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FOOD SCIENCE & TECHNOLOGY | RESEARCH ARTICLE

Nutritional quality of wastes emanating from processing of genotypes of freshwater Prawn, *Macrobrachium vollenhovenii*

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Abstract: Wastes from processing of chelipede genotypes of *Macrobrachium vollenhovenii* were analysed for nutritional quality. The analysed genotypes were individuals possessing equal length of left and right sides arms/chelipedes, longer left side and shorter left side (EA, LL, and SL). Wastes (combined head, exoskeleton, walking appendages and chelipede) from production of fleshy tissues of each genotype were assessed for indices of nutritional quality, proximate composition (PC): dry matter—DM (% wet weight), crude protein (CP), crude fibre (CF), ether extract (EE), ash (AC), and carbohydrate (CHO), expressed in % DM, and *in vitro* digestibility (I_vD) at 3:1 sample to digestive enzyme (pepsin and trypsin). Data were analysed for differences ($p < 0.05$) across genotypes. Significant differences occurred in PC of genotypes' wastes. DM ranged between 35.70 ± 1.41 (SL) and 49.90 ± 0.28 (EA). The CP, CF, EE, AC and CHO ranges were: 40.53 ± 0.56 (LL)–53.73 ± 0.24 (SL), 7.64 ± 0.13 (LL)–10.98 ± 0.12 (SL), 7.11 ± 0.09 (EA)–9.02 ± 0.17 (LL), 11.36 ± 0.07 (EA)–19.56 ± 0.08 (SL) and 6.08 ± 0.03 (SL)–31.37 ± 0.56 (EA). The SL differed from EA and LL in 100%PC; EA differed from LL in 42.9% PC. Digestibility (pepsin plus trypsin): 41.7 ± 0.84% (LL), 52.86 ± 1.31% (EA) and 55.02 ± 0.53% (SL) were dissimilar, LL and EA digested better in pepsin, while SL digested better in trypsin. Wastes showed

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Dr Oyediran O. Oyebola is a lecturer in the Department of Aquaculture and Fisheries Management, University of Ibadan, Ibadan, Nigeria. I teach, conduct research and carry out community development in Aquaculture and Fisheries Management with special interest in fish genetics, breeding and biotechnology. The current research is part of the nutri-genetics and nutri-genomic aspect of my research activities in broad area involving application of genetics and biotechnology tools in aquaculture development and aquatic environments' fisheries resources identification, management, conservation, improvement and utilization. My research covers finfish and shellfish resources. The current research is a step further in finding sustainable use for wastes from shellfish industry, while assessing nutritional differences in the wastes from discovered genotypes of freshwater prawn, *Macrobrachium vollenhovenii.*

PUBLIC INTEREST STATEMENT

Prawns are common and populous shellfishes which serve nutritional and socio-economic roles in global communities. However, one of the main challenges of shellfish resources is the environmental and financial issues associated with disposal of wastes emanating from processing of their fleshy tissues. Therefore, for the purpose of utilization in human food supplement and or in livestock feed, the nutritional values of wastes processed from particular genotypes of freshwater prawn, *Macrobrachium vollenhovenii* were investigated, using chemical composition and digestibility as indices. It was found that the wastes had qualities of proteineous food (40.53–53.73% crude protein in dried form) and were digestible (41.7–55.02%). All the genotypes had desirable qualities for exploration in human and livestock nutrition. These information highlights food relevance of the wastes, thus paving the way for reduction in environmental and financial challenges of wastes disposal in the shellfish industry.

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qualities of proteineous food. However, differences in genotypes' nutritional qualities would influence their relative utilities.

Subjects: Bioscience; Nutrition; Food Science & Technology; Food Chemistry; Nutrition

Keywords: shellfish; nutritional utilization of wastes; wastes' digestibility; human and livestocks' food

1. Introduction

Prawns are among the commonest and most populous shellfishes. Diverse indigeneous species of the prawns are available in local global communities, where they serve nutritional and socio-economic roles. Fleshy tissues of the shellfishes are nutritious and they are essential export materials, especially in Asia countries. However, one of the main challenges of shellfish resources is the need to find sustainable means of disposal of wastes emanating from processing of the fleshy tissues. Every year, some 6–8 million tonnes of wastes are produced globally from shellfishes (Yan & Chen, 2015) and large quantity of wastes is produced from shrimps companies around the world (Udo & Opeh, 2013). An estimated 43% of shellfish production ends up as products for human consumption while the remaining percentages is classed as waste (Archer, Boutton, & Hibbard, 2001).

India and Bangladesh produce sizeable quantity of shellfishes, but with challenges of waste production. Processing of penaeid prawns resulted in more than 50% of the raw material as waste in India (Nair & Prabhu, 1989), while production of exportable frozen products through processing industries produced 40–80% wastes in Bangladesh, depending upon species and process (Irianto & Giyatmi, 1997; Suparno & Poernomo, 1992). These wastes, which comprise mainly the head, tail, vein/viscera and shell, are usually dumped outside the factory premises, paving the way for serious environmental pollution (Khan & Nowsad, 2012).

