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Physico-chemical properties of extruded aquafeed pellets containing black soldier fly (*Hermetia illucens*) larvae and adult cricket (*Acheta domesticus*) meals

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RESEARCH ARTICLE

Abstract

Fish farming is faced with the challenge of high cost of feeds because of the cost of high quality protein needed for formulation of the feeds. Thus, there is urgent need for alternative protein sources. The effects of substituting freshwater shrimp meal (FWSM) with black soldier fly larvae meal (BSFM) or adult cricket meal (ACM) on physico-chemical properties of hot-extruded fish feed pellets were investigated. The FWSM protein in a 26 g/100 g protein fish feed formulation was substituted at 0, 25, 50 and 75%, and moisture content of the formulated blends adjusted to 10, 20 or 30 g/100 g prior to extrusion. Floatability, expansion rate, bulk density, durability index, water absorption index, water solubility index, and water stability of extruded pellets were determined. Sinking velocity and the total suspended and dissolved solids in water were determined for the optimal pellets. Pellet floatability was not influenced by the type of insect meal but the interaction between level of inclusion and moisture content of the feed at extrusion. Pellets with high floatability >90% were produced from all feed blends at 30 g/100 g moisture content. Expansion ratio, was not influenced by type of insect meal or the level of inclusion but by the moisture content whereby feed blends extruded at 30 g/100 g moisture gave pellets with high expansion ratio ~60%. Bulk density was influenced by the interaction of the three factors. Pellet durability and water absorption indices were not influenced by the investigated factors or their interactions. Processed pellets were generally highly durable (99%) out of water, but the stability in water was significantly influenced by the interaction of type of insect meal level of inclusion and moisture content at extrusion. Water solubility increased with increasing extrusion moisture. Overall, it was possible to process good quality extruded pellets with 75% BSFM or 75% ACM at 30 g/100 g feed moisture.

Keywords: aquafeed, extrusion, edible insects, processing

1. Introduction

Aquaculture is one of the fastest growing food-producing sectors in the world (Halden *et al.*, 2014); it supplies about 50% of all the fish that is consumed. Thus, fish farming is a venture that has great potential for making nutritious food available, creating employment and enhancing income generation in developing countries. However, fish farmers are faced with the challenge of high cost of feeds, which hinders profitability. In Africa, and other developing countries, cost of feed is dependent on the cost of the key ingredients utilised (Cocker, 2014), which is estimated at 80-90% of the total cost of fish feed production (Rana *et al.*, 2009). In East Africa, aquafeed manufacturers use silver cyprinid, *Rastrineobola argentea* (Pellegrin) and fresh water shrimp, *Caradina nilotica* (P. Roux), as protein source ingredients in fish feeds (Munguti *et al.*, 2014); the inclusion of these high quality protein ingredients promotes feed efficiency and growth rate (Barroso *et al.*, 2014). However, due to high cost and competing uses, the search for cheaper alternative protein sources has become important.

Insects have high crude protein content ranging from 35-73 g/100 g and other important nutrients; the nutritional composition has been regarded comparable to that of fish meal. Edible insects are therefore increasingly being considered attractive and suitable protein ingredients to replace fish meal (St-Hilaire *et al.*, 2007; K.K.M. Fiaboe *et al.*, personal communication). Insect production is more sustainable and is associated with a much smaller ecological footprint for many reasons: lower greenhouse gas and ammonia emissions (Oonincx *et al.*, 2010), less land area (Oonincx and De Boer, 2012), more efficient feed conversion (Van Huis, 2013), and potential to be grown on organic by-products of which 1.3 billion tons is produced globally per annum (Durst *et al.*, 2010; FAO, 2011). Insect production using organic by-products reduces the water footprint of the feed (Rumpold and Schlüter, 2013) due to their high feed conversion efficiency. These insects form part of the natural diet of many edible fish species (Makkar *et al.*, 2014; Ravindran and Blair, 1993).

The work of Bondari and Sheppard (1981) revealed the possibility of using soldier fly larvae in commercial fish production. Crickets are also believed to be potential sources of nutrients for fish (Makkar *et al.*, 2014). However, majority of the studies used live insects or the ground meal without further processing. Extrusion processing is an effective modern way of adding value especially for commercial undertakings. Traditionally, aqua feed were prepared by mixing the ingredients into a simple mash. Then, pelletization by simple pressing techniques was later introduced to make fish pellets (Moscicki, 2011). However, these two methods had limitations related to efficiency of utilisation, handling and storability of the product. According to Rokey *et al.* (2010), extrusion was first used to produce dry expanded pet foods in the 1950s. The method has so far been widely accepted, and is used to produce many tons of animal food throughout the world. It is an efficient method of producing both floating and sinking aqua feed (Moscicki, 2011). Other reasons that have contributed to the rise in use of extrusion process include versatility of the process and reduced cost of production when large quantities of feed are to be produced for commercial reasons. In addition, extrusion enhances digestibility and bioavailability of nutrients, improves safety of the final product and is also an environmentally-friendly process. The aim of this study was to investigate the effects of substituting fresh water shrimp meal by different levels of black soldier fly meal and cricket meal on the physico-chemical properties of extruded fish feed pellets.

2. Materials and methods

Research site

Extrusion trials were performed using a small-scale fish feed extruder at a cottage fish feed processing facility owned by a group of small-scale fish farmers in Vihiga County, Kenya. This cottage facility formulates and processes the feed using locally obtained ingredients that include rice bran, wheat pollard, wheat bran, maize germ, maize bran, cotton seed cake, soybean, sunflower cake, fresh water shrimps and cassava for its group members who farm tilapia, *Oreochromis niloticus* (L.) and catfish, *Clarias gariepinus* (Burchell). Laboratory analyses were carried out at the Department of Dairy and Food Science, Egerton University, Kenya.

