Effect of dietary protein level on growth performance, carcass characteristics, intestinal microflora, hematological Parameters and some biochemical changes of growing Japanese quail

Osama A. Saker., * Gmal ., I Mazyad., ** Adel M. El-Gamal** and Mohamed M Elseify. **

* Department of biochemistry, nutritional deficiency diseases and toxicology Animal Health Research Institute Kafr El-Sheikh.&** Department of microbiology animal health research institute Kafr El Sheikh

Abstract

A total of one hundred and twenty 1-day-old apparently healthy Japanese quail were selected and randomly divided into 4 groups. Treatments were designed with four levels of crude protein (22.17, 24.2, 26.2 and 28%) respectively to study the effects of dietary crude protein level on growth performance, carcass characteristics, intestinal microflora, hematological parameters and some biochemical changes of growing Japanese quail. Results showed that growth performances parameters significantly improved as dietary crude protein level increased .The results indicated that dietary protein level beyond 24% has no beneficial effect on growth. Carcass traits results revealed that dressing percentage, breast muscles and leg muscles were increased significantly as crude protein level increased in the diet but weights of liver, heart gizzard and abdominal fat percentage were decreased significantly. Intestinal microflora count revealed that total aerobic bacteria, Coliform bacteria, Lactic acid bacterium and *Escherichia coli* increased significantly as crude protein level increased. Hematological examinations results demonstrated that hemoglobin concentration, red blood cells count, total leucocytic count, monocytes and lymphocytes were increased significantly as crude protein increased but basophils, eosinophils and heterophils were showed that total protein, albumin and decreased significantly. Serum analysis globulin in serum increased significantly as crude protein increased. It could be concluded that dietary level of 24% crude protein is recommended for growth performances and economic efficiency of growing Japanese quail because of increased dietary protein level beyond 24% % has no beneficial effect on growth.

Keywords: Blood chemistry , Carcass yield, Dietary protein., Feed efficiency ,Hematological examination ,Japanese quail

Introduction

The growing demand of animal protein could be met quickly by rearing quail commercially within a short period (**Thomas and Ahuja1989**). The quails meat has a more reduced calories level and a higher protein level than chicken and duck meat. The fat content is lower in quail than in chicken and duck meat and the proportions of vitamins (A, C, B2, B6, B12) and minerals (Ca, Fe, P, K, Zn, Cu) are higher. (**Ionita** *et*

al., 2011). Body weight daily weight gain and feed conversation ratio improved (P<0.05) for birds fed medium and high crud protein compared with birds fed low crud protein in diet (Sharifi et al., 2011). Dressed carcass as a percentage of live body weight was maximum in quails offered high protein (Mosaad et al., 2009) The muscle (breast and drumstick) yields were significantly higher in birds fed the high protein diet compared with those of the medium protein and Low protein diets(Laudadio et al., 2012). Higher protein level has positively influenced the participation of the trenched parts in the whole carcass structure (breast, wings, thighs and shanks (Marcu et al., 2011). Broiler fed low levels of protein had heavier liver weight than those fed diets with high level. (Golian et al., 2010). Percentage of fat pad decreased as crude protein increased in the diet (Ghahri et al., 2010). Total protein, albumin and globulin in serum tended to decrease with declining dietary protein levels (Tian et al., 2014). Increasing dietary protein level caused increase (P<0.05) in leucocytes which attribute to increase (P<0.05) in lymphocytes(Tollba, and El-Nagar2008). Japanese quail diets containing sub-optimal energy and protein levels helped in reducing microflara count, particularly pathogens and in turn, improved quail performance and immunity. (Mohamed 2009). The higher dietary protein level enhanced the ability of the birds to face undesirable circumstances. (Fathi et al., 2000) . Dietary protein levels appear to influence the immune response levels of broiler chickens (Cheema et al., 2003). Therefore the present study was perfomed to study the effects of dietary protein level on growth performance, carcass characteristics, intestinal microflora, hematological Parameters and some biochemical changes of growing Japanese quail.

Materials and Methods

2.1. Experimental groups:

A total of 120 one day old apparently healthy Japanese quail were used in this experiment. They were obtained from the General Egypt Poultry Organization. They were divided into 4 equal groups. Each group was housed in a separate compartment. Each compartment was bedded by fresh clean wood shave forming a deep litter of 4 cm depth and changed every week. Each compartment was provided with continuous lightening program, suitable feeder and water supply. Prophylactic antibiotics program measures against the most common infectious bacterial and viral diseases were carried out.

