AMINO ACID DIGESTIBILITY IN BROILER BIRDS FED WITH PREMIUM PALM KERNEL CAKE BASED DIET

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ABSTRACT. To investigate premium palm kernel cake (PKC) based diet's effect on amino acid digestibility, this study conducted a feeding trial. Four groups of 150 day-old broiler chickens in every group were given four isocaloric diets using these formulation: Diet 1 - commercial feed for both starter and grower phase Diet 2 - broiler feed that contains 30% PKC for starter phase + commercial grower, Diet 3 - commercial starter + broiler feed that contains 45% PKC for grower phase (and Diet 4 - broiler feed that contains 30% PKC for starter phase + broiler feed that contains 45 % premium PKC for grower phase. During the experiment's day 31, bird faeces' samples were collected based on group diets in order to estimate the availability of amino acid as well as the digestibility of amino acids using balance experiment. Ultra-Performance Liquid Chromatography with photo-diode array detector was used to analyse amino acid. The findings showed no significant difference in digestibility of amino acids with $p<0.05$ in the feed-broiler chickens' comparison with commercial feed in terms of different levels of PKC based diet. This study shows that by including 30-45% premium PKC in poultry diet, it could contribute as protein source in broiler feed production with suitable supplementation of essential amino acid. Further studies are encouraged to combine the analysis of excreta, ileal digesta and growth performance in order to achieve more concrete findings.

Keywords: amino acid, digestibility, broiler chicken, poultry, palm kernel cake

INTRODUCTION

Palm kernel cake (PKC) is known as one of non-conventional feed that is derived from extraction of palm oil that has been proven to be good source of feed for various livestock. PKC is widely used as ruminant (beef and dairy cattle) feed because of its source of energy, protein, minerals, and vitamins (Pimentel *et al*., 2015). In poultry, PKC is used as alternative feed resource in order to conserve conventional ingredients in feed of the poultry diets like maize and soybeans, due to their low price and availability (Sharmila *et al*., 2014). However, in poultry feed, PKC usage is limited because its nature to have high fiber content while there is also wide variation in PKC used in poultry feed's optimum level. According to Alimon (2004), the usage of PKC in poultry feed varies based on the poultry's class, sex, age as well as the origin and oil and shell content variation of the PKC that is being used.

The chemical composition of the palm kernel cake varies depending on the type of fruit palm, the sample source and the method of oil extraction (solvent or mechanical extraction) (Ahmed and Mostafa, 2018). Due to the reason that production cost is lesser than solvent extraction, in most cases in Malaysia, PKCs are produced by extracting from the expeller (Alimon, 2004). In Malaysia, the average chemical composition of palm kernel cake is 88–94.5%

for dry matter, 13-20% for crude fibre, 10-19.8% for crude protein, 5-8% for ether extract, 3 -12% for total ash, 0.2-0.3% for calcium, 0.48-0.7% for phosphorus, 2.68% for lysine, and 1.75% for methionine respectively (Alimon, 2004).

Based on the study of McNab (1994), about 25% practical poultry diets' cost can be accounted for amino acids, hence dietary amino acids (in the form of proteins) must be paid attention to despite the fact that dietary components like amino acids, minerals and vitamins are essential components to formulate poultry's feed. Over the years, many studies were conducted to verify requirements of amino acid in poultry industry from numerous points of view such as economic as well as scientific's perspective. However, depending on the nature of the study and characteristic of the situations, there were variance in the reported values as there are different factors affecting amino acid's requirement. In the study of Ishibashi (1990), the findings classified the factors into four different categories which are environmental factors, animal status, genetic background, and also dietary factors. In dietary factors, different factors were identified which are metabolizable energy, antagonism and imbalance between amino acids, availability of dietary amino acids, palatability, deficiency and excess of amino acids, and diet. The requirement is defined by previous studies as the rate at which the bird must be fed with nutrients or energy so that it can perform as the necessary protein synthesis, maintenance, or to produce eggs within required rates (Emmans and Fisher, 1986; Ji-Hyuk Kim, 2015). The study of Buttery and D'Mello (1994) mentioned that essential amino acid deficiency would cause performance decrease. However, if the diets contain too much amino acids which exceeds the requirements, the excess will be excreted. This will then cause pollution. On top of that, according to McNab (1994), animal welfare problems will also occur with problems like defects in carcass, breast blisters as well as hock burn. The standard practise to quantify and qualify dietary feed is by conducting growth performance as well as digestibility study among the birds.

