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Lysine Requirements of Female Japanese Quails Base on Performance and Carcass Variables from Twenty-One to Forty-Two Days of Age

Research Article

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ABSTRACT

The present research was conducted to estimate the dietary lysine (Lys) requirement of growing female quail base on performance and carcass constitutes at 24 to 42 d of age. A dose-response diet mainly based on corn, corn gluten and soybean meal was used. Lys content was minimized in dose-response diet (10 g/kg) while assuring the minimum levels of all other essential amino acids in a manner that would meet or exceed recommendation. This experiment was carried out in a complete randomized design arrangement with 6 dietary Lys level; 10.0, 11.5, 13.0, 14.5, 16.0 and 17.5 g/kg, from 21 to 42 d of age. Each treatment was consisted of 5 floor pens as replicate with 50 quail chicks. As a result of this study increasing dietary Lys level, body weight, body weight gain and feed conversion ratio improved significantly (P<0.05). Feed intake, carcass weight, breast weight and yield, and thigh weight were significantly (P<0.05) affected by dietary Lys. The thigh yield at 42 d of age were not influenced significantly (P>0.05) by dietary Lys. Our results suggested that Lys requirement is 13.5 and 14.7 g/kg when feed conversion, body weight (BW) gain are considered and is at least 13.6 and 15.0 g/kg when and breast and thigh meat properties are taken into account in female Japanese quails at 21-42 d of age for second order polynomial and quadratic broken-line models respectively. These results indicated that the Lys requirement of 13.0 g/kg (total basis) is not adequate from 21 to 42 d of age for female Japanese quails. Future research, however, should determine lysine needs for immune system functions of quails reared in conventional environments or during an infectious challenge.

KEY WORDS lysine, performance, quail, requirement.

INTRODUCTION

Japanese quail (*Coturnix coturnix japonica*) belongs to the order *Galliformes* and the family *Phasianidae* (Karaalp, 2009). These birds are mainly produced for their eggs and meat, but are also used as laboratory animals. The preference for the Japanese quail production is justified by the fact that the quail has several advantages, such as rapid growth, early sexual maturity and a short generation interval (Kaur *et al.* 2006). Essential amino acid recommendations by NRC (1994) for Japanese quail are largely based on researches conducted at least 5-6 years before 1994's however; the meat production performance of Japanese quails has also been improved during recent years due to genetic selection. Therefore, there is need of updating optimal nutritional requirements of Japanese quails with the improvement in genetic makeup to exploit production potentiality (Kaur *et al.* 2008). Lysine (Lys) is the second limiting amino acid after methionine in a maize-soybean diet and furthermore, the ideal protein concept used in formulation of amino acid levels was based on Lys that developed at Illinois University in 1950 s to 1960 s. The Agricultural Research Council in the United Kingdom was the first to propose an ideal protein for pigs in which lysine was used as the reference amino acid. Several reports are available on lysine requirements in chicken and turkey, but information on the lysine requirement of Japanese quails is scanty (Kaur *et al.* 2006; NRC, 1994; Hajkhodadadi *et al.* 2013). Thus this study was conducted to determine the Lys requirement of female quail at 21 to 42 d of age based on performance variables. For this purpose, dietary Lys was supplemented in diet from deficient level to an excessive level for female Japanese quail.

Many different curves that relate animal response to feed composition can be fitted to data from animal feeding trials. The most realistic curve for each set of data is difficult to choose because of the variability in the responses of different individuals, or pens of individuals, fed on the same diet in broiler (Pesti *et al.* 2009) therefore in this research, different equations were used for determining Lys requirements of female Japanese quails.

MATERIALS AND METHODS

Birds

3000 one-day old growing Japanese quail chicks were provided from Tehran University hatchery and divided into 30 wood shaving floor pens (220 cm×200 cm) with 100 birds per pen and they were raised with common starter diet up to 21 d of age.

The common starter diet contained was formulated by UFFDA software based on NRC (1994) nutrient recommendation for Japanese quail and consisted 240 g/kg protein and 12.1 MJ ME/kg. During this period, water and feed were provided *ad libitum*.

