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Evaluation of cockroach (*Periplaneta americana*) powder as a potential feed ingredient for ruminants: chemical composition, fatty acids profile and ruminal degradability

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Abstract This experiment was aimed at determining the chemical composition, fatty acid (FA) profile and degradability of American cockroach (*Periplaneta americana*) powder (ACP) in comparison with soybean meal (SBM), fish meal (FM), and poultry byproduct meal (PBM). The cockroaches were stored for 2h at -20 °C, transferred to liquid nitrogen and subsequently grinded. Other samples were dried at 60 °C for 48 h. Experimental diets were: (1) control diet (only SBM), 2) diet containing 3% FM, 3) diet containing 3% ACP, and 4) diet containing 3% PBM. Two fistulated Holstein heifers were used for estimation of the ruminal degradability of protein sources and experimental diets. The results indicated that the ACP contained 55.05, 24.55, 3.76, 8.68, and 5.60% crude protein (CP), ether extract, ash, and neutral and acid detergent fiber, respectively. The ACP was rich in monounsaturated and polyunsaturated FAs. There were significant differences in dry matter (DM) and CP degradability among protein sources. The degradability of soluble fraction (a) of SBM and ACP was significantly higher than other protein sources. The potentially degradable DM (b) for SBM was significantly higher. The CP washable fraction 'a' was significantly higher for FM and PBM. In contrast, the SBM contained larger 'b' which was smaller in FM and PBM. The estimated effective degradability of CP at all rumen passage rates was significantly higher in ACP than other protein sources. No significant differences were observed between the experimental diets in DM degradability coefficients (a, b and c). The control and ACP diets contained higher CP fraction 'b' than PBM diet. This experiment clearly showed that the ACP can be a good source of protein and mono-unsaturated fatty acids for ruminants.

Keywords: American cockroach, protein source, degradability, fatty acid

Introduction

With the high rate at which the world population is growing, food production may not meet demand (Wang et al., 2005). The search for fast and innovative feed solutions to improve the sustainability of the livestock sector and to provide sufficient food for the world's growing population in a more sustainable way is a major global challenge for the near future (FAO, 2014). A-

ccordingly, high quality protein is needed to sustain livestock production (Beski et al., 2015). Animal-based proteins such as fish meal (FM) and animal by-products are valuable feedstuff with high digestibility, but they are associated with cost fluctuations, pathogenic contamination, and environmental impacts. Therefore, plant-based proteins are used, but they have the disadvantages of inapp-

ropriate amino acid profiles, anti-nutritional factors, and mycotoxin contamination (Kovitvadhi et al., 2019).

In this perspective, insects have been addressed as a possible alternative feed for animals. The production of insects as feed has interesting characteristics. Indeed, they generate low greenhouse gas and ammonia emissions, have a favorable feed conversion ratio as cold-blooded animals, and require less water and soil to grow (Makkar et al., 2014). For instance, in one day, some of insect larvae can reduce 30 tons of food waste to 10 tons, while producing 930 kg of dry biomass (Salomone et al., 2017). Moreover, they can provide animal feed bio-converting food wastes thus ultimately not competing with humans for natural resources (Diener et al., 2011; Makkar et al., 2014). Edible insects have been suggested as a potential sustainable alternative source for livestock feed because they are a source of energy, protein, fat, minerals, and vitamins, and cause comparably low environmental impact (Bovera et al., 2016). Mean estimates show energy levels to be around 400–500 kcal per 100 g of DM, making it comparable with other protein sources (Payne et al., 2016). Protein is a significant component of edible insects, comprising between 30% and 65% of the total DM. Over the past years, edible insects have gained recognition for their potential as an alternative protein source (Van Huis, 2016; Ojha et al., 2021). After protein, fat is another main component of insects. The insect unsaturated fatty acids (FA) profile is similar to poultry and white fish but contains more PUFAs than either poultry or red meat (Rumpold and Schluter, 2013). In addition to the nutritional value, the insect-based feed could have a further advantage in improving the taste of final meat products (Schiafone et al., 2017).

