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EFFECT OF BLACK SEED (*NIGELLA SATIVA*) AND GARLIC (*ALLIUM SATIVUM*) FEED SUPPLEMENTS ON PRODUCTIVE PERFORMANCE AND SOME PHYSIOLOGICAL AND IMMUNOLOGICAL RESPONSES OF JAPANESE QUAIL

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SUMMARY

A total number of 160 one day-old Japanese quails were used to evaluate some herbal feed additives (black seed and garlic) as growth promoters for quails, Birds were divided into four equal groups. The treatments were control (0% additives), 3% black seed, 3% garlic and 3% black seed + 3% garlic. Birds were exposed to16 hours light daily; feed and water were given ad-libitum. The experiment lasted for 10 weeks, during which some productive, physiological and immunological parameters were taken. Results indicated that using black seed and garlic improved body weight during 0-6 weeks of age. Black seed and garlic alone or in combination significantly decreased the feed intake at 4, 5, 6 weeks of age and increased feed conversion. Using black seed and garlic increased egg number, egg weight and egg mass. Adding black seed and garlic decreased serum cholesterol, LDL, HDL and total lipids in the treatments compared with control group. All treatments increased serum total protein, albumin and globulin. However, yolk cholesterol, LDL, HDL and total lipids were decreased by using all treatments compared to control group. Primary and secondary responses to Newcastle disease virus were higher in garlic group than black seed and control groups. Also using black seed and garlic have increased relative weights of spleen, bursa and thymus. It can be concluded that using black seed and garlic in quail diets improve the productive performance and some physiological and immunological parameters for Japanese quail and produce more healthy food for human consumption.

Keywords: *Japanese quail – garlic – black seed – production – immunity – blood biochemistry.*

INTRODUCTION

Feed additives are important materials that can improve the efficiency of feed utilization, animal performance and enhance immune response. In this century the medical properties of garlic and black seed have some considerations. Nowadays, there is an increase demand for using the natural biological feed additives instead of using synthetic drugs which may have many adverse effects (Fluck *et al.,* 1976).

There is a general agreement that using black seed and garlic as feed additives improve the performance, health and immunity in poultry. Moreover, they affect serum constituents and protect liver and kidney function. Supplementation of *Nigella sativa* succeeded to reduce the concentration of serum cholesterol, triglycerides (Mandour *et al.*, 1995), serum total lipids and liver cholesterol (Abdo, 1998) in broiler. Also, *Nigella sativa* reduced the fat content, serum total lipids and serum cholesterol in ducks (Ghazala and Ibrahim, 1996).

Nigella sativa seeds (NSS) is one of the most important medical plants. It contains compounds that possess antimicrobial, antifungal, antitoxic and pharmacological activities (Galil *et al.,* 1994). Moreover, Durrani *et al.* (2007b) explained that black seed had an antibacterial activity against different pathologenic bacteria. Similarly, Toghyani *et al.* (2010) and Khan *et al.* (2012) reported that increasing the level of black seed in the diet improved the immune response of chicks and induced marked increase in the weight of lymphoid organs. While, El-Ghamry *et al.* (1997) noticed no harmful effect in blood plasma constituents by feeding hens diet containing *Nigella sativa* meal. In addition, Abou-El-Soud (2000) found that weight gain, feed intake and feed conversion were higher for quails received diet supplemented with 2% black cumin seeds. Abou-Egla *et al.* (2001) noted that mortality tended to be lower in birds fed diets containing Nigella sativa meal.

Garlic (*Allium sativum)* is widely distributed and used in all parts of the world. It has antibacterial, antifungal, antitoxic, antiparasitic and antioxidant properties (Ertas *et al.,* 2005). Moreover, garlic is considered as hypo-cholesterolemic in rats (Sharma, 1986) and in normal persons. Abdo (1998) revealed that garlic at 3 % in broiler diets recorded the best finding of growth. Similarly, Mohamed *et al.* (2000) repoeted that garlic increases body weight, feed consumption and egg production in Fayoumi hens. El-ghamry *et al.* (2002) reported that fresh garlic leads to preferable performance parameters, increased immunity and viability of broiler chicks.

