

Substitutions of Soybean Meal with Enriched Palm Kernel Meal in Laying Hens Diet

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ABSTRAK

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Serangkaian penelitian dilakukan untuk menggantikan bungkil kedelai (SBM) dengan bungkil inti sawit (PKC) dalam ransum ayam petelur. Tahap pertama dilakukan untuk meningkatkan kandungan protein dan asam amino BIS melalui proses fermentasi dan dilanjutkan dengan penambahan enzim untuk meningkatkan pencernaan asam amino. Selanjutnya dilakukan uji biologis untuk mengetahui efektifitas PKC yang sudah difermentasi (FPKC) dan ditambahkan enzim (EFPKC) untuk menggantikan SBM didalam ransum ayam petelur. Nilai energy (AME) dari PKC, FPKC dan EFPKC diukur dengan menggunakan ayam broiler dan dilanjutkan dengan pengukuran nilai asam amino tercerna pada ileal (IAAD). Nilai AME dan IAAD dari EFPKC kemudian digunakan untuk meramu ransum penelitian. Ransum diberikan pada ayam petelur umur 51 minggu selama 8 minggu. Lima (5) jenis ransum disusun dengan kandungan gizi yang sama, tetapi SBM diganti dengan EFPKC secara bertingkat. Ransum perlakuan terdiri dari 1. Kontrol (tanpa EFPKC), 2. 25% SBM dalam ransum Kontrol diganti dengan EFPKC, 3. 50% SBM dalam ransum Kontrol diganti dengan EFPKC, 4. 75% SBM dalam ransum Kontrol diganti dengan EFPKC and 5. 100% of SBM dalam ransum Kontrol diganti dengan EFPKC. Setiap ransum perlakuan diberikan pada 24 ekor ayam (6 ulangan, 4 ekor/ulangan). Hasil penelitian menunjukkan bahwa fermentasi PKC meningkatkan protein kasar dan asam amino, kecuali threonin dan arginin, tetapi menurunkan AME. Penambahan enzim pada FPKC meningkatkan nilai IAAD. Akan tetapi hanya enzim BS4 yang dapat meningkatkan nilai AME pada EFPKC. Uji biologis menunjukkan bahwa sekitar 25 hingga 50% bungkil kedelai didalam ransum dapat diganti dengan bungkil inti sawit yang sudah difermentasi dan ditambahkan enzim tanpa menyebabkan gangguan yang berarti pada performan ayam petelur.

Kata Kunci: Bungkil Kedelai, Bungkil Inti Sawit, Fermentasi, Enzim, Produksi Telur

ABSTRACT

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A series of experiment was conducted in order to substitute soybean meal (SBM) with palm kernel cake (PKC) as a protein source in laying hens diet. First experiment was to increase its protein and amino acids content by fermentation process and followed by enzymes supplementation to improve nutrient digestibilities. Second experiment was conducted to evaluate the effectiveness of enzyme- supplemented fermented palm kernel cake (EFPKC) to replace SBM in laying hens diet. The energy (AME) of the PKC, the fermented PKC (FPKC) and the EFPKC was measured by ileal amino acids digestibility (IAAD) in broilers. The AME and the IAAD values of the EFPKC were used for diet formulation in the feeding trial. A feeding trial was performed in laying hens, aged 51 weeks for 8 weeks egg production. Five diets with different levels of substitution of SBM with EFPKC but similar nutrient contents were formulated, i.e.: 1. Control (without EFPKC), 2. 25% of SBM in control diet substituted with EFPKC, 3. 50% of SBM in control diet substituted with EFPKC, 4. 75% of SBM in control diet substituted with EFPKC and 5. 100% of SBM in control diet substituted with EFPKC. Each diet was fed to 24 hens (6 replicates of 4 birds/replicate). Results of the experiment showed that the fermentation of PKC increased the crude protein and most of the amino acids contents except the threonine and arginine, but decreased its AME. Supplementation of enzymes (BS4 or CE) improved the ileal amino acid digestibilities of the fermented PKC. However, only BS4 enzymes increased the AME of the EFPKC. About 25% to 50% of the SBM in the diet could be substituted with the EFPKC without any detrimental effect on the performances of laying hens.