In developing countries, such as Africa, waste shells are often dumped in landfill or the sea, where they constitute environmental menace; shells disposal can cost up to US$150 per tonne in developed countries, such as Australia (Yan & Chen, 2015). To this effect, waste generation and disposal are increasingly penalized and international policies are now addressing sustainable production and utilization of fisheries resources in a manner that will reduce waste emissions to the environment. Depending on species, these wastes could be harnessed into useful products for human and livestock nutrition.

Macrobrachium vollenhovenii are indigenous freshwater prawn species of Africa. This shellfish is distributed in ponds, lakes, rivers, and irrigation ditches in West Africa (Udo & Opeh, 2013). The *M. vollenhovenii* is one of the largest species of *Macrobrachium* known (New, 2002), and it is of the highest commercial potential among the *Macrobrachium* species (Ajuzie & Fagade, 1992). The species has potential for commercial aquaculture and they hold promise as means to mitigate food insecurity, especially, in riverine communities (Marioghae, 1982), where they support artisanal fisheries in many developing countries in Africa (Nwosu & Wolfi, 2006). Awareness on aquaculture development of the freshwater prawn is increasing. However, its aquaculture development would increase waste generation, which would heighten the challenge of finding an avenue for management and utilisation or disposal of the wastes that would be generated.

Prawn wastes may vary based on diversity of forms, conditioned by genetic and or environmental factors. Earlier reports indicated that prawn wastes vary, depending on species and processing methods (Irianto & Giyatmi, 1997; Suparno & Nurcahya, 1984; Suparno & Poernomo, 1992; Suparno & Susana, 1984). An assessment of genetic diversity of *M. vollenhovenii* has revealed genotypic diversity in the species. A recent study (Oyebola, Jacob, Omorodion, Akinsola, & Akinade, 2016) reported that the genotypes of *M. vollenhovenii* had differences in chelipede morphology, which correlated with differences in allozyme fingerprints. Three strains were identified: those that possess shorter left compared to right arm/chelipede (SL); those with longer left compared to right sides' chelipede (LL); and those with equal length of left and right sides' chelipede. The SL was the earlier reported strain of the freshwater prawn (Bello-Olusoji, Bankole, Sheu, & Oyekanmi, 2006; Bello-Olusoji, Omolayo, & Arinola, 2004; Holthuis, 1980; Jimoh, Clarke, Whenu, Anetekhai, & Ndimele, 2012; Lawal-Are & Owolabi, 2012), while the rest were newly observed.

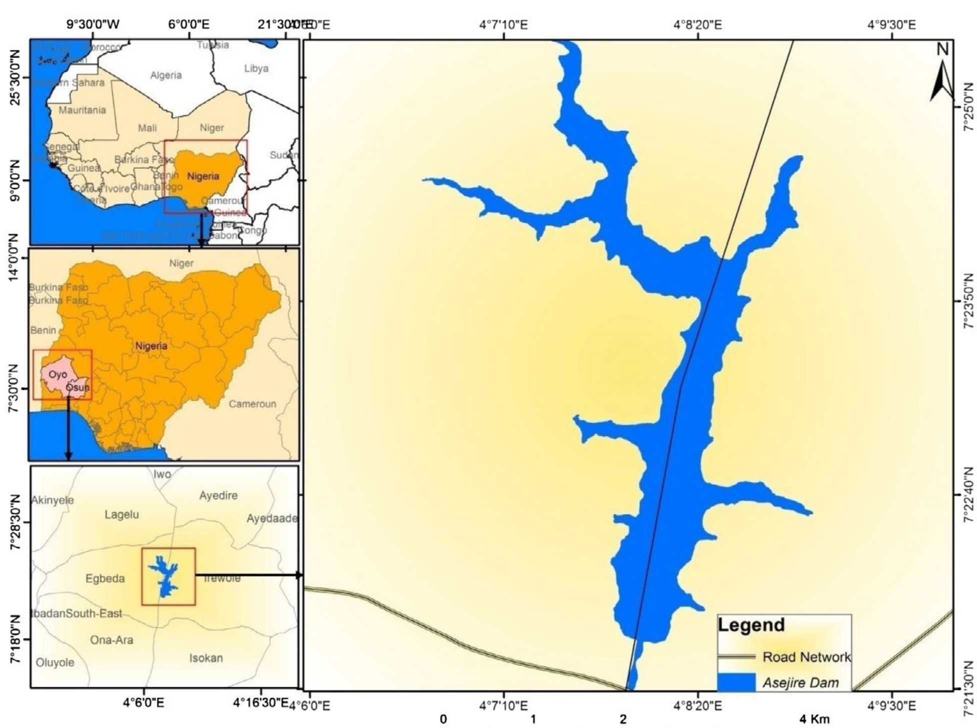
Nutritional assessment of the processed fleshy tissues from the genotypes showed that all the strains were of good quality as protein food for man, while the genotypes showed differences in proximate quality and *in vitro* digestibility of fleshy tissues (Oyebola, Adelaja, & Adelani, 2016). It is opined that availability of information on the nutritional quality of the strains would increase interest on aquaculture production of these strains with consequence on increased challenges of management and disposal of their wastes. The wastes from processing of the fleshy tissues of the genotypes could be of desirable quality for human consumption and or in livestock feeds, but this has to be established. There is dearth of information on the potential impact of the observed genetic variability in the species on the quality of their wastes, especially with respect to relevance for human and or livestock nutrition. Therefore, this study was carried out to evaluate the values and the differences in proximate composition and *in vitro* digestibility of wastes emanating from processing of the fleshy tissues of the three chelipede genotypes of *M. vollenhovenii*, with the intention of exploring the wastes for the purpose of human consumption or livestock feeds.