The cockroach species *Periplaneta americana*, commonly known as the American cockroach, is considered edible amongst most cockroach species especially in countries like China where they are bred in captivity, sold and supplied to farmers who use them as livestock feed (Sikkema, 2015; Sule et al., 2020). According to Sule et al. (2020), American cockroach proximate composition contained crude protein 53.10±0.09%, fat 10.56±0.11%, fiber 11.69±0.23%, ash 8.37±0.13%, and metabolizable energy 1.48±0.093 MJ kg⁻¹. According to Jiang et al. (2012), various research works reported *P. americana* to have a variety of pharmacological attributes such as being analgesic, anti-viral, anti-tumor, anti-inflammatory, improving immunity and promoting tissue repair. Recent studies indicated that insect meal can be an excellent replacement for FM or SBM in animal feed (Biasato et al., 2019; Iaconisi et al., 2017; Onsongo et al., 2018; Van der Fels-Klerx et al., 2018). Therefore, this study was designed to compare the chemical and digestive properties of American cockroach with common protein sources used in animal nutrition.

Materials and methods

Protein sources

Native adult American cockroaches were obtained from an artificial rearing insect farm (Arvinmealworm, Mashhad, Iran). All live insects were starved for about 24 h to clear their gastrointestinal tract of any residual food. The insects were then stored for 2 h in a freezer at -20 °C. Next, the frozen insects were transferred to liquid nitrogen and subsequently grinded using a blender (LBC15 laboratory model, USA). The frozen-grinded insects were freeze-dried to stable weight and the moisture content was determined. Insects were stored at -20 °C for later use. Fish meal was obtained from southern Iran (Jask Young Fishermen Co., Hormozgan, Iran), poultry byproduct meal (PBM) from Gonbad city (Qaboos Co., Golestan, Iran), and SBM was Brazilian produce. Samples of protein sources were dried at 60 °C for 48 h, grinded to pass through a 1-mm sieve (Wiley mill) for FA and chemical analysis, and the 2-mm size for ruminal *in sacco* incubation, and then stored at -20 °C.

Experimental diets

Four isocaloric and isonitrogenous diets (Table 1), including; (1) control diet (containing 18% SBM), 2) diet containing 3% FM, 3) diet containing 3% ACP, and 4) diet containing 3% PBM, were formulated according to NRC (2001). In fact, the protein sources in diets 2, 3 and 4 replaced SBM at 3% level.

Chemical analysis

The DM, crude protein (CP), ash, and ether extract (EE) were measured based on AOAC (2005), and NDF and ADF according to Van Soest et al. (1991). Analysis of FAs was carried out according to IUPAC (1979) using an Autosystem Gas Chromatograph (3400 Varian Star; Varian Inc., Palo Alto, CA) equipped with CP-SIL-88 capillary column (60 m×0.25 mm, Varian) with helium as the carrier gas. The samples were incubated at 100 °C for 60 min and extracted in 5 mL of hexane. The column temperature was initially 50 °C for 1 min, which was increased by 4 °C/min to 190 °C. The injector and detector temperatures were 280 °C and 300 °C, respectively.

In sacco measurements

Ruminal *in sacco* degradation of the protein sources and experimental diets was carried out according to Orskov and McDonald (1979) to measure the kinetics of DM and CP degradation.

The 2-mm grinded samples were used for rumen degradation measurement and subjected to standard rumen degradability procedures using two fistulated Holstein heifers (approximately 400 kg live weight). The cows were fed a TMR diet containing 1.8 kg alfalfa hay, 1.8 kg concentrate, 0.5 kg corn silage, and 1.8 kg wheat straw twice a day (0800 and 1600 h). The cows had free access to fresh water and mineral salt licks. Dacron bags (10 × 15 cm) with 45–50 µm pore size containing appro-

ximately 4 g samples were incubated, in duplicate, in each heifer for each of the testing time periods: 0, 2, 4, 8, 16, 24, 48, 72, and 96 h. At the end of each incubation time, the bags were removed from the rumen and washed under running tap water until the rinsing water was colorless (approximately 1 min). Zero-time disappearances (washing losses) were obtained by washing unincubated bags in a similar fashion. The bags were then dried in an oven at 60 °C for 48 hours. Degradability (P) of DM and CP was calculated using the equation of Orskov and McDonald (1979):

$$P = a + b(1 - e^{-ct})$$

where, *P* is the disappearance of DM and CP during time *t*, *a*: soluble fraction which is rapidly washed out of the bags, *b*: insoluble but potentially degradable fraction, *c*:

the degradation rate of fraction *b* per hour, *t*: degradation time, and *e*: base for natural logarithm.