Key Words: Soybean Meal, Palm Kernel Cake, Fermentation, Enzyme, Egg Production

INTRODUCTION

Soybean meal (SBM) is commonly used as a protein source in poultry feed due to its high protein and amino

acids contents and digestibilities. The increasing demand for SBM due to the increasing world poultry feed production, stimulate the increase of SBM and the feed prices. Therefore, some attempts have been

reported to replace the SBM with some non conventional feedstuffs such as rapeseed meal (Leeson et al. 1987; Ciurescu 2009), sunflower seed meal (Shi et al. 2012), fermented cotton seed meal (Azman & Yilmaz 2005) and peanut meal (Pesti et al. 2003).

Palm kernel cake (PKC) is produced abundantly in tropical countries such as Indonesia, Malaysia, Nigeria and other countries. The world PKC production is estimated at 8,345 million ton in year 2014. The PKC is a by product of palm kernel oil production, mostly produced by mechanical extraction of the oil from the palm kernel seed. The nutritive values of the PKC, including its digestible amino acids values have been reported by some authors (Nwokolo et al. 1977; Onwudike 1986; Sue and Awaludin 2005; Sundu et al. 2006). The protein, amino acids and the digestible amino acids of the PKC are much lower than the SBM. The crude protein, lysine and methionine contents of SBM varies from 42.74-50.71%, 2.0-3.36% and 0.58-0.90 %, respectively (Clarke and Wiseman 1999) while the crude protein, lysine and methionine contents of the PKC varies from 14-21%, 0.59-0.69% and 0.30-0.47 %, respectively. Therefore, the replacement of SBM with PKM in poultry diet formulation will disturb the nutrient balances of the diet, unless other feed ingredients or synthetic nutrients such as essential amino acids are adjusted.

Some efforts have been reported to increase the protein and amino acids contents of the PKC and its nutrients digestibility in order to increase the inclusion of PKC in the diet and reduce the inclusion of the SBM in poultry diet. Iyayi & Aderolu (2004) improved the crude protein of the PKC from 16.0 to 21.11% and the metabolizable energy (ME) from 2610 kcal/kg to 2840 kcal/kg by fermentation with *Trichoderma viridae*. Supplementation of enzyme in the diet containing PKC have been reported to increase the TME_n of the diets (Iyayi & Davies 2005; Chong et al. 2008). Recently, Saenphoom et al. (2013) reported that enzyme supplementation (mainly cellulase and mannanase) increased the TME and TME_n in PKC. In order to improve the nutrient contents and digestibility of the PKC, a study was designed to ferment the PKC, followed with exogenous enzyme supplementation. The inclusion of the processed PKC in poultry diet is expected to reduce the inclusion level of SBM in the diet.

MATERIALS AND METHODS

Improvement of the nutritive values of the palm kernel cake

Palm kernel cake (PKC) used in this study was obtained from a commercial feed mill in Bekasi, West

Java. Prior to the treatments, the PKC was sieved with 2 mm mesh in order to reduce its shell contents as described by Sinurat et al. (2013) and then fermented with *Aspergillus niger* in attempt to increase its protein and amino acids content following the procedures described by Pasaribu (2013 unpublished). The PKC and the fermented PKC (FPKC) were supplemented with 2 (two) kind of enzymes, i.e., a crude enzymes produced in our laboratory (BS4) at 20 ml/kg feedstuff and a commercial multi enzymes (CE) at 2 g/kg feedstuff. The commercial enzymes according to the official brochure consists of cellulase 6,000 U/g, xylanase 10,000 U/g, glucanase 700 U/g, phytase 400 U/g, α -amylase 700 U/g, protease 3,000 U/g, pectinase 70 U/g and lipase 5 U/g while the BS4 enzymes consist of β -mannanase, CMCase (cellulase), β -mannosidase, β -glucosidase and α -galactosidase (Purwadaria et al. 2003). Assay on saccharification activity on palm kernel meal of the commercial enzyme and the BS4 enzyme was similar, i.e., 632.1 u/ml and 641.1 unit/g, respectively (Pasaribu et al. 2009). Previous report indicated that 2 g CE enzyme/kg or 20 ml BS4 enzyme/kg were effective to improve the dry matter and protein digestibility as well as metabolizable energy of PKC (Sinurat et al. 2013).