The objective of the present work is to use the natural feed additives in Japanese quail diets to increase its productive, immunological and physiological performance, and to avoid the bad side effects of chemical additives. Also to evaluate the possibility of using these additives in Japanese quail diets to obtain egg with low cholesterol content.

MATERIALS AND METHODS

Birds, diets and experimental design:

A total of 160 Japanese quail *(Coturnix coturnix japonica)* one-d old chicks were distributed into four groups with two replications, each of 20 chicks. The chicks were reared in starter batteries. They were maintained on a light cycle of 16L: 8D until the end of experiment. Chicks were fed on commercial starter and grower basal rations until six weeks of age. Thereafter, they were fed on the breeding ration till 10 wk old (Table 1). Both diets were formulated to meet NRC (1994) requirements. Chicks of the 1st group was fed on the basal diet without any supplements and considered as the control group. While the other three groups received the basal diet supplemented with 3% of powder Black Seed (Nigella sativa) and 3% powder Garlic (Allium sativum) and their combination in the 2nd, 3rd, and 4th, groups, respectively. The black seed and garlic were purchased from local market; black seeds were crushed and thoroughly mixed with basal diet. At three and five weeks old all chicks were vaccinated with Newcastle disease virus (NDV) by Lasota Strain Vaccine to detect the primary and secondary humoral immune response respectively.

Chicks were individually weighed at biweekly intervals until the sixth week of age. Live body weight, feed intake and feed conversion ratio were recorded during these periods.

Mortality was recorded daily; the accumulative mortality rate was calculated.

1- Physiological traits:

A- Blood hematological and biochemical determinations:

Blood samples were collected from randomly selected birds at four and six week of age. The clotted blood was centrifuged at the speed of 4000 r.p.m for 15 minutes. Sera were stored at –20C^{o} until biochemical and antibodies determinations were done.

Serum total protein (g/dl) was determined according to the method described by Henry (1964), the determination of serum albumin was carried out according to the method of Doumas *et al*. (1971), and globulin was calculated by subtraction of serum albumin from total serum protein. Moreover, total lipids, cholesterol, LDL- and HDL, were determined using chemical analysis by available commercial kits.

B- Yolk cholesterol and lipids content:

After measuring the egg quality, yolk samples for each treatment were separated from the broken eggs, calculated and extracted to determine cholesterol and total lipids according to Folch et al., (1957).

2- Immune Responses (humoral immunity):

A- Lymphoid organs weight and blood cells :

Ten birds were randomly selected from each treatment at four and six week old. Birds were slaughtered by severing the carotid artery and jugular vein. Lymphoid organs; bursa fabricius, thymus and spleen were excised from each bird, weighed, and their weights were recorded.

B- Antibody production against Newcastle disease virus (NDV):

Serum samples of the slaughtered birds that were immunized artificially with NDV as an active acquired immunity antibodies were collected at seven days after the first and second immunization (at

four and six weeks old for each hen) to detect the primary and secondary antibody hemagglutination (clumping of red blood cells)according to Hitchner *et al*. (1980). Serum samples from each treatment were tested for total antibodies response directed specifically against the triggering NVD.

White cells were counted and differentiated into lymphocytes (L), heterophils (H) and other types, and the ratio of H to L was calculated.

C- Cell-mediated immunity:

Five birds 6 wk old from each treatment were injected in the right wing with 0.1ml of phytohemagglutinin –L(sigma chemical co., st. louis, u 063178).(each 1 ml contains 100 µg PHA-L dissolved in sterile saline). The swelling of the wings were measured with micrometer before injection and post injection by 24, 48 and 72 hrs. While the left wing was injected with 0.1 ml of the sterile saline and kept as control birds.

3- Reproductive traits:

Egg production traits:

Eggs were collected daily from quails of each treatment from the first oviposition till the 10^{th} wk of age. Averages weekly egg number, and egg weight were calculated.

4- Statistical analysis:

The obtained data on immunological parameters, and blood parameters were analyzed using statistical analysis system (SAS, 1996). In order to determine significant differences between all possible mean comparisons Duncan`s multiple range test (1955) was applied. Statistical significance was accepted at a probability level of 0.05.

