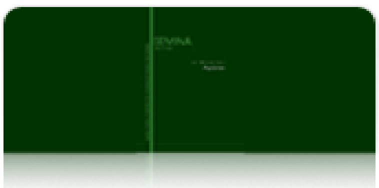

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Diet metabolisability and performance of slow-growing chickens fed with cottonseed cake

Metabolizabilidade das dietas e desempenho dos frangos de crescimento lento alimentados com torta de algodão

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Abstract

The aim of this study was to evaluate poultry performance and diet metabolisability in slow- growing naked neck chickens fed different substitution levels of soybean meal protein with cottonseed cake protein. The trial was conducted in a completely randomized design with five treatments (0, 10, 20, 30 and 40% substitution of soybean meal protein with cottonseed cake protein), six replicates and fifteen birds per replicate. The birds and diets of each experimental unit were weighed at one day old. At 30, 60 and 85 days old, the chickens and leftovers were weighed again to estimate the feed intake (FI), weight gain (WG), feed conversion (FC) and final weight (FW) from 1 to 30 days, from 1 to 60 days and from 1 to 85 days old. To evaluate diet metabolisability, a three-day total excreta collection (26 to 28 days of age) was performed. A quadratic effect was reported for diet metabolisability, in which the minimum substitution level was 17.45% and 17.69% for apparent metabolisable energy (AME) and nitrogen-corrected apparent metabolisable energy (AMEn), respectively. Based on performance, it is recommended to replace 40% soybean meal protein with cottonseed cake protein.
Key words: Alternative feedstuff. Nutrition. Naked neck.

Resumo

O objetivo desse estudo foi avaliar o desempenho das aves e a metabolizabilidade das dietas de frangos de crescimento lento, da linhagem pescoço pelado, alimentados níveis distintos de substituição da proteína do farelo de soja pela proteína da torta de algodão. O experimento foi conduzido em delineamento inteiramente casualizado com cinco tratamentos (0, 10, 20, 30 e 40% de substituição da proteína do farelo de soja pela proteína da torta de algodão), seis repetições e quinze aves por repetição. Com um dia

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de idade foram pesadas as aves e as dietas de cada unidade experimental. E aos 30, 60 e 85 dias de idade as aves e as sobras das dietas foram pesadas novamente para contabilizar o consumo de ração (CR), ganho de peso (GP), conversão alimentar (CA) e peso final (PF) das aves no período de 1 a 30 dias, de 1 a 60 dias e de 1 a 85 dias de idade. Para avaliação da metabolizabilidade das dietas foi realizada a coleta total de excretas por três dias (26 a 28 dias de idade). Foi identificado efeito quadrático no ensaio de metabolizabilidade das dietas em que o nível mínimo de substituição foi de 17,45% e 17,69% para EMA e EMAn, respectivamente. Com base no desempenho recomenda-se 40% de substituição da proteína do farelo de soja pela proteína da torta de algodão.

Palavras-chave: Alimento alternativo. Nutrição. Pescoço pelado.

Introduction

The small farmer has shown interest in raising slow-growing chickens since there has been an increasing consumer demand for meat from broilers raised in free-range systems. This niche market considers the organoleptic characteristics of the meat described as tastier and visually more attractive by the more intense pigmentation.

The increased production of slow-growing chickens boosts demand for feedstuff for feed production. Their feeding is diversified, with the need to include alternative feedstuffs (SAKOMURA et al., 2014), although the main ingredients for feed production are corn and soybean meal, which are responsible for most of the production cost. To meet the need for using alternative feedstuffs and to reduce the costs of feeding chickens raised in a free-range rearing system, we seek co-products that can partially or totally substitute these ingredients in the diet.

Cottonseed is the third most protein-producing vegetable ingredient in the world, the first being soybean meal and the second, canola meal. Cottonseed cake is a by-product obtained after hydraulic pressing or chemical extraction to isolate the seed oil. The classification of cottonseed by-product is based on the fat percentage: cake has a content of 5% residual fat, whereas meal has a residual fat content of less than 2% (BELTRÃO et al., 2000).

