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Ideal ratio of digestible methionine plus cystine to digestible lysine for growing Japanese quails¤

Proporción ideal de metionina más cistina digestibles a lisina digestible para codornices japonesas en crecimiento

Relação ideal de metionina mais cistina digestível e lisina digestível na dieta de codornas japonesas na fase de crescimento

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Summary

Background: methionine is considered essential for maintenance, growth, and feather development. Methionine supply during the growth phase can influence the weight at sexual maturity and egg size. **Objectives:** to evaluate the effects of several digestible methionine + cystine/digestible lysine ratios for growing Japanese quails (1 to 40 days of age) with repercussions on the early stage of production (41 to 110 days of age). **Methods:** a total of 1,000 one-day-old Japanese quails were randomly distributed to five digestible (methionine + cystine)/digestible lysine ratios, with 10 replicates and 20 birds per experimental unit. A basal diet formulated to meet all nutrient requirements was added with graded levels of DL-methionine replacing glutamic acid. **Results:** weight of the birds at 40 days, weight gain until 40 days, feed intake, carcass weight, fat, and body protein deposition, methionine + cystine intake and birds viability in the growing phase presented a linear trend. A quadratic effect was observed for feed conversion and uniformity at 40 days of age, and percentage of feathers did not vary. A linear increase occurred in the laying phase for feed intake and weight gain of the birds. No effects of the assessed ratios were observed on the other variables. **Conclusions:** a 0.73 ratio of digestible (methionine + cystine) to digestible lysine in the growing phase provided uniform growth and satisfying performance of Japanese quails during the laying phase.

Keywords: *Coturnix japonica, ideal protein, productive performance, sulfur amino acids.*

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Resumen

Antecedentes: la metionina es esencial para el mantenimiento, crecimiento y desarrollo de las plumas. El suministro de metionina durante la fase de crecimiento puede influir en factores tales como el peso corporal a la madurez sexual y el tamaño del huevo. **Objetivo:** evaluar los efectos de varias proporciones de metionina + cistina digestibles y lisina digestible en la dieta de codornices japonesas en fase de crecimiento (1 a 40 días de edad) y su efecto en la producción inicial de huevos (41 a 110 días de edad). **Métodos:** se utilizaron 1.000 codornices japonesas de un día de edad, en un diseño aleatorizado con cinco proporciones de (metionina + cistina) digestible/lisina digestible, con 10 replicaciones y 20 aves por unidad experimental. La dieta basal formulada se complementó con cinco niveles de DL-metionina en sustitución del ácido glutámico. **Resultados:** el peso de los animales al día 40, la ganancia de peso hasta el día 40, la ingesta de alimento, el peso de la carcasa, la deposición de grasa y proteína corporal, la ingesta de metionina + cistina y la viabilidad de las aves en la fase de crecimiento apresentaram uma tendência linear. Se observó un efecto cuadrático para conversión de alimento y uniformidad a los 40 días de edad y el porcentaje de plumas no varió. Durante el período de producción de huevos hubo un aumento lineal para consumo de alimento y ganancia de peso de las aves. Não foram observados outros efeitos proporções em relação a outras variáveis. **Conclusiones:** una proporción de 0,73 entre metionina + cistina digestible/lisina digestible en la fase de crecimiento provee un desarrollo uniforme y un rendimiento satisfactorio en la producción de huevos de la codorniz japonesa.

Palabras clave: *aminoácidos azufrados, Coturnix japonica, proteína ideal, rendimento productivo.*

