JOURNAL OF FOOD QUALITY & HAZARDS CONTROL
SHAHID SADOUGHI UNIVERSITY OF MEDICAL SCIENCES & HEALTH SERVICES

Journal of Food Quality and Hazards Control 10 (2023) 62-69

Evaluation of the Amount of Some Metals, Fatty Acid and Microbial Load of African Palm Weevil Larvae *Rhynchophorus phoenicis* (Coleoptera: Curculionidae) Collected from Ondo State, Nigeria

J.M. Adesina [1*], A.R. Jose [2], K.D. Ileke [3], T.E. Mobolade-Adesina [2], O.Y. Komolafe [2]

1. Department of Crop Production Technology, Rufus Giwa Polytechnic, P.M.B. 1,019, Owo, Ondo State, Nigeria
2. Department of Science Laboratory Technology, Rufus Giwa Polytechnic, P.M.B. 1,019, Owo, Ondo State, Nigeria
3. Department of Biology, Federal University of Technology, P.M.B 704, Akure, Ondo State, Nigeria

HIGHLIGHTS:

- *Rhynchophorus phoenicis* is a good source of fat, protein, and crude fiber.
- Linoleic acid, oleic acid, and palmitic acid made up most of the fatty acid concentrations of *R. phoenicis*.
- Fe, Mn, and Zn are the most important metal compounds of *R. phoenicis*.
- Consumption of larvae might be used to combat malnutrition deficiencies.

Article type
Original article

Keywords
Fatty Acids
Larva
Metals
Coleoptera
Weevils
Nigeria

Article history
Received: 8 Jan 2023
Revised: 4 Apr 2023
Accepted: 10 May 2023

Acronyms and abbreviations
CFU=Colony Forming Unit
MC=Moisture Content
NA=Nutrient Agar
PDA=Potatoe Dextrose Agar
TA=Tryptone Agar

ABSTRACT

Background: Edible insects are rich in protein, amino acids, fat, vitamins, and trace elements. However, they are the potential carriers of toxicants, allergenic substances, anti-nutrients, and pathogens. The present study aims to determine the proximate and nutritional, fatty acid, metal composition, and microbial load of palm weevil larvae (*Rhynchophorus phoenicis* Fabricius, (Coleoptera: Curculionidae)), an insect species commonly consumed in Nigeria.

Methods: Twenty five *R. phoenicis* were randomly collected in April, 2021 from different local farms. The insects were exterminated by freezing and thereafter defrosted at room temperature in the laboratory; with the exception of the samples for moisture analysis, they were oven dried to a constant weight at around 65 °C for 24 h, grounded, and analyzed for proximate content, fatty acids, metals, and microbial load following standard laboratory procedures.

Results: The results show that *R. phoenicis* contained 45.60% crude fat, 15.79% crude fiber, and 5.25% crude protein by weight. Linoleic acid, oleic acid, and palmitic acid made up most of the fatty acid concentrations at 54.13, 23.86, and 14.19%, respectively. Iron (Fe) content was the highest metal (4.923 ppm), followed by manganese (Mn; 2.767 ppm) and zinc (Zn; 1.04 ppm). The isolated microorganisms were mold and yeast (5×10^{-5} Colony Forming Unit (CFU)/g), *Staphylococcus* sp. (33×10^{-5} CFU/g), and *Micrococcus*/*Bacillus substilis* sp. (5×10^{-5} CFU/g).

Conclusion: The high nutritional composition present in *R. phoenicis* evaluated in this study, compared to the dietary protein value obtained from other animal food sources, suggests the need for their adoption as animal protein and essential fatty acid sources in human diets.

* Corresponding author (J.M. Adesina)
E-mail: moboladesina@rugipo.edu.ng
ORCID: https://orcid.org/0000-0003-1174-9095

To cite: Adesina J.M., Jose A.R., Ileke K.D., Mobolade-Adesina T.E., Komolafe O.Y. (2023). Evaluation of the amount of some metals, fatty acid and microbial load of african palm weevil larvae *Rhynchophorus phoenicis* (Coleoptera: Curculionidae) collected from Ondo state, Nigeria. *Journal of Food Quality and Hazards Control*. 10: 62-69.

DOI: 10.18502/jfqhc.10.2.12668

Journal website: http://jfqhc.ssu.ac.ir

Introduction

Insects make up the largest animal category in the world, accounting for 76% of all species (Stork, 2018). They are crucial elements of ecosystems as well as killers of agricultural crops, disease carriers, and good sources of nutrients. In the industrialized world, insects are seen as a suitable substitute for animal products in both rural and urban settings (Chia et al., 2019). Eighteen orders totaling more than 2,300 species, of which five are at least well-documented, are edible (Tang et al., 2019). There have been reports of edible desert locusts, cutworms, termites, blue butterfly larvae, backstroke eggs, and insect fly maggots from Africa, Asia, and Latin America (Yin et al., 2017).

"Entomophagy" is the name given to the practice of eating insects. According to Tang et al. (2019) and Zabentungwa et al. (2021), entomophagy may be a viable approach to helping people in several nations throughout the world have better nutritional health. Depending on the region, the type of insects accessible, and the ethnic group involved, the ritual may appear different (Johnson, 2010). Edible insects are mostly significant biological resources that are rich in dietary protein, amino acids, fat, carbs, different vitamins, and trace minerals (Alamu et al., 2013; Xiaoming et al., 2010).

