Egypt. Poult. Sci. Vol (37) (I): (213-231) (2017) **(1711)**

Egyptian Poultry Science Journal

http://www.epsaegypt.com

ISSN: 1110-5623 (Print) – 2090-0570 (On line)

EFFECT OF BEE POLLEN AT DIFFERENT LEVELS AS NATURAL ADDITIVES ON IMMUNITY AND PRODUCTIVE PERFORMANCE IN RABBIT MALES

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Received:16/01/2017 Accepted:02/03/2017

ABSTRACT: This study was designed to evaluate the effect of using bee pollen (BP) at different levels as a natural growth promoter on the productive performance, digestibility, carcass traits, caecal microbial activity and some blood parameters of growing rabbits. Sixty New Zealand White (NZW) male rabbits, 5 weeks old were randomly divided into four equal groups, with 695.75± 10.01 g BW. In the 1st group, rabbits were fed without any supplementation and considered as control group. While, those in the 2nd, 3rd, and 4th groups were daily treated with BP at 200, 500 and 700 mg/kg BW, respectively and served as low, medium and high treatment levels. From these findings could be noted that the growing rabbits treated with BP significantly ($P \leq 0.05$) increased final body weight and total weight gain and decreased feed intake (DM) as well as improved in feed conversion as compared with control group. Both of digestibility coefficients and nutritive values of treated rabbits were significant ($P \leq 0.05$) improved than that of the control. The dressing weight, carcass weight and kidney fat were significantly ($P \leq 0.05$) increased with increasing BP levels.

The immunoglobulin concentration (total antioxidants capacity, IgG, IgM and IgA) increased significantly ($P \leq 0.05$) with increasing BP levels. The amylase, lipase, and protease activities were significantly ($P \leq 0.05$) increased in the pancreatic homogenate and intestinal contents in the rabbits treated with high levels of BP as compared to control group. The best economical efficiency was obtained at the high BP level compared to other treatment groups.

Conclusively, from the obtained results it could be concluded that the supplementation of BP at any levels studied had positive effects on rabbit's performance, blood lipid regulation which reflecting their antioxidant of growing rabbits.

Keywords: Bee pollen, rabbits, performance, carcass, digestibility, blood parameters.

INTRODUCTION

In recent years, commercial rabbit production has been gaining much attention due to their rapid growth, small body size, as well high prolificacy and meat production. Therefore, rabbits can convert about 20% of the protein in their diet to meat, which is higher (8-12%) than beef (Basavaraj et al., 2011). Nowadays, it's well known that the feed additives supplementation can be used safely in rabbit rations to improve their performance, which added to the diets with small quantities. There are several properties of bee pollen (BP) in the nutritional and medical purposes as an anti-aging substance and antioxidant to improve animal's immune functions and the growth (Tu et al. 2015). The positive effect of feed additives such as BP can be expressed through better appetite and improved feed conversion as well as immune system activation and increased vitality (Perić, et al., 2009). In last two decades, BP has been used as antibiotics substitutes to improve the performance and immune response in the animals (Wang et al., 2007 and Attia et al. 2011b).Bee pollen is an agglomerate of pollen grains, which collected by bee workers and then mixed with nectar and the secretion from the hypo pharyngeal glands such as β-glycosidase enzymes. Bee-pollen used as a natural growth promoter due to rich source of proteins, carbohydrates, vitamins, lipids and minerals in addition to flavonoids, carotenoids, enzymes or coenzymes, antioxidant substances, polyphenolics as flavonoids and carbohydrates, polyphenols and other healthy compounds and digestive enzymes added by the bees (Khojasteh and Shivazad, 2006, Wang et al., 2007 Attia et al., 2014a and Taha, 2015). Bee pollen contains 10-40% proteins, 13-55% carbohydrates, 1-20% lipids, 0.5-3% minerals, 0.02-0.1% vitamins, 0.04-3% flavonoids, 3-8% water and other compounds such as resins and antibiotic substances (Carpes et al., 2007 and Taha, 2015).Bee pollen has been used as antibiotics substitutes in the rabbit rations, medical science, and bio-cosmetology because of its biological properties such as antioxidant, antimicrobial and antiseptic activities, apitherapy and antiviral, anti-fungal, antibacterial, anti-inflammatory, antitumour, anti-ulcer and immunity activation and local anaesthetic properties (Song et al., 2005 and Hajkova et al., 2013). The findings of Attia et al. (2011a and 2014a) showed that bee pollen increased body weight gain and survival rate as well as improved feed intake and feed conversion ratio of offspring up to 12 weeks of age. Also, Hedia, et al. (2007) showed that treatment with BP caused marked increases in plasma total protein, albumin, and glucose. Similarly, El-Hanoun et al. (2007) and Attia et al. (2011b) found that growing rabbits supplemented with BP at 250 and 500 mg/kg BW significantly increased growth and survival rates from weaning till mature age. El-Neney and El-Kholy (2014) and Zeedan and El-Neney (2014) reported that improved rabbits performance, BWG, oxidative status, immunity response and reduced feed intake and improved FCR. Also, they found an improvement of immunity response, blood biochemical, economic efficiency, digestibility coefficient and nutritive values. Therefore, study was designed to investigate the effect of supplementation of BP as feed additives on the productive performance, digestibility, carcass traits, cecum microbial activity and blood biochemical as well as certain digestive enzymes activity in pancreatic tissue and intestinal content and economic efficiency of growing NZW rabbits.

