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Residual Feed Intake and its Effect on Cell-Mediated Immunity in Laying Hens Given Different Propolis Levels

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Abstract: The main aim of this study was to investigate the relationship between residual feed intake and some hematological parameters in laying hens fed different level of Propolis. Residual feed intake is used as a trait to distinguish efficient and non-efficient laying chicken. A Phytohemagglutinin-P (PHA-P) assay and white blood cells differentiation were used to evaluating the immune response of the laying hens fed different propolis levels. The present result indicated that the laying hens fed different levels of Propolis had significantly hyper responder to PHA-P injection, higher lymphocytes count and lower heterophils count when compared with control-group. With respect to residual feed intake, it was generally noticed that the observed feed consumption values were somewhat closely to expect feed consumption values for hens fed diet containing 100 or 150 mg Propolis compared to remaining treatments. Also, the results revealed that the equations calculated for laying hens fed diet adding 100 or 150 mg Propolis had a better rate of determination (R^2) compared to other fed 0 or 50 mg Propolis. In accordance to correlation coefficients, it could be noticed that the wattle swelling measured at 24, 48 and 72 h post PHA-P injection were significantly positive correlated with residual feed consumption in supplemental propolis at 100 mg. Similar trend was observed in 150 mg supplemental propolis laying at 72 h post PHA-P injection. With respect to white blood cells differentiation, the present results showed that significantly negative relationship between RFC and heterophils percentage in all treatment groups. Inversely, the relationship between RFC and lymphocytes percentage was significant and positive in all treatment groups. Finally, it could be concluded that the Propolis supplementation improve the immune status of laying hens via minimizing the residual feed intake.

Key words: Residual feed intake, immunological parameters, laying hens

INTRODUCTION

Laying hens can be considered as being genetically programmed for efficient Egg Mass (EM) production. There is, however, a lot of variation in efficiency between individuals within a population, as shown by variation in feed consumption that cannot be explained by metabolic Body Weight (BW), body weight gain and Egg Mass (EM). This variation in feed consumption is reflected in differences in Residual Feed Intake (RFI) (Luiting and Urff, 1991). RFI is defined as the difference between observed Feed Intake (FI) and FI predicted from metabolic BW (representing maintenance) and BW gain and EM (both representing production). RFI is, thus, a measure for feed efficiency: chickens with low RFI (R-) need less feed to reach the same BW and production level and are, therefore, more efficient producers than chickens with high RFI (R+). The phenotype of an animal is a function of its genotype and present and past environments. Environmental factors such as nutrition, feeding management and temperature may influence growth and modify sensitivity to infectious agents and immunoresponsiveness (Zulkifli *et al.,* 1994). Immunocompetence and growth are influenced by genetic and non-genetic factors, only recently has evidence appeared showing a negative relationship between body weight and immunocompetence in Leghorns (Martin *et al.,* 1990), broilers (Qureshi and Havenstein, 1994) and brown egg layers (Kreukniet *et al*., 1994). In broiler breeding there is a further complication because although broilers are reared under *ad libitum* feeding, the management of broiler breeders involves restriction in feed intake to control over-consumption and its deleterious effects on health and reproduction (Katanbaf *et al.,* 1989). As a result of this husbandry, research results have accumulated on effects of various feeding regimens on body weight (Robinson *et al.,* 1993) and feed intake and diet selection (Forbes, 1995). Immune stimulation (Klasing *et al.,* 1987) and injection of chickens with IL-1-containing supernatant from bacterial endotoxin stimulated chicken HD11 macrophages (Klasing *et al.,* 1987) induced a reduction of growth and feed utilization in chickens and accelerated muscle protein degradation. In this study, we evaluate the relationship between residual feed intake and cell mediated immunity of laying Hy-line hens fed different levels of Propolis.

MATERIALS AND METHODS

This experiment was carried out at poultry breeding farm, Poultry Production Department, Faculty of Agriculture, Ain Shams University. One hundred and twenty 46 -week-old, Hy-line White strain were randomly assigned to 4 groups of 30 hens each. The hens were individually housed in individually cages, on a 16 h light schedule. Chickens in all groups were reared under the same environmental, managerial and hygienic conditions. Feed and water were supplied *ad libitum*. The hens received a typical layer diet containing 2800 ME kcal/kg and 18% CP to meet or slightly exceed the nutrient requirement recommended by NRC (1994). The hens were fed basal diet (control) or basal diet containing 50, 100 and 150 mg Propolis/kg.

