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The Effect of Diet Propolis Supplementation on Ross Broiler Chicks Performance

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Abstract: An experiment was conducted to investigate the effects of Alcoholic Extract of Propolis (AEP) on Ross (308) broiler chicks performance. This experiment , was carried out in a completely randomized design with 6 treatments (different levels of propolis including 0, 50, 100, 150, 200 and 250 mg/kg diet) for 6 weeks. Weight gain, feed consumption , feed conversion ratio and mortality rate at the end of 3rd, 6th wk and whole periods (0 to 6 wk) were compared statistically. Results indicated that in all periods, average weight gain, feed consumption, feed efficiency were significantly higher for propolis fed birds and inclusion of Propolis also reduced mortality rate in comparison to control diet.

Key words: Ross (308) broiler chicks, Propolis, alcoholic extract, mortality rate

Introduction

Propolis (Bee glue) is a complex resinous hive product and mixture of wax , sugars and plant exudates collected by bees from certain plant sources. More than 300 constituents have been identified in different propolis samples(Banskota *et al*., 2001). In general, propolis composition is directly related to that of bud exudates collected by bees from various trees:poplar (Populus spp.), birch (Betula alba), beech (Fagus stylvatica), horse chestnut (Aesculus hippocastanum), alder (Alnus glutinosa) and various conifers (Mochida *et al.,* 1985; Ghisalberti, 1979; Amoros *et al*., 1992; Bankova *et al*., 2000). Literature survey revealed that flavonoids, aromatic acids, diterpenic acids and phenolic compounds appear to be the principal components responsible for the biological activities of propolis samples. The contents depend on the collecting location , time and plant source (Greenaway *et al*., 1991; Markham *et al*., 1996). The ethanolic extract of propolis has some activities such as antibacterial (Mochida *et al*., 1985; Ghisalberti, 1979; Velikova *et al*., 2000; Pepeljnjak *et al*., 1985), antifungal (Dimov *et al*., 1991; Schneidewind *et al*., 1979; Murad *et al*., 2002) , antiviral (Amoros *et al*., 1992; Amoros *et al*., 1994), Local-anaesthetic (Paintz and Metzner, 1979), anti-inflammatory (Strehl *et al*., 1993; Miyataka *et al*., 1997), antioxidant (Sun *et al*., 2000; Isla *et al*., 2001), hepatoprotective (Gonzales *et al*., 1995), immunostimulating (Dimov *et al*., 1991), and cytostatic (Frenkel *et al*., 1993; Banskota *et al*., 2001). Propolis are used by worker bees to line the inside of nest cavities and all brood combs, repair combs, seal small cracks in the hive, reduce the size of hive entrances, seal of inside the hive any dead animals or insects which are too large to be carried out and perhaps most important of all, to mix small quantities of propolis with wax to seal brood cells (Krell, 1996). Flavonoids are a group of polyphenolic compounds diverse in chemical structure and characteristics. They occur naturally in fruit, vegetables, nuts, seeds, flowers, and bark and are an integral part of the human diet (Middleton and Kandaswami, 1993; Hackett *et al*., 1986). They have been reported to exhibit a wide range of biological effects , including antibacterial, antiviral (Hanasaki *et al*., 1994), anti-inflammatory, antiallergic (Middleton Kandaswami, 1993; Hanasaki *et al*., 1994), and vasodilatory actions (Duarte *et al*., 1993). For 12 months 4000 Hubbard Golden comb hens, initially aged 5.5 months and of mean weight 1850 g , were given a meal mixture without and with propolis (10 , 20 and 30 mg/Kg diet). 30 mg/Kg propolis in diet significantly increased egg production, egg weight, feed utilization and weight gain by 6.07 , 1.27 , 5.46 and 6% respectively, compared with controls (Bonomi *et al*., 1976). In other experiment ,effect of some bee products on immune response of chicken infected with virulent NDV was studied. Mortality rate was reduced in groups infected with virulent NDV and subsequently treated either with propolis or honey in compared with the infected groups only (Hegazi *et al*., 1995). Ghisalberti (1979) reported that inclusion of 500 ppm Propolis in broilers improved body weight (20 %) for propolis fed birds in comparison to control group. Although numerous reports concerning the biological activities of propolis collected in certain countries have been documented, information concerning the characteristics of Iranian propolis is still quite limited. Therefore, the present study investigated the effect of 96% ethanolic extract of propolis samples collected from different regions of Ardebile on performance of Ross (308) broiler chickens.

