Effect of Enzyme Supplementation on Nutritive Values of Fermented Palm Kernel Cake Used to Substitute Soybean Meal in Broiler Diet

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(received 27-06-2015; revised 03-08-2015; accepted 21-09-2015)

ABSTRAK

Suatu penelitian dirancang untuk meningkatkan nilai gizi bungkil inti sawit (PKC) dengan proses fermentasi yang diikuti dengan penambahan enzim untuk menggantikan penggunaan bungkil kedelai dalam ransum ayam broiler. Percobaan pertama menggunakan rancangan faktorial 2 x 2 untuk mengetahui pengaruh proses fermentasi (PKC dan PKC terfermentasi atau FPKC) dan pengaruh penambahan enzim (tanpa enzim dan ditambah enzim BS4). Kecernaan bahan kering, energi metabolis (AME) dan kecernaan asam amino (IAAD) keempat bahan tersebut diukur dengan menggunakan ayam broiler jantan. Sebanyak 7 ulangan digunakan untuk mengukur kecernaan bahan kering dan AME, sedangkan untuk IAAD hanya 3 ulangan. Percobaan kedua dirancang untuk menggantikan bungkil kedelai dengan FPKC yang ditambah dengan enzim BS4 (EPFKC). Empat jenis ransum percobaan, yaitu kontrol (tanpa EFPKC) dan penggantian 10%, 20 dan 40% bungkil kedelai dengan EFPKC disusun dengan kandungan gizi yang sama dan memenuhi kebutuhan gizi ayam broiler. Ransum diberikan pada ayam broiler umur 1 hingga 35 hari, masing-masing dengan 6 ulangan. Bobot badan, konsumsi ransum, FCR dan mortalitas diukur selama percobaan. Pada akhir penelitian, diukur persentase karkas, lemak abdomen, bobot hati dan rempela. Hasil menunjukkan bahwa fermentasi PKC meningkatkan kecernaan bahan kering (P<0.05) dan AME meskipun tidak nyata (P>0.05). Penambahan enzim tidak nyata (P>0.05) mempengaruhi kecernaan bahan kering dan AME. Penambahan enzim nyata (P<0.05) menurunkan AME. Namun, penambahan enzim tidak nyata (P>0.05) mempengaruhi IAAD asam amino essensil. Penggantian bungkil kedelai dengan EFPKC menyebabkan penurunan konsumsi pakan dan pertumbuhan pada ayam broiler.

Kata Kunci: Bungkil Inti Sawit, Fermentasi, Enzim, Broiler, Bungkil Kedelai

ABSTRACT

Sinurat AP, Purwadaria T, Purba M. 2015. Effect of enzyme supplementation on nutritive values of fermented palm kernel cake used to substitute soybean meal in broiler diet. JITV 20(3): 184-192. DOI: http://dx.doi.org/10.14334/jitv.v20i3.1185

Two experiments were designed to improve nutritional values of palm kernel cake (PKC) by biofermentation process, followed by enzyme supplementation to substitute soybean meal (SBM) in broilers diet. A factorial of 2 x 2 design was applied in the first experiment, i.e. fermentation process (non fermented PKC and fermented PKC) and enzyme supplementation (no enzyme and +BS4 enzyme). Dry matter (DM) digestibility, AME and amino acids ileal digestibility (IAAD) of the treatment ingredients were measured in broiler chickens. Seven replicate were applied for the DM and AME assays and 3 replicate for IAAD assay. Second experiment was designed to study the effect of SBM substitution with enzyme supplemented FPKC (EPFKC). Four diets were formulated, i.e., control diet without EFPKC, 10%, 20 and 40% SBM substituted with EFPKC. All diets were formulated to meet the nutrient requirements of broilers. Each diet was fed to broilers from 1 to 35 d. Body weight, feed consumption, FCR and mortalities were measured. Carcass yield, abdominal fat and weight of liver and gizzard were measured at the end of experiment. Results showed that fermentation of PKC increased the DM digestibility, the AME was also increased but not significant. Enzyme supplementation did not affect the DM digestibility and AME of PKC. Fermentation process significantly (P<0.05) decreased IAAD of some indispensable amino acids. However, supplementation of enzyme did not affect the IAAD of indispensable amino acids. Substitution of soybean meal with EFPKC reduced the feed intake and growth rate of broilers.