Statistical analysis

The General Linear models procedure of SAS (2003) was used to determine statistical differences between protein sources or experimental diets as a completely randomized design. The Tukey's test was used to compare the means. Effects were considered significant at $P < 0.05$. Data were analyzed using the following statistical model:

$$Y_{ij} = \mu + T_j + e_{ij}$$

where, Y_{ij} is the dependent variable, μ : the overall mean, T_j : treatment effect, and e_{ij} : residual error.

Table 1. Ingredients and chemical composition of the experimental diets fed as total mixed ration

	Experimental diets [†]			
	C	FM	ACP	PBM
Ingredients (% of DM)				
Alfalfa hay	20.0	20.0	20.0	20.0
Corn silage	20.0	20.0	20.0	20.0
Barley grain	10.0	10.0	10.0	10.0
Corn grain	20.0	20.0	20.0	20.0
Whole cottonseed with lint	5.0	5.0	5.0	5.0
Soy bean meal	18.0	14.0	14.5	14.5
Fish meal	0.0	3.0	0.0	0.0
American cockroach powder	0.0	0.0	3.0	0.0
Poultry byproduct meal	0.0	0.0	0.0	3.0
Wheat bran	5.5	6.5	6.0	6.0
Calcium carbonate	0.5	0.5	0.5	0.5
Vitamin-mineral Mix [‡]	0.8	0.8	0.8	0.8
Salt	0.2	0.2	0.2	0.2
Chemical composition				
Metabolizable energy (Mcal/kg DM)	2.45	2.46	2.47	2.46
Crude protein (%)	17.2	17.2	17.2	17.2
Ether extract (%)	4.4	4.4	3.6	3.5
Neutral detergent fiber (%)	31.0	31.0	31.2	31.3
Acid detergent fiber (%)	19.6	19.6	19.6	19.8
Non-Fiber carbohydrates (NFC) [§] (%)	43.2	43.3	43.1	43.7
Calcium (%)	0.9	0.9	0.9	0.8
Phosphorus (%)	0.6	0.6	0.6	0.5

[†]C: control (based soybean meal, FM: diet containing 3% fish meal, ACP: diet containing 3% American cockroach powder, PBM: diet containing 3% poultry by-product meal.

[‡] Contained (/kg of premix): 330,000 IU of vitamin A, 60,000 IU of vitamin D, 1,000 IU of vitamin E, 160g Ca, 85g P, 63g Na, 45g Mg, 2,100 mg Zn, 1,500 mg Mn, 535 mg Cu, 12 mg Se, 45 mg.

[§]NFC: calculated as 100 – (CP + Ash + NDF + EE).

Results

Chemical composition of protein sources

A large variation in chemical composition between the protein sources was found in this study (Table 2). The DM content was not different. The highest OM level was recorded in ACP and PBM, and the lowest one in FM. The CP content of The FM contained the highest CP level and SBM the lowest ($P < 0.01$). There was no significant difference between the CP value of ACP and PBM. The EE content of ACP was significantly highest among the protein sources. The SBM contained the lowest EE concentration. The NDF and ADF values were

the highest in SBM and the lowest in PBM. The ash content was the lowest in ACP and PBM and the highest in FM ($P < 0.01$).

Fatty acid profile of protein sources

The FA profile was significantly ($P < 0.01$) affected by the protein sources (Table 3). Myristic, palmitoleic and palmitic acid concentrations were highest in FM, and pentadecanoic and palmitoleic acid in PBM. The lowest percentage of stearic acid was found in ACP and SBM. Vaccenic acid percentage was highest in SBM and PBM, and lowest in ACP and FM. The highest percentage of oleic acid (40.5 %) was recorded in ACP, while linoleic a-

nd linolenic acid contents were highest in SBM. Overall, the highest percentage of saturated FAs (SFA) was four-

nd in FM, the highest percentage of mono-unsaturated FAs (MUFA) in ACP and PBM and the highest percentage of poly-unsaturated FAs (PUFA) in SBM.