Cottonseed cake is commonly used to fed ruminants because of its high fiber content, but it also has between 30 and 43% protein (ROSTAGNO et al., 2017). For that reason, cottonseed cake has also been studied in poultry diets to evaluate the metabolisability and chicken performance (CARVALHO et al., 2010; MIRANDA et al., 2017; SANTOS e al., 2013).

The use of alternative feedstuffs is related to its nutritional value and cost, as well as the economic viability in poultry diets. The industrial production in large quantities favors the acquisition price, but only the diet cost analysis can prove whether it compensates for the farmer to use the product or not (FRANZOI et al., 1998).

The aim of this study was to evaluate the performance and diet metabolisability in slow-growing naked neck chickens fed four substitution levels of soybean meal with cottonseed cake.

Material and Methods

The experiment was conducted at the Aviculture Sector of the Federal University of Tocantins - UFT, School of Veterinary Medicine and Animal Science - EMVZ, according to the guidelines of the Ethics Committee on Animal Use (CEUA-UFT-EMVZ), protocol no. 2301.002965/201599, from January 30, 2017 to April 25, 2017. The daily temperature and humidity data throughout the experimental period were provided by the meteorological station located on the University campus. From the data, the averages of maximum, minimum and mean temperature, and humidity were calculated. The analyses of nutritional composition of diets were performed at the Laboratory of Animal Nutrition of the Federal University of Tocantins - UFT, School of Veterinary Medicine and Animal Science - EMVZ, Araguaina campus.

A total of 450 one-day old naked neck chickens of both sexes with a mean weight of 36.26 ± 2.5 grams were used. The experiment was conducted in a completely randomized design (CRD) with repeated measures. The treatments consisted of four substitution levels of soybean meal crude protein with cottonseed cake crude protein (0, 10, 20, 30 and 40%) with six replicates per treatment and fifteen birds per replicate. Repeated measures were performed for three evaluation periods: 1 to 30, 1 to 60 and 1 to 85 days of age.

The diets were formulated to meet the nutritional requirements of chickens in the starter (1 to 30 days), grower (31 to 60 days) and finishing phases (61 to 85 days) according to the chemical composition of the cottonseed cake, maize and soybean meal (Table 1), following the recommendations of Sakomura et al. (2014) for naked neck strain. The electrolyte balance of diet was calculated according to Mongin (1981): (mg.kg^{-1} Na^+ from feed/22.990) + (mg.kg^{-1} K^+ from feed/39.102) - (mg.kg^{-1} Cl^- from feed/35.453) (Table 2).

Table 1. Cottonseed cake, maize and soybean meal composition used in experimental diets.

Chemical composition	Cottonseed cake[1]	Maize[1]	Soybean Meal[1]
Dry matter (DM) %	94.20	89.08	89.49
Crude protein (CP) %	36.21	9.79	44.48
Mineral matter (MM) %	5.18	1.17	5.97
Ether Extract (EE) %	6.27	2.48	1.23
Crude fiber (CF) %	15.19	1.34	5.42
Neutral detergent fiber (NDF) %	42.00	9.17	12.87
Acid detergent fiber (ADF) %	23.49	2.73	5.39
Gross Energy (GE) cal.g^{-1}	4771.17	3911.93	4142.12

[1]Analyses performed at the Laboratory of Animal Nutrition at the School of Veterinary Medicine and Animal Science of the Federal University of Tocantins.

The chickens and diets of each experimental unit were weighed at one day of age. At 30, 60 and 85 days of age, the birds and leftovers were weighed again to estimate the feed intake (FI), weight gain (WG), feed conversion (FC) and final weight (FW).

From 1 to 30 days of age, the chicks were housed in batteries (1.0 m x 1.0 m x 0.40 m) and assigned to experimental cages equipped with trough waterers and feeders and a lighting system for heating until the 14th day of life. At 31 days, following the same distribution of treatments, the chickens were transferred to paddocks with an area of 10 m x 5 m, containing a small handmade shelter equipped with tubular feeder and waterers, where they were reared up to 85 days of age.

