EFFECT OF FUNCTIONAL FEED ADDITIVES ON EGG PRODUCTION, HATCHABILITY AND HEMATOLOGICAL TRAITS OF JAPANESE QUAILS DURING SUMMER CONDITION

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ABSTRACT
The present study was conducted to investigate the effect of using functional feed additives on egg production, hatchability and hematochemical traits of Japanese quails under hot weather condition. A total of 288 a day old of Japanese quail were allocated to six treatments with three replicates containing T0: control, T1: Optifeed® 500g/ton feed, T2: Oleobiotec® 100g/ton feed, T3: Force®6 100g/ton, T4: VêO® premium 250g/ton feed and T5: mixing of (Optifeed® 250g + Oleobiotec® 50g+ Force®6 (50 g)+VêO® premium 125 g/ton). The results were summarized that the functional feed additives were significantly higher in body weight, body weight gain in T1 and T5, high economic profit in T1, T4 and T5 and FCR improved in T1, T3, T4 and T5 at age 35 day old, egg production percentage and economic profit and food conversion ratio (FCR) more improvement in the all treatments of feed additives, feed intake in T1 and T3 at ages (36-70) day old. Also higher Haugh unit, egg weight, eggshell thickness, eggshell strength, egg yolk lipids, RBCs, Hb, PC.

Keyword: feed additives, egg production, hatchability, hematochemical traits, Japanese quails, hot weather.

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The results were summarized that the all treatments of feed additives, feed intake in T1 and T3 at ages (36-70) d, total (1–17) d, body weight gain in T1 and T5 at age 35 day old, egg production percentage and economic profit and food conversion ratio (FCR) more improvement in the all treatments of feed additives, feed intake in T1 and T3 at ages (36-70) day old. Also higher Haugh unit, egg weight, eggshell thickness, eggshell strength, egg yolk lipids, RBCs, Hb, PC.

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INTRODUCTION
Nutrition is obviously a critical determinant in the growth of the gut in the layer quails, beneficial effects of dietary additives such as herbs extracts or organic acids, on the energy and protein utilization of poultry (19). Spices and herbs of various plants extract have appetizing and digestion stimulating property and antimicrobial effects (1 - 2). When considering supplementing the feed with herbs and spices or their extracts to stimulate the appetite, we have to know the taste preferences of different animal species (11). The ban on nutritive antibiotic use increased awareness of the consumers triggered a need for natural and safe feed additives to achieve better production results on farm animals. Plant extracts are used in animal nutrition as appetite and digestion stimulants, stimulants of physiological functions, for prevention and treatment of certain pathological conditions, as colorants and antioxidants (8). So, there is a tendency to use herbs and probiotics as natural feed additives to avoid the residual cumulative effect for either antibiotics or synthetic drugs in final products of poultry, which has a negative effect on the human health (15). Antioxidants compounds like herbs are substances present in lower concentrations and significantly delay or prevent oxidation of substrates such as protein, lipids, carbohydrates and DNA (17).

The objective of this trial was to check the effect of Optifeed® (appetite stimulation), Oleobiotic® (health & performances improvement), VêO® Premium (stress management) and Force®6 (vectorized curcumin as antioxidant) in local quails productive and reproductive performances and some blood physiological analysis.