MATERIALS AND METHODS

Animals and experimental design:

This study was carried out at the Industrial Rabbitry, Tohoria village belonging to near Shebeen El-Kanater city, Qalubia Province, Egypt. Bee pollen (mixture of Maize, Clover and Bean) was obtained from an apiary belonging to Agriculture Research Center, Plant Protection Institute, El-Qaunatir, Qalyoubia Governorate.

A total of 60 healthy male New Zealand White (NZW) rabbits, with 695.75 ± 10.01 g BW, 5 weeks of age were raised under normal managerial and hygienic conditions throughout experimental period from 5 to 15 weeks of age. Rabbits were individually raised in wire galvanized battery cages supplied with feeders and automatic stainless steel nipples. Fresh drinking tap water was automatically available all the time in each cage.

All rabbits were randomly divided into four equal groups. In the 1st group, rabbits were fed standard ration without any supplementation and served as control, while the 2nd, 3rd, and 4th groups were daily offered BP at 200, 500 and 700 mg/kg BW and considered as low, medium and high doses treatments.

All rabbits were fed ad-libitum on the commercial pellets ration contained 17.20 % crude protein, 2.69 % crude fat, 14.10 % crude fiber and 2520 DE, Kcal /kg diet (Table 1) according to NRC (1977). Rabbits in treated groups were daily orally given BP as the water suspension.

Interior ambient temperature (°C) and relative humidity (%) in the rabbitry farm were measured four times each month. Averages of ambient temperature, relative humidity were 30-32°C and 50-65%, respectively.

Productive performance:

The rabbits in all groups were individually weighed, while weight gain (BWG) was calculated by subtracting the initial live body weights from final ones of each period. Feed consumption was weekly recorded, while feed conversion ratio (FCR) was calculated as g feed/g gain. Both of protein and digestible energy intakes averages were measured and calculated by multiplying the feed intake by the percent of protein and digestible energy contents of experimental diets.

Digestibility trial:

At the end of the experiment, six males from each group were taken in digestibility trials over the period of 7 days to estimate both of nutrient digestibility coefficients and nutritive values of the tested diets. Both of feed intake and feces for males were daily weighed and recorded. The representative feces samples were dried at 60°C for 48 hrs ground and stored for analysis. The feed and feces were chemically analyzed according to AOAC (2000). The digestible crude proteins (DCP) as well as total digestible nutrients (TDN) values were calculated according to the formula of Cheeke et al., (1982). The digestible energy was calculated by using the following equation: DE (kcal/kg) =TDN × 44.3 according to Schneider and Flatt (1975).

Carcass characteristics:

At the end of the experiment, six rabbits per each group were randomly taken to determine carcass characteristics. Before slaughtering, all rabbits fasted for 12 hours and then individually weighed as pre-slaughter weight. The fasted rabbits were slaughtered by cutting the jugular vein. The slaughter rabbits were weighed, skinned and eviscerated after complete bleeding. Also, the carcass, giblets edible parts (liver, heart, and kidney), cecum, and kidney fat were immediately weighed. All carcass traits were calculated as the percentage of the pre-slaughter weight.

Cecum samples:

Samples of cecum content were taken individually from six rabbits of each group and cecum contents were obtained after slaughtering and filtrated to estimate pH and cecum micro flora. Total bacterial count of E. coli was estimated according to Collins et al. (1995). Ammonia-nitrogen and total volatile fatty acids (VFA's) content in cecum samples were measured according to Ahmed (1976) and AOAC (2000), respectively.

Blood parameters:

At the slaughter blood samples (six representative rabbits from each treatment) were taken in heparinized tubes and then centrifuged at 3000 rpm for 20 minutes and kept at -20 C until analysis. Plasma total protein, albumin, total lipids, glucose, triglycerides, urea-N concentration, cholesterol, and creatinine were determined using specialized commercial kits. Also, plasma IgG, IgM, and IgA and total antioxidant capacity (mmol/L) were determined using commercial kits.