Resumo

Antecedentes: a metionina é considerada essencial para manutenção, crescimento e desenvolvimento das penas nas codornas. O fornecimento nutricional de metionina durante a fase de crescimento pode influenciar fatores como peso corporal para maturidade sexual e tamanho do ovo. **Objetivo:** avaliar os efeitos entre metionina + cistina digestível e lisina digestível na dieta para codornas japonesas na fase de crescimento (1 a 40 dias de idade) com repercussão na fase inicial de postura (41 a 110 dias de idade). **Métodos:** foram utilizadas 1.000 codornas japonesas com 1 dia de idade, distribuídas em delineamento inteiramente casualizado com 5 relações entre (metionina mais cistina) digestível/lisina digestível, 10 repetições e 20 aves por unidade experimental. Foi formulada uma ração basal suplementada com cinco níveis de DL-metionina em substituição ao ácido glutâmico. **Resultados:** o peso corporal das aves até os 40 dias, ganho de peso até os 40 dias, consumo de ração, peso de carcaça, deposição de gordura e de proteína corporal, consumo de metionina + cistina e viabilidade das aves na fase de crescimento houve aumento linear para. Efeito quadrático foi observado para conversão alimentar e uniformidade aos 40 dias e a porcentagem de penas não variou. Na fase de postura, ocorreu aumento linear para consumo de ração e ganho de peso das aves. **Conclusões:** a relação metionina + cistina e lisina digestível de 0,73 na dieta de recria proporcionou crescimento uniforme e desempenho satisfatório de codornas japonesas na fase de postura.

Palavras chave: *aminoácidos sulfurosos, Coturnix japônica, proteína ideal, rendimento produtivo.*

Introduction

Quail farming is very important for providing jobs and producing high quality protein at a low cost. Nutrition during the growing phase of the birds can influence their performance during the production phase. Most research has focused on the nutritional requirements of quails during the laying phase and there is little information about requirements during the growing phase. Quails are characterized by early sexual maturity (40 days), demanding feeding programs that maximize growth by combining body development with sexual maturity, thus allowing uniformity of breeding stock and productivity (Pinto *et al.,* 2003).

The ideal protein concept implies feeding the best ratios between lysine and other amino acids, thus reducing the crude protein content of the diet. This is possible due to the availability of industrial amino acids in the market. Methionine, the first limiting amino acid for quails (Mandal *et al.,* 2005) is essential for maintenance, growth and feather development. High protein ingredients (e.g. soybean) are commonly used to meet the methionine + cystine requirements of quails during the initial phase. Nevertheless, this practice increases production costs, deviate amino acids for functions not related to growth, worsens environmental conditions, and increases pollution. The use of synthetic amino acids is an interesting alternative to minimize those problems (Silva and Costa, 2009). Therefore, the objective of this study was to evaluate the ratio between digestible methionine + cystine to digestible lysine [d(Met + Cys)/dLys] in the diet for growing Japanese quails.

Materials and methods

Ethical considerations

This study was aproved by the Ethics Commitee for the Use of Animals of the Departament of Animal Science, Universidade Federal de Viçosa (Brazil), in September 13th, 2011 (process number 13/2011).

Growing phase (from 1 to 40 days of age)

The experiment was conducted in the Poultry Faculty of the Department of Animal Science (Departamento de Zootecnia) at the Federal University of Viçosa (Universidade Federal de Viçosa). A total of 1000 one-day-old female Japanese quails (*Coturnix japonica*) were used (body weight 7.59 ± 0.15 g). The animals were randomly distributed into five diets with increasing d(Met + Cys)/dLys ratios. The experimental unit consisted of 20 birds per cage, with 10 replicates. Each cage measured 50 cm^2 (width and length) and had a wooden floor covered by 4 cm of shavings, providing 125 cm^2/bird of floor space. One sixty-watt light bulb was placed in each cage to warm the birds until they reached 15 days of age.

The cages were set inside a shed. They were made of galvanized wire and covered and managed under a raffia curtain for the first 15 days of the birds' life to keep the place warm. This was also achieved by hanging curtains on the laterals of the shed. One glass pressure drinker was installed per cage. Quails were guided to the water right after being housed. The glass pressure drinkers were replaced by nipple drinkers at 12 days of age. One tray feeder (diameter: 18 cm) was used per cage until birds were 12 days of age and was then replaced by a gutter feeder placed in the front of the experimental units.

The units had 24 hours a day of artificial light (with the same light bulbs used for warmth) until birds were 15 days old. Then, only natural light was provided until birds were 40 days old to avoid premature sexual maturity. Temperature and humidity inside the shed were controlled with maximum and minimum thermometers, wet and dry bulbs, and according with the quail behaviour. Temperature was recorded at 4:00 p.m. and humidity at 9:00 a.m. and 3:00 p.m.