Table 2. Chemical composition of the experimental protein sources (n=5)

Chemical composition (% DM)	Protein sources [†]				SEM	P value
	SBM	FM	ACP	PBM		
Dry matter	90.14	89.40	85.77	89.61	2.17	NS
Organic matter	95.14 ^b	94.38 ^c	96.24 ^a	96.14 ^a	0.11	**
Crude protein	44.54 ^c	65.55 ^a	55.05 ^b	54.04 ^b	0.55	**
Ether extract	1.66 ^d	10.07 ^c	24.55 ^a	22.53 ^b	0.43	**
Neutral detergent fiber	12.58 ^a	1.73 ^d	8.68 ^b	2.58 ^c	0.14	**
Acid detergent fiber	9.02 ^a	1.42 ^c	5.60 ^b	1.73 ^c	0.12	**
Ash	4.86 ^b	5.62 ^a	3.76 ^c	3.86 ^c	0.11	**

[†] SBM: soybean meal, FM: fish meal, ACP: American cockroach powder, PBM: poultry by-product meal

SEM= Standard error of the mean

NS= Non-significant

a,b: Within rows, mean with common superscript(s) are not different (P> 0.05)

Table 3. Fatty acids composition of the protein sources (n=5)

Fatty acid methyl-ester (%)	Protein sources [†]				SEM	P value
	SBM	FM	ACP	PBM		
C10: 0 (Capric acid)	0.09	0.04	0.06	0.05	0.015	NS
C12: 0 (Lauric acid)	0.03	0.08	0.04	0.09	0.020	NS
C14:0 (Myristic acid)	0.14 ^d	5.16 ^a	1.34 ^b	0.77 ^c	0.201	**
C14:1 (Myristoleic acid)	0.70	0.37	0.19	0.29	0.178	NS
C15:0 (Pentadecanoic acid)	0.34 ^c	4.06 ^b	0.41 ^c	5.48 ^a	0.429	**
C15:1 (Pentadecenoic acid)	0.33	0.15	0.22	0.14	0.057	NS
C16:0 (Palmitic acid)	11.86 ^d	25.42 ^a	21.27 ^b	17.48 ^c	1.271	**
C16:1 (Palmitoleic acid)	0.13 ^c	5.28 ^a	2.25 ^b	5.15 ^a	1.052	**
C18:0 (Stearic acid)	4.08 ^b	11.29 ^a	3.63 ^b	10.52 ^a	1.845	**
C18:1 trans-9 (Vaccenic acid)	1.20 ^a	0.46 ^b	0.33 ^b	1.18 ^a	0.204	*
C18:1 cis-9 (Oleic acid)	13.09 ^d	19.02 ^c	40.52 ^a	32.85 ^b	2.010	**
C18:2 cis-6 (Linoleic acid)	50.58 ^a	14.01 ^c	21.90 ^b	12.92 ^c	2.502	**
C18:3 (Linolenic acid)	10.09 ^a	6.24 ^b	2.59 ^c	3.15 ^c	1.030	**
C20:0 (Arachidic acid)	0.59	0.91	0.65	1.01	0.169	NS
SFA	16.82 ^d	47.06 ^a	27.29 ^c	35.40 ^b	2.205	**
MUFA	16.10 ^c	24.68 ^b	43.41 ^a	40.54 ^a	2.501	**
PUFA	60.67 ^a	21.85 ^b	23.19 ^b	17.57 ^c	1.254	**
Identified FA	94.09	93.18	93.89	93.51	2.030	NS

[†] SBM: soybean meal, FM: fish meal, ACP: American cockroach powder, PBM: poultry by-product meal

SFA: saturated fatty acids, MUFA: monounsaturated fatty acids, PUFA: polyunsaturated fatty acids

SEM= Standard error of the mean

NS= Non-significant

a,b: Within rows, mean with common superscript(s) are not different (P> 0.05)

In situ degradability measurement

Table 4 shows the *in situ* DM and CP degradability characteristics of the protein sources. Kinetic analysis of DM degradation showed that the soluble fraction (a) of SBM and ACP was highest among the protein sources (P<0.01). The potentially degradable DM fraction (b) was highest in SBM and FM, and lowest in PBM. The degradation rate of DM (c) in FM was highest among the protein sources. The estimated effective degradability of DM at the rumen passage rates of 0.03, 0.06, and 0.09/h SBM was highest in SBM, and lowest in PBM. The CP washable fraction 'a' was higher in FM and PBM than in SBM and ACP. In contrast, the SBM contained higher 'b' than ACP and this fraction in FM and PBM was the lowest. The degradation rate of CP (c) in FM and ACP was higher than that of SBM and PBM. The estimated -

effective degradability of CP at all rumen passage rates in ACP was significantly higher than other protein sources.