Measurements and observations: Body weight was individually recorded at 46, 50 and 54 weeks of age. Also, number and weight of eggs were recorded daily from the 46-54 weeks of age. Internal and eggshell quality was determined at 54 weeks of age. The egg length (long axis) and width (short axis) were measured with the electronic caliper. The width to length ratio was shown in percentage points and constituted the egg shape index. The height of thick albumen (H) and the egg Weight (W) were used to calculate Haugh units from the formula of Williams (1997):

$$HU = 100 \log (H + 7.7\text{-}1.7\ W^{0.37})$$

where:

H = Thick albumen height.
W = Egg weight.

Yolk diameter along the chalazae line was determined with the caliper (mm). The eggshell, after the removal of the egg content, was dried. Subsequently the eggshell was weighed to the nearest 0.01 g. Eggshell thickness without inner membranes was measured (mm) with the micrometer. The albumen weight was calculated from the difference between the entire egg weight and the yolk and eggshell weight. The contents of yolk, albumen and the eggshell were expressed as percentages from the weight of a fresh egg. The breaking strength was measured according to Fathi and El-Sahar (1996) which assessed the resistance of the egg to crushing.

At 54 weeks of age, blood samples were taken from the brachial vein into heparinized tubes for all birds. Plasma was obtained from the blood samples by centrifugation for 10 min at 4000 rpm and was stored at -20°C until the time of analysis. The frozen plasma was allowed to thaw at room temperature prior to analysis. Plasma total protein, albumin and cholesterol were determined by enzymatic colorimetric methods using available commercial kits. The plasma globulin was calculated as the difference between plasma total protein and albumin.

A Phytohemagglutinin-P (PHA-P) injection assay (Cheng and Lamont, 1988) was used to evaluate *in vivo* T-cell-mediated immune response of Hy-Line laying hens. Birds were injected intradermally in the wattle with 0.5 mg of PHA-P (Sigma Chemical Co., St. Louis, Missouri) in 0.1 mL of Phosphate Buffered Saline (PBS) after marking the injection site. The thickness of wattle was measured (to nearest 0.01 mm]) at 0, 24, 48 and 72 h after PHA-P injection. Wattle swelling was calculated as the difference between the thickness of the wattle prior to and after injection of PHA-P.

At 54 week of age, blood samples were obtained from each treatment for Heterophil (H) and Lymphocyte (L) enumeration based on the procedures of Gross and Siegel (1983). Briefly, one drop of blood being smeared on each of glass slides. The smears were stained using Wright's stain. Two hundred leukocytes, including granular (heterophils) and nongranular (lymphocytes) ones, were counted on different microscopic fields representing 200 cells and the heterophil to lymphocyte ratio was calculated.

Computing data and statistical analysis: The feed consumption for hens was predicted to derive regression equation according to treatment. Residual Feed Consumption (RFC) was calculated as the Difference Between Observed (OFC) and Expected Feed Consumption (EFC) for each experimental hen. Each treatment had its own regression coefficients according to the following equation:

$$EFC = aBW_i^{0.75} + bEM_i + c)W_i + d$$

where:

EFC = Expected feed consumption of hen I grams).
$Bw_i^{0.75}$ = Mean metabolic body weight of hen I ($kg^{0.75}$).
Em_i = Egg mass production of hen I (grams).
$)w_i$ = Body weight gain.
a, b and c = Partial regression coefficients.
d = Intercept.

All calculations and analyses were made using General Linear Models (GLM) procedure of SAS User's Guide, 2001. Correlation coefficients of RFC with productive traits were estimated for each strain using the PROC CORR procedure of SAS.