A number of methods were conducted by Wayne and Xiuhua (2010) in order to conclude the availability of amino acids availability (referred as amount of dietary amino acids that are acceptable for absorption, digestion, and use) by utilizing *in vitro* (enzymatic and chemical assays), indirect (plasma amino acids or microbiological) or direct (growth and digestibility assays). To estimate amino acid availability, digestibility assay is always chosen as the technique because digestibility is known to be the greatest single determinant of the amino acid's availability. The digestibility assays are divided into two main categories which are excreta and ileal. Excrete digestibility is pertaining to collecting excreta from caecectomized or intact birds, whereas to measure ileal digestibility, the digest will be collected from the ileum's distal part. The second method is more complex in terms of technicality, but by using that method, it can eliminate some confounding factors (Lemme *et al.*, 2004).

The aim of this study is to determine the digestibility of amino acids in broiler birds fed a diet based on a premium palm kernel cake (PKC) compared to commercial chicken feed.

MATERIALS AND METHOD

Location

Trials were conducted in the Institute of Poultry Technology, Masjid Tanah Melaka. The period of the experiment was between September to October 2017.

Experimental Diets

Four groups of experimental diets were used in this experiment, one based on commercial diet (control) and others based on PKC diets (Table 1). Commercial diets (starter and grower) were purchased from local suppliers while PKC diets formulated by MPOB were purchased from feed producers (Banyan Agri).

Table 1. Experimental diet by group.

Group	Experimental Diets
1	Commercial starter + commercial grower
2	Broiler starter with 30% PKC + commercial grower
3	Commercial starter + Broiler grower with 45% PKC
4	Broiler starter with 30% PKC + Broiler grower with 45% PKC

Experimental Birds and Housing

A total of 600 day-old Ross 308 broiler chicks were obtained from a local hatchery and raised on the wood shaving floor brooders. The birds were randomly allotted to four treatment groups consisting of 150 birds per treatment from day-old to 35th day. Two types of feed were offered to experiment birds: starter feed from day-old until day 14 of age and grower feed from day 15 to the end of the experiment. Feed was supplied according to the broiler manual Aviagen (2014) and MPOB Manual Table Feed. Clean water offered *ad libitum* throughout the experimental period. Environmental temperature in the first week of life was 35°C and decreased to 27°C until the end of the experiment. Light schedule was 24 hours of light throughout the experimental period. Birds were vaccinated against IB-ND (Infectious Bronchitis Newcastle Disease) for the first week and IBD (Infectious Bursal Disease) for the second week of age via drinking water. ND-Lasota Strain was given on the 2-4 weeks via drinking water.

Laboratory Analysis

The proximate composition of the feed ingredients was carried out in accordance with AOAC (1990). Total amino acids in feeds and faeces were extracted by acid hydrolysis while tryptophan was extracted by alkaline hydrolysis (AOAC 2000). Before acid hydrolysis, performic acid oxidation was conducted for methionine and cysteine. In order to find amino acids in hydrolysate, reversed phase Ultra Performance Liquid Chromatography (Waters Aquity H-Class Systems with photodiode/fluorescence detector) with pre-column derivatization were conducted.

Data Collection

The birds were weighed daily from day one until the experiments ended. By subtracting the birds' final body weight after feeding trial ended from their initial body weight which was later changed to daily weight gain by dividing by seven, the birds' weekly body weight gains during the experiment were obtained. Meanwhile, efficiency of the daily feed was considered as daily feed intake ratio to daily body weight gain. Feed intake was monitored by feeding weighed quantities of feed daily and subtracting the left-over from the quantity fed the previous day. Feed Conversion Ratio (FCR) was derived as the ratio of feed consumed to weight gain. The faeces of the birds were collected daily according to group diets and recorded before being stored at 4°C. When the experimental period ended (day 31), the samples were polled and mixed. Faecal collection (triplicate) was weighed. And then, it was dried to achieve constant weight at 65°C in 48 hours. The dried faeces were milled through 1 mm sieve prior to chemical analysis. Using the below mentioned formula, amino acid digestibility (%) was calculated (Lemme *et al.*, 2004).

Amino acid digestibility (%) =

(Amino acid intake (consumed) – Amino acid in output (faeces)) / (Amino acid intake (consumed)) x 100 %

Statistical Analysis

The data in this study were subjected to analysis of variances by utilizing general linear model (GLM). Means of variables in this research were compared based on Duncan's multiple range test during the observation of significant differences. The significance level was established at $P<0.05$.