2.2. Experimental feeding program:

The present feeding trial was lasted 6 weeks The diets were formulated according to N.R.C. (1994) for Japanese quail (table 1) and the applied experimental feeding design according dietary crude protein level (table 2)

Physical composition	protein22.17%	protein24.2%	protein26.2%	protein28%
Yellow corn	60.2	56	51.7	49
Soybean meal	31.76	33	34.22	32.5
44%	4.5	7.57	10.7	15.12
Corn glutine	.9	.9	.9	.9
62%	1.3	1.3	1.3	1.3
Dicalcium	033	0.33	033	033
phosphate	0.25	0.2	,15	.15
Lime stone	.0.4	0.4	0.4	0.4
Choline 60%	0.3	0.3	0.3	0.3
Lysine				
Common salt				
Premix **				
Chemical				
composition%				
ME Kcal/kg *	2903.8	2902.6	2900	2935.6
Crude protein	22.17	24.2	26.2	28
Calcium	0.8	0.8	0.8	0.8
Available	0.3	0.3	0.3	0.3
phosphorus	1.3	1.3	1.3	1.3
Lysine	0.75	0.75	0.75	0.75
Methionine +	2000mg/1kg	2000mg/1kg	2000mg/1kg	2000mg/1kg
cystine				
Choline				

Table (1): calculated Physical and chemical composition of the experimental diets used for feeding Japanese quail.

.** The used premix (*Multivita Co.*) composed of vitamin A 12000000 IU, vitamin D_3 2200000 IU, vitamin E 10000 mg, vitamin K_3 2000 mg, vitamin B_1 1000 mg, vitamin B_2 5000 mg, vitamin B_6 1500 mg, vitamin B_{12} 10 mg, Niacin 30000 mg, Biotin 50 mg, Folic acid 1000 mg, Pantothenic acid 10000 mg, Iron 30000 mg, Manganese 60000 mg, Copper 4000 mg, Zinc 50000 mg, Iodine 1000 mg, Cobalt 100 mg, Selenium 100 mg, calcium carbonate (CaCO₃) carrier to 3000g

Group	Diet
1	protein deficient feed group (22.17% protein). i.e. diet contain protein
1	less than recommended level of N.R.C. (1994) act as negative control.
2	protein adequate feed group(24.2% protein) i.e. diet contain protein
	recommended level of N.R.C. (1994) act as positive control.
3	First higher level feed group(26.2% protein)
4	Second higher level feed group (28% protein)

Table : (2) The applied experimental design during the experimental period.

2.3 Experimental Parameters:

2.3.1 Growth performance measurements:

Feed intake and body weight of Japanese quail were recorded weekly and in cumulative manner for measuring growth performance. Body weight was calculated according to (Vohra and Roudybush, 1971). Body weight gain was calculated according to (Castell and Tiews, 1980) and the feed conversion ratio was calculated according to (Tacon, 1987).

2.3.2. carcass traits measurements:

At the end of the growing period (6 weeks), five birds were taken randomly from each group, weighed and slaughtered to complete bleeding and weighed to determine dressing percentage, breast muscle, leg muscle, Abdominal fat, head, liver, heart, gizzard weight and their relative weights to body weight were recorded.

2.3.3 Isolation and enumeration of intestinal microflora:

Caecal contents of slaughtered five quails per group were transferred under aseptic conditions into a sterile plastic bag, diluted with saline solution (1:10) and homogenized in prepared asepsis physiological saline, and diluted from 10^{-1} to 10^{-8} fold portions after dilution, 100 µl of each sample was plated onto the specific media. Nutrient agar medium was used for total aerobic Bacterial count , after incubation aerobically at 37 °C from 24 to 48 h (**Swanson** *et al.*,**1992**).MacConkey agar medium was used for Coliforms bacteria at 37c aerobically for 24 hours (**Oxoid**, **1992**).Eosin methylene blue (EMB) agar was used for E. coli incubated aerobically at 37 °C from 8 to 12 h (**Anonymous**, **1992**).De Man-Rogosa-Sharpe agar (MRS) was used for lactobacilli, cultivated in a 3% CO2 atmosphere at 37 °C for 48 h. (**Guban** *et al.*, **2006**).Numbers of colony forming units (cfu) were expressed as log 10 cfu per gram. The results are given as arithmetical means ± Standard Error (S.E.) The intestinal bacterial content was enumerated by pour plate method (**Quinn** *et al.*,**1992**). **S.S.agar** for salmonella cultivation at 37c aerobically for 24 hours.

