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IMPACT OF PROPOLIS SUPPLEMENTIONS ON GROWTH PERFORMANCE AND PHYSIOLOGICAL RESPONSES IN JAPANESE QUAIL

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ABSTRACT: To investigate the response of feeding Japanese quail chicks to propolis supplementation on growth performance, blood constituents, hormones, immunity, antioxidant, carcass characteristics and bacterial count, one hundred and twenty, one-week-old Japanese quail chicks were divided to four dietary groups with three replicates (10 birds). The first group fed a basal diet, while the groups 2^{nd},3^{rd} and 4^{th} were fed with 200, 400 and 600 mg of propolis/kg diet until the age of six weeks. The results showed a significant improvement in feed intake, body weight, body weight gain and feed conversion with increasing of propolis supplementation. All treatments increased significantly ($P<0.01$) percentage of dressing and internal organs than the Control birds. Propolis supplementation significantly ($P<0.01$) increased blood components; Hb, RBC, PCV, WBCs, lymphocyte, total protein, globulin, albumin, glucose, high density lipoprotein, total antioxidant capacity, IgM, IgG, thyroid hormones ($x10^3$) as compared to control group and decreased total lipids, plasma cholesterol, LDL, ALT, AST. Moreover, propolis decreased the Intestinal total anaerobic, aerobic microflora counts and total coliform compared to control group. There was no effect on monocytes, eosinophils and neutrophils as compared to control group. The results showed that supplementation of propolis at levels of 200, 400 and 600 mg of propolis/kg feed improves growth performance, blood components, antioxidant indices, thyroid hormones, immune parameters and carcass characteristics.

Keywords: propolis - Japanese Quail- growth - blood - immunity.

INTRODUCTION

Studies are being conducted on alternative products that can promote growth, improve feed utilization, and maintain gut health (Zhang *et al*. 2005). One alternative might be to incorporate propolis into in bird diets. Propolis is a resin that honey bees (Apis mellifera caucasica) collect from buds and bark of certain trees and plants. (Moreno, *et al*. 2000; Dimov *et al*. 1991). Several of chemical compounds contained in Propolis like inorganic compounds, steroids, amino acids and polyphenols (phenolic aldehydes, aglycone, phenolic acids, alcohols, esters and ketones). Several biological properties, including antibacterial, antiviral, antifungal, antioxidant, hepatoprotective and immunostimulating activities of propolis have been reported and therefore propolis supplements are used as an additive in poultry feed (Tatli Seven, 2008).This has been confirmed by Mathivanan *et al*. (2013) that propolis has a beneficial effect on feed intake, conversion and daily gain in poultry. Also, beneficial effect against a variety of Gram-negative bacteria and some Gram-positive bacteria using propolis according to Velikova *et al*., 2000. It is clear that propolis can be considered as substitutional to the use of food antibiotics (Itavo *et al*., 2011). Indeed, several researchers (Cetin *et al*., 2010; Fischer *et al*., 2010) have demonstrated that propolis is capable of inducing immunological effects in animals via the synthesis of antibodies, affecting macrophage activation and lymphoid organ weight. Numerous studies have recorded the useful effect of propolis on the immune response and growth in poultry (Shalmany and Shivazad, 2006; Tatli Seven *et al*., 2008; Babaei *et al*., 2016). Positive effect of the supplemental propolis added to chick feed led to an improvement in growth performance as reported by (Sherif and El-Saadani 2017; El-Naggar and Abdel-Khalek 2019). Due to the lack of information about the use of propolis in growth feed for quail birds. The aim of this study is to determine the impact of propolis supplementation added to quail feed on growth performance and physiological responses in Japanese quail.

MATERIALS AND METHODS

The experiment was carried out on a private farm in El Kharga city, New Valley Governorate, Egypt from September to October 2018 .