RESULTS AND DISCUSSION

Cell-mediated immunity, some hematological parameters and productive traits of laying hens fed different levels of Propolis are summarized in Table 1. The PHA intradermally reaction, a T-lymphocyte-dependent response, has been well researched and has been shown to be a reliable indicator of *in viva*

Table 1: Cell-mediated immunity, some hematological parameters and productive traits of laying hen fed different levels of Propolis

	Propolis level (mg/kg)					
	0	50	100	150	Pooled S.E.M	Prob.
Cell mediated-immunity						
24 h post PHA-P injection	0.31^{d}	0.40^{c}	0.54^{b}	0.65^{a}	0.02	0.001
48 h post PHA-P injection	0.21^{d}	0.36^{c}	0.41^{b}	0.51^{a}	0.03	0.001
72 h post PHA-P injection	0.09^{d}	0.19^{c}	0.30^{b}	0.36^{a}	0.02	0.01
White blood cells						
Heterophils	35.16^{a}	34.82^{b}	32.14^{bc}	30.27^{c}	0.52	0.01
Lymphocytes	64.84^{c}	65.18^{c}	67.86^{b}	69.73^{a}	0.67	0.01
Hematological parameters						
Total plasma protein (g/dl)	5.67^{b}	5.68^{b}	6.18^{a}	6.25^{a}	0.12	0.05
Globulin (g/dl)	2.10^{b}	2.24^{b}	2.67^{a}	3.02^{a}	0.15	0.02
Cholesterol (mg/dl)	132.14^{a}	131.12a	125.46^{b}	119.18^{c}	2.14	0.001
Productive parameters						
Observed feed consumption	6559^{b}	6368^{c}	6652^{a}	6636^{a}	30.25	0.01
Feed conversion ratio	2.45^{a}	2.31^{a}	2.13^{b}	2.07^{b}	0.10	0.001
Egg number	43.56^{c}	45.08^{b}	50.41^{a}	51.35^{a}	0.85	0.01
Egg weight	61.53^{b}	61.21^{b}	62.26^{a}	62.83^{a}	1.32	0.04
Albumen (%)	60.27	60.18	60.11	59.59	2.15	ns
Yolk (%)	29.76	29.83	29.74	30.09	0.74	ns
Shell (%)	9.97^{b}	10.00^{b}	10.15^{a}	10.32^{a}	0.14	0.03
Shell thickness	0.311^{b}	0.323^{b}	0.362^{a}	0.365^{a}	0.02	0.01
Breaking strength	3.15^{b}	3.29^{b}	3.54^{a}	3.65^{a}	0.16	0.001

$^{a-d}$Means with different letters within row are significantly differed

cellular immunity in poultry (Goto *et al.* 1978). The skin response reflects a complex series of physiological events such as mitogen-receptor and lymphocyte-macrophage interactions, release of chemical mediators, cellular proliferation and changes in vascularity (Chandra and Newberne, 1977). Histologically, PHA is strongly mitogenic to T-lymphocytesa and intradermali injections elicit macrophage infiltration and dense perivascular accumulations of lymphocytes 24 h post-injection in chickens (Goto *et al.,* 1978; McCorkle *et al*., 1980). The increased infiltration by basophils and eosinophils 24 h post-injection has been described as a cutaneous basophil hypersensitivity response (Stadeckerm *et al*., 1977). The present result showed that the specific responses to PHA-P injection at all times showed marked differences between treatment groups, with supplemental Propolis laying hens at all levels reaching much higher responder than control group. Propolis according to research has shown to be effective against a variety of bacteria, viruses, fungi and molds. It has been shown to be a non-specific immunostimulant. The delayed hypersensitivity skin test using propolis as sensitizing antigen showed specific stimulation to propolis after 72 h after inoculation with specific antigen. Egyptian propolis gave the typical delayed hypersensitivity when inoculated to the sensitized chickens. The thickness index was 0.90 mm thickness if compared with non-sensitized control group 0.12 mm thickness (Hegazi *et al*., 1996). Hegazi *et al*. (1995) studied the effect of some bee products on immune response of chicken infected with virulent NDV. They found that, the mortality rate was reduced in-groups infected with virulent NDV and subsequently treated either with propolis or honey if compared with the infected groups only. It was clear that, propolis acts actively as antiviral agent than honey. The treatment with propolis and honey of NDV infected chicken groups induced increase in the antibody titres and phagocytic percentage. The inoculation of different antigens in the footpad of sensitized and non-sensitized chickens induced different degrees of footpad thickness as well as cellular and vascular reaction depending on the type of inoculation with NDV antigen. In accordance to white blood cells differential, it could be noticed that the Propolis levels at 100 and 150 mg significantly decreased heterophils count and significantly increased lymphocytes count compared to control-fed group. In birds, the heterophil are phagocytic cells whose main is protection against invading microorganisms, whereas primary functions of lympho-involve cell-mediated and humoral immunity. Heterophils increase and lymphocytes decrease when are stressed, so that the ratio between them is a index of response to a stressor (Gross and Siegel, 1985). In accordance to H/L ratio, our results showed that the propolis supplementation at 2 and 3g/kg diet significantly increased the H/L ratio of laying hens. The H/L ratio is a recognized measure of stress in birds (Maxwell, 1993) that has become a valuable tool in stress research especially when combined with the convenience and repeatability of automated blood cell counts.

