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Digestible Lysine Requirements the Performance, Carcass Traits and Breast Meat Quality of Slow-Growing Broilers

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ABSTRACT

Three experiments were conducted to estimate the digestible lysine requirements of slow-growing broilers on their performance, carcass traits, and breast meat quality. Different broilers were evaluated in each experiment. In Experiment 1, broilers were evaluated from 29-49 days of age (grower phase I), in Experiment 2, from 50 to 69 days old (grower phase II), and in Experiment 3, from 70-84 days old (finisher phase). A completely randomized design with five treatments of four replicates each was applied in all experiments. The following dietary digestible lysine values were investigated: 0.871, 1.011, 1.151, 1.291 and 1.431% in Experiment 1; 0.803, 0.943, 1.083, 1.223 and 1.363% in Experiment 2; and 0.766, 0.906, 1.046, 1.186 and 1.326% in Experiment 3. In all three experiments, digestible lysine values quadratically affected feed intake, weight gain, and feed conversion ratio. Lysine intake linearly increased with increasing digestive lysine values, whereas lysine utilization efficiency linearly decreased. Lysine requirements for maximum feed intake (1.298, 1.109, 1.150%), weight gain (1.183, 1.199, 1.162%), and feed conversion ratio (1.203, 1.162, 1.126%) were estimated in Experiments 1, 2 and 3. Digestible lysine requirement for carcass yield were estimated as 1.162, 1.068 and 1.107% in experiments 1, 2 and 3, respectively. Lysine influenced the physical-chemical parameters broiler breast meat. Digestible lysine levels of 1.203, 1.162 and 1.126% are recommended in the diets of Redbro Plume broilers during the phases 29-49, 50-69 and 70-84 days of age to optimize feed conversion ratio.

INTRODUCTION

Lysine is an essential amino acid and it is often the second limiting amino acid in diets based on corn and soybean meal. Lysine has considerable quantitative importance in the diet and is directly involved in the body development of poultry. It is an essential amino acid with a significant physiological function in muscle protein synthesis (Costa *et al.*, 2001; Lana *et al.*, 2005). In addition, lysine is a constituent of basic structures, such as collagen because it is precursor of the hydroxylysine (Sandel & Daneil, 1988), as well as of elastin, histones, carnitine (Champe *et al.,* 2009) and digestive enzymes.

Lysine is the main amino acid taken into account in feed formulations based on the ideal protein concept, where the requirements of other amino acids are estimated in proportion to lysine (Pedroso *et al.*, 2003). Therefore, determining the proper lysine requirement is essential for the formulation efficient feeds, with no limitations or excess of amino acids. Lysine -limiting diets have direct negative effects on muscle development, thus affecting the performance, carcass characteristics, and body composition of broilers (Oliveira *et al.*, 2013).

Several studies have been carried out to evaluate the responses of broilers to different lysine concentrations in the diet and to determine broiler lysine requirements for efficiency, performance, and carcass composition (Alam *et al.*, 2012). However, the formulation of diets for slow-growing broilers are typically based on the nutritional requirements established for genetically-improved broilers with high growth potential, as there is little information regarding the growth requirements for slow-growing broilers, which may limit the efficiency of feed utilization and compromise the final profitability.

The CobbSasso150 (Cobb-Vantress, 2008) slow-growing broiler strain manual recommends digestible lysine values of 1.22; 1.08 and 1.00% for the starter, grower and finisher phases, respectively, and Nascimento *et al.* (2009) recommended 1.041, 1.006 and 0.760% of digestible lysine in the starter, grower and finisher diets of ISA Label chickens.

One of the main parameters used to determine broiler nutritional requirements is the breast, the most expensive carcass part and considered a sensitive indicator of dietary nutrient supply. According to Sklan & Noy (2004), about 7.5% of all carcass protein is composed of lysine, and therefore further studies on the evaluation of digestible lysine values in feeds, particularly on its requirement for meat quality, are needed, considering the importance of lysine for the synthesis of muscle protein.

The RedBro Pluméis a slow-growth broiler strain that presents feathered neck, yellowish legs, intense red feathers, tail with black feather, and well-developed breast (Miranda *et al.*, 2005). It reaches an average final body weight of 2,200 kg between 70 and 80 days of age with a feed intake of about 5,700-6,300 kg. It is commonly reared in semi-intensive or extensive systems, and is highly valued in the chicken meat market.

The objectives of the present study were to estimate the digestible lysine requirements of broilers with low genetic growth potential between 29 and 49 days, 50 to 69 days, and 70 to 84 days, using performance evaluations, carcass characteristics, and breast meat quality.