Enzymes activity:-

The pancreas and GIT segments sampling procedure was estimated by using the method according to Uni et al. (1999). Both of pancreatic and tissue samples were homogenized in ice-cold 0.2 MTris -HCl buffer, pH 8.0, containing 0.05 M NaCl in the ratio 1:4 (wt. /vol.). The homogenate sample was centrifuged at 3000xg for 15 minutes at 4°C, the supernatant was stored frozen (–70°C) for enzymes assay. The homogeneous intestinal digesta samples were collected by massaging the tract from the distal end of the duodenum to the ileocecal junction according to the method of Jin et al. (2000).

All samples were diluted 10-folds, based on the weight of the sample, with ice-cold PBS (pH 7.0), and homogenized for one minute and solicited for 1 min with 3 cycles at 30-s intervals. All samples were centrifuged at 18000x g for 20 min at 4°C. The supernatants were classified into small portions and then stored at -70°C to enzymes assay. Amylase activity in sera, tissues, and digesta was determined using the method of Somogyi (1960). The activity of lipase was assayed by using the method described by Tietz and Fiereck (1966), while the activity of Protease was analyzed by using the method of Lynn and Clevette-Radford (1984).

Statistical analysis:

The obtained data were statistically analyzed by using SPSS (2012) computer program using the following fixed model: $Y_{ij}=\mu + T_i + e_{ij}$

Where: Y_{ij} = The observation; μ = Overall mean; T_i = Effect of treatments groups; e_{ij}= Random error component assumed to be normally distributed. Duncan's multiple range tests was performed (Duncan, 1955) to detect significant differences among means.

RESULTS AND DISCUSSION

Chemical composition of bee pollen:-

The chemical composition of BP (Table 2) indicate that BP is a rich source of carbohydrates (61.71%), proteins (29.94%), Fiber (1.17%), ash (2.83%) and fats (4.35%) on DM basis. The most prevalent amino acids in BP were leucine, alanine, glutamine, arginine glycine, aspartic acid, valine and lysine and minerals (e.g., Na, Ca, Mg, P, Zn, Mn and Fe).

Productive performance:

Data presented in Table 3, showed that the rabbit in treated groups had significantly ($P \leq 0.05$) increased body weight and weight gain compared to control group. This increase in body weight and weight gain could be attributed to improving their crude protein digestibility, which led to improving nutrient and protein utilization and resulting in higher protein anabolism and intestinal absorptive capacity. These positive improvements could be due to the nutritive value of the bee-pollen as a rich source of protein (29.94%), essential amino acids (e.g., Leucine, Alanine, Glutamine and lysine), fat (4.35%), carbohydrates (61.71%) and minerals (e.g., Na, Ca, Mg, P, Zn, Mn and Fe) as presented in Table 2. These results agree with those of El-Hanoun et al. (2007), Wang et al. (2007), El-Neney and El-Kholy (2014) and Soha and El-Rayes (2016). Similarly, the findings of Babaei et al. (2016) showed that addition of 5000 mg kg^{-1} pollen could be beneficial in improving the performance of quail chicks. Contrary, the findings of Dias et al., (2013) showed indicated that BP supplementation of growing rabbits was not sufficient to improve the performance from weaning up to slaughter age.

Feed intake and feed conversion:

The means DM, protein intake and digestible energy intakes in treated groups were slightly decreased ($P<0.05$), while no significant effects DCP and TDN intakes than that of the control (Table 3). Feed intake for males in treated groups was lowered ($P\leq0.05$) by about 4.41 and 5.73% for medium and high doses compared to control one.

This result may suggest that the use BP in diets for growing rabbits might enhance the growth of lactic acid fermenting bacteria in the gut and improved the food digestibility and utilization of ammonia due to BP supplementation. The decreased feed intake in rabbits supplemented with BP could be explained by the increase in nutrients such as minerals and vitamins could accelerate nutrients metabolism and increase energy digestibility. These results agree with the findings of El-Neney and El-Kholy (2014), Zeedan and El-Neney (2014) for rabbit and Soha and El-Rayes (2016) and Babaei et al. (2016) for Japanese quails. El-Hanoun et al. (2007) indicated that the feed efficiency was improved of growing NZW rabbits supplemented with low doses (250) and high doses (500) mg BP per kg BW compared to control group during winter and summer seasons from weaning up to mature age. On contrary, the results of Han et al. (2010) and Haščík et al. (2012) indicated that bee-pollen increased feed intake of broiler chickens.

The findings in Table3 indicated that feed conversion (g intake/ g gain) of DM, DCP and TDN were improved of growing rabbits supplemented with different levels of bee pollen compared to control. The improvement in feed conversion of treated rabbits may be due to the role of BP in decreasing NH_3–N, pathogenic bacteria counts and increasing nutrients digestion. Also, this may be due to high levels of amino acids, vitamins, minerals in bee pollen as well as enzymes and coenzymes, which added by bees during formation of BP pellets, which play important role in good digestion and cell growth. These findings agree with those of Wang et al., (2007), Hedia et al. (2007) and Zeedan and El-Neney (2014).