To evaluate diet metabolisability, the total excreta was collected for three days (26 to 28 days of age) and feed intake was recorded during this period (RODRIGUES et al., 2005; SAKOMURA; ROSTAGNO, 2016; SIBBALD; SLINGER, 1963) with feed and water *ad libitum*. For excreta collection, metallic trays were covered with plastic and placed under the cages. The samples were collected twice a day, at 8 a.m. and at 4 p.m., thus avoiding fermentation and possible loss of nutrients from the excreta. After collection, the excreta were stored in plastic bags, identified by experimental unit and kept in freezer for one week (SAKOMURA; ROSTAGNO, 2016).

The excreta were thawed at room temperature, weighed and homogenized to take one sample from each experimental unit. Samples were placed in a forced ventilation oven at 55º C for 72 hours for pre-drying to determine the dry matter content. Then, they were ground in a Willey mill using 1-mm sieve (SILVA; QUEIROZ, 2006).

Table 2. Centesimal composition of the starter (1 to 30 days), grower (31 to 60 days) and finishing diets (61 to 85 days) with different substitution levels of soybean meal (SBM) crude protein (CP) with cottonseed cake (CSC) crude protein.

Ingredients	Substitution levels of SBM CP with CSC CP														
	---------------1 to 30 days old-------------					--------------31 to 60 days old ------------					-----------------..61 to 85 days old --------------				
	0	10	20	30	40	0	10	20	30	40	0	10	20	30	40
Corn	55.42	55.76	55.61	54.00	52.19	62.98	63.08	63.18	62.45	60.49	74.75	73.70	73.00	72.55	72.00
Soybean meal	37.71	34.08	30.01	26.75	23.70	31.90	28.69	25.48	22.42	19.50	21.42	19.27	17.13	14.99	12.85
Cottonseed c.	0.00	4.62	9.74	13.88	18.52	0.00	3.98	7.97	11.95	15.94	0.00	2.66	5.33	7.98	10.64
Inert	2.52	1.56	0.34	0.00	0.00	2.51	1.55	0.59	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Dicalc. Phosp.	1.85	1.88	1.92	1.90	1.65	0.88	0.91	0.94	0.98	1.01	0.91	0.91	0.91	0.91	0.91
Limestone	0.97	0.98	0.99	0.99	0.81	0.89	0.90	0.90	0.91	0.91	0.92	0.60	0.92	0.92	0.92
Premix[a b]	0.24	0.24	0.24	0.24	0.24	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40
Salt	0.50	0.50	0.50	0.49	0.49	0.38	0.39	0.39	0.40	0.41	0.42	0.42	0.42	0.42	0.42
DL-methion.	0.63	0.37	0.40	0.43	0.45	0.02	0.06	0.10	0.15	0.19	0.17	0.10	0.15	0.17	0.15
L-lysine	0.08	0.00	0.25	0.38	0.40	0.00	0.00	0.00	0.06	0.17	0.11	0.00	0.18	0.18	0.19
L-threonine	0.07	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.00	0.05
Soybean oil	0.00	0.00	0.00	0.94	1.55	0.00	0.00	0.00	0.24	0.89	0.86	1.92	1.53	1.44	1.45
Total	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Nutritional composition															
ME(kcal/kg)	2750	2750	2750	2750	2750	2850	2850	2850	2850	2850	3100	3100	3100	3100	3100
CP%	21.48	21.48	21.48	21.48	21.48	19.39	19.39	19.39	19.39	19.39	15.48	15.48	15.48	15.48	15.48
CF%	2.95	3.47	4.03	4.46	4.95	2.78	3.21	3.65	4.08	4.50	2.42	2.70	2.98	3.26	3.54
NDF%	11.81	13.29	14.86	15.96	17.27	11.90	13.14	14.38	15.55	16.58	11.82	12.55	13.29	14.07	14.81
ADF%	4.90	5.72	6.58	7.24	8.02	4.69	5.37	6.05	6.71	7.35	4.24	4.66	5.09	5.52	5.95
EE%	2.66	2.90	3.14	4.23	5.01	2.83	3.03	3.23	3.64	4.42	3.93	5.08	4.81	4.82	4.93
CSCEE%	0.00	0.28	0.61	0.87	1.16	0.00	0.24	0.49	0.74	0.99	0.00	0.16	0.33	0.50	0.66
Calcium	0.97	0.97	0.97	0.97	0.97	0.68	0.68	0.68	0.68	0.68	0.67	0.54	0.65	0.65	0.64
Avai. Phosp.	0.45	0.45	0.45	0.45	0.45	0.27	0.27	0.27	0.27	0.27	0.27	0.26	0.26	0.25	0.25
Chlorine	0.35	0.35	0.34	0.34	0.33	0.27	0.27	0.27	0.28	0.28	0.29	0.29	0.29	0.29	0.29