Table (1): Composition of Japanese quail starter, grower and breeder basal rations.

Ingredient	*Starter and Grower, %*	Breeder, %
Yellow corn	55.20	59.10
Soybean meal (44%)	36.00	27.00
Corn gluten (60%)	5.80	5.20
Bone meal	1.22	1.80
Plant oil	0.15	1.23
Limestone	0.86	4.94
Methionine	0.104	0.11
Lysine	0.125	0.07
Premix*	0.30	0.30
Salt	0.25	0.25
Total	100	100
Calculated analysis		
Crude protein %	24.012	20.02
Metabolizable Energy (kcal/kg)	2906.16	2884
Crude fiber %	3.86	3.37
Crude fat %	2.44	2.51
Calcium (%)	0.8	2.5
Available phosphorus (%)	0.3	0.35

**: Premix:Each kilogram of diet contains vitamins and trace elements as follow = A, 12000 I.U., D3, 2500 I.U., E, 10mg., B1, 2mg., B2, 5mg., B6, 4mg., B12, 10µg., Niacin, 25mg., Pantothenic acid, 10mg., Biotin, 50µg., Folic acid, 1000µg., and Coline chloride, 255mg. Selenium, 350µg., Copper, 10mg., Iodine, 1.0mg., K, 2.0mg., Iron, 33mg., Manganese, 60mg.and , 100mg. Zinc.*

RESULTS AND DISCUSSION

1. Productive Trait:

A. Body weight:

Results presented in table (2) showed that 3% black seed improved body weight at ages 4-5-6 wk compared with control.

This result agrees with those reported by Abel-Latif *et al.* (2002) and Abou El-Soud (2000) who showed that quails fed dietary black seed recorded the highest values of body weight. Similar results were obtained with layers (Nofal *et al.*, 2006 and El-Kaiaty *et al.*, 2002a).

The improvement of the body weight may be due to the antimicrobial effect of the black seeds (Hanafy and Hatem, 1991 and Soltan, 1999) and the high amount of unsaturated fatty acids in the black seeds which are very essential to the poultry and this may explain the significant effects of dietary herbal feed additives in improving metabolic process (Abd El-Malak *et al.*, 1995) which is also in agreement with Samanta and Dey (1991) and Mahmood *et al.* (2009), who concluded that powdered garlic may be incorporated as growth promoter in the ration of Japanese quails. In addition, there are other pharmacological positive effects of black seed on growth performance may be attributed to its content of volatile oil or essential oil has certain biological functions that could act not only as antibacterials and antioxidants but also as a stimulant of digestive enzymes in the intestinal mucosa and pancreas that improve the digestion of dietary nutrients and feed efficiency subsequently increasing the growth rate (Abu-Dieyeh and Abu- Darwish, 2008), these results are in agreement with Ismail (2011).

Table (2): Body weight (g) of Japanese quail chicks fed on diet supplemented with 3% black seed, 3% garlic and their combination.

Age (wk)	Treatment					
	Control	Black seed 3%	Garlic 3%	Black seed 3% + garlic 3%	MSE	Pro
1	27.29	27.55	27.91	26.01	0.767	0.116
2	60.50	61.86	58.40	58.59	2.775	0.652
3	100.48	106.07	104.43	103.77	2.982	0.812
4	135.39 [b]	145.89 [a]	142.45 [a]	143.03 [a]	3.354	0.031
5	171.01 [c]	191.15 [a]	184.95 [b]	187.64 [b]	4.993	0.041
6	203.15 [c]	225.44 [a]	216.40 [b]	215.50 [b]	4.878	0.021

Means within a row with different superscripts are significantly different (P≤0.05).

B. Feed intake and feed conversion:

It is clear from tables (3 and 4) that feed intake and feed conversion were significantly decreased by using 3% black seed alone and 3% black seed + 3% garlic compared with control group and group fed 3% garlic alone at 5 and 6 wk of age for Japanese quail.