MATERIAL AND METHODS

Birds and housing

Three experiments were conducted at the Poultry Production Integrated Center of the Universidade Federal Rural do Rio de Janeiro (UFRRJ), located at latitude 22° 45 ‘S and longitude 43° 41’ W. The experiments were approved by the Ethics Committee on Animal Use (ECAU) of the Institute of Animal Science of UFRRJ, process number 23083.011133 / 2014-48.

A total of 1,200 one-d-old RedbroPlumé male broiler chicks (average weight = 40.4 g) were housed in a conventional broiler house. Birds were vaccinated in the hatchery against Marek’s disease and fowlpox. Water and feed were supplied *ad libitum* while broilers remained in the house before being transferred to the experimental facilities.

Three experiments were carried out. Experiment 1 corresponds to grower phase I (29 to 49 days), experiment 2 to grower phase II (50 to 69 days), and experiment 3 to the finisher phase (70 to 84 days). In each experiment, different birds were used.

The experimental shed was divided in 20 pens (4 m × 5 m each), equipped with a bell drinker and two tube feeders. The floor was covered with wood shavings (± 8 cm thick). Feed and water were provided *ad libitum* throughout the experiment. The lighting regime of 16 hours of light and 8 hours of darkness was applied in accordance with the recommendations of the Normative Instruction N. 46 of the Brazilian Ministry of Agriculture (Brasil, 2011).

Prior to the start of each experiment, the feeds supplied were formulated according to Rostagno *et al.* (2011).

Diets and experimental design

The experiments were conducted according to a completely randomized design, with five treatments with four replicates each, totaling 20 experimental units. Each replicate consisted of 22 broilers in experiment 1; 20 in experiment 2; and 18 in experiment 3, totaling 440 birds in experiment 1, 400 animals in experiment 2 and 360 in experiment 3. Birds were selected according to body weight uniformity, and all birds remained in the shed until the end of each experiment.

For the chemical and physical-chemical analysis of breast meat (experiments 2 and 3), the breast of two broilers per replicate were collected after slaughter, totaling eight breasts per treatment.

The basal diets (Table 1) were formulated to contain all nutrients as recommended by Rostagno *et al.* (2011), except for lysine, calcium and phosphorus. The criterion to determine the nutritional requirements was average body weight (BW), instead of age as described in the tables. The calcium and available phosphorus requirements used for feed formulation were based on Pinheiro *et al.* (2011a) and Pinheiro *et al.* (2011b). Corn gluten, an ingredient that is limiting in lysine, was included in all experimental diets with the aim of obtaining a basal lysine value without decreasing the crude protein value of the diet.

Table 1 – Ingredients and calculated nutritional composition of the basal diets in each experiment.

Ingredient (g kg^{-1})	Experiments		
	1	2	3
Corn (7.88% CP)[1]	63.253	66.166	66.975
Soybean meal (46.90% CP)[1]	28.493	25.893	24.369
Soybean oil	2.257	2.689	3.034
Corn gluten (60.09% CP)[1]	2.037	1.299	1.369
Dicalcium phosphate	1.195	1.223	1.378
Limestone	0.843	0.842	1.014
Corn starch	0.900	0.900	0.900
DL-Methionine (98.5%)	0.247	0.234	0.219
L-Threonine (98.5%)	0.057	0.059	0.054
Mineral mix[2]	0.100	0.100	0.100
Vitamin mix[3]	0.100	0.100	0.100
Salt	0.457	0.444	0.438
Choline chloride (60%)	0.050	0.040	0.040
Antioxidant (BHT)[4]	0.010	0.010	0.010
Total	100.00	100.00	100.00
Nutritional composition (%)[5]			
Crude protein	19.761	18.322	17.700
Metabolizable energy (Kcal/kg)	3,100	3,150	3,175
Linoleic acid	2.680	2.928	3.112
Available phosphorus[6]	0.350	0.335	0.320
Calcium[7]	0.780	0.735	0.690
Sodium	0.200	0.195	0.192
Digestible methionine+cystine	0.796	0.745	0.717
Digestible methionine	0.523	0.490	0.469
Total lysine [8]	0.987	0.912	0.874
Digestible lysine	0.871	0.803	0.776
Digestible threonine	0.709	0.663	0.638
Digestible valine	0.825	0.763	0.738
Digestible isoleucine	0.748	0.687	0.661
Digestible arginine	1.157	1.070	1.026

[1]Values determined at the Animal Nutrition Laboratory DNAP / IZ.

[2]Guaranteed levels/kg product: 50g iron; 8,500mg copper; 1,000mg cobalt; 1.000mg iodine; 70g manganese, 60g zinc.

