the nutritional potential of insects. According to them, in general as a food group, insects are nutritious, rich in protein and fat, providing ample quantities of minerals and vitamins, the essential amino acids are often present, but the protein quality of each insect should be considered in relation to the dietary staple. The fiber content (chitin from the exoskeleton) is higher than in conventional meat but comparable to that of cereal grains. All food insects are a significant source of short chain polyunsaturated fatty acids, a good source of iron, calcium and B vitamins. The amino-acid composition is in most cases better than that of grains and legumes.

In Africa, despite of the substantial efforts made in order to increase food production, there is a very serious worldwide nutritional problem due to foodstuff deficiency affecting low income groups living in rural areas and slum zones urban cities, however there are many sources of staple not enough investigated in spite of their enormous importance for a very large group of people. Entomophagy cultural tradition in Africa might be a good option to improve nutritional health to habitants of those regions.

The aim of this study is to assess macronutrients of palm grub *Rynchophorus palmarum* L. edible insects available and consume at Côte d'Ivoire, to inform population the benefits of health they provide and promote their intake on a daily diet.

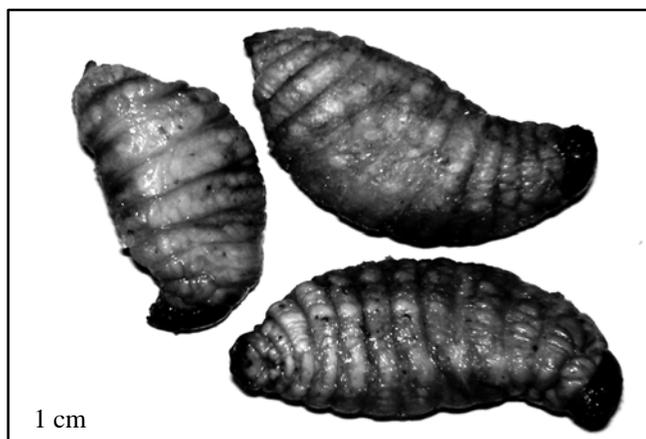
## **EXPERIMENTAL**

### **Materials and methods**

#### **Larvae, sampling and processing**

Live larvae of *Rynchophorus palmarum* L. (Figure 1) were collected in Anyama (Côte d'Ivoire) palm grove. After capture, insect larvae were put in an icebox and were

transported to the laboratory and were used within 24 hours of collection. The species were specifically identified in the entomology department of Université Nangui Abrogoua (Côte d'Ivoire).



**Fig. 1:** Larvae of *Rhynchophorus palmarum* L.

### **Proximate composition analyses**

Crude protein was analyzed using the Kjeldahl method<sup>9</sup> with nitrogen analyzer (Gerhardt Vapodest, 50, Germany). Dry matter was determined gravimetrically after drying sample overnight at 105°C. Lipid content was determined using the method of Bligh and Dyer<sup>10</sup>. Ash was quantified after incinerating the sample in the muffle furnace at 550°C overnight and minerals (potassium, sodium, calcium, magnesium, iron, phosphorus) were analyzed by atomic absorption spectrophotometry (Perkin Elmer, Model 1100, Paris, France) following the method used by Idouraine et al<sup>11</sup>. Carbohydrates content was determined through the method used by Samant and Rege<sup>12</sup>. Energetic value was calculated using the Atwater factors 4, 4 and 9, for carbohydrates, proteins and lipids respectively<sup>13,14</sup>. Lipid class were determined according the method used by Gbogouri et al.<sup>15</sup>

### **Determination of amino acid composition**

Total amino acid composition of samples was determined after hydrolysis in 6 M HCl with phenol (1%) at 150°C for 60 min, in Pico-Tag system (Waters, Milford, Mass., U.S.A.). The phenylisothiocyanate (PITC®) amino acid derivatives were eluted on HPLC Applied Biosystems Model 172 A (Applied Biosystems, Foster City, Calif., U.S.A.) equipped with a PTC RP-18 column (2.1 mm × 22 cm). Sodium acetate (45 mM, pH 5.9) and sodium acetate (105 mM, pH 4.6; 30%), and acetonitrile (70%) were used as buffers.