The results was in agreement with those of Soltan (1999) who found that the addition of 1% Nigella sativa seeds to the diet of quail improved feed conversion ratio, Zeweil (1996) in quail, EL-Kaiaty et al (2002a) in layer and Osman and EL-Barody (1999) in broilers.

Table (3): Feed intake (g) of Japanese quail chicks fed on diet supplemented with 3% black seed, 3% garlic and their combination.

Age (wk)	Treatment					
	Control	Black seed 3%	Garlic 3%	Black seed 3% + garlic 3%	MSE	P<0.05
1	52.14	55.02	50.98	54.38	2.63	0.8591
2	77.03	65.47	77.56	74.44	3.13	0.5308
3	142.37	138.21	139.21	140.55	3.48	0.3542
4	150.11	144.84	149.17	145.95	4.23	0.4650
5	172.86 [a]	149.15 [c]	155.95 [b]	152.64 [bc]	2.26	0.0411
6	211.31 [a]	186.44 [c]	191.40 [b]	189.50 [bc]	4.43	0.0302

Means within a row with different superscripts are significantly different (P≤0.05).

The improvement in feed conversion in Nigella sativa seeds and fresh garlic groups may be due to the antibacterial, antifungal, antitoxic and other active compounds.

Abdo (1998) reported that adding fresh garlic or black seed in broiler diets gave the best findings of growth, meat quality and immunity of broilers. While, Soliman *et al.* (1999) reported that fresh garlic leads to preferable performance parameters and increased immunity and viability of broiler chickens.

Tolba and Hassan (2003) found that garlic improved broiler growth, feed conversion and decreased mortality rate, these results are in agreement with Saeid *et al.* (2013).

The mortality rates through the period from 2 till 10 wk of age are presented in (Table 4). The mortality rate recorded for control birds, was significantly higher than those received 3% Black Seed or 3% Garlic and their combination. This indicates that the addition of Black Seed and Garlic improved livability of the chicks.

The present results indicated that body weight of quails was improved significantly at 4 to 6 weeks of age while feed intake and feed conversion were improved significantly by the feed supplements at 5 and 6 weeks of age but not at younger ages.

Table (4): Feed conversion ratio (FCR) and mortality rate (%) of Japanese quail chicks fed on diet supplemented with 3% black seed, 3% garlic and their combination.

Trait	Age (wk)	Treatment				MSE	Pro
		Control	Black seed 3%	Garlic 3%	Black seed 3% + garlic 3%		
FCR	1	2.33	2.54	2.40	2.43	0.154	0.667
	2	2.30	1.84	2.32	2.37	0.189	0.759
	3	3.45	2.65	2.55	3.16	0.347	0.351
	4	4.37	4.07	4.33	3.97	0.633	0.751
	5	6.31 [a]	4.41 [c]	5.62 [b]	5.10 [b]	0.128	0.0001
	6	8.61 [a]	6.26 [b]	6.95 [b]	6.72 [b]	0.903	0.0003
Mortality rate, %	1-10	4.00 [a]	2.90 [b]	2.50 [b]	2.81 [b]	0.437	0.0001

Means within a row with different superscripts are significantly different (P≤0.05).

C- Egg production (egg number, egg weight and egg mass):

Tables (5, 6 and 7) indicated that the egg number, egg weight and egg mass were increased in groups fed 3% black seed, 3% garlic and their combination compared with control.

These results agree with those of Soltan (1999) who found that the addition of Nigella Sativa seeds to the diet of quail improved egg production percentage, egg mass. Also Khodary *et al.* (1996) showed that broilers fed on diet supplemented with black seeds increased egg production as was found by Hassan *et al.* (2007).

Table (5): Egg number per hen per week of Japanese quail breeder hens fed on basal diet supplemented with 3% black seed, 3% garlic and their combination.

Age (wk)	Treatment				MSE	Pro
	Control	Black seed 3%	Garlic 3%	Black seed 3% + garlic 3%		
7	2.23	2.75	2.50	2.83	0.393	0.0711
8	3.50 [c]	4.50 [b]	4.75 [b]	5.50 [a]	0.448	0.0157
9	3.80 [c]	5.25 [b]	5.01 [b]	6.25 [a]	0.478	0.0268
10	4.61 [c]	6.13 [b]	6.20 [b]	7.21 [a]	0.419	0.0048

Means within a row with different superscripts are significantly different (P≤0.05).