[3]Guaranteed levels/kg product: 12,000,000 IU Vitamin A (i);2,250,000 IU Vitamin D3;25,000 IU Vitamin E (i); 3,000 mg Vitamin K3; 2.400mg Vitamin B1 (thiamine); 12g Vitamin B2 (riboflavin);2,000 mg vitamin B6 (pyridoxine);24,000 mg vitamin B12 (i);42g niacin (me), 15g calcium pantothenate; 1,800 mg folic acid; 50 mg BHT;180mg biotin; 180mg selenium.

[4]BHT (butylated hydroxytoluene).

[5]Brazilian Tables for Poultry and Swine.

[6,7]Pinheiro *et al.* (20011a, 2011b).

[8]Analyzed amino acid.

In all experiments, the treatments consisted of five diets with increasing digestible lysine values, obtained by the addition of L-Lysine HCl (78% purity) to the basal diet in replacement of cornstarch. The treatments were as follows: 0.871, 1.011, 1.151, 1.291 and 1.431% digestible lysine in experiment 1; 0.803, 0.943, 1.083, 1.223 and 1.363% digestible lysine in experiment 2; 0.766, 0.906, 1.046, 1.186 and 1.326% digestible lysine in experiment 3.

Measurements

The variables evaluated in each experiment were feed intake, weight gain, feed conversion ratio, lysine intake, and lysine utilization efficiency (related to digestible lysine intake). Carcass and parts yields (breast, wing, thigh + drumstick and back) and relative weights of edible viscera (gizzard, liver and heart) and of abdominal fat. Carcass yield and the relative weights of edible viscera and abdominal fat were determined as their weight relative to slaughter weight. Parts (breast, wing, thigh + drumstick and back) yield was determined as part weight relative to cold carcass weight. Breast meat quality parameters (chemical and physical-chemical composition) were also measured in experiments 2 and 3.

At the end of each experiment, four broilers per replicate were selected based on the representative average weight of the replicates, totaling 16 chickens per treatment. Birds were subjected to eight-hour fasting, after which they were weighed, sacrificed by cervical dislocation, bled, plucked, and their viscera, head, neck and feet were removed. Carcasses were then chilled, dripped for 5 min to reduce the excess of water absorbed in the previous step, and submitted to standard commercial cut-up. The breasts were stored at ± 4 °C until analyses.

Chemical and physical-chemical analyses

The analyses of the chemical composition of the breast meat were carried out in the Laboratory of Animal Nutrition of the Institute of Animal Science (IZ / UFRRJ); and the physical-chemical analyses in the Laboratory of Food Technology (IT / UFRRJ).

The frozen breasts were thawed in a refrigerator at approximately 10 °C for a period of 24 hours, and all visible fat and skin were removed before chemical and physical analyses.

The muscular portion of the breast (*pectoralis major*) of two broilers per experimental unit was removed to determine its chemical composition, including moisture, crude protein, ether extract, and ash contents, according to Silva & Queiroz (2006).

The following physical-chemical parameters of the breast meat were determined: thawing loss (DL), cooking weight loss (CL), shear strength (SS), pH and color.

In order to determine thawing loss, samples were weighed on a semi-analytical scale before and after thawing, and DWL was calculated as the difference between frozen and thawed sample weights multiplied by 100.

Cooking loss was determined by weighing the thawed breast samples were weighed and then both sides of the sample were cooked on plate placed on top of electric resistance until reaching an internal temperature of 82 to 85 °C. The samples were cooled at room temperature and weighed. Cooking loss was as the difference between the thawed sample weight and cooked sample weight sample multiplied by 100.

Shear strength was determined in the samples used for cooking loss determination. Cylinders of approximately 1.5 cm diameter and 2.5 length were cut each breast sample. Each cylinder was placed with the fibers oriented perpendicularly to the blade of a texturometer (TA.XT plus Texture Analyzer, Stable Micro Systems, UK) and cut. The result was expressed in kilograms force per gram (kgf/g).

Meat pH was determined using a pHmeter. The electrode was immersed in mixture consisting of a breast meat sample (± 10 g) and distilled water (± 10 ml). The pH values were recorded when the reading was stabilized.

Breast meat color was determined using a spectrophotometer and the CIELab system. The parameters L* (lightness), a* (redness) and b* (yellowness) were recorded at four different points of the surface of the *pectoralis major* muscle.

Statistical analysis

The obtained data were submitted to analysis of variance using the statistical program SISVAR (version 5.6) (Ferreira, 2011). Responses were considered significant when at $p<0.05$. The responses to dietary lysine levels were studied by analysis of regression. Estimates of digestible lysine requirements were determined, when possible, using a quadratic model.

RESULTS AND DISCUSION

Experiment 1 - Performance and carcass traits during grower phase I (29-49 days of age)

There was a quadratic influence of dietary digestible lysine levels on feed intake, weight gain and feed conversion ratio, and linear influence on lysine intake and utilization efficiency (Table 2). The greatest weight gain (1149 g) and best feed conversion ratio (2.24) were obtained with 1.183 and 1.203% dietary digestible lysine levels, respectively. The results also that the treatment the lowest digestible lysine level (0.871%) limited the growth performance of the evaluated birds, as shown by the worst weight gain and feed conversion ratio responses.