Experimental Design

One hundred and twenty, Seven-day old chicks of Japanese Quails were having nearly equaled live weights and randomly distributed into four treatments groups housed in 3 replicate pens (each contained 10 chicks). Chicks were raised under similar managemental and hygienic conditions. Feed and water were supplied *ad libitum* throughout the experimental period which ended at 6 wks of age. The basal diet (control) has been formulated to meet the nutrient requirements of chicks and fed 23 % CP and 2800 Kcal. The composition of the basal diet is given in Table (1a) and the chemical composition of propolis recorded in Table (1b). Chicks in the first group were fed on a basal diet and considered as a control group, while the other three groups 2, 3 and 4 were fed on a basal diet supplemented with 200, 400 and 600 mg propolis/kg feed respectively .At the

beginning of the experiment (1 week) birds were individually weighed (g) and at the end of the experiment (6 weeks). Feed intake for each replicate, body weight gain (BWG) and feed conversion (FCR) were recorded as g feed/g gain.

At 6 weeks of age, five birds from each treatment were selected randomly, weighed and slaughtered for carcass dressing. The carcass organs weights (carcass, heart, liver, gizzard, spleen and intestinal) were expressed as percentage of the live weight. Intestinal anaerobic and aerobic microflora counts were determined. Total anaerobic count, total coliform count and aerobic plate count (APC) were carried out according to American Public Health Association (A.P.H.A, 1985). Ten blood samples were collected from each experimental group at the time of slaughter and divided into two parts. The first part was collected in heparin tubes while the second part was collected in non-heparin tubes to obtain serum. Fresh blood aliquots were used to determine hematological parameters [white blood cells (WBCs), red blood cells (RBCs), hemoglobin (Hb), and Packed cells volume (PCV)]. Serum was obtained from blood samples stored at 20 °C for later analysis. Blood biochemical parameters such as: total protein, albumin, globulin, total lipids, cholesterol, glucose concentration, low-density lipoprotein (LDL), high-density lipoprotein (HDL), alanine aminotransferase ALT, aspartate aminotransferase (AST) and total antioxidant capacity TAC in blood serum were determined using a commercial kit (Biolabosa As. Frances). Blood hormones, thyroxine (T4), triiodothyronine (T3) and immune parameters; Immunoglobulin G (IgM) and immunoglobulin G (IgG) were determined by enzyme immunoassay using commercial kits.

Statistical analysis data

Statistical analysis data obtained from this study were analyzed using the Statistical Analysis System (SAS, 2002) general linear model procedure, using one-way ANOVA as in the following model: $Yik= \mu+ Ti + eik$ Where, Y is the dependent variable; μ is the general mean; T is the effect of experimental treatments; and e is the experimental random error. Significant differences between means were defined at $P<0.05$ compared using Duncan's multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

Growth performance and carcass

Propolis supplementation effect on feed intake, body weight, body weight gain, and feed conversion ratio for Japanese quail chicks are shown in Table (2). Propolis supplementation improved feed conversion ratio and increased significantly ($p< 0.01$) feed intake, body weight and body weight gain comparing to control.Supplementation of propolis at 200, 400 and 600 mg propolis/kg of diet led to an increase in body weight by 4.03, 8.85 and 18.9 %, feed intake by 1.66, 3.06 and 8.46 %, body weight gain by 5.05, 11.04 and 23.58 %, of the control group, respectively. Result showed that birds fed diet supplemented with 600 mg propolis/kg of diet had the higher feed intake, body weight, body weight gain and the best in feed conversion compared with the other dietary treatments. This improvement in the current study may be due to a reduction bacterial load causing disease in the intestine and an

improvement in the status of the intestinal lumen, resulting in increased absorption and utilization of nutrients. moreover, the high content of phenolic acids and flavonoids in propolis has positive effects on health and metabolism because it improves beneficial microbes in the gut (Viuda-Mattos *et al*., 2008). As pointed out by Moreno, *et a*l. 2000; Dimov *et al*. 1991. Several of chemical compounds contained in Propolis like inorganic compounds, steroids, amino acids and polyphenols (phenolic aldehydes, aglycone, phenolic acids, alcohols, esters and ketones).Many biological properties, including antibacterial, antiviral,antifungal, antioxidant, hepatoprotective and immunostimulating activities of propolis have been reported. The results are consistent with those reported by Sherif and El-Saadani (2017) which showed that Increasing propolis level increased body weight (BW) and body weight gain (BWG). On the other hand, El-Naggar and Abdel-Khaleq (2019) showed that ducklings fed diet supplemented with different levels of propolis had significantly greater BW, BWG and better feed conversion as compared to control.