Materials

Sunflower cake, maize germ, wheat pollard, dried fresh-water shrimps and dried cassava chunks were purchased from a local vendor in Luanda town, Vihiga County. Blanched and sundried black soldier fly larvae (*Hermetia illucens* L.) were obtained from Sanergy Limited, a commercial insect rearing farm based in Nairobi, Kenya, while blanched and sundried adult house crickets (*Acheta domesticus* (L.) were purchased from farmers in Homa Bay County, Kenya. Each ingredient was separately ground into fine meal using hammer mill (model 4, Arthur H. Thomas, Philadelphia, PA, USA) and passed through a 1.0 mm aperture sieve.

Experimental design

A 2×4×3 factorial arrangement in a completely randomised design was used. The factors investigated were: insect type (2 levels: black soldierly larvae and adult cricket), level of freshwater shrimp meal (FWSM) protein substitution with black soldier fly larvae meal (BSFM) or adult cricket meal (ACM) (4 levels: 0, 25, 50 and 75%), and feed moisture content (3 levels: 10, 20 and 30 g/100 g on wet weight basis). The experiment was replicated four times.

Determination of proximate composition of ingredients

Proximate composition was determined according to AOAC (2000) methods: crude protein was determined by AOAC method 984.13, crude fat by AOAC method 920.39 and crude ash by AOAC method 942.05. Crude fibre was determined according to ISO 6865.2000 (ISO, 2000), while carbohydrate content was determined by difference.

Formulation of insect meal containing blends

Based on the proximate composition of ingredients, the required quantities to formulate 5 kg of isoproteinous blends (26.44±0.75 protein) were calculated using Microsoft Excel® function on Windows 2007 (Microsoft Corporation, Redmond, WA, USA). The amount of water needed to achieve desired moisture content was calculated, weighed, and added to the other ingredients in 20 l plastic bucket. These were mixed manually by hands for 2 min, then transferred to a multi-vane paddle mixer (model MX-25; Unitech, New Delhi, India) and mixed further for 2 min at moderate speed. Compositions of the various blends are given in the results section.

Extrusion process

A low-cost single screw extruder (model DOLLY; Unitech), with a screw length to diameter ratio of 9.1 and operating at a speed of 200 rpm was used. The extruder was equipped with a pre-conditioning chamber where the incoming feed was first preheated with steam at an inlet pressure of 4 bars. The blended ingredients were introduced into the steam conditioning chamber through a manual hopper at a feed rate of 1 kg/min. The feed was conditioned by passing through the conditioning chamber for approximately 2 min after which the mixture was channelled into the extrusion jacket that housed a single screw with a barrel length and diameter of 55 and 6 cm, respectively. The extruder was set to operate at 120 °C. The extruded material exited through a 2 mm-diameter die and cut into pellets by a rotating blade. The pellets were dried in a solar tent to constant weight. About 500 g of the dried pellets were sampled in duplicate and packed into zip-lock bags for analysis.

Determination of physico-chemical properties of pellets

Floatability

For each sample, 10 randomly selected pellets were put into 250 ml beakers containing 200 ml of distilled water at room temperature. This was done in 3 replicates and average number of pellets that were found floating after 20 min was recorded. Floatability was calculated as the number of floating pellets after 20 min divided by the total number of pellets introduced in the water multiplied by 100 (Khater *et al.*, 2014; Umar *et al.*,2013).

Expansion ratio, surface area and volume

Expansion ratio, surface area and volume were determined as outlined by Khater *et al.* (2014). For each sample, the diameter (D) and length (l) of 10 randomly selected pellets were measured using a digital Vernier caliper (SR No. 09070705763; Mars, McLean, VA, USA) and their average value recorded. Expansion rate (ER) was calculated using the expression:

$$ER(\%) = ((D - D_i) / D_i) \times 100$$

where D_i is the die diameter. The surface area (SA) and volume (V) of pellets were estimated using the expressions:

$$SA = \pi D(l + D / 2)$$

$$V = \pi D^2 l / 4$$

Bulk density

The procedure outlined by Fallahi *et al.* (2012) was used. Extruded pellets were milled using a laboratory-scale grinder and passed through a 1 mm aperture sieve. A 50 ml graduated measuring cylinder was tarred and gently filled with 50 g of the powder. The bottom of the cylinder was repeatedly tapped gently until there was no further reduction in sample volume. Bulk density was calculated as weight of the sample divided by the respective volume (g/l).

Pellet durability index

Pellet durability index was determined as outlined by Umar *et al.* (2013). About 15 g of each sample was sieved on a 2.36 mm sieve in triplicate. The pellets that were retained on the sieve were weighed (W_i) and placed in a flask mounted on a Lab-Line shaker (SR No. 0486-0002; Lab-Line Instruments, Inc, Melrose Park, IL, USA), which was then shaken for 20 min at 260 rpm. The pellets were then sieved and re-weighed (W_r) and the pellet durability index (PDI) calculated using the expression:

$$PDI(\%) = (W_r / W_i) \times 100$$

Water absorption index and water solubility index

The water absorption index (WAI) and water solubility index (WSI) were determined as described by Rosentrater *et al.* (2009). About 0.625 g of each sample (W_i) was ground and suspended in 8 ml distilled water in a tarred 12 ml centrifuge tube. The contents were shaken vigorously for 3 min and then centrifuged (using a bench-top centrifuge, model DSC-200T; Digisystem Laboratory Instruments Inc., New Taipei City, Taiwan) at 2,500 rpm for 10 min. The supernatant was decanted and transferred into a tarred aluminium dish and placed in a hot air oven (SR No. WOF 500 80502004; Daihan Scientific Co, Ltd, Seoul, Korea) maintained at 135 °C for 2 h. The dish and its contents was cooled in a desiccator and re-weighed on a sensitive weighing scale (model DJ-1505-S; Shinko Denshi Co Ltd, Tokyo, Japan), and the difference in weight (W_s) obtained. The mass of the gel remaining in the centrifuge tube (W_g)

was obtained as well. The WAI and WSI were calculated using the expressions:

$$WAI(\%) = (W_g / W_i) \times 100$$

$$WSI(\%) = (W_s / W_i) \times 100$$

Water stability

The procedure described by Umar *et al.* (2013) was used. About 4 g of each sample was weighed and put on a 0.5 mm wire mesh screen in 3 replicates. The screen with the sample was immersed into a 250 ml beaker containing 200 ml distilled water for 20 min. The pellets retained on the wire mesh were then dried in hot-air oven (105 °C) for 24 h. The percentage ratio of weight of pellets retained on the wire mesh to the initial weight gave the water stability.