Key Words: Palm Kernel Cake, Fermentation, Enzyme, Broilers, Soybean Meal

INTRODUCTION

Modern chickens (broilers and layers) require high quality feed to support their rapid growth, high productivity and high efficiency in feed utilization. Soybean meal (SBM) is one of good quality feedstuff that commonly used as a protein and or amino acids source in chickens diet. Its inclusion in broiler’s diet varies between 15 to 30%, depends on the presence of other protein sources included in the diet. Even in countries
with no SBM production such as Indonesia, SBM is commonly used in poultry diet. Increases of national broiler production causes increases in importation of SBM and price of feed. Indonesia imported 3.069 million ton SBM in 2010 and 4.250 million ton in 2014 (USDA 2015). In order to achieve self sufficiency in feed ingredients, it is important to seek alternatives to limit or replace SBM with local feed ingredients.

Some studies have been reported to reduce SBM in broiler’s diet with the more cheaper ingredients such as rapeseed meal (Taraz et al. 2006; Riyazi et al. 2008; Ciurescu 2009), sunflower seed meal (Shi et al. 2012), fermented cotton seed meal (Azman & Yilmaz 2005) and peanut meal (Dhawale 2005).

As a world leading producer of palm oil, Indonesia produces palm kernel cake (PKC)- a by product of palm kernel oil production in a large quantity. The Indonesian PKC production in 2014 was estimated 4.55 million ton (USDA 2015). Its nutritive values including its digestible amino acids have been reported by some authors (Nwokolo et al. 1977; Onwudike 1986; Sue & Awaludin 2005; Sundu et al. 2006; Sinurat et al. 2014). The protein, amino acids and the digestible amino acids of the PKC are much lower than the SBM. Replacing the SBM with PKC as such for a protein source in poultry diet will deteriorate their performances. Sinurat et al. (2014) reported that the PKC could be fermented with Aspergillus niger to increase its crude protein from 14.76% to 20.04%. Some amino acids (except threonine and arginine) content increased varies between 5.2-117.2% and also its protein and ileal amino acids digestibility.

Although, the protein and amino acids contents and the ileal amino acids digestibility of the fermented PKC were higher than the PKC, it is still not comparable with those of the SBM. The feeding trial on laying hens showed that only 25-50% of the SBM could be substituted with enzyme supplemented fermented PKC without significantly affecting egg production and feed efficiency (Sinurat et al. 2014). In order to increase protein and amino acids content of PKC, Sinurat et al. (2015) have modified the fermentation method by adding cassava leaf meal as protein sources prior to PKC fermentation. This new method was able to increase protein content of fermented PKC from 21.91 to 28.97% as compared to the previous method (Sinurat et al. 2014). The methionine, lysine, tryptophan and threonine levels were also increased from 0.290 to 0.317%, 0.643 to 0.740, 0.150 to 0.273 and 0.31 to 0.273%, respectively.

Enzymes have been widely used to increase nutrients availability of poultry feedstuff or feed. Enzyme complex (consist of xylanase, beta-glucanase and cellulose) have been reported to increase the crude protein digestibility and the AME of feed containing paddy and rice bran (Kang et al. 2013). Khan et al. (2006) also reported an increase on the dry matter-, organic matter-, crude protein and energy-digestibilities of feed containing sunflower meal caused by supplementation of commercial enzymes. A multi enzyme produced by Eupenicillium javanicum has been developed in our laboratory (Purwadaria et al. 2003; Pasaribu et al. 2009). The enzyme, called BS4 has been reported effectively to improve nutrient digestibility of palm oil by products such as palm oil sludge (Pasaribu et al. 2009) and palm kernel cake (Sinurat et al. 2013). Sinurat et al. (2014) also showed that the enzyme improved the protein- and the amino acids-digestibilities of fermented PKC.

