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#### Abstract

A large number of insect species are potential sources of valuable nutrients for humans and animals. Inadequate information and advocacy on them seem to make this valuable source under-utilized in nutritional biochemistry. This study was conducted to determine the lipid composition profiles of processed larvae (palm grubs) of *Rhynchophorus phoecinis* and *Rhynchophorus palmarum* using standard methods. Average body weight was: *R. phoenicis* (19.3 ± 1.85g), *R. palmarum* (9.27 ± 3.83g). Lipid analyses results were varied in the two grubs. Crude fat was 6.24 - 7.48 g/100 g; SFA was 28.5 - 31.3% of total fat; MUFA was 37.4 - 39.9%; PUFA was 28.8 - 34.1%; PUFA/SFA was 0.92 - 1.20; AA/DGLA was 0.75 - 9.96; EPA/DHA was 0.17 - 0.17; LA/ALA was 5.59 - 34.6; EPSI was 0.72 - 0.91. The only significant sterol was cholesterol with levels of 77.9 - 103 mg/100g. Total phospholipid range was 386 - 437 mg/100g with phosphatidylcholine (192 - 219 mg/100g) predominating. Significant differences existed in the fatty acid levels of the samples at  $r_{=0.05}$ .

Keywords: Lipid Composition; Rhynchoporus palmarum; Rhynchoporus phoenicis

# Abbreviations

FA: Fatty Acid; LDL: Low Density Lipoprotein; SFA: Saturated Fatty Acid; MUFA: Monounsaturated Fatty Acid; PA: Petroselinic Acid; CV: Coefficient of Variation; LA: Linoleic Acid; ALA: Alpha-Linolenic Acid; AA: Arachidonic Acid; EPA: Eicosapentaenoic Acid; DPA-3: Docosapentaenoic Acid; DHA: Docosahexaenoic Acid; PUFA: Polyunsaturated Fatty Acid; GLA: Gamma-Linolenic Acid; CLA: Conjugated Linolenic Acid; DGLA: Dihomo-Gamma-Linolenic Acid; EPSI: Essential PUFA Status Index; r<sub>xy</sub>: Correlation Coefficient; R<sub>c</sub>: Regression Coefficient; C<sub>A</sub>: Coefficient of Alienation; IFE: Index of Forecasting Efficiency

# Introduction

A number of insects or their products were used as food in the past by some West African tribes, and are to a certain extent still eaten as tit-bits, or exclusively by children. Insects were also used in compounding poisons and curative medicines and palliatives; and some possible associations between them and diseases can be found in folklore.

Insects eaten are mostly those which can be collected in large numbers, such as locusts in the gregarious phase, emerging alate termites, caterpillars, and the large African cricket *Brachytrypes*. Also eaten occasionally and sometimes regarded as delicacies, are fatty 'grubs' such as the enormously distended queen termite, larvae and pupae of scarabaeid beetles, and of the African silk worm *Anaphe* spp. [1].

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The eating of insects, or entomophagy, is not confined to Africa. It is still being practiced in all the five continents, and has been throughout the course of history, and in all past cultures, including those of ancient China, Mexico, Egypt, Israel and Greece. The late Professor Bodenheinmer of Jerusalem has written a 352-page book on the subject [2].

Weevils are most diverse family in the world, occurring from the poles to the tropics, and feeding on all types of plants. They may range in size from around 1 - 50 mm long, and palm-associated weevils span the entire range. Most can be easily recognized by the elongated rostrum, or snout, bearing small mandibles at the tip. Larvae possess a large head capsule with correspondingly large mandibles, are legless, and move peristatically. They are usually whitish or cream-coloured, but often turn an orange-yellow colour prior to pupation. Late instar (http://itp.lucidcentral.org/id/palms/sap/glossary.htm#instar) larvae may be significantly larger than the imago (http://itp. lucidcentral.org/id/palms/sap/glossary.htm#imago), up to 64 mm long and 25 mm wide.

Worldwide there are ten species of *Rhynchophorus* that feed on palms. The *R. ferrugineus* and *R. palmarum* are essentially found in most warm areas around the world where there are palms, particularly the Mediterranean. You might call them...Med-weevil... *Rhyn-chophorus* (rhin-KOH-for-us) is Greek meaning "snout bearing" and *cruentatus* (krew-en-TAT-us) is Latin for blood coloured.

In their book on Cameroon cuisine, Grimaldi and Bikia [3] described their recipe for "coconut larvae" as a favourite dish offered only to good friends.

The flavour of "palmworms" (fat, legless larvae of the weevil genus *Rhynchophorus*) has been appreciated throughout the tropical world for centuries. There are number of species, but the major ones from the standpoint of wide distribution and use as food are *Rhynchoporus palmarum* in the Western Hemisphere, *R. phoenicis* in Africa, and *R. ferrugineus* in Asia.

The scientific classification is: Kingdom (Animalia), Phylum (Arthropoda), Class (Insecta), Order (Coleoptera), Family (Curculionidae), Genus (*Rhynchophorus*), Species (*R. palmarum* and *R. phoenicis*), Binomial name (*Rhynchophorus palmarum* and *Rhynchophorus phoenicis*).