MATERIAL AND METHODS
This experiment was carried out at the Poultry Farm, Department of Animal Resources / College of Agriculture / University of Salahaddin-Erbil during (28 June to 5 September 2015). A total of 288 day old unsexed Japanese quails were reared in cages for 70 days. Chicks were randomly distributed into 6 treatments, each treatment with 3 replicates: 48 chicks (36 females and 12 males) per each replicate, (T0= Control, T1= Optifeed® (500g/ton feed), T2=Oleobiotic®100g/T feed, T3= Force®6 100 g/T, T4=VêO® premium 250 g/T feed and T5= (Optifeed® 250g + Oleobiotec® 50g+ Force®6 (50 g)+VêO® premium 125 g)/Ton, the quantity of additives used as a recommendation of FODE laboratory-France. Feed and water were supplied ad libitum, the feed content 3200, 3100 and 3050 kcal/kg metabolizable energy, 23.2, 22.4 and 21.8% crude protein in starter, grower and layer diet respectively. The temperature in quail house was as average (33±2)°C. The body weight, body gain, feed intake, feed conversion ratio, mortality and economic profit were measured at the age 35 and 70 days, also at the age 70 day egg production (H.D%), egg weight, haugh unit, shell thickness, shell weight, eggshell strength, total lipid cholesterol and malondialdehyde in egg yolk by using commercial kits. At the end of 70th d, blood samples from the brachial vein of 15 birds from each treatment were collected in EDTA tubes to measure the total RBC (106/mm3) were determined by using Natt & Herrick staining solution (12) in a haemocytometer chamber. Differential leukocyte count (heterophil and lymphocyte) made on slides stained with Wright-Giemsas and observed in an optical microscope (100x) to determined H/L ratio. Hemoglobin level (g/100 ml) was measured by the cyanmethemoglobin method and hematocrit (PCV) (%) was determined using a micro-hematocrit capillary, serum was harvested after blood centrifuged to measure the total cholesterol (mg/l), high density lipoprotein (HDL), low density lipoprotein (LDL), very low density lipoprotein (VLDL) concentration and determined serum antioxidant enzymes concentration of antioxidant activity (AOA), malondialdehyde (MDA) and glutathione peroxidase (GPx) using commercial kits from Bio-compare. At the beginning of 70th d for 7 days a total of 1200 fertile eggs, 200 eggs for each treatment incubated and hatched during 17 days. At hatching all live and dead chicks were counted and the percentages of (fertility, hatching of fertile and total eggs, (early 1-7 d, mid 8-14 d, late 15-17 d and total 1-17 d) dead embryos, disabled chicks and hatched chicks weight. The antibody titer of Newcastle Disease (HI), Infectious Bursa Disease (IBD) or Gumboro...
and Infectious Bronchitis Viral (IBV) were measured by ELISA. All data were analyzed by using CRD (Complete Randomize Design) by SAS (16), as per variance, significant differences among treatment means were determined by Duncan’s multiple range tests (6).

RESULTS AND DISCUSSION

Body weight (BW) and body weight gain (BWG) were significantly (p<0.05) higher in the treatments T1 and T5, also high economic profit (EP) in T1, T4 and T5 and feed conversion ratio (FCR) improved in T1, T3, T4 and T5 at age 35 day old. Hen housed egg production percentage and economic profit were achieved highly significant (p<0.05) and FCR more improvement in the all treatments of feed additives compared to the control, also feed intake was significantly (p<0.05) higher in T1 and T3 compared with the other treatments at ages (36-70) day old. But, mortality was not significant among all treatments at ages 35 and 70 days old (Table 1). Aromatic plants and their extracts have a positive effect on growth performance in chickens (10), weight gain and feed intake were increased whereas the feed gain ratio was lowered when compared to control, also dietary essential oils and spices or their active components have a positive effect of on digestion process. The initial effect of adding spice plant extracts to animal or bird feeds is one of stimulating appetite; aroma can stimulate the olfactory nerves and gustatory papillae (13). Feed supplements with growth promoting activity increase stability of feed and beneficially influence the gastrointestinal ecosystem mostly through growth inhibition of pathogenic microorganism’s growth due to improved health status of digestive system, animals are less exposed to the toxins of micro-biological origin, consequently herbs and spices help to increase the resistance of the animals exposed to different stress situations and increase the absorption of essential nutrients, thus improving the growth of the animals (18).

Table 1. Effect of functional feed additives on body performances and economic profit of Japanese quails at ages 35 and 70 days.

| Treatments | IBW (g) | BW (g) | BWG (g) | FI (g) | FCR | Mortality% | EP IQD/quail | Egg Production (HD %) | F1 (g) | FCR | Mortality% | EP IQD/quail |
|------------|---------|--------|---------|--------|-----|-----------|-------------|----------------------|--------|-----|-----------|-------------|
| 35 day old | T0      | T1     | T2      | T3     | T4  | T5        | T0          | T1                   | T2     | T3  | T4        | T5          |
| T0= Control, T1= Optifeed® (500g/ton feed), T2=Oleobiotech®100g/T feed, T3=Force®6: 100 g/T, T4=VêO® premium 250 g/T feed and T5= (Optifeed®250g+Oleobiotech®50g+ Force® (50 g)+VêO® premium 125 g)/T/on, BW =body weight, BWG= body weight gain, FCR= feed conversion ratio, FI= feed intake, HD= hen day. The same superscripts within rows means non-significant, a, b, c Means within rows with different superscripts differ significantly at (P≤ 0.05).