With respect to hematological parameters, it could be speculated that the laying hens fed high Propolis levels (100 and 150 mg) were significantly increased total plasma protein and plasma globulin compared to

control-fed group. Inversely, the Propolis supplemental at high levels significantly decreased plasma cholesterol. The low level of Propolis was intermediated in all cases. 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. Also, Propolis stimulated mammalian tissue regeneration, as it caused strong activation of mitosis of cells cultured *in vitro* and it enhanced protein biosynthesis (Gabrys *et al*., 1986).

The laying hens fed either 100 or 150 mg Propolis consumed more feed compared to other fed control-fed group. However, the 50 mg Propolis level was intermediated. Concerning feed conversion ratio, the present result indicated that the Propolis supplementation at all levels had significantly improved feed conversion ratio compared to control-fed group. The egg number was significantly affected by Propolis supplementation. Moreover, the egg number was gradually increased with Propolis level increasing. The laying hens fed diet containing 100 and 150 mg Propolis significantly increased egg weight compared to control fed-group. The increase feed intake and egg mass in Propolis groups, resulting in significantly improve feed conversion ratio compared to control-group. Shalmany and Shivazad (2006) reported that chicks fed Propolis containing diets consumed significantly higher feed. Bonomi *et al.* (1976) found an increase in feed intake when laying hens were fed Propolis versus control groups. Also, 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 resin, 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.

Concerning internal and eggshell quality, it could be noticed that there was no significant difference among treatment groups for both albumen and yolk percentage. However, the eggs produced from laying hens fed diet containing either 100 or 150 mg Propolis were significantly higher eggshell thickness compared to other groups. Also, the supplemental Propolis at high level (100 or 150 mg) significantly increased eggshell breaking strength compared to remaining groups.

Multiple regression equation of feed consumption: Observed feed consumption, egg mass, change in body weight and metabolic body weight for each hen within each treatment was used to estimate the regression coefficients are listed in Table 2. The results revealed that the equation calculated for laying hens fed diet adding 100 or 150 mg Propolis had a better rate of determination (R^2) compared to other fed 0 or 50 mg Propolis. That is mean, the figures of RFC calculated from these equations are more reliable and have a highly applicable prospective. The present results revealed that the regression of body weight on feed intake may differ between treatment groups, but the accuracy of feed intake predictions is independent of the power used to express metabolic body weight. Pirchner (1985) reported that differences in Observed Feed Consumption (OFC) and Expected Feed Consumption (EFC) are caused by variability in several factors, such as composition of product (eggs), body weight change, food spillage, metabolic rate and in the ability to synthesis egg and body constituents. Figure 1 illustrates the observed against expected feed consumption for each hen. While, Fig. 2 depicts the residual feed consumption value for each hen. It was generally noticed that the observed values were somewhat closely to expected values for hens fed diet containing 100 or 150 mg Propolis compared to remaining treatments. This adjacency was reflected on RFC, where it was more consistent to zero line. The negative values of RFC are desirable rather than positive ones. The efficient hens which have negative RFC figures were more frequent than inefficient ones. The results of El-Sayed and El-Hakim (1994) and Hussein *et al.* (2000) confirmed that the efficient birds were less actives, less heat production, spent more time resting and less time standing than inefficient bird. The last result explained by van Eerden *et al.* (2004). They reported that selection for higher efficiency of food use in laying hens might result in birds that one less frustrated prior to laying at less stressfully. If reduction in feed intake is an indicator of stress, then the observation that feed intake of the R+ chickens decreased considerably after transportation, whereas feed intake of R-chickens remained almost unaffected.