Data presented in Table (3) showed that influence of dietary different levels of propolis supplementation on the relative weights of dressing, heart, liver, gizzard, spleen and intestinal. Results showed that relative weight of the internal organs of Japanese quail of treated groups were significantly ($p<0.01$) increased with increase of propolis level and higher than control group. In addition, the treated with 600 mg propolis/kg of diet had the best carcass weight compared with other dietary treatments. The improvement from dressings in propolis-supplemented treatments could be attributed to the increase in body weight at slaughter as it has been suggested that the high value of the weight of the living body is attracted and can be related to the physiological state of the high value of the carcass weight (Ojewole *et al*., 2000).This important production of immune cells may also be due to the antioxidant activity of propolis which is a rich source of vitamins (Moreira, 1986), enzymes (Khalil and El-Sheikh, 2010) and other biological components including fatty acids, amino acids and flavonoids. (Wagh, 2013) who showed that propolis can be used as a natural growth promoter in poultry (Attia *et al*., 2015).The results are consistent with those reported by Sherif and El-Saadany (2017) which showed that the relative carcass weight and lymphoid organ weights were significantly improved ($p < 0.01$) by increasing the level of propolis. Similar results were confirmed by Elnaggar and Abd El-khalek (2019) who showed that ducklings fed a basal diet supplemented with different levels of propolis resulted in a significant increase in the ratio of dressing and total edible parts compared to the control.

Blood indices

The data in Tables (4,5) show that different supplemental dietary levels of propolis have a significant ($P < 0.01$) increase in the hematological traits of quail parameters (Red Blood Cells , Hemoglobin (Hb),Packed cells volume (PCV), white blood cells (WBCs)) and lymphocyte compared to the control. Feeding diets with different levels of propolis supplement had a significant ($P<0.01$) increase in the blood

biochemical parameters of quail (total protein, albumin, globulin, glucose concentration, HDL and T_4, T_3 hormone while total serum lipids, cholesterol, LDL, ALT, and AST were significantly ($P < 0.01$) decreased compared to the control group. Moreover, antioxidant indices (total antioxidant capacity TAC) and immune indices (IgG), (IgM) were higher in birds of Japanese quail fed basal diets supplemented with different levels of propolis compared to the control group. Furthermore, no significant effects of different levels of propolis were detected on monocytes, eosinophils and neutrophils as compared to control group. Red blood cells are responsible for the manufacture of hemoglobin, and the transport of carbon dioxide and oxygen in the blood, so higher values indicate greater potential for these functions and better health (Olugbemi *et al*. 2010).