## Gas chromatography analyses

Fatty acid methyl esters (FAME) were obtained by transmethylation of lipid aliquots (100 mg)<sup>16</sup>: samples were dissolved with 1.5 mL of hexane and 1.5 mL of borontrifluoride in methanol (8%, w/v), and heated at 100°C under nitrogen for 1 h. After cooling, the fatty acid methyl esters were extracted in hexane under nitrogen.

FAME were analyzed by gas chromatography on Perichrom™ 2000 system (Saulx-les-Chartreux, France), equipped with a flame ionisation detector (FID) and fused silica capillary column (50 m × 0.25 mm × 0.5 µm, BPX70 SGE Australia Pty Ltd). Temperatures were set as follows: 2 min initial period at 120°C, increasing at 40°C/min to the second step at 180°C for 8 min, and flowing out at 3°C/min to the final period at 220°C for 45 min. Injection and detector ports were maintained at 230°C and 260°C, respectively. Fatty acids were identified by comparing their relative retention time with appropriate vegetable standards and marine PUFA 2 standards from Supelco (Supelco Park, Bellefonte, PA 16823-0048 USA). The results, made in triplicate, were displayed as percent of total identified fatty acids.

## Differential scanning calorimetry analysis

Thermal characteristics of oil were measured using Differential Scanning Calorimeter (DSC 204 F1 Phoenix®, Netzsch-Gerätebau GmbH, Germany). Purified nitrogen (99.9%) was the purge gas and flowed at 20 mL/min. The calorimeter was calibrated according to the standard procedures established by the manufacturer user book using indium and water. Approximately, 8-10 mg of sample were weighed into aluminum pans and covers were hermetically sealed into place while an empty hermetically sealed aluminum was used as reference. The sample and the reference pans were then placed inside the calorimeter and subjected to the following temperature program: heating to 50°C and holding for 5 min, cooling to -50°C at the rate of 5°C/min and holding for 5 min, heating from -50°C to 50°C at 5°C/min. The transition temperatures, enthalpies of fusion or of crystallization (J/g) were calculated using the Proteus® Software for thermal analysis (DSC 204 F1 Phoenix® Netzsch-Gerätebau GmbH, Germany).

## Statistical analysis

Results are expressed as the mean ± standard deviation of several sample with Kyplot (version 2.0 beta 15, ©1997-2001, Koichi Yoshioka) statistical software. The data were statistically analyzed by one way analysis of variance (ANOVA). Means were compared by Turkey's test. Differences were considered statistically significant at  $P < 0.05$ .

## RESULTS AND DISCUSSION

### Proximate composition

Table 1 shows the proximate composition of *Rhynchophorus palmarum* L. larva. The moisture content of this insect (61.4 g/100 g) is comparable to the moisture content of *Rhynchophorus phoenicis* larva reported by Ekpo and Onigbinde<sup>2</sup>. This value is less than the reported value of fish<sup>17,18</sup> and the value of cow milk, egg<sup>19</sup>. The moisture of food is used as a measure of stability and susceptibility to microbial contamination<sup>20</sup>, so that the meal can be improved by process such as sun-drying, frying, or roasting to extend the keeping quality. On the other hand, the high moisture content may imply that most of the essential nutrients in the larva will be in solution and in forms that are easily available to the body when the larva is consumed as food.