The results of Haugh unit, egg weight, eggshell thickness, eggshell strength and egg yolk lipids concentration were significantly (P<0.05) higher in the treatments which were given the feed additives compared to the control. While, egg yolk cholesterol and malondialdehyde concentrations were significantly (P<0.05) lower in the treatments of feed additives compared to the control. However there were no significant differences concerning between control and experimental treatments in the relative weight of eggshell (Table 2).
Table 2. Effect of functional feed additives on Haugh unit, egg weight, some eggshell quality and some egg yolk measurement of Japanese quails at age 70 days.

| Traits                          | T0            | T1              | T2              | T3              | T4              | T5              | SEM   | S.L   |
|---------------------------------|---------------|-----------------|-----------------|-----------------|-----------------|-----------------|-------|-------|
| Haugh unit %                    | 72.73<sup>a</sup> | 82.24<sup>b</sup> | 80.55<sup>ab</sup> | 76.95<sup>b</sup> | 77.82<sup>b</sup> | 80.04<sup>ab</sup> | 1.76  | **    |
| Egg weight (g)                  | 10.89<sup>c</sup> | 12.43<sup>ab</sup> | 11.28<sup>b</sup> | 12.03<sup>a</sup> | 11.75<sup>b</sup> | 12.14<sup>ab</sup> | 0.58  | **    |
| weight %                        | 8.41<sup>a</sup> | 8.615<sup>a</sup> | 8.630<sup>a</sup> | 8.790<sup>a</sup> | 8.509<sup>a</sup> | 8.904<sup>a</sup> | 0.217 | NS    |
| Thickness (mm)                  | 0.224<sup>c</sup> | 0.305<sup>a</sup> | 0.276<sup>b</sup> | 0.290<sup>ab</sup> | 0.274<sup>b</sup> | 0.297<sup>a</sup> | 0.031 | **    |
| strength (g/cm<sup>2</sup>)     | 607<sup>b</sup> | 691<sup>a</sup>  | 689<sup>a</sup>   | 685<sup>a</sup>   | 699<sup>a</sup>   | 716<sup>a</sup>   | 21.6  |       |
| Total lipids (mg/yolk)          | 34.76<sup>b</sup> | 35.47<sup>a</sup> | 37.25<sup>a</sup> | 36.32<sup>a</sup> | 35.89<sup>ab</sup> | 36.93<sup>a</sup> | 0.69  |       |
| cholesterol (mg/yolk)           | 84.36<sup>a</sup> | 78.62<sup>b</sup> | 76.33<sup>b</sup> | 77.90<sup>b</sup> | 77.23<sup>b</sup> | 77.05<sup>b</sup> | 0.65  |       |
| MDA (µg/g yolk)                 | 0.23<sup>a</sup> | 0.16<sup>b</sup> | 0.14<sup>b</sup> | 0.13<sup>b</sup> | 0.17<sup>b</sup> | 0.14<sup>b</sup> | 0.022 |       |

T0= Control, T1= Optifeed® (500 g/ton feed), T2= Oleobiote® 100 g/T feed, T3= Force®6: 100 g/T, T4= VeO® premium 250 g/T feed and T5= (Optifeed®250g+Oleobiote®50g+ Force®6 (50 g)+VeO® premium 125 g)/Ton, MDA= malondialdehyde. The same superscripts within rows means non-significant<sup>a–c</sup>. Means within rows with different superscripts differ significantly (P≤ 0.05).

Results for red blood cells count (RBC), hemoglobin (Hb), packed cell volume (PCV), high density lipoprotein (HDL), Antioxidant activity (AOA) and glutathione peroxidase concentrations were significantly higher (P<0.05), as well as improvement was found in H/L ratio in the treatments of feed additives compared with the control. Whereas, total cholesterol (TCH), low density lipoprotein (LDL), very low density lipoprotein (VLDL) and malondialdehyde (MDA) concentrations were significantly lower (P<0.05) in the treatments of feed additives compared with the control (Table 3). Herbs and spice could be inhibit the nutrition stress or any stress that cause the increase in H/L ratio, because the stress could increase the stimulation of adrenal gland to produce some hormones such as estrone which has a direct effect to analyze a lymphatic cell which causes increase in H/L ratio (9).

Table 3. Effect of functional feed additives on some whole blood, serum biochemical and antioxidant traits of Japanese quails at age 70 day.