Our results are in agreement with that of Attia *et al*. (2014) which showed that it increases erythrocytes and hemoglobin by continuous or intermittent addition of propolis in broiler rations at 300 mg/kg. The same results for Sasso chickens obtained by Omar *et al*. (2014), Sherif and El-Saadani (2016) For laying hens. The beneficial effect of propolis on protein fractions may be due to the hepatoprotective effect which appears to preserve the body protein from degeneration and anabolic effect in favor of protein synthesis. In the current study, it may be that the improvement in protein fractions and globulin concentration that has been observed may be because chicks livers will be able to synthesize enough globulin for immune action that keeps the body protein from degeneration (Khalil, 2006). These results are consistent with the results of Abdel-Kareem and ElSheikh (2015) who showed that the addition of propolis in the layer ration (250, 500 and 1000 mg/kg diet) increases total protein, globulin and albumin.Similarly, propolis has shown positive effects on the immune response and antibodies in poultry (Yang *et al*., 2008; PopielaPleban *et al*., 2012). This may be attributed not only to the specific antibacterial effect of propolis with positive effects on metabolism (Aygun *et al*., 2012), but also to its antifungal (Sforcin, 2007), antiparasitic (Freitas *et al*., 2006), and antiviral (Gekker *et al*., 2005), immunomodulatory (Dimov *et al*., 1992), anti-inflammatory (Dobrowolski *et al*., 1991), and antioxidant (Krol *et al*., 1990) effects. These results agree in part with those of Kacániová *et al*. (2012). Additional evidence was found to improve the health of the presented chicken by lowering serum cholesterol, triglycerides, creatinine, urea and asparatate aminotransferase compared to the control, indicating improved kidney and liver function and lipid metabolism. This may be due to the effect of the propolis on the catabolic muscles. In addition, Omar *et al* (2002) found that the improvement in erythrocytes, hemoglobin, PCV and protein by propolis could be due to the direct effect of the anabolic action of the synthetic protein, which can protect the protein body from degeneration. The effect of propolis on plasma metabolites can be attributed to its vitamin, mineral and phospholipid contents (Leja *et al*., 2007) and its antioxidant effects (Šarić *et al*., 2009). The effects of propolis on cholesterol are in agreement with those reported by Fuliang *et al*. (2005). This may be due to

the effect of propolis on lipid metabolism (Matsui *et al*., 2004), and there was a significant ($p < 0.01$) decrease in the liver enzymes, AST and ALT by increasing propolis in quail rations. Galal *et al*. (2008) noted the same finding that ALT and AST activities were significantly reduced by adding propolis to the diet layer at 100 and 150 mg/kg diets. Similarly, Abdel-Kareem and El-Sheikh (2015) found that when fed chickens with diet containing propolis at 250, 500 and 1000 mg/kg, liver enzymes (AST and ALT) decreased.The present results regarding the decreased activities of transaminases in the blood can be attributed to the high biological activity resulting from the addition of Propolis to its content of nutritional values that may prevent lipid oxidation. These results agreed with Cetin *et al*. (2010) and Freitas *et al*. (2011), who found an increase in the concentration of IgM and IgG due to the addition of propolis compared to the control group and the improvement in the immune status may be due to propolis because it contains flavonoid components that raise cytokines. These cytokines stimulate the activities of B lymphocytes that will be able to produce immunoglobulins (Fritas *et al*., 2011).

Our results were confirmed by Mahmoud *et al*. (2015) who found a significant increase in total antioxidant capacity (TAC) as a result of propolis supplementation in broiler rations of 250, 500 and 750 mg/kg, which may be due to their higher flavonoid conte. The same result was observed with Shreif and El-Saadany (2017) who showed that there is a positive effect of supplementing propolis to the chicks' diet, especially on the immune status, physiological status and antioxidants. Moreover, adding propolis/kg diet can improve the health of chicks. The same results were obtained by Elnaggar and Abd El-khalek (2019) for ducklings.

Bacteria count

The results of the intestinal microbial count of Japanese quail are presented in Table (6). All propolis levels led to a decrease in the total anaerobic count, total aerobic count and total microform compared with the control group. In this study, the beneficial effect on the number of gut microbes may be due to the addition of propolis to bird feed because it contains flavonoid and phenol components, as well as to its antimicrobial activity (Tatli Seven *et al*., 2009). Moreover, the ability of propolis can be explained by the strong antibacterial effect and its content of micro-nutrients and beneficial which may have positive effects on the health of birds (Canogullari *et al*., 2009; Shreif and El-Saadany, 2017; Elnaggar and Abd El-khalek, 2019).

CONCLUSION

It can conclusion using propolis for growing Japanese quail diet could be highly benfecal and improves performance, blood components, hormones, immunity parameter, antioxidant indices and carcass characteristics, therefore, propolis could be an available feed additives supplementation with 600 mg /kg will gine the quail diet effect**.**

Table (1a): Composition of the experimental diet- through the growing period.