**Table 1: Chemical and nutritional value of *Rhynchophorus palmarum* L. larva (g/100 g of wet product)**

Parameter	g/100 g of wet product
Moisture	61.4 ± 2.4
Crude proteins	7.1 1.0
Lipids	21.8 ± 0.7
Carbohydrates	9.0 ± 0.1
Ash	0.8 ± 0.2
Energetic density (kcal/100 g of wet product)	260.6
Mineral	mg/100 g of wet product
Potassium	443.0 ± 10.0
Sodium	85.4 ± 4.2
Calcium	49.4 ± 5.2
Magnesium	40.5 ± 3.5
Iron	60.3 ± 4.0
Phosphorus	10.5 ± 2.3

Mean ± standard deviation of three determinations

The crude protein content (7.1 g/100 g) is higher than the content of cow milk<sup>19</sup>, less than fish, egg, meat and poultry<sup>19,18</sup>. This protein value was comparable to *rhynchophorus phoenicis* larva<sup>2</sup>.

A relatively high carbohydrates content (9%) is observed when compared with the reported values for meat and fish<sup>19</sup>. This value was also higher than the value of *rhynchophorus phoenicis* larva (2.20 %).

Minerals are among the most important components of edible insects because they are always available in plenty relative to most other foods. Table 1 indicates that the insect is specifically rich in potassium, sodium and iron. About 100 g of the insect is able to provide a reasonable amount of iron, and potassium for these vulnerable groups.

The Table 1 also shows a fairly high level of fat content (21.8 g/100 g), probably one of the reasons why the insect is a favorite of many in the community. From the visual appearance (Figure 1) of the insect, one would not expect so much of fat content from the head, the high level of fat was located in the abdomen. According to Ayieko et al.<sup>21</sup>, the abdomen of black ants which the villagers of Kenya prefer is approximately 50% fat content, and was comparable to the fat content from both head and thorax.

### **Amino acid composition**

The quality of the protein of the larva is determined by its content of the essential amino acids. As shown in Tables 2 and 3, all the essential amino acids were present in the protein of the *Rhynchophorus palmarum* larva in the proportion higher than the egg protein, comparable to that of the fish meal. The larva protein was particularly rich in histidine, threonine, phenylalanine, lysine, isoleucine, those can meet the minimum daily requirement (Table 3) according to McGilvery and Golstain<sup>22</sup> and FAO/WHO/UNU<sup>13</sup>. These essential amino acids are limiting in wheat, rice cassava and maize based diets that prevalent in developing countries<sup>23</sup>.

The limiting amino acids were leucine, valine and methionine with scores were 53.20, 68.60 and 73.85, respectively. These values were lower than values for *Rhynchophorus phoenicis* in which only valine was the limiting amino acid<sup>3</sup>.

The *Rhynchophorus palmarum* larva may constitute a cheaper source of protein and essential nutrients (amino acids) that is easily available and affordable to the native within the localities where the insect larva are found.

**Table 2: Amino acids composition of *Rhynchophorus palmarum* L. larva compared to the conventional food**

Amino acid	<i>R. palmarum</i> L. larva (g/100g of protein) <sup>a</sup>	Whole egg (g/100 g of protein) <sup>19</sup>	Fish meal (g/100 g of protein) <sup>19</sup>
Aspartic acid + asparagine	15.70	ND	ND
Glutamic acid + glutamine	19.72	ND	ND
Serine	3.66	0.95	3.10
Glycine	3.77	0.40	5.35
Histidine*	3.05	0.30	1.93
Arginine	5.00	0.75	4.61
Threonine*	4.86	0.64	3.18
Alanine	2.80	0.73	4.94
Proline	3.00	0.52	3.18
Tyrosine*	2.58	0.52	2.32
Valine*	3.30	0.85	3.81
Methionine*	1.13	0.42	2.05
Cysteine*	0.04	0.30	0.92
Isoleucine*	3.80	0.78	3.23
Leucine*	3.74	1.091	5.42
Lysine*	4.81	0.86	5.81
Phenylalanine*	3.56	0.71	2.90
Total essential amino acids	30.87	ND	ND
Total amino acids	84.52	ND	ND

