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Abstract

Insects are potential ingredients for animal feed and human food. Their suitability may be influenced by species and nutritional value. This study was aimed at determining the nutritional profile of four insects: Dipterans; black soldier fly (Hermetia illucens Linnaeus) family stratiomyidae and blue calliphora flies (Calliphora vomitoria Linnaeus) family Calliphoridae; and orthopterans; crickets (Acheta domesticus Linnaeus) family Gryllidae and grasshoppers (Ruspolia nitidula Linnaeus) family Tettigoniidae to establish their potential as alternative protein sources for animals (fish and poultry) and humans. Gross energy, crude protein, crude fat, crude fiber, carbohydrates, and total ash were in the ranges of 2028.11-2551.61 kJ/100 g, 44.31-64.90, 0.61-46.29, 5.075-16.61, 3.43-12.27, and 3.23-8.74 g/100 g, respectively. Hermetia illucens had the highest energy and ash content; C. vomitoria were highest in protein and fiber content, R. nitidula were highest in fat, whereas A. domesticus had the highest carbohydrate content. All insects had essential amino acids required for poultry, fish, and human nutrition. The arginine to lysine ratios of H. illucens, C. vomitoria, A. domesticus, and R. nitidula were 1.45, 1.06, 1.06, and 1.45, respectively. The fatty acids comprised of polyunsaturated fatty acids (PUFAs) and saturated fatty acids (SFAs). Palmitic acid (23.6-38.8 g/100 g of total fat) was the most abundant SFA, exception R. nitidula with 14 g/100 g stearic acid. Linoleic acid (190-1,723 mg/100 g) and linolenic acid (650-1,903 mg/100 g) were the most abundant PUFAs. Only C. vomitoria had docosahexaenoic acid. The study indicates that the insects studied are rich in crude protein and other nutrients and can potentially be used for human and animal (fish and poultry) feeding.

Key words: insect, nutritional value, black soldier fly, blue calliphora fly, cricket

Insects are generally nutritious and therefore have potential for use in human and animal feeding (Klunder et al. 2012). They are rich sources of first-class protein (20-76 g/100 g dry matter), fat content (2-50 g/100 g dry basis), carbohydrates (2.7-49.8 mg per kg fresh weight), up to 70 g/100 g total fatty acids could be polyunsaturated fatty acids and minerals such as calcium, zinc, potassium, iron, manganese, and phosphorus (Kourimska and Adamkova 2016). For example, grasshoppers (Ruspolia nitidula Linnaeus) family Tettigoniidae contains 36-40 g/100 g crude protein, 41-43 g/100 g fat, 10-13 g/100 g dietary fiber, and 2.6-3.9 g/100 g ash on dry matter basis (Ssepuuya et al. 2016). Insects, therefore, have tremendous nutritional potential when either used as primary sources of human food or intermediate products such as animal feeds for poultry and fish. This study focused on insects recommended for food: R. nitidula and crickets [Acheta domesticus (Linnaeus) family gryllidae] and for animal feed: blue calliphora flies [Calliphora vomitoria (Linnaeus) family Calliphoridae], a close relative to the common house fly and black soldier fly (Hermetia illucens

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Linnaeus) family stratiomyidae (EFSA 2015). Insects that have been researched for use in animal feeds include the common house fly (Musca domestica), black soldier flies (Hermetia illucens), mealworms (Tenebrio molitor), locusts (Locusta migratoria, Schistocerca gregaria, Oxya spp., and others), and silkworms (Bombyx mori and others; Makkar et al. 2014, Stamer 2015, Veldkamp and Bosch 2015). Hermetia illucens pre-pupae are either commercially reared on organic waste such as kitchen food waste or naturally found in pig, poultry, and cattle manure waste (Veldkamp and Bosch 2015). Calliphora vomitoria are harvested from open damping sites or reared (Nakiyemba 2016). There is no competition for the use of H. illucens pre-pupae and C. vomitoria between humans and animals because these insect species are currently not consumed by humans. Acheta domesticus and R. nitidula can be directly consumed as human food. Although both A. domesticus and R. nitidula can be reared, R. nitidula are also seasonally abundant in African countries, especially Eastern Africa where they are obtained by harvesting from the wild (Kelemu et al. 2015).

Whereas the nutritional composition of *A. domesticus* has been widely researched, a few studies (Mbabazi et al. 2009, Ssepuuya et al. 2017) on the nutritional composition of *R. nitidula* have excluded the amino acid profile. Other studies (Kinyuru et al. 2010, Ssepuuya et al. 2019) have focused on *Ruspolia differens*, a close relative. Similarly, for feed insects, *Hermetia illucens* pre-pupae has been widely studied (Makkar et al. 2014, Spranghers et al. 2016), but there is no current research on the nutritional composition and quality of *C. vomitoria*. This study was, therefore, aimed at confirming the nutritional characteristics of some insects such as *A. domesticus* as well as determining the nutritional potential of unresearched/less researched insects such as *C. vomitoria* for use as food and feed.