The *in situ* DM and CP degradability characteristics of the experimental diets are shown in Table 5. No significant differences were observed between the experimental diets in degradability coefficients (a, b and c) for DM. Also, the effective DM degradability at all passage rates was not significantly affected by the dietary treatments. There was no significant difference in CP fractions 'a' and 'c' between the experimental diets. In contrast, control and ACP diets contained higher CP fraction 'b' than PBM diet. The effective CP degradability of ACP and control diets were always highest (P<0.05) and it was always lowest for FM diet at all passage rates (passage rate, k per hour=0.03, 0.06 and 0.09).

Table 4. Ruminal degradation parameters and effective degradability of DM and CP of protein sources

Item	Protein sources [†]				SEM	P value
	SBM	FM	ACP	PBM		
Degradability coefficients for DM						
a (%)	36.0 ^a	32.0 ^b	33.5 ^{ab}	27.7 ^c	0.9	**
b (%)	61.9 ^a	29.1 ^c	56.2 ^b	31.0 ^c	1.0	**
c (h ⁻¹)	5.4 ^b	6.0 ^a	5.1 ^b	4.6 ^c	0.1	**
Effective Degradability for DM						
ED, 0.03 (%)	75.9 ^a	51.5 ^c	69.1 ^b	46.6 ^d	0.4	**
ED, 0.06 (%)	65.4 ^a	46.7 ^c	59.5 ^b	41.2 ^d	0.5	**
ED, 0.09 (%)	59.3 ^a	43.8 ^c	54.0 ^b	38.3 ^d	0.6	**
Degradability coefficients for CP						
a (%)	12.0 ^c	41.3 ^a	34.8 ^b	41.2 ^a	1.1	**
b (%)	81.9 ^a	18.2 ^c	49.5 ^b	17.8 ^c	1.2	**
c (h ⁻¹)	4.1 ^b	7.2 ^a	6.8 ^a	2.6 ^c	0.3	**
Effective Degradability for CP						
ED, 0.03 (%)	64.3 ^b	54.1 ^c	69.3 ^a	49.8 ^d	0.4	**
ED, 0.06 (%)	48.9 ^c	51.2 ^b	61.3 ^a	46.7 ^d	0.6	**
ED, 0.09 (%)	40.5 ^d	49.3 ^b	56.3 ^a	45.3 ^c	0.7	**

[†] SBM: soybean meal, FM: fish meal, ACP: American cockroach powder, PBM: poultry by-product meal

SEM= Standard error of the mean

a,b: Within rows, mean with common superscript(s) are not different (P> 0.05)

Table 5. Ruminal degradation parameters and effective degradability of DM and CP of the experimental diets

Item	Experimental diets [†]				SEM	P value
	C	FM	ACP	PBM		
Degradability coefficients for DM						
a (%)	34.1	32.4	34.0	32.9	2.1	NS
b (%)	47.6	49.1	47.0	47.8	1.9	NS
c (h ⁻¹)	12.2	12.0	12.9	11.7	0.5	NS
Effective Degradability for DM						
ED, 0.03 (%)	72.4	71.7	72.1	72.0	0.4	NS
ED, 0.06 (%)	61.6	65.1	66.1	65.6	0.6	NS
ED, 0.09 (%)	61.6	60.5	61.7	61.0	0.7	NS
Degradability coefficients for CP						
a (%)	33.8	34.7	33.7	36.3	1.8	NS
b (%)	51.1 ^a	46.8 ^{ab}	50.9 ^a	44.4 ^b	1.9	*
c (h ⁻¹)	10.2	10.4	11.1	10.3	0.8	NS
Effective Degradability for CP						
ED, 0.03 (%)	74.0 ^a	70.7 ^b	73.9 ^a	70.5 ^b	0.4	*
ED, 0.06 (%)	66.6 ^a	64.1 ^b	66.9 ^a	64.2 ^b	0.5	*
ED, 0.09 (%)	61.5 ^{ab}	59.6 ^b	61.9 ^a	59.9 ^{ab}	0.6	*

[†]C: control (based soybean meal), FM: diet containing 3% Fish meal, ACP: diet containing 3% American cockroach powder, PBM: diet containing 3% poultry by-product meal.

SEM= Standard error of the mean

NS= Non-significant

a,b: Within rows, mean with common superscript(s) are not different (P> 0.05)

Discussion

There have been several reports on the nutritional aspect of edible insects, but less attention has been paid to the American cockroach. On the other hand, due to the biodiversity of insects, different results have been obtained in relation to the nutritional aspects. The chemical composition of ACP and other protein sources in this study was in line with other experiments, showing a large variation between insect species and other protein sources (Kovitvadhi et al., 2019). The moisture content of the ACP was relatively low (14.23%), in agreement with Boate and Suotonye (2020) and Abulude et al. (2017). This reflects the fact that cockroach meal has a longer shelf life and can be stored for a long time. Low moisture content reduces microbial activities and deterioration of food during storage (Siulapwa et al., 2014).