Therefore, an experiment was designed to test the biological values of the fermented PKC (produced with the new method) supplemented with the BS4 enzyme and the possibility of using the product to substitute SBM in broiler’s diet as reported in this paper.

**MATERIALS AND METHODS**

**Effect of enzyme supplementation on nutritive values of fermented palm kernel cake used to substitute soybean meal in broiler diet**

Two steps of experiments were carried out in order to test biological values of fermented PKC. The first experiment was to determine AME and ileal amino acids digestibility of the FPKC produced with new fermentation method as described by Sinurat et al. (2015). The PKC was pretreated by autoclaving and supplemented with 10 % cassava leaf meal (9:1) before fermented with A. niger.

For the digestibility (dry matter, protein, ileal amino acids digestibility and AME) study, a commercial broiler feed was used as basal diet (B). Basal diet was mixed with ingredients tested 50:50 and added 2% acid insoluble ash (celite) as an indicator as described in Ravindran et al. (2005) and Sinurat et al. (2014). The experiment was arranged in a 2 X 2 factorial design. The first factor was fermentation process, i.e., non fermented PKC and fermented PKC (FPKC) and the second factor was enzyme supplementation (no enzyme and with enzyme). The enzyme supplemented was BS4 enzyme (150 U/kg) as reported by Sinurat et al. (2014). Parameters measured were dry matter digestibility, apparent metabolisable energy (AME) and ileal amino acids digestibility (IAAD).

One hundred (100) broiler chicks were reared on litter pens with standard rearing management from one day to 28 days old. At 28 d, 35 male chicks were selected and placed in individual wire cages. Each treatment diet was fed ad libitum to 7 (seven) chicks, and considered as replications. Treatment diets were fed for 5 days and samples of excreta was collected at the last 3 days for AME determination. After 5 days
feeding the tested diet, 35 birds were sacrificed by CO₂ asphyxiation and the digesta in the ileal was collected into plastic containers. Samples from 2 or 3 birds fed with the same diet were pooled to make 3 sample replications for each treatment. The digesta were immediately kept in the freezer for further chemical analyses.

The feed and the tested ingredients were analysed for dry matter, nitrogen (protein), amino acids and acid insoluble ash (AIA) contents. The excreta collected was analysed for the dry matter, nitrogen (protein), gross energy and AIA contents while the ileal digesta were analysed for dry matter, amino acids and AIA contents. The dry matter and the nitrogen were analysed following procedures described by AOAC (2005), while the amino acids were analysed by HPLC method at Bogor Agricultural University laboratory. All data were subject to analyses of variance, followed by least significance difference when the ANOVA was significant (P<0.05) according to procedure described by Steel & Torrie (1997).

Table 1. Composition of experimental diets in the feeding trial

| Ingredients (%) | Control  | SBM-10  | SBM-20  | SBM-40  |
|-----------------|---------|---------|---------|---------|
| Maize           | 62.18   | 58.78   | 57.58   | 57.54   |
| Soy bean meal   | 26.85   | 24.17   | 21.2    | 14.27   |
| Meat and bone meal | 3.93   | 3.3     | 3.83    | 7.0     |
| Corn gluten meal| 3.0     | 3.0     | 3.0     | 3.0     |
| Limestone       | 0.47    | 0.54    | 0.44    | 0.31    |
| Crude palm oil  | 1.5     | 2       | 2       | 1.5     |
| Fermented PKC + Enzyme | 0 | 5.9 | 9.67 | 14.5 |
| DL Methionine   | 0.35    | 0.37    | 0.3     | 0.43    |
| L-lysine        | 0.35    | 0.41    | 0.46    | 0.55    |
| Threonine       | 0.07    | 0.1     | 0.13    | 0.18    |
| Tryptophan      | 0       | 0       | 0.01    | 0.05    |
| Mono calcium phosphate | 0.76 | 0.9 | 0.76 | 0.1 |
| Sodium bicarbonate | 0.1    | 0.1     | 0.1     | 0.1     |
| Vitamin mix     | 0.025   | 0.025   | 0.025   | 0.025   |
| Mineral mix     | 0.05    | 0.05    | 0.05    | 0.05    |
| Salt            | 0.2     | 0.2     | 0.2     | 0.2     |
| Choline Chloride| 0.1     | 0.1     | 0.1     | 0.1     |
| Coccidiostat    | 0.05    | 0.05    | 0.05    | 0.05    |
| Fungistat       | 0.05    | 0.05    | 0.05    | 0.05    |
| Total           | 100.0   | 100.0   | 100.0   | 100.0   |

