

Effect of energy to protein ratio using alternative feed ingredients on growth performance and nutrient digestibility in broilers

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ABSTRACT

Present study was conducted to evaluate the different levels of energy to protein ratios (EPR) using food waste and black soldier fly larvae meal (FWBSFL) on growth performance and nutrient digestibility of broilers. A total of 160 one-day old broiler chicks were divided randomly to four groups and each group had 8 replicates with 5 chicks per replicate. The control diet was formulated using conventional feed ingredients with EPR of 154 for the starter period and 167 for the finisher period. The other treatments were diets with normal, low, and high EPR (154, 143, and 166 for the starter period; 167, 155, and 177 for the finisher period) using FWBSFL. Feed consumption and body weight gain as well as digestibility of crude protein, crude fiber, and fat were decreased due to feeding of FWBSFL diets. Different levels of EPR using FWBSFL decreased growth performance and nutrient digestibility of broilers.

Key words: Black soldier fly larvae meal, Broiler, Energy to protein ratio, Food waste.

INTRODUCTION

Feed accounts for the largest part of the total cost of poultry production. The black soldier fly (*Hermetia illucens* Linnaeus) belongs to the Stratiomyidae family. It is abundant and widespread in the tropical areas. Black soldier fly larvae (BSFL) can be easily cultivated using organic waste (Newton *et al.* 2005). They contain high amount of protein (39.16 to 64%) (Atteh and Ologbenla 2015) that can therefore be used as a protein source in the animal diets. Globally, 1.3 billion tons of food were wasted each year. Food waste can be used as an alternate feed resource in poultry feed, partially or completely substituting conventional feed resources (Omoikhoje *et al.* 2011). Food waste is generally used as energy source in broiler diets (Chen *et al.* 2007). The objective of this study was to investigate the effect of different levels of energy to protein ratio (EPR) using food waste (rice waste and meat and bone waste) and BSFL on growth performance and nutrient digestibility of broilers.

MATERIALS AND METHODS

Management of Broiler chicks: A total of 160 Cobb 500 one-day-old broiler chicks were weighed and tagged individually. The chicks were allocated randomly into 32 cages to form four treatments. Each treatment had eight replicates and there were 5 chicks per replicate. The experiment was conducted in a traditional open sided house provided with artificial light at night. Feed and water were

provided *ad libitum* throughout the experimental period. All birds were vaccinated against Newcastle disease and Gumboro disease at 7 and 21 days of age. Handling of birds was performed according to the protocol approved by the Institutional Animal Care and Use Committee (AUP-RO82/2016).

Experimental Diets: Food waste was done by pooling fresh food waste collected daily from restaurants in Serdang, into two parts. One part contained rice waste (RW), while the second part contained meat and bone waste (MBW). The food waste were then processed daily by washing using tap water, immersing in hot water at 90°C for 10 minutes, and then oven-drying at 60°C for two days (Hossein and Dahlan 2015). Live BSFL less than seven days old were bought and placed in the oven at 60°C for five days. Food waste-Black Soldier fly larvae (FWBSFL) was used as ingredients in the alternate diets. The conventional ingredients consisted of corn and soybean meal were included in the formulation of control diet based on a normal EPR of 154 for starter and 167 for finisher diets (Table 1). Three different levels of EPR using FWBSFL were formulated to form three dietary treatments (for the starter period 154, 143, and 166, and for the finisher period 167, 155, and 177). The FWBSFL diets contained less nutritional value with higher energy content compared to the control diets.

Growth performance: Body weight and feed intake of birds were recorded weekly. Subsequently, feed conversion ratio

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(FCR) was determined. The metabolizable energy (ME) of BSFL was determined according to the formula by Janssen (1989). Samples of BSFL, RW, and MBW were analyzed for proximate composition (AOAC 2005). BSFL was analyzed for amino acid composition (Rozan *et al.* 2000).