continue

continuation

Potassium	0.85	0.82	0.78	0.76	0.73	0.77	0.71	0.65	0.59	0.54	0.61	0.57	0.53	0.49	0.45
Sodium	0.22	0.22	0.22	0.22	0.22	0.19	0.19	0.19	0.19	0.19	0.20	0.20	0.20	0.20	0.20
Digestible Lys.	1.18	1.16	1.31	1.11	1.14	1.04	0.95	0.86	0.83	0.83	0.87	0.72	0.80	0.74	0.68
Diges. Met+Cys.	0.68	0.69	0.69	0.70	0.70	0.66	0.66	0.66	0.66	0.66	0.72	0.61	0.63	0.63	0.57
Diges. Threonine	0.85	0.84	0.83	0.83	0.82	0.77	0.71	0.66	0.60	0.59	0.62	0.58	0.54	0.50	0.51
*EB mEq.kg^{-1}	210.99	202.19	191.88	182.71	175.21	189.10	181.01	172.94	165.00	156.92	148.38	142.21	136.35	130.61	124.82

Composition per kilogram of premix for starter phase: vitamin A 2,000,000 IU; vitamin D3 600,000 IU; vitamin E 5000 IU; vitamin K3 450 mg; vitamin B1 500 mg; vitamin B2 1500 mg; Pantothenic acid 3,500 mg; vitamin B6 700 mg; vitamin B12 2500 mcg; nicotinic acid 9,000 mg; folic acid 250 mg; biotin 15 mg; choline 80 mg; iron 10 mg; copper 2,500 mg; manganese 20 mg; zinc 18 mg; iodine 535 mg; selenium 75 mg; [b]Composition per kilogram of premix for growing birds: folic acid 120 mg; cobalt 179 mg; copper 2,688 mg; choline 108 mg; iron 11 mg; iodine 537 mg; lysine 49 mg; manganese 31 mg; mineral matter 350 mg; methionine 321 mg; calcium pantothenate 1,920 mg; vitamin A 1,500,000 IU; vitamin B1 300 mg; vitamin B12 2800 mcg; vitamin B2 960 mg; vitamin B6 450 mg; vitamin D3 300,000 IU; vitamin E 3,000 IU; vitamin H 0,20 mg; vitamin K 400 mg; zinc 22 mg.
*EB=electrolytic balance calculated according to Mongin (1981).

The diets and excreta samples were analyzed for crude protein, ether extract, mineral matter, dry matter, gross energy, crude fiber, neutral detergent fiber and acid detergent fiber (SILVA; QUEIROZ, 2006).