Nutrient composition

| Crude protein, % | 21.8 | 21.8 | 21.8 | 21.8 |
| Metabolisable energy, kcal/kg | 3000 | 3000 | 3000 | 3000 |
| Digestible lysine, % | 1.185 | 1.185 | 1.185 | 1.185 |
| Digestible methionine, % | 0.633 | 0.665 | 0.679 | 0.697 |
| Digestible meth. + cys, % | 0.908 | 0.905 | 0.905 | 0.905 |
| Digestible tryptophan, % | 0.208 | 0.202 | 0.202 | 0.202 |
| Digestible threonine, % | 0.758 | 0.758 | 0.758 | 0.758 |
| Calcium, % | 0.90 | 0.90 | 0.90 | 1.05 |
| Available P, % | 0.46 | 0.46 | 0.46 | 0.46 |
Substitution of soybean meal with enzyme supplemented fermented palm kernel cake (EFPKC) in broilers diet

The second experiment was designed to study the effect of substitution of SBM with the EFPKC on the performance of broilers. Four dietary treatments with graded levels of SBM substitution with EFPKC were formulated. All diets were formulated to meet the nutrient requirement of broilers according to Cobb (2012), i.e., crude protein 21.8%, metabolizable energy 3000 kcal/kg, digestible amino acids (lysine 1.185%, methionine + cystine 0.905%, tryptophan 0.202%, threonine 0.758%), calcium 0.90% and available phosphorus 0.46%. The ingredients and nutrient composition of the experimental diets are shown in Table 1.

The nutrient values of the EFPKC (ME and digestible amino acids) obtained from experiment 1, was used for the formulation of the treatment diets. The treatments consist of:
1. Standard diet (Control = C) with SBM level as in normal broiler diet
2. Diet with 10% of the SBM substituted with EFPKC
3. Diet with 20% of the SBM substituted with EFPKC
4. Diet with 40% of the SBM substituted with EFPKC

Each diet was fed to 60 broilers (6 replicates and 10 birds per replicate) from 1 to 35 days old. The birds were reared in pens with rice hulls as litter. Feed and water were given ad libitum. Parameters observed were feed intake, body weight, mortalities, and feed conversion ratio. At the end of the feeding trial, 2 birds (1 male and 1 female) from each pen were slaughtered to determine carcass and abdominal fat. Data were analysed with analyses of variance (ANNOVA) in a completely randomized design (4 treatments X 6 replicates). Duncan’s multiple range test were applied to show difference between treatment means when the ANOVA was significant at P<0.05 (Steel & Torrie 1997).

RESULTS AND DISCUSSION

Nutrients digestibility of PKC and FPKC as affected by enzyme supplementation

Effect of fermentation process and BS4– enzyme supplementation on dry matter digestibility and metabolisable energy (AME) of PKC and FPKC are presented in Table 2. The dry matter (DM) digestibility of PKC was only significantly (P<0.05) affected by fermentation (F) process, but not by enzyme supplementation (E) nor by the interaction of F x E. The DM digestibility of PKC in this study was 37.3% while previous study (Sinurat et al. 2013) showed a higher DM digestibility, i.e. 56.8%.

The AME of the PKC was not significantly (P>0.05) affected by fermentation process (F), enzyme supplementation (E) nor by the interaction of F x E. The AME of the PKC obtained in this experiment was 2079 kcal/kg, almost similar to the previous study i.e., ME 2074 Kcal/kg (Sinurat et al. 2014) and 2091 kcal/kg (Sinurat et al. 2013). However, Saenphoom et al. (2013) reported a lower TME of PKC i.e., 4.71 MJ/kg (or 1126 kcal/kg), which may be due to different process applied in the production of the PKC.