Nutrient digestibility: Two birds from each replicate at the ages of 18 and 39 days were transferred in clean cages for sampling. Feed and water were provided at *ad libitum*. Three days pre-slaughter, titanium oxide (TiO₂) was mixed in all diets at 5 g/kg as an indicator for nutrient digestibility. The birds were slaughtered at the end of 21 and 42 days of age. Ileal digesta samples were collected starting from the Merckel's diverticulum to the ileal-cecal-colon junction. The digesta samples were oven-dried at 60°C for two days and then stored at -20°C. Feed and digesta samples were dried and analyzed for nutrients composition (AOAC 1995) and TiO₂ content (Short *et al.* 1996). Gross energy of diets and digesta was determined by using a bomb calorimeter IKA C 200. Apparent nutrient digestibility was determined using the formula i.e. Apparent nutrient digestibility = $100 - [100 \times (\% \text{ TiO}_2 \text{ in feed} / \% \text{ TiO}_2 \text{ in digesta}) \times (\% \text{ nutrient in digesta} / \% \text{ nutrient in feed})]$ (Maynard and Loosli 1969).

Statistical analysis: Data were analyzed using the general linear models of SAS (Statistical Analysis System, version 9.4) in a completely randomized design. The differences among means were compared using Duncan's new multiple range test.

RESULTS AND DISCUSSION

Composition of alternate feed ingredients: BSFL and MBW contained high amount of protein, 55.99 and 50.9%, respectively (Table 2). The protein content of BSFL was higher compared to reported values of 40 to 42% (Arango Gutiérrez *et al.* 2004; St-Hilaire *et al.* 2007). BSFL contained low amount of fat (1.86%), whereas MBW had high fat content of 20.39%. Black soldier fly larvae can be stored for a long time due to their low-fat percentage. The fat content of BSFL was lower compared to reported level of 30 to 35% (Arango Gutiérrez *et al.* 2004; St-Hilaire *et al.* 2007). The energy and protein contents were affected by the drying method and larvae age (Aniebo and Owen 2010; Driemeyer 2016). BSFL in the current study had high crude protein and low fat because the BSFL used was less than 7 days old and was dried using the oven. BSFL generally contained high protein because the exoskeleton of BSFL contained chitin,

Table 1: Composition of experimental diets.

Ingredients, %	Starter diet				Finisher diet			
	T1 (control)	T2 (normal EPR)	T3 (low EPR)	T4 (high EPR)	T1 (control)	T2 (normal EPR)	T3 (low EPR)	T4 (high EPR)
Rice bran	4	4	4	4	7	7	7	7
Palm kernel cake	5	5	5	5	2	2	2	2
Meat and bone waste	0	19	23	15	0	17.1	20.9	14.4
Rice waste	0	56.5	52.5	60.5	0	59.4	55.6	62.1
Black soldier fly larvae	0	6	6	6	0	5	5	5
Corn	51.6	0	0	0	54.6	0	0	0
Soybean	23.5	0	0	0	20.6	0	0	0
Fish meal	6	0	0	0	6	0	0	0
Methionine	0.2	0	0	0	0.2	0	0	0
Lysine	0.2	0	0	0	0.1	0	0	0
Palm oil	5	5	5	5	5	5	5	5
Dicalcium phosphate	4	4	4	4	4	4	4	4
Salt	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3
Vitamin premix	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Calculated analysis								
ME (kcal/kg)	3025.50	3064.56	3085.36	3043.76	3106.72	3124.6	3144.37	3110.57
Crude Protein (%)	19.63	19.9	21.56	18.3	18.58	18.67	20.22	17.57
Energy to protein ratio (EPR)	154	154	143	166	167	167	155	177
Ash (%)	6.37	8.29	8.73	8.49	7.16	8.70	9.36	8.16
Fat (%)	7.27	8.72	9.41	8.52	7.68	10.42	10.44	9.35
Crude fiber (%)	2.66	1.99	2.34	2.41	2.89	1.83	2.11	2.31
Calcium (g kg ⁻¹)	11.2	10.3	10.6	10.0	11.1	10.2	10.5	10.0
Phosphors (g kg ⁻¹)	14.5	9.4	9.4	9.4	14.8	9.7	9.7	9.7
Methionine (g kg ⁻¹)	6.3	5.1	5.3	4.9	5.8	4.4	4.5	4.3
Lysine (g kg ⁻¹)	13.5	10.7	11.6	9.8	11.2	9.4	10.3	8.8