The apparent metabolisable energy (AME) and nitrogen-corrected apparent metabolisable energy (AMEn) were determined based on the data of feed intake, excreta production, dry matter, nitrogen (%) and GE (kcal.kg^{-1}) of diets and excreta, according to the equations described by Matterson et al. (1965):

AME

AME control diet = GE ing - GE exc / DM ing

AME test diet = GE ing - GE exc / DM ing

AME feedstuff = AME ref + ((AME test - AME ref)/ (g feedstuff/g diet))

AMEn

NB = Ning - Nexc

AMEn control diet = (GE ing - GE exc ± 8.22 * NB)/ DM ing

AMEn test diet = (GE ing - GE exc ± 8.22 * NB)/ DM ing

AMEn feedstuff = AMEn ref + ((AMEn test - AMEn ref)/(g feedstuff/g diet))

From the data above, the coefficients of apparent metabolisability of dry matter (CAMDM), crude protein (CAMCP) and gross energy (CAMGE) were determined by the ratio of AMEn and gross energy (GE) of the diets, according to the formula:

CMNT% = [(NTING - NTEXC)/ NTING] x 100, where:

CMNT: coefficient of nutrient/energy metabolisability;

NTING: amount of nutrient/energy ingested in grams;

NTEXC: amount of nutrient/energy excreted in grams.

For the statistical analysis, normality and homoscedasticity tests were performed. Given these assumptions, data were submitted to analysis of variance according to the statistical model: $Y_{ijk} = \mu + CSC_i + k + e_{ijk}$, where: Y_{ijk} = the dependent variable value for the jth observation in the ith substitution level of soybean meal with cottonseed cake, in the kth time; μ = overall mean; CSC_i = effect of the ith substitution level of soybean meal with cottonseed cake; k = the effect in the kth time; e_{ijk} = experimental error.

The data were submitted to regression analysis using first and second order polynomial models, considering the level of significance of the F test for the adjustment of models, the coefficient of determination (R^2 = SS model / SS treatments) and the linearity deviation. When the linearity deviation was not significant ($P>0.05$), the model was accepted, whereas when it was significant ($P<0.05$) the model was only accepted if the coefficient of variation was less than 10%. When significant ($P<0.05$) and with a coefficient of variation above 10%, the model was rejected.

SISVAR software was used for the statistical analysis, considering the significance level of 5%.

Results and Discussion

Throughout the experimental period, the average, maximum and minimum temperatures were 25.5° C, 30.9° C, 22.4° C, with a relative humidity of 85%.

There was a negative linear effect for feed conversion (FC) from 1 to 30, 1 to 60 and 1 to 85 days of age. For the other variables evaluated at the different ages, no significant effect was found (Table 3).

Table 3. Average feed intake (FI), weight gain (WG), feed conversion (FC), and final weight (FW) of slow-growing chickens from 1 to 30, 1 to 60 and from 1 to 85 days of age fed diets with different substitution levels of soybean meal (SBM) crude protein (CP) with cottonseed cake (CSC) crude protein.

Variables	Substitution of SBM CP with CSC CP						P			
	0 %	10 %	20%	30%	40%	Mean	LE	QE	LD	CV
1 to 30 days										
FI (g)	1269	1266	1208	1262	1197	1232	0.27	0.80	0.23	5.9
WG (g)	855	846	841	857	879	856	0.41	0.33	0.98	6.5
FC ($g.g^{-1}$)	1.48	1.44	1.43	1.47	1.36	1.44	0.02	0.29	0.12	4.5
FW (g)	893.8	886.2	883.5	896.1	924.4	896.8	0.34	0.33	0.98	6.4
1 to 60 days										
FI (g)	4190	4052	3866	4062	3873	4008	0.17	0.60	0.46	8.6
WG (g)	1808	1767	1717	1877	1907	1815	0.17	0.18	0.54	9.3
FC ($g.g^{-1}$)	2.32	2.29	2.25	2.16	2.04	2.20	0.00	0.17	0.96	5.2
FW (g)	1852	1813	1769	1923	1960	1863	0.14	0.18	0.58	8.9
1 to 85 days										
FI (g)	7822	7602	7444	7563	7181	7522	0.28	0.97	0.85	10.6
WG (g)	2778	2735	2640	2832	2795	2756	0.70	0.46	0.56	9.5
FC ($g.g^{-1}$)	2.83	2.77	2.82	2.66	2.56	2.72	0.01	0.33	0.70	7.2
FW (g)	2826	2782	2694	2882	2847	2806	0.67	0.46	0.56	9.1

CV = coefficient of variation;
P = probability of type I error by the F test;
LE = linear effect; QE = quadratic effect; LD = linearity deviation;
Equation FC (1 to 30 days) = -0.002CSC+1.48 (p=0.02; R^2=0.51);
Equation FC (1 to 60 days) = -0.006CSC+2.35 (p=0.00; R^2=0.91);
Equation FC (1 to 85 days) = -0.006CSC+2.85 (p=0.01; R^2=0.79).