2. Materials and methods

2.1. Experimental site

Samples of freshwater prawn, *M. vollenhovenii* were collected from Asejire Lake, Nigeria and utilised for this study. Asejire Lake lies at the borderline between Oyo and Osun States of Nigeria, on latitude 04° 07′E and 07° 21′N at an altitude of 137 m above sea level. It is a major artificial dam constructed on River Osun which links the Ogun River and drains ultimately to the Lagos Lagoon in south western Nigeria. Asejire River is one of the series of West African rivers that do not drain into Niger system but discharge into coastal lagoons and creeks bordering the Atlantic Ocean (Omoike, 2004). The map of the sampling location, Asejire Lake, is presented in Figure 1.

Figure 1. Map showing the sampled location of the *M. vollenhovenii* genotypes, the Asejire Lake, Nigeria.



2.2. Sampling procedure

Samples were purposively collected from the Asejire Lake, based on the reported availability of the chelipede strains (Oyebola, Jacob, et al., 2016). Fresh and healthy captured samples of the freshwater prawn were collected from fisherfolks at the main landing site of the lake in June–October, 2015. Samples of *M. vollenhovenii* were transported inside iced containers to the Department of Aquaculture and Fisheries Management, University of Ibadan, Nigeria, where the genotypes were identified and separated to the genotypic sub-groups. Samples were separated to the chelipede genotypic subgroups following the morphologic and molecular description provided by Oyebola, Jacob, et al. (2016). Morphologically, samples possessing equal length of chelipede on the left and right sides, referred to as Equal Arm, were denoted as EA; those with longer left side, referred to as Long Left were denoted as LL; and those with shorter left side, referred to as Short Left, were denoted as SL. The LL, SL, EA were further identified following the earlier molecular descriptions using allozyme marker (12.5% SDSPAGE) in which allozyme fingerprints of LL were 18.4 and 50.0 kDa bands, while 20.0 kDa separated SL from EA. Samples of each of the genotypic groups were preserved in separate container and kept in freezer at −4°C prior to chemical analysis. An image of the chelipede genotypes is presented in Figure 2.

2.3. Sample preparation

Preserved samples of the LL, SL and EA (11.6–15.6 cm length and 22–32 g weight) were thawed and deveined (the exoskeleton was removed) to separate the fleshy edible tissues from the wastes. Pooled samples of the wastes from each of the genotypic subgroups were collected into separately containers. The wastes composed of the head, the exoskeleton, the walking legs and the chelipedes, thus following the earlier description by Adeyeye-Tayo and Ogunjobi (2008), but chelipedes were included in the current study. Image of the parts of the prawn that constituted the wastes are presented in Figure 3. The wastes from each of the genotypes were subjected to proximate analysis and *in vitro* digestibility test, following standard laboratory procedures.

Figure 2. Image of the Chelipede genotypes of *M. vollenhovenii* (Oyebola, Jacob, et al., 2016).

Notes: EA indicate the genotypes having equal length of left and right side chelipedes, LL indicate genotypes having longer left side chelipede, SL indicate genotypes having shorter left side chelipede.

Figure 3. Components of the analysed wastes (head, walking appendages, chelipede, telsons and carapace obtained from processing of *M. vollenhovenii*).