RESULTS AND DISCUSSION

The experimental diets' composition of nutrient was analyzed before being fed to birds according to group as shown in Table 2. Laboratory tests showed that nutrient composition of PKC diets was better in crude protein and metabolizable energy (ME) compared to commercial feed (grower feed). The values of crude protein and ME in grower feed with 45% PKC were 21.2% and 3110.9 kcal/kg respectively, while in commercial growers, the crude protein and ME was lower 18.1% and 3067.6 kcal/kg respectively. Interestingly, the laboratory results also revealed that the composition of crude fat in the PKC diet were both better than in the commercial diet. The values of crude fat in starter PKC diet and grower PKC diet were 9.1% and 9.0% respectively, while the values of crude fat in commercial starter diet and commercial and grower diet were 3.6% and 6.2% respectively. Other researchers also found that the fat content makes PKC an energy feed. However, the laboratory test showed higher percentage in crude fiber for both PKC diet (starter and grower). The values of crude fibre in the PKC diet were 22.9% and 7.3% respectively, while the values of crude fibre in the commercial diet were 3.4% and 2.2%. Dietary fibre has always been considered an antinutritional factor that interferes with nutrient retention thus reducing growth performance in poultry. However, according to Jha and Mishra (2021), by including sufficient dietary fibre's amount in poultry diets, it helps to promote gastrointestinal tract development as well as improving growth performance, nutrient utilization, and the parameters of gut health.

Based on physiological requirements, muscle and egg protein synthesis need 20 amino acids (Ravindran and Bryden, 1999). The laboratory results revealed that the value of amino acids in the grower PKC diet was better than in commercial growers. From the viewpoint of physiology, both essential and non-essential amino acids are important for the synthesis of different proteins in the body. The total values of essential amino acids and non-essential amino acid in grower PKC diet were 10.53% and 10.46% respectively, while in commercial grower, essential amino acid as well as non-essential amino acid's values were 8.26% and 7.55% respectively. This result indicates that the PKC diet has good potential to be widely used in poultry feeds.

The analysed amino acid composition in birds' faeces is shown in Table 3. Total amino acid composition in faeces with PKC diet was shown higher than control diet but it did not show significant difference for both of the essential as well as non-essential amino acids ($p>0.05$). In

Table 2. Nutrient composition of experimental diets.
*Metabolizable energy value was calculated using the method 37 x % CP + 81 x % EE + 35.5 x % NFE for poultry (Fisher and Boorman, 1986)

Nutrient Composition (%)	Starter Diet		Grower Diets	
	Commercial Starter	PKC 30%	Commercial Grower	PKC 45%
Dry matter	89.9	91.3	88.7	89.7
Crude protein	22.9	22.8	18.1	21.2
Crude fat	3.6	9.1	6.2	9.0
Crude fiber	3.4	22.9	2.2	7.3
Crude ash	11.1	7.1	8.8	7.2
Nitrogen Free Extract (NFE)	48.9	29.4	53.4	45.0
*Metabolizable Energy (kcal/kg ME)	2874.9	2624.4	3067.6	3110.9
Calcium (Ca)	0.61	1.0	0.48	1.2
Phosphorus (P)	0.44	0.5	0.30	0.64
Natrium (Na)	0.15	0.1	0.13	0.19
Essential amino acids				
Histidine	0.54	0.54	0.43	0.39
Arginine	2.86	2.63	1.95	2.65
Threonine	1.07	0.9	0.64	0.85
Valine	1.25	0.99	0.87	1.11
Methionine	0.2	0.32	0.13	0.35
Isoleusine	2.82	1.98	1.92	2.42
Leusine	0.95	0.52	0.62	0.64
Phenylalanine	1.13	1.18	0.68	0.91
Tryptophan	0.22	0.14	0.18	0.13
Lysine	1.34	0.85	0.84	1.08
Non-essential amino acids				
Aspartic acid	2.07	1.5	1.18	1.89
Glutamic acid	4.52	3.21	3.1	4.02
Serine	0.92	2.03	0.65	1.19
Glycine	0.79	0.75	0.5	0.76
Alanine	1.15	0.86	0.69	1.07
Proline	1.57	0.94	1.04	1.05
Tyrosine	0.38	0.54	0.35	0.43
Cystine	0.03	0.08	0.04	0.05

Table 3. Amino acid composition in faeces between groups.