There was an improvement in feed conversion of broilers from 1 to 30, 1 to 60 and 1 to 85 days old in function of substitution of soybean meal protein by cottonseed cake levels.

The cottonseed cake levels increased diet ether extract in the different phases of production. This increase in ether extract content may have led to the extra caloric effects of fat, with improvement in the use of non-lipid dietary components. Fat stimulates the release of cholecystokinin that, in addition to increasing pancreatic secretion, also acts at the satiety center, decreasing the rate of gastric emptying of the digestive tract (SAKOMURA et al., 2004). According to Hetland et al. (2004), the insoluble fiber in broiler diets has the opposite effect to that of the fat, because it increases the passage rate. However, a moderate amount of fiber in the diet can increase starch digestibility since it is retained longer in the gizzard until the particle size decreases to pass into the duodenum. In addition, higher fiber concentrations in the diet lead to gastric distension due to water retention, causing a reduction in appetite and feed intake (OLIVEIRA et al., 2012). Therefore, the high fiber content and ether extract in the diet may have contributed to the improvement in feed conversion of chickens.

The influence of fat on passage rate of feed through the digestive tract in poultry was studied by Andreotti et al. (2004), evaluating soybean oil levels (0.0, 3.3, 6.6, 9.9) on the transit time in broilers at 22 and 42 days of age. The authors observed a linear reduction in transit time, i.e. higher passage rate, at both ages, but they attributed this effect to rice husk levels (0.0, 4.7, 9.4, 14.1), finely ground, added to the diet as an inert material, since it increased the fiber content that may have negatively influenced the transit time.

The retention time of feed in the intestine is also influenced by the age of the bird; the older the chicken, the more developed the digestive tract and the greater fermentation in the cecum. Age implies a longer retention time of feed and better nutrient utilization (ANDREOTTI et al., 2004; BRUMANO et al., 2006), which may have contributed to the improvement in feed conversion during the total rearing cycle (from 1 to 85 days of age).

Another important factor is that the cottonseed cake has a high amount of fiber, and the utilization of this fiber depends on the age of the animals and fiber solubility. Carvalho (2012) characterized the fiber of the cottonseed cake and verified that it is composed of 55.5% of total carbohydrates, being 16.9% of non-fibrous carbohydrates that are comprised of 3.7% sugar, 1.7% of starch and 95.1% of pectin.

Sugar and starch are components of the cellular content, whereas the pectin composes the cell wall, but it is highly soluble in neutral detergent, then largely degraded by microorganisms. Therefore, the complete development of the digestive tract, mainly of the ceca in the adult chicken, can influence the utilization of the pectin in diets containing cottonseed cake.

For diet metabolisability, there was a quadratic effect for AME on an organic matter basis= $0.65CSC^2-22.69CSC+2735.22$ ($p=0.00$; $R^2=0.44$); AMEn on an organic matter basis= $0.69CSC^2-24.42CSC+2575.31$ ($p=0.00$, $R^2=0.47$), coefficient of apparent metabolisability of gross energy= $0.0077CSC^2-0.4105CSC+77.35$ ($p=0.02$, $R^2=0.33$). For the other variables, there was no significant effect (Table 4).

Table 4. Apparent metabolisable energy (AME) and nitrogen-corrected apparent metabolisable energy (AMEn) and coefficients of apparent metabolisability of dry matter (CAMDM), crude protein (CAMCP) and gross energy (CAMGE) of slow-growing chickens fed diets with different substitution levels of soybean meal (SBM) crude protein (CP) with cottonseed cake (CSC) crude protein[1].