Birds were fed a corn-soybean meal basal diet deficient in methionine + cystine, with 205 g CP/kg, 12.14 MJ/kg, 10.5 g of digestible lysine/kg (sub-optimal level) and 5.67 g digestible methionine + cystine/kg, corresponding to a d(Met + Cys)/dLys ratio of 0.54 (Table 1).

This basal diet was supplemented with five graded levels of DL-methionine (99%), replacing glutamic acid by a protein equivalent corresponding to the d(Met + Cys)/dLys ratio of 0.54 (5.67 g digestible methionine + cystine/kg, with no supplementation); 0.60 (6.3 g digestible methionine + cystine/kg); 0.66 (6.93 g digestible methionine + cystine/kg); 0.72 (7.56 g digestible methionine + cystine/kg); and 0.78 (8.19 g digestible methionine + cystine/kg). The diets were iso-caloric and iso-nitrogen. The ratios between amino acids and lysine proposed by NRC (1994) were maintained, except for methionine + cystine to lysine, which varied according to the recommendation by Pinto *et al*. (2003). To make sure the diet was not deficient in any amino acid, 3% was added for each amino acid requirement, except for methionine + cystine and digestible lysine.

When quails reached 40 days they had their beaks trimmed before moving them to egg laying cages. This was to prevent cannibalism and was in accordance with the guidelines of the Conselho Nacional de Controle de Experimentação Animal – CONCEA and was approved by the Ethics Committee of the Department of Animal Science at the Federal University of Viçosa.

Laying phase (from 41 to 100 days of age)

Birds were moved to laying cages at 41 days of age keeping the same birds from each experimental unit

Table 1. Composition (dry matter basis) and nutritional value of the diets for Japanese quails in the growing phase.

Ingredient (g/kg)	Methionine + cystine/lysine ratio				
	0.54	**0.60**	**0.66**	**0.72**	**0.78**
Corn	628.8	628.8	628.8	628.8	628.8
Soybean meal (45%)	326.0	326.0	326.0	326.0	326.0
Limestone	11.8	11.8	11.8	11.8	11.8
Dicalcium phosphate	10.7	10.7	10.7	10.7	10.7
Salt	3.5	3.5	3.5	3.5	3.5
Mineral mixture[1]	0.5	0.5	0.5	0.5	0.5
Vitamin mixture[2]	1.0	1.0	1.0	1.0	1.0
Choline chloride (60%)	3.8	3.8	3.8	3.8	3.8
Antioxidant[3]	0.1	0.1	0.1	0.1	0.1
Coccidiostat[4]	0.5	0.5	0.5	0.5	0.5
L- Lysine (78.8%)	1.1	1.1	1.1	1.1	1.1
DL- Methionine (98.2%)	0.0	0.6	1.2	1.8	2.4
L- Threonine (98%)	1.8	1.8	1.8	1.8	1.8
L- Isoleucine (98%)	0.4	0.4	0.4	0.4	0.4
L-Glutamic	5.0	4.4	3.8	3.2	2.6
Starch	5.0	5.0	5.0	5.0	5.0
Total	1000.0	1000.0	1000.0	1000.0	1000.0
Calculated composition					
Metabolizable energy (MJ/kg)	12.14	12.14	12.14	12.14	12.14
Crude protein g/kg	205.00	205.00	205.00	205.00	205.00
Calcium g/kg	8.00	8.00	8.00	8.00	8.00
Available phosphorus g/kg	3.00	3.00	3.00	3.00	3.00
Sodium g/kg	1.50	1.50	1.50	1.50	1.50
Crude fiber g/kg	28.50	28.50	28.50	28.50	28.50
Amino acids					
Lysine g/kg	10.50	10.50	10.50	10.50	10.50
Methionine + Cystine g/kg	5.67	6.3	6.93	7.56	8.19
Tryptophan g/kg	2.09	2.09	2.09	2.09	2.09
Threonine g/kg	8.55	8.55	8.55	8.55	8.55
Arginin g/kg	10.39	10.39	10.39	10.39	10.39
Isoleucine g/kg	8.23	8.23	8.23	8.23	8.23
Valine g/kg	7.98	7.98	7.98	7.98	7.98

[1]Composition/kg of the product: manganese: 160g (as MnO_2); iron: 100g (as $FeSO_4$ -$7H_2O$); zinc: 100g (as $ZnSO_4$); copper: 20g (as $CuSO_4$ - $5H_2O$); cobalt: 2g (as $CoSO_4$ $7H_2O$); iodine: 2g (as KI); excipient q.s.p.: 1000 g.