In this experiment, each ingredient (PKC and FPKC) was mixed with a basal diet with the ratio 50:50 and 2% ash insoluble ash (celite) as indicator. Five (5) dietary treatments were mixed for the digestibility trial, i.e., 1. Basal diet, 2. PKC without enzyme supplementation, 3. FPKC without enzyme supplementation, 4. FPKC + 20 ml BS4 enzyme/kg and 5. FPKC + 2 g commercial enzyme/kg. Each diet was fed ad libitum to 6 (six) male broiler chickens aged 28 d reared in individual wire cages.

Three days after feeding the test diets, excreta were collected by placing plastic trays underneath each cage in order to measure the metabolisable energy (AME) of test ingredients. The excreta was dried in oven at 70 °C and its gross energy was determined with bomb calorimeter. Six (6) replicates were assigned for each test diet for the AME determination.

The digestibility of the amino acids in the PKC and the FPKC were measured following the procedure described by Ravindran et al. (2005). Six (6) days after feeding the test diet, the animals were sacrificed by CO₂ asphyxiation and the digesta in the ileal was collected into plastic containers, pooled within the same diet and immediately kept in the freezer for further chemical analyses. The frozen ileal digesta were freeze-dried prior to analyses of amino acids. Diets and the digesta were analysed for dry matter, AIA and crude protein according to procedures of AOAC (2005) while amino acids analyses were carried out by HPLC at Bogor Agricultural University laboratory.

Calculations

Apparent ileal amino acid digestibility and AME were calculated following the formula:

Amino acid digestibility coefficient of the diet:
 $= ((AA/AIA)_d - (AA/AIA)_i) / (AA/AIA)_d \times 100\%$

Where:

$(AA / AIA)_d$ = ratio of amino acid to acid-insoluble ash in the diet,

$(AA/AIA)_i$ = ratio of amino acid to acid-insoluble ash in the ileal digesta.

The amino acid digestibility of test ingredient:

$= (2 \times \text{AA digestibility of test diet}) - \text{AA digestibility of basal diet}$

And the calculation of the metabolizable energy (AME) was calculated as follow:

AME of diet = GE diet - ((AIA in diet / AIA in Excreta) X GE in Excreta)

The AME of the test ingredient:

$= (2 \times \text{AME of test diet}) - \text{AME of the basal diet}$

Where:

ME = Apparent metabolisable energy (kcal/kg)

GE = Gross energy (kcal/kg)

AIA = Acid insoluble ash (%)

Substitution of soybean meal with fermented PKC in laying hens diet

A feeding trial was carried out to study the effect of substituting soya bean meal with enzymes-supplemented FPKC (EFPKC) in the diet on the performance of laying hens. The FPKC was supplemented with BS4 enzymes at 20 ml/kg FPKC and its nutritive values (i.e., the AME and the ileal digestible amino acids values obtained from the digestibility trial) were used in the formulation of the diets, while nutrient values of other ingredients were based on the normal values used in commercial diet formulation in Indonesia. Five dietary treatments, i.e., 1. Control diet (without EFPKC), 2. Diet with 25% substitution of SBM (substituted with EFPKC), 3. Diet with 50% substitution of SBM (substituted with EFPKC), 4. Diet with 75% substitution of SBM (substituted with EFPKC) and 5. Diet with 100% substitution of SBM (substituted with EFPKC). In order to maintain the similar nutrient values of the treatment diets, reduction of SBM in diets were followed by an increase in the EFPKC level. All diets were formulated with similar nutrient levels, i.e., ME 2650 kcal/kg, crude protein 17%, Ca 4.1%, Available P 0.35%,

