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Relationship of arginine with lysine in diets for laying Japanese quails

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ABSTRACT - To determine the relationship of arginine with lysine for Japanese quails during the period of production, an experiment was conducted using 360 subspecies of Japanese quails (*Coturnix coturnix japonica*) with 162 days of age, distributed in a completely randomized design. Diets were formulated with corn, soybean meal, sorghum and wheat bran containing 20.0% crude protein and 2,800 kcal ME/kg. The basal diet contained suboptimal level of lysine equal to 1% and was supplemented with five levels of L-arginine 99% (0.032; 0.083; 0.134; 0.185 and 0.236%) to replace the glutamic acid, corresponding to the relationship of arginine with digestible lysine of 1.16, 1.21, 1.26, 1.31 and 1.36. The parameters studied were: feed intake, egg production per hen/day, egg production per hen housed, commercial egg production, egg weight, egg mass, feed conversion by egg mass, feed conversion per dozen eggs, weight and percentage of components of the eggs (yolk, albumen and shell) and specific gravity. There was no significant effect on the relationship of arginine with digestible lysine in the diet of Japanese quails for any of the parameters examined. The arginine/lysine ratio of 1.16, which corresponds to a daily intake of 288.84 mg of arginine, provides satisfactory performance and egg quality of Japanese quails.

Key Words: *Coturnix coturnix japonica*, digestible amino acid, egg quality, performance

Introduction

According to the IBGE (2007), quail raising has shown considerable growth in recent years: the squab has grown 5.3% compared with 2006, while egg production has totaled about 131 million dozen. However, little is known about the nutrition parameters for quail; quail growers often uses foreign tables of nutritional requirements that can over-or underestimate the nutritional requirements of these birds in Brazilian conditions. Evolution of the genetic characteristics of the quail has been observed; these birds are generally heavier, more productive and laid larger eggs (Oliveira, 2007). With the introduction of different genotypes in the market, the need to update the nutritional requirements of these birds has become bigger.

Among the studies involving the nutrition of Japanese quails, many comprehend minerals and amino acids, especially lysine and methionine, and a few, threonine and tryptophan. However, little is known about the requirement of other amino acids such as arginine. Arginine is an essential amino acid, because since they lack the urea cycle, birds have the functional requirement of arginine greater than mammals (Baker, 1994). There is evidence that there is antagonism between arginine and lysine-induced imbalance in the relationship between these two amino acids, so that excess lysine stimulates arginase increased renal catabolism of arginine in the body, causing symptoms of deficiency of this amino acid, which can cause a decrease in productive responses. The increased level of arginine to lysine-rich diet relieves the depressant effect caused by this antagonism (Bertechini, 2006). A deficiency of arginine may cause changes in the structure of primary feathers, resulting in appearance described in the literature as “bird helicopter” (Cook et al, 1984).

Arginine is an amino acid available for studies of nutritional requirements, but its supplementation in diets is not yet common, although it is considered an essential amino acid and plays important functions in mammals and birds (Cynober et al., 1995). Currently, the recommended use of arginine in diets for quail are based on the NRC (1994), as there are no studies with this amino acid found in Brazil. Knowing that climatic conditions and management are factors that may influence the use of this recommendation, this research aimed to evaluate different relationships of arginine whit lysine in the diet of laying Japanese quails.

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Material and Methods

The experiment was conducted in the Setor de Avicultura do Departamento de Zootecnia da Universidade Federal de Viçosa, and lasted 63 days, in the period from January to March 2009.

Three hundred and sixty Japanese quails at 65 days of age were distributed in a completely randomized design, consisting of five treatments, nine replicates and eight birds per experimental unit.

A basal diet (Table 1) was made, supplemented with five levels of L-arginine 99% (0.032, 0.083, 0.134, 0.185, 0.236%), replacing the glutamic acid in protein equivalent, corresponding to the ratio of arginine to lysine in ratios of 1.16, 1.21, 1.26, 1.31 and 1.36. Differences arising from the balancing of protein equivalent to the arginine and glutamic acid in different ratios of arginine to lysine in evaluation were offset by the starch.

The relationships of digestible threonine, tryptophan and methionine plus cystine with lysine adopted in this research were the same used by Umigi et al. (2008), Pinheiro et al. (2008) and Reis et al. (2009), respectively. As the other relationships of amino acids were not determined on the digestible basis, they were seen maintaining the relationship of total amino acid with lysine as recommended in the NRC (1994). The energy level of the diet used was 2,800 kcal/kg, as determined by Moura et al. (2008); the levels of calcium and phosphorus were corrected for energy density of the diet according to the same authors. The other requirements were met in accordance with those recommended by the NRC (1994).

