



Determination of Crude Protein and Metabolisable Energy of Japanese Quail (*Coturnix coturnix japonica*) during Laying Period

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ABSTRACT

This study was carried out to determine the energy and protein requirements of laying Japanese quails. A completely randomized design of treatments comprising four dietary protein levels (18, 20, 22 and 24%) and three levels of metabolisable energy (3000, 3100, 3200 kcal/kg) in a 4 × 3 factorial arrangement was used. 144 Japanese quails aged 7 weeks were randomly divided into 12 dietary treatments with 3 replicates per treatment and each replicate with 4 birds. The experiment lasted for five weeks. The results of the study showed that there was no significant ($P > 0.05$) effect of protein, energy or their interaction on feed intake, feed conversion ratio, hen day production, egg weight and egg number. However, protein as a single variable had a significant effect ($P < 0.05$) on feed intake. There was also no significant ($P > 0.05$) effect of protein, energy or their interaction on egg quality traits (yolk colour, yolk weight, albumen weight, shape index, shell thickness, shell weight, and haugh unit). However, birds fed 20% crude protein and 3000 kcal/kg metabolisable energy had better hen day production, number of eggs per bird and egg quality traits compared with birds on the other groups. Dietary protein increased egg production and egg weight, augmented by energy. The yolk colour was increased with increasing energy level. Therefore, the results of the experiment revealed that 20% crude protein and 3000 kcal/kg metabolisable energy could be used to obtain the best production performance and good egg quality traits of Japanese quails at the laying phase.

Key words: Japanese quail, Production performance, Egg quality traits, Metabolisable energy, Protein.

INTRODUCTION

One of the major sectors of the livestock industry in Nigeria is poultry production. This sector has hitherto been dominated by the rearing of chickens. However in recent years, Japanese quail has gained worldwide importance as a laboratory animal (Baumgartner, 1994) and productive bird because of its advantageous attributes such as small body size, rapid growth rate, and early sexual maturity (Siyadati, 2011) in 5 to 6 weeks of age, high rate of reproduction, ability to produce 3 to 4 generations in a year, relative ease of maintaining the colony (Shim and Vohra, 1984), lesser space and feed requirements compared with the domestic chickens, cheaper cost of production, hardiness and ability to thrive in small cages. Therefore they are suited for commercial rearing, egg and meat production under intensive management.

Despite these attributes, the major constraint of quail production in Nigeria is the continually review of the nutritional requirements of Japanese quail for production

over time by Beane and Howes (1966), Vohra (1971) and NRC (1994), the non availability of economical and efficient rations (Barque et al., 1994), poor documentation of energy and protein requirements, the efficiency of feed utilization for quails (Monica et al., 2010) and lower productivity of quail fed based on nutritional requirements data obtained in other countries with different climatic conditions (Soares et al., 2003). However Alaganawy et al. (2014) reported that adequate amino acid balance is the most important nutrient for Japanese quails. Other research reported energy and crude protein levels of 2900 kcal ME/kg and 22%, respectively (Reda et al., 2015) and levels of 3000 kcal ME/kg and 26% respectively (Jahanian and Edriss 2015).

For optimum productivity of Japanese quail in Nigeria, the energy and crude protein requirements are important. The objective of this study was to determine the energy and protein requirement of Japanese quail birds in the laying period and also investigate the effects of

different dietary levels of protein and energy on egg production.

MATERIALS AND METHODS

Experimental diets and management of birds

The experiment was carried out at the poultry unit of the teaching and research farm, university of Ibadan, Ibadan, Nigeria. The experiment lasted for five weeks between August and September 2014. Birds were acclimatized to the experimental diets by the first week, while the remaining 4 weeks were used for egg quality data collection, feed intake data collection, hen day production and for measurement of internal egg quality parameters. The experimental diets as shown on table 1, were corn-soybean based with four dietary protein levels (18, 20, 22 and 24%) and three levels of metabolisable energy (3000, 3100, 3200 kcal/kg). A total number of 144 Japanese quails aged 7 weeks were used for the experiment. The birds were housed in a 36 compartments cage with 4 birds in each compartment. These contained the twelve experimental units with three replicates each. The birds were allocated to 12 diets in a completely randomized design. The hens received the diets from 7 weeks of age till 12 weeks of age and were provided water *ad libitum*.