Table 1. Ingredients of the experimental diets in the feeding trial (%)

Ingredients	Control	25% SBM Substituted	50% SBM Substituted	75% SBM Substituted	100% SBM Substituted
Maize	48.45	52.41	46.790	50.58	47.98
Hominy/corn bran	17	7.86	9.730	0.84	1
Calcium carbonate	9.37	9.00	8.480	7.96	7.19
Soya bean meal	20	15.00	10.000	5	0
Fermented PKC + Enzyme (EFPKC)	0	9	15	23	27.7
Meat and bone meal	3.61	5.00	7.000	9	12
DCP	0.42	0.00	0.00	0.00	0.00
Vegetable oil	0.5	0.85	2.000	2.5	3
DL Methionine	0.18	0.26	0.290	0.31	0.37
L-Lysine	0	0.15	0.240	0.34	0.29
Vitamin mixture	0.03	0.03	0.03	0.03	0.03
Mineral mixture	0.04	0.04	0.04	0.04	0.04
Sodium Bicarbonate	0.1	0.1	0.1	0.1	0.1
Salt	0.2	0.2	0.2	0.2	0.2
Choline Chloride	0.1	0.1	0.1	0.1	0.1
Total	100.0	100.0	100.0	100.0	100.0

digestible lysine 0.702 %, digestible methionine 0.387%, digestible methionine + cystine 0.600% and digestible tryptophane 0.164%. The crude fibre of the diets according to the calculated values were 3.9, 4.7, 5.7, 6.4 and 7.1% for diet 1, 2, 3 and 4, respectively. The composition of the experimental diets is presented in Table 1.

Each diet was fed to 24 (6 replicates of 4 birds/replicate) Isa Brown laying hens aged 51 weeks old. All birds were placed in individual cages, but each four birds were equipped with one feeder. Therefore, 4 birds were considered as a replicate. Feed and water were given ad libitum and the trial was lasted for 8 (eight) weeks. Performance of the laying hens such as egg production (HD dan HH), feed intake, and feed conversion ratio were recorded weekly. Egg qualities (yolk color score, HU and egg shell thickness) were determined at 5 weeks after treatment. Body weight changes due to the treatments were also determined by weighing the birds individually before and after treatment. The data were analysed with analyses of variance in a completely randomized design (5 treatments x 6 replicates). Differences between treatments were examined with Duncan multiple range test when the analyses of variance showed a significant difference at $P < 0.05$ (Steel & Torrie 1997).

RESULTS AND DISCUSSION

Enrichment of nutritional values of palm kernel cake (PKC) by fermentation and enzyme supplementation

As shown in Table 2, fermentation of PKC with *A. niger* reduced the crude fibre slightly, increased the crude protein from 14.70 % to 20.04 % or 36.3 % improvement. Most of the amino acids such as aspartic acid, glutamic acid, serine, histidine, alanine, tyrosine, methionine, valine, phenylalanine, isoleucine, leucine, lysine and tryptophan contents were also increased after fermentation. However, threonine and arginine contents were decreased by fermentation process. This result was in contrast to the report of Muhammad and Oleyede (2009) which showed that fermentation of *Terminalia catappa* seed meal with *A. niger* reduced almost all the amino acids content, except the glutamic acid, alanine and phenylalanine. Improvements on the nutrient values of the FPKC was mainly as the effect of *A. niger* cells proliferation and some metabolites such as enzymes produced during the growing process (Supriyati et al. 1998).