[2] Composition/kg of the product: retinol acetate: 12.000 mg, cholecalciferol: 3.600 mg, DL-tocopheryl acetate: 3.500 mg, thiamine hydrochloride: 2.500 mg, riboflavin-5-phosphate sodium: 8.000 mg, pyridoxine hydrochloride: 5.000 mg, D-calcium pantothenate: 12.000 mg, D-biotin: 200 mg, menadione nicotinamide bisulphite: 3.000 mg, folic acid: 1.500 mg, nicotinic acid: 40.000 mg, cyanocobalamin: 20.000 mg, selenium: 150 mg (as Na_2SeO_3-$5H_2O$); vehicle q.s.p.: 1.000g.

[3] Butyllated hidroxytoluene.

[4] Salinomycin 12%.

together to evaluate growth and development during the laying phase. The experimental units were 50 x 34 cm galvanized wire cages in a ladder arrangement, equipped with gutter feeders in the front section and corresponding nipple drinkers. The stocking density per experimental unit was 106.2 cm^2/bird. Light was provided 16 hours a day during the experimental period. Light supply was controlled with a timer, allowing the lights to be turned on and off at night and early morning, in accordance with common procedures in commercial poultry farms.

Temperature and humidity inside the shed were daily recorded. The quails were fed a single diet based on corn and soybean (Table 2) in the laying phase. This diet was formulated to meet the nutritional requirements proposed by Rostagno *et al*. (2011).

Evaluated parameters

Feed intake was weekly measured and the average intake of died birds was subtracted to calculate the real consumption per experimental unit. Feed conversion in the growing phase was calculated by dividing feed intake by the body weight gain accumulated in the period (kg of diet/kg weight gain). All birds were weighed at the beginning and at the end of the growing phase and at the laying phase to determine weight at 40 and 110 days, and weight gain in each phase. Mortality was daily recorded to determine bird viability in the growing and laying phases. All birds were weighed to determine uniformity of experimental units and these weights were expressed as a percentage of the individual weights, which were within 10% of the mean.

On reaching 40 days, two birds that were within the average weight of each experimental unit were slaughtered. Birds were slaughtered according to the protocol for animal use in the Department of Animal Science at the Federal University of Viçosa. Birds were dry-plucked and feathers were weighed for feathering calculations. Next, birds were eviscerated, cooled, frozen, and ground to determine the contents of dry matter, ether extract and crude protein according to the AOAC (2000). Body fat and protein deposits were calculated by slaughtering an additional group of 50 one-day-old quail. They were compared to body fat and protein deposits of the quail slaughtered at the end of the growing phase.

Table 2. Composition (dry matter basis) and nutritional value of the diet for quails in the laying phase.

Ingredient (g/kg)	Composition (g/kg)
Corn	581.2
Soybean meal	313.8
Limestone	71.1
Dicalcium phosphate	11.0
Soybean oil	12.0
Common refined salt	3.3
Mineral mixture[a]	0.5
Vitamin mixture[b]	1.0
DL- Methionine	3.0
L-Lysine HCL	2.0
Antioxidant[c]	0.1
Choline chloride	1.0
Total	1000.0
Calculated composition	
Metabolizable energy (MJ/kg)	11.72
Crude protein (g/kg)	193.10
Lysine (g/kg)	10.80
Methionine + cystine (g/kg)	8.64
Tryptophan (g/kg)	2.26
Threonine (g/kg)	5.93
Calcium (g/kg)	30.90
Available phosphorus (g/kg)	3.00
Sodium (g/kg)	1.45
Crude fiber (g/kg)	27.00

[a]Provided g/kg diet: manganese: 0.08, iron: 0.05, zinc: 0.05, copper: 0.01, cobalt: 0.001, iodine: 0.001, excipient q.s.p.: 1000 g.
[b]Provided g/kg diet: retinol: 36.0 mg, cholecalciferol: 0.89 mg, tocopherol: 23.33 mg, thiamine: 2.5 mg, riboflavin:8.0 mg, pyridoxine: 5.0 mg, pantothenic acid: 12.0 mg, biotin: 2.0 mg, naphthoquinone: 3.0 mg, folic acid: 1.5mg, nicotinic acid: 40.0 mg, cobalamin: 20.0 mg, selenium: 1.5 mg, vehicle q.s.p.: 1.000g.
[c]Butylated hydroxytoluene.