Table (6): Egg weight (g) per hen per week of Japanese quail breeder hens fed on basal diet supplemented with 3% black seed, 3% garlic and their combination .

Age (wk)	Treatment				MSE	Pro
	Control	Black seed 3%	Garlic 3%	Black seed 3% + garlic 3%		
7	8.17	8.50	8.30	8.40	0.106	0.081
8	9.33 [b]	10.45 [a]	10.85 [a]	10.92 [a]	0.113	0.015
9	9.87 [c]	12.60 [b]	12.40 [b]	13.20 [a]	0.052	0.026
10	10.34 [c]	13.15 [b]	13.35 [b]	14.23 [a]	0.073	0.004

Means within a row with different superscripts are significantly different (P≤0.05).

Table (7): Egg mass (g) per hen per week of Japanese quail breeder hens fed on basal diet supplemented with 3% black seed, 3% garlic and their combination .

Age (wk)	Treatment				MSE	Pro
	Control	Black seed 3%	Garlic 3%	Black seed 3% + garlic 3%		
7	18.219	23.37	20.75	23.77	3.57	0.071
8	32.655 [b]	47.02 [a]	51.54 [a]	60.06 [a]	4.794	0.0005
9	37.506 [c]	66.15 [b]	62.12 [b]	82.5 [a]	2.489	0.0001
10	47.667 [c]	80.60 [b]	82.77 [b]	102.59 [a]	4.887	0.0003

Means within a row with different superscripts are significantly different (P≤0.05).

2- Immunological traits:

A- Relative lymphoid organs weight:

All of treatments showed the same trend of increase in thymus and bursa than control, but no significant differences in spleen values. It can be noticed that black seed, garlic or combination of black seed + garlic had more effect on thymus, bursa than other studied groups in Table (8).

This agrees with that reported by Abdo (1998), El-Kaiaty *et al*. (2002a), Hassan *et al.* (2007) and Osman and El-Barody (1999) who reported that supplementation of black seed had significant effect on lymphoid organs and immune response.

Table (8): Relative Lymphoid organ weights of 4 and 6 wk old Japanese quail chicks fed on basal diet supplemented with 3% black seed, 3% garlic and their combination.

Trait	Treatment				MSE	Pro
	Control	black seed 3%	Garlic 3%	Black seed 3% + garlic 3%		
At 4 wk						
Thymus	0.105 [b]	0.291 [a]	0.313 [a]	0.326 [a]	0.0040	0.0001
Bursa	0.082 [b]	0.186 [a]	0.210 [a]	0.203 [a]	0.0056	0.0001
Spleen	0.071	0.160	0.192	0.183	0.0009	0.0800
At 6 wk						
Thymus	0.086 [b]	0.163 [a]	0.182 [a]	0.191 [a]	0.025	0.0435
Bursa	0.073 [b]	0.161 [a]	0.173 [a]	0.182 [a]	0.310	0.0490
Spleen	0.056	0.057	0.062	0.065	0.006	0.045

Means within a row with different superscripts are significantly different (P≤0.05).

B- Immune response (Antibody production)

Humoral immunity (Ab's production) against NDV vaccination was tested at 4 and 6 weeks of age during treatments. Geometric means of Ab's production were determined and shown in Table (9)

At primary and secondary response, using 3 % garlic gave the highest value; also, all treatments were higher than control group. These effects, may be, due to both effective substances Nigllone (in black seed) and Allicin (in garlic) which activate the secondary lymphoid organs and serum globulin. Secondary response was higher when compared with primary. The same result was obtained by El-Kaiaty *et al.* (2002b). Increasing secondary response values may be due to the age of hen. This finding was in complete agreement with that of Vanderzijp *et al*. (1983) and lamont and Smyth (1984). Trout *et al.* (1988) indicated that on the early step in the initiation of humoral immunity (Ab's production); there is an increase in serum hormones (especially, corticosterone, T_3 and T_4). Also, physiological status may play an important role in immune response.