According to Beckerman [4] and Bedford [5], palm grubs (larvae of African palm weevil) live and feed on the starchy pulp of the trunk of the raphia palm (*Rhynchophorus phoenicis* or African raphia palm weevil) and oil palm (*Rhyncophorus palmarum* or African oil palm weevil), which are both common in the tropical rainforest of Nigeria. Though African palm weevils are pests as they destroy valuable plant materials, the grubs are highly valued delicacies in Western and Niger Delta regions of Nigeria, where they are either eaten raw or after cooking by boiling, roasting or frying while some are used for medicinal purposes [6,7]. Palm grub has been shown to be high in crude protein (23.4 %), fatty acids, minerals (zinc and iron) and vitamins (thiamine and riboflavin) [8]. Hence, the need to exploit the nutrient potentials of palm grubs in order to bridge the gap between animal protein supply and consumption. This line of thought has therefore generated the present research interest in determining the lipid composition profiles (crude fat, fatty acids, sterols and phospholipids) of *R. palmarum* and *R. phoenicis* on comparative basis.

#### **Materials and Methods**

#### **Collection of Sample**

Samples of raphia and oil palm tree grub (*Rhyncophorus phoenicis* and *Rhyncophorus palmarum*) were collected from Igbo-Orisa in Ikole –Ekiti, Ekiti State, Nigeria. About 15 whole samples of each type were collected together with pulpy centre of decaying palm tree trunk, put in a rubber container (with cover), sprayed with distilled water (to maintain a moist environment) and brought to the laboratory.

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#### Sample Treatment

The mature larvae were sorted out (eight in number). The weight, length and diameter of the eight samples were taken when fresh. The samples were then properly identified as *R. phoenicis* and *R. palmarum*. The samples were then oven-dried at about 80°C till constant weight was achieved and later dry-milled, the flour was sieved with 200 mm mesh. The sieved samples were kept in the deep freezer in plastic containers pending analysis.

About 0.25g of each sample was weighed into the extraction thimble and fat extracted with petroleum ether (40-60°C boiling range) using a Soxhlet apparatus [9]. The extraction lasted 5 - 6h.

The crude fat extracted was converted to the methyl ester using the boron trifluoride method [9]. The gas chromatographic conditions for the analyses of FAME (fatty acid methyl esters) were as follows: The GC was the HP 5890 powered with HP ChemStation rev A09.01 [1206] software [GMI, Inc, Minnesota, USA] fitted with a flame ionization detector (FID). A split injection with split ratio of 20:1 was used. GC inlet temperature was 250°C with an oven programme of initial temperature at 60°C, first ramping at 10°C/min for 20 min (maintained for 4 min), second ramping at 15°C/min for 4 min (maintained for 10 min) and detector temperature at 320°C. A capillary column (30 m x 0.25 mm) packed with a polar compound (HPINNOWAX) with a diameter (0.25 µm) was used to separate the esters. The peaks were identified by comparison with standard fatty acid methyl esters. Carrier gas used was nitrogen.

The sterol analysis was as described by AOAC [9]. The aliquots of the processed fat were added to the screw-capped test tubes. The samples were saponified at 95°C for 30 min, using 3 ml of 10 % KOH in ethanol, to which 0.20 ml of benzene had been added to ensure miscibility. Deionised water (3 ml) was added and 2 ml of hexane was added in extracting the non-saponifiable materials. The extractions, each with 2 ml of hexane, were carried out for 1h, 30 min and 30 min respectively. The hexane was concentrated to 1 ml in the vial for gas chromatography analysis and 1µl was injected into the injection pot of GC. The GC conditions of analyses were similar to the GC conditions for methyl esters analyses. The peaks were identified by comparison with standard sterols.

The method of Raheja., *et al.* [10] was employed in the analysis of the phospholipids content determination. The GC conditions for analyses of phospholipids were similar to FAME analyses except in the following: Column type was HP5, oven programme initial temperature at 50°C, second ramping at 15°C/min for 4 min, maintained for 5 min and the detector was pulse flame photometric detector (PFPD).

For the purpose of ensuring the accuracy of the results obtained, the followings were prepared: sterols, phospholipids and fatty acid methyl esters which were then compared with respective analytical results; calibration curves were prepared for all the standard mixtures and correlation coefficient determined for fatty acids, sterols and phospholipids. Correlation is a statistical index that shows the quality assurance of the calibration curve performed. It was prepared with the Hewlett Packard Chemistry (HPCHEM) software (GMI, Inc 6511 Buncker Lake Blud Ramsey, Minnesota, 55303, USA).

Statistical analyses [11] were carried out to determine mean, standard deviation, and coefficient of variation in per cent. Further statistical analysis was carried out by calculating the linear correlation coefficient ( $r_{xy}$ ), linear regression coefficient ( $R_{xy}$ ), coefficient of determination ( $r_{xy}^2$ ), coefficient of alienation (CA), index of forecasting efficiency (IFE) in per cent and subjection of the  $r_{xy}$  to standard Table value at  $r_{=0.05}$  and two degrees of freedom.

# **Results and Discussion**

#### Results

The body size distribution is shown in Table 1. For *R. palmarum*, mean weight (g) was 9.27 ± 3.83 and range of 6.78 - 17.6 (= 10.8)

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Statistics*	R. palmarum	R. phoenicis
Live weight (g)		
Mean	9.27	19.3
Range	(6.78 - 17.6) = 10.8	(16.1 - 21.6) = 5.52
Standard deviation (SD)	3.83	1.85
Coefficient of variation (%)	41.4	9.58
Length (cm)		
Mean	5.64	8.93
Range	(4.80 - 6.50) = 1.70	(8.10 - 10.2) = 2.10
Standard deviation (SD)	0.61	0.68
Coefficient of variation (%)	10.7	7.66
Diameter (cm)		
Mean	1.68	2.86
Range	(1.30 - 2.70) = 1.40	(2.60 - 3.10) = 0.50
Standard deviation (SD)	0.47	0.18
Coefficient of variation (%)	27.8	6.45

with a coefficient of variation (CV) of 41.4 %; mean length (cm) was 5.64 ± 0.61 and range of 4.80 - 6.50 (=1.70) with CV% of 10.7; mean diameter (cm) was 1.68 ± 0.47 and range of 1.30 - 2.70 (= 1.40) with CV % of 27.8.