The broilers fed the diets with lower digestible lysine values (0.871%) presented higher feed intake (2930 g), probably in an attempt to compensate for amino acid limitation. On the other hand, the higher feed intake obtained as digestible lysine levels increased may be due to the gradual improvement in the ratio between lysine and other amino acids in the diet, since dietary amino acid balance has a direct effect on voluntary feed intake (Gonzales, 2002).

Table 2 – Feed intake, weight gain, feed conversion ratio, lysine intake and lysine utilization efficiency of slow-growing broilers in experiment 1 (29 to 49 days old).

	Digestible lysine (%)						
Variables	0.871	1.011	1.151	1.291	1.431	Regression*	CV[2] (%)
Feed intake, g	2930	2830	2715	2753	2733	Q	1.63
Weight gain, g	956	1055	1108	1205	979	Q	2.60
Feed conversion ratio	2.87	2.52	2.31	2.16	2.61	Q	3.63
Lysine intake, g	25.52	28.61	31.25	35.54	39.11	L	1.60
LUE[1] (%)	37.48	36.89	35.47	33.91	25.04	L	1.98

[1]LUE, lysine utilization efficiency. [2]CV (%), coefficient of variation; *L, Linear effect; *Q, quadratic effect ($p<0.05$). The equations are shown in Table 10.

Nascimento *et al.* (2009) studied the digestible lysine requirements of Isa Label chickens reared in semi-intensive conditions and estimated a 1.056% digestible lysine to achieve optimal weight gain. However, Rosa *et al.* (2014) estimated that 0.908% digestible lysine should be supplemented into the diet of male broiler chickens at 28-56 days old.

Dietary lysine values had a quadratic effect on carcass, breast and wing yields and a linear effect on leg (thigh + drumstick) yield (Table 3). The highest carcass (73.62%) and wing (27.72%) yields obtained at estimated digestible lysine values of 1.162 and 1.147%, respectively. The estimated value for breast yield is not recommended because the effect was negative.

Table 3 – Relative weights and yields of carcass cuts and edible offal of slow-growing broilers slaughtered at 50 days of age in experiment 1.

	Digestible lysine (%)						
Variables	0.871	1.011	1.151	1.291	1.431	Regression*	CV[1] (%)
Yield (%)							
Carcass	67.21	71.19	72.68	74.08	66.98	Q	3.26
Breast	29.14	28.35	28.48	26.79	29.26	Q	6.49
Leg	32.15	32.35	32.59	33.82	33.14	L	3.40
Wing	12.98	13.12	13.64	13.10	12.96	Q	8.58
Back	29.95	27.18	28.08	26.65	29.85	NS	10.29
Relative weight (%)							
Heart	0.44	0.43	0.46	0.45	0.46	NS	11.06
Liver	1.72	1.66	1.85	1.60	1.79	NS	10.10
Gizzard	1.95	1.84	1.84	1.77	1.94	NS	15.96

[1]CV, Coefficient of variation;

*NS, Not significant; *L, Linear effect; *Q, quadratic effect ($p<0.05$). The equations are shown in Table 10.

The quadratic effect obtained for carcass, breast, and wing yields suggests that digestible lysine levels higher than the estimated requirements possibly reduced lysine utilization efficiency.

The observed increases in carcass and parts yields may be explained by the fact that the main function of lysine is protein deposition in the carcass, and it is one of the main amino acids in muscle proteins. According Sklan & Noy (2004) about 7.5% of all carcass protein is composed of lysine.

Rosa *et al.* (2014) evaluated diets with increasing digestible lysine values fed to male broilers and did not find any effects on carcass and breast yields, obtaining an average carcass yield of 65.44%, which is lower than that found in this study (73.62%).

Experiment 2 - Performance, carcass traits, and breast meat quality results obtained in grower phase II (50 to 69 days old)

Performance and carcass traits

Dietary lysine values had a quadratic effect on feed intake, weight gain and feed conversion ratio and a linear effect on the lysine intake and utilization efficiency (Table 4). The requirement for maximum weight gain (907 g) was estimated at 1.199% digestible lysine, and for best feed conversion ratio (3.21) was estimated at 1.162% digestible lysine. Considering these requirements, when compared with the treatment with the lowest lysine value, weight gain improved in 8.71% and feed conversion ratio in 5.65%. The lowest feed intake (3492g) during this period was obtained at an estimated digestible lysine level of 1.109%.