Materials and Methods

Hermetia illuccens pre-pupae were reared on millet brew waste due to better colony performance and high protein content when compared with other substrates such as swine waste (Supp Tables S1-S3 [online only]). They were harvested at the pre-pupae stage (fifth-sixth instar). Adult C. vomitoria were harvested using bottle traps placed around garbage dumps in Makerere University in Kampala, Uganda, with decomposing chicken offal as the lure. Ruspolia nitidula were harvested from the wild using light traps in Masaka and Kampala districts in Uganda during the November-January season. Acheta domesticus were reared on a mixed feed containing banana peels, cassava peels, cassava leaves, sweet potato peels, and sweet potato leaves at inclusion levels of 18, 12, 43, 7, and 20%, respectively (Supp Table 3 [online only]) and then harvested as adults after 3 mo. For all the four insects, random sampling from the rearing/harvesting sites was done three times to obtain a total of 750-g representative laboratory samples.

Nutritional Analyses

Moisture content, crude protein, crude fat, ash, and crude fiber were determined by using methods described by AOAC (2012). Moisture content was determined by the draft oven method (934.01). Crude protein by the Kjeldhal method (976.06), crude fat by the Sohxlet method (991.36), crude fiber by the acid detergent fiber method (962.09), total ash by ashing in a carbolite furnace at 550°C for 2 h (942.05) and total carbohydrates determined by subtracting other proximate parameters from 100% (Reis et al. 2012). Gross energy was determined by the bomb calorimetry method as described by Smit et al. (2004).

Fatty acid composition was determined by preparation of fatty acid ester derivatives (Christie 1993) and the fatty acid methyl esters were analyzed by gas chromatography/mass spectrometry (GC/ MS) on a 7890A gas chromatograph (Agilent Technologies, Inc., Santa Clara, CA) linked to a 5975 C mass selective detector (Agilent Technologies, Inc., Santa Clara, CA) as elaborated by Musundire et al. (2016). Fatty acids were identified as their methyl esters by comparison of gas chromatographic retention times and fragmentation patterns with those of authentic standards and reference spectra published by the library-MS databases: National Institute of Standards and Technology 05, 08, and 11. Serial dilutions of the authentic standard octadecanoic acid (0.2-125 ng/µl) were analyzed by GC/MS in full scan mode to generate a linear calibration curve (peak area vs concentration) with the following equation: $[y = 7E + 06x - 4E + 07 (R^2 = 0.9757)]$, which was used for the external quantification of the different fatty acids.

Instrument Conditions

Inlet temperature was 270°C, transfer line temperature was 280°C, and column oven temperature programmed from 35 to 285°C with the initial temperature was maintained for 5 min and then increased by 10°C/min to 280°C and held at this temperature for 20.4 min. The GC was fitted with an HP 5MS low bleed capillary column (30 m × 0.25 mm i.d., 0.25 µm; J &W, Folsom, CA). Helium at a flow rate of 1.25 ml/min was used as the carrier gas. The mass selective detector was maintained at ion source temperature of 230°C and a quadrapole temperature of 180°C. Electron impact mass spectra were obtained at the acceleration energy of 70Ev. A 1.0-µl aliquot of sample was injected in the splitless mode using an auto sampler 7683 (Agilent Technologies, Inc., Beijing, China). Fragment ions were analyzed over $40-550 \, m/z$ mass range in the full scan mode. The filament delay time was set at 3.3 min. The amino acid profile was determined using the method described by Musundire et al. (2016). Chromatographic separation was done using a Waters ACQUITY UPLC (ultra-performance liquid chromatography) I-class system (Waters Corporation, Milford, MA). The UPLC was fitted with an ACE C18 column (250 mm × 4.6 mm, 5 μm, Aberdeen Scotland), with the heater turned off and the autosampler tray cooled to 5°C. Mobile phases used were water (A) and acetonitrile (B) each with 0.01% formic acid. Gradient elution was used at a constant flow rate of 0.7 ml/min. The injection volume was 1 µl. The following gradient was used: 0 min, 5% B; 0-3 min, 5-30% B; 3-6 min, 30% B; 6-7.5 min, 30-80% B; 7.5-10.5 min, 80% B; 10.5-13.0, 80-100% B, 13-18 min, 100% B; 18-20 min, 100-5% B; 20-22 min, 5% B. The flow rate was held constant at 0.7 ml/min. The injection volume was 1 μl. Leucine encephalin, a mass spectrometry standard was used as the reference compound.

Data Analysis

Data for proximate analysis, fatty acid profile, amino acid profile, and minerals were analyzed and presented as means \pm SD. Means for proximate analysis, fatty acid profile, and amino acid profile were analyzed using one-way analysis of variance to test for significant differences (P < 0.05). Means were separated using Tukey's honest significant difference (P < 0.05). The statistical package for social scientists' software (SPSS Inc., Released 2007, SPSS for Windows, Version 16.0., Chicago, IL) was used for data analysis.