\*Essential amino acids, <sup>a</sup> Present study, ND. not determined

**Table 3: *Rhynchophorus palmarum* L. larva protein content as per WHO reference protein pattern (Source<sup>19,22</sup>)**

Amino acid	Reference pattern (g/100 g) protein	Chemical score (%)
Lysine	5.17	93.04
Histidine	1.77	172.32
Threonine	3.47	140.05
Valine	4.81	68.60
Methionine	1.53	73.85
Isoleucine	4.19	90.69
Leucine	7.03	53.20
Phenylalanine	3.01	118.72

### Lipid characteristics

Table 4 showed the lipid class and fatty acid compositions. Lipid class composition showed values met in most conventional sources of lipid. Neutral and polar lipid fractions were about 98% and 2%, respectively (Figure 2). The unsaponifiable fraction which includes sterols and fat soluble vitamins was about 0.95%. Palmitic (38.8%) and oleic (44.6%) acids are the major fatty acids in the larval oil. These values are slightly different from those found by Due et al.<sup>24</sup> on different parts of the *Rhynchophorus palmarum* larva : in the skin fat and digestive fat content, palmitic acid was about 39.87% and 40.44%, respectively, and oleic acid was about 45.62% and 46.71%, respectively. Essential fatty acid (linoleic and linolenic acids) were about 4.3% and 1.4%, respectively. What gives a value of n-6/n-3 equal to 3.1, value very close to that recommended in human nutrition (4-10). Proportions of monounsaturated fatty and saturated fatty acids were similar. Proportion of total unsaturated fatty acids (52.4%) of larva oil is higher than reported value for lard, cocoa butter, palm oil, cucurbit seed oil but less than fish oil, olive oil, almond oil<sup>15,25-27</sup>. This high content of unsaturated fatty acids and the good value of n-6/n-3 implies that this oil could be advised to reduce risks of cardiovascular disease, to lower serum cholesterol and triglyceride levels<sup>14</sup>. According to Martirosyan et al.<sup>27</sup>, about 10% of the unsaturated fatty acids is made up of the essential fatty acids, linoleic and linolenic acids enhances the advantage.

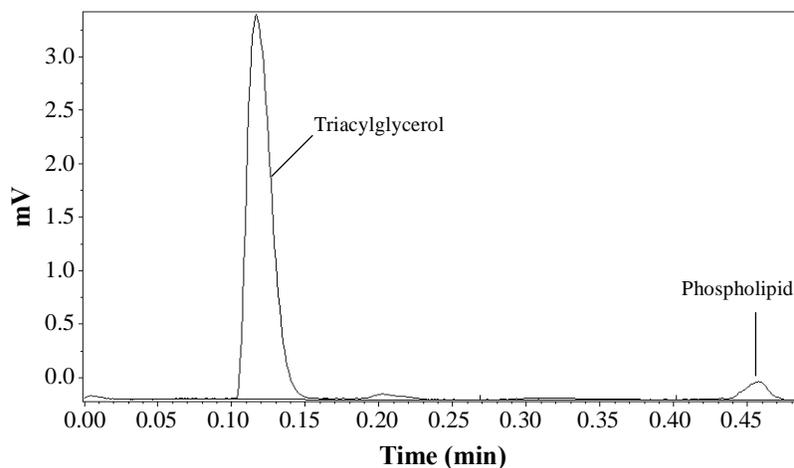


Fig. 2

**Table 4: Lipid class and fatty acids composition of lipids of *Rhynchophorus palmarum* L. (% of total fatty acids)**

	Content (%)
Lipid class	
Neutral lipid	98.0 ± 0.0
polar lipid	2.0 ± 0.1
unsaponifiable matter	0.95 ± 0.12
Fatty acid	
Myristic acid (14 : 0)	2.9 ± 0.1
Palmitic acid (16 : 0)	38.8 ± 0.3
Palmioleic acid (16 : 1 n-7)	2.1 ± 0.0
Stearic acid (18 : 0)	4.9 ± 0.1
Oleic acid (18 : 1 n-9)	44.6 ± 0.3
Linoleic acid (18 : 2 n-6)	4.3 ± 0.1
Linolenic acid (18 : 3 n-3)	1.4 ± 0.0
Others	1.0 ± 0.0
TSFA	46.6 ± 0.6
MUFA	46.7 ± 0.2

Cont...