According to estimates, 81.4% of all proteins are of vegetable origin, with cereals alone accounting for 57% of those proteins and oil seeds and nuts for 16.8% (Oso and Ashafa, 2021). These natural protein resources are being overharvested due to the population surge, and imported food is becoming more expensive (Tang et al., 2019). Therefore, it is vital to carry out research to find local alternative protein sources globally. It has been believed that since insects are frequently consumed by many organisms, including humans, they could be a desirable replacement (Banjo et al., 2006; Shantibala et al., 2014). Most edible insects are widely accessible, affordable, and have a high-quality protein content that is required to supplement the cereal-based meals eaten in developing nations.

Villagers in Owo, Ondo State, and other states in Nigeria use *Rhynchophorus phoenicis* Fabricius, 1801, (Coleoptera: Curculionidae), also known as African palm grubs or palm weevils and frequently referred to as "Ekuku" in Yoruba dialect, as a good substitute for meat in the soup. Treatment of children's coughs was also thought to benefit from its consumption by Ogoni tribe of Nigeria (Okaraonye and Ikewuchi, 2008). These mature larvae are typically collected by farmers or agricultural households from inside the trunks of fallen palm trees, where adults lay their eggs and raise their young. It measures more than 2 cm in width, and 5 cm in length, and resembles a squishy, cream-colored grub (Okoli et al., 2019).

The development of nutritious food supplements can benefit from knowledge about the nutritional value of these edible insects (Shantibala et al., 2014). Despite the nutritional potential of edible insects, there are safety risks associated with insect consumption. However, there is limited information about the likelihood that it may contain harmful metals and bacteria. From the point of view of food safety, humans may consume food borne pathogens, allergens, toxicants, and anti-nutrients through entomophagy (Rumpold and Schlüter, 2013), which may be responsible for food related diseases and human or animal death. Assessment of microbial loads in food source is commonly used in the evaluation of food microbiological quality or to certify the possible safety of foods (Ebenebe and Okpoko, 2015). So, the current study aimed to characterization *R. phoenicis* proximate analysis, fatty acid, metals, and microbial load compositions to determine its safeness for consumption.

Materials and methods

Sample collection and preparation

Twenty five *R. phoenicis* samples were randomly taken in April, 2021 from an oil palm plantation in Owo, Ondo State. Following Banjo et al. (2006) procedure, the insects were exterminated by freezing. The ice-covered samples were next allowed to defrost at room temperature in the laboratory, and then, with the exception of the samples for moisture analysis, they were oven dried to constant weight at around 65 ^{o}C for 24 h. Before being processed into a powder through an electric grinder (Binatone model, Japan), the larvae were surface sterilized. Each grounded sample was stored in clearly marked, airtight plastic containers, and stored in the same air-dried oven that was used for all laboratory tests.

Moisture Content (MC) determination

A clean empty crucible was weighed as W_1 and 5 g of pulverized *R. phoenicis* sample was added to it and then reweighed as W_2. This was dried for 24 h at 105 °C in a hot air oven. To ensure full drying, the dried sample was chilled in a desiccator before being placed back in the oven. Readings were made using the same timing until constant values were obtained, and the weight was taken as W_3 (AOAC, 2012).

The MC percentage was obtained by the following formula:

$$\%MC=[(W_2–W_3)/(W_2–W_1)\times100]$$

Proximate analysis

The samples were examined in triplicate for moisture, ash, and crude fiber using the AOAC (2012) recommended techniques using the micro Kjeldahl method to measure nitrogen. The nitrogen content was transformed into protein by multiplying it by a ratio of 6.25. The difference was used to determine carbohydrates.

[DOI: 10.18502/jfqhc.10.2.12668]

Ash content determination

The crucible was initially weighed and recorded as W_1, and 2 g of the crushed *R. phoenicis* sample was then added, resulting in a new weight of W_2. The crucible was then heated to 500 °C in a muffle furnace (Gilson Company Inc, USA) for 6 h, after which the sample was allowed to cool and weighed once again as W_3 (AOAC, 2012). Percentage ash content was calculated as:

$\%Ash=[(W_3–W_1)/(W_2–W_1)\times100]$

Crude fiber determination

A 1 L of conical flask marked "W_1" was filled with 2 g of the defatted sample. Then, 200 ml of boiling, 1.25% sulfuric acid (H_2SO_4) (Avondale Laboratories, UK), and 200 ml of water were carefully added, and the mixture was heated for 30 min while keeping the volume constant with a cooling finger. After being filtered using muslin fabric and washed with distilled water, the heated sample was filled into the conical flask with a spatula.

About 200 ml of 1.25% boiling sodium hydroxide (NaOH) (Meru Chem PVT. Ltd. Mumbai, India) was added to the sample and heated at a fixed temperature sustained with cooling finger for 30 min. The residue was completely washed with hot distilled water and then once with 10% hydrochloric acid (HCl) before the sample was filtered using a poplin towel. It was then allowed to cool in a desiccator and weighed as W_2 after being dried in an oven overnight at 105 °C. After that, it was burned to ash at 550 °C for 90 min in the muffle furnace. After cooling, it was weighed as W_3 and recorded (Ademola and Abioye, 2017).

The fiber percentage was achieved by the following formula:

$\%Fiber=[W_2–W_3/W_1\times100]$

Crude fat determination

A clean washed 250 ml extraction flask was oven dried at 105 °C and weighed. About 30 g of the grounded sample was then weighed into a labelled permeable thimble. Thereafter, the thimble mouth was sealed with white clean cotton wool. Two hundred ml petroleum ether (Avondale Laboratories, UK) was added into a 250 ml extraction bottle (AOAC, 2012).