Sinking behaviour

The procedure outlined by Umar *et al.* (2013) and Rosentrater *et al.* (2009) was used with modification. Ten pellets from each sample were put into a 250 ml measuring cylinder containing 200 ml distilled water. The number of pellets sinking at onset (time 0), 2, 4, 8, 24 and 48 h were counted and a graph of the percentage number of sinking pellets against time plotted.

Total suspended solids and total dissolved solids in water

Total suspended solids (TSS) and total dissolved solids (TDS) were determined according to AOAC (1998). Triplicate samples of 5 g were weighed and introduced into a 250 ml measuring cylinder containing 200 ml distilled water. Aliquots (5 ml) of the supernatant were drawn before introducing the pellets (time 0), and after 4, 8, 24 and 48 h, and filtered through a pre-weighed Whatmann® filter paper No. 2 (110 mm diameter; Whatmann International Ltd, Maidstone UK). The residue was dried at 105 °C for 1 h. The filtrate was poured into a pre-weighed aluminium dish and evaporated at 180 °C for 2 h. Both the dried residue and filtrate were cooled in a desiccator and then weighed. TSS was the percentage difference in dry weight of the filter paper while TDS was the percentage difference in dry weight of the aluminium dish.

Statistical analysis

Analysis of variance was performed using General Linear Model procedure (PROC GLM) of the Statistical Analysis System (SAS) software version 9.1.3 (SAS Institute Inc., Cary, NC, USA). Means were separated using Tukey's HSD test at 95% confidence level.

3. Results

Composition of ingredients and formulated blends

Proximate composition of the various ingredients is shown in Table 1. The ACM had highest protein content, which was about 15% higher than that of FWSM. The BSFM contained highest amount of fat, which was 2.4 times higher than that contained in FWSM while the fat content of ACM was about 1.3 times that of FWSM. The amounts of fibre contained in ACM and BSFM were double that contained in FWSM, whereas FWSM had more than double the amount of ash contained in ACM and BSFM. The gross composition of formulated blends is given in Table 2. Substituting fish meal with BSFM resulted in a slight decrease in protein content of feed blends, whereas substituting with ACM progressively increased the protein content. All the blends containing insect meal had higher contents of fat and fibre than the control. Blends containing BSFM particularly had relatively higher fat content. The control formulation had higher ash content than the insect meal containing formulations. Carbohydrate contents of the blends were fairly equal.

Table 1. Proximate composition of ingredients (g/100 g DM).[1]

Ingredient	Component				
	Crude protein	Crude fat	Crude fibre	Ash	Carbohydrate
Sunflower cake	20.60±0.67	21.29±0.51	31.89±0.74	4.78±0.97	21.44±1.31
Maize germ	13.81±0.16	9.87±0.05	16.17±1.20	5.89±0.14	54.26±0.09
Wheat pollard	16.01±0.33	8.50±0.49	11.49±1.34	3.44±0.03	60.56±0.43
FWSM	53.98±1.52	10.53±1.44	4.18±0.23	22.34±2.02	11.97±1.26
BSFM	41.77±0.65	24.95±0.35	8.81±0.35	10.8±0.03	13.67±0.31
ACM	62.35±1.03	13.34±0.19	8.62±0.08	8.36±0.14	7.30±1.10
Cassava	1.96±1.12	0.26±0.20	1.9±0.03	2.34±0.02	93.54±0.25

[1] FWSM = fresh water shrimp meal; BSFM = black soldier fly meal; ACM = adult cricket meal.

Table 2. Gross composition (g/100 g DM) of formulated experimental blends.[1]

Ingredient	Control	Fish meal substituted formulations					
		BSFM25	BSFM50	BSFM75	ACM25	ACM50	ACM75
Sunflower cake	19.0	18.5	18.1	17.6	19.2	19.4	19.6
Maize germ	19.0	18.5	18.1	17.6	19.2	19.4	19.6
Wheat pollard	28.5	27.8	27.1	26.5	28.8	29.1	29.4
FWSM	28.5	20.9	13.6	6.6	21.6	14.5	7.3
BSFM	–	9.5	18.4	27.1	–	–	–
ACM	–	–	–	–	6.3	12.5	19.1
Cassava flour	5.0	4.9	4.8	4.6	5	5.1	5.2
Proximate							
Protein	26.6	26.2	25.7	25.3	26.9	27.1	27.4
Fat	11.4	12.7	14	15.2	11.6	11.7	11.9
Fibre	13.7	13.9	14.1	14.5	14.1	14.4	14.8
Ash	9.5	8.7	8	7.3	8.5	7.5	6.5
Carbohydrate	38.8	38.5	38.2	37.7	38.9	39.3	39.4

[1] FWSM = fresh water shrimp meal; BSFM = black soldier fly meal; ACM = adult cricket meal; BSFM25 = BSFM substitutes 25% of the protein supplied by FWSM in control; BSFM50 = BSFM substitutes 50% of the protein supplied by FWSM in control; BSFM75 = BSFM substitutes 75% of the protein supplied by FWSM in control; ACM25 = ACM substitutes 25% of the protein supplied by FWSM in control; ACM50 = ACM substitutes 50% of the protein supplied by FWSM in control; ACM75 = ACM substitutes 75% of the protein supplied by FWSM in control.