Ingredients	Percentage in diet
Corn	50.70
Soybean meal	42.52
Vegetable oil	2.00
Di-Calcium phosphate	0.72
Calcium carbonate	1.25
Salt	0.33
Vitamin premix1	0.25
Mineral premix2	0.25
DL-methionine	0.13
L-Threonine	0.11
Sand	1.74
Nutrient composition	
Crude protein (%)	23.00
Metabolizable Energy (Kcal kg-1)	2800
Available phosphorus (%)	0.29
Calcium (%)	0.77
Sodium (%)	0.15
Methionine (%)	0.48
Threonine (%)	0.98
Methionine + Cysteine (%)	0.85
Lysine (%)	1.28

1 Provides per kg of diet, Vitamin A: 3125 µg, Vitamin K3: 5 mg, Vitamin D3: 75 µg, α-tocopherol acetate: 50 mg, Vitamin B1: 3 mg, Vitamin B2: 6 mg, Vitamin B6: 5 mg, Vitamin B12: 0.003 mg, Niacin: 50 mg, Folic acid: 1 mg, Pantothenic acid: 10 mg, 12Biotin: 0.1 mg. 2 Provides per kg of diet, Cu: 5 mg, I: 2 mg, Co: 0.5 mg, Se: 0.15 mg, Mn: 90 mg, Fe: 50 mg, Zn: 70 mg.

Table (1b): The major compounds of Egyptian propolis

Proximate analysis of propolis	
Crude protein	1.9
Carbohydrates	1.7
Ash	4.1
Fat	1.4
Essential oils	4.1
Stearic%	7.2
Palmitic%	12.9
Oleic%	13.3
Linolenic%	0.79
Linoleic%	1.9
Palmitoleic%	9.1
Flavonoids (Total) %	27.9

Table (2): Effect of propolis on body weight, body weight gain, feed intake and feed conversion of Japanese quail.

Treatments / **Traits**	**control**	**Propolis 200 mg/kg feed**	**Propolis 400 mg/kg feed**	**Propolis 600 mg/kg feed**	**Pooled SEM**	**P value**
Initial weight (g)	37.14	37.13	37.04	37.03	0.14	0.895
Final body weight (g)	192.12 ^{d}	212.01 ^{c}	230.96 ^{b}	251.06 ^{a}	0.24	0.001
Feed intake (g)	560.84 ^{d}	598.43 ^{c}	617.03 ^{b}	637.84 ^{a}	1.22	0.001
Body weight gain (g)	154.97 ^{d}	174.87 ^{c}	193.92 ^{b}	214.02 ^{a}	0.19	0.001
Feed conversion(g feed/g gain)	3.62 ^{a}	3.42 ^{b}	3.18 ^{c}	2.98 ^{d}	0.01	0.001

a,b,c. means with the different letters in the same row are significantly different ($P \leq 0.01$) ;
SEM= Standard error of means.

Treatments / Traits	control	Propolis 200 mg/kg feed	Propolis 400 mg/kg feed	Propolis 600 mg/kg feed	Pooled SEM	P value
Dressing (%)	64.73 ^{c}	69.84 ^{b}	73.56 ^{a}	75.28 ^{a}	0.59	0.001
Heart %	0.79 ^{d}	0.81 ^{c}	0.83 ^{b}	0.85 ^{a}	0.01	0.001
Liver %	1.77 ^{d}	1.96 ^{c}	2.02 ^{b}	2.18 ^{a}	0.01	0.001
Gizzard %	1.85 ^{d}	1.96 ^{c}	2.23 ^{b}	2.58 ^{a}	0.01	0.001
Spleen (%)	0.043 ^{d}	0.045 ^{c}	0.053 ^{b}	0.055 ^{a}	0.0004	0.001
Intestinal %	2.94 ^{c}	2.98 ^{b}	2.98 ^{b}	3.06 ^{a}	0.01	0.001

Table (3): Effect of propolis on some relative carcass characteristics of Japanese quail

a,b,c. means with the different letters in the same row are significantly different (P≤0.01) ; SEM= Standard error of means

Table (4): Effect of propolis on some hematological blood, antioxidant indices and immune indices of Japanese quail.