Data and sample collection

Performance parameters (feed intake, egg production, egg weight, and feed conversion ratio) were calculated during the course of the trial. To determine the cholesterol and fatty acid profile of egg yolk, three eggs were randomly sampled at week six of the experiment from each treatment respectively. Egg quality parameters (yolk weight, yolk index, albumin weight, haugh unit, shell weight, and shell thickness) were measured at week six, using five eggs from each treatment.

Performance evaluation

Daily egg production per replicate was recorded and number of eggs per hen per week was calculated. Eggs laid per replicate were weighed daily and average weight for that particular week was calculated. The data thus generated (egg production and egg weight) was used to calculate egg mass/bird/week (weekly egg no. in replicate x average egg weight). Weekly feed intake was determined (total feed offered during a week - Feed refused at the end of week). Data on feed intake and egg mass were used to calculate feed conversion (feed intake/egg mass; g/g).

Egg quality evaluation

External qualities: Egg weight was measured using Mettler top-loading weigh balance. The length and width (cm) of each egg was measured using Vernier caliper. The width was measured as the distance between two ends of the egg at the widest cross sectional region using Vernier caliper. The length was measured as the distance between the broad and narrow ends of the eggs.

Egg shape index (ESI) was calculated as the percentage of the egg breadth (width) to the egg length (Panda, 1996). The formula that was used is as follows:

$$\text{Egg shape index} = \frac{\text{Width of egg (mm)} \times 100}{\text{Length of egg (mm)}}$$

The thickness of individual air-dried shells is measured to the nearest 0.01mm using micrometer screw gauge (Chowdhury, 1987). Eggshells were air-dried in the crates. The relative shell weight was calculated by relating the shell weight to the weight of the egg. Shell thickness was measured using a micrometer gauge (in mm).

Internal qualities: Yolk height, yolk width and yolk diameter (cm) were measured using a Vernier caliper. **Albumen height:** The egg was gently broken and the maximum albumen height was measured with tripod micrometre (Doyon *et al.*, 1986) Albumen weight is the difference between the egg weight and the sum of weight of yolk and dry eggshell expressed as a percentage of the whole egg. Percentage of Albumen weight was calculated as the percentage of the albumen weight to other egg weight. Yolk weight was measured using Mettler top-loading weighing balance. Percentage Yolk weight was calculated as the percentage of the yolk weight to the egg weight.

Yolk index was estimated from ratio of yolk height to yolk width. Visual yolk colour was determined with a yolk colour fan (scale 1 to 15).

Haugh unit (HU) is a relationship between egg weight and height of thick albumen surrounding yolk. This was calculated using the values obtained from the egg weight and albumen height as expressed by Haugh (1937) in the formula shown below:

$$\text{HU} = 100 \log [H + 7.57 - 1.7 W^{0.37}]$$

Where, H = Albumen Height (mm) and W = Weight of the egg (g).

Data collection

Data of feed offered and body weight were recorded weekly and used to calculate feed intake, weight gain, feed conversion ratio and protein efficiency ratio.

Statistical analysis

Data were subjected to analysis of variance (ANOVA) using SAS statistical package (SAS, 2003) as a

4×3 factorial arrangement in a completely randomized design. Significant means were separated using Duncan multiple range test at $P < 0.05$.