Table 2. Crude fibre, Protein and amino acids composition of palm kernel cake (PKC) and fermented PKC (FPKC) with *A. niger* (% dry matter)

	Palm kernel cake (PKC)	Fermented palm kernel cake (FPKC)	Improvement due to fermentation (%)
Crude Fibre	15.09	14.76	- 2.2
Crude Protein	14.70	20.04	36.3
Aspartic acid	1.251	1.525	21.9
Glutamic acid	2.869	3.148	9.7
Serine	0.669	0.758	13.4
Histidine	0.333	0.394	18.3
Glycine	0.424	0.757	78.2
Threonine	0.695	0.473	-32.0
Arginine	1.191	1.137	-4.6
Alanine	0.631	0.983	55.7
Tyrosine	0.311	0.602	93.3
Methionine	0.205	0.216	5.2
Valine	0.731	0.857	17.3
Phenilalanine	0.677	0.766	13.3
Isoleucine	0.475	0.559	17.6
Leucine	0.865	0.968	12.0
Lysine	0.614	0.678	10.4
Tryptophan	0.102	0.221	117.2

The ileal amino acids digestibility (IAAD) of palm kernel cake (PKC) and fermented palm kernel cake (FPKC) with or without enzyme supplementation are shown in Table 3. Fermentation of the PKC increased the crude protein digestibility and most of the IAAD, except the IAAD of serine, threonine, alanine, valine and phenylalanine. Muangkeow and Chinajariyawong (2009) also reported an increase in the true amino acids digestibility except for arginine of the PKC after fermentation. Supplementation of the BS4 enzyme or the commercial enzyme into the FPKC improved the protein digestibility and all the amino acids digestibilities. The ileal crude protein digestibility of the FPKC were greater when supplemented with BS4 enzyme (91.39%) as compared to the commercial enzyme (65.26%). Some amino acids, particularly the indispensable amino acids (arginine, histidine and leucine) digestibility values were greater than 100%, which is difficult to explain. Perhaps, it could be explained that enzyme supplementation in to the FPKC also increased the amino acids digestibility of other feedstuffs included in the basal diet, since the tested ingredients were mixed with a basal diet (50 : 50) in this assay and these increments were counted as the

improvement of the FPKC. More studies or assays should be performed to explain this. An IAAD assay which use the tested ingredient as the sole source of protein in the diet as reported by Huang et al. (2006) could be a good tool to explain this.

The results showed that the tryptophan digestibility of the PKC and the FPKC were negatives, i.e., -25.26% and -40.24%, respectively. It could not be explained why this happened. Perhaps, more replicates of laboratory analyses should be performed to validate the values. Nwokolo et al. (1976) and Muangkeow & Chinajariyawong (2009) did not report the tryptophan digestibility value of the PKC in poultry. Sulabo et al. (2013) reported high values of tryptophan apparent ileal digestibility (82.3-84.1%) of palm kernel cake measured in growing barrows. However, the FPKC supplemented with enzymes, either BS4 or the commercial enzyme showed high tryptophan digestibilities, i.e., 107.53 and 92.11 %, respectively.

As shown in Table 3, the AME value of the PKC was decreased (P<0.05) after the fermentation. The AME of the fermented PKC was increased when supplemented with the BS4 enzyme, but not with the

Table 3. Metabolisable energy (AME), Ileal protein- and amino acids digestibility coefficient of palm kernel cake (PKC) and fermented PKC supplemented with enzymes

Nutrients	PKC*	FPKC*	FPKC+BS4*	FPKC+CE*
AME [#] (Kcal/kg)	2074 ^a	1788 ^b	1867 ^{ab}	1752 ^{ab}
Protein digestibility, %	34.08	49.79	91.39	65.26
Aspartic acid, %	33.39	49.19	85.42	95.94
Glutamic acid, %	48.07	65.43	89.46	101.21
Serine, %	49.10	44.81	80.89	92.24
Histidine, %	56.73	71.45	96.83	106.07
Glycine, %	48.21	55.24	81.64	96.37
Threonine, %	42.65	30.87	77.05	88.50
Arginine, %	74.16	66.14	87.85	102.61
Alanine, %	55.84	47.45	77.98	88.43
Tyrosine, %	37.31	57.36	88.65	98.51
Methionine, %	44.42	64.83	71.17	99.83
Valine, %	66.82	61.67	85.36	96.01
Phenylalanine, %	67.98	62.45	81.49	93.34
Iso-leucine, %	30.20	91.92	116.97	134.64
Leucine, %	23.16	91.03	114.60	131.14
Lysine, %	60.39	62.20	80.23	84.44
Tryptophan, %	-25.56	-40.24	107.53	92.11