Treatments / Traits	control	Propolis 200 mg/kg feed	Propolis 400 mg/kg feed	Propolis 600 mg/kg feed	Pooled SEM	P value
RBC(106/mm3)	3.23 ^{d}	3.42 ^{c}	4.01 ^{b}	4.06 ^{a}	0.01	0.001
HB(g/dl)	16.40 ^{d}	17.00 ^{c}	18.23 ^{b}	19.37 ^{a}	0.14	0.001
PCV%	36.63 ^{d}	39.13 ^{c}	42.17 ^{b}	44.30 ^{a}	0.17	0.001
WBC(103/mm3)	41.45 ^{c}	43.39 ^{b}	44.69 ^{ab}	45.09 ^{a}	0.30	0.001
Lymphocytes, (%)	51.77 ^{b}	51.83 ^{b}	52.33 ^{b}	53.27 ^{a}	0.26	0.022
Neutrophils (%)	35.53	35.37	34.87	33.96	0.41	0.078
Eosinophils, (%)	2.93	2.93	2.83	2.67	0.07	0.137
Monocytes, (%)	9.77	9.87	9.97	10.10	0.21	0.657
TAC(mg/dl)	413.21 ^{c}	415.59 ^{c}	422.83 ^{b}	430.50 ^{a}	0.73	0.001
IgG (mg/100 ml)	818.53 ^{c}	820.33 ^{bc}	822.90 ^{b}	834.96 ^{a}	0.84	0.001
Igm (mg/100 ml)	224.42 ^{d}	228.30 ^{c}	236.36 ^{b}	247.30 ^{a}	0.94	0.001

a,b,c. means with the different letters in the same row are significantly different (P≤0.01) ; SEM= Standard error of means;GPX = glutathione peroxidase; GSH = glutathione; SOD = superoxide dismutase; TAC= total antioxidant capacity; Immunoglobulin G (IgG), IgM= Immunoglobulin.

Table (5): Effect of propolis on some blood constituents of Japanese quail.

Treatments / Traits	control	Propolis 200 mg/kg feed	Propolis 400 mg/kg feed	Propolis 600 mg/kg feed	Pooled SEM	P value
Total protein (g/dl)	3.57 ^{d}	3.76 ^{c}	4.22 ^{b}	4.37 ^{a}	0.01	0.001
Albumin (g/dl)	1.64 ^{d}	1.75 ^{c}	2.09 ^{b}	2.11 ^{a}	0.01	0.001
Globulin (g/dl)	1.94 ^{d}	2.01 ^{c}	2.13 ^{b}	2.26 ^{a}	0.01	0.001
Glucose, (mg/dl)	176.66 ^{d}	184.33 ^{c}	191.67 ^{b}	199.00 ^{a}	1.03	0.001
Total lipids (mg/dl)	297.33 ^{a}	286.33 ^{b}	276.33 ^{c}	267.00 ^{d}	1.09	0.001
Cholesterol (mg/dl)	208.00 ^{a}	203.33 ^{b}	201.33 ^{b}	189.33 ^{c}	0.88	0.001
HDL (mg/dl)	67.33 ^{b}	74.67 ^{c}	80.33 ^{b}	87.67 ^{a}	0.96	0.001
LDL(mg/dl)	118.33 ^{a}	110.00 ^{b}	102.67 ^{c}	95.33 ^{d}	0.88	0.001
AST (U/L)	55.67 ^{a}	47.67 ^{b}	44.33 ^{c}	41.00 ^{d}	0.80	0.001
ALT (U/L)	20.90 ^{a}	19.40 ^{b}	18.23 ^{c}	17.70 ^{c}	0.28	0.001
T4 (ng/ml)	8.07	8.63	9.73	10.50	0.10301	0.001
T3, (ng/ml)	1.98 ^{d}	2.02 ^{c}	2.09 ^{b}	2.32 ^{a}	0.01	0.001

a,b,c. means with the different letters in the same row are significantly different ($P \leq 0.01$) ;
SEM= Standard error of means

Table (6): Effect of propolis on on total anaerobic, aerobic count and total coliform of bacteria in intestine of Japanese quail.