The covered permeable thimble was then placed into the condenser and the apparatus was gathered for extraction, which lasts for 6 h after the permeable thimble was removed and the extraction flask was placed in the water bath to remove traces of petroleum ether from the sample (AOAC, 2012). Then the weight was recorded as W_3.

Fat percentage was determined as follows:

$\%Fat=[(W_3–W_2)/W_1\times100]$

W_1: weight of ground sample; W_2: weight of empty extraction flask; W_3: weight of extraction flask+oil.

Protein content determination

About 0.5 g of pulverized sample was weighed into a dried 500 ml macro-Kjeldahl flask (Thomas Scientific, USA) and mixed with 20 ml of distilled water. The flask was spun at 1,000 rpm for 15 min and permitted to rest for about 30 min. Two tablets of mercury catalyst (UNICAT Catalyst Technologies, USA) were added with 30 ml of conc. H_2SO_4 (Avondale Laboratories, UK). The flask was boiled carefully under controlled heat on the digestion stand till a pure digest was achieved and was heated for another 5 h. The flask was cool down and distilled water (100 ml) was gently added to it. Thereafter, 10 ml of the digest was placed into an uncontaminated 750 ml macro Kjeldahl flask. The residue was washed four times with 50 ml of distilled water and an aliquot was poured into the flask. About 20 ml of H_3BO_3 indicator solution (Fisher Scientific, USA) was added into a 250 ml erlenmeyer (conical) flask placed under the distillation apparatus with 750 ml Kjeldahl flask attached to the distillation apparatus. Then, 150 ml of 10 N NaOH was gradually emptied into the flask and permitted to cool below 30 °C, by letting enough cold water to run through and also modifiable heat to reduce bubbling and avert suck back. About 40 ml of the concentrate was collected and the distillation was stopped. To determine NH_4-H, the distillate was titrated using a 25 ml burette graduated at 0.1 ml intervals with 0.01 N standard HCl (Ademola and Abioye, 2017).

Nitrogen percentage was calculated by the formula below:

$\%Nitrogen= [(NA\times TV)\times(0.014\times DF)/volume\ of\ aliquot\times weight\ of\ sample)]\times100$

NA: normality of acid (0.01 N); TV: titer value; DF: dilution factor; volume of aliquot: 10 ml.

Fatty acid analysis

For about 5 min, 50 mg of extracted fat content of the sample was esterified (saponified) in dry methanol at 95 °C with 3.4 ml 0.5 M potassium hydroxide (KOH) and the mixture was neutralized with 0.7 M HCl. Thereafter, to attain a complete methylation process, the mixture was heated for 5 min at 90 °C with adding 3 ml of 14% boron trifluoride in methanol. Fatty acid methyl esters were removed from the mixture with redistilled n-hexane thrice (Rashid et al., 2008). The sample was concentrated to 1 ml prior to gas chromatography analysis and 1 µl was introduced into the injection port of the Hewlett Packard 6,890 Gas Chromatograph-Mass Spectrometer (GC-MS) attached with Hewlett Packard 5,973 mass spectroscopy detector (Botineștean et.al., 2012).

Metals test

One g of the sample was weighed into a crucible and ashed at 550 °C for 5 h in a muffle furnace and then cooled in a desiccator. The ashed samples were dissolved in 1 ml nitric acid (HNO_3) and 1 ml HCl, respectively and then made up to 100 ml. Afterward, metals were detected using Atomic Absorption Spectrophotometry (AAS), (Model UV

[DOI: 10.18502/jfqhc.10.2.12668]

4.1, Thermo Scientific, United Kingdon) (Iwegbu and Igene, 2022).

Microbial load determination

-Media

The media used were Eosin Methylene blue (EMB) (Neogen, USA), MacConkey Agar (Biolife, Nigeria), Potatoe Dextrose Agar (PDA) (Titan Biotech Ltd., India), Tryptone Agar (TA) (Biolife, Nigeria), and Nutrient Agar (NA) (Biolife, Nigeria). All the media were pre-stored according to the manufacturer's prescription.

-Isolation and identification of organisms

Five g of the sample (*R. phoenicis*) was milled aseptically, transferred into 45 ml of sterile distilled water, and then mixed thoroughly (Ebenebe and Okpoko, 2015). One ml of the dilution was transferred into another tube containing 9 ml of sterile distilled water to obtain 10^{-1}, the dilution protocol continued to obtain 10^{-9} (Braide and Nwaoguikpe, 2011). One ml was inoculated from dilution 10^{-6} into the sterile plate and the prepared sterilized media was poured on it (pour plate method). It was allowed to congeal and incubated at a temperature of 37 °C for 48 h (Nwaehujor et al., 2022). Colonies developed on the corresponding media plates were counted and used in estimating the bacterial loads in the sample after incubation (Nwaehujor et al., 2022). The bacterial count was done with the use of a digital colony counter (Labtech, model AVI-659, India). Cultural physiognomies, color change in media, and morphological features studied on culture plates were used to identify the bacteria (Hemalata and Virupakshaiah, 2016). Authentication of bacterial isolates identities were achieved by further referencing their physiognomies with those found in the Bergy's manual of determinative biology (Prescott et al., 2011).

Data analysis

Data were subjected to Microsoft excel to find the mean and standard deviation of triplicate samples. Statistics for Social Science Packages (SPSS) version 21 was also used for Pearson correlation ($p \leq 0.05$).