2.4. Proximate analysis

The wastes were chemically assessed for proximate values using the official methods of chemical analysis (AOAC, 2000). Samples were oven-dried at 105°C for 5 h to obtain dry matter (DM) content. Crude protein was analysed using the Kjeldahl method. Analysis of crude protein content followed digestion, distillation, and titration procedures to determine total nitrogen in sample (N), which was converted to percentage crude protein (N multiplied by a constant of 6.25). Samples were ashed in furnace at 600°C for 2 h to determine ash content. Crude fat content was assessed through the Soxhlet extraction method, following standard procedures. To determine the fibre content, samples were macerated, homogenized and digested in 100 ml of trichloroacetic acid digestion reagent (500 ml glacier acetic acid, 450 ml of water, 50 ml conc. Nitric acid), boiled, filtered in Whatman paper, dried overnight at 105°C, weighed and ashed at 600°C (overnight) in muffle furnace. Data were collected on dry matter; crude protein, crude fat, crude ash and crude fibre contents, expressed as percentage of dry matter (% DM). Carbohydrate was taken as difference between DM and the sum of values of other analysed parameters (Ehigiator & Oterai, 2012).

2.5. Determination of in vitro digestibility

The wastes were analyzed for *in vitro* digestibility at 3:1 (6 g of tissue sample: 2 g of digestive enzyme). Pepsin and trypsin are mostly used as simulate enzymes for *in vitro* digestibility tests (Joint FAO/WHO/UNU, 1985). Hence, pepsin and trypsin were purposively selected for the digestibility study, based on their respective role in gastric and duodenal digestion respectively. The gastric medium digestion was maintained at pH 2.5, while duodenal digestion was maintained at pH, 6.5, as described by Clare Mills et al. (2013). The enzyme treated tissue samples from each of the strains were digested following the standard laboratory procedures earlier described. Digestibility of tissues from each of the genotypes was determined separately for the pepsin and trypsin media.

2.6. Statistical analysis

Data were analysed using descriptive statistics: mean, standard deviation, and percentages. Data were compared across genotypes using one-way analysis of variance (ANOVA) of the PAST statistical software (Hammer, Harper, & Ryan, 2005).

3. Results and discussions

3.1. Proximate composition of wastes from processed chelipede genotypes of M. vollenhovenii

All the genotypes wastes showed potentials as proteineous food sources and they were significantly different ($p < 0.05$) with respect to their proximate composition (Table 1). The SL was significantly different from EA and LL in all analysed parameters, while EA and LL were dissimilar in 42.9%.

Table 1. Proximate quality of wastes of the chelipede genotypes of *M. vollenhovenii*

Parameters (%)	Genotype		
	LL	EA	SL
Moisture	53.92 ± 0.76[b]	49.14 ± 0.68[c]	62.48 ± 0.41[a]
Dry matter (DM)	44.90 ± 0.42[b]	49.90 ± 0.28[a]	35.70 ± 1.41[c]
Crude protein (CP)	40.53 ± 0.56[b]	41.18 ± 0.34[b]	53.73 ± 0.24[a]
Crude fibre (CF)	7.64 ± 0.13[b]	7.98 ± 0.04[b]	10.98 ± 0.12[a]
Ether extract (EE)	7.26 ± 0.43[b]	7.11 ± 0.09[b]	9.02 ± 0.17[a]
Ash content (AC)	13.39 ± 0.14[b]	11.36 ± 0.07[c]	19.56 ± 0.08[a]
Carbohydrate (CHO)	29.50 ± 0.42[a]	31.37 ± 0.56[a]	6.08 ± 0.03[b]

Notes: Means with the same superscript (a, b or c) along the same row are not significantly ($p > 0.05$) different.

LL = genotype having longer left sides chelipede, EA = genotype having equal length of left and right sides chelipede, SL = genotype having shorter left sides chelipede. CP, CF, EE, AC and CHO were expressed as % dry matter.

3.2. Dry matter content

Dry matter ranged from 35.70 ± 1.41 (SL) to 49.90 ± 0.28 (EA), and moisture contents were between 49.14 ± 0.68 (EA) and 62.48 ± 0.41% (SL). This indicated that the samples were high in moisture content. These values were lower than the 77.43% reported for moisture content in shrimp wastes (Nair & Prabhu, 1989). It is expected that shell-containing materials would not be high in moisture, since shell contains more of hardy calcium-containing materials. However, the high moisture in the samples could be a reflection of the contributions of the fleshy materials within the shells of the analysed wastes, because, fleshy tissues are usually locked up within the hard shells, especially in the chelipedes. The values were higher than the reported values in Ehigiator and Oterai (2012), in which 10.25 ± 0.08% moisture content occurred in exoskeleton and appendages, while 38.40 ± 0.32% moisture level occurred in the carcass (exuviate with the limbs, and other uneaten parts) of the same species. The difference in the reported values in the earlier and the current studies could be traced to diversity of waste composition, waste sources and phenotypic variations of the genotypes within the species. The physical or nutritional characteristics of an organism are referred to as phenotypes and this can be controlled by genetic factors conditioned by presence of genotypes.

