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EFFECT OF DIETARY SUPPLEMENTATION OF POMEGRANATE PEEL POWDER AND BUTYLATED HYDROXY TOLUENE ON SOME PRODUCTIVE, PHYSIOLOGICAL AND IMMUNOLOGICAL PARAMETERS OF JAPANESE QUAIL

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SUMMARY

This experiment aimed to study the effect of pomegranate peel powder (PPP) and butylated hydroxy toluene (BHT) on some productive, physiological and immunological parameters of Japanese quails. One hundred and eighty Japanese quails aged 11 weeks old were randomly divided into four treated groups, each group contain 3 replicates 15 bird each. The first group was fed control diet without any supplementation. While, the second and third groups were fed on diet containing 10 and 15 g PPP/kg respectively, and the fourth group were fed on diet containing 125 g/ton BHT. The results indicated that: 1- Pomegranate peel treatments powder significantly increased body weight gain, while, feed intake and feed conversion were significantly lower in all treated groups compared with control group. 2-Egg production traits (egg number, egg weight and egg mass) were significantly increased in PPP and BHT supplemented groups compared to control. 3-Egg shell weight was significantly higher in treated groups. While, yolk diameter was significantly longer in control than treated groups. While, yolk color and albumin were higher at (22 wks. of age). 4-Plasma total lipid TP, Cholesterol, low density lipoprotein LDL and high density lipoprotein HDL, creatinine, uric acid, AST and ALT significantly decreased by PPP addition .While, plasma TP was significantly increased in PPP and BHT treated groups. 5-Deftherdbody weight after slaughter was significantly increased in groups fed on 10 PPP g/kg diet. While, heart percentage of weight was significantly lowered in quails supplemented with 15 g PPP /kg diet. 6- Supplementation of PPP and BHT didn't affect the total micro bacterial count in instant. 7-Total phenolicswere significantly increased. While, the TBARS values was significantly decreased in birds fed 15 g PPP/kg compared with that received BHT supplemented diet and the control group. In conclusion, PPP addition by 15g PPP/kg diet can improve productive and physiological parameters and also can extend meat shelf-life.

Keywords: Butylated hydroxy toluene - Japanese quails- pomegranate peel powder

INTRODUCTION

Antioxidants play a major role in poultry performance due to the prevention of oxidative destruction of dietary fats and to provide enhanced protection for the longer chain polyunsaturated fatty acid. Many studied investigate the waste of pomegranate different parts as peel, seeds and leaf extracts the antioxidant capacity was respectively 55.3, 35.7 and 16.4%. Therefore, it seems that high antifungal and antioxidant activity of peel and seeds of pomegranate due to the high percentage of phenolic compounds in these plant parts extracts (Salahvarzi *et al*., 2011). In addition, pomegranate peel extract with an abundance of flavonoids and tannins has been shown to have a high antioxidant activity (Abdel Moneim *et al*., 2011). Pomegranate peel extract have more potential as a health supplement rich in natural antioxidants than the pulp extract (Li *et al*., 2006). Negi *et al*. (2003) studied the polyphenolic compounds in pomegranate peel (PP) as it is a natural source of such as ellagic tannins, ellagic acid and gallic acid. Negi and Jayaprakasha (2003) studied the action of phenolics as an antioxidant by donating electrons and reacting with free radicals to convert them to more stable products and terminate free radical chain reactions. Some studies showed the medical effect of pome granate on immune system Gracious Ross *et al*. (2011) found that 100 mg/kg Punic agranatum fruit rind powder (PGFRP) orally stimulate the cell-mediated and humoral components and increasing antibody titer to typhoid-H antigenin rabbits. Also, inhibit antimicrobial activity Yahia *et al*. (2011).

Synthetic antioxidants have been widely used as food or feed preservatives because oft heir low cost and effectiveness. The most common used synthetic antioxidants, butylated hydroxyl anisole (BHA) and butylated hydroxyl toluene (BHT). European Food Safety Authority, (2012) mentioned that BHT (E 321) authorised as a food additive in the EU that was previously evaluated by the EU Scientific Committee for Food (SCF) in 1987 and the Joint FAO/WHO Expert

Committee on Food Additives (JECFA) several times, the latest in 1996.

Natural phenolic antioxidants in vegetables and fruits had strong activity and low toxicity compared with those of synthetic phenolic antioxidants, such as butylated hydroxyl toluene (BHT) (Marinova and Yanishlieva, 1997).

The main target of this study was to evaluate the effect of adding pomegranate peel powder (PPP) as natural antioxidants compared with BHT as a synthetic antioxidant to Japanese quails diets on productive performance, egg quality traits, physiological and immunological parameters.