Results

The proximate analysis showed that *R. phoenicis* contained 45.60±3.31% weight of crude fat and 15.79±0.74% crude fiber. The MC was 22.00±2.55% while ash content, crude protein, and carbohydrate were 1.99±0.71, 5.25±0.30, and 9.36±0.18%, respectively.

The larvae of *R. phoenicis* contained nine fatty acids. Linoleic acid 54.13±0.001%, oleic acid 23.86±2.17%, and palmitic acid 14.191±0.15% had the highest fatty acid concentrations, respectively. Stearic acid concentration was 5.77±0.74%, arachidic acid 1.79±0.22%, myristic acid 0.09±0.03%, palmitoleic acid 0.05±0.01%, margaric acid 0.11±0.01%, and linolenic acid concentration was 0.005±0.00%.

The highest metal composition of *R. phoenicis* was iron (Fe) (4.923 ppm). Manganese (Mn) composition was 2.767±0.54 ppm, zinc (Zn) 1.038±0.10 ppm, lead (Pb) 0.003±0.00 ppm, and copper (Cu) composition was 0.291±0.09 ppm. However, chromium (Cr) and nickel (Ni) composition was measured 0.001±0.00 ppm. Cadmium (Cd) was not detected in the palm weevil larvae.

Microbes were only detected in the samples cultured with PDA, NA, and TA. The microbial count for PDA was 5×10^{-5} Colony Forming Unit (CFU)/ml and the organism isolated was bacterium. For NA, the count load was 33×10^{-5} CFU/ml and *Staphylococcus* was isolated. TA recorded 5×10^{-5} CFU/ml and *Micrococcus* and *Bacillus subtilis* were isolated. No fungus was observed in the sample.

Pearson correlation revealed that yeast and *B. substilis* had a significant relationship ($p<0.05$) with Ni, Cu, ash content, crude fat, and crude fiber. Meanwhile, *Staphylococcus* had a significant relationship ($p<0.05$) with Mn and Fe (Table 1).

Table 1: Pearson correlation of microbial load with metals and proximate contents

	Nickel (Ni)	Manganese (Mn)	Iron (Fe)	Copper (Cu)	Chromium (Cr)	Ash Content	Crude Fat	Crude Fiber
Yeast	0.991 **	0.317	0.167	0.883 *	0.991 **	0.899 *	0.901 *	0.888 *
p-value	0.001	0.603	0.788	0.047	0.001	0.038	0.037	0.044
Staphylococcus	0.623	0.977 **	0.933 *	0.854	0.623	0.082	0.834	0.849
p-value	0.262	0.004	0.021	0.065	0.262	0.895	0.079	0.069
Bacillus substilis	0.991 **	0.317	0.167	0.883 *	0.991 **	0.899 *	0.901 *	0.888 *
p-value	0.001	0.603	0.788	0.047	0.001	0.038	0.037	0.044

*: Correlation is significant at the 0.05 level (2-tailed).
**: Correlation is significant at the 0.01 level (2-tailed).

[DOI: 10.18502/jfqhc.10.2.12668]

Discussion

The crude protein value (5.25%) observed in the present study is not comparable to the mean value of 21.91% reported by Okoli et al. (2019) and is lower than the value of 10.51% (late larvae stage) reported by Omotoso and Adedire (2007) for the larvae of the same insect species collected from different locations in Bayelsa State, Nigeria and Igbokoda, Ondo State, Nigeria. These differences could be due to variations in the dietary habits of the insects, age (stage of development), or different ecotypes (Okoli et al., 2019). Nevertheless, the protein content of the *R. phoenicis* larvae recorded in this study could be a good source of dietary protein when compared with the protein content percentage (3.8-4%) of milk (Jacob et al., 2013; Onyeike et al., 2005). Likewise, Ehounou et al. (2019) stated that the protein content of *R. phoenicis* larvae (29.9 mg/100 g) collected from Ebimpé in Anyama, Côte d'Ivoire is higher than that of N'dama beef (22.16 mg/100 g) and tuna fish (25.56 mg/100 g) examined. However, the crude fat content in the present study was high and close to 50.23% recorded by Tang et al. (2019) in edible Lepidopteran and Heteropteran larvae and higher than 33.05 and 21.54% recorded in fresh and roasted *Rhychophorus ferruginous* larvae collected from Ondo State, Nigeria (Owolabi and Ajiboyede, 2022). The 15.35% crude fat content found in *Oryctes rhinoceros* obtained from Ese-odo area of Ondo State, Nigeria (Offiah et al., 2019) was lower than 66.61 and 62.15% present in fresh *R. phoenicis* larvae sourced from Akwa Ibom State, Nigeria and Warri, Delta State, Nigeria documented by Edijala et al. (2009) and Ekpo et al. (2009), respectively. This indicates that the insect could serve as an essential source of fat for developing countries that have low access to seafood and as dietary fat supplements in animal feed formulation for husbandry in developed countries (Hlongwane et al., 2020). Furthermore, the carbohydrate value was consistent with 6.71 and 15.98% carbohydrate content of sting bugs and cicada, respectively (Mlcek et al., 2014; Raksakantong et al., 2010), and higher than 7.82% carbohydrate content of *R. phoenicis* (Omotoso and Adedire, 2007). However, the value obtained in the present study was lower than that of (25.87%) in roasted larvae found by Edijala et al. (2009). In the present study, the carbohydrate content of *R. phoenicis* probably suggests that insects are an energy-giving food and should be used as food rather than a supplement (Ekpo et al., 2009). The ash content of 1% observed in this study was significantly lower than 8.02-11.83% obtained from insect larvae of *R. phoenicis* and *O. rhinoceros* in an earlier investigation (Offiah et al., 2019; Omotoso, 2015). The difference in the results might be due to differences in food substrate, insect developmental stage, and agro-ecological zone, as stated by Opara et al. (2012).