The result of the current study showed that the samples for the *M. vollenhovenii*, which were obtained from the same habitat and composed of similar waste materials had significant differences in moisture content of wastes from their chelipede genotypes. Moisture content is inversely related to dry matter and this is reflected in the result of this study. Dry matter is an indication of meat yield after drying process. The values of dry matter followed the pattern EA > LL > SL. This pattern indicates that one of the novels, the EA, would have the highest dried fish meat yield, while the least would occur in the earlier reported strain, the SL. However, the pattern of dry matter content in the wastes ran contrary to the reported pattern in the fleshy tissues (Oyebola, Adelaja, et al., 2016), in which the LL had the superior dry matter content, while the EA had the lowest

3.3. Crude protein content

Proteins are an important component of food items, and crustacean shells are 20–40% protein (1). Wastes from all the chelipede genotypes of *M. vollenhovenii* showed desirable protein contents. The crude protein content was between 40.53 ± 0.56% DM and 53.73 ± 0.24% DM. These values were above the reported range presented by Yan and Chen (2015). High levels of protein, comprising 40–50% dry matter, were found in the shell wastes of shrimp (Khan & Nowsad, 2012; Nair & Prabhu, 1989); 44–52% crude protein occurred in shrimp's discard, (Revankar, 1978; Shahidi, 1994); and 50.3% crude protein were reported for processing waste powder of *M. rosengergii* (Babu & Nath, 2011). The crude protein content of the wastes from the chelipede genotypes of the analysed *M. vollenhovenii* compared favourably with the high protein content reported in the closely related species. This high level of protein indicate the potential for a wider use of the wastes from the *M. vollenhovenii* genotypes as proteineous food source for humans.

The potential for widened scope of utilization of valuable nutrient as functional nutritive component in human food, in addition to chitin and chitosan, have been reported for shrimp wastes (Yan & Chen, 2015). The protein content in wastes from shrimp and that of the analysed freshwater prawns' genotypes were similar. Hence, the wastes from the indigenous *M. vollenhovenii* could compete with the most popular and export valuable shellfish, the shrimp, as cheaper alternative sources of valuable protein nutrients as functional nutritive component in human food. The genotypes showed variation in their crude protein content. The LL and EA had similar protein contents, which were significantly lower than the value in SL. This indicates that, despite the good protein content in the wastes from all the genotypes, superior protein yield per dried waste sample is obtainable in the SL. This is interesting because the pattern was contrary to the values in the fleshy tissue, in which the EA was superior.

It is rational to expect that the two equal arms/chelipedes of the EA would yield more fleshy materials and this would contain more protein but the current result indicated a contrary trend. Hence, it would be of interest to understand which of the other proximate qualities actually contributed to the highest value of dry matter content (49.90 ± 0.28% wet weight) obtained in the EA. However, the result showed that, overall, the SL was superior with respect to derivable protein from its wastes.

3.4. Crude fibre content

Fibre has tangible contribution to tenderness of meat and it has relationship with digestibility of ingested food material. In this study, the obtained crude fibre was between 7.64 ± 0.13% DM and 10.98 ± 0.12% DM. These values occurred in LL and SL respectively. These values were higher than those of wastes from *M. vollenhovenii*, as reported in Ehigiator and Oterai (2012). The deviation in these values could be attributed to physiological factors emanating from composition of the wastes. Significant differences occurred in crude fibre content of the analysed genotypes; the LL and EA had similar values, which were significantly lower than the value in SL.

Conventionally, genetic and environmental factors could influence physiological variation in species, and these would reflect in expression of their phenotypic characteristics, such as the nutritional qualities. Since the chelipede genotypes were obtained from the same habitat, and were composed of similar materials, it could be inferred that the differences would mainly be as a result of physiological variations necessitated by variation in their genetic characteristics. However, the result indicated that the waste from SL would produce the least tender prawn wastes' meat and this would contribute to its digestibility; while the LL and EA would produce the most tender prawn wastes' meat with relatively low digestibility. The values of crude fibre content in the wastes were higher than the reported 2.40 ± 0.01% DM to 2.49 ± 0.16% DM obtained in the fleshy tissue (Oyebola, Adelaja, et al., 2016). The pattern of superiority of fibre content in the genotypes was also different, as EA > LL > SL occurred in fleshy tissue, while SL > EA > LL was obtained in pattern of crude fibre content in wastes from the genotypes.

3.5. Ether extract

Ether extract is an indicator of fat content in food substances. The values of ether extract obtained in the current study (7.11 ± 0.09% DM in EA and 9.02 ± 0.17% DM in LL) were higher than the one reported by an earlier study on *M. vollenhovenii* (Udo & Opeh, 2013), and the 2.8% DM fat, obtained in processing powder wastes of *M. rosenbergii* (Babu & Nath, 2011). However, it was similar to the values obtained in the head and maxillary case of shrimp waste (Khan & Nowsad, 2012). This indicated that the genotypes were rich in fat. The report is in agreement with the opinion that *M. vollenhovenii* seem to be rich in fat (Udo & Opeh, 2013).