Variables	Substitution of SBM CP with CSC CP					Mean	P			
	0%	10%	20%	30%	40%		LE	QE	LD	CV
AME² ($kcal.kg^{-1}$)	2637.2	2781.7	2504.4	2481.3	2954.5	2671.8	0.04	0.00	0.00	4.5
AMEn² ($kcal.kg^{-1}$)	2478.1	2608.6	2320.4	2311.9	2786.0	2501.0	0.02	0.00	0.00	4.1
CAMDM² (%)	69.84	67.18	69.15	69.22	69.05	68.89	0.92	0.60	0.52	5.6
CAMCP² (%)	56.64	55.07	55.59	57.19	58.32	56.56	0.50	0.50	0.95	8.3
CAMGE² (%)	75.05	73.49	72.06	71.16	74.08	73.17	0.32	0.02	0.66	4.4

[1]Expressed on an organic matter basis
[2]Analyzes performed at the Laboratory of Animal Nutrition at the School of Veterinary Medicine and Animal Science of the Federal University of Tocantins
CV = coefficient of variation;
P = probability of type I error by the F test at 5% probability;
LE = linear effect; QE = quadratic effect; LD = linearity deviation;
AME equation ($kcal.kg^{-1}$) = $0.65CSC^2-22.69CSC+2735.22$ ($p=0.00$; $R^2=0.44$);
AMEn equation ($kcal.kg^{-1}$) = $0.69CSC^2-24.42CSC+2575.31$ ($p=0.00$; $R^2=0.47$); where CSC is the substitution level of soybean meal protein with cottonseed cake protein.

It can be observed from the regression equations that the minimum substitution level of soybean meal with cottonseed cake is 17.45% for AME and 17.69% for AMEn. So, after the minimum substitution level, there was better use of dietary energy.

As the substitution level of soybean meal with cottonseed cake increased, so did the fiber content. According to Jorgensen et al. (1996), the metabolisable energy intake decreases with the higher fiber content in the diet, resulting in a better use of energy.

According to Choct (1997), high fiber content acts as a physical barrier that limits the accessibility of enzymes in the gastrointestinal tract, inhibiting the flow of nutrients, reducing digestibility and metabolisable energy. However, the dietary fiber contents (Table 2) are within the tolerable limit for broilers, and according to Larbier and Leclerq (1994), dietary insoluble fiber increases linearly the excretion of endogenous nitrogen and bacterial mass in the excreta, causing an increase in the amount of endogenous and exogenous substrates available to the bacterial fermentation in the cecal region.

The influence of fiber on bacterial production in the digestive tract was also observed by Sun et al. (2013) when evaluating the inclusion of fermented cottonseed meal on the bacterial microbiology of the ileum and the cecum of broilers. The results indicated that the microbiological composition of the ileum and cecum was considerably affected by the diet, with an increase in the population of lactobacilli and a decrease in the number of *Escherichia coli* when fermented cottonseed meal was added. Therefore, cottonseed meal promoted the growth of beneficial bacteria in the intestinal tract.

The substitution of soybean meal with cottonseed cake also increased the dietary fat content, which may have influenced the better use of energy due to the extra caloric effects of fat (SAKOMURA et al., 2014). Fiber and fat have antagonistic effects on the digestive tract in poultry: fiber increases the passage rate while

fat reduces it, which may have balanced the rate of gastric emptying and decreased nutrient flow. The mechanism of utilization of the fibrous fraction by the digestive tract in poultry is still little known, and more detail is needed to understand its physiological mode of action.

Conclusion

Based on broiler performance, the substitution of 40% soybean meal protein with cottonseed cake protein is recommended. A quadratic effect was verified in the metabolisable assay of rations with a minimum level substitution of 20.70%, 20.09% and 26.65%, respectively, for EMA, EMAn and apparent metabolisable coefficient of crude energy.

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