Generally, the improvement in growth performance of rabbits supplemented with BP could be reflecting better absorption of amino acids or/and due to antibacterial properties and enzymes or coenzymes of Bee pollen.

Mortality rate (%):

Data presented in Table 3 showed no mortality rate in treated rabbit groups, which decreased remarkably by about 13.3% than that of the control. The decreased mortality rate may have been due to the decreased pH, NH_3–N; pathogenic bacteria count in the cecum leading to decreasing diarrhea of rabbits. Also, increased survival rate in treated groups could be due to enhancing immune functions, promote the growth of an animal, protect the intestinal tract health and consequently improve animal products quality and security (Liu et al., 2010 and Attia et al., 2011a, b). Also, BP itself has been improve of immune system also, showed to exhibit antifungal, anti-inflammatory, antibacterial activity, and/or an antimicrobial agent (Proestos et al., 2005 and Hajkova, et. al., 2013). These findings agree with those of El-Neney and El-Kholy (2014), they found that the mortality rate in rabbits treated with BP decreased remarkably than that of the control.

Digestibility and nutritive values:

Data presented in Table 4 indicated that nutrient digestion coefficients and nutritive values were significantly ($P<0.05$) higher in treated rabbits than control. The highest values of digestibility coefficients in BP for almost all the tested nutrients suggest more efficient utilization of the nutrients in the intestinal tract, which leads to more favorable for feed conversion ratio compared to rabbits in the control group.

These results agree with El-Neney and El-Kholy (2014), Zeedan and El-Neney (2014) and Tu, et al. (2015). Also, the findings of Khojasteh and Shivazad (2006) and Wang et al., (2007) indicated that the highest digestibility of all nutrients in treated groups can be discussed from the point that BP contains digestive enzymes from the bees may be associated with improvement in digestibility of all nutrients. The increased digestibility may be due to the reduction in feed intake that occurred, therefore could be documented that the digestibility of the diet increases with decreasing intake (NRC, 1977). So, the improved digestibility of nutrients and nutritive value may be reflected in better growth performance, while the increased DCP could be attributed to the improvement of CP digestibility. The findings of Wang et al. (2006) showed that the layer hens supplemented with BP at 1.5% in the diet improved organizational structure of the digestive tract leads to enhancing the digestion and absorption. Also, Song et al. (2005) found that broilers supplemented with 1.5% bee pollen in diets promoted the early intestine development, enhanced immune functions and promoted liver, pancreas, and small intestine development.

Carpes et al. (2007) reported that BP may cause the better absorption of amino acids because consists of 11 enzymes or coenzymes or/and due to antibacterial properties of BP. Also, the findings of Hajkova et al. (2013) indicated that the addition of BP in diet had demonstrable concentration-dependent effects on the mucosa of the small intestine, which have a positive effect on improving the absorptive mucosal surface and therefore could affect the usability for the received nutrients in food. Leja et al. (2007) and Saric et al. (2009) reported that the positive effect of BP on BW and BWG can be due to the presence of a lot of nutrient factors (as antioxidants, vitamins, mineral, essential fatty acids, vitamins, amino acids, enzymes etc.) and protective agents, such as flavonoids, carotenoids, and phenolic constituents in the BP which are able to improve nutrient value of the feed as well as feed digestibility and absorption.

Results in this study showed that the use of supplemented BP improved nutrients digestibility, absorption as well as the characteristics of intestinal morphology, this response can lead to increased feed utilization and improved performance. Amino acids, vitamins and trace elements of BP are nutritionally beneficial for improving intestinal absorption due to the stimulus intestinal cells development, differentiation and proliferation and because they improve the environmental conditions for the intestinal microbial ecosystem. Also, these findings suggested that these supplements render the feeds more available for utilization by positively affecting the microflora population and improving feed utilization through slowing feed rate of passage through the digestive tract, which reflecting the better absorption.

Carcass characteristics:

Data presented in Table 5 showed that the rabbits treated with BP had the heaviest mean carcass characteristics than the control group. The significant ($P<0.05$) increase in carcass traits for treated groups may be mainly related to the increase in growth performance and digestibility of treated rabbits, which use as an indicator for good healthy status of rabbits. These findings agree with those of Haščík, et al. (2012) and El-Neney and El-Kholy (2014). In contrast, the findings of Dias et al. (2013) and Attia et al. (2014b) showed that the carcass yield of rabbits was not influenced by BP supplementation.

The relative weight of heart, kidney, liver, and cecum in rabbits received a bee pollen were insignificantly increased than that of the control. These findings agree with Zeedan and El-Neney (2014) and Hosseini et al. (2016). The lowest kidney fat percentage was obtained in treated groups may be due to the lower digestible energy intake by rabbits compared to the control. This decrease could be attributed to the reduction in plasma total lipids as well as cholesterol. These results agree with those of Botsoglou et al., (2010), who found that the decreased fat content reflect bee pollen flavonoids content, which led to decreased plasma lipid levels and improved glucose tolerance as well as attenuate obesity.