Some reports have shown different results on the effect of fermentation process on metabolisable energy of PKC. Sinurat et al. (2014) showed that fermentation of PKC with A. niger significantly decreased the AME (from 2074 to 1788 kcal/kg). Similar patterns were also reported by Muangkeow & Chinajarajyawong (2009) after fermented with A. wentii. Dairo & Fasuyi (2008) also reported a decrease in the ME of PKC after fermentation without addition of a fungus as inoculum. In contrast, Bintang et al. (1999) and Iyayi & Aderolu (2004) showed an increase in the ME of PKC after fermented with A. niger and Trichoderma viride, respectively.

In this study, PKC was mixed with 10% cassava leaf meal (CLM) prior to fermentation as described by Sinurat et al. (2015). Despite the ME of CLM, (i.e. 1160 kcal/kg according to Darma et al. 1989), was much lower than the ME of PKC, the fermentation process was still able to increase the ME of the PKC, although the differences were not significant (P>0.05).

Enzyme supplementation did not significantly (P>0.05) affect the DM digestibility nor the AME of the PKC nor the FPKC, although it increased from 37.3% to 46.8% or 25.5% improvement. Previous study showed a similar but significant improvement (26.2%) in the DM digestibility of the PKC due to enzyme supplementation (Sinurat et al. 2014). The DM digestibility of the PKC was increased from 56.8% to 71.7% as affected by the enzyme supplementation. Although statistically not significant, the AME of the PKC was increased from 2079 to 2385 kcal/kg or 15% improvement, while previous study showed an increase from 2091 to 2317 kcal/kg or 11% improvement. The AME of the FPKC was only slightly increased as the effect of enzyme supplementation, i.e., from 2496 to 2554 kcal/kg or 2.3% improvement. Previous study (Sinurat et al. 2014) showed a similar trend, i.e., 4.4% improvement due to addition of similar enzymes. Saenphoom et al. (2013) showed a high (60.3%) improvement on the TME of PKC as effect of enzymes (cellulase and mannanase) supplementation. In their study, the PKC was soaked in water, added with the enzyme, and incubated for 18 hours before the digestibility study. Present results showed that the enzyme improved DM digestibility and AME less on fermented PKC as compared to the effect on non-
fermented PK. Less increase of AME due to the enzyme supplementation to FPKC compared to PKC might occur due to the saccharification activity of fibernolytic enzymes produced during fermentation. The FPKC may contain shorter fiber molecules than that of PKC, therefore they were less digested by the enzyme addition. Purwadaria et al. (1998) reported that fermented palm oil sludge with A. Niger produced mannanase and cellulase in the course of fermentation. Those enzymes might actively digest the fiber.

Although the DM digestibility and the AME were increased due to fermentation process and enzyme supplementation, the effects were not significant (P>0.05) statistically. Perhaps, this is due to the high variability on the parameter observed in this study as shown by the coefficient of variation (CV), i.e., 22.14% and 14.8% for the DM digestibility and AME, respectively (Table 2).

The level of digestible amino acids in a feedstuff is calculated by multiplying the amino acids concentration in the feedstuff with the digestibility coefficient (percentage) of the amino acids. These values are commonly used for poultry ration formulation. The digestible amino acids of the PKC and FPKC as affected by enzyme supplementation are presented in Table 4.

Effect of enzyme supplementation on ileal amino acids digestibility (IAAD) of PKC and FPKC is presented in Table 3. The IAAD of essential (or indispensable) amino acids were not significantly (P>0.05) affected by enzyme supplementation nor by interactions between fermentation process (F) and enzyme supplementation (E). Some indispensable amino acids, i.e., arginine, histidine, isoleucine, leucine, phenylalanine and valine were significantly (P<0.05) decreased by fermentation process. These results do not agree with the results of Muangkeow & Chinajariyawong (2009) which showed that true amino acids (except for arginine) digestibility of the PKC were increased after fermented with A. wentii. Sinurat et al. (2014) also reported an increase in IAAD of indispensable amino acids except the arginine, threonine, tryptophan and valine. Supplementation of cassava leaf meal in PKC fermentation process may have changed IAAD profiles of FPKC. The IAAD values of PKC obtained in this study were much higher than IAAD of the PKC used in the previous experiment (Sinurat et al. 2014) which indicate the difference in quality of raw material (PKC) used in fermentation.