**Table 1:** Body size distribution of oil palm and raphia palm tree grubs(Rhyncoporus palmarum and Rhynchoporus phoenicis) (wet weight).\*Eight studies.

The results of the crude fat content and the energy content of the samples are shown in Table 2. The fat content was low at 7.48 - 6.24 g/100 g and corresponding energy levels of 284 - 237 kJ/100 g. The lower values of crude fat and energy were due to *R. phoenicis* and the higher values due to *R. palmarum*.

Parameter	R. palmarum	R. phoenicis	Mean	SD	CV %
Crude fat (g/100 g)	7.48	6.24	6.86	0.88	12.8
Energy (kJ, kcalories)	284 (67.3)	237 (56.2)	261 (61.7)	33.3 (7.89)	12.8 (12.8)

Table 2: Crude fat and fat energy values from R. palmarum and R. phoenicis (dry weight).

In Table 3, the levels of saturated fatty acid (SFA) and monounsaturated fatty acid (MUFA) are shown. The following fatty acids (FAs) were not detected: C2:0, C3:0, C4:0and C5:0 in the two samples. Also, the following FAs recorded 0.00 % level in the samples: C6:0, C8:0, C10:0, C12:0 and C18:1 (trans-11).

Table 4 depicts the polyunsaturated fatty acid (PUFA) n-6 and n-3 composition of grub samples.

Table 5 contains the summary of the results in Tables 3 and 4 as well as some ratio values.

The statistical analysis of the results in Tables 3 and 4 as presented in Table 6 shows that correlation coefficient  $(r_{xy})$  was high at 0.9180.

Fatty Acid	R. palmarum	R. phoenicis	Mean	SD	CV%
C2:0 Acetic	_ <sup>a</sup>	_ <sup>a</sup>	-	-	-
C3:0 Propionic	-	-	-	-	-
C4:0 Butyric	-	-	-	-	-
C5:0 Valeric	-	-	-	-	-
C6:0 Caproic	0.00	0.00	0.00	_b	_b
C8:0 Caprylic	0.00	0.00	0.00	-	-
C10:0 Capric	0.00	0.00	0.00	-	-
C12:0 Lauric	0.00	0.00	0.00	-	-
C14:0 Myristic	0.38	0.33	0.36	0.04	9.96
C16:0 Palmitic	23.8	26.4	25.1	1.84	7.32
C18:0 Stearic	3.30	3.63	3.47	0.23	6.73
C20:0 Arachidic	0.48	0.42	0.45	0.04	9.43
C22:0 Behenic	0.45	0.39	0.42	0.04	10.1
C24:0 Lignoceric	0.055	0.048	0.052	0.005	9.61
SFA	28.5	31.3	29.9	1.98	6.62
C14:1( <i>cis</i> -9) Myristoleic	0.16	0.14	0.15	0.014	9.43
C16:1( <i>cis</i> -9) Palmitoleic	4.25	5.13	4.69	0.62	13.3
C18:1(trans-6) trans-Petroselinic	0.17	0.15	0.16	0.014	8.84
C18:1( <i>cis</i> -6) Petroselinic	16.4	18.3	17.4	1.34	7.74
C18:1(trans-9) Elaidic	0.016	0.014	0.015	0.001	9.43
C18:1(cis-9) Oleic	15.5	15.4	15.5	0.07	0.46
C18:1(trans-11) Vaccenic	0.00	0.00	0.00	-	-
C20:1( <i>cis</i> -11) Gondoic	0.61	0.53	0.57	0.06	9.92
C22:1( <i>cis</i> -13) Erucic	0.15	0.13	0.14	0.014	10.1
C24:1 ( <i>cis</i> -15) Nervonic	0.055	0.048	0.052	0.005	9.61
MUFA	37.4	39.9	38.7	1.77	4.57
trans-	0.19	0.17	0.18	0.014	7.86
cis-	37.2	39.7	38.5	1.77	4.60

**Table 3:** Saturated and monounsaturated fatty acids composition of R. palmarum and R. phoenicis(dry weight) (% levels of total fat).

SFA: Saturated fatty acid; MUFA: Monounsaturated fatty acid; a = not detected; b = not determined.

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Fatty Acid	R. Palmarum	R. phoenicis	Mean	SD	CV%
C18:2( <i>cis</i> -9, 12) Linoleic ( <i>n</i> -6)	30.2	15.3	22.8	10.5	46.3
C18:2( <i>cis-</i> , trans-11) Rumenic ( <i>n</i> -6)(CLA)	0.20	0.18	0.19	0.014	7.44
C18:3(cis-6, 9, 12) GLA (n-6)	0.81	2.74	1.78	1.36	76.9
C20:2( <i>cis</i> -11, 14) Eicosadienoic ( <i>n</i> -6)	0.068	0.060	0.064	0.006	8.84
C20:3 (cis-8, 11, 14) DGLA (n-6)	0.56	0.50	0.53	0.042	8.00
C20:4 ( <i>cis</i> -5, 8, 11, 14) Arachidonic ( <i>n</i> -6)	0.42	4.94	2.68	3.20	119
C22:2( <i>cis</i> -13, 16)Docosadienoic ( <i>n</i> -6)	0.055	0.048	0.052	0.005	9.61
C18:3( <i>cis</i> -9, 12 15) Alpha linolenic ( <i>n</i> -3)	0.87	2.73	1.80	1.32	73.1
C20:3 ( <i>cis</i> -11, 14, 17) Eicosatrienoic ( <i>n</i> -3)	0.30	0.26	0.28	0.028	10.1
C20:5( <i>cis</i> -5,8,11,14,17) Timnodonic (EPA) ( <i>n</i> -3)	0.055	0.048	0.052	0.005	9.61
C22:6( <i>cis</i> -4,7,10,13,16,19) Cervonic (DHA) ( <i>n</i> -3)	0.32	0.28	0.30	0.028	9.43
PUFA	34.1	28.8	31.5	3.75	11.9
<i>n</i> -6	32.3	23.7	28.0	6.08	21.7
n-3	1.83	5.10	3.47	2.31	66.7