The improvement in feed conversion ratio as digestible lysine values increased up to the requirement level is probably due to the positive association between the efficiency of amino acid utilization and the increase in muscle mass or nitrogen retention, since the estimated requirement for weight gain and feed conversion ratio were very close and similar to the results obtained by Almeida (2010).

The manual of the slow-growing broiler strain CobbSasso150 (Cobb-Vantress, 2008) recommends 1.08% digestible lysine during the growth phase, which is lower than those determined in the present study.

Lysine values influenced in a quadratic manner carcass, breast, leg and wing yields, and the relative weight of abdominal fat (Table 5).

Table 4 – Feed intake, weight gain, feed conversion ratio, and lysine intake and utilization efficiency of slow-growing broilers in experiment 2 (50 to 69 days old).

	Digestible lysine (%)						
Variables	0.803	0.943	1.083	1.223	1.363	Regression*	CV[2] (%)
Feed intake, g	3800	3521	3523	3544	3641	Q	4.17
Weight gain, g	828	849	890	940	877	Q	3.82
Feed conversion ratio	3.64	3.49	3.21	3.14	3.40	Q	5.70
Lysine intake, g	30.51	33.20	38.15	43.35	52.10	L	4.67
LUE[1] (%)	36.31	39.27	35.98	33.04	22.35	L	4.99

[1]LUE, lysine utilization efficiency

[2]CV (%), coefficient of variation; *L, Linear effect; *Q, quadratic effect ($p<0.05$). The equations are shown in Table 10.

Table 5 – Carcass and parts yields and relative weights of edible offal and abdominal fat of slow-growing broilers slaughtered at 70 days of age in experiment 2.

	Digestible lysine (%)						
Variables	0.803	0.943	1.083	1.223	1.363	Regression*	CV[1] (%)
Yield (%)							
Carcass	69.38	69.68	73.63	75.89	64.30	Q	2.91
Breast	26.30	28.40	28.25	28.68	27.02	Q	6.32
Leg	27.14	28.27	29.15	29.06	27.36	Q	8.85
Wing	12.85	13.08	13.30	13.12	13.10	Q	5.11
Back	25.79	32.48	26.72	25.62	28.99	NS	8.59
Relative weight (%)							
Heart	0.37	0.37	0.36	0.35	0.38	NS	12.81
Liver	1.50	1.35	1.38	1.37	1.40	NS	14.15
Gizzard	1.84	1.71	1.76	1.77	1.65	NS	19.89
Abdominal fat	1.64	1.49	1.26	1.33	1.78	Q	40.24

[1]CV, Coefficient of variation;

*NS, Not significant; *L, Linear effect; *Q, quadratic effect ($p<0.05$). The equations are shown in Table 10.

The highest carcass yield (74.24%), breast yield (28.74%), leg yield (29.15%) and wing yield (13.21%) were obtained at estimated digestible lysine values of 1.068, 1.107, 1.101 and 1.142%, respectively. The lowest relative weight of abdominal fat (1.31%) was achieved at an estimated digestible lysine value of 1.075%.

The lowest abdominal fat relative weight (1.31%) was calculated at an estimated digestible lysine value of 1.075%. The dietary excessive supply of amino acids can lead to imbalances, limiting lean tissue growth and increasing fat deposition. The supply of diets with protein in excess of the requirements or low-digestibility proteins results in amino acid imbalance. Therefore, the excessive amino acids that are not utilized by the body need to be deaminated, resulting in carbohydrates that are used as energy source, increasing the potential for fat deposition (Lesson, 1995), which may explain the increase in abdominal fat in the broilers fed the diet containing the highest digestible lysine value (1.363%).

Breast meat quality

The breast meat quality parameter results are shown in Table 6.

Breast meat moisture and crude protein contents linearly increased as dietary digestible lysine increased in 1.39% and 6.23%, respectively. The linear response observed in breast meat moisture content may be partially explained by the increase in crude

Table 6 – Chemical and physical-chemical composition of the breast meat of slow-growing broilers slaughtered at 70 days of age in experiment 2.

	Digestible lysine (%)						
Variables	0.803	0.943	1.083	1.223	1.363	Regression*	CV[7] (%)
Chemical composition (%)**							
Humidity	73.07	73.00	73.21	73.89	74.10	L	7.89
Crude protein	22.13	22.71	22.97	23.38	23.60	L	6.54
Ether extract	1.43	1.34	1.42	1.09	0.85	L	7.00
Ash	1.44	1.43	1.45	1.40	1.40	NS	5.43
Physical-chemical composition							
pH	6.21	5.89	6.03	6.13	5.97	NS	3.25
TWL[1] (%)	4.92	5.02	5.56	7.65	6.54	L	3.14
CWL[2] (%)	18.63	18.98	19.21	19.14	19.91	L	2.67
SS[3] (kgf/g)	3.33	3.14	3.24	3.45	3.29	NS	18.21
L*[4]	46.98	47.32	46.34	46.12	47.24	NS	24.54
a*[5]	5.70	5.88	6.01	5.90	6.10	L	20.78
b*[6]	9.75	9.78	10.23	9.89	10.06	NS	29.97