Among the indispensable amino acids, only digestible arginine, lysine and threonine were affected by treatments significantly (P<0.05). The digestible arginine level was significantly (P<0.05) reduced by the fermentation (F), but not significantly (P>0.05) affected by the enzyme (E) supplementation nor by their interaction (F x E). The digestible lysine was significantly (P<0.05) affected by the interaction (FxE), in which the enzyme supplementation increased the digestible lysine in PKC but not significantly (P>0.05) affected when applied in the FPKC. The digestible threonine was significantly (P<0.01) higher in the FPKC as compared to the PKC. In general, data on Table 4 showed that supplementation of enzyme did not increase the digestible amino acids in the FPKC. Solid substrate fermentation of palm kernel cake using A. niger USM F4 produced mannanase, a hemicellulytic enzyme for digesting NDF in PKM (Syarifah et al. 2012). Since the fermentation process of PKM in this experiment also produced enzymes, therefore supplementation of exogenous enzyme may have been in excess. As shown by Pourreza et al. (2007) supplementation of enzymes to a certain levels or activities improves the nutrients (dry matter, energy and protein) digestibility of feed, but supplementation of enzyme in a higher levels did not. Their data also showed that excess exogenous enzymes supplementation reduced nutrients digestibility of the feed.

| Table 2. Dry matter digestibility and metabolisable energy of PKC and FPKC fermented PKC with enzyme supplementation |
|---------------------------------------------------------------------------------------------------------------|
| **Ingredients** | **Dry Matter (DM) Digestibility,** % | **Metabolisable Energy (AME), kcal/kg** |
| Palm kernel cake (PKC) | 37.3±12.5 ** | 2079±389 ** |
| PKC + BS4 enzyme | 46.8±4.35 ** | 2385±152 ** |
| Fermented PKC (FPKC) | 50.64±9.1 ** | 2496±371 ** |
| FPKC + BS4 enzyme | 51.4±13.6 ** | 2554±511 ** |

* Values in the same column with different superscript significantly different (P<0.05)
** Standard deviation. Coefficient variation (CV) of the DM digestibility: 22.14%; AME: 14.8%
Table 3. Ileal amino acids digestibility of PKC and fermented-PKC with enzyme supplementation (%)

| Amino acids          | Basal* | PKC   | PKC+ Enzyme | FPKC | FPKC+ Enzyme | Significance (P) |
|----------------------|--------|-------|-------------|------|--------------|------------------|
|                      |        |       |             |      |              | Fermentation (F) | Enzyme (E) | F x E |
| Arginine             | 93.50  | 92.86 | 87.44       | 76.68| 77.70        | 0.005           | 0.538     | 0.374 |
| Histidine            | 89.28  | 87.11 | 70.35       | 56.55| 65.86        | 0.016           | 0.537     | 0.054 |
| Isoleucine           | 88.97  | 91.03 | 84.02       | 75.56| 74.37        | 0.013           | 0.327     | 0.480 |
| Leucine              | 90.67  | 91.77 | 84.08       | 77.31| 76.89        | 0.023           | 0.325     | 0.375 |
| Lysine               | 91.79  | 67.34 | 78.09       | 72.21| 72.41        | 0.921           | 0.204     | 0.219 |
| Methionine           | 93.05  | 87.18 | 75.24       | 81.05| 83.69        | 0.893           | 0.594     | 0.410 |
| Phenylyalanine       | 90.15  | 95.13 | 84.43       | 73.52| 74.78        | 0.005           | 0.285     | 0.184 |
| Threonine            | 86.41  | 82.45 | 72.51       | 68.58| 71.61        | 0.109           | 0.424     | 0.153 |
| Valine               | 89.22  | 86.91 | 80.48       | 72.06| 72.16        | 0.024           | 0.470     | 0.456 |

*Basal diet is a commercial broiler starter diet. The data were used for calculation of the IAAD