	Content (%)
PUFA	5.7 ± 0.1
TUFA	52.4 ± 0.3
n-6/n-3	3.1 ± 0.1

Mean ± Standard deviation of three determinations

TSFA, Total saturated fatty acid

MUFA, Total monounsaturated fatty acid

PUFA, Total polyunsaturated fatty acid

TUFA, Total unsaturated fatty acid

### Thermal properties of lipids

Thermal properties were performed using DSC to assess physical properties of *Rhynchophorus palmarum* larva oil. Figure 3 shows the DSC melting and cooling curves for the larva oil. All DSC data is summarized in the Table 5. During the melting process, there is only one endothermic peak (melting point) at 22.5°C with the melting enthalpy about 16 J/g. At this temperature all the fat is in fluidized form. At Contrary, the cooling process showed two endothermic phenomenon; two crystallized peaks appear at 1.6°C and -19.6°C showing two types of triacylglycerols: saturated triacylglycerols crystallized at 1.6°C and the highly unsaturated triacylglycerols at -19.6°C. This is in agreement with the work of Gbogouri et al.<sup>26</sup> The data obtained during cooling process are useful to control fractionation of oil during industrial production heating process.

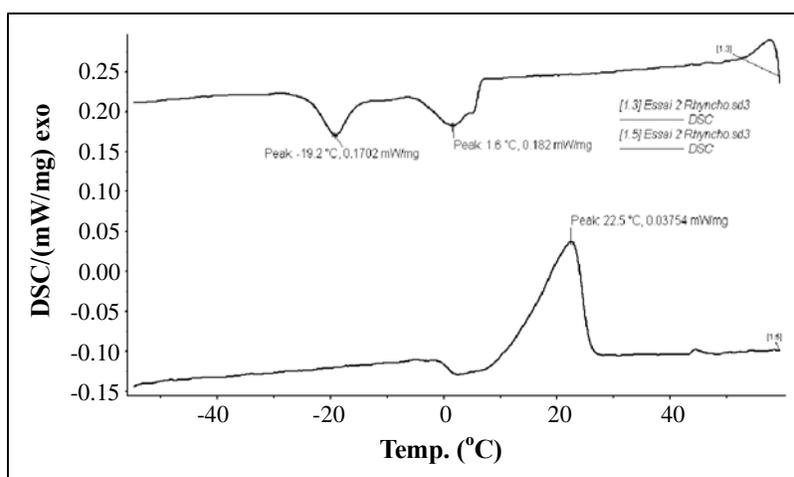


Fig. 3

**Table 5: DSC data obtained from heating and cooling thermograms of lipid of *Rhynchophorus palmarum* larva**

Curve	Transition temperature (°C)		T <sub>on</sub> (°C)	T <sub>off</sub> (°C)	ΔH (J/g)
	1	2			
Melting	22.5		Nd	26.0	16.0
Crystallization	1.6	-19.2	7.0	-25.0	-7.1
Nd Not detected					

## CONCLUSION

The result of the present study show that *Rhynchophorus palmarum* larva is a rich source of good quality protein, essential fatty acids (linoleic and linolenic acids), potassium, calcium, sodium, iron and magnesium. This insect may constitute a cheaper source of essential nutrients that is easily available and affordable to the natives within the localities where the insect larva are found. We recommended that the larva oil must be protected from light and oxygen and stored at low temperatures (< 5°C) to avoid peroxidative changes.

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