| Treatments | T0            | T1              | T2              | T3              | T4              | T5              | SEM   | S.L   |
|------------|---------------|-----------------|-----------------|-----------------|-----------------|-----------------|-------|-------|
| TRBC (10<sup>6</sup>cells/mm<sup>3</sup>) | 3.091<sup>b</sup> | 3.982<sup>a</sup> | 3.557<sup>ab</sup> | 3.849<sup>a</sup> | 3.625<sup>a</sup> | 3.977<sup>a</sup> | 0.13  | *     |
| Hb (g/dl)  | 11.72<sup>c</sup> | 15.33<sup>a</sup> | 14.10<sup>ab</sup> | 14.64<sup>ab</sup> | 13.48<sup>b</sup> | 15.58<sup>a</sup> | 0.49  | **    |
| PCV (%)    | 37.45<sup>b</sup> | 40.88<sup>a</sup> | 38.71<sup>ab</sup> | 41.00<sup>a</sup> | 39.43<sup>b</sup> | 41.63<sup>a</sup> | 0.75  |       |
| H/L ratio  | 0.478<sup>a</sup> | 0.279<sup>b</sup> | 0.238<sup>b</sup> | 0.288<sup>b</sup> | 0.301<sup>b</sup> | 0.225<sup>b</sup> | 0.0071 |       |
| TCH (mg/dl)| 183.3<sup>a</sup> | 161.5<sup>b</sup> | 143.2<sup>c</sup> | 153.2<sup>b</sup> | 155.2<sup>c</sup> | 148.4<sup>b</sup> | 1.81  | **    |
| HDL (mg/dl)| 31.88<sup>d</sup> | 41.72<sup>a</sup> | 40.82<sup>ab</sup> | 46.89<sup>b</sup> | 41.95<sup>d</sup> | 51.63<sup>a</sup> | 0.841 |       |
| LDL (mg/dl)| 117.3<sup>a</sup> | 90.18<sup>b</sup> | 72.93<sup>c</sup> | 85.74<sup>b</sup> | 82.76<sup>c</sup> | 73.82<sup>d</sup> | 1.08  |       |
| VLDL (mg/dl)| 34.12<sup>a</sup> | 29.60<sup>b</sup> | 22.25<sup>c</sup> | 20.57<sup>c</sup> | 30.49<sup>b</sup> | 22.95<sup>c</sup> | 0.36  |       |
| AOA (mmol/L)| 1.48<sup>b</sup> | 1.93<sup>a</sup> | 1.83<sup>a</sup> | 2.13<sup>a</sup> | 1.88<sup>a</sup> | 2.04<sup>a</sup> | 0.058 |       |
| GPx (mg/ml)| 74.69<sup>c</sup> | 81.52<sup>b</sup> | 86.91<sup>ab</sup> | 89.13<sup>a</sup> | 86.77<sup>ab</sup> | 88.23<sup>a</sup> | 0.82  |       |
| MDA (µmol/ml)| 3.37<sup>b</sup> | 2.03<sup>b</sup> | 2.35<sup>ab</sup> | 1.52<sup>d</sup> | 2.27<sup>b</sup> | 1.94<sup>b</sup> | 0.04  |       |

T0= Control, T1= Optifeed® (500 g/ton feed), T2= Oleobiote®100 g/T feed, T3= Force®6: 100 g/T, T4= VeO® premium 250 g/T feed and T5= (Optifeed®250g+Oleobiote®50g+ Force®6 (50 g)+VeO® premium 125 g)/Ton, TRBC= total red blood cells count, Hb= hemoglobin, PCV= packed cell volume, TCH= total cholesterol, HDL= high density lipoprotein, LDL= low density lipoprotein, AOA= Antioxidant activity, GPx= glutathione peroxidase, VLDL= very low density lipoprotein, MDA= malondialdehyde.<sup>a–d</sup> Means within rows with different superscripts differ significantly (P≤ 0.05).

Many active components of herbs and spices can prevent lipid peroxidation through quenching free radicals or through activation of antioxidant enzymes like superoxide dismutase, catalase, glutathione peroxidase and glutathione reductase. Main molecules responsible for the antioxidative properties of herbs and spices are phenolic substances (flavonoids, hydrolysable tannins, phenolic acids, phenolic terpenes) and some vitamins (E, C and A). Often used herbs rich in phenolics are: rosemary, thyme, oregano, sage, green tea, chamomile, etc. (3, 5, 7). It has been hypothesized that the increased de-conjugation of bile acids, results in greater excretion of bile acids from the intestinal tract, the increased excretion of bile acids stimulates the replacement of bile acids from cholesterol.
The results of hatched eggs are summarized in (Table 4) the treatments of feed additives were significantly (P<0.05) higher in the percentages of fertility, hatchability of fertile egg, hatchability of total egg and hatched chicks weight compared to the control. Whereas; feed additives treatments were significantly (P<0.05) lower in the percentages of early dead embryos (1-7) d, mid dead embryos (8-14) d, late dead embryos (15-17) d, total dead embryos (1-17) d and disabled chicks compared to the control.