The birds received 24 h of light/day during the 24 days of age (Hajkhodadadi *et al.* 2013). The temperature was kept at 35 ˚C from 1 to 3 d of age, and then it was reduced by 2.5 ˚C/week until the birds were 3 weeks old (Hajkhodadadi *et al.* 2013). At 21 day of age, 1500 female quail chicks were selected by phenotypic method and weighed individually and were randomly allotted to 30 floor pens with 50 female birds per pen to achieve the similar average weight of birds in each pen. There were 6 dietary treatments each containing 5 replicate pens (250 birds). All birds reared on wood-shaving floor pen (100 cm×150 cm) in temperature controlled house during 21-42 days of age. The temperature was 29 ˚C until the birds were 6 weeks old. Food and water were available *ad libitum* during the grower period.

Diets

Before starting the experiment, all protein-containing ingredients were analyzed for basal chemical composition (AOAC, 1995) and amino acid (Andrews and Baldar, 1985) content. Then, the experimental diets were formulated using these determined actual values. A dose-response diet mainly based on corn, corn gluten and soybean meal was used. Lys content was minimized in dose-response diet (10 g/kg) while assuring the minimum levels of all other essential amino acids in a manner that would meet or exceed NRC (1994) recommendations. The calculated content of AME in dose-response diet was 12.1 MJ/kg, and the analyzed concentrations of CP and lysine were 243 and 10.5 g/kg, respectively (Table 1).

The lysine level was gradually increased in 5 further isoenergetic and isonitrogenous diets by the stepwise inclusion of L-lysine.HCl at the expense of L-glutamine to achieve the following lysine concentrations; 10.0, 11.5, 13.0, 14.5, 16.0 and 17.5 g/kg (Table 1). L-lysine, DL-methionine and L-threonine were feed grade quality. All ingredients with the exception of the variable ones (L-glutamine, L-lysine and inert filler) were mixed as a single lot and then mix were divided into 6 parts. L-glutamine, L-lysine and inert filler were added separately in the respective proportions to each diet and mixed again. All chicks received feed from 21 to 42 of age in mash form.

Measurements

Performance parameters

At 42 d of age, body weight (BW), feed consumption from each pen was measured weekly then the average body weight gain (BWG) was calculated by initial body weight and final body weight for all replicate during the experiment. The mortality was recorded daily. Feed conversion ratio (FCR) was corrected for mortality and represents grams of feed consumed by birds in a pen divided by grams of weight gain (Hajkhodadadi *et al.* 2013).

At 42 of age, the quails were starved for 6 h, but drinking water was supplied then 4 quails selected from each pen were weighed individually before slaughter. The birds were removed from their pens and then transferred in boxes (4 birds in each box) to a dissection room. Quails were slaughtered by severing the jugular vein. The quails plucked mechanically after hot water scalding then carcasses were eviscerated by hand. All weights were individually recorded for the carcass, thigh and breast. The yield of different carcass specifications were expressed as a percentage relative to final live body weight (Fatufe *et al.* 2004).

Statistical analysis

Data in this experiment were evaluated by one-way ANOVA in a completely randomized design (CRD).

Table 1 Ingredient and calculated composition of experimental diets (g/kg, as fed basis)