These results were in complete agreement with that of Bachman and Mashaly (1986). Also the increase in WBC may be due to an increase in the membrane protection from autoxidation, because there are numerous reports on the significant role of black seed in increasing immunity and inhibiting non-enzymatic peroxidation (Houghton et al., 1995). Similarly, Durrani et al. (2007a) reported that black seed and garlic stimulates the immune system.

The Birds fed on diet supplemented with 3% black seed, 3% garlic and their combination had significantly lower heterophils to lymphocytes ratio H/L at four and six wk of age (Table 9). The highest values of H/L ratio were for control group, compared with the other experimental groups. While the lowest value of H/L ratio were for chicks fed on diet supplemented with 3% garlic. These results are in agreement with those reported by Ather (2009) who showed that the lowest H/L ratio was observed in the1% black seed treated group, this suggests that black seed is possible to be used as an anti stress factors when added to feed. Ali *et al.* (2014) found the same results.

Table (9): Heterophils to lymphocytes ratio (H/L ratio) and Humoral immunity against NDV of 4 and 6 wk old Japanese quail fed on basal diet supplemented with 3% black seed, 3% garlic and their combination.

Trait	Treatment				MSE	Pro
	Control	black seed 3%	Garlic 3%	Black seed 3% + garlic 3%		
At 4 wk						
H/L ratio	0.42 [a]	0.32 [b]	0.20 [c]	0.25 [c]	0.0137	0.0432
1st Ab titer	3.05 [c]	4.72 [b]	5.85 [a]	5.10 [b]	0.4430	0.0001
At 6 wk						
H/L ratio	0.45 [a]	0.36 [b]	0.22 [c]	0.28 [c]	0.0543	0.0313
2nd Ab titer	3.41 [c]	5.72 [b]	6.43 [a]	6.14 [b]	0.3234	0.0002

Means within a row with different superscripts are significantly different (P≤0.05).
1st Ab: primary response of antibody production at 4 wk of age.
2nd Ab: secondary response of antibody production at 6 wk of age.

C- Cell-mediated immunity:

It is well known that phytoheamagglutinin is a lectin isolated from red kidney bean and stimulates T-cell proliferation with minimal effect on B cells. In vivo it is considered a good measure of T-lymphocyte function (Qureshi *et al*., 1997). Cell-mediated immunity (CMI) plays a major role in the response against intracellular bacteria and virus. It is clear from Table (10) that, injection of PHA-L significantly increased the wing-web swelling of chicks received diet supplemented with 3% of Black Seed, 3% Garlic and their combination. It is clearly observed that chicks received diet with no supplementation had the lowest wing-web swelling 24 h post PHA-L injection compared to the other experimental groups.

Table (10): Wing-web swelling (µm) in response to PHA-L injection (as indicative of cellular immunity) of Japanese quail chick fed on basal diet supplemented with 3% black seed, 3% garlic and their combination.

Time Post- injection (hour)	Treatment				MSE	Pro
	Control	black seed 3%	Garlic 3%	Black seed 3% + garlic 3%		
24	0.864 [b]	2.033 [a]	2.432 [a]	2.251 [a]	0.0044	0.0001
48	0.821 [b]	1.027 [a]	1.043 [a]	1.032 [a]	0.0035	0.0001
72	0.762	0.846	0.885	0.862	0.0059	0.231

Means within a row with different superscripts are significantly different (P≤0.05).

3- Physiological traits:

A- Serum cholesterol high density lipoprotein (HDL) and low density lipoprotein (LDL):

Results in table (11) showed that supplemented diets with feed additives decreased serum cholesterol level, while HDL serum cholesterol was higher in control.

It can be concluded that the best supplementation for reducing serum cholesterol and HDL is 3% garlic + 3% black seeds.

Results explained that the depression of serum HDL due to suppression HMG Co-A that was essential for the synthesis of the cholesterol in liver. Both Nigllone in black seed and Allicine in garlic were the effective substances suppressed the synthesis of HMG Co-A in liver (El-Kaiaty *et al.*, 2002b).