Table 2: Coefficients of partial regression and constant for laying hens fed different Propolis levels

Propolis (mg/kg)	Constant	EM	)W	$BW^{0.75}$	R^2
0	6572.91	-1.20	0.54	12.85	0.44
50	-3935.82	2.02	-0.06	19.39	0.45
00	10542.04	-2.78	1.34	19.00	0.63
50	-4885.78	4.02	2.84	-7.03	0.78

EM = egg mass,)W = body weight change, $BW^{0.75}$ = metabolic body weight, R^2 = rate of determination

Correlation coefficient between RFC, cell-mediated immunity and some productive traits: Calculation RFC by phenotypic multiple regression analysis is an

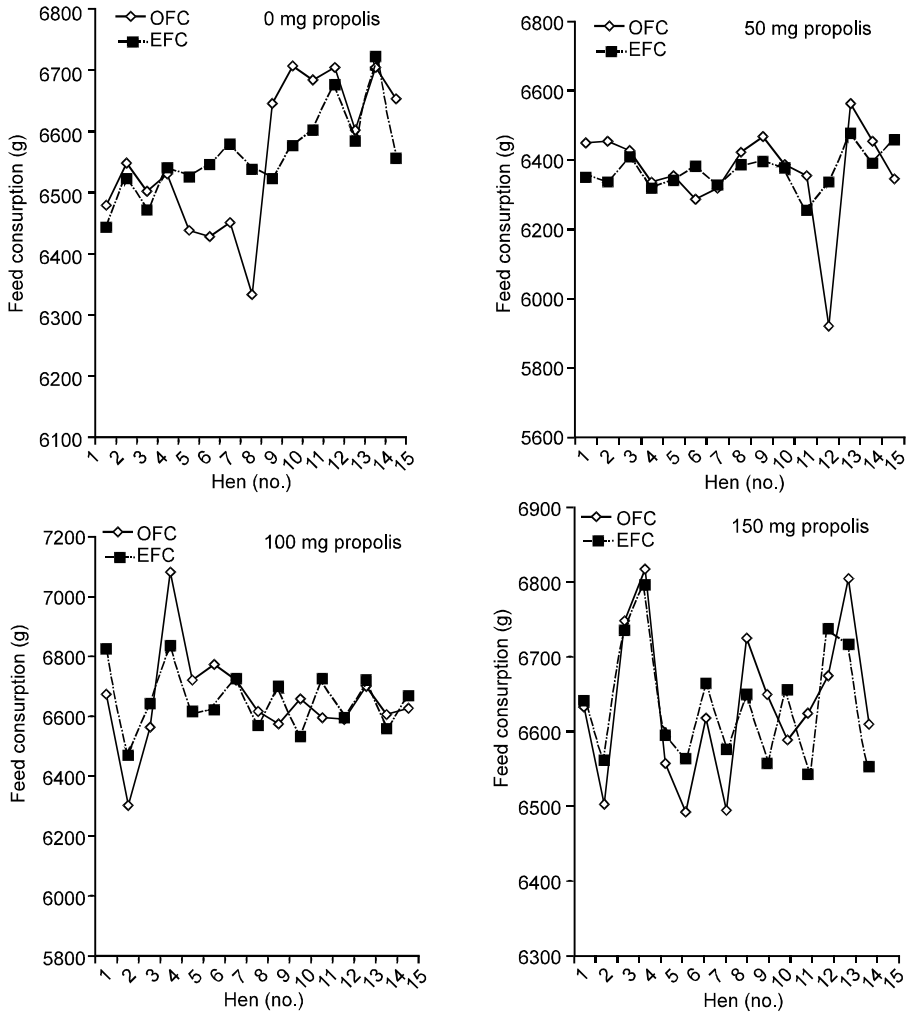

Fig. 1: Observed vs. expected feed consumption of laying hens fed different levels of propolis