Item	Graded level of lysine (g/kg)					
	10.0	11.5	13.0	14.5	16.0	17.5
Ingredient, g/kg						
Ground maize	523.4	523.4	523.4	523.4	523.4	523.4
Soybean meal (CP 44%)	262.4	262.4	262.4	262.4	262.4	262.4
Maize gluten meal (CP 61%)	151.6	151.6	151.6	151.6	151.6	151.6
Calcium carbonate	13.6	13.6	13.6	13.6	13.6	13.6
Dicalcium phosphate	8.8	8.8	8.8	8.8	8.8	8.8
Sodium chloride	1.6	1.6	1.6	1.6	1.6	1.6
Sodium bicarbonate	1.9	1.9	1.9	1.9	1.9	1.9
Vitamin premix[1]	2.5	2.5	2.5	2.5	2.5	2.5
Mineral mineral[2]	2.5	2.5	2.5	2.5	2.5	2.5
L- Lysine.HCl	0	1.9	3.9	5.9	8.0	10.0
Glutamine	7.5	6.0	4.5	3.0	1.5	0
L-threonine	1.7	1.7	1.7	1.7	1.7	1.7
DL-methionine	0.7	0.7	0.7	0.7	0.7	0.7
Inert filler[3]	21.8	21.4	20.9	20.4	19.8	19.3
Total	1000	1000	1000	1000	1000	1000
Calculated composition						
Apparent metabolizable energy, MJ/kg	12.1	12.1	12.1	12.1	12.1	12.1
Crude protein, g/kg	240	240	240	240	240	240
Lysine, g/kg	10.0	11.5	13.0	14.5	16.0	17.5
Methionine, g/kg	5.5	5.5	5.5	5.5	5.5	5.5
Methionine + cysteine, g/kg	8.2	8.2	8.2	8.2	8.2	8.2
Threonine, g/kg	11.2	11.2	11.2	11.2	11.2	11.2
Nonphytate P, g/kg	3.0	3.0	3.0	3.0	3.0	3.0
Calcium, g/kg	8.0	8.0	8.0	8.0	8.0	8.0
Sodium, g/kg	1.5	1.5	1.5	1.5	1.5	1.5
Analyzed composition						
Crude protein (CP), g/kg	240.2	240.0	245.0	241.0	243.0	240.0
Total lysine, g/kg	10.5	11.9	13.5	14.9	16.5	18.0

[1] Vitmin premix include per kg of diet: vitamin A: 1242 µg; vitamin E: 7.7 mg; vitamin B_{12}: 0.01 mg; Cholecalciferol (D_3): 0.2 mg; Menadione: 0.8 mg; Folic acid: 0.4 mg; Choline chloride: 170 mg; D-pantothenic acid: 12 mg; Riboflavin: 2.6 mg; niacin, 4 mg; Biotin: 0.2 mg; Thiamin: 0.7 mg; Pyridoxine: 2. mg and Butylated hydroxytoluene: 125 mg.

[2] Mineral premix supplied per kg of diet: Manganese: 16 mg; Zinc: 15 mg; Iron: 8 mg; Copper: 4 mg; Iodine: 1.6 mg; Selenium: 0.08 mg and Butylated hydroxy-toluene: 125 mg.

[3] Filler (sand) represents inert space in the diet to which L-lysine.HCl was added to derive the projected lysine level.

All data were examined for normality distribution for ANOVA then were analyzed by the GLM procedure of (SAS, 1996). Percentage data corresponding to mortality, carcass defects ratio were transformed to arcsine √% for analysis. Lysine effect ($P<0.05$) was separated using the Duncan multiple range test with α of 0.05 by SAS software (SAS, 1996).

Estimating requirements

Second order polynomial equation

A second order equation could fit the increase and decrease in performance of Japanese quail. For instance, for the second order polynomial equation (Pesti *et al*. 2009):

$$Y= a + bX + cX^2$$

Where:

Y: the dependent variable.

X: the dietary lysine concentration.

a, b and c: constants.

Lys need were expressed as 95% of dietary lysine level at maximum response.

Two-quadratic broken line equation

The most commonly applied model to nutritional response experiments is the broken line model with the quadratic ascending portion a function of dietary nutrient concentration. The quadratic-broken line nutritional response models are a subset of spline models where the slope of one line is equal to 0. Two Quadratic-broken line models assume that as concentrations of a nutrient are increased in the diet, there is a change in response up to some point, where the maximum (or minimum) response is reached There is some plateau level above the requirement where the nutrient is neither helpful, nor toxic, for the Two-quadratic broken line (Pesti *et al*. 2009):

$$Y= L + U \times (R-X)^2 + V \times (X-R)$$

Where:

Y: the dependent variable.
X: the dietary Lys concentration.
L: the ordinate.
R: the abscissa of the breakpoint.
L, U and V: constants and the value R- is zero at values of x > R (Pesti *et al.* 2009).