Values of protein, fat and energy vary across insect species and also within species depending on the diet, stage of development, sex and environmental factors (Ramos-Elorduy et al., 2002; Finke and Oonincx, 2014; Ademolu et al., 2010). Protein is a significant component of edible insects, comprising between 30% and 65% of the total DM (Dobermann et al., 2017). Insect proteins have favorable protein profiles and can replace/complement the traditional sources of feed (Zielinska et al., 2015). The concentration of proteins in an insect also depends on the metamorphic stage of the insect. Adult wasps have been reported to have more protein than pupa and larva stage (Yin et al., 2017). The CP content of the ACP in the present study (55.05%) is almost identical to the reported value (53%) by Boateng et al. (2018) and Bernard and Allen (1997) in *P. americana*. Ramos-Elorduy et al. (1997) reported a CP value of 65.60% in this cockroach. Zielinska et al. (2015)

stated that, the quality of the insect proteins in comparison to other animal and plant proteins has to be assessed through the amino acids content. Similar to our results, Sayed et al. (2019) reported that the CP content of SBM and insect meal (*Bactrocera zonata*) were 44.0 and 58.1%, respectively. In another study, Taufek et al. (2018) compared the chemical compositions of FM vs cricket meal and reported their CP levels as 53.61 and 57.02%, respectively. Wang et al. (2005) reported that the CP percentage of insect meal (Field cricket) was 58.3% on a DM basis, comparable with those of the conventional protein feed supplements, SBM (46.8%), meat and bone meal (48.5%), and FM (60.2%). It has been reported that, measured amounts of nitrogenous substances of insects may be higher than their actual protein content since some nitrogen is also bound in the exoskeleton (Klunder et al., 2012). Kamalaka et al. (2005) reported that the level of CP in FM and poultry slaughterhouse waste powder was 63.8 and 55.6 % of DM, respectively, which was the same as the results obtained in this experiment.

Lipid content and types of lipids in insects vary according to their species and life stage (Tzompa-Sosa et al., 2014). Insect lipids can supply energy and essential FAs (Ramos-Elorduy, 2008). In this experiment the EE content of ACP was 24.55%. Fat content in some edible insects ranged from 12.97% to 24.7% (Zielińska et al., 2015). Fat content in *P. americana* was 28.20% (Ramos-Elorduy et al., 1997) and 26.93% (Boateng et al., 2018). Sayed et al. (2019) reported that the fat content of SBM and insect meal (*Bactrocera zonata*) were 1.9 and 25.3%, respectively. These results are consistent with our results. In Wang et al. (2005) study, the fat content of insect meal (field cricket), FM, meat and bone meal and SBM were 10.3, 4.11, 8.71 and 1.84%, respectively. Kamalaka et al. (2005) reported that the fat level of FM and poultry slaughterhouse waste powder was 8.1 and 13.8 %, respectively. The value of EE in PBM in this study was similar to the results obtained by Narang and Lal (1985). In this experiment, the ACP fat content was slightly lower than that (28.4%) reported by Bernard and Allen (1997) and higher than that in Abulode and Folonus (2003) report (21.21%). Edible crickets contain, on average, 4.30 to 33.44% of lipids in DM basis (Magara et al., 2021). Although the high fat level of ACP could be useful for energy production in the animal, this high level of fat could be a disadvantage in fiber digestion and ruminal fermentation function. Therefore, lower levels of this protein source may find use in ruminant nutrition.

Apparently, exoskeleton of insects contributes to such high fiber contents. Chitin (a polymer of N-acetyl glucosamine), the main component of the insect exoskeleton (Chaudhari et al., 2011), is considered as fiber (Finke, 2007). The NDF and ADF contents of ACP were 8.68 and 5.60%, respectively. Zielińska et al. (2015) reported that in some edible insects, the average fiber contents ranged from 1.97% for *T. molitor* to 3.65%

for *G. sigillatus*. Contrary to these results, Jayanegara et al. (2017), comparing three insect species (including *Gryllus assimilis*, *Tenebrio molitor* and *Hermetia illucens*) with SBM, reported that all insect meals had higher NDF and ADF than that of SBM. The ADF content is related to chitin content in insects (Marono et al., 2015). Similar to our results, Finke (2002) reported that cricket nymphs and adult crickets contained intermediate levels of fiber (9.6 and 10.2% ADF in DM basis) and adult mealworms contained high levels of ADF (20.4% DM basis). He noted that insects with a hard exoskeleton do contain more fiber. The crude ash value of ACP was 3.76% is in line with data of other authors (Boate and Suotonye, 2020; Kulma et al., 2016) using different species of cockroaches. Magara et al. (2021) reported that edible crickets contained 2.96 to 20.50% ash per dry weight.