This result agreed with Mandour *et al.* (1998) who reported a decrease in plasma HDL concentration resulted from feeding chicken with 0.50% of black cumin. Phytosterol compound found in black cumin can inhibit the formation of micelles due to the absorption of bile acids into the intestine. This is in accordance with the finding of Bonsdorff (2005) that the mechanism of phytosterols is believed to inhibit cholesterol absorption and reabsorption of endogenous cholesterol derived from the ration in the digestive tract. Therefore, phytosterols increase the spending time of the excess cholesterol absorption, and causes a decrease in serum cholesterol levels. It is assumed that cholesterol reduction would be offset by the decreased levels of HDL in the blood serum.

Furthermore, Galal *et al.* (1997), Mohamed *et al.* (2000) and Helal and Mohamed (2001) showed that garlic had a significant effect on lowering the level of serum cholesterol of either chicks or rabbits and the reduction is positively related to the level of garlic in the diet.

The main reason of cholesterol and triglyceride reduction in blood may be due to substances like carvacol and thymol present in herbs (Akiba and Matsumoto, 1982). These substances decrease cholesterol and triglyceride concentration in blood (Zargari, 2001).

B- Serum total lipids (STL):

It is clear that all feed additives decreased significantly serum total lipids compared with control.

Table (11): Serum constituents of 6 wk old Japanese quail chicks fed on basal diet supplemented with 3% black seed, 3% garlic and their combination.

Traits	Treatment					
	control	Black seed 3%	Garlic 3%	Black seed 3% + garlic 3%	MSE	Pro
Cholesterol (mg/dL)	225.76 [a]	171.53 [c]	185.67 [b]	173.34 [c]	1.342	0.0001
LDL (mg/dL)	168.32 [a]	143.51 [b]	147.21 [b]	142.31 [b]	1.251	0.030
HDL (mg/dL)	47.50 [a]	34.71 [c]	39.02 [b]	32.21 [c]	1.73	0.0001
Total lipids (mg/dL)	862.21 [a]	763.54 [c]	789.56 [b]	768.34 [c]	1.821	0.0001

Means within a row with different superscripts are significantly different (P≤0.05).

C- Serum total protein (TP), Albumin (ALB) and Globulin (G):

It can be observed from table (12) that serum total protein, albumin and globulin were increased by supplemented 3% black seed, 3% garlic and their combination compared with control .Increase of globulin may be due to the immunostimulant effect of Nigella sativa (Cole *et al.*, 1994).

This result agrees with that reported by Abd El-latif *et al.* (2002) who showed that adding black seed to quail diet enhanced plasma total protein, albumin and improved plasma globulin at 6 weeks of age. Also Tollba *et al.* (2005) and Abou El-Soud (2000) showed that adding black seed for diets increased total protein, albumin and immunoglobulin.

This conclusion agrees with the finding of Khodary *et al.* (1996) and Nagwa and Fathy (1997) which confirms the previous reports about the immune stimulant effect of *Nigella Sativa* and its high content of essential amino acids (Khalifah, 1995) and this may explain the significant effects of dietary herbal feed additives in improving metabolic process.

Table (12): Serum concentrations of total protein, albumin and globulin g/dl of 6 wk old Japanese quail chicks fed on diet supplemented with 3% black seed, 3% garlic and their combination .

Traits	Treatment					
	control	Black seed 3%	Garlic 3%	Black seed 3% + garlic 3%	MSE	Pro
Total protein (g/dl)	4.61 [c]	6.92 [a]	6.28 [b]	6.31 [b]	0.383	0.003
Albumin (g/dl)	2.40 [b]	3.81 [a]	3.45 [a]	3.65 [a]	0.239	0.001
Globulin (g/dl)	2.21 [c]	3.11 [a]	2.83 [ab]	2.66 [b]	0.409	0.004

Means within a row with different superscripts are significantly different (P≤0.05).