Table 4. Effect of functional feed additives on hatching traits of Japanese quails at age 70 days.

| Traits                         | T0     | T1     | T2     | T3     | T4     | T5     | SEM     | S.L     |
|-------------------------------|--------|--------|--------|--------|--------|--------|---------|---------|
| Fertility %                   | 83.45a | 94.68a | 88.77a | 93.25a | 90.51a | 93.70a | 1.83**  |         |
| Hatchability of fertile egg % | 74.16a | 90.25a | 83.09b | 89.05ab| 84.36b | 89.62a | 1.08**  |         |
| Hatchability of total egg %   | 71.31c | 89.50b | 81.76b | 88.30b | 82.81b | 88.62a | 1.22**  |         |
| Early dead embryo (1-7) d %   | 4.72a  | 2.33b  | 2.78b  | 1.95b  | 2.65b  | 1.75b  | 0.087   |         |
| Mid dead embryo (8-14) d %    | 2.67a  | 1.35b  | 1.40b  | 1.25b  | 2.00b  | 1.33b  | 0.024   |         |
| Late dead embryo (15-17) d %  | 1.90a  | 0.75b  | 1.50b  | 1.00b  | 1.50b  | 1.00b  | 0.015   |         |
| Total dead embryos (1-17) d % | 9.29a  | 4.43b  | 5.68b  | 4.20b  | 6.15b  | 4.08b  | 0.031   |         |
| Disabled chicks %             | 2.85a  | 0.75b  | 1.33b  | 0.75b  | 1.55b  | 1.00b  | 0.027   |         |
| Hatched chicks weight (g)     | 8.04b  | 8.85a  | 8.75a  | 8.93a  | 9.05a  | 0.055  |         |         |

T0= Control, T1= Optifeed® (500g/ton feed), T2= Oleobiotec®100g/T feed, T3= Force®6: 100 g/T, T4= VêO® premium 250 g/T feed and T5= (Optifeed®250g+ Oleobiotec®50g+ Force®6 (50 g)+VêO® premium 125 g)/Ton

a–b Means within rows with different superscripts differ significantly (P≤ 0.05).

The results of immunological ELISA titer against some diseases of hatched quail chicks in (Table 4) observed that the all treatments of functional feed additives were significantly higher (P<0.05) in Newcastle (ND), Gamboro (IBD) and Infectious bronchitis (IBV) diseases compared to the control T1. The immune system generally benefits from the herbs and spices rich in flavonoids, vitamin C and carotenoids. The plants containing molecules which possess immune-stimulatory properties, these plants can improve the activity of lymphocytes, macrophages and NK cells; they increase phagocytosis or stimulate the interpheron synthesis (4). Herbs feed additives have unusual components that provide unique health benefits by decreasing free radical and increasing the antioxidant capacity of the blood and improves the general healthy conditions of poultry that may be reflected in increased immune response (15).

Table 5. Effect of functional feed additives on immunological ELISA titer against some diseases of hatched quail chicks

| Traits    | T0     | T1     | T2     | T3     | T4     | T5     | SEM     | S.L     |
|-----------|--------|--------|--------|--------|--------|--------|---------|---------|
| ND (μg/ml)| 5437c  | 6722a  | 6909a  | 8011a  | 7215ab | 7983a  | 132**   |         |
| IBD (μg/ml)| 3167b | 4089a  | 4008a  | 4562a  | 4307a  | 4121a  | 122*    |         |
| IBV (μg/ml)| 1766c | 2124b  | 2178b  | 2339ab | 2417a  | 2295ab | 98**    |         |

T0= Control, T1= Optifeed® (500g/ton feed), T2= Oleobiotec®100g/T feed, T3= Force®6: 100 g/T, T4= VêO® premium 250 g/T feed and T5= (Optifeed®250g+ Oleobiotec®50g+ Force®6 (50 g)+VêO® premium 125 g)/Ton.

ND= Newcastle disease, IBD = Gamboro disease and IBV= Infectious bronchitis disease.

a–b Means within rows with different superscripts differ significantly (P≤ 0.05).

The results in this study showed that functional feed additives (500g Optifeed®, 100g Oleobiotec®, 100g Force®6, 250g VêO® premium and (250g Optifeed® + 50g Oleobiotec® + 50g Force®6 +125g VêO® premium) /Ton dietary treatments group gave the best performance in most measured parameters, benefit profit per quail, egg qualities (external and internal), improves some whole blood, serum biochemical and antioxidant traits of quail parameters. This finding also demonstrated for the effect of different feed additives of diets increased the fertility and hatchability and immunological ELISA titer against some diseases of hatched quail chicks tremendously.

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