Due to the fact that each of the protein sources has different origin and types (plant, insect, etc.), the results of FAs profile showed many differences between the experimental groups. The unsaturated FAs profile of insects is similar to that of poultry and white fish but contains more PUFAs than red meat (Rumpold and Schluter, 2013). Several studies have been published on FAs composition of insects (Bukkens, 1997; Rumpold and Schluter, 2013). It has been reported that, some insects such as *P. americana* and *A. domesticus* are able to synthesize PUFAs. On the other hand, it must be noted that the fat profiles of insects are highly dependent on their feedstuff (Dobermann et al., 2017). For example, one study has shown that levels of eicosapentaenoic and docosahexaenoic acids can be increased in black soldier flies by feeding them fish offal (St-Hilaire et al., 2007). Womeni et al. (2009) reported a fat percentage of 6-7% in several insect species, which was rich in PUFAs and contained essential FAs such as linoleic and α -linoleic acids. Similar trends in insect FA composition were reported by other researchers (Chakravorty et al., 2014; Ghosh et al., 2017; Zielińska et al., 2015; Akullo et al., 2018). All these studies also reported a higher concentration of PUFA and MUFA than SFAs, which are good for health. The concentration of PUFA and MUFA was more than 50% of total fat in *Rhynchophorus phoenicis* (Raphia weevil), *Zonocerus variegates* (grasshopper), *Homorocoryphus nitidulus* (cricket), *Protaetia brevitarsis* (beetle), and *Teleogryllus emma* (cricket) species (Ghosh et al., 2017; Womeni et al., 2009; Akullo et al., 2018). The SFA in ACP was 27.29%. The three main components of the SFA are myristic (1.34%), palmitic (21.27%) and stearic (3.63%) acids. Similar to our results, Zielińska et al. (2015) showed that the palmitic acid was 23% in edible insects tested, but stearic acid ranged between 7.35 to 9.27%. According to Yang et al. (2006), SFAs in some edible insects were between 26.4 to 39.2%. In this experiment only two MUFA, palmitoleic acid (2.25%) and oleic acid (40.52%) were detected in the ACP. Yang et al. (2006) also detected only these two MUFAs in the tested insects.

Similar to our results, Zielińska et al. (2015) reported t-

he highest concentration of oleic acid in *T. molitor* (40.86%). They reported MUFA levels in some species of edible insects ranged from 34.33 to 43.27%, which is consistent with our results for ACP (43.41%). The two main components of PUFAs in ACP were linoleic acid (20.40%) and linolenic acid (2.79%). In insect powder, similar to the plant oils, the linoleic acid content was higher than linolenic acid (Makkar et al., 2014). In our experiment, the PUFA value for ACP was 23.19%. According to Zielinska et al. (2015), similar content of PUFA was shown in *S. gregaria* (26.28%). Similar PUFA values were reported by Yang et al. (2006) in the Spur-throated grasshopper (24.23%) and Giant water bug (25.43%). It has been reported that the concentration of PUFAs in insect meals was comparable with red meat and some fish (Sinclair et al., 1992; Li et al., 2002) and more than that found in vegetables (Pereira et al., 2001). In ACP, small amounts of other FAs were also measured (Table 3) as also found by Yang et al. (2006). The FAs of insects are generally comparable to those of fish and poultry in their degree of unsaturation, but contain more PUFA (Zielinska et al., 2015). Similar to our results, Magara et al. (2021) reported that the SFA, MUFA, and PUFA in poultry tissues were in the range of 30.9-32.2, 48.0-49.1 and 19.1-20.4%, respectively.