D- Yolk contents:

Results in Table (13) indicated that adding 3% black seed, 3% garlic and their combinations significantly decreased the yolk cholesterol, LDL, HDL and total lipids compared with control group. This agreed with that reported by Hassan *et al.* (2007), Nofal et al. (2006), Tollba *et al.* (2005) and El-Kaiaty *et al.* (2002a) who observed that adding black seed or garlic for diets decreased significantly the yolk cholesterol and total lipids and may produce egg with low cholesterol and total lipids content.

Table (13): Some yolk parameters of Japanese quail breeder hens fed on basal diet supplemented with 3% black seed, 3% garlic and their combination.

Traits	Treatment					
	control	Black seed 3%	Garlic 3%	Black seed 3% + garlic 3%	MSE	Pro
Cholesterol (mg/g yolk)	15.66 [a]	10.82 [b]	11.72 [b]	10.62 [b]	0.35	0.003
LDL (mg/g yolk)	9.73 [a]	6.43 [b]	6.83 [b]	6.72 [b]	0.41	0.001
HDL (mg/g yolk)	5.84 [a]	4.33 [c]	4.95 [b]	4.21 [c]	0.23	0.004
Total lipids (mg/g yolk)	237.12 [a]	186.41 [c]	193.52 [b]	182.31 [c]	0.72	0.002

Means within a row with different superscripts are significantly different (P≤0.05).

CONCLUSION

The results of the present study indicate that 3% black seed and 3% garlic either alone or in combination can improve body weight, feed conversion, egg production, livability and immunity. Moreover, it reduces the total lipids and cholesterol in egg of Japanese quails and produces more healthy food for human consumption.

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تأثير اضافة حبة البركة والثوم على الآداء الانتاجي وبعض الاستجابات الفسيولوجية والمناعية فى السمان اليابانى

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استخدم فى هذه الدراسة عدد 160 كتكوت سمان عمر يوم وتم تقسيمهم الى 4 مجاميع بكل مجموعة 40 طائر وكانت المجموعات على النحو التالى:

المعاملة الاولى : العليقة الاساسية بدون أى اضافة

المعاملة الثانية : العليقة الاساسية + 3% حبة بركة مطحونة

المعاملة الثالثة : العليقة الاساسية + 3% مسحوق ثوم

المعاملة الرابعة : العليقة الاساسية + 3% حبة بركة مطحونة + 3% مسحوق ثوم

الطيور تعرضت الى 16 ساعة اضاءة / يوم ، العلف والماء قدم بصورة حرة.

النتائج فى هذه الدراسة أوضحت أن هناك فروق معنوية فى وزن الجسم ومعدل تحويل العلف وكمية العلف المستهلك وكل المعاملات قللت العلف المستهلك وزاد وزن الجسم وكفاءة التحويل الغذائى. كل المعاملات حسنت وزن البيض وكتلة البيض.

وكانت أعلاها هى المعاملات المحتوية على الثوم + حبة البركة ، كوليسترول سيرم الدم والصفار انخفض فى كل المعاملات ، زاد البروتين الكلى والالبيومين والجلوبيولين فى السيرم ، الاستجابة المناعية الاولية والثانوية كانت أعلى فى كل المعاملات عن مجموعة االكنترول.

ارتفع مستوى الاجسلم المضادة ضد مرض النيوكاسل (المناعة السائلية) وزيادة تورم وسادة الجناح بعد الحقن بمادة PHA-L (المناعة الخلوية) وكانت أعلاها هى المعاملات المحتوية على الثوم ، زاد وزن الاعضاء الليمفاوية فى كل المعاملات بالنسبة لمجموعة الكنترول.

توصى هذه الدراسة باستخدام الاضافات الغذائية الطبيعية مثل حبة البركة أو الثوم أو خليطهما بنسبة 3% الى علائق السمان اليابانى لتحسين الآداء الانتاجى والفسيولوجى والمناعى والذى يمكن أن ينعكس على زيادة وزن الجسم والانتاج وقلة استهلاك العليقة ، وانتاج بيض منخفض الكوليسترول والدهون الضارة بصحة الانسان مع تحسين الحالة المناعية والصحية للسمان اليابانى بالاضافة لانتاج بيض ولحم أمن صحياً.