Cecum traits:-

Data presented in Table 6 indicated that cecal pH value and E. coli count for treated groups were significantly ($P<0.05$) decreased than that of the control (Table 6). The decreased cecal pH and E. coli count for rabbits treated with BP could be attributed to more fermentation of the caecal leading to reduce caecal pH as a result of increasing volatile fatty acids production. Also, cecal pH value in rabbits depends on some factors such as the diet amount and composition. The fluctuations in the pH value could reflect the changes of organic acids accumulated in the ingesta.

These results agree with those of El-Neney and El-Kholy (2014) and Zeedan and El-Neney (2014). Therefore the BP supplementation that is probably the most interest to rabbit's producers is as an antifungal and antimicrobial, which exhibiting the highest scavenging capacity and antioxidants activity tend to be those that contains high levels of flavonoids as well as phenolic acid derivatives (Proestos et al., 2005 and Carpes et al., 2007).

Referring to cecal VFAs concentrations, these findings indicated that the cecal VFAs concentrations in treated groups were increased in comparison with control group, that point to greater cecal microbial activity. The total VFA concentration in rabbit received high BP significantly ($P<0.05$) increased than that of the control. The increased total VFA could be attributed to the decreased pH, NH3–N and increased the digestible protein and TDN with high BP (Table 4). These results are in accordance with those of El-Neney and El-Kholy (2014), who found that the addition BP in growing rabbits reduced the numbers of the total bacterial count in cecum content and increasing TVFAs as well as decreasing pH and NH3-N. When absorbed, VFAs produced in the cecum can cover about 40% of rabbit maintenance requirement so; higher VFAs production could be beneficial with regard to better energy supply and better body weight gain as a consequence. Additionally, VFA provide the main metabolic fuel for the mucosa of the large intestine (Marty and Vernay 1984). Also, the results of Falcao, et al. (2007) showed that the supplementation of rabbit rations with BP increases volatile fatty acids in the cecum of rabbits and decreasing the concentration of cecum ammonia. Also, decreasing the concentration NH3–N may be attributed to the conversion of ammonia-N into microbial protein for the benefit of rabbits which characterized by the pseudo-rumination and improvement in the digestion of nutrient as shown in Table 4. The VFAs are actively absorbed through the cecal and colonic walls and utilized by the rabbit as energy sources, as in ruminants. Results of cecum activity indicated that supplementation of BP improved the microbial fermentation by increasing utilizing ammonia nitrogen and FVAs and reducing pH values as compared to the control diet.

Biochemical parameters:-

Plasma blood constituents:-

Data presented in Table 7 indicated that plasma total protein, albumin, and globulin concentrations in treated rabbits increased significantly ($P<0.05$) than that of the control. The increased blood proteins in rabbits received a high dose of BP may be associated with improvement of crude protein digestibility as well as to the high level and good quality of protein contents in bee pollen. These results are in agreement with the findings of Hedia et al. (2007), Zeedan and El-Neney (2014) and Attia et al. (2015) they noted that the supplementation of bee pollen for rabbits has a positive effect on blood proteins. Also, Attia et al. (2014a) and Soha and El-Rayes (2016) observed the same trend in broiler chicks.

Referring to glucose level, these findings showed that plasma glucose concentration increased significantly ($P<0.05$) for treated groups than that of the control. The increased plasma glucose in treated groups may be reflecting the increasing energy availability (sugars) for the physiological and biochemical functions. Also, Wei et al., (2008) stated that the increased plasma glucose in treated groups could be attributed to increased pollen from the glucose contents (8.2 -13.1), sucrose (15.8 - 18.4%) and fructose (19.9%). These findings agree with those of Zeedan and El-Neney (2014) and Attia et al. (2015).

Plasma lipid functions:-

Data presented in Table 7 showed that the plasma total lipids, cholesterol and triglycerides levels in treated groups significantly ($P<0.05$) decreased than that of the control group. The decreased triglycerides and cholesterol values may be due to unsaturated fatty acids in BP as oleic, linoleic and linolenic (14.20, 10.14 and 16.64%, respectively), that play role in inhibits accumulation of lipid peroxidation product(Soha and El-Rayes, 2016).

The significant decreased in plasma cholesterol of growing rabbits supplemented with BP could be attributed to reduced absorption and/or synthesis of cholesterol in the gastrointestinal tract by BP supplementation. Similarly, the decreased cholesterol levels in treated groups may be directly related to the influence of BP on lipid metabolism. These results are in agreement with the findings of Zeedan and El-Neney (2014), Attia et al. (2015) and Soha and El-Rayes (2016) who found that total lipids and cholesterol significantly ($P<0.05$) decreased in rabbit or broiler treated with BP.