The physiological role of crude fiber in sustaining appropriate peristaltic movement of the intestinal tract cannot be underscored (Awobusuyi et al., 2020). Constipation and intestinal problems may result from a low-fiber diet (Bessa et al., 2020). The crude fiber content (15.79%) observed in the present study were higher than 0.2-3.3% reported by Banjo et al. (2006) and Ekop et al. (2010) from 14 dried common edible insects in South western Nigeria and *Gymnogryllus lucens*, *Heteroligus meles*, *R. phoenicis*, and *Zonocerus variegatus* collected from different locations in Akwa Ibom State, Nigeria, respectively. Also higher than 0.18 and 3.70% recorded by Owolabi and Ajiboyede (2022) in fresh and roasted *R. phoenicis* larvae collected from Okitipupa in Ondo State, Nigeria and 9.72% in *Oryctes rhinoceros* larvae sourced from Ese Odo area of Ondo State, Nigeria (Offiah et al., 2019). Hence, *R. phoenicis* based diets could help empty gastrointestinal content, control weight, reduce body fat, and also promote satiety when consuming small amounts of food. However, the high MC (22%) increases the risk of microbial contamination, deterioration of the larvae, and reduction of shelf life, except after a reduction from processing (Banjo et al., 2006).

Feng et al. (2017) reported that, unlike other animal fats, edible insect fats possess higher essential fatty acid content, which is necessary for the human body, and hence edible insect fat has a high nutritional value. The oleic acid value detected in this study was less than 35.05% mentioned by Tang et al. (2019) for *R. phoenicis* but higher than the oleic acid that can be found in beef (10.52%). Oleic acid belongs to the monounsaturated fatty acid family, and it is very important in the human diet because it helps reduce blood pressure and cure inflammatory, immune, and cardiovascular diseases (Sales-Campos et al., 2013). The highest value of fatty acid content recorded was linoleic acid (54.13%), indicating that the studied *R. phoenicis* larvae have an appreciable value compared to pork (7.29%), chicken (14.00%), and beef (36.10%). Linoleic acid is the major ingredient of polyunsaturated fatty acids in insects and has been proven to be anti-inflammatory, acne-reductive, and skin-lightening. With the risk of saturated fatty acids overtaking unsaturated fatty acids as food in pork and beef, matured larvae or adults of edible insects could be a better source of monounsaturated fatty acids and polyunsaturated fatty acids, which are healthier for the human diet (Tang et al., 2019).

R. phoenicis has been documented to have appreciable amounts of mineral elements such as calcium (Ca), phosphorus (P), magnesium (Mg), potassium (K), and sodium (Na), which are necessary for the human body system (Ehounou et al., 2019; Elemo et al., 2011). The ability of *R. phoenicis* larvae to contain metals and their amounts are of importance. All metals detected were in

[DOI: 10.18502/jfqhc.10.2.12668]

appreciable and moderate amounts in this study. Pb was detected and was not above the toxic level of 0.35 ppm (Kumar et al., 2020). This makes the insect ideal for consumption, but its accumulation in the human body over time could be of concern. The Fe amount recorded in this study was far below what Ehounou et al. (2019) and Tang et al. (2019) recorded for the same species. *R. phoenicis* larvae could serve as a Fe supplement for iron-deficient people, especially pregnant women. As Fe is very important in the functioning of cells and is one of the essential components of red blood cells (Ehounou et al., 2019), when it is above the required limit, it becomes toxic by increasing free radicals in the body or damaging the liver (Ekop et al., 2010). Similarly, Mn, Zn, and Cu prevent cardiomyopathy, muscle degeneration, growth retardation, impaired spermatogenesis, immunologic dysfunction, and bleeding disorders and strengthen the immune system (Akullo et al., 2018; Saris et al., 2000; Thomas, 2018). Zn is necessary for the proper functioning of the immune system. It is also essential in cell formation, wound healing, and the breakdown of glucose (Lin et al., 2018). The Mn content of the larvae in the study is relatively low compared to the 7.17 ppm in roasted larvae recorded by Owolabi and Ajiboyede (2022). Chaney (2006) stated that the presence of Mn in palm weevil larvae is a constituent of arginase and pyruvate carboxylase. Therefore, *R. phoenicis* larvae can supply necessary nutritive elements together with other foods that are rich in other essential minerals for healthy human diets and growth (Alamu et al., 2013).

Some insects act as vectors or submissive intermediate hosts of vertebrate pathogens such as bacteria, protozoa, viruses, or helminths (Reineke et al., 2012). It has been found that *R. phoenicis* carries an appreciable microbial flora, mainly bacteria. The bacterial flora seen in *R. phoenicis* included *Staphylococcus* spp., *Micrococcus,* and *B. substilis.* The Pearson correlation showed that there was a positive relationship between the isolated microbes and some metals, as well as their proximate contents. The presence of metals in *R. phoenicis* larvae lends credence to the findings of Tan (2022), who documented that metals such as Cd and Pb generally accumulate in soil and are absorbed by crops grown in such soil, which are in turn consumed by the insect feeding on such a plant or crop. There was a positive correlation between the isolated bacteria and the proximate and metal composition of *R. phoenicis* larvae, since the larvae are typically enriched in these mineral elements, thus providing a conceivably favourable environment for the microbes to live and breed (Tang et al., 2019). There is, however, no fear about the effects of this bacterium, since humans are healthy carriers of the organisms (Geraldine and Beth, 2008). They can also be found in fish, pork, meat, or any other food in most countries, including Nigeria. Moreover, processing under good hygiene would further reduce their presence (Banjo et al., 2006; Ebenebe and Okpoko, 2015).