Treatments / Traits	control	Propolis 200 mg/kg feed	Propolis 400 mg/kg feed	Propolis 600 mg/kg feed	Pooled SEM	P value
Total anaerobic count $x10^2$	0.80 ^{a}	0.67 ^{a}	0.36 ^{b}	0.27 ^{b}	0.05	0.002
Aerobic plate count $x10^3$	4.37 ^{a}	2.70 ^{b}	2.46 ^{b}	1.30 ^{c}	0.12	0.001
Total coliform Count $x10^3$	20.00 ^{a}	14.67 ^{b}	9.33 ^{c}	3.00 ^{d}	0.68	0.001

a,b,c. means with the different letters in the same row are significantly different ($P \leq 0.01$) ;
SEM= Standard error of means

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propolis - Japanese Quail- growth - blood - immunity.

الملخص العربى

تاثير اضافة البروبوليس على أداء النمو والاستجابات الفسيولوجية في السمان الياباني

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تمت دراسة تأثيراضافة البروبوليس لاعلاف السمان اليابانى على اداء النمو وبعض مكونات الدم والهرمونات والمناعه والخصائص المضادة للأكسدة والذبيحة والعد البكتيرى، تم تقسيم مائة وعشرين كتكوت من السمان الياباني عمراسبوع إلى أربع مجموعات غذائية ، كل مجموعة تحتوى على ٣مكررات بكل مكرر ١٠ كتاكيت. كانت المجموعة الأولى تتغذى على العليقة الاساسية ، بينما تم تغذية المجموعات الثانية والثالثة والرابعة بـ ٢٠٠ و ٤٠٠ و ٦٠٠ مجم من البروبوليس / كجم علف حتى عمر ٦ اسابيع. اوضحت النتائج تحسن معنوي في تناول العلف ووزن الجسم ووزن الجسم المكتسب وتحويل العلف بزياده مستوى البروبوليس فى العلف. اضافه البروبوليس ادى الى زياده معنويه فى نسبه الذبيحه كما تحسنت نسب الاعضاء الداخلية للسمان اليابانى، كما سجلت المعاملات زياده معنوية فى صفات الدم : عدد كرات الدم الحمراء والهيموجلوبين ونسبة المكونات الخلوية للدم (الهيماتوكريت) وعدد كرات الدم البيضاء ونسبه خلايا lymphocyte وبروتينات الدم الكلي والالبيومين والجلوبيولين وجلوكوز الدم و HDL وإنزيمات الاكسدة TAC وجلوبيولينات المناعه IgM و IgG وهرمون الغده الدرقيه T3و T4 وانخفضت الدهون الكليه و نسبه الكوليسترول و LDL وانزيمات الكبد AST و ALT علاوة على ذلك ، قلل البروبوليس من مجموع البكتيريا اللاهوائية والهوائية وبكتيريا الكوليفورم مقارنة بمجموعة الكنترول. لم يكن هناك تأثير على نسبه خلايا monocytes و eosinophils و neutrophils مقارنة بمجموعة الكنترول.
اوضحت النتائج أن إضافة البروبوليس بمستويات ٢٠٠ و ٤٠٠ و ٦٠٠ ملجم من البروبوليس / كجم علف حسن أداء النمو ومكونات الدم و مضادات الأكسدة وهرمونات الغدة الدرقية والاستجابه المناعية وخصائص الذبيحة.