Kidney functions:-

Data presented in Table 7 revealed that the urea and creatinine levels were significantly ($P<0.05$) lower in the treated groups than that of the control. The decreased plasma urea and creatinine with increasing BP levels may be an interaction between BP and the activity of some microbial strains. It can be hypothesized that BP has interfered to some extent on decreasing the harmful bacteria and increase protein synthesis with a consequent reduction in ammonia production and/or utilization. These findings are in accordance with those of El-Hanoun et al. (2007), Attia et al. (2014b) and El-Neney and El-Kholy (2014), they found that the rabbits fed diet supplemented with BP reduced urea-N and creatinine concentrations as compared with the control group. In contrast, the findings of Attia et al. (2014b) showed that bee pollen had significantly increased creatinine concentration, while plasma urea was significantly ($P<0.05$) decreased.

Immunological parameters and Immunoglobulin concentration:-

As shown in Table 7 the lowest (1.34 mmol/l) total antioxidants capacity was recorded in control group, while highest values (1.84, 2.87 and 3.90 mmol/l) were obtained in treated groups with significant differences.

The increased total antioxidants capacity in treated groups may be due to amino acids and vitamins as well as trace elements of bee pollen absorption through the stimulation of intestinal cells development, proliferation and differentiation, which improve the environmental conditions for the intestinal microbial ecosystem. Also, BP can improve the cell immune responses, antibody production speed and reinforce the immunological system (Song et al., 2005). These results agree with those of Attia, et al. (2014b), Zeedan and El-Neney (2014) and El-Neney and El-Kholy (2014) they found that the addition of BP caused a significant increase of total antioxidants capacity as compared to control diet.

Regarding immunoglobulin (IgG, IgM, and IgA) concentrations in the plasma of rabbits (Table 7), the concentrations of IgG, IgM and IgA of rabbits were higher for rabbit supplemented with BP at high and medium doses followed by low doses, while the lower concentrate was recorded in the control group. The improved blood components as a result of treatment with BP may be due to improvement in the immune response (minerals and flavonoids have a role in enhancing immune system). These results agree with those of Attia et al., (2011a, b) who found that the immune functions for rabbits treated with bee pollen enhanced than that of the control.

Enzymes activity:

Data presented in Table (8) showed that the activity of lipase, amylase and protease were significantly ($P<0.05$) increased in pancreatic homogenate and intestinal contents of treated rabbits than that of the control. This improvement at high BP level on the activity of enzymes may be due to the supplementary role on the overall digestive kinetic action causing an enhancement of the titers of digestive enzymes in pancreatic tissue and intestinal content. Also, another possible for increased enzymatic activity could be the 'favorable change in pH of GI tract' mediated by exogenous enzymes (Abdel-Rahman et al., 2010). These results agree with those of Attia et al. (2012), who stated that the activity of enzymes for growing rabbit supplemented with BP in the diets with exogenous digestive enzymes (Kemzyme) than that of the control group. Moreover, the results of Murai et al., (2000) reported that the increased availability of nutrients in the GIT that caused increased the activity and secretion of the digestive enzymes from the glandular mother cells as a positive feedback response which stimulates hormone secretion in the gut. Also, the findings of Morgavi et al. (2001) showed that the exogenous enzymes can survive in the intestine and exert their action on available substrates.

On the other hand, the results of Song et al., (2005) showed that the antimicrobial properties of bee pollen could be caused by weakens the growth of pathogenic bacteria and leads to optimal enzyme activity.

Economical efficiency (%):

Calculations were carried out according to the costs diet, management, and meat production of the rabbit as shown in Table 9. From these results could be noted that the best economical efficacy was obtained (1.70) in rabbits fed bee pollen at high dose followed by 1.68 and 1.67% for low and medium doses, while the lowest one (1.60) was obtained for rabbits in the control group. The relative economical efficacy amounted 105, 104 and 107% for low, medium and high groups, respectively than that of the control. The differences in the diet and bee pollen prices are undoubtedly regarded to the common selling prices in different countries. Similar results were obtained by Hedia, et. al. (2007), Attia, et al. (2014b), El-Neney and El-Kholy (2014) and Zeedan and El-Neney (2014) showed that rabbits treated with high level of BP recorded high value of EE and REE compared to control group.

CONCLUSIVELY

From the obtained findings could be concluded that the bee pollen is confirmed an interesting resource, able to improve the productive performance, BWG, and reduced daily feed intake and resulting in an improved FCR of growing rabbits. Also, immunological parameters, blood biochemical profiles, and kidney functions were improved in treated groups than in control.

Moreover, bee pollen can represent an important resource to promote the performance of rabbit. From the obtained results could be noted that the best level of bee pollen in rabbit ration was 700 mg/kg BW. However, further studies are needed with more numbers of animals and with more different doses.

Table (1): Composition and calculated analysis of the experimental diets.