Conclusion

The current study showed that *R. phoenicis* larvae could be a good source of dietary protein, fiber, carbohydrate, mineral elements such as Fe, Zn, and Mn, and moderately rich fatty acids. These findings revealed the potential nutritional quality of the larvae in mitigating nutritional deficiency and promoting good health. The relatively high nutrient value of *R. phoenicis* indicates its promising role in combating human nutritional imbalance and highlights the need for its incorporation in human diets as an alternate source of animal protein and essential fatty acids. It is, therefore, recommended that *R. phoenicis* mass rearing be encouraged and increased in order to bridge the animal protein supply gap in developing countries. The larvae in this study, on the other hand, have a high MC, and their short shelf life can be resolved with prompt processing. More attention should be focused on assessing the risk factors in the edible insect groups. This study did not examine the anti-nutritional factor and vitamin constituents of the larvae; therefore, further studies are recommended for the exploration of the larvae anti-nutritional factor and vitamin constituents.

Author contributions

J.M.A. conceived and designed the study, checked grammar, and proofread the manuscript; A.R.J. conceived, monitored the conduct of the study, and prepared the manuscript draft; K.D.I. analyzed and interpreted data, and conducted a plagiarism check; T.E.M-A. sourced for *R. phoenicis*, prepared the larvae sample for nutritional composition analysis and sourced for relevant pieces of literature; O.Y.K. carried out the microbial load estimation protocol. All authors read and approved the manuscript.

Conflicts of interest

The authors declare no conflict of interest.

Acknowledgements

This study did not receive any specific grant from funding agencies in the public, commercial, or non-profit sector.

References

Ademola O.A., Abioye O.R. (2017). Proximate composition, mineral content and mineral safety index of *Lablab Purpureus* seed flour. *International Journal of Science and Healthcare Research*. 2: 44-50.

[DOI: 10.18502/jfqhc.10.2.12668]

Akullo J., Agea J.G., Obaa B.B., Okwee-Acai J., Nakimbugwe D. (2018). Nutrient composition of commonly consumed edible insects in the Lango sub-region of northern Uganda. *International Food Research Journal*. 25: 159-166.

Alamu O.T., Amao A.O., Nwokedi C.I., Oke O.A., Lawa I.O. (2013). Diversity and nutritional status of edible insects in Nigeria: a review. *International Journal of Biodiversity and Conservation*. 5: 215-222. [DOI: 10.5897/IJBC12.121]

AOAC. (2012). Official methods of analysis. Association of Official Analytical Chemists, Washington D.C, USA.

Awobusuyi T.D., Pillay K., Siwela M. (2020). Consumer acceptance of biscuits supplemented with a sorghum–insect meal. *Nutrients*. 12: 895. [DOI: 10.3390/nu12040895]

Banjo A.D., Lawal O.A., Songonuga E.A. (2006). The nutritional value of fourteen species of edible insects in southwestern Nigeria. *African Journal of Biotechnology*. 5: 298-301. [DOI: 10.5897/AJB05.250]

Bessa L.W., Pieterse E., Sigge G., Hoffman L.C. (2020). Insects as human food; from farm to fork. *Journal of the Science of Food and Agriculture*. 100: 5017-5022. [DOI: 10.1002/jsfa.8860]

Botineștean C., Hădărugă N.G., Hădărugă D.I., Jianu I. (2012). Fatty acids composition by gas chromatography–mass spectrometry (GC-MS) and most important physical-chemicals parameters of tomato seed oil. *Journal of Agroalimentary Processes and Technologies*. 18: 89-94.

Braide W., Nwaoguikpe R.N. (2011). Assessment of microbiological quality and nutritional values of a processed edible weevil caterpillar (*Rhynchophorus phoenicis*) in Port Harcourt, southern Nigeria. *International Journal of Biological and Chemical Sciences*. 5: 410-418. [DOI: 10.4314/ijbcs.v5i2.72059]

Chaney S.G. (2006). Principles of nutrition II: micronutrients. In: Devlin T.M. (Editor). Textbook of biochemistry with clinical correlation. 6th edition. John Wiley and Sons, New York. pp: 1091-1120.

Chia S.Y., Tanga C.M., Van Loon J.J., Dicke M. (2019). Insects for sustainable animal feed: inclusive business models involving smallholder farmers. *Current Opinion in Environmental Sustainability*. 41: 23-30. [DOI: 10.1016/j.cosust.2019.09.003]

Ebenebe C.I., Okpoko V.O. (2015). Microbiological quality of raw and roasted African palm weevil (*Rhynchophorus phoenicis*) consumed in the south eastern Nigeria. *Animal Research International*. 12: 2159-2165.

Edijala J.K., Egbogbo O., Anigboro A.A. (2009). Proximate composition and cholesterol concentrations of *Rhynchophorus phoenicis* and *Oryctes monoceros* larvae subjected to different heat treatments. *African Journal of Biotechnology*. 8: 2346-2348.