acceptable alternative in a breeding program of no reliable estimates of genetic correlation are available (Luiting and Urff, 1991). This suggestion was reported because the author found the estimates of genetic correlation of RFC with the economic traits did not clearly differ from zero. Also, Tixier-Boichard *et al.* (1995) found that the genetic correlation between RFC and the independent variables used in the prediction equation (metabolic body weight, change in body weight and egg mass) were generally low, which confirms the validity of the selection on a phenotypic assessment of RFC. Data presented in Table 3 showed that the correlation coefficients for some quantitative traits with Residual Feed Consumption (RFC) and cell mediated immunity. The wattle swelling measured at all times were negatively correlated, with statistically significant at 48 h post PHA-P injection, with RFC in control-group. Similar, trend was observed in 50 mg supplemental Propolis. The wattle swelling measured at 24, 48 and 72 h post PHA-P injection were significantly positive correlated with residual feed consumption in supplemental Propolis at 100 mg. Similar trend was observed in 150 mg supplemental Propolis laying at 72 h post PHA-P injection. With respect to white blood cells differentiation, it could be noticed that significantly negative relationship between RFC and heterophils percentage in all treatment groups. Inversely, the relationship between RFC and lymphocytes percentage was significant and positive in all treatment groups. Van Eerden *et al.* (2004) concluded that a population of chickens from a commercial breed shows considerable variation in RFI. Specific antibody production against KLH, *M. butyricum* and *S. enteritidis* lipopolysaccharide, however, is not influenced by efficiency in terms of RFI. R+ animals may have a higher level of non-antigen specific antibodies, as indicated by the higher antibody response to *Salmonella* protein.

Heterophils count was significantly negative correlated with residual feed intake in all treatment groups. Conversely, there was significantly positive relationship between lymphocytes count and residual feed consumption in laying hens fed diet containing 100 or

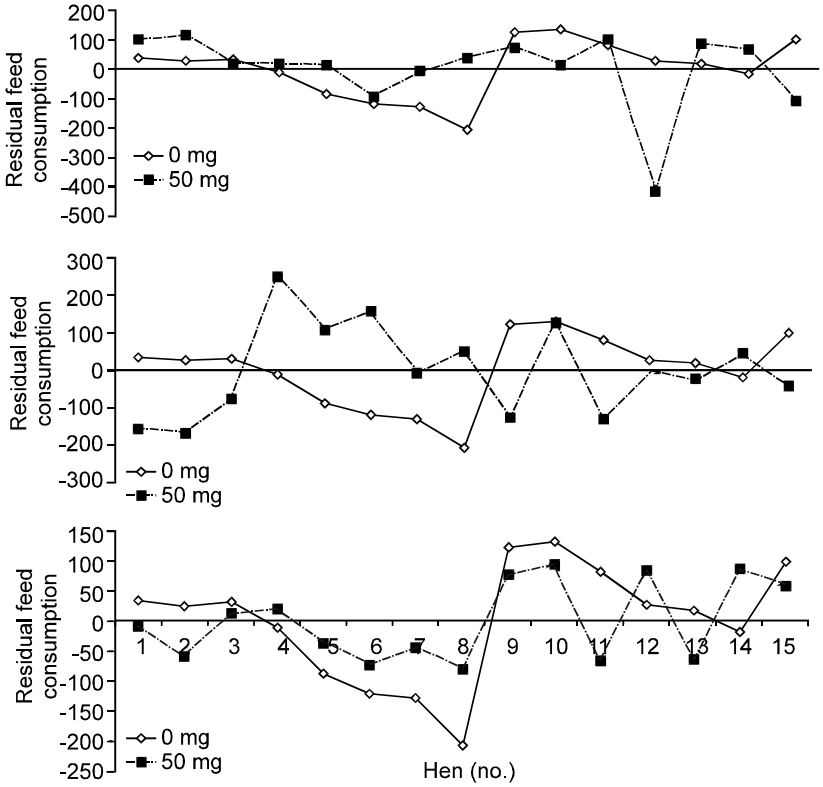

Fig. 2: Residual feed consumption of laying hens fed different levels of propolis

Table 3: Phenotypic correlation coefficients among residual feed consumption, cell-mediated immunity and some productive parameters