MATERIAL AND METHODS

Experimental birds and treatments:

The present study was carried out at Fayiom Animal Research Station, Animal Production Research Institute, Ministry of Agriculture, Egypt. The chemical analyses were carried out at Laboratories of the Animal Production Research Institute (APRI), Ministry of Agriculture, Egypt. Ground PPP was obtained from Dokki Market, Giza Governorate, Egypt. The PPP was obtained in dried form with moisture content of 9-10%. Butylated Hydroxy Toluene (BHT) was obtained from poultry breeding laboratory (APRI). A total number of 180 one day old Japanese quails were randomly divided into four treated groups, each group contain 3 replicates 15 bird each. The first was served as control. While, the second and third groups were fed on diet containing 10 and 15 g ppp/kg and the fourth group were fed on diet containing 125 g/ton Butylated Hydroxy Toluene (BHT) as shown in Table (1).All birds were fed ad.libitum, body weight and feed intake were measured daily, data on Table (2) showed the chemical composition of pomegranate peel (dried and fresh base).

Table 1. Composition and calculated analysis of the quail's diet

Ingredients	Treatments			
	T1 (Control)	T2(10ppp)	T3(15ppp)	T4(BHT)
Yellow corn	58.45	58.45	58.45	58.45
Soyabeen meal 44%	25.80	25.80	25.80	25.80
Corn glutten meal 62%	6.70	6.70	6.70	6.70
Vegetable oil	1.30	1.30	1.30	1.30
Dicalcium phosphate	1.10	1.10	1.10	1.10
Limestone	5.70	5.70	5.70	5.70
Common salt (NaCl)	0.34	0.34	0.34	0.34
Premix/kg**	0.30	0.30	0.30	0.30
dl-Methionine	0.05	0.05	0.05	0.05
L-Lysine	0.06	0.06	0.06	0.06
Choline chloride	0.20	0.20	0.20	0.20
Pomegranate peel powder (g/kg)	___	10.00	15.00	___
Butylated Hydrox Toluene (BHT) g/ton	___	___	___	125
CALCULATED analysis*				
C.P%	20.01			
ME (kcal/kg)	2890			
C.A%	2.5			
C.F%	3.36			
A.v phosphorus	0.35			

****premix:** Supplied per kg diet: Vit. A, 7040 IU; Vit.D3, 2000 IU; Vit. E, 8.8 IU; Vit. K3, 1.76 mg; Biotin, 0.12 mg; Thiamine, 1.2 mg; Riboflavin, 3.2 mg; Pantothenic acid, 6.4 mg; Pyridoxine, 1.97 mg; Niacin, 28 mg; Vit. B12, 0.008 mg; Choline, 320 mg; Folic acid, 0.38 mg; Mn, 60 mg; Fe, 60 mg; Zn, 51.74 mg; Cu, 4.8 mg; I, 0.69 mg; Se, 0.16 mg. * The values were calculated from NRC (1994).

Table 2: Chemical composition of pomegranate peel (g/kg DM, except DM g/kg fresh base)

	DM	CP	NDF	ADF	ASH
1-Fresh pp	962	36	208	151	54
2-Dried pp%	94.7	3.37	18.2	12.6	4

where: PP= pomegranate peel, DM= dry matter, CP= crude protein , NDF= neutral detergent fiber ADF= acid detergent fiber. 1- (Mirzaei-Aghsaghali *et al*, 2011), 2- (Taher-Maddah *et al*., 2012)

Productive performance:

Body weight and feed consumption were measured biweekly; while mortality rate were recorded daily. Body weight gain, feed conversion (g feed intake/g body weight gain) and mortality rate percent were calculated at the end of the experiment.

Egg production and egg quality traits:

Egg number were recorded daily and weighed to calculate egg mass throughout the experiment egg mass= average egg number/ day X average egg weight (g). A total of 24 eggs (3 eggs/treatment) were collected from every treatment at two periods of time (16 and 22 weeks of age) to estimate egg quality parameters in poultry breeding laboratory as egg weight (g), egg length (cm), egg diameter (mm), yolk weight (g), yolk diameter (mm), yolk color, albumin %, shell weight (g) and shell thickness (mm). Albumen and yolk heights and widths were measured for each egg. Then yolk index was calculated using the following formula according to Romanoff and Romanoff (1949): YI= YH / YD × 100

Where: YI: yolk index. YH: yolkheight. YD: yolk diameter.