Table 1 - Percentage and calculated compositions in the experimental diets, as fed basis

Ingredients	T1	T2	T3	T4	T5
Corn	31.741	31.741	31.741	31.741	31.741
Soybean meal (45%)	28.503	28.503	28.503	28.503	28.503
Sorghum	23.500	23.500	23.500	23.500	23.500
Wheat bran	3.000	3.000	3.000	3.000	3.000
Soybean oil	1.988	1.988	1.988	1.988	1.988
Limestone	6.657	6.657	6.657	6.657	6.657
Dicalcium phosphate	1.039	1.039	1.039	1.039	1.039
Salt	0.322	0.322	0.322	0.322	0.322
DL-methionine (99%)	0.349	0.349	0.349	0.349	0.349
L-lysine HCL (79%)	0.193	0.193	0.193	0.193	0.193
L-isoleucine (99%)	0.185	0.185	0.185	0.185	0.185
L-valine (99%)	0.124	0.124	0.124	0.124	0.124
L-tryptophan (99%)	0.007	0.007	0.007	0.007	0.007
L-arginine (99%)	0.032	0.083	0.134	0.185	0.236
Starch	0.100	0.216	0.333	0.449	0.567
Glutamic acid	2.000	1.833	1.665	1.498	1.329
Choline chloride (60%)	0.100	0.100	0.100	0.100	0.100
Vitamin supplement[1]	0.100	0.100	0.100	0.100	0.100
Mineral supplement[2]	0.050	0.050	0.050	0.050	0.050
Antioxidant[3]	0.010	0.010	0.010	0.010	0.010
Total	100.00	100.00	100.00	100.00	100.00
Calculated composition					
Metabolizable energy (kcal/kg)	2.800	2.800	2.800	2.800	2.800
Crude protein (%)	19.97	19.97	19.97	19.97	19.97
Digestible lysine (%)	1.000	1.000	1.000	1.000	1.000
Total lysine (%)	1.080	1.080	1.080	1.080	1.080
Digestible arginine (%)	1.160	1.210	1.260	1.310	1.360
Digestible methionine + cystine (%)	0.840	0.840	0.840	0.840	0.840
Digestible threonine (%)	0.605	0.605	0.605	0.605	0.605
Digestible tryptophan (%)	0.210	0.210	0.210	0.210	0.210
Total isoleucine (%)	0.976	0.976	0.976	0.976	0.976
Total valine (%)	0.997	0.997	0.997	0.997	0.997
Calcium (%)	2.900	2.900	2.900	2.900	2.900
Available phosphorus (%)	0.300	0.300	0.300	0.300	0.300
Sodium (%)					
Crude fiber (%)	0.1402.920	0.1402.920	0.1402.920	0.1402.920	0.1402.920

[1] Composition/kg of product: vit. A - 12,000,000 U.I.; vit. D3 - 3,600,000 U.I.; vit. E - 3,500 U.I.; vit. B1 - 2,500 mg; vit. B2 - 8,000 mg; vit. B6 - 5,000 mg; pantothenic acid - 12,000 mg; biotin - 200 mg; vit. K - 3,000 mg; folic acid - 1,500 mg; nicotinic acid - 40,000 mg; vit. B12 - 20,000 mg; Selenium - 150 mg; vehicle qs - 1,000 g.
[2] Composition/kg of product: Mn - 160 g; Fe - 100 g; Zn - 100 g; Cu - 20 g; Co - 2 g; I - 2 g; vehicle qs - 1,000 g.
[3] Butylated hydroxy toluene - BHT (99%).
T = Treatment.

The composition and nutritional value of ingredients used to formulate the diet were calculated according to Rostagno et al. (2005).

Birds were housed in galvanized wire cages equipped with feeders and nipple drinkers. Each cage provided a space of 106 cm^2/bird.

Feed and water were provided *ad libitum* throughout the experimental period.

The daily management was to collect and to count the eggs (including eggs broken, cracked, with a soft shell and shelled), to record the temperatures (°C) and relative humidity in the experimental shed once a day, at 4 p.m. by using thermometers and to record maximum and minimum dry and wet bulb, positioned at a central point at the level of the birds. The lighting program consisted of providing 17 hours of light per day, controlled by an automatic timer that allowed the lights to turn on and off automatically overnight, according to the procedure adopted on commercial farms.