Results

Proximate Composition

There were significant differences in the nutritional composition of insects commonly used for human food (*R. nitidula* and *A. domesticus*). Similarly, insects used for animal feeds (*H. illucens* and *C. vomitoria*) also had significant differences in their nutritional composition (Table 1). Gross energy values ranged from 2,028.11 kJ/100 g for *C. vomitoria* to 2,548.27 kJ/100 g for *H. illucens* prepupae. Dry matter content ranged from 26.44 g/100 g in *C. vomitoria* and *R. nitidula* to 47.71 g/100 g in *A. domesticus*. Crude protein ranged from 40.79 g/100 g in *R. nitidula* to 62.57 and 64.90 g/100 g in crickets and *C. vomitoria*, respectively. Crude fat content was lowest in *C. vomitoria* (0.67 g/100 g) and highest in *R. nitidula* (46.29 g/100 g). Crude fiber ranged from 5.07 to 8.04 g/100 g (crickets, *H. illucens* and grasshopper) to 16.61 g/100 g in *C. vomitoria*. Carbohydrates ranged from 3.43 g/100 g (*H. illucens* pre-pupae)

to 12.23–12.27 g/100 g (*C. vomitoria* and *A. domesticus*). Total ash ranged from 3.32 (*R. nitidula*) to 8.74 g/100 g (*H. illucens* pre-pupae).

Fatty Acid Profile

The fatty acid profile of *C. vomitoria*, *H. illucens* pre-pupae, crickets, and *R. nitidula* is summarized in Table 2.

The fatty acid composition varied significantly (P < 0.001) between the insect species for human food (R. nitidula and A. domesticus), except for lauric acid (F = 3.975; df = 3, 8; P = 0.053). Similarly, insect species for animal feed (C. vomitoria and H. illucens pre-pupae) had significant variations in fatty acid composition (P < 0.001) except palmitoleic acid (F = 3.203; df = 3, 8; P = 0.944), linolenic acid (F = 3.203; df = 3, 8; F = 0.0.896), and docosahexaenoic acid (DHA; F = 3.203; df = 3, 8; F = 1.0). The content of polyunsaturated fatty acids ranged from 950 mg/100 g (5 g/100 g of all fatty acids) in crickets to 3,626 mg/100 g (17 g/100 g of all fatty acids) in H. illucens pre-pupae.

The total amount of saturated fatty acids (SFA) ranged from 2,345 mg/100 g (*C. vomitoria*) to 8,846 mg/100 g (*H. illucens* prepupae). Palmitic acid, stearic acid, and myristic acid were the main saturated fatty acids detected in black soldier flies, *C. vomitoria*, *A. domesticus*, and *R. nitidula*. The main SFA in these insects was palmitic acid ranging from 5,860 mg/100 g (38.8 g/100 g of all fatty acids) in black soldier flies to 3,053 mg/100 g (25.2 g/100 g of all fatty acids) in crickets. However, the main SFA in *R. nitidula* was stearic acid (Table 2).

The concentration of total monounsaturated fatty acids (MUFAs) ranged from 2,638 mg/100 g (in *H. illucens* pre-pupae, constituting 12.3 g/100 g of the total fatty acid content) to 8,328 mg/100 g (*R. nitidula*, constituting 38.2 g/100 g of the total fatty acid content). The main monounsaturated acid (MUFA) in *A. domesticus*, *R. nitidula*, and *C. vomitoria* was oleic acid contributing 40.7, 62.7, and 45.0 g/100 g of the total fatty acid content, respectively, whereas that main monounsaturated acid in black solider fly pre-pupae was palmitoleic acid contributing 13.1 g/100 g of the total fatty acids (Table 2).

Hermetia illucens pre-pupae, C. vomitoria, crickets, and R. nitidula had appreciable amounts of alpha-linolenic acid ranging from 100 mg/100 g (C. vomitoria) to 1,903 mg/100 g (H. illucens pre-pupae). Only C. vomitoria contained DHA (Table 2).

Amino Acid Profiles

The amino acid content of *C. vomitoria*, *H. illucens* pre-pupae, *A. domesticus*, and *R. nitidula* is summarized in Table 3. All insects contained both essential and nonessential amino acids.

Twelve amino acids were determined including methionine, the most limiting for fish and poultry diets as well as those commonly limiting in human diets such as lysine and methionine Generally, there were significant (P < 0.001) differences in amounts of amino acids, except for proline (F = 2.726; df = 3, 8; P = 0.114), serine (F = 0.934; df = 3, 8; P = 0.468), and leucine (F = 0.897; df = 3, 8; P = 0.483) in the different insect species. The nonessential amino acid values ranged from 3.53 g/100 g (serine in H. illucens and

Table 1. Proximate composition of Calliphora vomitoria, Hermetia illucens pre-pupae, Acheta domesticus, and Ruspolia nitidula

Insect	Gross energy ¹ (kJ/100 g)	Dry matter	Proximate components (g/100 g)*					
			Crude protein	Crude fat	Crude fiber	Carbohydrates	Total ash	
Calliphora vomitoria	2,028.11 ± 9.6°	26.44 ± 0.4^{c}	64.90 ± 1.6°	0.67 ± 0.1^{d}	16.61 ± 2.4 ^a	12.23 ± 2.1 ^a	5.59 ± 0.4^{b}	
Acheta domesticus	$2,056.73 \pm 8.4^{b}$	47.71 ± 0.9^{a}	62.57 ± 0.1^{b}	12.15 ± 0.0^{c}	8.04 ± 0.4^{b}	12.27 ± 0.6^{a}	4.97 ± 0.2^{c}	
Hermetia illucens	$2,551.61 \pm 11.7^{a}$	36.29 ± 1.5^{b}	$44.31 \pm 0.3^{\circ}$	31.88 ± 1.3^{b}	5.07 ± 1.1^{b}	3.43 ± 0.4^{b}	8.74 ± 0.0^{a}	
Ruspolia nitidula	$2,069.53 \pm 6.7^{b}$	26.44 ± 0.4^{c}	40.79 ± 0.3^{d}	46.29 ± 0.2^{a}	5.88 ± 0.4^{b}	3.73 ± 0.7^{b}	3.32 ± 0.1^{d}	
Leuven stat (P value)	0.888	0.341	0.111	0.085	0.200	0.228	0.163	