	Propolis (mg/kg)			
	0	50	100	150
Cell mediated-immunity				
24 h post PHA-P injection	-0.09	-0.11	0.50*	0.40
48 h post PHA-P injection	-0.58*	-0.13	0.52*	0.10
72 h post PHA-P injection	-0.09	-0.32	0.56*	0.53*
White blood cells				
Heterophils	-0.51*	-0.66**	-0.70**	-0.68**
Lymphocytes	0.45	0.41	0.67**	0.70**
Hematological parameters				
Total plasma protein	0.14	0.27	0.25	0.31
Globulin	-0.26	-0.41	-0.35	-0.51*
Cholesterol	0.20	0.31	-0.45	-0.37
Productive parameters				
Observed feed consumption	0.81***	0.92***	0.76**	0.63**
Feed conversion ratio	0.58*	0.93***	0.62**	0.79**
Egg number	0.40	0.58*	0.48	0.44
Egg weight	-0.07	-0.05	0.06	0.04
Albumen (%)	0.38	0.14	-0.13	0.18
Yolk (%)	0.53*	0.60**	0.68**	0.59**
Shell (%)	-0.48	-0.09	-0.22	-0.15
Shell thickness	0.46	0.38	0.33	0.23
Breaking strength	0.39	0.30	0.14	-0.05

*p<0.05, **p<0.01, ***p<0.001

150 mg Propolis level. Similar trend, but not significant, was noticed in 0 or 50 mg Propolis level. There was significant negative relationship between plasma globulin and residual feed consumption in laying hens fed diet containing 150 mg Propolis. Similar trend, but not statistically significant, was noticed in remaining groups. Both observed feed consumption and feed conversion ratio was significantly positive correlated with RFC in all treatment groups. Positive relationship between egg number and RFC was observed in all groups. Tixier-Boichard *et al.* (1995) indicated that positive correlation between RFC and egg number. Significant positive relationship between yolk percentage and RFC was observed in all treatment groups. Similar result was obtained by Fathi and Galal (2007). They reported that the yolk percentage was significantly positive correlated with RFC in both brown and white egg-type strains. El-Sayed and El-Hakim (1994) calculated significant and high positive association between RFC and the yolk percent in full-sib normal and dwarf hens. The author showed that the positive correlation probably reflects the increase in dry matter percentage and energy content in the egg when the proportion of yolk increases.

REFERENCES

Bonomi, A., F. Morletto and M. Binachi, 1976. Propolis in feeds for laying hens. Avicoltura, 54: 43-54.

Buhatel, T., S. Vesa, A. Dimitrin and I. Moldovan, 1983. Contribution to knowledge of the stimulative effect of propolis on piglet and pullets. Buletinul Institului Agronomic Cluj Napoca Zoot. Med. Vet., 37: 45-48.

Chandra, R.K. and P.M. Newberne, 1977. Nutrition, immunity and infections. Plenum Press, New York.

Cheng, S. and S.J. Lamont, 1988. Genetic analysis of immunocompetence measures in a white Leghorn chicken line. Poult. Sci., 67: 789-995.

El-Sayed, T.M. and A.E. El-Hakim, 1994. Residual feed consumption. The Second Scientific Conf. Poult. Sep. Kafr El-Sheikh, Egypt, pp: 52-67.

Fathi, M.M. and A. Galal, 2007. Estimating residual feed consumption in two genetic lines of egg-type chicken under Egyptian environmental conditions. 4th World's Poultry Conference, Sharm El-Sheikh, March, 27-30, pp: 187-198.

Fathi, M.M. and E.A. EL-Sahar, 1996. Determining the strength of eggshell by using an appropriate apparatus and an equation to calculate egg surface depending on its dimensions. Egypt. Poult. Sci., 16: 285-303.

Forbes, J.M., 1995. Voluntary food intake and diet selection in farm animals, CAB International, Wallingford, U.K.

Gabrys, J., Z. Konecki, W. Krol, S. Scheller and J. Shani, 1986. Free amino acids in bee live product (propolis) as identified and quantified by gas-Liquid chromatography. Pharmacol. Res. Commun., 18: 513-518.

Ghisalberti, E.L., 1979. Propolis: A review. Bee World, 60: 59-84.

Giurgea, R., V. Toma, H. Popescu and C. Linicencu, 1981. Effects of standardized propolis extracts on certain blood constituents in chickens. Clujul-Med., 54: 151-154.

Goto, N., H. Kodama, K. Okada and Y. Fujimoto, 1978. Suppression of phytohemagglutinin skin response in thymoectomized chicken. Poult. Sci., 57: 246-250.

Gross, W.B. and P.B. Siegel, 1983. Evaluation of the heterophil/lymphocyte ratio as a measure of stress in chickens. Avian Dis., 27: 972-979.