The regression coefficient (R<sub>c</sub>) had a value of 0.82 meaning that for every I unit increase in the parameters determined in *R. palmarum* there was a corresponding increase of 0.82 units in the *R. phoenicis*.

# **Table 4:** Polyunsaturated fatty acid (PUFA) levels (% total fat) of R. palmarum and R. phoenicis (dry weight) CLA: Conjugated linoleic acid; GLA: Gamma-linolenic acid; DGLA: Dihomo-gamma linolenic acid.

The spread of various fatty acids was more pronounced in the *R. palmarum* than in *R. phoenicis* as shown by these respective values: mean value of  $3.85 \pm 8.10$  and  $3.85 \pm 6.94$ . The coefficient of alienation (C<sub>A</sub>) was 39.65% while the index of forecasting efficiency (IFE) per cent was 60.35. The IFE is a reduction in the error of prediction of relationship between the two samples. The r<sub>xy</sub> level at r = 0.05 was 0.388 which showed a significant difference in the fatty acids levels in the two grubs.

Parameter	R. palmarum	R. phoenicis	Mean	SD	CV%
SFA	28.5	31.3	29.9	1.98	6.62
MUFA	37.4	39.9	38.7	1.77	4.57
trans-	0.19	0.17	0.18	0.014	7.86
cis-	37.2	39.7	38.5	1.77	4.60
PUFA	34.1	28.8	31.5	3.75	11.9
<i>n</i> -6	32.3	23.7	28.0	6.08	21.7
<i>n</i> -3	1.83	5.10	3.47	2.31	66.7
PUFA/SFA	1.20	0.92	1.06	0.20	18.7
MUFA/SFA	1.31	1.27	1.29	0.028	2.19
<i>n-6/n-3</i> PUFA	17.7	4.65	11.2	9.23	82.6
LA/ALA	34.6	5.59	20.1	20.5	102
AA/DGLA	0.75	9.96	5.36	6.51	122
EPA/DHA	0.17	0.17	0.17	0.00	-
EPSI	0.91	0.72	0.82	0.13	16.5

 Table 5: Some parameters calculated from Tables 3 and 4.

 LA: Linoleic acid; ALA: Alpha linolenic acid; AA: Arachidonic acid; DGLA:

 Dihomo-gamma-linolenic acid; EPA: Eicosapentaenoic acid; DHA: Docosa 

 hexanoic acid; EPSI: Essential PUFA status index.

Statistics	Values
Correlation coefficient $(r_{xy})$	0.9180
Coefficient of determination $(r_{xy}^{2})$	0.84
Intercept (Regression coefficient, Rc)	0.82
Mean of fatty acids due to R. palmarum	3.85
Standard deviation of fatty acids due to R. palmarum	8.10
Mean of fatty acids due to <i>R. phoenicis</i>	3.85
Standard deviation of fatty acids due to R. phoenicis	6.94
Coefficient of alienation ( $C_A$ ) %	39.65
Index of forecasting efficiency (IFE) %	60.35
Degrees of freedom = $n-2 = 26-2$	24
r <sub>= 0.05</sub>	0.388
Statistical decision	Significant

Table 6: Statistical analysis of the results in Tables 3 and 4.

The sterol results in Table 7 showed the values of cholesterol to be high in all the samples ranging from 103 - 77.9 mg/100g representing percentage levels of 99.3 - 99.1% making other types of sterols very irrelevant since they constituted 0.70 - 0.93% of the total sterols in the samples.

Sterol	R. palmarum	R. phoenicis	Mean	SD	CV%
Cholesterol	102.50324	77.85154	90.177397	17.4	19.3
Cholestanol	4.73441e-4	6.32826e-4	5.531335e-4	1.13	20.4
Ergosterol	1.96914e-3	2.16821e-3	2.068675e-3	0.14	6.80
Campesterol	7.08256e-1	7.07107e-1	7.076815e-1	0.008	0.118
Stigmasterol	1.66947e-3	1.69171e-3	1.68059e-3	0.016	0.936
5-Avenasterol	8.85468e-3	8.85747e-3	8.856075e-3	0.002	0.022
Sitosterol	6.43315e-3	6.40140e-3	6.417275e-3	0.022	0.350
Total	103.23090	78.57840	90.90465	17.4	19.2

Table 7: Sterol levels (mg/100 g) of the grub samples (dry weight).

Phospholipid	R. palmarum	R. phoenicis	Mean	SD	CV%
Phosphatidylethanolamine	116 (26.5 %)	107 (27.7 %)	112	6.36	5.71
Phosphatidylcholine	219 (50.1 %)	192 (49.7 %)	206	19.1	9.29
Phosphatidylserine	78.4 (17.9%)	71.0 (18.4 %)	74.7	5.23	7.00
Lysophosphatidylcholine	3.15 (0.72 %)	1.08 (0.28 %)	2.12	1.46	69.2
Phosphatidylinositol	19.5 (4.46 %)	14.9 (3.86 %)	17.2	3.25	18.9
Total	437	386	412	36.1	8.76

Table 8: Phospholipid levels (mg/100 g) of the grub samples (dry weight).