[1]TWL, Thawing weight loss; [2]CWL, Cooking weight loss; [3]SS = Shear Strength

[4]Lightness; [5]Redness; [6]Yellowness

[7]CV, Coefficient of variation; *NS, Not significant; *L, Linear effect ($p<0.05$)

**Values determined in fresh meat.

protein and the reduction in ether extract contents. Trindade Neto *et al.* (2011) observed a quadratic response in carcass water deposition up to 1.10% dietary digestible lysine, and an increase in protein deposition rate was also observed. According to those authors, this suggest an improvement of the efficiency of dietary nutrient utilization, which may also have occurred in the present study, as shown by the increasing breast meat moisture and crude protein contents observed. The linear reduction (40.56%) in breast meat ether extract content may reflect the fact that lysine is primarily used for protein synthesis, and some authors have highlighted its effect in reducing abdominal fat.

Increasing dietary digestible lysine increased TWL and CWL. From the meat quality perspective, this behavior is not desirable. According to Dabés (2001), when the muscle tissue presents low water retention, its moisture loss and subsequent weight loss during further-processing steps are higher, indicating loss of meat nutritional value from the released exudates, and resulting in meat juiciness and tenderness losses. Anadón (2002) mentioned that meat texture, as determined by shear force, is closely related to intramuscular water content and meat water holding capacity, and therefore, the higher the water content of the muscle, the greater the tenderness of the meat. Despite the linear increase breast in moisture content observed in the present study, shear force was not significantly different among treatments (p>0.05), likely reflecting the linear increase in TWL and CWL.

Relative to breast meat color, meat redness linearly increased with increasing dietary digestiblelysine levels. This result may be due a significant increase in oxidative muscle fibers (red color) (Ono *et al.*, 1993). In addition, the higher b*values (yellowness) in relation to a* values (redness) obtained in the present study are consistent with the report of Souza *et al.* (2004), who observed thatthe breast muscle of poultry tends to present higher yellowness (higher b* values relative to a* values) compared with the meat of other animal species.

Experiment 3 - Performance, carcass traits, and of breast meat quality results obtained in the finisher phase (70 to 84 days old)

Performance and carcass traits

Dietary digestible lysine values influenced in a quadratic manner feed intake, weight gain and feed conversion ratio, and in a linear manner lysine intake and utilization efficiency (Table 7). The best weight gain (1007 g) and feed conversion ratio (2.87) results were obtained at estimated dietary digestible lysine levels of 1.162 and 1.126%, respectively (Table 10). These results are consistent with those obtained by Nascimento *et al.* (2016), who evaluated the effects of increasing digestible lysine values on the performance of broilers of the slow-growing strain Embrapa 041 during the period of 35-84 days of age and estimated digestible lysine requirements of 1.196 and 1.078% for optimal weight gain and feed conversion ratio, respectively.

Table 7 – Feed intake, weight gain, feed conversion ratio and lysine intake and utilization efficiency of slow-growing broilers in experiment 3 (70 to 84 days old).

	Digestible lysine (%)						
Variables	0.766	0.906	1.046	1.186	1.326	Regression*	CV[2] (%)
Feed intake, g	2941	2886	2826	2843	2853	Q	6.47
Weight gain, g	928	949	990	1040	977	Q	5.91
Feed conversion ratio	3.23	3.10	2.88	2.81	3.03	Q	7.66
Lysine intake, g	23.92	26.03	27.80	32.84	38.79	L	5.28
LUE[1] (%)	36.22	31.71	36.83	26.63	21.41	L	6.45

[1]LUE, lysine utilization efficiency

[2]CV (%), coefficient of variation; *L, Linear effect; *Q, quadratic effect (p<0.05). The equations are shown in Table 10.

The lowest feed intake (2832g) obtained in this phase was obtained at an estimated digestible lysine of 1.150%. In all experiments, feed intake decreased to a minimum and then increased as a function of dietary digestible lysine levels, which may be due to an improvement in the ratio of lysine to the other amino acids present in the diet (Conhalato *et al.*, 1999) and subsequent worsening of this relationship, when lysine was supplied in excess of the requirements.

Dietary digestible lysine values had a quadratic effect on carcass, breast and wing yields, and a linear effect on leg yield (Table 8). The best carcass yield (69.34%), breast yield (26.96%) and leg yield (31.95%) results were obtained at estimated dietary digestible lysine of 1.065, 1.195 and 1.107%, respectively.