Values are means \pm SD. Means with different superscripts in the column are significantly different (P < 0.05). In all cases, P < 0.0001 for ANOVA, n = 3. ¹Dry matter basis.

Table 2. Fatty acid profile of Calliphora vomitoria, Hermetia illucens pre-pupae, Acheta domesticus, and Ruspolia nitidula

Fatty acids	Fatty acid content (mg/100 g) in different insect species (dry basis)					Leuven stat
	Hermetia illucens	Calliphora vomitoria	Acheta domesticus	Ruspolia nitidula	0.053 <0.0001 <0.0001 <0.0001 0.018 <0.0001	(P value)
Lauric acid (C12:0)	63.0 ± 8.9^{a}	160.0 ± 20.0^{a}	170.0 ± 80^{a}	100.0 ± 30.0^{a}	0.053	0.189
Myristic acid (C14:0)	2526.0 ± 443.0^{a}	$128.0 \pm 43.0^{\circ}$	1410.0 ± 330.0^{b}	$500.0 \pm 23.0^{\circ}$	< 0.0001	0.156
Palmitic acid (C16:0)	5860.0 ± 160.0 a	$2,067.0 \pm 503.0^{b}$	$3053.0 \pm 508.0^{\circ}$	0.0 ± 0.0^{d}	< 0.0001	0.202
Stearic acid (C18:0)	397.0 ± 26.0^{a}	563.0 ± 32.0^{a}	1057.0 ± 136.0^{b}	$1745.0 \pm 221.0^{\circ}$	< 0.0001	0.192
Palmitoleic acid (C16:1)	1983.0 ± 880.0^{a}	797.0 ± 100.0^{ab}	547.0 ± 188.0^{b}	751.0 ± 89.0^{b}	0.018	0.109
Oleic acid (C18:1)	655.0 ± 511.0^{a}	$3,933.0 \pm 702.0^{b}$	4927.0 ± 980.0^{b}	$7577.0 \pm 626.0^{\circ}$	< 0.0001	0.823
Linoleic acid (C18:2)	1723.0 ± 407.0^{a}	0.0 ± 0.0^{b}	190.0 ± 59.0^{b}	$760.0 \pm 130.0^{\circ}$	< 0.0001	0.110
Linolenic acid (C18:3)	1903.0 ± 145.0^{a}	$1,000.0 \pm 330.0^{b}$	760.0 ± 20.0^{b}	650.0 ± 140.0^{b}	< 0.0001	0.235
DHA	0.0 ± 0.0^{b}	96.0 ± 27^{a}	0.0 ± 0.0^{b}	0.0 ± 0.0^{b}	< 0.0001	0.052
Total saturated	8,846.0	2,918.0	5,690.0	2,345.0		
Total unsaturated	6,264.0	5,826.0	6,424.0	9,738.0		
Total MUFA	2,638.0	4,730.0	5,474.0	8,328.0		
Total PUFA	3,626.0	1,096.0	950.0	1,410.0		

Values are means ± SD (n = 3). Means with different superscripts in a row are significantly different (P < 0.05). DHA (docosahexaenoic acid); MUFA (monounsaturated fatty acids); PUFA (polyunsaturated fatty acids).

Table 3. Amino acid profile (% crude protein) of Calliphora vomitoria, Hermetia illucens pre-pupae, Acheta domesticus, and Ruspolia nitidula)