[#] AME = Apparent metabolisable energy (measured with 6 replicates) and different superscript indicated significantly different (P<0.05)

*PKC = palm kernel cake; FPKC = fermented PKC; BS4 = BS4 enzyme; CE = commercial (multi) enzyme

commercial enzyme. This result indicated that the BS4 enzyme is more suitable to improve the AME values of the FPKC. Muangkeow & Chinajariyawong (2009) reported also that the AME of the palm kernel meal was decreased from 2202 to 2080 kcal/kg, after fermented with *A. wentii*. Similar trend was also occurred on the TME values as the effect of the fermentation. However, Bintang et al. (1999) reported an increase on the AME of the PKC after fermented with *A. niger* measured on growing ducks. Similar process of fermentation was also reported to increase the AME values of palm oil sludge or solid heavy phase measured on laying hens (Sinurat et al. 2007).

Both enzymes (the BS4 and the commercial enzyme) have been reported effectively to increase the dry matter digestibility and the TME of palm oil sludge, but only the BS4 enzyme was effective to increase the protein digestibility (Pasaribu et al. 2009).

The effect of substitution of dietary soybean meal with enzyme- supplemented fermented palm kernel cake (EFPKC) on the performance of laying hens

The performance of the laying hens during the 8 (eight) weeks production period is presented in Table 4, 5 and 6. As shown in Table 4, the body weight of birds before the treatments were similar ($P>0.05$). However, after eight (8) weeks feeding trial the body weight of the birds were significantly ($P<0.001$) affected by the dietary treatments. All the birds showed a reduction in body weight or a negative body weight changes. The more the soybean meal substituted by the EFPKC, the more the reduction in body weight ($P<0.002$), although only significant at 100% SBM substitution.

Unlike in the broiler production, the reduction in body weight on laying hens is not an issue, as far as the egg production and the feed efficiency are not impaired. The reduction in body weight is a reflection of inadequate nutrients, especially the energy and the protein or amino acids intake. In this experiment, the highest body weight loss (-309.3 g/bird/8 weeks) was found on birds fed without soybean meal or 100% substitution of soybean meal with the EFPKC. All the

experimental diets were formulated to have a similar nutrient contents such as the AME, crude protein, digestible amino acids, Ca and available P that met the requirement of laying hens. Therefore, the body weight lost during the experiment may be due to a reduction in feed intake, hence nutrients intake (Table 5). There are many factors that affect the feed intake in laying hens. Some of the dietary factors include the nutritional balance, flavor and the presence of an antinutritional substances. Results of this study showed that the more soybean meal substituted with the EFPKC, the less the feed intake. It seems that the flavor of the EFPKC reduced the palatability of the diet. Some reports showed that inclusion of high levels of fermented feed (Engberg et al. 2009) or fermented feedstuffs such as palm kernel meal (Iyayi & Aderolu 2004), cassava peel (Osei et al. 1990), palm oil sludge (Fenita et al. 2010) in the diet reduced the feed intake of laying hens. Although some authors also reported that inclusion of fermented feedstuffs in the diet did not affect the feed intake of laying hens (Loh et al. 2007; Dairo & Fasuyi 2008).

The average hen-day egg production, feed intake and the feed conversion ratio of the birds were significantly ($P<0.001$) affected by the substitution of the soy bean meal with the EFPKC. As shown in Table 5, the results showed that the substitution of 25% soybean meal in the diet did not significantly ($P>0.05$) impair the HD egg production, the FCR and the egg weight, although the feed intake was significantly ($P<0.05$) decreased. Further substitution of the soybean meal to 50% has shown a slightly reduction in the HD egg production, FCR and the egg weight, although the differences with the control were not statistically significant ($P>0.05$). However, substitution of 75% and 100% soybean meal with the EFPKC has shown a significantly ($P<0.05$) reduction in the HD egg production and impaired the FCR, but not on the the egg weight ($P>0.05$). The results also showed that the feed intake was significantly ($P<0.001$) reduced when the soybean meal was substituted with the EFPKC. The more the soybean meal was substituted with the EFPKC, the more the feed intake was reduced.