Table 1. Composition of experimental diet fed to Japanese quails during laying for the determination of crude protein and metabolisable energy

Ingredients, g/kg	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6	Diet 7	Diet 8	Diet 9	Diet 10	Diet 11	Diet 12
Corn	654.7	665.7	657.7	635.7	635.7	623.7	578.7	578.7	558.7	513.7	508.7	488.7
Soybean meal	262.0	262.0	262.0	300.0	300.0	300.0	352.0	352.0	352.0	410.0	410.0	410.0
Soybean oil	12.0	23.0	39.0	8.0	19.0	35.0	18.0	28.0	48.0	25.0	40.0	60.0
Wheat bran	30.0	8.0	0	15.0	4.0	0	10.0	0	0	10.0	0	0
Dicalcium phosphate	17.5	17.5	17.5	17.5	17.5	17.5	17.5	17.5	17.5	17.5	17.5	17.5
Limestone (38% Ca)	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0
Salt	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0
Vitamin-mineral premix	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0
DL-Methionine	3.8	3.8	3.8	3.8	3.8	3.8	3.8	3.8	3.8	3.8	3.8	3.8
L-Lysine.HCl	2.9	2.9	2.9	2.9	2.9	2.9	2.9	2.9	2.9	2.9	2.9	2.9
Threonine	1.1	1.1	1.1	1.1	1.1	1.1	1.1	1.1	1.1	1.1	1.1	1.1
Total	1000	1000	1000	1000	1000	1000	1000	1000	1000	1000	1000	1000
Nutrient content												
Protein, g/kg	185.6	183.0	181.0	201.9	200.2	198.5	221.5	219.9	218.2	242.5	242.1	240.1
ME, kcal/kg	3007.5	3109.5	3205.7	3024.6	3102.7	3190.2	3031.2	3102.2	3200.8	2992.9	3101.6	3200.2

RESULTS AND DISCUSSION

Table 2 shows the overall effects of protein, energy and their interaction on production performance of laying Japanese quails. The result obtained shows that there were no significant effects ($P > 0.05$) of protein, energy or their interaction on the performance of laying quails, except the feed intake, that was significantly affected ($P < 0.05$) by protein.

Feed intake

Different levels of dietary energy did not affect performance parameters significantly ($P > 0.05$). For different levels of protein, there was a significant difference ($P < 0.05$) in feed intake. 20% and 22% crude protein (CP) inclusion levels differ significantly from 18% and 24% inclusion level. However, 20% inclusion level had the highest value of feed intake.

Feed conversion ratio

The FCR was not significantly affected by different dietary levels of protein as shown in table 3. However, inclusions at 20% CP led to the highest value of FCR while inclusion at 18% had the lowest value. It shows that

inclusion at 18% had the best FCR. Also different levels of energy did not affect FCR significantly, although birds fed with the dietary energy of 3,000 kcal/kg had the lowest and the best value of FCR. This result agreed with the findings of Jahanian and Edriss (2015) which reported 26% Crude Protein and energy levels of 3000 kcal ME/kg as adequate for Japanese quails.

The combination of 24% CP and 3200kcal had the highest value and significantly different ($P < 0.05$) from 18% CP and 3000, 3200kcal/kg metabolisable energy (ME) and 24% CP and 3000, 3200 kcal ME, but significantly different from other combinations. However, the combination of 24% CP and 3100kcal/kg ME had the lowest and the best value of FCR.

Hen-day production

Protein as a variable did not affect hen-day production (HDP) at different inclusion levels. Also different energy levels did not significantly affect hen day production. However, birds fed with 3000 inclusion level had the highest value while birds on 3200kcal inclusion level had the lowest value.

The combined effect of 20% CP and 3000kcal had the highest value of hen-day production and significantly

different ($P < 0.05$) from inclusion level at 18% CP and 3100kcal/kg ME but not significantly different from other combinations.

Number of eggs/bird

Both energy and protein at different levels of inclusion had no significant ($P > 0.05$) difference on the number of eggs laid per bird respectively. The combination of 20% CP and 3000kcal ME had the highest number of egg/bird and is significantly different from the combination of 18% CP and 3100kcal ME. However, the combined effect of 20% CP and 3000 ME did not differ significantly from other combinations though it has the highest value.

Average egg weight

24% CP had a significant difference from other levels of CP on the average weight of egg though the weight are similar in value with difference not greater than 0.22. The effects of different levels of energy on the average weight of egg were not significantly different. The combined effects of 18% and 24% CP each with 3000kcal ME is significantly different from the combined effects of groups that had 3100 and 3200 kcal ME each with 18% CP and 3000 and 3100 kcal ME each with 20% CP and 22% CP with 3000 kcal ME.