Variation in lipid content in shrimp shell was related to seasonal variations, species, physiological status, diet as well as sexual maturity (Gordon & Roberts, 1977; Jacquot, 1961; Kinsella, 1988). The genotypes showed variation in the ether extract, indicating differences in lipid content. The variation in ether extract of the genotypes could be linked with any of the factors observed in the cases of the shrimps. However, the variation in fat content of the *M. vollenhovenii* genotypes compared to that of other shrimp wastes, especially that of *M. vollenhovenii*, reported in Gordon and Roberts (1977), may be due to any of the highlighted environmental and or genotype-induced phenotypic variations. The pattern of relationship between ether extracts of the genotypes' fleshy tissue was dissimilar to that of the wastes. The pattern SL > EA > LL occurred in fleshy tissues, while SL > LL > EA occurred in the wastes.

3.6. Ash content

Ash content is a gross indicator for availability of minerals in food compounds. The obtained values of ash content ranged from 11.36 ± 0.07% DM to 19.56 ± 0.08% DM. The result indicated that ash was high in all the analyzed wastes from the chelipede genotypes. Ash contents as high as 14.6–29.0% DM have been reported for shrimp waste discards (Revankar, 1978; Shahidi, 1994). The result of the current study is in agreement with the findings of these authors. However, these values were higher than the reported values by Udo and Opeh (2013) as well as Ehigiator and Oterai (2012), when wastes of *M. vollenhovenii* were analysed Ash contents were significantly different ($p < 0.05$) across the genotypes.

The lowest value (11.36 ± 0.07% DM) was obtained in EA, while the highest (19.56 ± 0.08% DM) occurred in SL. This followed the pattern: SL > LL > EA. However, the pattern was contrary to that of the fleshy tissues, in which EA > LL > SL was reported. Since ash content is an indicator of mineral content, the current result implies that the EA had comparatively low mineral content, while the SL would contain relatively higher minerals.

It is important to investigate the reasons for the lowest ash content obtained from the EA in the current study. This is necessary because the morphological feature of EA suggests that it would have comparative advantage in having more calcareous materials emanating from the mass of shell in its two long chelipedes, while the other genotypes would have comparatively smaller mass of shell due to the relatively smaller length of either the left or the right side chelipede. This is especially necessary in this case, in which EA had the highest dry matter content above the rest two genotypes. The low ash content in EA may indicate that the ash is not the main contributor to the superior dry matter content observed in it, just as it was observed that crude protein was not the main contributor to the superior dry matter content of the genotype. It is of interest to document the class of nutrient that contributed to the superior dry matter content of the EA.

3.7. Carbohydrate content

Carbohydrates are one of the most important sources of energy and dietary fibre in many foods, where they contribute to the sweetness, appearance and textural characteristics of the material. Indigestible carbohydrates form part of a group of substances known as dietary fibre. The proteins, polysaccharides and dietary fibre are insoluble in alcohol, whereas monosaccharides and oligosaccharides are soluble in alcoholic solutions. One of the most commonly used methods of extracting these low molecular weight carbohydrates is to boil a defatted sample with an alcohol solution. However, this approach was not followed in the current study, because of the desire to compare the findings of the study with that of Ehigiator and Oterai (2012), who studied the same species, in the same region. These authors determine the carbohydrate content of the freshwater prawn, *M. vollemhovenii* as the percent of dry matter remaining after the contributions from other components (crude protein, crude fibre, ether extract and ash contents) have been removed.

The wastes from the chelipede genotypes had high carbohydrate contents. The values were even higher than those of crude fibre in some instances. Crude fibre was between 6.08 ± 0.03% DM and 31.37 ± 0.56% DM, while 7.64 ± 0.13% DM to 10.98 ± 0.12% DM were the observed carbohydrate contents in the genotypes. Moreover, these high carbohydrate values were obtained after the crude fibre had been separately determined. The utilized analytical method did not involve specific protocol for extracting the low molecular weight carbohydrates, that is, the monosaccharides and polysaccharides. Therefore, it could be inferred that the deduced carbohydrates in this study, were the alcohol soluble class of carbohydrates that were not extracted by the fibre content analytic technique. Hence, wastes from processing of the *M. vollenhivenii* genotypes probably contained high concentration of carbohydrates of the alcohol soluble monosaccharides and polysaccharides classes. However, detailed analysis of the carbohydrate profile of the genotypes of *M. vollenhovenii* would be necessary in future research. It has been observed that carbohydrate can be as high as 54.5% DM in prawns (Babu & Nath, 2011). The carbohydrate contents of the assessed genotypes was lower than the 43.57 ± 0.75% DM carcass carbohydrate reported by Gordon and Roberts (1977), but it was much higher than the 0.58 ± 0.17 and 2.54 ± 0.56% DM obtained in combined exoskeleton and appendages, and in shells of *M. vollenhovenii* (Ehigiator & Oterai, 2012). The variation in these reports could be linked with the observed and discussed trends in the earlier sections on the other proximate qualities.