In Table 9, the  $r_{xy}$  was low (0.0979) with high  $R_c$  (34.4). The two mean values were close but scattered within each sample with values as 87.3 ± 86.6 (*R. palmarum*) and 39.1 ± 47.4 (*R. phoenicis*). The  $C_A$  was high (99.52%) and IFE was very low (0.48%) thereby making the prediction of relationship between the phospholipids level of the grubs difficult. The reduction in the error of prediction of relationship was just 0.48%. The  $r_{xy}$  value at  $r_{=0.05}$  was 0.878 showing result was not significant differently between the grub phospholipid values.

Statistics	Samples Values
Γ <sub>xy</sub>	0.0979
r <sub>xy</sub> <sup>2</sup>	0.0096
Rc	34.4
Mean due to <i>R. palmarum</i>	87.3
Standard deviation due to R. palmarum	86.6
Mean due to R. phoenicis	39.1
Standard deviation due to R. phoenisis	47.4
C <sub>A</sub> (%)	99.52
IFE (%)	0.48
Degrees of freedom = $n-2 = 5-2$	3
r <sub>= 0.05</sub>	0.878
Statistical decision	Not significant

Table 9: Statistical analysis of the phospholipid levels (mg/100 g) of the grub.

On pair-wise comparisons the *R. palmarum* was less than *R. phopenicis* in all the parameters determined. However, the range in all the parameters was least in each case for *R. phoenicis* but on the reverse, the CV % in the *R. palmarum* was greater than the *R. phoenicis* on pair-wise basis. Adeyeye and Aye [12] reported mean body weight of 15.7g, mean length of 9.47 cm and mean body diameter of 2.0 cm in *R. phoenicis* whereas in *R. palmarum* mean body weight was 30g, mean body length was 3 - 4 cm and mean body diameter was 1.0 - 1.5 cm [13]. Dufour [14] reported a live weight of 3 - 16g for the grubs. While the larva of *R. phoenicis* was white, that of *R. palmarum* was yellow.

The crude fat values were close to the level in *R. phoenicis* flesh (9.37 g/100g) and viscera (6.27 g/100g) [12]. The crude fat content in the present samples was close and therefore had a low CV% of 12.8.

Palmitic acid (C16:0), was the overall highest saturated FA in both samples ranging between 23.8 - 26.4% total fat with a trend of *R. phoenicis* > *R. palmarum*. This acid accounts for 27% of the FAs in beef. There is a very strong evidence that C16:0 raises serum cholesterol levels [15] and that this occurs predominantly by increasing bad cholesterol (LDL) levels. This FA accounts for most cholesterol-raising activity from fat sources, thereby increasing the risk of atherosclerosis, cardiovascular diseases and stroke [16]. Stearic acid (C18:0) is the second highest SFA with range value of 3.30 - 3.63% with a trend of *R. phoecinis* > *R. palmarum*. The acid accounts for about 18 % of the FA in beef. Several studies have shown that the C18:0 effects on total cholesterol is minimal and not detrimental to human health [17]. For practical purposes, stearic acid is essentially neutral in its effects on serum total cholesterol, similar to oleic acid [15]. It is not clear why stearic acid does not raise cholesterol level as do other SFAs. A possible reason could be that it is rapidly absorbed into tissue compared with other SFAs [15]. Lauric (C12:0) and myristic (C14:0) FAs are responsible for raising bad cholesterol levels in blood serum [15] and have been shown to be strongly correlated with early heart attack [18]. However, the percentages of lauric (0.00 - 0.00%) and myristic (0.38 - 0.33%) in the samples were low. These results suggest that the grubs have no more cholesterol-raising effect than chicken or fish and therefore, grubs need not to be eliminated from cholesterol-lowering diets [19]. Other minor SFAs were arachidic (C20:0), behenic (C22:0) and lignoceric (C24:0) which were all low in values with respective levels of 0.48 - 0.42%, 0.45 - 0.39% and 0.055 - 0.048%. The lowest CV% was recorded in total SFAs (6.62%) and highest in behenic acid (10.1%) and others between 6.73 - 9.96% (for SFAs having percentage values greater 0.00 %).

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Petroselinic acid (C18:1 cis-6) occurs up to a level of 50% or more in seed oils of the Umbelliferae family, including carrot, parsley and coriander. Kleiman and Spencer [20] gave the levels of petroselinic acid (PA) as 24 - 37% in the various species of Umbellifere and Araliaceae. The unusual position of the double bond in this FA provides an opportunity for production, through cleavage of this bond, of valuable raw materials-lauric and adipic acids. Petroselinic acid range in the present samples was 16.4-18.3 %. The data of Weber, *et al.* [21] on fat absorption and fecal excretion indicate that petroselinic acid from dietary triacylglycerols is absorbed as readily as oleic acid. Petroselinic acid was the highest concentrated of all the MUFA FAs.

Trans-18:1 levels in the samples were trans petroselinic acid (C18:1 trans6) (0.17 - 0.15%), elaidic acid (C18:1 trans-9) (0.016 - 0.014%), vaccenic acid (C18:1 trans-11) (0.00 - 0.00%). Tissues of ruminant animals, such as cows, sheep and goats, can contain a number of different 18:1 isomers like C18:1trans-9 (5.0 %) and C18:1 cis-9 (85 %), C18:1 trans-11 (0.47 %) and C18:1 cis-11 (47 %) [22]. Although cis-vaccenic acid was not in these results, however its biosynthetic precursor C16:1(cis-9) palmitoleic acid had values range of 4.25 - 5.13 % as the third highest level of the MUFA FAs. Gondoic acid C20:1 (cis-11) is a common if minor constituent of animal tissues and fish oils, often accompanied by the 13-isomer. It is also found in rapessed oil and seed oils of related species; it had a range of 0.61 - 0.53% in the grubs. Erucic acid [C22:1 (cis-13)] was 0.15 - 0.13% in the grubs.