Table 8 – Carcass and parts yields and relative weights of edible offal and abdominal fat of slow-growing broilers slaughtered at 85 days of age in experiment 3.

	Digestible lysine (%)						
Variables	0.776	0.906	1.046	1.186	1.326	Regression*	CV[1] (%)
Yield (%)							
Carcass	65.47	67.49	68.31	70.60	65.21	Q	4.51
Breast	26.39	26.76	26.87	26.93	26.92	Q	5.90
Leg	30.91	31.15	31.97	32.17	31.29	Q	10.67
Wing	12.14	12.32	12.19	12.34	12.55	NS	8.56
Back	24.60	25.38	25.31	25.77	24.15	NS	12.95
Relative weight (%)							
Heart	0.40	0.39	0.41	0.41	0.40	NS	9.19
Liver	1.59	1.62	1.63	1.66	1.71	NS	16.21
Gizzard	2.06	2.00	2.00	2.02	2.06	NS	21.01
Abdominal fat	2.45	3.15	2.97	3.68	4.05	L	51.42

[1]CV, Coefficient of variation;

*NS, Not significant; *L, Linear effect; *Q, quadratic effect ($p<0.05$). The equations are shown in Table 10.

These results are consistent with the findings of Rezaei *et al.* (2004), who described that dietary lysine levels may affect meat production, particularly breast meat yield, as this cut represents a large percentage of the carcass and contains significant amounts of lysine. According to Leclercq (1998), the breast muscle is the tissue that most profits from high dietary lysine values, as lysine is the main amino acid in breast muscle fibers. Breast development is directly related to body weight and muscle protein deposition and contributes to increase carcass weight, as reported by Trindade Neto *et al.* (2009) and Rosa *et al.* (2014), who observed that protein deposition in the breast muscle increased as a function of increasing dietary digestible lysine levels, resulting in higher breast yield, and consequently, higher carcass yield.

Giblet relative weights were not affected by the treatments ($p>0.05$). On the other hand, abdominal fat relative weight linearly increased as a function of dietary digestible lysine values. This result may be partially explained by the fact that feeding amino acids in excess of the requirements may limit lean tissue growth and increased fat deposition, as discussed in experiment 2. This effect is undesirable for both production and economic reasons: in addition of reducing carcass yield and feed efficiency (Gaya & Ferraz, 2006), chicken carcasses and cuts presenting excessive fat are rejected by the consumers (Meza *et al.*, 2015).

Breast meat quality

No significant effects of dietary digestible lysine values on the chemical composition of breast meat were detected in broilers slaughtered at 84 days of age (Table 9). These results are in agreement with those of Takeara *et al.* (2010) and Oliveira *et al.* (2013), who did not find any influence of dietary digestible lysine levels on the chemical composition of broiler carcasses.

The same effect of the dietary treatments on TWL of broilers slaughtered at 70 days of age (Experiment 2) was observed in Experiment 3. Although fresh meat moisture content was not different among treatments, significant TWL were observed, and the reasons for this result were previously discussed in experiment 2.

Meat texture is directly related to the amount of intramuscular water content and meat water-holding capacity; therefore, the higher the water content in muscle, the greater the meat tenderness (Anadón, 2002). However, meat tenderness, as determined by shear strength, was not significantly affected. The linear increase in TWL and CWL observed as dietary digestible lysine levels increased did not contribute reduce shear strength. These effects did not negatively influence meat tenderness because the mean value obtained in the present study was 3.90 kgf/g, and the reference value adopted by many authors for hard-boiled breast meat is 7.5 kgf/g, according to Lyon *et al.* (1985).

In general, the lysine requirements estimated in the present study are higher than those calculated in previous studies conducted with broilers of low genetic potential for growth (Nascimento *et al.*, 2009; Oliveira *et al.*, 2013; Rosa *et al.*, 2014). Moreover, the obtained values are also higher than those used in reference tables, such as the Brazilian Tables for Poultry and Swine (Rostagno *et al.*, 2011), or in manuals of genetic strains, such as the recommendations for Cobb broilers (Cobb-Vantress, 2013) and for Cobb Sasso 150 broilers (Cobb-Vantress, 2008).

Table 9 – Chemical and physical-chemical composition of the breast meat of slow-growing broilers slaughtered at 85 days of age in experiment 3.