Floatability, expansion ratio, surface area, volume and bulk density of pellets

Insect type did not significantly influence (P=1.0000) pellet floatability (Table 3). The effect of interaction between level of FWSM substitution and moisture content was, however, significant (P=0.0198). In all formulations, pellet floatability increased with increasing moisture content (Table 4), but seemingly decreased with increasing level of insect meal inclusion when moisture content of formulation was maintained at 10 and 20 g/100 g. All blends with 30 g/100 g moisture content produced floating pellets with over 93% floatability and the values were not significantly different (P=0.05). Expansion ratio, surface area and volume of pellets were not influenced by insect type (expansion ratio: P=0.2800; surface area: P=0.6050; volume: P=0.5728). Level of FWSM substitution also did not influence these parameters (25% substitution: P=0.0658;

Table 3. Summary of ANOVA test outputs.[1,2]

Variation source	df	*F*	ER	SA	V	BD	DI	WAI	WSI	WS
IT	1	0.00	41.24	300.81	189.57	4,530.13	0.11	0.06	11.24	7.59
LS	3	3,201.74**	92.46	1,933.21	1,074.71	44,716.59***	0.07	0.03	6.71	17.76**
MC	2	22,474.35***	126.08*	3,753.04*	2,198.23*	282,984.17***	0.26	0.04	13.47*	48.07***
IT×LS	3	551.74	5.54	2,014.58	1,058.46	1,795.16	0.34	0.09	4.43	22.98***
IT×MC	2	37.89	8.24	269.82	114.14	2,443.92	0.03	0.01	2.18	43.09***
LS×MC	6	1,084.94*	21.22	385.55	245.64	10,700.08	0.37*	0.04	5.79	31.02***
IT×LS×MC	6	808.89	63.69	934.91	489.58	2,348.22	0.12	0.02	3.34	6.59*
Error		343.23	33.80	1,094.73	579.85	4,552.123	0.15	0.03	3.46	2.75
R^2 (%)		89.15	57.85	51.69	52.89	87.89	57.25	50.93	61.19	89.06
CV		28.97	10.24	28.15	31.85	11.71	0.39	4.61	16.05	1.99

[1] IT = insect type; LS = level of insect meal inclusion; MC = moisture content; df = degrees of freedom; F = floatability; ER = expansion ratio; SA = surface area; V = volume; BD = bulk density; DI = durability index; WAI = water absorption index; WSI = water solubility index; WS = water stability.

[2] ***P<0.001; **P<0.01; *P<0.05; R^2 = coefficient of determination; CV = coefficient of variation.

Table 4. Effect of insect type, level of insect meal inclusion and feed moisture content on pellet floatability (F), expansion ratio (ER), surface area (SA), volume (V), and bulk density (BD).[1,2]

Property	MC (g/100 g)	Formulation						
		Control	BSFM25	BSFM50	BSFM75	ACM25	ACM50	ACM75
F (%)	10	68.8±6.3^{a,y}	0.0±0.0^{e,y}	20.3±1.3^{c,z}	7.5±0.5^{d,y}	46.3±6.3^{b,y}	25.0±5.0^{c,z}	0.0±0.0^{e,z}
	20	98.8±1.6^{a,x}	95.0±4.0^{ab,x}	71.3±6.3^{c,y}	12.5±2.5^{e,y}	95.0±0.0^{b,x}	42.5±4.5^{d,y}	51.3±6.3^{d,y}
	30	100.0±0.0^{a,x}	100.0±0.0^{a,x}	98.8±1.3^{a,x}	98.8±1.3^{a,x}	93.8±6.3^{a,x}	93.8±6.4^{a,x}	96.3±3.8^{a,x}
ER (%)	10	54.5±1.8^{b,y}	61.7±4.8^{a,x}	48.4±2.6^{c,y}	49.4±3.5^{bc,y}	51.2±1.5^{bc,y}	53.5±1.2^{b,y}	57.7±3.2^{ab,x}
	20	59.5±3.6^{ab,x}	58.6±6.1^{ab,x}	54.9±1.2^{b,y}	52.5±3.6^{b,xy}	67.8±5.0^{a,x}	56.4±3.3^{b,xy}	54.0±2.3^{b,x}
	30	52.6±7.8^{a,y}	60.7±2.8^{a,x}	61.7±4.5^{a,x}	55.7±1.5^{a,x}	62.4±6.1^{a,x}	59.5±1.2^{a,x}	54.5±3.7^{a,x}
SA (mm^2)	10	88.5±7.0^{b,y}	115.4±8.5^{a,y}	94.2±10.4^{b,z}	73.3±8.7^{b,y}	85.3±6.3^{b,y}	122.3±7.6^{a,y}	121.6±14.2^{a,y}
	20	118.0±7.2^{c,x}	136.7±7.8^{b,x}	120.3±5.0^{c,y}	122.8±0.3^{c,x}	111.8±11.2^{c,x}	167.8±16.9^{a,x}	91.9±7.6^{d,z}
	30	116.9±11.3^{b,x}	149.9±14.6^{a,x}	151.1±13.0^{a,x}	93.3±8.7^{c,y}	112.7±5.8^{bc,x}	154.6±8.7^{a,x}	140.7±16.3^{ab,x}
V (mm^3)	10	53.9±3.2^{b,y}	75.8±9.3^{a,z}	56.9±6.6^{b,z}	42.5±5.1^{b,y}	50.9±4.1^{b,y}	77.5±8.5^{a,y}	78.4±8.4^{a,y}
	20	76.7±8.2^{c,x}	89.8±3.0^{b,y}	76.7±3.5^{c,y}	77.6±0.8^{c,x}	75.3±7.9^{c,x}	111.9±5.3^{a,x}	55.9±5.3^{d,z}
	30	72.9±7.9^{b,x}	100.4±8.3^{a,x}	102.5±9.4^{a,x}	57.8±7.4^{b,y}	73.2±6.8^{b,x}	103.5±7.9^{a,x}	91.9±4.7^{a,x}
BD (g/l)	10	570.5±49.2^{d,x}	720.4±51.5^{bc,x}	719.3±44.1^{c,x}	861.3±14.3^{a,x}	771.9±97.3^{ab,x}	713.8±54.7^{c,x}	822.9±24.8^{a,x}
	20	466.3±3.9^{c,y}	502.5±40.2^{b,y}	517.1±39.2^{b,y}	676.8±34.3^{a,y}	545.2±78.4^{b,y}	564.2±71.5^{ab,y}	597.7±58.5^{ab,y}
	30	453.8±5.2^{b,y}	456.7±13.7^{b,y}	427.7±14.4^{e,y}	428.1±17.7^{e,z}	513.4±47.9^{a,y}	457.5±18.5^{b,y}	520.7±32.0^{a,z}