The results shown in table 3 indicated that there were no significant effects ($P > 0.05$) of protein, energy or their interaction on the performance of laying quails, except feed intake, that was significantly affected ($P < 0.05$) by protein. However, the mean effect of protein and energy on feed intake of experimental quail birds fed on 20% and 3000kcal had the highest feed intake, 34.28g/bird/day, while quails on 18% CP and 3200kcal had the lowest feed intake. These results agree with Tuleun et al. (2013) who reported a similar result.

Feed conversion was lowest in birds fed with 24% CP and 3000 kcal, though it was not significantly different ($P > 0.005$) from birds fed with 22%, 20% and 18% CP. This support the findings of Murakami et al. (1993) who reported that dietary protein had no significant influence on feed efficiency when laying Japanese quails were fed on diets with different protein levels. The groups fed with 3000 kcal ME had a better feed conversion ratio. This shows that the dietary energy level at 3000kcal helped improved production, directing the use of crude protein in the diet, instead of energy generation.

Hen day production was higher in quails fed 18% and 20% CP and 3000 kcal/kg ME. This result agrees with the result of Khosro et al. (2011) who reported higher egg production in birds fed 20% CP but a lower energy level

(2950kcal/kg). They are also similar to the 20% CP suggested by NRC (1994) and Garcia et al. (2005) with 2,850 and 2,950kcal/kg ME respectively. The lowest hen day production was obtained in quails fed 24% CP and 3200kcal/kg ME. This also is in line with the results of Khosro et al. (2011). Egg production is costly in terms of energy and protein. The required energy for egg formation may be derived from daily feed intake or from the body reserve. Daily feed intake is a more important source of nutrient for small birds like quail than body reserve. If energy or protein is limiting, birds can compensate by reducing egg size or the number of eggs laid or by increasing the laying interval and spreading the loss of egg formation over a longer period (Brand et al., 2003).

Egg number per bird was higher in birds fed 20% CP and 3000kcal/kg ME, with the value, 6.06g compared with 5.17g from birds fed on 18% CP and 3200kcal/kg ME.

Average egg weight was higher in birds fed 24% CP and 3000 kcal/kg ME. This result is similar to the findings of Garcia *et al.* (2005) who reported a quadratic effect of protein level on the produced egg weight and maximum egg production with 23.1% CP in the diet. However, Khosro *et al.* (2011) reported a higher egg mass with 20% CP and 2,900 kcal ME in the diet. The result of this study shows that egg size and weight depend greatly on daily crude protein intake augmented by adequate dietary energy (3000 kcal/kg) since layers do not store a large amount of protein.

Egg quality traits

Table 4 shows the overall effects of protein, energy and their interaction on egg quality traits of laying Japanese quails. The result obtained shows that there were no significant effects ($P > 0.05$) of protein, energy or their interaction on egg quality traits of laying birds

Table 5 shows the mean effects of protein and energy on egg quality traits. Data obtained indicated that there were no significant ($P > 0.05$) differences among the treatment groups for most of the egg quality traits except haugh unit and shell weight for protein effect and yolk weight for energy effect.

Egg weight, shape index, shell thickness and shell weight

The combination of 22% CP and 3000kcal ME has the highest value and is significantly different ($P < 0.05$) from all other combinations of different protein and energy levels. The combination of 18% CP and 3000 kcal ME had the highest significant value but not significantly different from other combinations except the combinations of protein and energy levels of 18%, 24% CP and 3200,

3000kcal ME respectively. The combination of 18% CP and 3200kcal ME is significantly different ($P < 0.05$) from the combination of 20% CP and 3100kcal ME and the combinations of 22% CP with the three levels of energy. The combination of 24% CP and 3200kcal ME had the highest value but not significantly different from other combinations except for the combinations of 22% CP and 3000kcal ME and 20% CP and 3200kcal ME, which are in turn not significantly different from other combinations.