Elaidic acid can raise bad cholesterol (LDL) in serum [23]. Trans-fatty acids do not have beneficial properties compared to cis-fatty acids. Trans-fatty acids may behave similar to SFAs. Palmitoleic acid is beneficial in reducing bad cholesterol (LDL), it also reduces the fat deposition in blood vessels and reduces blood clot formation [15]. Oleic acid [C18:1 (cis9)] is the primary MUFA in beef and accounts for about 33 % of the FA in beef. In the present report oleic acid had a range of 15.5 - 15.4% with the trend: *R. palmarum* > *R. phoenicis* values. Oleic acid does not raise serum cholesterol [19]. In several studies on the relative carcinogenicity of fatty acid or their ability to suppress the immune system, oleic acid is the FA with the least negative effect [15]. One reason why oleic acid may not raise serum cholesterol concentrations is because it is a favoured substrate for the liver enzyme that converts cholesterol to an inactive form (the Acyl CoA transferase: cholesterol acyltranferase) [15]. Oleic acid formed the second most concentrated MUFA FA in the samples. While the total MUFA (trans) was 0.19 - 0.17 %, the MUFA (cis) was 37.2 - 39.7% in the samples. In both the SFA and the MUFA FAs, the trend was *R. phoenicis* > *R. palmarum*.

The major important polyunsaturated FAs found in beef are linoleic acid (LA) (C18:2) (about 3.5%), alpha-linolenic acid (ALA) (C18:3) (1.5%), arachidonic acid (AA) (C20:4) (about 1%), eicosapentaenoic acid (EPA) (C20:5) (< 1%), docosapentaenoic acid (DPA-3) (C22:5) (< 1%) and docosahexaenoic acid (DHA) (C22:6) (< 1%) [24]. ALA (C18:3) is classified as a short-chain omega-3 FA and is also found in nuts and seeds. EPA, DPA-3 and DHA are found predominantly in foods of marine origin and are classified as long-chain omega-3 FAs. Both linoleic acid (C18:2) and AA (C20:4) belong to the omega-6 family of FAs. The present levels of LA were higher than the literature value cited above, the range was 30.2 - 15.3%, this was also the case in AA with values of 0.42 - 4.94%. For the n-3 FAs, EPA range was 0.055 - 0.048%, DHA is 0.32 - 0.28%, ALA was 0.87 - 2.73 and eicosatrienoic acid (GLA) with value range of 0.81 - 2.74%.

The omega-3 FAs, like ALA (C18:3), appear to have little direct value for human health. However, the human body can add 2 or 4 carbons to these 18carbon chains fats to produce 20- or 22-carbon chains omega-3 FA. Thus, ALA is a precursor for EPA (C20:5) and DHA (C22:6) FAs, which are important for human health. It has been suggested that ALA has a beneficial effect on cardiovascular heart disease [25]. ALA (C18:3) may help balance LA (C18:2) and be beneficial.

For many years linoleic acid (LA) (C18:2; omega-6) was thought to be the preferable FA for the diet because it was considered to be the most effective cholesterol-lowering FA. However, despite an increase in LA intake (from about 4% to 7%), there has been a growing reservation about recommending its consumption, due to no proven long-term safety [15]. Current recommendations have been moderated and now caution that intakes of this fatty acid should not exceed current concentrations (about 7% of total energy intake) [15]. However,

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recent information from the American Heart Association indicates that LA has a noticeable effect on lowering cholesterol further than oleic and palmitic acids when plasma cholesterol levels are high (> 200 mg/dl). They suggest that at a 10% calorie intake in the form of PUFAs, LA achieves a maximal effect on cholesterol lowering. It also has been suggested [26] that a higher intake of LA appears to protect against stroke, possibly through potential mechanism of decreased blood pressure, reduced platelet aggregation and enhance deformability of erythrocyte cells.

LA and ALA are FAs that can be transformed to CLA (conjugate linolenic acid) by bacteria in the rumen [27] hence, rumenic acid. Interest in CLA research has increased in the past few years as a result of reports of CLA consumption providing several health benefits [20]. Because plants do not synthesize CLA, ruminant fats in milk or meat are the primary dietary CLA sources for humans. It has been found that CLA is an antioxidant, which also reduces circulating cholesterol in mice [28]. Other literature reports that CLA has a positive effect by reducing cardiovascular risk, protects against atherosclerosis, is anti-carcinogenic, reduces body contents of adipose tissue and lipid, and enhances the immune system [28]. The level of CLA in the grub samples ranged from 0.20 - 0.18% with the trend as *R. palmarum* > *R. phoenicis*.

It has been suggested that AA (C20:4) is detrimental to human health [29]. However, it promotes inflammation that is an important protective response when one is injured. It also forms the basis of antiinflammatory prostaglandins that the body uses, to reduce inflammation [30]. The amount of AA in beef is very low (< 0.5% of total fat); thus great amounts of beef have to be consumed to detect any contradictory effect. AA in the grubs was much higher than 0.5% in the beef, values ranged from 0.42 - 4.94% with *R. palmarum* < *R. phoenicis*.