Amino acids	Amino ac	P values	Leuven stat (P value)			
	Hermetia illucens	Calliphora vomitoria	Acheta domesticus	Ruspolia nitidula		
Essential						
Lysine	3.88 ± 0.00^{b}	5.83 ± 0.00^{a}	5.78 ± 0.82^{a}	3.88 ± 0.37^{b}	0.001	0.095
Valine	3.19 ± 0.01^{a}	2.64 ± 0.00 ab	2.52 ± 0.35^{b}	3.19 ± 0.3^{a}	0.012	0.124
Methionine	26.26 ± 0.03^{a}	7.83 ± 0.00^{b}	7.83 ± 1.11^{b}	26.60 ± 2.57^{a}	< 0.0001	0.096
Tyrosine	9.69 ± 0.01^{a}	9.69 ± 0.00^{a}	9.72 ± 1.38^{a}	0.20 ± 0.00^{b}	< 0.0001	0.052
Isoleucine	19.01 ± 0.01^{a}	19.01 ± 0.01^{a}	19.23 ± 2.75^{a}	7.90 ± 0.76^{b}	< 0.0001	0.079
Leucine	24.01 ± 0.03^{a}	22.03 ± 0.01^{a}	22.32 ± 3.19^{a}	24.07 ± 2.35^{a}	0.483	0.117
Phenylalanine	7.68 ± 0.00^{a}	3.61 ± 0.00^{b}	3.52 ± 0.59^{b}	7.68 ± 0.74^{a}	< 0.0001	0.116
Nonessential						
Arginine	5.61 ± 0.00^{a}	6.16 ± 0.00^{a}	6.12 ± 0.43^{a}	5.62 ± 0.54^{a}	0.152	0.116
Glutamine	6.04 ± 0.00^{a}	5.06 ± 0.00^{a}	4.99 ± 0.70^{a}	6.04 ± 0.58^{a}	0.031	0.117
Serine	3.53 ± 0.0^{a}	3.87 ± 0.00^{a}	3.78 ± 0.53^{a}	3.53 ± 0.33^{a}	0.468	0.113
Glutamic acid	4.61 ± 0.01^{a}	6.10 ± 0.00^{b}	6.05 ± 0.86^{b}	4.61 ± 0.44^{a}	0.006	0.103
Proline	7.04 ± 0.00^{a}	8.15 ± 0.00^{a}	8.15 ± 1.16^{a}	7.04 ± 0.68^{a}	0.114	0.106

Values are means \pm SD (n = 3). Means with different superscripts in a row are significantly different (P < 0.05).

R. nitidula) to 8.15 g/100 g (proline in *C. vomitoria*). Essential amino acids ranged from 0.2 g/100 g (tyrosin) to 26.6 g/100 g (methionine) in *R. nitidula*. The four insects were mainly rich in methionine, leucine, isoleucine, tyrosine, and phenylalanine.

Discussion

Determining the nutritional composition of potential food and feed ingredients is an important step in developing dietary recommendations to prevent or treat malnutrition. Prior to this study, there was limited research into the nutritional composition of some edible insects and thus their evaluation for use as food and feed in Uganda.

According to Mlcek et al. (2014), the energy values of insects are high but vary with insect species and locality as shown in this study for differences among species. The obtained energy values for R. nitidula (2,069.53 kJ/100 g) and A. domesticus (2,056.73 kJ/100 g) are comparably higher than 1,783.64 kJ/100 g reported for orthoptera (grasshoppers, locusts, and crickets; Rumpold and Schlüter 2015). This could be attributed to environmental conditions such as geographical location, as the insects were reared/harvested in Uganda. Gross energy is mainly influenced by the macronutrients (protein and fat) composition of the insect, which is also influenced by other factors such as diet and sex (Kulma et al. 2019). However, the studied insects (R. nitidula and A. domesticus) were collected without considering the sex of individual insects. The high fat content of H. illucens pre-pupae (31.88 g/100 g) and grasshoppers (46.29 g/100 g) explains the high gross energy values. In contrast, for C. vomitoria and A. domesticus, the high energy values can only be explained by the high protein values, 64.9 and 62.57 g/100 g, respectively. Because insects are poor sources of carbohydrates, the contribution of carbohydrates to gross energy values is negligible. Ruspolia nitidula and A. domesticus contained more energy than conventional animal protein sources such as lean fried beef and pork, 213 and 974.87 kJ/100 g, respectively, and a wide range of livestock products (690.36-2,949.72 kJ/100 g; Heinz and Hautzinger 2010, Mlcek et al. 2014). Therefore, R. nitidula and A. domesticus could make great contributions to human dietary energy intake in addition to high-quality animal protein such as beef, pork, fish, and others; however, for insects whose gross energy content comes mainly from fat, there is need to regulate fat consumption to avoid fat-related

health problems such as cardiovascular diseases. It is also imperative to note that the chitin content of insects may not be fully utilized for energy, but rather useful in reducing the glycemic load when insects are eaten with other foods.

In animal production, low energy intake affects growth and productivity. For example, in laying hens restricted energy intake reduces egg size, rate of egg production, and weight gain (Kingori et al. 2014). In swine production, energy is regarded as the most expensive feed ingredient that influences carcass characteristics such as lean or fatty pork (Velayudhan et al. 2015). Edible insects are high in energy; most of which comes from their high protein content. Velayudhan et al. (2015) demonstrate that high protein feeds for swine increase energy retention (net energy) in form of lean meat up to the genetic capacity of the animal beyond which fat will be deposited. Therefore, high protein ingredients such as edible insects could help in lean pork production provided feeds are formulated to meet growth and production requirements of swine.

The energy values for *C. vomitoria* (2,028.11 kJ/100 g) and *H. illucens* pre-pupae (2,551.61 kJ/100 g) were higher than for conventional protein sources used in animal feed; for instance, cyprinid fish meal and high-quality Chilean low-temperature (anchovy) fish meal contain 2,011.25 and 2,013.34 kJ/100 g, respectively (Maina et al. 2002). Insects such as *C. vomitoria* and *H. illucens* pre-pupae, which are not consumed by humans, could be a potential ingredient in animal feeds.