Blood samples and physiological parameters:

Three birds from each treatment at the end of the experiment were randomly chosen in order to slaughter, blood were collected in heparinized tubes and centrifuged at 3000 rpm for 15 minutes and stored frozen at -20 ºC until the time to analysis. Blood plasma glucose (mg/dl), total protein (mg/dl), cholesterol (mg/dl) were determined according to Richmond (1973) , LDL (mg/dl), HDL (mg/dl) , Aspartate Amino Transferase AST (U/ml), Alanine Amino Transferase ALT (U/ml) were assayed by the method of Reitman and Frankel (1957) , TAOC (mm/l), uric acid and creatinine (mg/dl) were determined using chemical kits at poultry breeding laboratory according to Aggoor *et al*. (2000).

After slaughtering birds were weighed then defthired and weighed after that the carcass weight were measured. Liver, spleen, intestine and heart were measured byelectric scale and relative weights were calculated as a percentage of the life body weight.

Microbiological analyses:

A total number of 12 samples from the small intestine were taken. The small intestine was tied with cotton third in two different places and cut it between the tided area then resaved in saline wateruntil it reach to microbiology laboratory at animal health research institute. The samples were plated for enumeration of total bacterial count (TBC), Coliform counts, and *Enterobacteriaceae* counts (Gilbert *et al*, 2000). All the samples were evaluated using 3M™ Petrifilm™ total bacterial count plates, coliform count plates, and *Enterobacteriaceae* count plates (3M Microbiology, St. Paul, MN, USA) according to the instructions of the manufacturer. Intestine samples were after that diluted by sterile distilled water (typically from 1:50 to 1:200) to enable colony enumeration. The diluted intestine sample was transferred to a sterile 3 M filtered homogenizer bag and homogenized for three minutes using a Stomacher machine (PBI International, Milan, Italy). The contents were allowed to settle, and then 1 mL of the liquid suspension was plated onto the appropriate Petrifilm™ according to the instructions of the manufacturer. The plates were incubated in stacks of no more than 20 plates at 35±1 °C for 48 hours for the aerobic count plates and for 24 hours for coliform and *enterobacteriaceae*count plates.

Thiobarbituric acid reactive substances (TBARS) measurement:

The 2-thiobarbituric acid test (TBA) was used to determine the extent of lipid oxidation in samples. The method for analysis was described by Du and Ahn (2002) as follow: five grams sample was weighed into a 50 ml test tube and homogenize (type PT 10/35) 13 for at the highest speed. One milliliter of the homogenate meat was transferred to test tube (3x100 mm), and butylated hydroxyanisole (50 µl; 7.2%) and TBA-trichloroacetic acid (TCA) (15 mm TBA-15% 2ml TCA) were added. The mixture was vortexed, incubated in a boiling water bath for 15 min to develop color, then cooled in cold water for 10 min, vortexed again, and centrifuged for 15 min at 2.500 xg. The absorbance of the resulting supernatant solution was determined at 531 nm against a blank containing 1 ml of deionized distilled water (DDW) and 2 ml of TBA-TCA solution. The amounts of TBA-reactive substance (TBARS) were expressed as milligrams of malonaldehyde per kilogram of meat.

Total phenolics:

Samples were analyzed for total phenolic using the Folin-Ciocalteus (F-C) assay (Escarpa and Gonzalez, 2001) with slight modifications. Five gram of cooked patty was homogenized with 25 ml of 70% acetone and kept overnight for extraction at refrigeration temperature. Suitable aliquots of extracts were taken in a test tube and the volume was made to 0.5 ml with distilled water followed by the addition of 0.25 ml F-C (1N) reagent and 1.25 ml sodium carbonate solution (20%). The tubes were vortexed and the absorbance recorded at 725 nm by using spectrophotometer (model: UV-VIS 5704 SS, ECIL, Hyderabad, India) after 40 min. The amount of total phenolics was calculated as tannic acid equivalent from the calibration curve using standard acid solution (0.1 mg/ml).

Statistical analysis:

The data were statistically analyzed using the general linear model procedure described by

Costate, (1986). The linear model included the main effect of PPP or BHT and age.

$Yij=\mu + Ai+Tj+eij$

Where: Y= response variable μ= overall mean Ai= age Tj= treatments eij= error, normally distributed. The statistical significant was declared at ($P\leq0.05$). Differences among means were tested using Duncun`s multiple range test (Duncun, 1955).