RESULTS AND DISCUSSION

Calculated and analyzed dietary Lys levels were in close agreement (Table 1). Body weight of female quails at initiation of experimentation was analyzed, there are no significant difference (P>0.05). Dietary Lysine was significantly affected FCR during the experiment so when dietary Lysine was in lowest level (10 gr/kg), feed conversion ratio (FCR) was in high amount (7.05) this may as a result of reducing in feed intake by means of unbalancing of amino acids in its diets. With increasing Lysine level to 1.45%, FCR was reached to optimum that was significantly differ (P<0.05) from other levels (Table 2). Mortality was in the normal range for the period between 21 and 42 d of age and was not affected by dietary Lys (P>0.05) (Table 2). In our study, supplementation of dietary Lys affected , BW and BW gain of female Japanese quail significantly at 21-42 d of age (P<0.05) (Table 2).

Dietary Lys supplementation affected breast, yield of female Japanese quails significantly (P<0.05) but not carcass and thigh yields (Table 3).

Lys requirement

BW and BW gain increased to peak and then decreased as dietary Lys increased, and this response provided a significant fit to a second order polynomial equation. According to this model, the lysine requirement for obtaining maximum BW and BW gain of female Japanese quails from 21 to 42 d of age were 13.3 g/kg respectively (Table 4). Gradient additions of dietary Lys improved FCR significantly (P<0.05) (Table 2). The Lys requirement for optimized FCR was 13.7 g/kg with second order polynomial equations (Table 4). Although dietary Lys affected feed intake significantly (P<0.05), but second order polynomial and two-quadratic broken line models did not fit to these data for determining dietary Lys requirement.

The two-quadratic broken line model estimated Lys requirement as 14.9 for BW and BW gain, and 14.5 g/kg for feed conversion ratio, at 21-42 d of age in female Japanese quails (Table 5).

Lys requirement, according to second order polynomial equation were 13.4 for carcass, and 13.5 g/kg for, breast and thigh weights. The second order polynomial equation not fitted (P>0.05) on carcass, breast and thigh yields significantly (Table 6).

With two-quadratic broken line model Lys requirement, for carcass, breast and thigh weight at 42 d of age, were 14.3, 15.1 and 15.4 g/kg, respectively (Table 7). In this study data of carcass, breast and thigh yields cannot use as a response to a Lys need determination by two quadratic broken-line model (Table 7).

The NRC (1994) recommended Lys requirement for Japanese quail was based on experiments that conducted at least 5-6 years before 1994 so after decade, in line with expanding quail production , it is obligatory for reevaluating the Lys requirement. There are different methods for determining the requirement of nutrients, but most commonly models were the second order polynomial and two-quadratic broken line (Robbins *et al.* 2006; Pesti *et al.* 2009).

With female Japanese quail, dietary Lys varied with the model used to estimate the requirement. In both second order polynomial, and two-quadratic broken line equations when Lys requirement was determined based on carcass traits as response, Lys requirement was higher than the Lys requirement estimated based on BW or BW gain. Our findings are similar with many studies and have led to the assumption that breast weight of broilers is improved when Lys is supplemented in the diets (Berri *et al.* 2008; Dozier *et al.* 2008; Ng'ambi *et al.* 2009). The finding is agree with so many study (Corzo *et al.* 2002; Coleman *et al.* 2003; Fatufe *et al.* 2004; Wang *et al.* 2006; Dozier *et al.* 2008; Kaur *et al.* 2008; Ayaşan and Okan, 2010) that conclude nutrient need for optimum breast and thigh weight is higher than weight gain as responses to requirement determination in broiler or turkey in different strain and sex based on different regression models.