Yolk colour, yolk weight and albumen weight

The combined effect of 20% CP and 3200 kcal ME was significantly different. However, this was not

significantly different from the combination of 22, 24, 24% CP and 3000, 3100, 3200kcal ME respectively, which in turn were not significantly different from other combinations. The combination of 18% CP and 3200kcal ME had the highest yolk weight. Statistically, it is significantly ($P < 0.05$) different from the combination of 22% CP and 3000kcal ME, but not significantly different from other combinations except the combinations of protein and energy. There is no significant effect of different combinations of energy and protein on albumen weight across the treatments. However, the combination of 20% CP and 3000kcal ME had the highest value of albumen.

Table 2. Effect of protein, energy and their interaction on performance of laying Japanese quails at 12 weeks of age

Parameters	Factors		
	Protein	Energy	Protein×Energy
Feed intake (g)	0.0094	0.6801	0.7294
Feed conversion ration (g/g)	0.8980	0.2836	0.2642
Hen day production (%)	0.7333	0.2179	0.7548
Egg/bird	0.6784	0.3104	0.7780
Egg weight (g)	0.1012	0.9619	0.0848

Table 3. The effects of protein and energy on production performance of laying Japanese quails at 12 weeks of age

Protein level (%)	Parameters					
	HDP (%)	Egg/bird	AEW (g)	FI (g)	FI/bird	FCR
18	81.05	5.43	10.77 ^b	867.36 ^b	30.98 ^b	3.80
20	80.56	5.69	10.76 ^b	943.14 ^a	33.68 ^a	3.96
22	78.67	5.61	10.88 ^{ab}	941.36 ^a	33.62 ^a	3.95
24	77.78	5.50	10.99 ^a	884.83 ^b	31.60 ^b	3.89
SEM	2.36	0.16	0.07	19.47	0.69	0.17
Energy level (kcal/kg)						
3000	82.29	5.73	10.84	909.60	32.49	3.72
3100	78.94	5.52	10.84	919.44	32.84	3.92
3200	77.31	5.43	10.86	898.48	32.09	4.05
SEM	2.05	0.14	0.06	16.86	0.61	0.14

*Means with the different superscript on the same column are significantly different ($P < 0.05$); HDP: hen day production, AEW: average egg weight, FI: feed intake, FCR: feed conversion ratio. SEM: Standard error of mean

Egg quality traits

Egg yolk and albumen weight: There was a significant effect ($P < 0.05$) of protein and energy on egg yolk weight (g) index (%). Improved yolk weight and index were obtained from birds fed CP levels of 24% and 3200kcal/kg ME and 18% CP and 3200kcal/kg ME, with values 4.04 ± 0.32 and 3.84 ± 0.26 ($P > 0.05$) respectively. Garcia et al. (2005) reported that protein levels had an effect on yolk percentage, which is consistent with the current study. Also the result of this study is in line with Khosro et al. (2011) who reported similar result.

For albumen weight, there were no significant differences ($P > 0.05$) with the levels of energy and protein across the group. However, the three energy levels with 20% CP had the highest value of albumen weight.

Yolk color: Dietary protein significantly affected yolk color ($P < 0.05$). These results suggest a relationship between dietary protein and egg yolk color. Increasing dietary energy increased egg yolk color, this is because of an increased in the inclusion of corn in the diet. This result is in agreement with the report of Khosro et al. (2011). It is known that egg yolk color is a result of dietary

carotenoids transferred to the egg yolk, and mostly the color is from xanthophylls, and partly carotene and cryptoxanthin. Yellow corn, corn gluten meal, etc., are dietary sources of xanthophylls. Corn gluten meal contains 5-8 times the xanthophylls of yellow corn. Therefore, in this study, it is presumed that corn intake contributed greatly to the lightening of the egg yolk color at 20% CP.

Haugh unit: The combination of 20% CP and 3200kcal/kg ME had the highest statistically significant value (88.96), with similar values to other levels of 20% CP, which are 88.59 and 87.44 combined with 3000, 3100 kcal/kg ME respectively. Haugh unit is used to determine the freshness and protein content of egg. Higher haugh unit value denotes better quality of the egg (fresher, higher quality eggs have thicker whites). The result obtained in this experiment shows that inclusion of 20% CP with any of the three levels of energy is suitable for good quality eggs and will be better in storage of egg.