At the end of the experimental period (63 days), the amount of feed consumed (g/bird/day) in each experimental unit was determined. To do so, feed scraps and waste products were weighed and subtracted from the amount of feed supplied during the experimental period. The number of dead birds was subtracted from the total number of birds in each experimental unit, which allowed obtaining the correct consumption per bird.

The egg production was expressed as a percentage of bird day (egg/bird/day) and birds were housed at the beginning of the experiment (egg/housed bird). These data were obtained by computing the daily number of eggs produced, including the broken, the cracked and abnormal (eggs with soft shell and shelled) eggs. The average number of marketable eggs during the experimental period was calculated by subtracting the broken, cracked and abnormal eggs from the total number of eggs produced.

To obtain the specific gravity of eggs at 16, 17 and 18 days from each of the three experimental periods, all intact eggs were immersed and evaluated in NaCl solutions with densities from 1.055 to 1.100 g/cm^3, with ranges of 0.005 g/cm^3 between them.

All intact eggs produced in each replicate were weighed on a 0.001-g precision balance on days 19, 20 and 21 of each of the three experimental periods to obtain the average weight of eggs. After weighing, four eggs were randomly selected from each repetition for quantification. Eggs selected for each repetition and each day were individually weighed on a 0.001-g precision balance. After weighing, they were identified and subsequently broken. The yolk of each egg was weighed and recorded, its shell was washed, and air dried for further weighing. The weight of albumen was obtained by the difference between the egg weight and yolk weight plus the weight of the shell.

The average weight of eggs was multiplied by the total number of eggs produced in the period, resulting in the total mass of eggs per period. Total mass of eggs was divided by the total number of birds of the period and by the number of days in the period, and finally expressed in grams of egg/hen/day. The conversion per dozen eggs, expressed as the total feed intake in kilograms divided by the dozen eggs produced (kg/ dozen), and conversion by egg mass, which was obtained by the feed intake in kilograms divided by the mass of eggs produced in kilograms (kg/kg) were evaluated.

The parameters were subjected to analysis of variance and regression, according to SAEG statistical package (Sistema para Análises Estatísticas, versão 9.1). Estimates for the determination of the ratio of arginine with lysine was satisfactory through linear regression and quadratic, as the best fit obtained for each parameter, considering the biological behavior of birds.

Results and Discussion

Maximum, minimum and dry bulb temperatures and relative humidity were checked daily during the experimental period (Table 2).

At the mature phase, the thermal comfort range of quail is from 18 to 22 °C, and the relative humidity ranges from 65 to 70% (Oliveira, 2004). Thus, according to the values recorded for the dry bulb thermometer (Table 2), it is possible to observe that the quails were in mild conditions of heat stress in part of the experiment.

There was no significant effect (P>0.05) for arginine/lysine ratio in the diet on feed intake (Table 3). The values obtained for this parameter are in agreement with Baker & Albino & Barreto (2003), who mentioned the average daily consumption of feed per adult Japanese quails between 23

Table 2 - Values of temperature and relative humidity (RH), recorded in the experimental shed

Schedule	Air temperature (°C)			RH (%)
	Maximum	Minimum	Dry bulb	
08:00 a.m.	-	-	27.6±1.7	79.3±5.2
04:00 p.m.	27.1±2.5	18.3±3.6	26.2±1.9	66.2±8.7

and 26 g. Lobato & Costa (2009), evaluating the arginine requirement for laying Japanese quails using the levels 1.01, 1.09, 1.17, 1.25 and 1.33% observed quadratic behavior for feed consumption, determining the requirement of 1.185% arginine to this parameter. Lima et al. (2007) evaluated the effect of arginine with lysine on the performance of brown-egg pullets and laying hens, using a 3 × 2 × 2 factorial arrangement, consisting of three levels of arginine (0.64, 0.72 and 0.79%) two lysine levels (0.71 and 0.78%) and two strains, and found no interference of arginine on the genotypes, or vice-versa. However, evaluating separately, the strains were different in feed intake.

It can be inferred that, in the present study, the increase in the arginine with lysine was not sufficient to cause amino acid imbalance that resulted in changing the plasma profile of the animal by activating the mechanisms regulating appetite, as described by Harper et al. (1970). The increase in the ratio of these amino acids probably did not provoke antagonism between arginine and lysine, which would be verified by reducing consumption.