Amino Acid Composition (%)	Group 1 (control)	Group 2 (broiler starter 30% PKC + commercial grower)	Group 3 (commercial starter + broiler grower 45% PKC)	Group 4 (broiler starter 30% PKC + broiler grower 45% PKC)
Histidine	0.0	0.0	0.0	0.0
Arginine	0.38	0.58	0.62	0.53
Threonine	0.13	0.18	0.17	0.13
Valine	0.07	0.16	0.15	0.10
Methionine	0.05	0.01	0.01	0.03
Isoleusine	0.13	0.13	0.08	0.11
Leusine	0.15	0.22	0.16	0.18
Phenylalanine	0.09	0.13	0.12	0.10
Tryptophan	0.03	0.03	0.03	0.03
Lysine	0.05	0.10	0.07	0.07
Total essential amino acid	**1.08**	**1.54**	**1.41**	**1.28**
Aspartic acid	0.18	0.27	0.29	0.25
Glutamic acid	0.43	0.64	0.67	0.58
Serine	0.0	0.0	0.0	0.0
Glycine	0.07	0.09	0.09	0.09
Alanine	0.10	0.09	0.13	0.12
Proline	0.13	0.15	0.12	0.13
Tyrosine	0.01	0.03	0.05	0.02
Cystine	0.05	0.02	0.07	0.03
Total non-essential amino acid	**0.97**	**1.29**	**1.42**	**1.22**

total, PKC diets' essential amino acids were 1.54%, 1.41%, and 1.28% compared to the control diet 1.08%. The total values of non-essential amino acid in PKC diets were 1.29% (group 2), 1.42% (group 3) and 1.22% (group 4) compared to the control diet 0.97% (group 1). The highest value was recorded for group 3 (PKC based diet) with arginine and glutamic acid, 0.62% and 0.67% respectively. The value of tryptophan was found similar (0.03%) in faeces among groups while no value was found for histidine and serine in faeces among diets.

In the initial days of research in the area of digestibility, excreta's determination digestibility research was most frequently conducted (Lemme *et al.*, 2004). Table 4 shows the amino acid digestibility in broiler birds among diets. In this study, the amino acid digestibility was

Table 4. Amino acid digestibility in broiler birds fed PKC based diets
*Digestibility (%) = [(AA intake– AA output)/AA intake] x 100 (Lemme *et al.*, 2004)

Amino Acid Composition (%)	Group 1 (control)	Group 2 (broiler starter 30% PKC + commercial grower)	Group 3 (commercial starter + broiler grower 45% PKC)	Group 4 (broiler starter 30% PKC + broiler grower 45% PKC)
Essential amino acids				
Histidine	100	100	100	100
Arginine	92	87	89	90
Threonine	93	88	91	93
Valine	97	91	94	95
Methionine	80	97	98	95
Isoleusine	97	97	99	98
Leusine	90	81	90	84
Phenylalanine	95	93	94	95
Tryptophan	94	91	92	90
Lysine	97	94	97	96
Non-essential amino acids				
Aspartic acid	94	90	93	93
Glutamic acid	94	90	92	92
Serine	100	100	100	100
Glycine	95	93	94	94
Alanine	95	94	94	94
Proline	95	92	95	93
Tyrosine	99	96	94	98
Cystine	24	83	17	17

in the range of 80 - 100% among diets except for cystine, the digestibility values were 24, 83, 17 and 17% in group 1, 2, 3 and 4 respectively. The highest digestibility amino acids were recorded in histidine and serine with similar value among diets (100%). The highest value of total digestibility amino acid comprising histidine, arginine, threonine, valine, methionine, isoleucine, leucine, phenylalanine, tryptophan, lysine, aspartic acid, glutamic acid, serine, glycine, alanine, proline, tyrosine and cysteine was observed in group 2. However, the values were not significantly different ($p>0.05$) in PKC based diets compared to commercial diet (control). A study by Sundu *et al.* (2006) showed that the availability of amino acid exceeds 85% which seems to be high except for glycine and valine. Based on requirements of young chicks, there are two amino acids that are viewed as essential which are methionine and lysine, which need to be considered when utilizing PKC based diet. Whereas tyrosine and cysteine viewed as

Table 5. Performance of broiler birds fed PKC based diets.

Performance Parameter	Diet 1 (control)	Diet 2 (broiler starter 30% PKC + commercial grower)	Diet 3 (commercial starter + broiler grower 45% PKC)	Diet 4 (broiler starter 30% PKC + broiler grower 45% PKC)
Average feed intake (kg)	2.81	2.71	2.65	2.54
Final body weight (kg)	2.0	1.9	1.7	1.7
Weight gain (kg)	1.6	1.6	1.3	1.3
Feed conversion ratio (FCR)	1.4	1.4	1.6	1.5

semi-essential amino acids due to the reason that these two types of amino acids can be synthesized from methionine and phenylalanine respectively. Out of the ten essential amino acids, there are three types that are the most limiting in poultry diets which are lysine, methionine as well as threonine (Ravindran and Bryden, 1999). Table 5 shows the performance of broiler birds fed PKC based diet from day-old to 35 days of age. Final body weights and feed conversion ratio (FCR) of broiler birds that were given PKC based diets were found to be not affected at $P>0.05$ but the average feed intake and weight gain of experimental birds were lower in PKC based diets compared to commercial diet (control). However, there was no significant values' difference found ($p>0.05$) in PKC based diets compared to commercial diet (control). Kidd *et al.* (2004) reported that feed conversion was improved by giving high amino acid density diets. Apart from that, broiler chickens' weight gain and breast meat yield also increase.