Table 4. Digestible amino acids of PKC and fermented-PKC with enzyme supplementation (%)

| Amino acids          | PKC    | PKC+ Enzyme | FPKC | FPKC+ Enzyme | Significance (P) |
|----------------------|--------|-------------|------|--------------|------------------|
|                      |        |             |      |              | Fermentation (F) | Enzyme (E) | F x E |
| Dispensable amino acids |      |            |      |              |                  |
| Arginine             | 1.494  | 1.443       | 1.337| 1.293        | 0.009           | 0.316     | 0.936 |
| Histidine            | 0.334  | 0.288       | 0.302| 0.308        | 0.764           | 0.332     | 0.216 |
| Isoleucine           | 0.732  | 0.702       | 0.755| 0.708        | 0.630           | 0.210     | 0.780 |
| Leucine              | 1.139  | 1.081       | 1.190| 1.131        | 0.285           | 0.218     | 0.991 |
| Lysine               | 0.436  | 0.532       | 0.614| 0.582        | 0.002           | 0.234     | 0.033 |
| Methionine           | 0.255  | 0.231       | 0.282| 0.279        | 0.125           | 0.551     | 0.631 |
| Phenylyalanine       | 0.809  | 0.743       | 0.821| 0.789        | 0.400           | 0.165     | 0.626 |
| Threonine            | 0.261  | 0.246       | 0.537| 0.518        | <0.001          | 0.413     | 0.943 |
| Valine               | 0.925  | 0.896       | 0.958| 0.904        | 0.613           | 0.320     | 0.762 |
| Dispensable amino acids |      |            |      |              |                  |
| Aspartic acid        | 0.982  | 1.243       | 1.426| 1.453        | 0.002           | 0.073     | 0.132 |
| Glutamic acid        | 2.415  | 2.618       | 2.562| 2.459        | 0.967           | 0.625     | 0.156 |
| Glycine              | 1.508  | 1.378       | 0.691| 0.594        | <0.001          | 0.010     | 0.636 |
| Proline              | 1.475  | 1.419       | 0.426| 0.389        | <0.001          | 0.014     | 0.552 |
| Serine               | 0.613  | 0.574       | 0.551| 0.551        | 0.056           | 0.344     | 0.336 |
| Tyrosine             | 0.404  | 0.334       | 0.485| 0.452        | 0.044           | 0.250     | 0.675 |
Table 5. Performances of broilers as affected by substitution of soybean meal with fermented palm kernel cake supplemented with enzyme

| Parameters          | Control | 10% SBM Substituted | 20% SBM Substituted | 40% SBM Substituted | Significance (P) |
|---------------------|---------|---------------------|---------------------|---------------------|-----------------|
| DOC body weight, g  | 42.4    | 43                  | 42.8                | 43.9                | 0.857           |
| Bodyweight 21 d, g  | 859.0   | 815.5               | 842.5               | 821.9               | 0.850           |
| Bodyweight 35 d, g  | 1929.6<sup>a</sup> | 1616.9<sup>b</sup> | 1753.2<sup>b</sup> | 1699.8<sup>b</sup> | 0.008           |
| Feed intake 1 – 21 d, g | 1331.7<sup>a</sup> | 1149.6<sup>b</sup> | 1250.6<sup>a</sup> | 1143.0<sup>b</sup> | <0.001          |
| Feed intake 1 – 35 d, g | 3116<sup>a</sup> | 2831.2<sup>b</sup> | 3026.5<sup>ab</sup> | 2825.5<sup>b</sup> | 0.043           |
| FCR 1 – 21 d        | 1.401   | 1.488               | 1.487               | 1.504               | 0.158           |
| FCR 1 – 35 d        | 1.657<sup>a</sup> | 1.804<sup>b</sup> | 1.743<sup>ab</sup> | 1.706<sup>b</sup> | 0.017           |
| Mortalities, %      | 3.33    | 6.67                | 10.0                | 16.7                | 0.059           |

<sup>*Different superscript in the same row showed significant difference (P<0.05)</sup>