Table 1: Composition and analysis of experimental diets

	Starter diets (mg/kg propolis)					
	%					
Ingredient	0	50	100	150	200	250
Corn	64.5	64.49	64.49	64.49	64.49	64.49
Soybean meal[1]	26	26.02	26.02	26.02	26.02	26.02
Fish meal[2]	6	6	6	6	6	6
Sun flower oil	0.7	0.7	0.7	0.7	0.7	0.7
Oyester shell	1.19	1.19	1.19	1.19	1.19	1.19
Dicalcium phosphate[3]	0.7	0.7	0.7	0.7	0.7	0.7
Salt	0.3	0.3	0.3	0.3	0.3	0.3
Premix[4]	0.5	0.5	0.5	0.5	0.5	0.5
D L – Methionine	0.1	0.1	0.1	0.1	0.1	0.1
Calculated analysis :						
AME_n (kcal/kg)	2965	2,965	2,965	2,965	2,965	2,965
CP(%)	20.8	20.84	20.84	20.84	20.84	20.84
Linoleic acids (%)	1.93	1.93	1.93	1.93	1.93	
Met (%)	0.49	0.49	0.49	0.49	0.49	0.49
Lys (%)	1.17	1.17	1.17	1.17	1.17	1.17
Ca (%)	0.92	0.92	0.92	0.92	0.92	0.92
$P_{available}$ (%)	0.4	0.4	0.4	0.4	0.4	0.4
	grower diets (mg/kg propolis)					
	%					
Ingredient	0	50	100	150	200	250
Corn	67.50	67.50	67.50	67.50	67.50	67.50
Soybean meal[1]	24.12	24.12	24.12	24.12	24.12	24.12
Fish meal[2]	3	3	3	3	3	3
Sun flower oil	2.11	2.11	2.11	2.11	2.11	2.11
Oyester shell	1.12	1.12	1.12	1.12	1.12	1.12
Dicalcium phosphate[3]	0.85	0.85	0.85	0.85	0.85	0.85
Salt	0.3	0.3	0.3	0.3	0.3	0.3
Premix[4]	0.5	0.5	0.5	0.5	0.5	0.5
D L – Methionine	0.5	0.5	0.5	0.5	0.5	0.5
Calculated analysis :						
AME_n (kcal/kg)	3,095	3,095	3,095	3,095	3,095	3,095
CP(%)	18.32	18.32	18.32	18.32	18.32	18.32
Linoleic acids (%)	2.8	2.8	2.8	2.8	2.8	2.8
Met (%)	0.34	0.34	0.34	0.34	0.34	0.34
Lys (%)	0.97	0.97	0.97	0.97	0.97	0.97
Ca (%)	0.81	0.81	0.81	0.81	0.81	0.81
$P_{available}$ (%)	0.35	0.35	0.35	0.35	0.35	0.35

[1]-44% Cp. [2]- 60% Cp. [3]- 22% Ca ; 18.7% P. [4]-provided per kg of diet : vitamin A , 9 , 000 lu ; vitamin D3 , 1,500 lu ; vitamin E , 10 lu ; vitamin K , 5 mg ; vitamin B_{12} , 0.007 mg ; thiamin , 0.4 mg ; riboflavin , 6 mg ; folic acid , 1 mg ; biotin , 0.15 mg ; pantathenic acid , 12 mg ; niacin , 35 mg ; pyridoxine , 4 mg ; choline , 1 , 000 mg ; Mn , 60 mg ; Cu , 5 mg ; Zn , 50 mg ; Se , 1 mg ; I , 0.35 mg ; ethoxyquin , 1,25 mg .

Materials and Methods

Propolis origins: propolis samples were collected from different locations in Ardebile province in may 2005 and pooled. Hand-collected propolis samples were kept in the dark up to their processing and stored at -20°C Preparation of Ethanolic Extracts of Propolis (EEP) Solution Propolis samples were cut into small pieces, ground to a fine powder and extracted with 96% ethanol. Let's see how a ethanolic extract of propolis can be made (Krell, 1996).

Take 30-40 grams of propolis powder and mix it with 70 ml of 90-96% alcohol. Leave for a minimum of 14 days, in a dark cool place (not in refrigerator), but stir also 3-6 times per day, or find a mechanical way to do this automatically. It is important to allow the alcohol molecules to come into contact with as many propolis compounds as possible, in order to extract them from the solid mass.

After two weeks (The longer the better) the solution can be filtered ; The liquid portion should be stored in a dark

Table 2: The effect of alcoholic extract of propolis supplementation on daily weight gain, daily feed consumption, feed conversion ratio and mortality