Ehounou G.P., Ouali-N'goran S.-W.M., Soro D., Bedikou M.E. (2019). Nutrient contributions of *Rhynchophorus phoenicis* Fabricius, 1801 (Coleoptera: Curculionidae), very appreciated larvae in Côte d'Ivoire compared with beef (N'Dama breed) and thon (*Thunnus thynnus*). *International Journal of Biological and Chemical Sciences*. 13: 2092-2103. [DOI: 10.4314/ijbcs.v13i4.16]

Ekop E.A., Udoh A.I., Akpan P.E. (2010). Proximate and anti-nutrient composition of four edible insects in Akwa Ibom state, Nigeria. *World Journal of Applied Science and Technology*. 2: 224-231.

Ekpo K.E., Onigbinde A.O., Asia I.O. (2009). Pharmaceutical potentials of the oils of some popular insects consumed in southern Nigeria. *African Journal of Pharmacy and Pharmacology*. 3: 051-057.

Elemo B.O., Elemo G.N., Makinde M.A., Erukainure O.L. (2011). Chemical evaluation of African palm weevil, *Rhychophorus phoenicis*, larvae as a food source. *Journal of Insect Science*. 11: 146. [DOI: 10.1673/031.011.14601]

Feng Y., Chen X.-M., Zhao M., He Z., Sun L., Wang C.-Y., Ding W.-F. (2017). Edible insects in China: utilization and prospects. *Insect Science*. 25: 184-198. [DOI: 10.1111/1744-7917.12449]

Geraldine O.C., Beth A.M. (2008). Bacteria in the intestine, helpful residents or enemies from within?. *Infection and Immunity*. 76: 3360-3373. [DOI: 10.1128/IAI.00187-08]

Hemalata V.B., Virupakshaiah D.B.M. (2016). Isolation and identification of food borne pathogens from spoiled food samples. *International Journal of Current Microbiology and Applied Sciences*. 5: 1017-1025. [DOI: 10.20546/ijcmas.2016.506.108]

Hlongwane Z.T., Slotow R., Munyai T.C. (2020). Nutritional composition of edible insects consumed in Africa: a systematic review. *Nutrients*. 12: 2786. [DOI: 10.3390/nu12092786]

Iwegbu A., Igene U.F. (2022). Assessment of proximate composition, minerals and vitamins of bush buck (*Gongronema latifolia*) leaf meal and leaf extracts. *Canadian Journal of Agricultural and Applied Sciences*. 2: 1-16. [DOI: 10.5281/zenodo.7032773]

Jacob A.A., Emenike A.F., Kayode A., Olusegun O., Uzoma A., Rukayat K.Q. (2013). Entomophagy: a panacea for protein-deficient-malnutrition and food insecurity in Nigeria. *Journal of Agricultural Science*. 5: 2013. [DOI: 10.5539/jas.v5n6p25]

Johnson D.V. (2010). The Contribution of edible forest insects to human nutrition and to forest management. In: Durst P.B., Johnson D.V., Leslie R.N., Shono K. (Editors). Forest insects as food: humans bite back. FAO Regional Office for Asia and the Pacific, Bangkok, Thailand. pp: 5-22.

Kumar A., Kumar A., Cabral-Pinto M.M.S., Chaturvedi A.K., Shabnam A.A., Subrahmanyam G., Mondal R., Gupta D.K., Malyan S.K., Kumar S.S., Khan S.A., Yadav K.K. (2020). Lead toxicity: health hazards, influence on food chain, and sustainable remediation approaches. *International Journal of Environmental Research and Public Health*. 17: 2179. [DOI: 10.3390/ijerph17072179]

Lin P.-H., Sermersheim M., Li H., Lee P.H.U., Steinberg S.M., Ma J. (2018). Zinc in wound healing modulation. *Nutrients*. 10: 16. [DOI: 10.3390/nu10010016]

Mlcek J., Rop O., Borkovcova M., Bednarova M. (2014). A comprehensive look at the possibilities of edible insects as food in Europe – a review. *Polish Journal of Food and Nutrition Sciences*. 64: 147-157. [DOI: 10.2478/v10222-012-0099-8]

Nwaehujor I.U., Inana M.E., Azeke E.A., Okoroafor C.H., Abdulbaki M.K., Okike O.O., Nwachukwu E.F. (2022). Microbial and fungal contamination of staple foods in Port Harcourt, Nigeria: special attention to high aflatoxin risk. *Journal of Food Quality and Hazards Control*. 9: 190-198. [DOI: 10.18502/jfqhc.9.4.11374]

Offiah C.J., Fasalejo O.F., Akinbowale A.S. (2019). Evaluation of nutritional and anti-nutritional values of *Oryctes rhinoceros* larvae in Ondo State, Nigeria. *Journal of Entomology and Zoology Studies*. 7: 204-207.

Okaraonye C.C., Ikewuchi J.C. (2008). *Rhynchophorus phoenicis* (F) larva meal: nutritional value and health implications. *Journal of Biological Sciences*. 8: 1221-1225. [DOI: 10.3923/jbs.2008.1221.1225]

Okoli I.C., Olodi W.B., Ogbuewu I.P., Aladi N.O., Okoli C.G. (2019). Nutrient composition of African palm grub (*Rhynchophorus phoenicis*) larvae harvested from raphia palm trunk in the Niger-delta swamps of Nigeria. *Asian Journal of Biological Sciences*. 12: 284-290. [DOI: 10.3923/ajbs.2019.284.290]

Omotoso O.T. (2015). Nutrient composition, mineral analysis and anti-nutrient factors of *Oryctes rhinoceros* L. (Scarabaeidae: Coleoptera) and winged termites, *Marcrotermes nigeriensis* Sjostedt. (Termitidae: Isoptera). *Current Journal of Applied Science and Technology*. 8: 97-106. [DOI: 10.9734/BJAST/