2.3.4 Hematological Parameters:

Blood samples were collected for estimation of total erythrocytic count (TEC), total leucocytic count (TLC) and packed cell volume (PCV) according to **Stoskopf** (**1993**). Haemoglobin percentage (Hb%) was assessed according to **Drubkin (1947**), and differential leucocytic count were determined according to **Schalm (1986**).

2.3.5 Serum traits:

Serum total protein was determined according to **Doumas** *et al.*, (1981), Serum albumin was determined according to **Reinhold** (1953), Serum globulin was determined by subtract the total serum albumin from total serum protein according to (Coles, 1974) and Khalil (2000). Albumin/ globulin ratio was determined according to (Saffinaz, 2001).

2.3.6 Statistical analysis:

Statistical analysis was made using Analysis of variance test (ANOVA) followed by Duncan Multiple Range test (DMRT) to test the significance between different groups in studied variable according to (SAS, 2004).

Results and Discussion

3.1Growth Performance:

The analysis of variance of obtained data showed in(table 3). The results indicated that the final live body weight, body weight gain and feed conversion ratio of the groups fed on dietary levels of (24.2, 26.2 and 28%) crude protein significantly improved when compared with the group fed on dietary levels of 22.17% crude protein .The results revealed that dietary level of 24% crude protein is recommended for growth performances and economic efficiency of growing Japanese quail. These results are similar to those observed by Sharifi et al., (2011) who found that body weight ,daily weight gain and feed conversation ratio improved for Japanese quail fed medium and high crud protein compared with birds fed low crud protein in diet. This results are in agreement with those obtained by Jeong and Ryu (2008) who fed broilers on 18, 19, 20, 21, and 22% crude protein and found that the feed conversion efficiency significantly improved in broilers fed with 21 and 22% crude protein when compared with 20% crude protein (P<0.05). The 22% crude protein treated group showed significant body weight increase when compared with other groups. The body weight gain was higher in 22% crude protein treated groups than those 21 and 18% crude protein treated groups. the present results are also in accordance with Fathi et al., (2000) who reported that there was a 6% increase in body weight associated with higher level of dietary protein in local breeds of chicken. Our results are confirmed by Khati et al., (2007) who observed that broiler quails fed on low protein diet recorded significantly low feed conversation ratio.

3.2Carcass Characteristics:

The statistical analysis of the obtained data are shown in(table 4). The results revealed that dressing percentage, breast muscles and leg muscles were increased significantly as crude protein level increased in the diet while weights of liver, heart gizzard and abdominal fat percentage were decreased significantly as crude protein level increased .The previous findings are in agreement with those obtained by Mosaad et al., (2009) who found that dressed carcass as a percentage of live body weight was maximum in quails offered high protein .These results were similar also to that recorded by Marcu et al., (2011) who reported that higher protein level has positively influenced the participation of the trenched parts in the whole carcass structure (breast, wings, thighs and shanks . The present results are also in accordance with El-Fiky et al., (2007) who observed that weights of liver, heart, gizzard and carcass for chicks fed on low protein diet increased compared to those fed the optimum protein diet. These results disagree with those obtained by Mosaad et al., (2009) who reported that dietary protein levels had no significant (P<0.05) effect on the internal organ weights. In contrast to Our results Bunchasak et al., (2005) fed hens on three levels of dietary protein (14, 16 and 18%) and found that liver weight of hens fed 14% CP diet was smaller than those of 16 and 18% CP diets (P<0.05) which may be related to age and stage of production.

3.3Intestinal microflora:

Intestinal microflora count are illustrated in(table 5). The results indicated that total aerobic bacteria, Coliform bacteria, Lactic acid bacterium and *Escherichia coli* increased significantly as crude protein level increased while Salmonella was Non-dectable. The previous result agree with those obtained by **Mohamed (2009)** who found that Japanese quail diets containing sub-optimal energy and protein levels helped in reducing microflara count, particularly pathogens and in turn, improved quail performance and immunity. The results were supported by **Zhou et al., (2015)** who reported that in recent years, many studies have shown that reducing dietary protein level and supplementing synthetic amino acids is a way to reduce the rate of diarrhea in weaning piglets, and protecting the intestinal structure and function after weaning. This article reviewed the effects of low protein diets with balanced amino acids on intestinal morphology, digestion and absorption function, immune function, and intestinal microflora in weaning piglets.