Table 6. The effect of substitution of soybean meal with enzyme fermented palm kernel cake on the carcass percentage and some organs weight

| Parameters          | Control | 10% SBM Substituted | 20% SBM Substituted | 40% SBM Substituted | Significance (P) |
|---------------------|---------|---------------------|---------------------|---------------------|-----------------|
| Dressed carcass, % BW | 77.20   | 76.00               | 76.50               | 77.70               | 0.413           |
| Abdominal fat, g/kg BW | 19.86   | 24.09               | 23.65               | 22.19               | 0.139           |
| Liver weight, g/kg BW | 14.18   | 12.24               | 15.95               | 15.23               | 0.081           |
| Gizzard weight, g/kg BW | 19.23   | 22.25               | 19.84               | 19.97               | 0.262           |

Effect of substitution of soybean meal (SBM) with the enzyme supplemented FPKC (EFPKC) on performance of broilers from day old to 35 days old is presented in Table 5. The feed intake of broilers at 1-21 d old was significantly (P<0.01) affected by treatments. The lowest feed intake was found when the SBM was substituted 10%, followed by 40% substitution. The feed intake was different significantly (P<0.05) between control (not substituted) with 10% and 40% substitution of SBM with EFPKC, however, substitution of 20% SBM did not show significant (P>0.05) difference with the control. Similar trend and significant (P<0.05) effect was also found on the feed intake during 1-35 d period. Since all dietary treatments were formulated with similar nutrient values, include the energy (ME) and the digestible amino acids, the difference on feed intake could not be due to the nutrient factors but may be there are some anti-nutrient substances in the EFPKC. If this assumption is true, then the more the SBM substituted, the more the EFPKC included in the diet. If the EFPKC contains some anti-nutrient substances, the lowest feed intake was expected to occur when 40% SBM was substituted with the EFPKC. This phenomenon has been reported by Sinurat et al. (2014) when the SBM was substituted with the fermented PKC (without enzyme supplementation) in laying hens diet. It could not be explained at this stage why this is not the case in this experiment.

Body weight of broilers at 21 d was not significantly (P>0.05) affected by the substitution of SBM with EFPKC. However, the body weight at 35 d was significantly (P<0.05) affected by the treatments. Substitution of SBM with EFPKC produced lighter birds at 35 d, as compared with the control. The body weight is a reflection of feed or nutrients intake. The heavier birds were found on control diet (859.0 g at 21 d and 1929.6 g at 35 d) which consumed the highest feed and the body weight was lighter as the feed intake reduced.

The feed conversion ratio (FCR) during 1 to 21 d was not significantly (P>0.05) affected by treatments, however the FCR during 1-35 d was significantly (P<0.05) affected by the treatments. The FCR data showed that birds fed with the control diet were the most efficient in feed utilization. However, statistical analyses showed the FCR was only significantly different (P<0.05) between birds fed the control diet and those fed with 10% SBM substitution.

The mortalities of birds during the experiment was not significantly (P>0.05) affected by treatments. The data, however, showed that the mortalities increased as...
the level of SBM substitution with EFPKC was increased.

The effect of substitution of SBM with EFPKC on carcass percentage, weight abdominal fat, liver and gizzard of broilers as affected by treatments is presented in Table 6. Statistical analyses showed that there was no significant (P>0.05) effect of treatments on the dressed carcass percentage, abdominal fat-, liver- and gizzard-weight of broilers. These results indicated that replacing the SBM with EFPKC did not change the metabolism of the birds.

CONCLUSION

This study concludes that fermentation process improved the nutrient values of the palm kernel cake. Supplementation of enzyme was not effective to improve AME and amino acids digestibility of fermented palm kernel cake. Supplementation of enzymes to fermented palm kernel cake was not recommended to substitute soybean meal in broiler diets, since it reduced feed intake and growth rate of broilers.

ACKNOWLEDGMENT

This study was funded by Kementerian Riset dan Teknologi Republik Indonesia through PKPP project. The authors thanks all the technician who helped to conduct this study. Mr. Helmi was in charge on fermentation process, Mrs. Emi Sujatmika was in charge on enzyme production and Mr. Kadiran was in charge on rearing the chickens.

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