[1] MC = moisture content; BSFM = black soldier fly meal; ACM = adult cricket meal; BSFM25 = BSFM replaces 25% of the protein supplied by fresh water shrimp meal (FWSM) in control; BSFM50 = BSFM replaces 50% of the protein supplied by FWSM in control; BSFM75 = BSFM replaces 75% of the protein supplied by FWSM in control; ACM25 = ACM replaces 25% of the protein supplied by FWSM in control; ACM50 = ACM replaces 50% of the protein supplied by FWSM in control; ACM75 = ACM replaces 75% of the protein supplied by FWSM in control.

[2] For each property, means followed by the same letters a-e along the same row, i.e. comparing the different formulations at the same moisture content, are not significantly different (P<0.05). Similarly, means followed by the same letters x-z down the same column for each property and formulation, i.e. comparing the different moisture levels, are not significantly different (P<0.05).

50% substitution: P=0.1805; 75% substitution: P=0.1645). The effect of moisture content was, however, significant (expansion: P=0.0389: surface area: P=0.0440; volume: P=0.0371). Blends containing 30 g/100 g moisture produced pellets with higher expansion ratio, surface area and volume compared to those containing 10 and 20 g/100 g moisture (Table 4). Also, at 10 and 20 g/100 g moisture contents, pellets processed from 50 and 75% ACM containing blends had higher expansion than the respective BSFM blends.

Bulk density of extruded pellets was not influenced by insect type, but by the level of inclusion (P<0.001), and moisture content (P<0.001). The high value of R^2 (87.90%; Table 3), however, shows that the three factors and their interactions, sufficiently accounted for bulk density. The BSFM and ACM containing blends had higher bulk densities than the control formulation at 10 and 20 g/100 g moisture contents, and increasing the level of insect meal inclusion from 25% to 75% gave a curvilinear relation with bulk density (Table 4). An increase in feed moisture content significantly (P<0.05) decreased bulk density. Moreover, at 30 g/100 g moisture, lower bulk densities were attained in pellets extruded from the BSFM containing blends (BMSF50, BMSF75) as compared to the control and the ACM containing ones.

Durability index, water absorption index, water solubility index and water stability

Insect type, level of substitution and moisture content of formulation, and their interactions did not influence pellet durability (Table 3). Processed pellets were generally highly durable. Similarly, WAI was not influenced by the investigated factors or their interaction. However, WSI was influenced (P<0.05) by moisture content; WSI increased with increasing moisture content (Table 5). Water stability of pellets was significantly influenced (P<0.05) by the interaction effect of insect type, level of insect meal inclusion and moisture content (Table 3), and the high value of R^2 (89.062%) and low value of CV (1.996) show that this interaction sufficiently explained the parameter.

Table 5. Effect of insect type, level of insect meal inclusion and feed moisture content on pellet durability index (DI), water absorption index (WAI), water solubility index (WSI), and water stability (WS).

Property	MC (g/100 g)	Formulation						
		Control	BSFM25	BSFM50	BSFM75	ACM25	ACM50	ACM75
DI (%)	10	99.8±0.2	99.4±0.5	99.7±0.0	99.1±0.0	99.9±0.0	99.5±0.2	98.9±0.1
	20	98.8±1.1	99.7±0.1	99.9±0.0	99.6±0.1	99.9±0.1	99.3±0.0	99.9±0.0
	30	99.7±0.2	99.6±0.2	99.9±0.1	99.9±0.1	99.8±0.1	99.6±0.2	99.8±0.1
WAI (%)	10	4.1±0.1^{a}	4.2±0.1^{a}	3.8±0.1^{b}	3.8±0.1^{b}	4.1±0.1^{a}	4.1±0.1^{a}	4.2±0.1^{a}
	20	4.1±0.1	4.0±0.0	3.9±0.2	4.1±0.1	3.9±0.1	4.2±0.0	4.2±0.1
	30	4.1±0.2	3.9±0.2	3.8±0.0	3.9±0.0	3.9±0.2	4.1±0.2	3.9±0.3
WSI (%)	10	12.8±2.0^{y}	10.2±0.7^{y}	10.5±0.7^{y}	10.3±0.0	11.2±0.5	13.4±2.6^{x}	9.5±0.8
	20	12.3±2.6^{b,y}	15.1±0.4^{a,x}	10.4±0.4^{b,y}	9.9±0.6^{b}	11.7±0.3^{b}	9.1±0.3^{by}	10.1±0.2^{b}
	30	14.1±1.8^{a,x}	13.6±0.4^{a,xy}	14.0±3.8^{a,x}	11.7±0.2^{b}	11.0±0.7^{b}	13.6±0.3^{a,x}	11.3±0.0^{b}
WS (%)	10	85.8±0.4^{a,x}	78.4±3.5^{bc,x}	77.6±0.0^{c,x}	74.1±1.0^{c,x}	86.1±1.3^{a,x}	80.1±0.6^{b,x}	79.6±0.3^{b,x}
	20	84.7±1.1^{bc,x}	83.4±0.0^{c,y}	87.0±0.7^{a,y}	80.3±0.4^{d,y}	86.3±2.7^{ab,x}	81.3±0.1^{b,xy}	85.6±0.4^{b,y}
	30	83.7±0.6^{b,x}	83.2±0.0^{b,y}	87.1±0.3^{a,y}	87.2±0.1^{a,z}	83.7±1.7^{b,x}	83.0±1.2^{b,y}	83.5±1.3^{b,y}