Furthermore NRC (1994) recommendations in Japanese quail was generally for growing quail male and female ones during the 0-42 d of age, so most producers need to use the requirement of female or male separately for raising Japanese quail . In contrast to our finding, in the present research, total dietary Lys requirement has been estimated as 13.6 g/kg (13.3-13.9) using second order polynomial equation for female Japanese quail from 21 to 42 d of age, which is slightly higher from NRC (1994) dietary Lys requirement (13.0 g/kg) for Japanese quails but With two-quadratic broken line equation Lys requirement, was determined as 15.0 g/kg (14.3-15.9) based on performance and carcass constitutes traits, and this value is more than NRC (1994) recommendation for Japanese quail. More Lysine requirement in both estimation models may occurred because of this reason that NRC (1994) recommendation for Lys estimation was based on performance traits only, so carcass specifications of birds were not considered. In the other hands Lys requirement is increased when carcass traits of birds are included (Ayasan and Okan, 2010).

Table 2 Performance of female Japanese quail from 21 to 42 d of age in response to dietary lysine[1]

Lys, g/kg	Body weight (g/bird)		Daily consumption		Feed conversion ratio (g/g)	Mortality (%)
	Final	Daily Gain	Feed (g/bird)	Lys (mg/bird)		
10.0	258.75[c]	82.52[c]	27.52[a]	239.40[d]	7.05[a]	2.13
11.5	297.63[ab]	123.75[a]	23.90[c]	254.60[dc]	4.09[c]	1.61
13.0	295.98[ab]	123.92[a]	25.12[bc]	273.71[bc]	4.27[c]	1.94
14.5	314.43[a]	130.03[a]	24.38[c]	283.60[b]	3.99[c]	1.49
16.0	297.85[ab]	113.96[ab]	27.16[ab]	325.40[a]	5.00[b]	1.24
17.5	275.13[bc]	98.24[bc]	21.84[d]	335.70[a]	4.70[bc]	1.62
SEM	3.76	3.54	1.17	4.3	0.670	0.762
Contrast						
Lys	0.0005	0.0003	0.0002	< 0.0001	< 0.0001	0.3029

The means within the same row with at least one common letter, do not have significant difference (P>0.05).
SEM: standard error of the means.

Table 3 Carcass constituents of female Japanese quail at 42 d of age

Lys, g/kg	Carcass	Breast	Thigh
	Yield (g/100 g LW[1])	Yield (g/100 g LW)	Yield (g/100 g LW)
10.0	69.39	27.56[ab]	14.72
11.5	69.65	26.68[b]	14.21
13.0	68.24	29.05[a]	15.34
14.5	65.95	28.64[ab]	14.71
16.0	68.07	28.46[ab]	15.32
17.5	69.16	27.98[ab]	14.84
SEM	1.740	1.161	1.095
Contrast			
Lys	0.569	0.0231	0.8085

LW: live weight.
SEM: standard error of the means.

Table 4 Lys requirement of female quail based on second order equations[1]

Response criterion	Equation[2]	R^2	CV	P-value	Requirement[3]
Final body weight (g)	$Y= -278.85 + 838.15 \times (Lys) - 298.57 \times (Lys)^2$	70.78	5.05	< 0.0001	13.3
Body weight gain (g)	$Y= -409.05 + 775.14 \times (Lys) - 278.63 \times (Lys)^2$	70.47	11.93	< 0.0001	13.3
Feed consumption (g)	$Y= 80.13 + 4.075 \times (Lys) - 5.50 \times (Lys)^2$	-	-	0.117	-
Feed conversion (g/g)	$Y= 29.06 - 34.91 \times (Lys) + 12.14 \times (Lys)^2$	68.23	29.06	0.0001	13.7

[1] R^2= multiple coefficient of determination × 100.
CV: coefficient of variation.
[2] Second order polynomial based on formulated dietary Lys.
[3] Dietary Lys requirement estimates 95% of asymptote.

Table 5 Lys requirement in female Japanese quail based on two-quadratic broken line equations for performance parameters[1]

Response criterion	Equation[2]	R^2	P-value	Requirement[3]
Final body weight (g)	$Y= 312.7 - 201.7 \times (R-X)^2 - 157.2 \times (X-R)$	61.27	0.0002	14.9
Body weight gain (g)	$Y= 133.7 - 323.1 \times (R-X)^2 - 96.72 \times (X-R)$	62.90	0.0001	14.9
Feed consumption (g)	Not met			
Feed conversion ratio (g/g)	$Y= 3.98 + 81.51 \times (R-X)^2 + 1.50 \times (X-R)$	81.61	< 0.0001	14.5

[1] R^2= multiple coefficient of determination × 100.
[2] The quadratic broken-line model is:

$y= L + U \times (R-x) \times (R-x) + V (R-X)$

Where:
L: the ordinate.
R: the abscissa of the breakpoint.
R: zero at values of x > R.