FWBSFL- Food waste-black soldier fly larvae; T1- normal EPR using conventional ingredients; T2- normal EPR using FWBSFL ; T3- low EPR using FWBSFL ; T4- high EPR using FWBSFL ; ME- Metabolizable energy; Vitamin premix-Supplied per kilogram of the diet: vitamin A (retinyl acetate), 8000 IU; vitamin D3 (cholecalciferol), 1000 IU; vitamin E (DL- α -tocopherol), 30 IU; vitamin K3 (menadione dimethylpyrimidinol, 2.5 mg; vitamin B1, 2 mg; vitamin B2, 5 mg; vitamin B6, 2 mg; vitamin B12, 0.01 mg; niacin, 30 mg; d-biotin, 0.045 mg; vitamin C, 50 mg; d-pantothenate, 8 mg, folic acid, 0.5 mg.

Table 2: Proximate composition of alternate feed ingredients.

Nutrient composition (%)	Rice waste	Black soldier fly larvae	Meat and bone waste
Dry matter	94.44± 0.34	95.00 ± 0.12	95.55 ± 0.21
Crude protein	10.1 ± 0.21	55.99 ± 0.49	50.9 ± 0.45
Crude fat	1.18 ±0.26	1.86 ± 0.10	20.39 ± 0.34
Ash	12.62 ± 0.02	0.81± 0.05	19.19 ± 1.12
Crude fiber	1.21 ± 0.24	Nil	Nil
ME (Kcal/Kg)	3402 ±0.88	696.3426 ±2.90	3789.42 ± 6.64

Values- mean ± standard error; ME- metabolizable energy.

Table 3: Amino acid composition of black soldier fly larvae.

Amino acids	(%)
Aspartic acid	4.808
Glutamic acid	6.581
Serine	3.288
Glycine	1.09
Histidine	1.476
Arginine	9.331
Threonine	2.236
Alanine	5.245
Proline	3.665
Tyrosine	2.697
Valine	2.193
Methionine	2.646
Cysteine	8.917
Isoleucine	1.222
Leucine	3.733
Phenylalanine	1.629
Tryptophan	0.049
Lysine	2.863

which contained nitrogen-hydrogen bonds (Waceko *et al.* 2016). The nitrogen content of BSFL meal increased the calculated protein content. BSFL contained 2.863% lysine and 2.646% methionine (Table 3).

Growth performance: Significantly ($P<0.05$) lower feed intake, body weight gain and live weight were observed in birds fed normal, high, and low EPR diets, respectively using FWBSFL compared to the control diets at day 21 (Table 1). Birds fed high EPR diets had higher ($P<0.05$) FCR compared to the control and low EPR diets. At day 42, birds fed normal, low, and high EPR diets had lower ($P<0.05$) feed intake, body weight gain and live weight compared to the control diet. FCR was not affected by the treatments. Growth performance in the overall period followed similar trend to those observed in birds at day 42.