Regarding crude fiber content, reared *H. illucens* pre-pupae, and harvested *R. nitidula* values were close to 5.06–13.56 g/100 g for Isoptera and Hemiptera (Rumpold and Schluter 2013) but higher than those of fish meal (0 g/100 g) and cyprinid fish meal (0.9 g/100 g; Maina et al. 2002, Abowei and Ekubo 2011).

Monogastric animals including fish and humans cannot digest crude fiber (Delbert 2010). However, in humans, consumption of fiber confers health benefits especially along the digestive tract such as prevention of colon cancer, constipation, alleviation of symptoms of irritable bowel syndrome and reduction of the risk for cardiovascular diseases (Anderson et al. 2009, Ottles and Ozgoz 2014). Ottles and Ozgoz (2014) recommended a daily dietary fiber intake of 28 and 36 g/day for adult women and men, respectively. The insects analyzed in this study contained 5.07–16.61 g/100 g of crude fiber. Therefore, consumption of 100 g of *A. domesticus* and *R. nitidula*

meals can provide 16 and 28.7% of the daily requirement for fiber, respectively.

In fish feeds, the maximum fiber inclusion is normally 7% to limit the amount of indigestible material (Delbert 2010). Dietary fiber intake may also reduce bioavailability of some minerals such as calcium except for highly fermentable fibers that improve mineral bioavailability (Ottles and Ozgoz 2014). The fact that *H. illucens* pre-pupae and *C. vomitoria* had higher crude fiber contents than fish meal (Maina et al. 2002, Abowei and Ekubo 2011) is an indication that insect meal could be less digestible than fish meal. Therefore, care has to be taken when formulating animal feeds to ensure optimal inclusion levels of insect meal for minimal effects on mineral bioavailability and digestibility.

As far as proteins are concerned, they influence the growth of humans and animals as well as productivity of animals such as chicken and fish. They play a key role in synthesis of body tissue, enzymes, and hormones (Beski et al. 2015). It is therefore important to evaluate the protein content of edible insects to determine whether the insects can provide adequate protein for human and animal (poultry and fish) feeding. The crude protein content of *R. nitidula* and *A. domesticus* are relatively similar to values earlier reported for Orthoptera (crickets, grasshoppers, and locusts; Rumpold and Schluter 2013). The crude protein content of the adult *C. vomitoria* is comparable to that of house fly larvae meal (40–60 g/100 g; Makkar et al. 2014), whereas that of *H. illucens* is in agreement with values reported by Diener et al. (2015).

Edible insects (*R. nitidula* and *A. domesticus*) have more protein than beans (23.5 g/100 g), lentils (26.7 g/100 g), and soy (35.5 g/100 g) moreover with all the essential amino acids present (Ramos-Elordy et al. 2012, Rumpold and Schluter 2015). In comparison with fresh edible portions of cattle and fish products (11–28 g/100 g protein; Bernard and Womeni 2017), the crude protein content of *A. domesticus* (41 g/100 g) and *R. nitidula* (23.5 g/100 g) on fresh weight basis were higher or comparable, respectively. Moreover, digestibility of insect protein is comparable to conventional animal protein sources such as beef, pork and others (Kinyuru et al. 2010). EFSA (2017) recommends adults to consume 0.66 g/kg body weight of protein per day. Thus, human protein requirements can be satisfactorily met by most edible insects (Rumpold and Schluter 2013), including *R. nitidula* and *A. domesticus*.

Production animals also require adequate protein levels in the diet for growth and productivity. Insects intended for use as feed for fish and poultry (H. illucens and C. vomitoria) were generally rich in protein. The crude protein content of C. vomitoria was similar to that of fish meal (60-80 g/100 g) and higher than 45-50 g/100 g of soy, whereas that of H. illucens pre-pupae was comparable to 45-50 g/100 g reported for soy meal (Sánchez-Muros et al. 2014). Therefore, C. vomitoria and H. illucens pre-pupae could substitute the expensive fish meal and soy meal. It is imperative to note that the protein content of the studied insects could have been affected by the rearing substrates for the reared types (Spranghers et al. 2016) and time/season of harvesting for the harvested types (Ssepuuya et al. 2017). For example, H. illucens pre-pupae fed on different substrates (millet brew waste, rotten ovacado fruits, chicken house waste, swine dung, and bovine dung) had protein contents ranging from 38.62 for ovacado waste to 55.71 g/100 g for swine dung (Supp Table 2 [online only]). Millet brew waste was used for H. illucens pre-pupae in this experiment because of its availability in Uganda, good performance in terms of pupae protein composition (Supp Table 2 [online only]) and its relatively high protein content (Supp Table 1 [online only]). Therefore, it is important to optimize rearing conditions for the reared insect types (A. domesticus and H. illucens pre-pupae) and also

identify the best harvesting seasons for the harvested types (*C. vomitoria* and *R. nitidula*) to ensure consistently high protein supply.