RESULT AND DISSECTION

From the data in Table (3) it could be observed that birds fed on diet supplemented with 10 and 15 g PPP/kg diet had significant ($P\leq0.01$) increase body weight gain (BWG) 267.7, 271.3 (g) compared to BHT supplemented diet and the control group 264 (g) respectively. While, there were a significant ($P\leq0.05$) decrease in feed consumption (Fi) and feed conversion (Fc) in all treated groups. However, Saki *et al*. (2014) mentioned that there were no significant increase onBWG, Fi and Fc to laying hensfed on pomegranate seed pulp (PSP) but it had higher BWG than the control group. Similar to Rajani *et al*. (2011) who found that broiler BW is not affected by pomegranate peel treatment. Treated normal rats with pomegranate peel extract resulted in non-significant increase in body weight after4-weeks (Enas, 2004). On the contrary, Lin *et al*., (1989) and Wang *et al*. (1997) who noted that BHA/BHT and ethoxyquin had significant increase in body weight of broilers. Mortality rate percent was lowerd in significantly in birds diets supplemented with15g PPP/kg diet and BHT compared with groups fed on 10g PPP/kg diet and the control. This result is in agreement with Rajani *et al*. (2011) who found that antioxidant feed additives reduced ascites mortality in comparison to control group ($P\leq0.05$).

Table 3. Productive performance as affected by addition of pomegranate peel powder (PPP) and butylated hydroxyle toluene (BHT) during the experiment period

Items	T1	T2	T3	T4	Sig.
	Initial body weight(11 wk)				
	257± 3.34	257 ±3.33	256 ±3.34	256 ±3.34	NS
	12-13 wks				
Bw (g)	260±0.578	261±0.581	262±0.560	260±0.545	NS
Fi(g/bird)	490.3^{a}±0.334	477^{b}±0.578	460.67d±0.333	467.67^{c}±0.340	***
Fc(gfeed/gain)	1.89^{a}±0.005	1.83^{b}±0.003	1.76^{d}±0.004	1.80^{c}±0.004	***
	14-15 wks				
Bw (g)	260.3^{b}±0.334	263^{b}±0.578	271^{a}±2.335	263^{b}±0.578	**
Fi(g/bird)	503^{b}±0.578	512^{a}±0.578	502.67^{b}±1.730	494.66^{c}±1.730	***
Fc(gfeed/gain)	1.93^{a}±0.003	1.95^{a}±0.004	1.85^{c}±0.016	1.88^{b}±0.003	***
	16-17 wks				
Bw (g)	264.67^{c}±0.334	269^{b}±0.579	273^{a}±1.01	266^{c}±0.576	***
Fi(g/bird)	612^{a}±0.578	585.7^{b}±0.046	562.3^{c}±0.879	552^{d}±0.579	***
Fc(gfeed/gain)	2.31^{a}±0.003	2.18^{b}±0.004	2.06^{d}±0.004	2.08^{c}±00003	***
	18-19 wks				
Bw (g)	265.33^{d}±0.303	270.3^{b}±0.345	274.2^{a}±0.334	267^{c}±0.578	***
Fi(g/bird)	689^{a}±0.578	681^{b}±0.577	665^{c}±1.730	663^{d}±1.734	***
Fc(gfeed/gain)	2.60^{a}±0.004	2.52^{b}±0.003	2.43^{d}±0.004	2.49^{c}±0.003	***
	20-22wks				
Bw (g)	270^{b}± 0.578	275^{a} ±0.330	276.3^{a} ±0.329	268^{c} ±0.329	***
Fi(g/bird)	720.3±0.879	717.7±0.329	652.0±33.538	688.3±0.334	NS
Fc(gfeed/gain)	2.67^{a}±0.003	2.61^{a}±0.004	2.36^{b}±0.004	2.57^{a}±0.003	*
	11-22 wks				
Bw (g)	264.04^{C}±1.283	267.7^{B}±1.853	271.3^{A}±2.257	264.9^{C}±1.223	**
Fi(g/bird)	602.9^{A}±0.689	594.7^{B}±0.522	568.5^{D}±7.64	573.13^{C}±1.024	***
Fc(gfeed/gain)	2.28^{A}±0.010	2.22^{B}±0.009	2.10^{D}±1.002	2.16^{C}±0.002	***
Mortality%	4.44	4.44	2.22	2.22	

a-d treatments within the (row-wise) with different superscripts are significantly different ($p\leq 0.05$). A-D within the same time (column-wise) with different superscripts are significantly different ($p \leq 0.05$); each value is a mean SE of three replicates.Bw= body weight (g), FI= feed intake (g), FC= feed conversion (g)

Data in Table (4) concluded that there were significant effects on yolk diameter and shell weight. Internal egg quality from birds receiving BHT contained a significantly ($P\leq0.005$) lower of yolk diameter. While, it was significantly ($P\leq0.005$) higher in shell weight than the control group and the other experimental groups.