Also, the result revealed that significant differences occurred in carbohydrate content of the chelipede genotypes. This may imply that latent genotype-induced variations could have been responsible for the observed differences in these earlier reported carbohydrate content, especially, the reports on *M. vollenhovenii.* Carbohydrate content was significantly higher in EA and in LL when compared with the value in SL, thus following the pattern EA > LL > SL. The pattern in the flesh tissue was EA > SL > LL (Oyebola, Adelaja, et al., 2016). The high carbohydrate content of the wastes of EA compared to other genotypes indicates that carbohydrates may have been the main nutrient that contributed to the superior dry matter content in EA. However, these carbohydrates would be the low molecular weight classes of the monosaccharides and the polysaccharides.

The pattern of proximate quality of the wastes from the genotypes, as observed in this study, showed deviations from that of the reported pattern for their fleshy tissues, indicating the need for caution in generalizing the trend of proximate quality of the fleshy tissues of the genotypes for that of their wastes. Also, the fact that wastes from SL was significantly different from EA and LL in all analysed proximate qualities, while EA and LL were dissimilar in 42.9% of the parameters indicates the need to consider the genotypes as separate entities with respect to their food quality.

All the genotypes had desirable quality as proteineous food substance, and they had reasonable fat and crude fibre contents. However, relatively, SL was the most proteineous and mineralized genotype; LL was intermediate in most of the analysed proximate qualities; while EA was the meatiest, but the meat would contain relatively higher carbohydrate content of the low molecular weight class. This characteristic indicates the relative importance of the wastes of the genotypes. The qualities would have influence on their digestibility and scope of utility.

3.8. In vitro digestibility of wastes of the chelipede genotypes

The role of proximate analysis in understanding the nutritional quality of food items is well known, as it reveals the gross nutrient contents. However, assessment of proximate quality and digestibility of novel shellfishes would be of great importance for its acceptance as an ideal food for human consumption. Digestibility test provides information on availability of nutrients in food for utilization by host consumers. The food relevance of any substance can be jeopardised by poor digestibility of its chemical composition. Shellfishes are important proteineous food but they could be attached to some allergen (Yadzir et al., 2012).

Digestibility would reflect allergenicity and this would have influence on availability and assimilation of nutrients from food items. Result on *in vitro* digestibility of the wastes from the chelipede genotypes of *M. vollenhovenii*, is presented in Table 2. The result indicated that significant differences ($p < 0.05$) occurred in the digestibility of the wastes from the genotypes. Pattern of wastes digestibility in pepsin and trypsin media were also different (Figure 4).

Digestibility was highest in SL, implying that the superior nutritional composition of this genotype would be relatively more utilizable. It also showed a correlation with its superior fibre content. The EA and LL were statistically similar in most of the proximate qualities but dissimilar in digestibility value. Pattern of digestibility in EA was dissimilar to that of LL. The significant differences in their

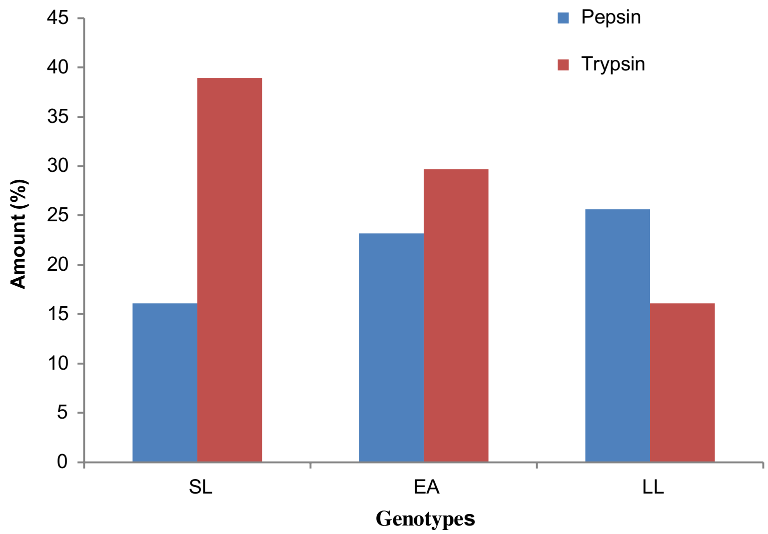
Table 2. Digestibility (%) of wastes of the chelipede genotypes of *M. vollenhovenii* in pepsin and trypsin digestion enzyme media

Parameters	Genotypes		
	LL	EA	SL
Pepsin	25.6 ± 0.56[a]	23.16 ± 0.33[b]	16.07 ± 0.32[c]
Trypsin	16.1 ± 0.28[c]	29.7 ± 0.98[b]	38.95 ± 0.21[a]
Total	41.7 ± 0.84[c]	52.86 ± 1.31[b]	55.02 ± 0.53[a]

Notes: Mean with the same superscript (a, b or c) along the same row are not significantly different ($p > 0.05$).