	Diet (mg/kg propolis)					
Parameter	0	50	100	150	200	250
Daily weight gain						
0 to 3 wk	23.953^{c}	24.263^{c}	24.890^{bc}	25.772^{b}	26.186^{b}	38.653^{a}
3 to 6 wk	73.100 ^{d}	72.779^{d}	73.991^{d}	75.586^{c}	78.066^{b}	85.055^{a}
0 to 6 wk	40.251^{d}	41.831^{dc}	41.605^{dc}	44.541^{c}	47.775^{b}	55.329^{a}
Daily feed consumption						
0 to 3 wk	43.565^{d}	44.421^{dc}	44.918^{c}	46.441^{b}	47.508^{b}	56.037^{a}
3 to 6 wk	131.965^{d}	130.701^{d}	131.713^{d}	136.077^{c}	141.514^{b}	149.367^{a}
0 to 6 wk	83.235^{c}	84.294^{c}	86.648^{b}	88.265^{b}	88.422^{b}	92.205^{a}
Feed conversion ratio						
0 to 3 wk	1.81^{a}	1.83^{a}	1.80^{a}	1.80^{a}	1.81^{a}	1.44^{b}
3 to 6 wk	1.80^{a}	1.79^{a}	1.78^{ab}	1.80^{ab}	1.81^{a}	1.74^{b}
0 to 6 wk	2.06^{ab}	2.01^{ab}	2.08^{a}	1.90^{ab}	1.85^{b}	1.66^{c}
Mortality, %						
0 to 3 wk	6.25^{a}	6.01^{a}	7.75^{a}	5.25^{a}	0.25^{b}	0^{c}
3 to 6 wk	2.05^{a}	1.97^{a}	1.75^{a}	1.65^{a}	0^{b}	0^{b}
0 to 6 wk	2.75^{a}	2.70^{a}	2.65^{a}	2.50^{a}	0.02^{b}	0^{b}

$^{a-d}$Value within a row with no common superscripts are significantly different ($p<0.01$)

green or dark brown bottle in a cool, dry and dark place. The ethanolic extract solution was then filtered through a whatman 1 filter paper or clean and very fine cloth and restored to the original volume with 96% ethanol.

Experiment design and treatments: 960 day–old commercial broiler chicks (Ross 308), were weighted individually and randomly allocated to dietary treatments so that each experimental unit had equal average weight and weight distribution. Chicks experimental diet were fed from day old to 42 d of age. Feed and water were available at libitum . Wood shavings were used as litter. Experiment was carried out in a completely randomized design with 6 treatments (different levels of propolis) and four replication of 40 birds per each treatment. Corn-soybean meal diets were formulated to meet or exceed National Research Council (1994) requirements. A starter diet with 2,966 kcal AME_n/Kg and different levels of propolis (0, 50, 100, 150, 200 and 250 mg/kg) was fed for the first 3 wk and a grower diet with 3,095 kcal AME_n/Kg and different levels of propolis same to the level used in starter period, from 3rd to 6th weeks of age (Table 1). Ethanolic extract of propolis was added to mixed diets. Weight gain and feed consumption were measured daily and then , feed conversion ratio (feed:gain) was calculated. Mortality rate were also measured.

Statistical analysis: Data from this experiment were evaluated by ANOVA using General Linear Models procedures (SAS Institute, 2001). Significant difference ($p<0.01$) between main effects were detected by Duncan (1955) multiple range test. Mortality data were converted by arcsin transformation before analysis, but for interpretation purposes the data are presented as percentage.

Results

Weight gain, feed consumption and feed conversion ratio are presented in Table 2. Chicks fed diets contain Propolis had an improved WG and feed efficiency compared to control diet but significant effects of Propolis inclusion were observed only in high levels (200 and 250 mg/kg). Mortality rate was affected in the same manner with WG and feed efficiency and only chicks fed diets containing 200 and 250 mg/kg Propolis had significantly lower mortality .

Discussion

Results obtained here indicated that addition of 250 mg/kg Propolis may be benificial in improving broiler chicks performance. Chicks fed Propolis containing diets consumed significantly($p<0.01$) higher feed . Bonomi *et al.* (1976) found an increase in feed intake when laying hens were fed propolis versus control groups. They concluded that increase in feed intake in the propolis groups may be due to improved birds health and higher palatability of propolis diets due to mixture of resine, wax, honey and vanillin content of propolis. Ghisalberti (1979) report additional weight gains for broiler chickens of up to 20% when 500 ppm of propolis was added to their diets. They said that this improved effect is partially due to its high content of flavonoids and increase feed intake of propolis diets than the control. Experimental work of Buhatel *et al.* (1983) showed that propolis supplementation to the ration of pullets

improved feed conversion. This effect is due to high content of flavonoids and healthy conditions of birds fed propolis. Increased feed intake obtained in our study is in good agreement with the results mentioned above. Significantly ($p<0.01$) higher weight gain in Propolis fed chicks observed here is probably asociated with higher feed intake. A marginal increase in feed intake and weight gain in the propolis groups, resulting in marginally superior feed efficiency than the control.

In this study , mortality rate were lower at the 3 wk and 0 to 6 wk of age when broilers were fed propolis extract specially more in high levels of propolis (200 and 250 mg/kg) versus control groups. Similar reports were drawn by Giurgea *et al*. (1981). They indicated that daily administration of propolis extract to chickens changed the blood concentration of cholesterol , total proteins and amino acid. It also stimulated the immune system which this effect resulted decreased in mortality as compared to the control. Results from this study indicate that using ethanolic extract of propolis (EEP) can result in improved broiler performance. Also , it may be concluded that propolis could be safely incorporated in broiler starter and grower diets up to 250 mg/kg level.

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