For the calculation of body fat and protein deposits, the value found in the sample was compared with the average weight of the live animal in each experimental unit, according to Pinto *et al*. (2003).

To obtain the average egg production in the period, the number of eggs produced, including cracked, broken and abnormal eggs, were expressed over the number of birds in the period (egg produced/bird/day) and over the number of birds housed at the beginning of the experiment (egg produced/housed bird). Feed conversion per dozen eggs was calculated as total feed intake in kilograms divided by a dozen eggs produced (kg/dz). Conversion per egg mass was calculated by dividing feed intake into kilograms by egg mass produced in kilograms (kg/kg). All the intact eggs produced per replicate in the last three days of each week were weighed and total weight was divided by the number of eggs weighed. The average weight of eggs was multiplied by the total number of eggs produced in the period. This total mass was divided by the total number of birds and also by the number of days in the period and expressed in grams of egg per bird, per day (g egg/bird/day).

Statistical analysis

The treatment effects were estimated by an analysis of variables ($p<0.05$) using linear and quadratic regression models and including the linear response plateau (LRP), in accordance with the best fit obtained for each variable and considering the biological response of birds. The SAS software (SAS Inst. Inc, Cary, NC, USA, 2014) was used for statistical analysis.

Results

Quails need a thermohygrometric environment of 35-37 °C with 65% relative humidity in the first week of life, and 32 °C with 60% relative humidity in the second week (15 days). Thereafter, birds no longer require artificial heating because they present good feathering at the optimum maximum and minimum temperatures of 31 °C and 19 °C, respectively, with 60 to 65% relative humidity (Vohra, 1971; Reis, 1980; Singh and Narayam, 2002; Pinto *et al.,* 2003; Oliveira, 2004). In the adult phase (data from 41 to 100 days of age) the thermal comfort zone of quails is within 18 and 22 °C and air relative humidity is between 65 and 70% (Oliveira, 2004).

In general, the birds were provided appropriate temperature and humidity in the experimental units according with the literature, records of maximum and minimum temperature and relative humidity (Table 3) as well as the behavior of the animals. Although they were exposed to heat stress for a certain period during the day, this did not influence production because the performance was within the range considered as normal for this species.

Table 3. Temperature and air humidity in the experimental shelter.

Age of the birds (days)	Air temperature (°C)	Relative humidity (%)	
		9h00 a.m.	3h00 p.m.
1 to 7	31.64 ± 2.13	89.07 ± 3.17	76.86 ± 13.46
8 to14	29.10 ± 2.03	84.50 ± 6.03	76.43 ± 10.75
15 to 40	25.12 ± 2.54	87.11 ± 6.07	72.31 ± 9.93
41 to 100	24.38 ± 2.17	86.94 ± 5.85	71.71 ± 8.97

The upper limit of the thermoneutrality zone is 27 °C (Vercese, 2010). This limit was not exceeded in the experimental units after birds presented complete feathering, thus preventing them from any potential negative environmental influence on performance.

During the growing phase, increasing levels of inclusion of methionine in the diet resulted in a linear increase ($p<0.01$) in body weight at 40 days ($\hat{Y}= 91.8470 + 54.9833 X$; $R^2 = 0.91$), in weight gain at 40 days ($\hat{Y} = 84.3500 + 54.8333 X$; $R^2 = 0.91$), feed intake ($\hat{Y} = 5.34000 + 7.66667 X$; $R^2 = 0.85$), methionine + cystine intake ($\hat{Y} = -33.3400 + 160.667 X$; $R^2 = 0.99$), carcass weight ($\hat{Y} = 63.4220 + 50.0333 X$; $R^2 = 0.99$), fat deposition ($\hat{Y} = 0.0350000 + 0.916667 X$; $R^2 = 0.80$), protein deposition in the carcass ($\hat{Y} = 0.240000 + 0.366667 X$; $R^2 = 0.99$), and viability ($\hat{Y} = 86.7000 + 15.0000 X$; $R^2 = 0.76$; Table 4).