LL = genotype having longer left sides chelipede, EA = genotype having equal length of left and right sides chelipede, SL = genotype having shorter left sides chelipede.

Figure 4. Pattern of *in vitro* digestibility of the EA, SL, and LL genotypes' processing wastes in the pepsin and trypsin medium.



digestibility and variation in pattern of digestibility at the acidic and the alkaline media may be the cumulative effect of the statistically insignificant differences in some of the analysed proximate qualities. For instance, although not significant, the EA had higher values of crude protein and crude fibre than the LL, while the ether extract and crude ash were comparatively higher in LL than EA. These statistically insignificant values could have culminated in the differences in their digestibility in the media. The pattern of digestibility of the wastes from the genotypes in pepsin and trypsin medium is presented in Figure 4. Digestibility in pepsin was higher than digestibility in trypsin for LL; but for EA, digestibility in trypsin was higher than digestibility in pepsin. Pattern of digestibility was similar in EA and SL. This pattern indicate that EA and SL would have better gastric digestion while duodenal digestion would be better in the LL.

Based on these results, it can be deduced that the earlier reported variation in chelipede morphology which correlated with genomic (allozyme) difference (Oyebola, Jacob, et al., 2016) agreed with the differential nutritional qualities of the wastes emanating from processing of the genotypes. The assessed phenotypes; the nutrient content, and its digestibility in pepsin and trypsin media, were different across the genotypes. The observations of the current study agrees with the postulation of Mayr (1969), who reported that genetic differences in living organisms could result in variation in other phenotypic characteristics and such are often accidentally encountered in agriculture. The study also showed that the wastes were digested but at different extents. The conducted *in vitro* test showed the pattern of digestibility and possibility of allergenicity in the analysed wastes from the *M. vollenhovenii* genotypes. This confirmed the relevance of *in vitro* digestibility tests in assessment of food quality of the analysed wastes.

Knowledge on gastro-duodenal digestion gives information on how a protein is presented to immune system in a physiological context, while pepsin resistance reveals biochemical stability of a protein, which may be predictive of allergenic potential (Astwood, Leach, & Fuchs, 1996). The *in vivo* digestibility tests gives understanding on bio-availability of nutrient content in consumed food items, but the challenges of cost and time for conducting *in vivo* digestibility test has increased the relevance of the *in vitro* digestibility assay (Clare Mills et al., 2013; Mills, 2014). Application of *in vitro* digestibility test is based on the observation that *in vitro* values are comparable to values obtained in *in vivo* techniques (Chong, Hashim, & Ali, 2002; Eggum, 1989; Wolzak, Elias, & Bressani, 1981) and *in vitro* methods have been used in analysis of digestibility in plant tissues (Carbonaro, Carnovale, & Vecchini, 1993; Chiou, Ku, & Chen, 1997), animals tissues (Darcy, 1984; Desrosiers, Bergeron, & Savoie, 1987), fish feeds and ingredients (Ali, Haque, Chowdhury, & Shariful, 2009; Sultana, Ahmed, Iqball, & Chisty, 2010) and fish by–products (Smichi et al., 2016). It has been used in the assessment of genotype effects on digestibility of food items (Fageer & El Tinay, 2004) and the technique has also been found to be useful for rapid and reliable evaluation of protein digestibility in fish (Tonheim, Nordgreen, Høgøy, Hamre, & Rønnestad, 2007). The wastes of the chelipede genotypes of *M. vollenhovenii* were digested by the *in vitro* enzymes, indicating that their digestion would not be resisted in the stomach of the host consumer. Also, the result showed differential expression of digestibility of the waste tissues from the genotypes, thus further providing confirmation that *in vitro* digestibility tests are viable preliminary tool in analysing digestibility of food items from diverse genotypes of *M. vollenhovenii.*

4. Conclusion

The wastes emanating from processing the genotypes of *M. vollenhovenii* demonstrated valuable proximate quality as proteineous food for human consumption and they were digestible. The *M. vollenhovenii* genotypes have thus shown potential for extensive utility in food for human. It will also be useful as proteineous sources for livestock and fish feed.

5. Recommendations

It is recommended that the knowledge from the current study should be used in promoting the wastes as a viable nutritional supplement for human and livestock. The obtained knowledge is useful for wastes reduction, especially those emanating from processing of the crustacean, such as the *M. vollenhovenii.* The revealed nutritional and food attributes of the genotypes would facilitate their marketing and utilities in agriculture and agro-based industries. This would consolidate the basis for expansion of their aquaculture for food security through enhanced livestock and human nutrition, especially in developing countries.

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