Table 4. Egg quality as affected by addition of pomegranate peel powder (PPP) and butylated hydroxyle toluene (BHT) during 2 periods of time (16 & 21 wks of age) and their interactions

Egg quality	T1	T2	T3	T4	Overall mean	Sig.
External egg quality:						
Egg weight 16wks (g)	13.04 ± 0.671	12.64 ± 0.965	13.39 ± 0.647	12.35 ± 0.601	12.85± 0.335	NS
Egg weight 21wks (g)	12.10 ± 0.251	12.0 ± 0.762	12.67 ±0.987	12.90 ± 0.699	12.41±0.329	NS
Overall main	12.57±0.398	12.32±0.566	13.03±0.553	12.63±0.432		
Egg Length 16wks(mm)	3.37 ± 0.145	3.23 ± 0.088	3.37 ± 0.088	3.37± 0.120	3.33±0.051	NS
Egg lenght21wks(mm)	3.2	3.27 ± 0.029	3.23 ± 0.126	3.27 ± 0.066	3.24±0.031	NS
Overall main	3.28 ±0.073	3.25±0.043	3.30±0.073	3.31 ±0.066		
Egg diameter 16wks(mm)	2.67 ± 0.033	2.57 ± 0.088	2.67 ± 0.033	2.60 ± 0.057	2.63 ±0.028	NS
Egg diameter 21wks(mm)	2.57 ± 0.033	2.57 ± 0.066	2.67 ± 0.033	2.70 ± 0.058	2.63± 0.028	NS
Overall main	2.62 ± 0.031	2.57± 0.050	2.67± 0.021	2.65± 0.043		
Egg shape16wks	79.43±2.56	79.65±4.84	79.36±2.91	77.39±2.798	78.95±1.47	NS
Egg shape21wks	80.21±1.04	78.57±1.76	82.63±2.13	82.66±0.88	81.02±0.84	NS
Overall main	79.82±1.25	79.11±2.32	80.99±1.78	80.02±1.77		
Internal egg quality:						
Yolk Height 16wks(mm)	1.13 ± 0.033	1.03±0.001	1.03 ± 0.089	1.00 ± 0.329	1.05±0.026	NS
Yolk Height 21wks(mm)	1.13 ± 0.088	1.07 ± 0.033	1.13 ± 0.033	1.20 ± 0.058	1.13±0.028	NS
Overall main	1.13± 0.042	1.03± 0.021	1.08±0.048	1.12±0.048		
Yolk diameter 16wks (mm)	2.63**a** ± 0.088	2.37**bc** ±0.664	2.57**ab** ±0.665	2.30 **c** ±0.058	2.46**A** ±0.054	*
Yolk diameter 21wks(mm)	2.83 ±0.033	2.83 ±0.145	2.67 ±0.088	3.10 ±0.251	2.86**B** ±0.080	NS
Overall main	2.73 ±0.061	2.60±0.127	2.61 ±0.054	2.70 ±0.214	***	
Yoil index16wks	43.05±0.497	42.32±1.156	40.44±4.243	44.98±1.795	42.70±1.130	NS
Yolk index2wks	40.07±3.474	37.83±2.237	42.58±1.647	39.41±4.468	39.97±1.439	NS
Overall main	41.56±1.711	40.08±1.512	41.51±2.094	42.20±2.496		
Yolk color 16wks	3.67 ±0.334	3.67 ±0.334	5.00 ±0.578	4.33 ±0.879	4.17**B** ±0.321	NS
Yolk color 21wks	6.00 ±0.578	6.00±0.578	4.67 ±0.334	6.33 ±0.334	5.75**A** ±0.250	NS
Overall main	4.83 ±0.602	4.83 ±0.602	4.83 ±307	5.33 ±0.616	***	
Yolk weight 16wks (g)	4.37 ±0.185	3.62 ±0.120	4.32 ± 0.246	3.99 ± 0.433	4.07±0.147	NS
Yolk weight 21 wks (g)	3.84 ± 0.351	4.01 ± 0.388	4.53 ± 0.066	4.1 ± 0.116	4.04±0.137	NS
Overall main	4.11 ±0.213	3.81 ±0.201	4.25 ±0.176	4.05 ±202		
Albumin Height 16wks(mm)	0.30	0.23± 0.066	0.17±0.333	0.23 ±0.333	0.23± 0.022	NS
Albumin Height 21wks(mm)	0.30	0.27 ± 0.033	0.33 ± 0.033	0.33 ± 0.087	0.31± 0.023	NS
Overall main	0.30	0.25 ± 0.034	0.25± 0.030	0.28 ± 0.034		
Albumin weight 16wks(g)	7.21±0.436	7.32±0.502	7.31±0.490	6.80±0.496	7.16 ±0.251	NS
Albumin weight 21wks(g)	6.89±0.166	6.46±0.371	6.96±0.681	7.14±0.535	6.86±0.218	NS
Overall main	7.05±0.247	6.89±0.428	7.13±0.385	6.97±0.334		
Albumin%16wks	4.20±0.272	3.43±1.150	2.34±0.560	3.48±0.595	3.36**B** ±0.366	NS
Albumin%21wks	4.36±0.104	4.10±0.370	4.86±0.543	4.55±0.875	4.47**A** ±0.249	NS
Overall main	4.28±0.136	3.77±0.561	3.60±0.664	4.02±0.529	*	
Shell weight 16wks(g)	1.47 ±0.106	1.70±0.202	1.77±0.093	1.55 ±0.076	1.62±0.066	NS
Shell weight 21wks(g)	1.37b ±0.046	1.53ab ± 0.069	1.51ab ± 0.051	1.67 a ± 0.088	1.52±0.042	*
Overall main	1.42 ±0.057	1.61 ± 0.103	1.63 ±0.075	1.61 ±0.058		
Shell thickness 16 wks(mm)	0.27 ± 0.009	0.32 ±0.029	0.32 ±0.027	0.29 ±0.033	0.30**B** ±0.013	NS
Shell thickness 21 wks(mm)	0.31 ±0.012	0.34 ±0.015	0.35 ±0.020	0.36 ±0.003	0.34**A** ±0.008	NS
Overall main	0.29 ±0.111	0.33 ±0.016	0.34 ±0.017	0.33 ±0.021	*	