3.4 Hematological studies:

Hematological examinations of the blood samples are presented in (table 6). The hematological examinations of results revealed that hemoglobin concentration, red blood cells count, total leucocytic count ,monocytes and lymphocytes were increased significantly as crude protein increased .while basophils, eosinophils and heterophils decreased significantly as crude protein increased. These results are in harmony with those recorded by **Tollba**, and **El-Nagar** (2008) who stated that increasing dietary

protein level for Egyptian laying hens caused increase (P<0.05) in leucocytes which was attribute to an increase (P<0.05) in lymphocytes. Our results agree also with those obtained by **Jahanian**. (**2009**) who found that an increase in dietary crude protein level of broiler chicks from 19 to 22.35% caused an increase (P<0.001) in the proportion of lymphocytes and consequently lower (P<0.05) heterophil-to-lymphocyte ratio. The previous findings was confirmed by **Fathi** *et al.*, (**2000**) who reported that the higher dietary protein level improved heterophil-to-lymphocyte ratio and enhanced the ability of the birds to face undesirable circumstances. These results disagree with those obtained by **Tollba, and El-Nagar(2008)**who observed that heterophil cells number did not significantly differ which may be related to species and age.

3.5 Serum traits:

Data concerned with the influence of dietary treatments on some serum traits are presented in(table 7) The analysis of variance of the present data showed that total protein, albumin and globulin in serum were increased significantly as crude protein increased. The previous findings are in agreement with those obtained by **Tian** *et al.*, (2014) who observed that total protein, albumin and globulin in serum tended to decrease with declining dietary protein levels. The present results are also in accordance with **Alagawany** *et al.*, (2011) who found that total protein and albumen of hens fed the high and moderate protein diet were significantly greater than those of hens fed the low-protein diet Our results was confirmed by **Bunchasak** *et al.*, (2005) who reported that serum total protein was tended to increase as protein levels increased.

CONCLUSION:

It could be concluded that dietary level of 24% crude protein is recommended for growth performances and economic efficiency of growing Japanese quail because of increased dietary protein level beyond 24% % has no beneficial effect on growth. The higher dietary protein level improved carcass characteristics, hematological parameters, serum total protein , plasma albumin and globulin concentration while intestinal microflora increased with higher protein level.

	Group				
parameters	<i>protein</i> deficient feed group 22.17% protein (Negative control)	<i>protein</i> Adequate feed group24.2% protein (Positive control)	First higher level feed group 26.2% protein	Second higher level feed group 28% protein	
Initial body weight	a	a	a	a	
	7.47+0.08	7.43+0.08	7.40+0.09	7.41+0.09	
Cumulative Final body	b 208.14±2.52	a	a	a	
weight		230.06±2.35	227.94±2.43	230.33±1.51	
Cumulative Total body gain	b	a	a	a	
	200.67±2.24	222.58±2.27	220.49±2.36	222.88±1.43	
Cumulative Average F.C.R.	a	b	b	b	
	3.32±0.04	2.86±0.06	2.97±0.07	2.88±0.08	

Table (3): Influence of dietary crude protein level on growth performance ofJapanese quail during experimental period:

Values are expressed as mean \pm standard errors. Means in the same row had different letters significantly differ at (**P** < **0.01**)Number=30

 Table (4): Influence of dietary crude protein level on carcass traits percentage of

 Japanese quail at the end of experimental period (Relative weight):

	Group			
Items	<i>protein</i> deficient feed group 22.17% protein (Negative control)	<i>protein</i> Adequate feed group24.2% protein (Positive control)	First higher level feed group 26.2% protein	Second higher level feed group 28% protein
Dressing %	с	b	a	a
	67.74±0.73	69.38±0.80	73.02±0.53	73.04±0.47
Head %	a	a	a	a
	4.95±0.14	5.00±0.15	4.99±0.08	4.95±0.08
Liver %	a	a	b	с
	2.40±0.20	2.25±0.11	1.90±0.14	1.76±0.19
Heart %	a	b	b	b
	1.02±0.0.02	0.91±0.03	0.92±0.03	0.91±0.02
Gizzard %	a	b	b	b
	2.03±0.08	1.98±0.09	1.95±0.07	1.96±0.08
Breast Muscles %	с	с	b	a
	18.24±0.16	18.77±0.19	19.30±0.27	20.03±0.13
Leg Muscles %	b	a	a	a
	12.87±0.47	13.71±0.44	13.38±0.32	13.58±0.19
Abdominal Fat %	a	b	b	b
	1.27+0.13	0.95+0.10	0.89+0.07	0.84+0.12

Values are expressed as mean \pm standard errors. Means in the same row had different letters significantly differ at (**P** < **0.01**)Number=5.