Two other omega-3 FAs in the samples, DHA (C22:6) and EPA (C20:5), have been reported to have health benefits. These omega-3 FAs have been shown to prevent cancer, cardiovascular disease, as well as being therapeutic for arthritis, autoimmune disease, inflammatory effects and depression [31]. DHA is also important during pregnancy for infant and brain development and reduces the incidence of premature birth. EPA lowers blood cholesterol and reduces blood clotting, allowing better circulation. Thus, there is a benefit from the production of additional DHA and EPA by the body's elongation and desaturation of shorter chain FAs (C18:3 omega-3; 18:2 omega-6; 18:1 omega-9), omega-9 in humans. In the grubs EPA ranged from 0.055 - 0.048% whereas DHA ranged from 0.32 - 0.28%.

Erucic acid (C22:1; about 1% in beef fat) is a fatty acid that is apparently responsible for a favourable response of persons with nervous system disorders [32]. The level of erucic acid in this report ranges from 0.15 - 0.13 %. The administration of erucic acid in the diet will reduce the serum levels and brain accumulation of very long chain SFAs (such as 26:0) responsible for demyelination [33]. Accumulation of certain long-chain FAs is associated with degenerative diseases of the central nervous system such as behenic acid (C22:0; about 1%) in beef fat) and lignoceric acid (C24:0; about 1%) as well as that of the unsaturated members of the C22 and C24 group. Behenic acid levels ranged from 0.45-0.39 %; lignoceric acid levels ranged from 0.055 - 0.48% in the grubs. Accumulation occurs because enzymes needed to maintain turnover of this FAs are lacking. Behenic acid has been detected to be a cholesterol-raising SFA factor in humans [34].

The relative amounts of PUFA and SFA in dietary oils is important in nutrition and health. The ratio of PUFA/SFA (P/S ratio) is therefore important in determining the detrimental effect of dietary fats. The higher the P/S ratios the more nutritionally useful is the dietary oil. This is because severity of atherosclerosis is closely associated with the proportion of the total energy supplied by SFAs and PUFAs [35]. The present PUFA/SFA levels ranged from 1.20 - 0.92 which is averagely good. Dietary studies on rats and animals have shown that ALA is a strong suppressor of n-6 FA metabolism, whereas 10 times as much LA is required to give an equal suppression of n-3 metabolism [36]. The n-6 and n-3 FAs have critical roles in the membrane structure and as precursors of eicosanoids, which are potent and highly reactive compounds. Since they compete for the same enzymes and have different biological roles, the balance between the n-6 and n-3 FAs in diet can be of considerable importance. The ratio of n-6 to n-3 or specifically LA to ALA in the diet should be between 5:1 and 10:1 [37] or 4-10 g of n-6 FAs to 1.0 g of n-3 FAs. As LA is almost always present in foods, it tends to be relatively abundant in animal tissues. This is supported in the present report as follows: C18:2 (n-6) ranged as follows: 30.2 - 15.3% whereas C18:3 (n-3) ranged as 0.87 - 2.73%. In

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turn, these FAs are the biosynthetic precursors in animal systems of C20 and C22 PUFAs, with 3 - 6 double bonds, via sequential desaturation and chain-elongation steps. On the overall n-6/n-3 ratios, the values of 17.7 to 4.65 follow the trend of LA/ALA of 34.6 to 5.59; only the values in the *R. phoenicis* fell within the standard. In the diet of *R. palmarum* as food source, it should be supplemented with n-3 source. The relative proportion of MUFA/SFA is an important aspect of phospholipids compositions and changes to this ratio have been claimed to have effects on such disease states as cardiovascular disease, obesity, diabetes, neuropathological conditions and cancer. The MUFA/ SFA levels in the samples ranged from 1.31 to 1.27 which were much better than in the PUFA/SFA particularly in *R. phoecinis*. For example, MUFA/SFA have been shown to have cytoprotective actions in pancreatic  $\beta$ -cells. Cis-Monoenoic acids have desirable physical properties for membrane lipids in that they are liquid at body temperature, yet are relatively resistant to oxidation.

A high ratio between AA and DGLA [dihomo-gamma-linolenic acid, C20:3n-6 (cis-8, 11, 14)], as an indicator of  $\Delta$ -5 desaturase activity in the skeletal muscle phospholipids has been related to good insulin sensitivity. Table 5 gives the AA/DGLA values range as 0.75 - 9.96 which were favourable to good insulin sensitivity. Both mead acid (C20:3n-9) (5z, 8z, 11z)- Eicosa-5, 8, 11- trienoic acid and osbond acid (C22:5n-6) are produced if insufficient essential PUFA are available to meet PUFA requirements in the synthesis of long-chain PUFA. Both mead and osbond acids are therefore essential PUFA status markers. The higher the EPA/DHA and EPSI (essential PUFA status index) the better the essential PUFA status.

Cholesterol is a high-molecular weight alcohol that is manufactured in the liver and in most cells. Along with SFA, cholesterol in the cell membrane gives cells necessary stiffness and stability.