a-d treatments within the same (row-wise) with different superscripts are significantly different (p≤ 0.05). A-D within the same time (column-wise) with different superscripts are significantly different (p ≤0.05); each value is a mean SE of three replicates.

From data in Table (5) it could be concluded that there were significant in plasma parameters as effect by supplemented diet with PPP and BHT. Plasma cholesterol, HDL, LDL, total lipids, AST, ALT, TAOC, createnine, uric acid and glucose were significantly (P≤0.001) decreased in all treated groups compared with the control one (Table 5). This result in agreement with Enas (2004) who investigated the role of Punicagrantum powder peel aqueous extract (PGPPE). She found that there was a significant decrease in plasma glucose in normal

rats treated with PPPE. While, plasma total protein were significantly (P≤0.05) increased in treated groups compared with the control. On the other hand, Fatma, (2009) demonstrated that hypercholestrolemic rats (positive control) administrated with PPP (5, 10 and 15 %) and its extracts (1, 2 and 3%) significantly decreased total cholesterol and HDL. While, PPP administration were significantly higher than the negative control.

Table 5. plasma parameters as affected by addition of pomegranate peel powder (ppp) and butylated hydroxyle toluene (BHT)

PLASMA	T1	T2	T3	T4	Sig.
Cholesterol(mg/dl)	216.52a±32.160	147.27b±2.751	144.70b±7.350	142.55b±2.325	*
HDL (mg/dl)	47.72a ±1.503	39.32b ±2.243	34.44b ±1.40	33.44b ±1.306	**
LDL (mg/dl)	113.13a±3.844	108.15ab±4.95	97.70bc±2.861	90.13c ±2.295	**
TL (mg/dl)	913a±11.857	658.48b±23.23	560.28c±9.047	516.77c±0.295	***
AST (U/ml)	0.880a ±0.027	0.69b ±0.043	0.52c ±0.010	0.46c ±0.018	***
ALT (U/ml)	0.87a ±0.032	0.66b ±0.023	0.62b ±0.070	0.50c ±0.018	***
TAOC(mM/L)	1.65a ±0.082	1.23b ±0.152	0.98b ±0.025	0.99b ±0.031	**
Albumen(mg/dl)	3.54 ±0.476	4.20 ±0.274	4.05 ±0.44	4.43 ±0.135	NS
Creatnine(mg/dl)	1.09a ±0.035	0.88b ±0.031	0.73c± 0.006	0.63c ±0.018	***
TP (mg/dl)	6.52d ±0.249	7.83c ±0.343	8.52b ±0.036	9.26a ±0.205	***
Uric acid (mg/dl)	5.90a ±0.150	4.85b ± 0.091	4.27c ±0.075	3.91c ±0.068	***
Glucose (mg/dl)	97.43a±0.859	87b±0.882	77.88c±1.282	72.52d±2.127	***

a-d treatments within the same (row-wise) with different superscripts are significantly different (p≤ 0.05).