[3] R: Lys requirement.

Table 6 Lys requirement in female Japanese quail based on second order equations[1]

Response criterion	Equation[2]	R^2	CV	P-value	Requirement[3]
Carcass weight (g)	Y= -83.67 + 414.13 × (Lys) - 147.17 × $(Lys)^2$	67.67	5.98	0.007	13.4
Carcass yield (g/g BW)	Y= 102.19 - 49.44 × (Lys) + 17.43 × $(Lys)^2$	-	-	0.228	-
Breast weight (g)	Y= -115.49 + 286.84 × (Lys) - 100.96 × $(Lys)^2$	74.5	6.38	< 0.0001	13.5
Breast yield (g/g BW)	Y= 14.28 + 19.32 × (Lys) - 6.54 × $(Lys)^2$	-	-	0.237	-
Thigh weight (g)	Y= -49.61 + 134.51 × (Lys) - 47.21 × $(Lys)^2$	70.10	7.21	0.0007	13.5
Thigh yield (g/g BW)	Y= 11.39 + 4.55 × (Lys) - 1.44 × $(Lys)^2$	-	-	0.7811	-

[1] R^2= multiple coefficient of determination × 100.
CV: coefficient of variation.
[2] Second order polynomial based on formulated dietary Lys.
[3] Dietary Lys requirement estimates 95% of asymptote.

Table 7 Lys requirement in female Japanese quail based on two-quadratic broken line equations for carcass constitutes at 42 d of age[1]

Response criterion	Equation[2]	R^2	P-value	Requirement[3]
Carcass weight (g)	Y= 208.9 - 139.3 × $(R-X)^2$ - 52.77 × (X-R)	65.93	0.027	14.3
Carcass yield (g/g BW)	Y= 66.86 + 12.70 × $(R-X)^2$ + 9.87 × (X-R)	14.50	0.362	-
Breast weight (g)	Y= 89.57 - 70.08 × $(R-X)^2$ - 54.52 × (X-R)	73.07	0.0001	15.1
Breast yield (g/g BW)	Y= 28.68 - 5.02 × $(R-X)^2$ - 3.81 × (X-R)	13.45	0.3978	-
Thigh weight (g)	Y= 46.80 - 29.01 × $(R-X)^2$ - 29.75 × (X-R)	70.85	0.002	15.4
Thigh yield (g/g BW)	Not met			

[1] R^2= multiple coefficient of determination × 100.
[2] The quadratic broken-line model is:

y= L + U × (R-x) × (R-x) + V (R-X)

Where:
L: the ordinate.
R: the abscissa of the breakpoint.
R: zero at values of x > R.

[3] R: Lys requirement.

In the present study, second order polynomial and quadratic broken-line were performed to determine the exact Lys requirement.

Our data suggested that Lysine requirement based on second order polynomial and quadratic broken-line models respectively, to be as 13.4, 14.7 g/kg when feed conversion ratio, BW gain are considered and may be at least 13.6, 15.0 g/kg when breast and thigh meat properties variables are taken into account in female Japanese quail at 21-42 d of age.

The two-quadratic broken line model may have overestimated the requirement for female quails when compared with second order polynomial equations, this result was agree with (Biswas *et al*. 2006; Dozier *et al*. 2008; Ayasan and Okan 2010) who concluded the same results in male broiler at 49-63 d of age.

CONCLUSION

These results indicated that the NRC (1994) Lys requirement of 13.0 g/kg (total basis) is not adequate from 21 to 42 d of age for female Japanese quails. Future research, however, should determine lysine needs for immune system functions of quails reared in conventional environments or during an infectious challenge.

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