[1] MC = moisture content; BSFM = black soldier fly meal; ACM = adult cricket meal; BSFM25 = BSFM replaces 25% of the protein supplied by fresh water shrimp meal (FWSM) in control; BSFM50 = BSFM replaces 50% of the protein supplied by FWSM in control; BSFM75 = BSFM replaces 75% of the protein supplied by FWSM in control; ACM25 = ACM replaces 25% of the protein supplied by FWSM in control; ACM50 = ACM replaces 50% of the protein supplied by FWSM in control; ACM75 = ACM replaces 75% of the protein supplied by FWSM in control.

[2] For each property, means followed by the same letters a-e along the same row, i.e. comparing the different formulations at the same moisture content, are not significantly different (P<0.05). Similarly, means followed by the same letters x-z down the same column for each property and formulation, i.e. comparing the different moisture levels, are not significantly different (P<0.05).

Sinking velocity, total suspended solids and total dissolved solids in water

Sinking velocity, TSS and TDS were determined on pellets processed from the control formulation and insect meal containing blends at 75% substitution (BSFM75 and ACM75) and 30 g/100 g moisture content because the three exhibited better properties in comparison to other treatments. Only <5% of the three differently processed pellets were able to sink in the first 8 h (Figure 1A). About 50 and 40% of pellets extruded from BSFM75 and control blends, respectively, had sunk after 24 h compared to 28% of the ACM75 pellets. This difference was significant (P<0.05). At 48 h, 90, 95 and 100% of the control, BSFM75 and ACM75 pellets, respectively, had sunk. Figure 1B shows that waters introduced with BSFM75 or ACM75 pellets did not differ in TDS levels. Moreover TDS of water introduced with these pellets was significantly lower (P<0.05) than that of water introduced with the control pellets. Figure 1C shows the TSS concentration overtime. At onset, water introduced with the control and ACM75 pellets had significantly higher (P<0.05) TSS concentration than that introduced with BSFM75 pellets. However, at 8 h TSS concentrations of water introduced with the three types of pellets were not significantly different (P<0.05). There was a sharp decrease in TSS concentration between 8-24 h and a slight increase from 24 h onwards in the waters introduced with BSFM75 and ACM75 pellets. A peculiar observation is that water introduced with the BSFM75 pellets had higher TSS than that introduced with the control or ACM75 pellets from 8 to 48 h.

4. Discussion

Proximate composition of the various ingredients agrees well with other values reported in literature (Finke, 2002; Munguti *et al.*, 2014; Tran *et al.*, 2015). The higher fat and fibre contents of BSFM and ACM compared to FWSM explain the higher fat and fibre contents of insect meal containing blends as compared to the control. FWSM had almost twice the amount of ash contained in BSFM and ACM, and was responsible for the higher ash content of the control. Overall, nutritional contents of BSFM and ACM blends were within that of the control indicating that the insect meals can effectively substitute FWSM as key protein source. Floatability determines whether an aqua feed is to be utilised by bottom, mid-level or top feeders that feed on sinking, slow sinking and floating feeds, respectively. It is related to structure formation, which is a function of composition of the feed material. Starch, and some proteins, play important role in structure formation during extrusion (Ayadi *et al.* 2011; Vijayagopal, 2004).