However, feed conversion and uniformity of birds at 40 days were quadratically influenced ($p<0.01$) and feed conversion ($\hat{Y} = 1.82400 + 15.1762 X - 10.7143 X^2$; $R^2 = 0.94$) worsened up to the estimated ratio of 0.71 and uniformity at 40 days ($\hat{Y}= -235.784 + 866.171 X - 595.635 X^2$; $R^2 = 0.99$) was improved by a 0.73 d(Met + Cys)/dLys. The ratios between d(Met + Cys)/dLys did not influence ($p>0.05$) feathering percentage.

In the laying phase, only feed intake, weight gain and feed conversion per dozen eggs were influenced ($p<0.01$) by the d(Met + Cys)/dLys ratios in the diets fed in the growing phase. A linear effect ($p<0.01$) occurred for feed intake ($\hat{Y} = 29.0620 + 9.1666 X$; $R^2 = 0.70$) and weight gain at 110 days of age ($\hat{Y} = 74.7900 + 49.5000 X$; $R^2 = 0.86$). Although feed conversion per dozen eggs had linearly improved, LRP had the best fit ($p<0.01$), estimating in 0.69 ($\hat{Y} = 0.6300 + 0.3333 X$, $R^2 = 0.99$; $y = 0.400$) the

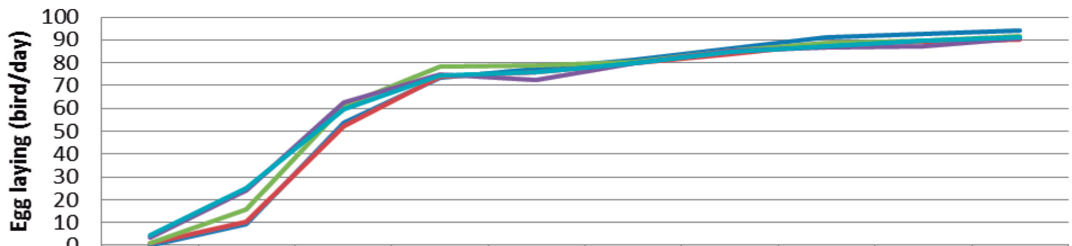
Table 4. Performance and laying of growing Japanese quails in function of the methionine + cystine/lysine ratio during the growing phase.

Parameter	Methionine + cystine:lysine ratio					CV (%)
	0.54	**0.60**	**0.66**	**0.72**	**0.78**	
Growing phase (1-40 days of age)						
Body weight at 40 days of age (g)[1]	119.6	126.7	129.6	130.8	134.0	3.7
Weight gain (g)[1]	112.0	119.1	122.0	123.2	126.4	3.9
Feed intake (g/bird/day)[1]	9.1	10.3	10.6	10.9	11.1	4.5
M+C intake (mg/bird/day)[1]	51.3	65.1	73.7	82.7	90.7	4.2
Feed conversion (g/g)[2]	3.23	3.47	3.49	3.55	3.50	4.5
Carcass weight (g)[1]	89.9	93.7	97.2	99.1	102.2	6.4
Feathering (g/kg)[ns]	68.0	67.0	65.0	56.0	60.0	8.5
Fat deposition (g/day)[1]	0.058	0.052	0.063	0.071	0.076	20.8
Protein deposition (g/day)[1]	0.44	0.46	0.48	0.50	0.53	9.1
Uniformity at 40 days (%)[2]	58.4	68.8	77.6	78.2	77.7	16.9
Birds viability (%)[1]	94.0	97.0	96.5	97.0	98.5	4.2
Egg laying phase (41–110 days of age)						
Egg production/bird/day[ns]	0.665	0.645	0.674	0.669	0.673	7.8
Egg production/housed bird/day[ns]	0.655	0.622	0.659	0.649	0.651	8.7
Feed intake (g/bird/day)[1]	24.8	22.9	22.9	22.0	22.4	7.5
Feed conversion/dozen (kg/dozen)[3]	0.45	0.43	0.41	0.39	0.40	9.5
Egg weight (g)[ns]	10.41	10.1	10.35	10.37	10.34	2.5
Egg mass (g/bird/day)[ns]	6.92	6.52	6.98	6.95	6.96	8.7
Conversion/egg mass (kg/kg)[ns]	3.30	3.92	3.63	3.75	3.74	13.4
Body weight at 110 days (g)[ns]	170.0	169.9	169.7	170.0	171.6	2.8
Weight gain (g)[1]	50.4	43.3	40.1	39.2	37.6	11.3
Uniformity at 110 days (%)[ns]	76.8	76.6	80.6	84.3	77.2	15.2
Bird viability (%)[ns]	97.4	95.9	94.0	96.1	93.7	5.6