There was no significant effect (P>0.05) on the relationship of arginine with lysine on egg production. However, Lobato & Costa (2009) found a quadratic effect of arginine levels for laying Japanese quails, and this is maximized at the level of 1.166% of arginine. Lima et al. (2007) observed that hens fed diets containing 0.79% arginine and 0.71% lysine had higher egg production, which corresponds to the arginine to lysine ratio of 1.13.

With respect to egg production per hen housed there was no significant effect (P>0.05) by relationships utilized. In the literature, there are few scientific studies determining the production of eggs per quail housed. This is an important parameter to be observed, since the mortality of these birds during the laying period is high, compared with mortality in laying hens.

The relationship of arginine with lysine in the diet did not influence (P>0.05) the quality of eggs to be commercialized.

There was no significant effect (P>0.05) in the relationship of arginine with lysine in the diet on egg weight. When working with laying hens, Lima et al. (2007) concluded that levels of 0.72% arginine and 0.78% lysine resulted in greater weight of eggs.

For parameters for mass and feed conversion per dozen eggs also no significant effect was found (P>0.05). However, working with layers, Lima et al. (2007) found better values of feed conversion in the lower lysine level (0.71%) and the highest level of arginine (0.79%) corresponding to the arginine to lysine ratio of 1.13.

In studies with broilers (Costa et al. 2001; Atencio et al., 2004) found no influence of arginine levels on the performance of growing birds. However, Brake et al. (1998), also working with broilers, observed improved feed conversion linearly with increasing the arginine/lysine ratio

Table 3 - Arginine/lysine ratio on performance parameters

Parameter[2]	Arginine/lysine ratio					CV (%)[1]
	1.16	1.21	1.26	1.31	1.36	
Feed intake (g)	24.90	25.50	24.80	25.13	25.33	2.95
Eggs/day (%)	92.30	94.50	90.99	89.52	93.61	4.80
Eggs/housed hen (%)	89.77	91.31	90.34	86.22	91.53	6.60
Commercial eggs (%)	96.78	97.44	97.11	96.11	98.00	1.99
Egg weight (g)	11.61	11.59	11.64	11.34	11.68	2.60
Egg mass (g/bird/ day)	10.72	10.95	10.60	10.16	10.93	5.81
Feed conversion (kg/kg)	2.33	2.33	2.36	2.48	2.32	5.81
Feed conversion (kg/dozen)	0.32	0.32	0.33	0.33	0.32	4.98

[1] CV = coefficient of variation; [2] Not significant, P>0.05.

Table 4 - Arginine/lysine ratio in the diet on the parameters of egg quality

Parameter[2]	Arginine/lysine ratio					CV (%)[1]
	1.16	1.21	1.26	1.31	1.36	
Yolk weight (g)	3.41	3.33	3.37	3.36	3.43	2.72
Yolk (%)	29.32	28,96	29.00	29.30	29.48	2.35
Albumen weight (g)	7.27	7.24	7.32	7.17	7.25	2.53
Albumen (%)	62.50	62.87	62.91	62.50	62.26	1.18
Shell weight (g)	0.95	0.94	0.94	0.93	0.96	3.28
Shell (%)	8.18	8.17	8.09	8.20	8.26	3.00
Specific gravity (g/cm^3)	1.073	1.073	1.073	1.073	1.074	0.16

[1] CV = coefficient of variation; [2] Not significant, P>0.05.

from 1.05 to 1.49 in the diet, at a temperature of 31 ºC; this result indicates that high temperatures require high ratios of arginine/lysine in the diet.

There was no significant effect (P>0.05) on the relation of arginine with lysine in the diet of Japanese quails for any parameters of egg quality (Table 4).

The relationship of arginine with lysine did not affect the specific gravity of eggs; this leads to the conclusion that this amino acid has no influence on external quality of egg shell as it also did not influence the weight of them. According to Hamilton (1978), egg weight is a factor of change in shell quality, because t heavier eggs tend to have weaker shells due to the bird inability to deposit calcium in the same proportion for growth and egg weight.

Conclusions

The arginine/lysine ratio of 1.16, which corresponds to a daily intake of 288.84 mg of arginine, provides satisfactory results of performance and quality of eggs of Japanese quails.

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