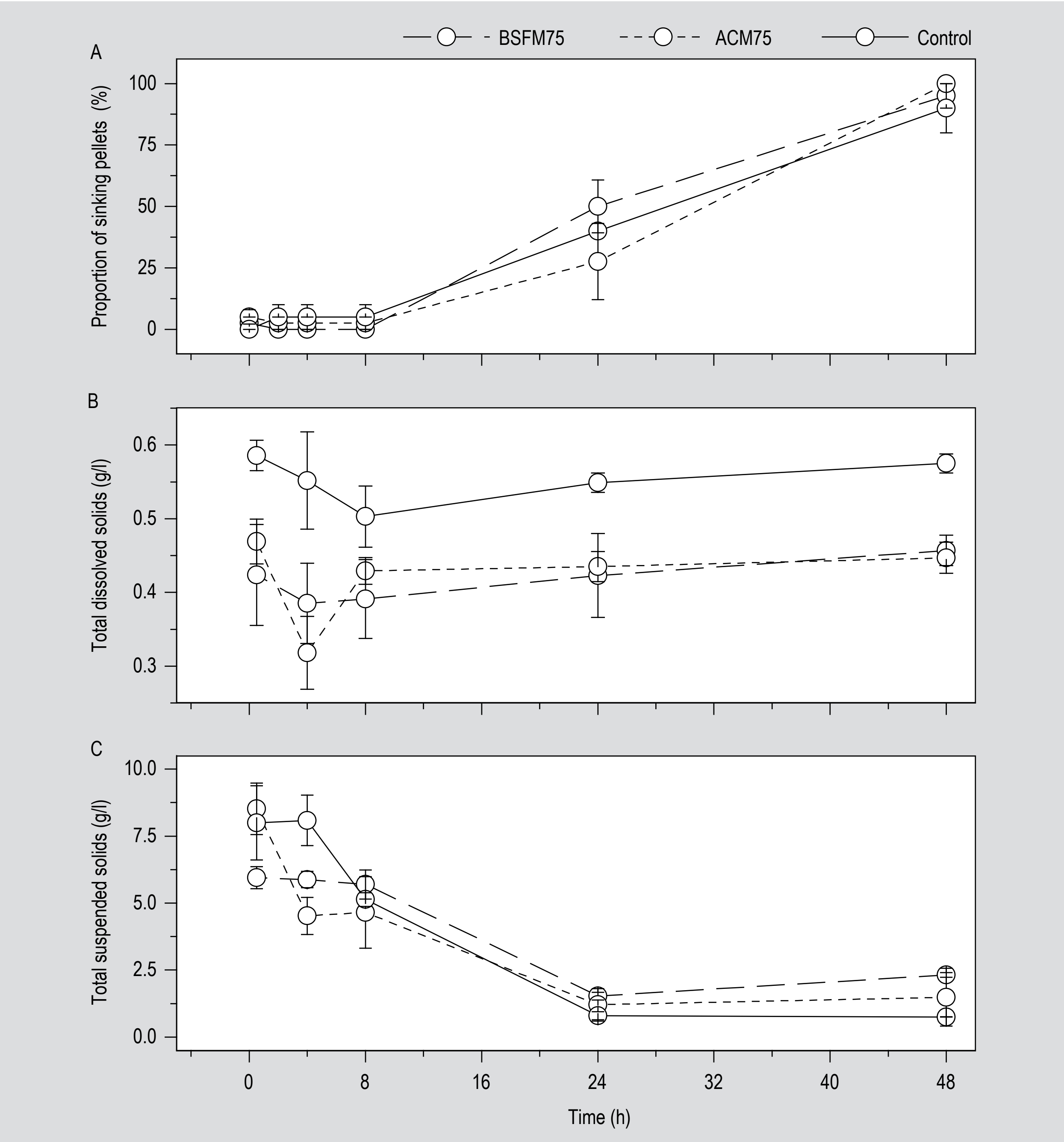

Figure 1. (A) Sinking profile of pellets, (B) total dissolved solids, and (C) total suspended solids of water introduced with pellets extruded from the control blend, black soldierly meal containing blend (75% substitution; BSFM75), and adult cricket meal containing blend (75% substitution; ACM75) adjusted to 30 g/100 g moisture content.

Feed moisture affects modification of macromolecules during the extrusion process but also lowers the energy consumption of the extruder (Levic and Sredanovic, 2010) by providing plasticisation effects, which is reason why appropriate feed moisture content is necessary. Depending on moisture content and temperature, starch granules are melted or gelatinised to form an amorphous continuous phase that traps the gases released at the extruder die enabling formation of expanded structures. In the present case, starch in various blends was supplied by wheat pollard, maize germ, sunflower cake and cassava, and these were not varied. The substitution of FWSM with BSFM or ACM, however, altered the composition of fat, and fibre. Contents of crude fibre and crude fat were particularly higher in BSFM and ACM than in FWSM. Increasing fat content may have been responsible for a decrease in pellet expansion and therefore floatability particularly for the BSFM containing blends. High fat content of feed causes reduced mechanical energy input during extrusion, which leads to an undercooked dough with low amount of dispersed starch hence a decreased overall expansion of the extruded pellets (Ilo *et al.*, 2000). Other ingredients such as fibre, depending on their polymeric nature, affect moisture binding of the feed or modify the behaviour or viscosity of the melt as it flows and exits through the die. The increase in floatability as moisture content was increased from 10 to 30 g/100 g could be attributed to increased starch gelatinization and a reduced melt viscosity that facilitated growth and maintenance of gas bubble growth (Singh *et al.*, 2014). Our findings are in agreement with the findings of Umar *et al.* (2013). Feed moisture content of 30 g/100 g offers the possibility to produce floating pellets at different levels of BSFM or ACM inclusion as may be desired.

In addition to floatability, expansion defines the brittleness of pellets (Rosentrater *et al.*, 2009) and also affects digestibility (Fallahi *et al.*, 2012); higher expansion ratios improve feed digestibility. Surface area and volume, which are related to expansion, give an indication of how the material will pack during storage and transportation (Khater *et al.*, 2014). As the plasticised dough exits the die under pressure, it swells, moisture flushes out, and cooling and relaxation occurs. These events affect the actual size and shape of extruded pellets. Expansion ratio, surface area and volume increased with moisture content because the water promoted starch gelatinisation leading to higher die expansion (Umar *et al.*, 2013). However, our finding somewhat contradicts the findings of Rosentrater *et al.* (2009) and Bandyopadhyay and Rout (2001) who found that an increase in feed moisture decreased the expansion ratio of extruded aqua feeds, but agrees well with the findings by Tumuluru (2013) who found that medium moisture content of 30-40 g/100 g increased die expansion. Fallahi *et al.* (2012) has also observed that moisture content, feed composition and the flow rate of the material through the extrusion barrel influences expansion rate. Although the effect of insect meal substitution on expansion ratio was not significant, a peculiar observation was that increasing the level of insect meal tended to decrease expansion of pellet expansion at each moisture content level, an observation that could be attributed to increase in lipids content in the formulated blends (Vijayagopal, 2004).