[1]Linear effect ($p<0.01$); [2]Quadratic effect ($p<0.01$); [3]LRP ($p<0.01$); [ns]Not-significant effect ($p\geq0.05$).

best d(Met + Cys)/dLys ratio during the growing phase.

Daily egg production per bird linearly increased ($p<0.01$) in function of amino acid ratios in the first two weeks of production according to the equations for the first week: $\hat{Y} = -10.9184 + 20.1133 X$; $R^2 = 0.91$; and 2nd week: $\hat{Y} = -37.1919 + 83.0017 X$; $R^2 = 0.96$. Egg production/bird/day only presented differences ($p>0.05$) from the third week (Figure 1). No effect was observed ($p>0.05$) for feathering in the growing phase or in the laying phase for egg production/bird/day or per housed bird, egg weight, egg mass, feed conversion/egg mass, viability, weight, or uniformity of birds at 110 days.



d(Met + Cys)/dLys ratios	1	2	3	4	5	6	7	8	9	10
0,54	0,13	9,249	53,757	73,52	77,317	81,299	86,21	91,043	92,749	94,018
0,60	1,116	10,21	52,239	74,078	75,872	79,793	83,611	88,095	89,14	90,306
0,66	1,061	15,792	60,985	78,536	79,05	80,242	85,316	88,544	89,759	91,838
0,72	3,548	23,879	62,759	74,712	72,233	79,843	85,233	86,709	87,428	90,707
0,78	4,245	24,938	59,416	74,427	75,732	79,715	84,876	87,323	89,486	91,016

Production Weeks

Figure 1. Egg production per week of Japanese quails in function of the ratios between digestible (methionine + cystine) to digestible lysine in the growing phase.

Discussion

Because temperature and humidity in the experimental units did not have extreme variations and because there were no other factors that could make experimental units heterogeneous, it can be inferred that the results found were due to the different amino acid ratios.

High methionine + cystine to lysine ratios provided greater growth of birds. Low ratios were related to low weight of the animals, increased feed ingestion, and greater weight gain during the production phase. Therefore, at the end of the growing period, lighter birds had greater diet intake during the laying stage, resulting in a compensatory gain with greater feed intake so all birds reached a similar weight at 110 days of age. Quails fed diets with the lowest d(Met + Cys)/dLys ratios (0.54 and 0.60) did not have a proper weight at 40 days due to greater egg production in the early phase of egg laying, extending the growing phase to the age when birds usually would be in the laying phase.

Similarly, Pinto *et al.* (2003) found a linear effect for body weight and weight gain in function of the methionine + cystine/lysine ratios in the diet of growing quails. According to these authors, a ratio lower than 0.68 between those amino acids may compromise weight gain in the growing phase.

Since there was no difference ($p>0.05$) for feathering, birds might have used energy and protein mostly for maintenance instead of growth. Total dietary protein and amino acids such as cystine, methionine, arginine, isoleucine, leucine, valine, lysine, serine, threonine, histidine, phenylalanine, tyrosine, and tryptophan are associated to feather formation.

A deficiency in crude protein could result in deficient feathering of birds. This allows us to infer that the protein reduction in this study with amino acids supplementation in isonitrogenic diets did not affect maintenance and feathering. However, regarding performance, a greater d(Met + Cys)/dLys ratio is required by quails.