It is recommended that the EPR of broiler diet is 154 for the starter diet and 167 for the finisher diet (Cobb Vantress 2013). Feed intake was lower in birds fed normal, low, and high level of EPR using FWBSFL than the control diet. The lower feed intake could be due to the reduced palatability of FWBSFL. It might be due to higher energy levels of FWBSFL diets compared to the control diet. The birds might have stopped eating once their energy needs were met leading to reduced feed intake. The decreased feed intake led to decreased final live weight and body weight gain in

birds fed FWBSFL diets. Yari *et al.* (2012) reported decrease of feed intake in birds fed high energy diets. In contrast, Dairo *et al.* (2010) reported a decreased feed intake in birds fed low-energy diets. Giachetto *et al.* (2003) found no effect of high-energy diets on bird live weight. Amino acid balance can also affect bird's growth performance in response to dietary energy (Classen 2017). Replacing corn and soybean meal with FWBSFL reduced methionine and lysine in the diets. Restriction of these amino acids in the FWBSFL diets resulted in reduced growth performance.

FWBSFL diets contained high levels of RW, which may contain anti-nutritional factors such as phytic acid (Noreen *et al.* 2009). Phytic acid in the rice was not affected by heat unlike other anti-nutritional factors (Juliano 1993). The presence of anti-nutritional factors in parboiled rice polish reduced the broiler growth performance (Azam and Howlader 1998; Islam 1996). In addition to this, MBW in the FWBSFL diets might have caused negative effects on the growth performance. In agreement with this finding, it was reported that inclusion of meat and bone meal in broilers diet up to 20% decreased growth performance (Wang and Parsons 1998). Eagleson (2015) also reported that broiler fed diets containing 10% meat and bone meal had decreased growth performance. On the other hand, Hossein and Dahlan (2015) reported that free-range village chicken could be fed diets containing 20% of dehydrated food waste without any negative effects on growth performance. Fast growing broiler chicken did not benefit from the high inclusion level of food waste.

Nutrient digestibility: FWBSFL diets contained less fiber compared to the control diet. Birds fed the normal and low EPR diets had increased ($P<0.001$) dry matter digestibility compared to the other treatments in the starter period at day 21 (Table 5). This finding was expected as the negative relation between nutrient digestibility and fiber content has been reported well by earlier workers (Ginindza and Norris 2017). Dietary treatments had no effect on digestibility of crude protein and crude fiber. Fat digestibility in birds fed FWBSFL diets was decreased ($P<0.05$) at day 21. However, Adeniji (2007) observed no effect on fat digestibility in broilers fed housefly maggot larvae at a level of 22%. Food waste may contain more saturated fat that was less digestible. Ash digestibility was higher ($P<0.001$) in birds fed normal, low and high EPR compared to the control diet.

Table 4: Effect of different levels of energy to protein ratio (EPR) on growth performance of broilers.

Parameters	T1(control)	T2(normal EPR)	T3 (low EPR)	T4 (high EPR)	SEM
0-21 days					
FI (g)	676.41 ^a	516.99 ^b	503.53 ^b	506.25 ^b	4.36 ^{***}
BWG (g)	453.48 ^a	306.38 ^b	332.75 ^b	283.03 ^c	4.37 ^{**}
LW (g)	759.54 ^a	565.11 ^b	591.17 ^b	554.50 ^c	1.38 ^{***}
FCR	1.50 ^b	1.69 ^{ab}	1.51 ^b	1.79 ^a	0.23 [*]
21-42 days					
FI (g)	2232.8 ^a	1603.8 ^b	1621.7 ^b	1400.6 ^b	1.33 ^{***}
BWG (g)	1177.04 ^a	857.25 ^b	907.95 ^b	797.38 ^b	1.13 [*]
LW (g)	2057.50 ^a	1692.86 ^b	1744.36 ^b	1603.07 ^b	2.17 ^{***}
FCR	1.90	1.87	1.79	1.76	0.11 ^{ns}
0-42 days					
FI (g)	2909.21 ^a	2118.55 ^b	1904.76 ^b	2127.17 ^b	1.17 ^{***}
BWG (g)	1627.1 ^a	1164.1 ^b	1247.1 ^b	1101.7 ^b	1.36 ^{**}
LW (g)	2057.50 ^a	1692.86 ^b	1744.36 ^b	1603.07 ^b	2.17 ^{***}
FCR	1.79	1.82	1.53	1.93	0.21 ^{ns}