Table 4. Body weight changes of laying hens after fed with enzyme supplemented-FPKC to replace soy bean meal for 8 weeks

Treatments	Initial body weight at 52 weeks of age, g	Final body weight at 60 weeks of age, g	Body weight change, g
Control (100% SBM)	1924	1854 ^a	69.7 ^a
25% SBM substituted	1920	1810 ^b	-101.3 ^a
50% SBM substituted	1916	1790 ^b	-127.0 ^a
75% SBM substituted	1910	1672 ^b	-185.7 ^a
100% SBM substituted	1857	1611 ^c	-309.3 ^b
Significance (P)	0.702	0.0001	0.002

Table 5. Performances of laying hens fed with soya bean meal substituted with enzyme supplemented- FPKC for 8 weeks

Treatments	Egg production (% HD)	Feed intake (g/b/d)	FCR (g/g)	Egg Weight (g/egg)
Control (100% SBM)	90.7 ^a	117.9 ^a	2.02 ^c	64.8 ^a
25% SBM substituted	90.6 ^a	111.4 ^b	1.97 ^c	62.6 ^a
50% SBM substituted	87.8 ^a	111.5 ^b	2.08 ^c	61.6 ^a
75% SBM substituted	75.9 ^b	106.6 ^b	2.36 ^b	60.1 ^a
100% SBM substituted	63.8 ^c	102.3 ^c	2.75 ^a	60.6 ^a
Significance (P)	0.001	0.0001	0.001	0.062

Different superscript in the same column indicated significantly different ($P < 0.05$).

Table 6. Egg quality of hens as affected by replacing soy bean meal with EFPKC in the diet

Treatments	Egg shell thickness, mm	HU	Yolk Color Score
Control (100% SBM)	0.34 ^a	93.5 ^a	6.3 ^b
25% SBM replaced	0.32 ^{abc}	96.8 ^a	8.2 ^a
50% SBM replaced	0.33 ^{ab}	94.8 ^a	7.9 ^a
75% SBM replaced	0.31 ^c	95.7 ^a	8.2 ^a
100% SBM replaced	0.32 ^{bc}	95.8 ^a	8.0 ^a
Significance (P)	0.013	0.348	0.001

Some reports have shown that substitution of 25 % (Ojewola & Ozuo 2006) or 50% (Dairo & Fasuyi 2008) and even 100 % (Mirnawati et al. 2011) soy bean meal with fermented palm kernel cake in the diet did not show negative effects on the performance of broilers. The present study, however, suggests that 25 to 50% of the soybean meal could be substituted with the EFPKC in the laying hens diet.

As shown in Table 6, the substitution of the soybean meal with the EFPKC in laying hens diet did not significantly ($P > 0.05$) affect the egg shell thickness and the HU, but significantly ($P < 0.05$) affect the yolk color scores. The yolk color scores were significantly improved when the EFPKC was included in the diet. Previous study also reported a similar trend when the PKC (Zanu et al. 2011) or the fermented PKC (Fenita et al. 2010) or other palm oil by-product such as palm oil sludge (Sinurat et al. 2007) are included in the diet of laying hens. Palm oil fruits contain high carotenoid as pigment sources (Sundram et al. 2003). Therefore, palm oil by products may also contain some carotenoids that contribute to improve the yolk color of eggs produced by hens fed with the fermented palm kernel cake.

CONCLUSION

This study concluded that the fermentation of palm kernel cake with *A. niger* increased the crude protein and most of the amino acids contents except the threonine and arginine, but decrease its AME. Supplementation of enzymes (BS4 or CE) improved the ileal amino acids digestibilities of the fermented PKC. However, only BS4 enzymes increased the AME of the fermented PKC.

About 25% to 50% of the soybean meal in the diet could be substituted by the enzyme- supplemented palm kernel cake (EFPKC) without any detrimental effect on the performances of laying hens.

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