[DOI: 10.18502/jfqhc.10.2.12668]

2015/15344]

Omotoso O.T., Adedire C.O. (2007). Nutrient composition, mineral content and the solubility of the proteins of palm weevil, *Rhynchophorus phoenicis* f. (Coleoptera: Curculionidae). *Journal of Zhejiang University Science B*. 8: 318-322. [DOI: 10.1631/jzus.2007.B0318]

Onyeike E.N., Ayalogu E.O., Okaraonye C.C. (2005). Nutritive value of the larvae of raphia palm beetle (*Oryctes rhinoceros*) and weevil (*Rhyncophorus pheonicis*). *Journal of the Science of Food and Agriculture*. 85: 1822-1828. [DOI: 10.1002/jsfa.2054]

Opara M.N., Sanyigha F.T., Ogbuewu I.P., Okoli I.C. (2012). Studies on the production trend and quality characteristics of palm grubs in the tropical rainforest zone of Nigeria. *Journal of Agricultural Technology*. 8: 851-860.

Oso A.A., Ashafa O.A. (2021). Nutritional composition of grain and seed proteins. In: Jimenez-Lopez J.C. (Editor). Grain and seed proteins functionality. Books on Demand, London, United Kingdom. pp: 31-50. [DOI: 10.5772/intechopen.97878]

Owolabi D.O., Ajiboyede O.K. (2022). Proximate and mineral analysis of red palm weevil *(Rhychophorus ferrugineus*) larvae. *International Journal of Entomology Research*. 7: 120-124.

Prescott L.M., Harley J.P., Klein D.A. (2011). Microbiology. 8th edition. WMC Brown, London.

Raksakantong P., Meeso N., Kubola J., Siriamornpun S. (2010). Fatty acids and proximate composition of eight Thai edible terricolous insects. *Food Research International.* 43: 350-355. [DOI: 10.1016/j.foodres.2009.10.014]

Rashid U., Anwar F., Moser B.R., Ashraf S. (2008). Production of sunflower oil methyl esters by optimized alkali-catalyzed methanolysis. *Biomass and Bioenergy*. 32: 1202-1205. [DOI: 10.1016/j.biombioe.2008.03.001]

Reineke K., Doehner I., Schlumbach K., Baier D., Mathys A., Knorr D. (2012). The different pathways of spore germination and inactivation in dependence of pressure and temperature. *Innovative Food Science and Emerging Technologies.* 13: 31-41. [DOI: 10.1016/j.ifset.2011.09.006]

Rumpold B.A., Schlüter O.K. (2013). Nutritional composition and safety aspects of edible insects. *Molecular Nutrition and Food Research*. 57: 802-823. [DOI: 10.1002/mnfr.201200735]

Sales-Campos H., Reis De Souza P., Crema Peghini B., Santana Da Silva J., Ribeiro Cardoso C. (2013). An overview of the modulatory effects of oleic acid in health and disease. *Mini-Reviews in Medicinal Chemistry*. 13: 201-210. [DOI: 10.2174/1389557511313020003]

Saris N.-E.L., Mervaala E., Karppanen H., Khawaja J.A., Lewenstam A. (2000). Magnesium: an update on physiological, clinical, and analytical aspects. *Clinica Chimica Acta*. 294: 1-26. [DOI: 10.1016/s0009-8981(99)00258-2]

Shantibala T., Lokeshwari R.K., Debaraj H. (2014). Nutritional and antinutritional composition of the five species of aquatic edible insects consumed in Manipur, India. *Journal of Insect Science*. 14: 14. [DOI: 10.1093/jis/14.1.14]

Stork N.E. (2018). How many species of insects and other terrestrial arthropods are there on earth?. *Annual Review of Entomology*. 63: 31-45. [DOI: 10.1146/annurev-ento-020117-043348]

Tan S. (2022). What are the health benefits of grasshoppers?. URL: https://www.webmd.com/diet/what-health-benefits-grasshoppers.

Tang C., Yang D., Liao H., Sun H., Liu C., Wei L., Li F. (2019). Edible insects as a food source: a review. *Food Production, Processing and Nutrition.* 1: 8. [DOI: 10.1186/s43014-019-0008-1]

Thomas C.N. (2018). Nutritional potentials of edible larvae of longhorned beetle (*Apomecyna parumpunctata Chev*.) (Coleoptera: Cerambycidae) in Niger delta, Nigeria. *International Journal of Agriculture and Earth Science.* 4: 46-51.

Xiaoming C., Ying F., Hong Z., Zhiyong C. (2010). Review of the nutritive value of edible insects. In: Durst P.B., Johnson D.V., Leslie R.N., Shono K. (Editors). Forest insects as food: humans bite back. FAO Regional Office for Asia and the Pacific, Bangkok, Thailand. pp: 85-92.

Yin W., Liu J., Liu H., Lv B. (2017). Nutritional value, food ingredients, chemical and species composition of edible insects in China. In: Mikkola H. (Editor). Future foods. Books on Demand, London, United Kingdom. pp: 27-54. [DOI: 10.5772/intechopen.70085]

Zabentungwa T.H., Rob S., Thinandavha C.M. (2021). Indigenous knowledge about consumption of edible insects in South Africa. *Insects*. 12: 22. [DOI: 10.3390/insects12010022]

[DOI: 10.18502/jfqhc.10.2.12668]