Bulk density measures the extent of puffing of the extruded material (Singh *et al.*, 2014), but unlike volume and surface area, takes into consideration the pores and voids that were developed during extrusion and therefore informs better on how the packaging material and storage facilities could be designed (Ayadi *et al.*, 2011; Chevanan *et al.*, 2007; Fallahi *et al.*, 2012). The decrease in bulk density with increase in moisture content is explained by the fact that moisture content favours expansion (Chevanan *et al.* 2007; Singh and Muthukumarappan, 2014, 2015). The inverse relationship between floatability and bulk density has also been reported (Adeparusi and Famurewa, 2011). Vijayagopal (2004) gives bulk densities for different categories of expanded aqua feeds: floating pellets <550 g/l, sinking pellets 550-650 g/l, and fast sinking pellets >650 g/l. Thus, from this study, BSFM and ACM can be used to produce pellets for all feeders by varying insect meal inclusion and feed moisture content. For the particular extruder used in this study, at 10 g/100 g moisture content, BSFM25, BSFM50, BSFM75, ACM25, ACM50 and ACM75 pellets would be suitable for bottom feeders such as catfish (*C. gariepinus*) while at 20 g/100 g moisture content, BSFM25, BSFM50, and ACM25 pellets are well suited for top feeders, ACM50 and ACM75 for mid-level feeders, while BSFM75 is suited for bottom feeders. At 30 g/100 g moisture content, BSFM25, BSFM50, BSFM75, ACM25, ACM50 and ACM75 pellets can all be used for top feeders such as tilapia (*O. niloticus*).

Durability index measured whether the extruded pellets can withstand mechanical handling (Singh and Muthukumarappan, 2015), and is dependent on factors such as the degree of heat treatment during extrusion, and the extent of starch transformation and its interaction with other macromolecules (Chevanan *et al.*, 2007, 2009; Rosentrater *et al.*, 2009). High values of durability index were obtained in this study and this could be attributed to the type of ingredients used in this study and the high level of temperature (120 °C) used during extrusion (Rosentrater *et al.*, 2009). The durability indices (>99%) indicate that the pellets produced can withstand major handling impacts characteristic of loading and unloading during transportation. Fallahi *et al.* (2012) also obtained >99% pellet durability index on extruded aqua feeds. The linear relationship between expansion ratio and durability index has also been reported by other studies. (Khater *et al.*, 2014; Kraugerud *et al.*, 2011). Our finding that moisture content did not influence durability index agrees with the finding of Singh and Muthukumarappan (2015), but negates the findings of Chevanan *et al.* (2007) and Foley and Rosentrater (2013) who observed significant effect of moisture content on durability of extruded aqua feeds containing distillers dried grains.

Water absorption and water solubility indices give the relationship that the pellets will have with water when introduced into fish ponds (Rosentrater *et al.*, 2009), and give an indication of the changes in ingredient content of the pellets in water (Fellows, 2000). Like our findings, Rosentrater *et al.* (2009) reported a curvilinear relationship between the inclusion levels of distillers dried grains and WAI and WSI of the extruded aqua feeds. Similarly, Fellows (2000) reported an inverse relationship between WSI and WAI. The increase in WSI with an increase in moisture content can be attributed to starch gelatinisation that increased with increase in moisture content of up to 30 g/100 g (Jackson *et al.*, 1990). It has also been reported that depolymerisation of macromolecules during extrusion produces simpler molecules that are more water soluble (Chiu and Solarek, 2009; Peluola-Adeyemi *et al.*, 2014). Several other studies have reported an increase in WAI and a decrease in WSI with an increase in moisture content (Badrie and Mellowes, 1991; Singh and Muthukumarappan, 2014). With respect to starch, WAI and WSI are also dependent on amylose and amylopectin (Moscicki *et al.*, 2012) and the ultimate ratio in the extruded product. An increase in WSI, as was the case in this study, is desirable in that it increases nutrient digestibility.

Water stability indicates how the product will withstand water dissolution (Ayadi *et al.*, 2011). Starch plays an important role in water stability (Vijayagopal, 2004), where its dextrinisation results in the products being able to absorb water fairly well. Similar findings were reported by Umar *et al.* (2013) and Foley and Rosentrater (2013) who found an increase in water stability with an increase in moisture content. Bandyopadhyay and Rout (2001) found an inverse relationship between moisture content and water stability. Tumuluru (2013) also reported that moisture content significantly affected water stability of extruded fish feeds. Vijayagopal (2004) observed that feed mixture needed to be wetted to moisture content of about 25 g/100 g for maximum water stability. The high water stability for pellets from all formulations extruded at 20 and 30 g/100 g moisture contents indicate that these products will resist nutrient leaching in water and thus allow better availability of nutrients for fish. In addition, minimal pollution of water would be experienced as a result of their use. For instance, FAO (2010) cites a water stability of 84% for extruded feeds intended for catfish, which are bottom feeders.

Sinking velocity gives an indication of how long the pellets would remain floating on water. The first 8 h after feed has been introduced into the pond is the time within which fish (top feeders such as tilapia) would be expected to have fed on the pellets. Expansion influences sinking velocity (Chevanan *et al.* 2007). The control, BSFM75 and ACM75 pellets had similar expansion ratios when extruded at 30 g/100 g moisture content. The three products thus had similar sinking characteristics within the first 8 h. Pellet sinking behaviour also coincides with changes in pond water quality. Suspended as well as the dissolved solids affect turbidity of water and thus light penetration (Hemalatha and Puttaiah, 2014). Within 0-8 h, the majority of the pellets would still be floating hence the high concentration of suspended solids. The drop in TSS between 8-24 h is explained by the fact that about half of the pellets are no longer floating while a slight increase in TSS between 24-48 h is because non-floating pellets begin to disintegrate. The high level of TSS in BSFM75 from 8-48 h is probably due to high fat content of BSFM making the pellets less intact. The TDS profiles imply that although all the pellets continued to lose small amounts of soluble nutrients into the water, BSFM75 and ACM75 pellets were more water stable than the control.

5. Conclusions and recommendations

Extruded aqua feeds with acceptable characteristics can be processed from blends containing BSFM or ACM as protein substitute. Up to 75% protein substitution level is possible but moisture content of the blends has to be increased progressively to about 30 g/100 g so as to obtain good quality pellets with regards to floatability, durability index, bulk density and water stability. Further work should investigate nutrient availability and digestibility as well as performance of the pellets in feeding trials.

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