CONCLUSION

The nutrient composition of PKC diets was better in crude protein and metabolizable energy (ME) with higher methionine and cystine value than in commercial feed. Overall, the amino acid digestibility ranged from 80 to 100% between diets except for cystine. The broiler birds' performance was not significantly different ($p>0.05$) in PKC based diets compared to commercial diet (control). It can be concluded that by including premium grade PKC at 30%-45%, it could be potentially used as broiler feed production's protein source since there is no substantial difference on amino acid digestibility ($p<0.05$).

REFERENCES

1. Ahmed M. Abdeltawab and Mostafa S.A. Khattab, (2018). *Utilization of Palm Kernel Cake as a Ruminant Feed for Animals: A Review*. Asian Journal of Biological Sciences. 11 (4): 157-164.
2. Alimon, A.R. 2004. *The nutritive value of palm kernel cake for animal feed.* Palm Oil Developments. 40:12-16.
3. AOAC, (1990). *Official methods of analysis (15th edition)*. AOAC.
4. AOAC, (2000). *Official methods of analysis (18th edition)*. AOAC.
5. Aviagen, T. (2014). *Ross 308 Broiler Nutrition Specifications*. Aviagen Group.
6. Buttery, P.J. and D'Mello J. P. F, (1994). *Amino acid metabolism in farm animals: An overview*. In: Farm Animal Nutrition, Ed. D' Mello J. P. F. pp 185-203 (CAB International). 1-10.

7. Emmans, G.C., Fisher,C. 1986. *Problem in nutritional theory*. In: Nutrient Requirements of Poultry and Nutritional Research, pp. 9-39. Ed. C Fisher, KN Boorman Butterworth.
8. Fisher, C and Boorman, K. N. 1986. *Nutrient requirement of poultry and nutritional research*: BPS, Symposium 19, Butterworths.
9. Ishibashi, T. 1990. Amino acid requirement of poultry. Japanese Poult Sci 27:1-15
10. Jha, R., Mishra, P. (2021). *Dietary fiber in poultry nutrition and their effects on nutrient utilization, performance, gut health, and on the environment: a review*. *J Animal Sci Biotechnol,* 12**,** 51. https://doi.org/10.1186/s40104-021-00576-0
11. Ji-Hyuk Kim, (2015). *Protein Quality and Amino Acid Utilization in Chickens*. KJPS, Vol.42 (1), 87-100.
12. Kidd, M. T., McDaniel, C. D., Branton, S. L., Miller, E. R., Boren, B. B., & Fancher, B. I. (2004). *Increasing amino acid density improves live performance and carcass yields of commercial broilers*. JAPR, 13(4), 593-604.
13. Lemme, A., V. Ravindran, V. and Bryden, W. L, (2004). *Ileal digestibility of amino acids in feed ingredients for broilers*. WPSJ, Vol. 60.
14. McNab, J.M. (1994). *Amino acid digestibility and availability studies with poultry*. In: D´ Mello, J.P.F. (Ed.) Amino acids in farm animal nutrition. Wallingford: CAB International. 63-98.
15. Pimentel, L.R., F.F. da Silva, R.R. Silva, A.R. Schio, E.S. de Oliveira Rodrigues and P.A. de Oliveira, (2015). *Feeding behavior of lactating cows fed palm kernel cake in the diet*. Acta Sci, Anim. Sci, 37: 83-89.
16. Ravindran, V. & Bryden, W.L. (1999). *Amino acid availability in poultry – in vitro and in vivo measurements*. Aust. J. Agric. Res, 50: 889–908.
17. Sharmila, A., Alimon, A.R., Azhar, K., Noor, H.M. and Samsudin, A.A. (2014). *Improving Nutritional Values of Palm Kernel Cake (PKC) as Poultry Feeds: A Review*. MJAS, 17(1):1 -1 8.
18. Sundu, B., Kumar, A., and Dingle.J. (2006). *Palm kernel meal in broiler diets: effect on chicken performance and health*. WPSJ.
19. Wayne L. Bryden1, Xiuhua Li. 2010. *Amino acid digestibility and poultry feed formulation: expression, limitations and application*. R. Bras. Zootec, 39: 279-287 (supl. especial).
20.

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