The ruminal degradation kinetics are affected by many factors such as origin of protein, feed processing, fistulated animals, pore size of bag and particle size (Nocek et al., 1979; Wadwa et al., 1998). In this study, the fractions 'a' and 'b' for DM of ACP were similar to the values for SBM. Similar to our results, at 96 h incubation time, DM disappearance of SBM was significantly higher than that of the FM and PBM (Kamalak et al., 2005). The highest DM degradability of SBM at all incubation time indicates that SBM is more susceptible to microbial attack in comparison to other protein sources (Khan et al., 1998). Gonzalez et al. (2002) reported different results for the fraction 'b' of SBM of approximately 70% of the DM. Others researchers reported lower values for fraction 'b', which ranged from 55.8% to 59.2% (Mondal et al., 2008; Maxin et al., 2013).

The effective DM degradability of ACP at 6% outflow rate was slightly lower than SBM and higher than FM and PBM. In line with our results, Kamalak et al. (2005) report that DM effective degradability at 6% outflow rate for SBM (52.2%) was higher than FM (47.2%) and PBM (41.4%). Limited data on the bioavailability of nutrients in insect products are available (Ojha et al., 2021). Bioavailability of a food is described as the fraction that is soluble and absorbable in the gastrointestinal tract (Cardoso et al., 2015).

The quality of a protein source is determined by both the composition of amino acids and the protein digestibility, expressed as a percentage of ideal protein (Belluco et al., 2013). Protein digestibility of some edible insects and the bioavailability of nutrients in edible insects have been examined in few studies (Churchward-Venne et al., 2017). In this study, the CP degradability coefficients 'a', 'b' and 'c' for ACP were

34.8%, 49.5% and 6.8 h⁻¹, respectively. The ACP 'a' was larger than SBM and smaller than FM and PBM values. The overall findings suggested that the water-soluble protein fraction was more easily digested than water-insoluble protein fraction during gastric and duodenal digestions (Ojha et al., 2021). Instead, the CP degradability 'b' for ACP was larger than FM and PBM and smaller than SBM values. The CP fraction 'c' for ACP was equal to FM and greater than PBM and SBM values. WenXiu et al. (2010) measured the degradability rate of CP in Japanese lateolabrax (worm larvae) and reported that the degradability rate of CP was 85%. The values for fractions 'a' and 'b' for SBM measured in our experiment are in agreement with Woods et al. (2003), who reported that these fractions were 12.9 and 83% respectively. Finke (2004) reported in a review that the protein digestibility of 50.2% in *Brachytrupes* sp. and 83.9% in *A. domesticus* crickets, which are slightly lower than the values in eggs (95%), beef (98%), and cow milk (95%). Marono et al. (2015) found a positive correlation between CP content and digestibility in insect meal (*Hermetia illucens*). Jayanegara et al. (2017) in an *in vitro* digestibility experiment, compared three insect species (including *Tenebrio molitor*, *Gryllus assimilis* and *Hermetia illucens*) with SBM and reported that all insect meals had lower DM and OM digestibility than that of SBM. They reported that, high fiber contents in insect-containing feeds reduced the DM and OM digestibility as compared to SBM.

Limited studies have been reported on the effects of chitin on ruminal methanogenesis. However, some experiments reported that chitosan (chitin deacetylation derivative) reduced methane production specifically by affecting the bacterial composition (Goiri et al., 2010; Belanche et al., 2016). Chumpawadee et al. (2005) showed that ruminal degradability rate of CP in SBM for rapid and slow degradable portions were 10.98 and 89.02%, respectively, which is similar to our results. The results of ruminal disappearance of DM and CP of experimental diets are shown in Tables 5. Replacing 3% FM, ACP, and PBM with SBM (control diet) had no significant effect on any of the degradability portions and effective degradability ratios of DM. Perhaps, the substituted amounts of these protein sources were too small to affect the DM degradability. Boateng et al. (2018) reported that feeding 2% and 4% cockroach meal (*P. americana*) had no effect on rats. Replacing 3% FM and PBM reduced the CP fraction 'b' compared to control and ACP diets. As the results in Table 4 show, the CP fraction 'b' in FM and PBM was much lower than in SBM and ACP, and this may have affected the CP degradability 'b' in the experimental diets. No other data are currently available regarding rumen degradability of insects.

Conclusions

Comparison of four different protein sources of different origins showed that insects are a good source of protein

and fat. They have balanced nutrient characteristics for ruminants and are high in MUFAs and PUFAs. There were significant differences between ACP and other protein sources in terms of DM and CP degradability. Future studies should be aimed at determining the palatability and *in vivo* effects of ACP in ruminants.

Conflict of interest statement

The authors certify that there is no conflict of interest.

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