Insects are also good sources of lipids the lipid contents of A. domesticus, R. nitidula, and black soldier flies were higher than 5.41-36.87 g/100 g reported for queen caste (Raksakantong et al. 2010), while that of harvested C. vomitoria was much lower than values reported for other insects. Such variations could be attributed to variations such as sex and the substrates on which these insects were fed (Kulma et al. 2019). Ruspolia nitidula had the highest lipid content on dry basis ($46.29 \pm 0.2 \text{ g}/100 \text{ g}$) followed by black soldier flies (31.88 \pm 1.3%). EFSA (2017) recommends that fat intake in adults should contribute between 20 and 35% of total energy intake. Therefore, this implies that they could greatly contribute to the energy requirements of humans and animals. Notably, the rearing conditions of insects such as feeding substrates need to be optimized to ensure consistent nutritional findings. The crude fat content of H. illucens falls within the range of 33-35 g/100 g reported by (Diener et al. 2015) for H. illucens pre-pupae. In comparison, the insects evaluated in this study, with the exception of C. vomitoria, had much higher fat contents than fish meal (3.5 g/100 g), anchovy (9.6 g/100 g), and cyprinid fish meal (12 g/100 g), respectively (Maina et al. 2002, Abowei and Ekubo 2011). However, there is a need to optimize feeding conditions of the domesticated insects to match the essential fatty acid profile of fish oil and essential fatty acid requirements such as omega 3 in terms of proportion for humans and animal feeding, as illustrated by Oonincx et al. (2019).

Regarding the carbohydrate content of the studied insects (3.43–12.27 g/100 g), is rather low compared with the main sources of energy for humans and animals such as maize, wheat, and rice. Notably though, the key source of energy in edible insects is fat.

The carbohydrate content of *C. vomitoria*, adult *A. domesticus*, reared *H. illucens* pre-pupae, and *R. nitidula* on dry basis lies within the range of 6.71 g/100 g for long stink bug to 15.98 g/100 g for cicada reported for edible insects (Raksakantong et al. 2010). Fish meal has 1.5 g/100 g carbohydrates, which is lower than that provided by *C. vomitoria*, *H. illucens* pre-pupae, and *A. domesticus* (Abowei and Ekubo 2011). Carbohydrates provide energy for metabolism in both humans and animals; however, the contribution of insects to dietary carbohydrate intake cannot sustain carbohydrate requirements of both animals (fish and poultry) and humans. Therefore, it is advisable to supplement insects with good carbohydrate sources such as cereals.

With respect total ash, the percentage total ash values on dry basis of harvested C. vomitoria, A. domesticus, reared H. illucens pre-pupae, and harvested R. nitidula falls within the reported range (2.94-25.95 g/100 g) for edible insects (Rumpold and Schluter 2013). The observation that H. illucens pre-pupae had the highest percentage of total ash is consistent with the findings of Finke et al. (2013) who evaluated the composition H. illucens larvae, tebo worm larvae, Turkestan cockroach nymphs, and adult house flies. However, values of total ash obtained in this study are lower than that of cyprinid fish meal (17.5 g/100 g) and anchovy fish meal (15.3 g/100 g) reported by Maina et al. (2002), thus implying that the insects evaluated could be low in mineral content However, the suitability of these insects for human and animal (fish and poultry) nutrition will depend on the presence of individual minerals such as iron, zinc, and calcium in sufficient quantities. Therefore, there is a need to investigate the presence of individual minerals in edible insects.

The content of polyunsaturated fatty acid (PUFA), MUFA, and SFA observed in this study is consistent with results reported previously by Yang et al. (2006), Elagbo (2015), and Bophimai and Siri (2010). Results of saturated fatty acids are consistent with those

of Yang et al. (2006). Results for MUFAs are similar to those of Yang et al. (2006), with only two MUFAs identified (palmitoleic acid and oleic acid). The concentration of total MUFAs ranged between 2683 mg/100 g in H. illucens pre-pupae to 8533 mg/100 g in R. nitidula. The amount of MUFAs in the analyzed insects lies within 714 to 5,889 mg/100 g for other insects (Yang et al. 2006) except R. nitidula with higher quantities. Linoleic acid and palmitic acid are among the dominant fatty acids in the insects evaluated in this study, consistent with data for cockroaches, tebo worm, and house flies (Finke et al. 2013). Black soldier flies have the highest levels of lauric acid, which is consistent with the findings by Finke et al. (2013) when compared with other insects including tebo moth, cockroach nymphs, and house flies. The main MUFA in A. domesticus, R. nitidula, and C. vomitoria was oleic acid consistent with the findings of Yang et al. (2006), Bophimai and Siri (2010), and Elagbo (2015) for other insects.

The ratios of saturated to unsaturated fatty acids were 1.24, 0.50, 0.97, and 0.24 for *H. illucens* pre-pupae, *C. vomitoria*, *A. domesticus*, and *R. nitidula*, respectively. Elagbo (2015) reported a ratio of 0.7 for edible migratory locust, which confirms that most insects have more PUFAs than SFAs (Finke et al. 2013). Therefore, despite the observed high total fat content in some edible insects, there could be less risk for cardiovascular diseases as most of the fat comprise healthy PUFAs.

Generally, terrestrial insects do not contain docosa hexanoic acid (DHA) and eicosapentanoic acid (EPA) (Bophimai and Siri 2010, Tran et al. 2015, Twining et al. 2016) but contain their molecular precursor alpha-linolenic acid, which is either converted into tissue or long-chain PUFAs to a minor degree by terrestrial insects (Torres-Ruiz et al. 2007, Bophimai and Siri 2010). However, the presence of DHA in *C. vomitoria* in this study could be attributed to the possibility of the insects having fed on a variety of substrates in the wild containing DHA (ST-Hilaire 2007, Torres-Ruiz et al. 2007). This implies that if edible insects are fed on EPA- and DHA-rich substrates, they could reduce deaths due to cardiovascular conditions in humans.