Data in Table (6) showed that there were no significant effects of PPP and BHT on bird's weights before and after slaughtered or the relative organs weights. While groups fed on diet contained PPP and BHT were significantly higher than control group on defithered birds weight also there were a significant decrease in relative heart relative weight in group fed 15 g/kg PPP compared to other groups. These results are in agreement with Chalfoun-Mounayar *et al*. (2012) who found that adding pomegranate molasses or juice to mice drinking water (4 ml/l) during 11 weeks leading to a significant decrease in the heart, lungs, and the liver. Rao *et al*. (2000) showed that short-term or subchronic exposure to BHT affects the liver of chickens, also showing histopathological changes in this organ.

Data in Table (7) concluded that there were no significant effects of PPP and BHT on micro bacterial count in the small intestine. This results are in contrary with Yahia *et al*. (2011) who concluded that combinations of pomegranate rind extract (PRE) with metal salt ZnSO4 and Vitamin C (1:1:1)exhibit enhanced antimicrobial effects against both Gram positive (Bacillus subtilis, Staphylococcus spp. and Brucella spp.) and Gram negative (E. coli).

Table 6. Bird and carcass weightas affected by addition of pomegranate peel powder (PPP) and butylated hydroxyle toluene (BHT)

traits	T1	T2	T3	T4	Sig.
LBW (g)	265.67±9.035	293 ±26.60	251.67 ±4.809	264.33 ±8.197	NS
SBW (g)	257.67±8.116	284 ±25.610	244.33 ±5.818	258 ±7.237	NS
DFEW	223b ±7.01	276a ±26.59	226b ±4.046	236.33ab ±6.936	**
CARCASS	203.1 ±9.28	241.3 ±34.15	123.87 ±60.69	167.67± 26.01	NS
LIVER%	3.01 ±0.566	2.54 ±0.474	3.18 ±0.377	3.56 ±0.230	NS
SPLEEN%	0.18 ±0.026	0.13 ±0.052	0.39 ±0.289	0.29 ±0.058	NS
INT.%	3.42 ±0.158	3.54 ±0.428	2.99 ±0.428	3.89 ± 0.386	NS
HEART%	0.74a 0.052	0.75 a ±0.069	0.54b 0.045	0.87a ± 0.054	*

a-d treatments within the same (row-wise) with different superscripts are significantly different (p≤ 0.05).

Table 7. micro-bacterial count as affected by addition of pomegranate peel powder (PPP) and butylated hydroxyle toluene (BHT)

MICRO	T1	T2	T3	T4	Sig.
TBC	1.673×10^5	2.441×10^6	1.716×10^6	1787×10^6	NSP=0.6
Entrobac.	8.00×10^2	2.053×10^3	1.217×10^4	5.900×10^3	NSP=0.5
Coliform c.	1.20×10^2	1.880×10^2	1.233×10^3	1.40×10^2	NSP=0.1

The means within the same row with at least one common letter, do not have significant difference (P>0.05).

Data in Table (8) concluded that there was significant increase in total phenolics. While, the TBARS values was significantly decreased in group fed 15 mg PPP/kg diet compared with BHT supplemented diet and the control group. The TBARS values significantly increased affected by the storage period. These results in agreement with Rajani *et al*. (2011) who mentioned that pomegranate peel treatment being the most effective (P≤0.01) introducing the Malondialdehyde (MDA) occurrence in meat of birds. MDA occurrence in meat is a popular assessment of lipid oxidation (Botsoglou *et al*., 1994). Although there was no study examining the antioxidative effect of dietary PP on stored meat, Swamy *et al*. (2011) reported the lower MDA in fresh liver samples of rats fed PP extract (25 mg/d polyphenols equivalent). The PP ability to lower MDA levels could be explained by antioxidant compounds including, (a) ellagitannins, a precursor of ellagic acid, which has been found to have antioxidative properties (Osawa *et al*., 1987; Mass *et al*., 1991). Also, Zeweil *et al*. (2013) reborted that lipid peroxide (malondialdehyde) levels decreased significantly to reach around 54% of the heat stressed bucks,' value of PP were 1.5, 3.0 and 4.5% of PP dietary used.