Ingredients, %	Dietary treatments
Yellow corn	5.20
Soybean meal (48%)	6.50
Wheat bran	29.52
Corn gluten meal (60%)	3.50
Clover hay	38.68
Barley	10.40
Molasses	3.00
L-Lysine HCl	0.05
Limestone	1.00
Dicalcium-phosphate	1.50
NaCl	0.35
Premix*	0.30
Total	100
Calculated analysis**	
DE, Kcal/ Kg	2520
Crude protein (CP)	17.20
Ether extract (EE)	2.69
NFE	56.08
Ash	9.93
Fiber fractions:	
Crude fiber (CF)	14.10
NDF %	35.76
ADF %	21.66
Hemicellulose	14.00
Cellulose	10.88
ADL%	10.47
Ca %	1.32
Total P %	0.54
Lys %	0.53
Meth %	0.18

* Each 3 kg of Vit and Min in Premix contain: 6000000IU Vit A, 900000 IU Vit D3 40000mg Vit E,2000mg Vit K, 2000mg Vit.B1, 4000mg Vit B2, 2000mg Vit B6, 10mg Vit B12, 50000mg Niacin, 10000 mg pantothenic acid, 50mg Biotin, 3000mg Folic acid, 250000 mg choline, 50000mg Zn, 8500mg Mn, 50000mg Fe, 50000mg Cu, 200mg I , 100mg Se and 100mg Co. ** According to NRC (1977).

Table (2): The chemical composition of bee pollen % (on DM basis).

Chemical analysis	**Bee pollen**
OM	97.17
CP	29.94
EE	4.35
NFE	61.71
CF	1.17
Ash	2.83
Amino acids, % of protein	
Arginine	2.14
Aspartic	5.31
Threonine	2.98
Serine	3.48
Glutamine	4.18
Glycine	5.81
Alanine	5.13
Valine	4.92
Leucine	5.28
Tyrosine	3.71
Phenylalanine	2..13
Lysine	5.50
Minerals, % of ash	
Ca	9
P	12
Fe	5.9
Zn	0.06
Mn	2.1
Mg	6.9
Na	1.5

Table (3): Effect of different levels of bee pollen on growth performance of growing rabbits.

BP level (mg/kg BW)→ Traits ↓	Control	Levels of bee-pollen (mg/kg BW)			SEM	Sig.
		Low	Medium	High		
Initial live body weight (g)	698	697	693	695	10.01	NS
Final live body weight (g)	2175[d]	2400[c]	2640 [b]	2844 [a]	12.42	*
Total weight gain (g)	1477[d]	1703[c]	1947[b]	2149[a]	14.11	*
Feed intake as:						
DM intake, g/head	7037[a]	6980[b]	6727[c]	6600[d]	15.50	*
DM intake, g/head/day	100.53[a]	99.71[b]	96.1[c]	94.29[d]	0.98	*
DCP intake, g/head/day	10.30	10.89	10.97	11.45	0.06	NS
TDN intake, g/head/day	61.69	63.45	64.09	64.34	2.20	NS
Protein intake (g)	1210[a]	1201[ab]	1157[bc]	1135[c]	7.50	*
Digestible Energy intake (DE) (kcal)	17733[a]	17590[b]	16952[c]	16632[d]	12.66	*
Feed conversion (g intake /g gain) of						
DM/g Total weight gain	4.76[a]	4.10[b]	3.45[c]	3.07[d]	0.01	*
DCP/ g Total weight gain	0.49[a]	0.45[b]	0.39[b]	0.37[b]	0.001	*
TDN/ g Total weight gain	2.92[a]	2.61[b]	2.30[c]	2.10[c]	0.01	*
Mortality rate%	13.33	0.00	0.00	0.00	-	-

[a, b, c,d]:Means in the same row bearing different superscripts are significantly different. *: $P \leq 0.05$, NS: not significant. Low dose=200, Medium dose=500 and High dose=700 mg/kg BW.

Table (4): Effect of different levels of bee pollen on digestibility coefficient and nutritive values of growing rabbits.

BP level (mg/kg BW)→ Traits ↓	Control	Levels of bee-pollen (mg/kg BW)			SEM	Sig.
		Low	Medium	High		
Digestion coefficient (%)						
DM	71.92[c]	72.87[c]	77.65[b]	79.95[a]	0.76	*
CP	59.61[d]	63.50[c]	66.42[b]	70.63[a]	1.22	*
EE	58.97[d]	61.62[c]	64.43[b]	67.88[a]	0.75	*
NFE	73.61[c]	75.70[b]	78.89[a]	79.84[a]	0.63	*
CF	44.51[b]	46.28[b]	50.55[a]	51.11[a]	0.75	*
Nutritive values						
TDN, **%***	61.36[d]	63.63[c]	66.69[b]	68.24[a]	0.90	*
DCP, **%****	10.25[c]	10.92[b]	11.42[b]	12.15[a]	0.05	*
DE (kcal/ kg)***	2718[d]	2819[c]	2954[b]	3023[a]	3.75	*

[A, b, c] Means in the same row bearing different superscripts are significantly different.
*: $P \leq 0.05$, NS: not significant. *TDN, % = DCP % + DCF % + DEE % (2.25) + DNFE %.
**DCP, % = Digestibility coefficient of CP × CP% of the diet.
***DE (kcal / kg) = TDN × 44.3. Low dose=200, Medium dose=500 and High dose=700 mg/kg BW.