Shell weight: Eggshell was significantly ($P < 0.05$) affected by different levels of protein. Birds fed 18% CP at 3200 kcal/kg ME showed higher egg shell weight than other birds. Yakout et al. (2000) suggested that eggshell percentage may be reduced by higher lysine levels. In chickens, Gardner and Young (1972) reported that increasing the dietary protein level from 12 to 18% produced a significant increase in the relative proportion of egg yolk, and a subsequent significant decrease in the proportion of eggshell. However, when comparisons were made among dietary protein levels from 9.3 to 20.5% (Fisher, 1969) and from 14 to 20% (Yamagami and Kobayashi, 1983), no significant differences were found in egg composition. The findings in present study are in consonance with the report of Fisher (1969) and Yamagami and Kobayashi (1983) in chicken and also it is in agree with Khosro et al. (2011) in quails.

Table 4. Mean effect of protein, energy and their interaction on egg quality of laying Japanese quails at 12 weeks of age

Parameters	P value		
	Protein	Energy	Protein×Energy
Egg weight (g)	0.4879	0.4353	0.3372
Yolk colour	0.7759	0.411	0.055
Yolk weight(g)	0.4212	0.3876	0.4522
Albumen weight (g)	0.2952	0.623	0.9615
Shape index (%)	0.3828	0.6552	0.3278
Shell thickness (mm)	0.421	0.3671	0.4368
Shell weight (g)	0.1959	0.2238	0.2237
Haugh unit	0.1034	0.2866	0.626

Means in the same column with different superscripts are significantly different ($P < 0.05$). Egg wt- Egg weight, ST- Shell thickness, HU- Haugh unit, Yolk wt- Yolk weight; Yolk wt- Yolk weight, Alb.wt- Albumen weight

Table 5. Effects of protein and energy on egg quality traits of laying Japanese quails at 12 weeks of age

Protein level (%)	Parameters						
	Egg weight (g)	Shape Index (%)	Shell thickness (mm)	Yolk weight (g)	Yolk colour	Albumen weight (g)	Haugh unit
18	11.8 ^{ab}	77.73 ^a	0.40 ^a	3.66 ^a	1.15 ^a	6.85 ^a	85.57 ^b
20	11.11 ^b	78.79 ^a	0.30 ^a	3.49 ^a	1.19 ^a	7.25 ^a	88.33 ^a
24	12.06 ^a	77.19 ^a	0.40 ^a	3.65 ^a	1.19 ^a	6.98 ^a	86.17 ^{ab}
SEM*	0.27	0.68	0.03	0.17	0.05	0.17	0.88
Energy							
3000	11.38 ^a	78.40 ^a	0.30 ^a	3.30 ^b	1.12 ^a	7.09 ^a	87.06 ^a
3100	11.50 ^a	77.83 ^a	0.30 ^a	3.52 ^{ab}	1.14 ^a	6.97 ^a	85.48 ^a
3200	11.67 ^a	77.68 ^a	0.30 ^a	3.75 ^a	1.19 ^a	6.87 ^a	86.85 ^a
SEM	0.21	0.58	0.20	0.14	0.15	0.15	0.77

*SEM: Standard error of mean

CONCLUSION

This study was done to know the best energy and protein level suitable for production of laying Japanese quail. From the results obtained in this research, it was observed that neither protein nor energy had a significant effect on both performance and egg quality traits of laying Japanese quails. However, it was observed that quails fed 20% CP with 3000 kcal/kg metabolisable energy had a better production performance and egg characteristics apart from yolk index and yolk colour than birds on other dietary treatments.

Thus, it can therefore be concluded that laying Japanese quail, *Coturnix coturnix japonica* requires 20% CP with 3000kcal/kg ME for optimum egg production and good egg quality characteristics in Nigeria.

Competing Interests

Authors have declared that there is no competing interest.

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