Table 8. Thiobarbituric acid reactive substances (TBARS) values (mg ofmalonaldehyde/kg meat) and the total phenolic content of (tannic acid eq) ug/g as affected by addition of pomegranate peel powder (PPP) and butylated hydroxyle toluene (BHT) during frozen storage at -20 C

Treatments/Storage period (month)	T1	T2	T3	T4	Sig.
0	0.307± 0.08 cA	0.271± 0.08bcA	0.103± 0.03aA	0.18± 0.05abA	**NS**
1	0.635± 0.09cB	0.266± 0.03bA	0.098± 0.01aA	0.388±0.17bAB	*****
2	1.016± 0.03cC	0.485± 0.16bB	0.140± 0.03aA	0.498± 0.31bB	*****
3	1.272± 0.13cD	0.763± 0.16bC	0.203± 0.04aB	0.896± 0.12bC	******
Total phenolics (as tannic acid eq)ug/g	153 ± 19.55 c	192 ± 10.60 b	225 ± 10.60 a	160 ±18.16c	*****

a-d treatments within the same storage conditions (row-wise) with different superscripts are significantly different (p≤ 0.05). A-D storage conditions within the same treatment (column-wise) with different superscripts are significantly different (p ≤0.05); each value is a mean SE of three replicates.

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تـأثير إضـافه مسـحوق قشـر الرمـان ومـاده بيوتايلـد هيدروكسـى التولـوين فـى العليقـه علـى بعـض القياسـات الانتاجيه والفسيولوجيه والمناعيه للسمان الياباني

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يهدف هذا البحث لدراسه تأثير إضافه مسحوق قشر الرمان المجفف و ماده بيوتايلد هيدروكسى التولوين (BHT) فى العليقه على الاداء الانتاجى والفسيولوجى والمناعى للسمان الياباني البياض. تم إستخدام عدد ١٨٠ سمانه عمر ١١ اسبوع قسمت الطيور عشوائياً الى ٤ مجاميع كل مجموعه قسمت الى ٣ مكررات (١٥ سمانه/مكرره). المجموعه الاولى : كنترول تغذت على العليقه بدون إضافات . المجموعه الثانيه: تغذت على عليقه مضاف اليها ١٠ جم مسحوق قشر الرمان /كجم عليقه. المجموعه الثالثه: تغذت على عليقه مضاف اليها ١٥ جم مسحوق قشر الرمان /كجم عليقة. المجموعه الرابعه: تغذت على عليقه مضاف اليها ١٢٥ جم ماده BHT /طن عليقه.

وقد تم الحصول على النتائج التاليه:

١- كان هناك زيادة معنوية فى وزن الجسم فى المعاملات المستخدم فيها مسحوق قشر الرمان بينما كميه الغذاء المستهلك ومعدل التحويل الغذائى انخفضا معنوياً فى كل المجموعات المعامله بالمقارنه بمجموعه الكنترول.

٢-وزن قشره البيضه كان اعلى معنوياً فى كل المجموعات المعامله بينما ارتفاع الصفار كان الاعلى معنوياً فى معامله الكنترول مقارناً بكل المجموعات المعامله ، سمك القشره وقياسات الصفار على عمر ١٦ اسبوع كانت اعلى معنوياً بينما لون الصفار والالبيومين كانا اعلى على عمر ٢٢ اسبوع.

٣- إنخفض كلا من الدهون الكليه فى البلازما والكوليستيرول و HDL و LDLواليوريك اسيد و الكرياتينين وانزيمات الكبد باضافه مسحوق قشر الرمان، بينما بروتينات البلازما الكليه زادت معنوياً فى مجموعات المعامله و مجموعه ال BHT.

٤- وزن جسم الطيور منزوعه الريش كان اعلى معنوياً فى المجموعات التى تغذت على ١٠ جم مسحوق قشر الرمان بينما النسبه المئويه لوزن القلب إنخفضت فى المجموعات التى تغذت على ١٥ جم مسحوق قشر الرمان.

٥- إضافه كلا من مسحوق قشر الرمان وماده BHT لم تؤثر فى العدد الكلى لميكروبات الامعاء.

٦- الفينولات الكليه زادت معنوياً بينما قيم TBARSوانخفضت معنوياً فى المجموعه التى تغذت على ١٥ جم مسحوق قشر الرمان مقارنه بمعامله ماده BHT والكنترول.

ويمكن التوصيه بإن إضافه ١٥ جم مسحوق قشر الرمان/ كم عليقه أدى الى تحسن فى الصفات الانتاجيه والفسيولوجيه وكذلك يمكن أن يطيل من مده حفظ وتخزين اللحم فى المبردات وحتى موعد استهلاكها.