	Group				
intestinal microflora	<i>protein</i> deficient feed group 22.17% protein (Negative control)	<i>protein</i> Adequate feed group24.2% protein (Positive control)	First higher level feed group 26.2% protein	Second higher level feed group 28% protein	
Total aerobic bacteria	11.40±0.68b	11.40±0.75b	11.60±0.60a	11.60±0.60a	
Coliform bacteria	8.20±0.49c	8.40±0.24b	8.60±0.68a	8.60±0.51a	
Lactic acid bacterium	7.40±0.24c	7.60±0.24b	7.60±0.24b	8.00±0.55a	
Escherichia coli	5.20±0.37 b	5.80±0.58 b	5.40±0.51c	6.20±0.37a	
Salmonella sp.	N.d	N.d	N.d	N.d	

Table (5): Influence of dietary crude protein level on intestinal microflora (log cfu/g) of Japanese quail at the end of experimental period:

Values are expressed as mean \pm standard errors. Means in the same row had different letters significantly differ at (P < 0.01)

Number=5 N.d: Non-dectable

	Group			
Items	<i>protein</i> deficient feed group 22.17% protein (Negative control)	<i>protein</i> Adequate feed group24.2% protein (Positive control)	First higher level feed group 26.2% protein	Second higher level feed group 28% protein
Hemoglobin Concentration (gm/dl)	10.93±0.07c	11.53±0.18bc	11.74±0.20ab	12.09±0.06a
Red Blood Cells	с	ab	bc	a
Count (m/cml)	3.84±0.05	4.05±0.03	3.96±0.05	4.11±0.04
Haematocrit	b	ab	ab	a
(gm/dl) %	35.27±0.21	35.63±0.07	35.64±0.17	35.84±0.10
Platelets count (/ cml)	$\begin{array}{c} & d \\ 20.321 \pm 80.69 \end{array}$	b 20.752 ±28.28	c 20.627 ±76.69	a 20.849 ±31.18
Total leucocytic	bc	c	b	a
count (/ cml)	28.439±127.84	28.709±85.96	28.985 ±43.02	29.543±95.13
MCV (u3)	с	b	b	a
	140.60±0.51	142.40±0.51	143.60±0.51	145.20±0.37
MCH (Pg.)	c	b	b	a
	35.97±0.21	36.73±0.10	36.90±0.03	37.20±0.03
MCHC (%)	b	ab	ab	a
	25.73±0.09	26.16±0.06	26.14±0.25	26.58±0.16
Basophils (%)	a	bc	b	c
	1.98±0.09	1.70±0.02	1.75±0.04	1.65±0.02
Eosinophils (%)	a	b	с	b
	6.15±0.09	5.84±0.03	5.57±0.05	5.76±0.04
Heterophils (%)	a 33.87±0.26	a 33.60±0.13	b 32.61±0.17	c 31.96±0.20
Lymophocytes	d	c	b	a
(%)	53.57±0.07	54.85±0.03	55.86±0.06	56.24±0.17
Monocytes (%)	b	b	a	a
	5.14±0.11	5.10±0.09	6.16±0.14	6.34±0.16

Table (6): Influence of dietary crude protein level on hemogram studies of Japanese quail at the end of experimental period:

Values are expressed as mean \pm standard errors. Means in the same row had different letters significantly differ at (**P** < **0.01**) Number=5

 Table (7): Influence of dietary crude protein level on some serum traits of

 Japanese quail at the end of experimental period:

Group Items	<i>protein</i> deficient feed group 22.17% protein (Negative control)	<i>protein</i> Adequate feed group24.2% protein (Positive control)	First higher level feed group 26.2% protein	Second higher level feed group 28% protein
Total Protein	c	с	b	a
g\dl	4.84±0.08	5.16±0.09	5.86±0.22	6.75±0.10
Albumin g\dl	d	cd	bc	a
	2.87±0.05	3.12±0.06	3.33±0.18	4.03±0.08
Globulins g\dl	b	b	a	a
	1.98±0.07	2.04±0.05	2.53±0.24	2.72±0.07
Albumin /	a	a	a	a
Globulin Ratio	1.46±0.07	1.53±0.04	1.37±0.14	1.48±0.05

Values are expressed as mean \pm standard errors. Means in the same row had different letters significantly differ at (**P** < **0.01**) Number=5

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