Gross, W.B. and P.B. Siegel, 1985. Selective breeding of chickens for corticosterone response to social stress. Poult. Sci., 64: 2230-2233.

Hegazi, A.G., H.F. El-Miniawy and F.A. El-Miniawy, 1995. Effect of some honeybee products on immune response of chicken infected with Virulent NDV. Egypt. J. Immuol., 2: 79-86.

Hegazi, A.E., F.A. El-Miniawy and F.K. Abd El-Hady, 1996. Influence of administration of propolis on chicken immune status. Egypt. J. Immunal., 3: 111-116.

Hussein, S.H., M.M. Fathi and A. Galal, 2000. Residual feed consumption in three local genotype of chicken segregating for naked neck gene. Egypt. Poult. Sci., 20: 311-326.

Katanbaf, M.N., E.A. Dunnington and P.B. Siegel, 1989. Restricted feeding in early and late-feathering chickens 1: Growth and physiological responses. Poult. Sci., 68: 344-351.

Klasing, K.C., D.E. Laurin, R.K. Peng and D.M. Fry, 1987. Immunologically mediated growth depression in chicks: Influence of feed intake, corticosterone and interleukin-1. J. Nutr., 117: 1629-1637.

Kreukniet, M.B., M.G.B. Nieuwland and A.J. van der Zijpp, 1994. Phagocytic activity of two lines of chickens divergently selected for antibody production. Vet. Immunol. Immunopathol., 44: 377-387.

Luiting, P. and E. M. Urff, 1991. Optimization of a model to estimate residual feed consumption in the laying hen. Livest. Prod. Sci., 27: 321-338.

Martin, A., E.A. Dunnington, W.B. Gross, W.E. Brilers, R.W. Briles and P.B. Siegel, 1990. Production traits and allonantigen systems in lines of chickens selected for high or low antibody response to sheep erythrocytes. Poult. Sci., 69: 871-878.

Maxwell, M.H., 1993. Avian blood leukocyte responses to stress. World's Poult. Sci. J., 49: 34-43.

McCorkle, F., I. Olah and B. Glick, 1980. The morphology of the phytohemagglutinin-induced cell responsein the chicken'sw attle. Poult. Sci., 59: 616-623.

National Research Council, 1994. Nutrient Requirements of Poultry. 9th Rev. Edn. National Academy Press, Washington, DC.

Qureshi, M.A. and G.B. Havenstein, 1994. A comparison of the immune performance of a 1991 commercial broiler with a 1957 randombred strain when fed "typical" 1957 and 1991 broiler diets. Poult. Sci., 73: 1805-1812.

Robinson, F.E., M.W. Wilson, G.M. Yu, G.M. Fasenko, and R.T. Hardin, 1993. The relationship between body weight and reproductive efficiency in meat-type chickens. Poult. Sci., 72: 912-922.

SAS Institute, 2001. SAS/STAT User's Guide Version 8.2 ed: Statistics. SAS Institute Inc., Cary, NC.

Shalmany, S.K. and M. Shivazad, 2006. The effect of diet Propolis supplementation on Ross broiler chicks performance. Int. J. Poult. Sci., 5: 84-88.

Stadeckerm, J., M. Luk, C.A. Dvorak and S. Leskowitz, 1977. The cutaneous basophil response to phytohemagglutininin chickens. J. Immunol., 118: 1564-1568.

Tixier-Boichard, M., D. Boichard, E. Groeneveld and A. Bordas, 1995. Restricted maximum likelihood estimates of genetic parameters of adult male and female Rhode Island Red chickens divergently selected for residual feed consumption. Poult. Sci., 74: 1245-1252.

Van Eerden, E., H. Van Der Brand, G.D.V. Reilingh, H.K. Parmentier, M.C.M. DeJong and B. Kemp, 2004. Residual feed intake and its effect on Salmonella enteritidis, infection among growing layer hens. Poult. Sci., 83: 1602-1609.

Williams, K.C., 1997. Some factors affecting albumen quality with particular reference to Haugh unit score. World's Poult. Sci. J., 48: 5-16.

Zulkifli, I., E.A. Dunnington, W.B., Gross and P.B. Siegel, 1994. Food restriction early or later in life and its effect on adaptability, disease resistance and immunocompetence of heat-stressed dwarf and nondwarf chickens. Br. Poult. Sci., 35: 203-213.