This is why serum cholesterol levels may go down temporarily when we replace SFA with PUFA in the diets [38]. Cholesterol acts as a precursor to vital corticosterols, hormones that help us deal with stress and protect the body against heart disease and cancer; and to the sex hormones like androgen, testosterone, estrogen and progesterone. Cholesterol is a precursor to vitamin D, a very important fat-soluble vitamin needed for healthy bones, teeth and nervous system, proper growth, mineral metabolism, muscle tone, insulin production, reproduction and immune system function. The bile salts are made from cholesterol. Bile is vital for digestion and assimilation of fats in the diet. Research shows that cholesterol acts as an antioxidant [39]. This is the likely explanation for the fact that cholesterol levels go up with age. As an antioxidant, cholesterol protects us against free radical damage that leads to heart disease and cancer. Cholesterol is needed for proper function of serotonin receptors in the brain [40]. Serotonin is in the body's natural "feelgood" chemical, low cholesterol levels have been linked to aggressive and violent behaviour, depression and suicidal tendencies. Mother's milk is especially rich in cholesterol and contains a special enzyme that helps the baby utilise the nutrient. Babies and children need cholesterol-rich foods throughout their growing years to ensure proper development of brain and nervous system. Dietary cholesterol plays an important role in maintaining the health of the intestinal wall. This is why low cholesterol vegetarian diets can lead to leaky gut syndrome and other intestinal disorders.

Cholesterol levels in literature from many animal protein sources are either lower or higher than the grubs cholesterol levels. Values in mg/100 g are: fish (50 - 60), egg yolk (1260), meat and poultry (60 - 120), brain (2000 - 3000), liver (300 - 350) [41]; others are rabbit, lean (71), others brain, sheep (2200), liver: ox (270), sheep (430), pig (260) and calf (370) [42]. Most authorities, but not all, recommend a reduction in dietary cholesterol to around 300 mg or less per day [41]; the two grubs had levels of cholesterol of 103-77.9 mg/100 g which means at least about 300 mg of *R. palmarum* or 311 mg of *R. phoenicis* will be consumed per day to meet this minimum level. The CV% of the cholesterol level was 19.3 which means the values were close in the two samples.

Phospholipids are not essential nutrients: they are just another lipid and, as such, contribute 9 kcalories per gram of energy. Minor contributor to the phospholipids level was lysophosphatidyl choline that contributed less than 1% in each of the grub samples (0.72 - 0.28%). Actual values for the lysophosphatidyl choline ranged from 3.15 - 1.08 mg/100g. The total phospholipids level ranged from 437 - 386 mg/100g showing the grubs to be moderately high in phospholipids content. The highest phospholipid was lecithin (phosphatidyl-choline) with values ranging from 219 - 192 mg/100g or 50.1 - 49.7%; this was slightly followed by phosphatidylethanolamine (cephalin,

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PE) with values ranging from 116 - 107mg/100g or 26.5 - 27.7% and for third position (phosphatidylserine): *R. palmarum* (78.4 mg/100g, 17.9%), *R. phoenicis* (71.0 mg/100g, 18.4%). The fourth position was occupied by Ptd Ins (PI) (phosohatidylinositol) with values of 19.5 mg/100g (4.46%) (*R. palmarum*) and 14.9 mg/100g (3.86%) (*R. phoenicis*). The CV% was generally low to high at 5.71 - 69.2.

Lecithin is usually the most abundant phospholipid in animals and plants, often amounting to almost 50 % of the total, and as such it is the key building block of membrane bilayers. This observation is true for lecithin values in these results with percentage values ranging from 50.1 - 49.7%. Phosphatidylcholines (PC) are a class of phospholipids that incorporate choline as a head group. They are a major component of biological membranes and can be easily obtained from a variety of readily available sources such as egg yolk or soy beans from which they are mechanically extracted or chemically extracted using hexane. They are also a member of the lecithin group of yellow-brownish fatty substances occurring in animal and plant tissues. At birth and throughout infancy, phosphatidylcholine concentrations are high (as high as 90% of the cell membrane), but it is slowly depleted to as low as 10 % of the cellular membrane in the elderly. As is such, some researchers in the fields of health and nutrition have begun to recommend daily supplementation of phosphatidylcholine as a way of slowing down senescence [43] and improving brain functioning and memory capacity [44]. Cephalin (PE, phosphatidylethanolamine) is found in all living cells, although in human physiology it is found particularly in nervous tissue such as the white matter of brain, nerves, neural tissue and in spinal cord. The US Food and Drug Administration (USFDA) have stated that consumption of PS may reduce the risk of cognitive dysfunction in the elderly [45]. Early studies of PS distilled the chemical from bovine brain.

Because of concerns about Bovine Spongiform Encephalopathy, however, modern and commercially available products are made from soybeans. PS has been demonstrated to speed up recovery, prevent muscle soreness, improve well-being, and might possess ergogenic properties in athletes involved in cycling, weight training and endurance running. In recent studies, PS has been shown to enhance mood in a cohort of young people during mental stress and to improve accuracy during tee-off by increasing the stress resistance of golfers [46]. PS has been shown to reduce specific immune response in mice [47]. The average daily PS intake from the diet in Western countries is estimated to be 130 mg.

# Conclusions

The findings of this study showed that the body size of mature grubs varied a lot. The crude fat levels were of slight unequal distribution. The SFA was less than the total unsaturated fatty acids in the grubs: 28.5% < 71.5% (*R. palmarum*) and 31.3% < 68.7% (*R. phoenicis*); the trans C18:1 was also generally low at 0.19 - 0.17% making the fats good for human health. The cholesterol was the only sterol of significance and was generally lower than the recommended daily intake of 300 mg per day by 1/3 - 1/4. The phospholipids was generally moderately high and will promote the health of grubs consumers. On the whole palm grubs will serve as good animal food in dietary fat sources.

#### **Quality Assurance**

The correlation determined for all the standards: fatty acids, phospholipids and sterols, all had values ranging as follows: 0.99833 - 0.99997 (fatty acids), 0.99909 - 0.99999 (phospholipids) and 0.99920 - 0.9994 (sterols); all the correlation values were greater than 0.95 which is the critical correlation for acceptance of these types of analytical results, thus attesting to the quality assurance of the determinations.

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