Means within the same row with different superscripts (a, b, c) are significantly different; ns- means within the same row with no superscripts are not significantly different; *P<0.05; **P<0.01; ***P<0.001; FWBSFL - food waste with black soldier fly larvae; T1- control diet; T2- normal EPR using FWBSFL; T3- low EPR using FWBSFL; T4- high EPR using FWBSFL. FI- feed intake; BWG- body weight gain; LW- live weight; FCR- feed conversion ratio.

Table 5: Effect of different levels of energy to protein ratio (EPR) on nutrient digestibility of broilers.

Nutrient (%)	T1(control)	T2(normal EPR)	T3 (low EPR)	T4 (high EPR)	SEM
0-21 days					
Dry Matter	85.15 ^c	89.15 ^a	89.40 ^a	88.13 ^b	0.17 ^{***}
Crude protein	83.40	89.08	88.81	85.56	0.37 ^{ns}
Ether extract	82.80 ^a	74.10 ^c	81.99 ^{ab}	77.17 ^{bc}	2.86 [*]
Crude fiber	69.24	77.22	73.31	72.67	0.92 ^{ns}
Ash	84.10 ^d	86.50 ^b	87.06 ^a	85.91 ^c	0.17 ^{***}
21-42 days					
Dry Matter	89.74	89.64	89.27	90.34	0.32 ^{ns}
Crude protein	90.24 ^a	87.63 ^b	87.72 ^b	87.04 ^b	1.06 [*]
Crude fiber	81.80 ^b	81.52 ^b	94.32 ^a	88.81 ^{ab}	0.32 ^{**}
Ether extract	94.65	94.37	94.63	93.52	1.04 ^{ns}
Ash	85.95 ^c	86.57 ^{bc}	88.15 ^a	87.30 ^{ab}	0.30 ^{***}

Means within the same row with different superscripts (a, b, c) are significantly different; ns- means within the same row with no superscripts are not significantly different; *P<0.05; **P<0.01; ***P<0.001; FWBSFL- food waste with black soldier fly larvae; T1- control diet; T2- normal EPR using FWBSFL; T3- low EPR using FWBSFL; T4- high EPR using FWBSFL.

At day 42, protein digestibility was reduced (P<0.05) in the birds fed FWBSFL diets. The high percentage of RW in the FWBSFL diets might have reduced the nutrients digestibility as RW contained anti-nutritive factors. The high content of ash in MBW might have caused the negative effect on protein digestibility (Johnson *et al.* 1998). Protein efficiency ratio decreased with increasing ash content of meat and bone meal from 24% to 34% (Johnson and Parsons 1997). In contrast to the results of present study, Chen *et al.* (2007) observed increased protein digestibility in chickens fed dehydrated food waste at 20% level in the diets. Digestibility of crude fiber in birds fed low EPR (high protein content) was the highest (P<0.01) compared to the other treatments at day 42. Similarly, Ratriyanto *et al.* (2014) also observed increased crude fiber digestibility in broilers fed high-protein diets compared to low-protein diets. Chen *et*

al. (2007) reported increased crude fiber digestibility due to the inclusion of dehydrated food waste in the diet of Taiwan native chickens. Ash digestibility was lower (P<0.001) in birds fed normal and high EPR diets compared to the control diets. However, dietary treatments had no effect on dry matter and fat digestibilities at day 42. The low fiber content in the FWBSFL diets and possible anti-nutritive factors in rice waste might have resulted in reduced nutrient digestibility.

CONCLUSION

Formulating broiler diets with different levels of EPR using FWBSFL decreased growth performance and nutrient digestibility. Therefore, it is necessary to consider the dietary balance of essential amino acid and fiber as well as safe level of anti-nutritive factors when using food waste and BSFL in formulating the broiler diets.

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