A quadratic effect in the methionine + cystine to lysine ratio was observed by Pinto *et al.* (2003) on bird feathering, which was increased to a ratio of 0.61. Because the ratios which have provided the highest feathering were lower than the best responses for weight gain and feed conversion, those authors stated that this parameter is not proper to set the requirements of methionine + cystine/lysine ratio for growing quails. Although a similar response was noted in this study, the best result for feathering was obtained using a lower ratio than that required for weight gain and feed conversion. Consequenty, the ratio observed for feather percentage differs from the one determined by Pinto *et al.* (2003).

Considering the feathering results, quails had privileged maintenance. Because of a large interaction between methionine and maintenance requirement, a deficiency of this amino acid might have occurred to other functions in birds fed lower ratios between methionine + cystine/lysine. Since methionine is an important sulfur amino acid in the translation of messenger RNA, being the first amino acid incorporated in the N-terminal position of all proteins, methionine requirement for maintenance and synthesis of feather proteins is as important as the need for body protein synthesis. Therefore, diets that did not meet the needs of sulfur amino acids of birds or did not meet the appropriate methionine + cystine/lysine ratio did not provide the best development conditions, reducing viability and uniformity in the growing phase.

The increase in amino acid ratios provided bird's greater feed intake and a methionine + cystine/lysine ratio in the growing phase. Quails fed greater ratios presented better development and weight gain, resulting in greater carcass weight and carcass protein deposition, which was also followed by greater fat deposition. Results different from the methionine + cystine/lysine ratios on the parameters of body composition were observed by Pinto *et al.* (2003) who found a quadratic variation with a maximum protein deposition rate at 0.746% methionine + cystine, corresponding to a 0.65 ratio.

In this study, the best d(Met + Cys)/dLys ratio for feed conversion was 0.71, although it was greater for the best weight gain. Pinto *et al.* (2003) reported 0.66 d(Met + Cys)/dLys ratio as the best response for feed conversion. The d(Met + Cys)/dLys ratios recommended for growing quails by Rostagno *et al.* (2011) and the total amino acids by the NRC (1994; 0.68 and 0.58, respectively), as well as the 0.54 recommended by Svacha *et al.* (1970), would not meet the requirements for maximum final weight, weight gain, and feed conversion estimated by recent studies and the present work. Nevertheless, heavier quails did not achieve greater egg production/bird/day or per housed bird, which does not justify the adoption of the ratio by weight gains of birds at 40 and 110 days.

Production parameters such as egg weight and egg mass as well as feed conversion/egg mass were not influenced by d(Met + Cys)/dLys ratios during the growing phase, but lighter birds achieved lower production in the first two initial weeks because of the deviation of dietary nutrients for weight gain. The 0.54 and 0.60 d(Met + Cys)/dLys ratios provided lower bird weight until six weeks than that reported by Leeson and Summers (2005), and Lima *et al.* (2011). Thus, quails fed diets with the lowest ratios delayed to start the egg-laying phase, presenting a production equal to the others only from the third production week. Feed conversion per dozen eggs was worst for quails fed the lowest ratios (0.54 and 0.60) in the growing phase due to a higher feed intake in the laying phase directed to weight gain. No reports on methionine + cystine/lysine ratios on the growing phase with repercussion on the laying phase of quails were found in the literature.

The best ratio for uniformity of quails was 0.73 at 40 days, so these birds maintained a uniform growth and would not need higher feed intake in the laying phase. This ratio is higher than 0.58, recommended by NRC (1994), than 0.66, estimated by Pinto *et al.* (2003) and higher than 0.68, recommended by Rostagno *et al.* (2011), although lower than 0.85, determined by Leeson and Summers (2005), and closer to 0.70, determined by Silva and Costa (2009).

In conclusion, the 0.73 d(Met + Cys)/dLys ratio (7.66 g of methionine + cystine/kg and 10.5 g of lysine/kg) in the growing diet, corresponding to 83.85 mg intake of methionine + cystine/bird/day provides uniform growth and better productive performance for Japanese quails during the early egg-laying phase.

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Conflicts of interest

The authors declare they have no conflicts of interest with regard to the work presented in this report.

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