For human feeding, the ratio of omega-6 to omega-3 fatty acids should be less than 4 (Scollan et al. 2003, Simopoulos 2008). The recommended ratio of polyunsaturated to saturated fatty acids (P/S ratio) for humans should be above 0.4 to reduce risks for cardiovascular disease, cancer, asthma among other diseases (Milicevic et al. 2014). Therefore, direct consumption of edible insects could provide a better balance of fatty acids essential for optimal health in humans.

In animal nutrition, consumption of omega-3 fatty acids results in improved animal health and production of healthier foods. In poultry, for example, omega-3 fatty acids improve disease resistance by moderating immune reactions and improving specific immunity (Pike 1999). The long-chain omega-3 fatty acids are also subsequently deposited in chicken products such as the eggs and meat, which are channeled into human diets. Modification of the fatty acid profile of animal products through the diet to match human targets could improve the quality of animal products such as poultry meat (Mlcek et al. 2014). Both C. vomitoria and black soldier fly pre-pupae contain linoleic and linolenic acids, which domestic hens, fresh water, and some marine fish are able to convert into DHA and EPA using elongase and desaturase enzymes (Kalakowska 2011, Hixson et al. 2015, Twining et al. 2016). Therefore, insects could greatly contribute to healthy fat requirements for humans either directly through consumption as food (A. domesticus and R. nitidula) or indirectly through consumption of fish and poultry fed on insects (C. vomitoria and black soldier fly pre-pupae).

Regarding the amino acid profile, edible insects are rich in both the essential and nonessential amino acids. The amount of essential amino acids in edible insects is generally higher than those found in conventional animal protein sources. For example, the amount of lysine and methionine obtained for *C. vomitoria* and pre-pupae (meant for fish and poultry feeds), *A. domesticus* and *R. nitidula* (meant for direct human consumption) were higher than those of beef meat (1.94 and 0.61 g/100 g), pork meat (0.59 and 1.8 g/100 g) and chicken meat (1.79 and 0.69 g/100 g; Amadi and Kiin-Kabari 2016). Because insects contain demonstrable high amounts of essential amino acids, they could replace the expensive skimmed milk in ready to use therapeutic foods (RUTF) for malnourished children. However, further research is needed to evaluate the technological potential of edible insects to replace the expensive RUTF.

Moreover, the amino acid profile of edible insects matches the essential amino acid requirements for animal feeding. The studied insects more than make up for the limiting amino acids in fish and poultry, which include cysteine, lysine, and arginine and methionine (Finke 2002). For example, the arginine content of black soldier fly pre-pupae (5.61) and R. nitidula (5.62) are slightly lower than that of fish meal (5.82), and arginine content of C. vomitoria (6.16) and A. domesticus (6.12) are higher than that of fish meal (5.82; Abowei and Ekubo 2011). In terms of amino acid balance for poultry and fish, edible insects are superior to fish meal. For example, the arginine to lysine ratio of fish meal is 0.74, which is lower than 1.18 and 0.84 recommended for leg horn chicks and cat fish, respectively (National Research Council 1993, 1994). The arginine to lysine ratios of H. illucens pre-pupae, C. vomitoria, A. domesticus, and R. nitidula obtained were 1.45, 1.06, 1.06, and 1.45. These values are higher than 0.74 for fish meal. Arginine to lysine ratios of H. illucens pre-pupae and R. nitidula were higher than 1.18 and 0.84 recommended for leg horn chicks and cat fish respectively, whereas arginine to lysine ratios of C. vomitoria and A. domesticus were higher than 0.84 required by cat fish and slightly lower than 1.18 required by leg horn chicks. The amino acid results indicate that insect meal is rich in essential amino acids critical for fish and poultry optimal growth and performance as well as human growth and maintenance.

In conclusion, H. illucens pre-pupae, C. vomitoria, A. domesticus, and R. nitidula are rich in protein and fat, most of the essential amino acids and fatty acids required for fish and poultry as well as humans. These insects, therefore, have the potential for utilization in human and animal feeding. Acheta domesticus and R. nitidula, which are already accepted for human consumption, can be used for production of value-added products such as packaged insects and insect meal for formulation and direct addition to food. Hermetia illucens pre-pupae and C. vomitoria can be recommended for use as a cost-effective alternative protein source in animal feed. There is, however, a need to determine the economic feasibility of producing the insects in quantities enough to justify their use as food. Furthermore, there is a need to evaluate the safety and/or develop processing protocols that ensure safe insect meal for human and animal feeding. For the less studied insects, especially C. vomitoria, more research is needed to evaluate their safety for feed as well as need for optimal rearing conditions other than wild harvesting. Finally, there is a need to develop and optimize suitable rearing protocols for the insects that were harvested from the wild, to avoid potential negative impacts of wild harvesting on the ecosystem.

Supplementary Data

Supplementary data are available at Journal of Insect Science online.

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