	Digestible lysine (%)						
Variables	0.766	0.906	1.046	1.186	1.326	Regression*	CV[7] (%)
Chemical composition (%)**							
Moisture	73.12	72.96	73.09	73.22	72.89	NS	5.67
Crude protein	23.39	23.42	23.34	23.38	23.31	NS	6.32
Ether extract	1.54	1.49	1.51	1.50	1.45	NS	6.78
Ash	1.49	1.55	1.49	1.53	1.48	NS	4.98
Physical-chemical composition							
pH	5.94	5.95	5.94	5.89	5.97	NS	2.98
TWL[1] (%)	3.87	4.22	4.51	6.12	5.54	L	3.02
CWL[2] (%)	15.39	15.76	16.68	16.43	17.12	L	3.45
SS[3] (kgf/g)	3.78	3.98	3.87	3.93	3.92	NS	20.32
L*[4]	48.56	49.54	48.67	48.67	49.14	NS	23.83
a*[5]	6.73	6.69	6.71	6.74	6.69	NS	18.34
b*[6]	8.16	8.45	9.65	8.43	9.23	NS	25.99

[1]TWL, Thawing weight loss; [2]CWL, Cooking weight loss; [3]SS = Shear Strength

[4]Lightness; [5]Redness; [6]Yellowness

[7]CV, Coefficient of variation; *NS, Not significant; *L, Linear effect (p<0.05)

**Values determined in fresh meat

The higher estimates calculated in the present study may partly be due to differences in the calculation of the amino acid requirements (percentage), as well as to genetic strain. According to Baker & Han (1994), conventional broiler strains require almost the double amount of amino acids (milligrams) compared with slow-growing strains; however, this difference is offset by the daily feed intake of chickens of conventional lines. This result suggests that, as a percentage, the requirements of broiler with lower growth genetic potential may be higher due to their lower daily feed intake compared with conventional broiler strains.

The summary of the digestible lysine requirements estimates according to rearing phase, as determined by quadratic regression, with their respective equations and coefficients of determination are described in Table 10.

Table 10 – Summary of digestible lysine requirements of slow-growing broilers.

Experiment 1 (29-49 days old)			
Item	Estimated value (%)	R^2	Quadratic regression[1]
Feed intake, g	1.298	0.93	$\hat{Y}$ = 1140.7x² – 2962.3x + 4645.9
Weight gain, g	1.183	0.74	$\hat{Y}$ = -2208.5x2 + 5223.9x - 1939.7
Feed conversion ratio	1.203	0.91	$\hat{Y}$ = 6.0496x2 – 14.555x + 10.995
Carcass yield (%)	1.162	0.87	$\hat{Y}$ = -81.086x2 + 188.4x - 35.814
Wing yield (%)	1.179	0.61	$\hat{Y}$ = -5.9038x2 + 13.548x + 5.6194
Experiment 2 (50-69 days old)			
Feed intake, g	1.109	0.90	$\hat{Y}$= 2809.8x2 – 6296.7x + 7019.4
Weight gain, g	1.199	0.73	$\hat{Y}$ = -579.45x2 + 1390.1x + 73.683
Feed conversion ratio	1.162	0.87	$\hat{Y}$ = 3.7536x2 – 8.7233x + 8.2735
Carcass yield (%)	1.068	0.60	$\hat{Y}$ = -92,821x2 + 198,23x - 31.598
Breast yield (%)	1.107	0.89	$\hat{Y}$ = -25,292x2 + 56,01x - 2.2733
Leg yield (%)	1.101	0.95	$\hat{Y}$ = -24,162x2 + 53,213x - 0.1475
Wing yield (%)	1.142	0.85	$\hat{Y}$ = -3,2799x2 + 7,936x + 8.4294
Abdominal fat (%)	1.075	0.88	$\hat{Y}$ = 5.4665x2 – 12.498x + 8.429
Experiment 3 (70-84 days old)			
Feed intake, g	1.150	0.95	$\hat{Y}$ = 754.37x2 – 1734.6x + 3829.2
Weight gain, g	1.162	0.73	$\hat{Y}$ = -579,45x2 + 1347,2x + 224.32
Feed conversion ratio	1.126	0.87	$\hat{Y}$ = 3,0977x2 - 6,9732x + 6.7933
Carcass yield (%)	1.065	0.68	$\hat{Y}$ = - 48,652x2 + 103,63x + 14.158
Breast yield (%)	1.195	0.98	$\hat{Y}$ = -2,9519x2 + 7,0539x + 22.741
Leg yield (%)	1.107	0.76	$\hat{Y}$ = -10,423x2 + 23,076x + 19.173

[1]Regression was significant (p<0.05).

CONCLUSIONS

Digestible lysine levels of 1.203, 1.162 and 1.126% are recommended in the diets of Redbro Plume broilers during the phases 29-49, 50-69 and 70-84 days of age, respectively, to optimize feed conversion ratio.

Increasing dietary digestible lysine values affect the chemical composition (moisture, protein and ether extract contents) of the breast meat of broilers slaughtered at 70 days of age and thawing and cooking weight loss of the breast meat of broilers slaughtered at 70 and 85 days of age.

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