Table (5): Effect of different levels of bee pollen on some carcass traits of growing rabbits.

BP level (mg/kg BW)→ Traits ↓	Control	Levels of bee-pollen (mg/kg BW)			SEM	Sig
		Low	Medium	High		
Pre -slaughter weight, kg	2.19[d]	2.44[c]	2.60[b]	2.81 [a]	0.38	*
Carcass weight, kg	1.08[d]	1.30[c]	1.40[b]	1.53[a]	0.21	*
Dressing,%	53.02[c]	57.17[b]	57.85[ab]	58.85[a]	0.25	*
Total giblets weight (%)	3.70	3.89	4.00	4.10	0.01	NS
Heart %	0.34	0.36	0.36	0.38	0.10	NS
Liver %	2.74	2.86	2.95	3.02	0.09	NS
Kidney %	0.62	0.67	0.69	0.70	0.01	NS
Cecum %	4.65	4.72	4.80	4.85	0.04	NS
Kidney fat weight %	0.70[a]	0.57[b]	0.40[c]	0.36[c]	0.001	*

[A, b, c] Means in the same row bearing different superscripts are significantly different. *: $P \leq 0.05$, NS: not significant.
Low dose=200, Medium dose=500 and High dose=700 mg/kg BW.

Table (6): Effect of different levels of bee pollen on cecum traits of growing rabbits.

BP level (mg/kg BW)→ Traits ↓	Control	Levels of bee-pollen (mg/kg BW)			SEM	Sig.
		Low	Medium	High		
Cecum pH	7.23[a]	6.24[b]	5.85[c]	5.12[d]	0.01	*
[1]Bacteria count E.coli × 10^4 CFU/g	15.27[a]	10.54[b]	5.22[c]	4.14[c]	0.06	*
NH_3–N (mmol/l)	10.07[a]	7.32[b]	5.93[c]	4.03[d]	0.08	*
Total VFA (mmol/l)	55.32[d]	69.98[c]	77.22[b]	82.01[a]	3.66	*

[A, b, c] Means in the same row bearing different superscripts are significantly different.
Low dose=200, Medium dose=500 and High dose=700 mg/kg BW.
*: $P \leq 0.05$, NS: not significant. [1] Number of bacterial cells / gm of cacum content (total count × 10^5).

Table (7): Blood picture and some blood plasma constituents (Within normal range) of growing NZW rabbits as affected by bee pollen treatment.

BP level (mg/kg BW)→ Traits ↓	Control	Levels of bee-pollen (mg/kg BW)			SEM	Sig.
		Low	Medium	High		
Plasma blood constituents						
Total protein (mg/ 100ml)	6.23^{d}	6.40^{c}	6.89^{b}	7.42^{a}	0.03	*
Albumin (mg/100ml)	2.93^{d}	3.09^{c}	3.29^{b}	3.57^{a}	0.01	*
Globulin (mg/100ml)	3.30^{c}	3.31^{c}	3.60^{b}	3.85^{a}	0.02	*
Glucose (mg/dl)	97.5^{d}	120.5^{c}	134.3^{b}	139.7^{a}	0.75	*
Plasma lipid function						
Total lipids (mg/dl)	466.9^{a}	427.6^{b}	402.5^{c}	370.2^{d}	15.50	*
Cholesterol (mg/dl)	212.1^{a}	197.8^{b}	195.5^{b}	190.1^{c}	0.10	*
Triglycerides (mg/dl)	187.7^{a}	160.5^{b}	145.2^{c}	135.5^{d}	0.22	*
kidney function						
Urea – N (mg /dl)	45.91^{a}	41.10^{b}	37.50^{c}	35.56^{d}	0.11	*
Creatinine (mg/dL)	1.47^{a}	1.28^{b}	1.08^{c}	0.88^{d}	0.01	*
Immunoglobulin concentration						
Total antioxidants capacity (mmol/l)	1.34^{d}	1.84^{c}	2.87^{b}	3.90^{a}	0.02	*
IgG (mg/ml)	3.52^{d}	4.15^{c}	4.50^{b}	4.85 ^{a}	0.10	*
IgM (mg/ml)	1.05^{d}	1.20^{c}	1.43^{b}	1.97^{a}	0.01	*
IgA (mg/ml)	0.452^{d}	0.500^{c}	0.635^{b}	0.674^{a}	0.04	*

$^{A, b, c}$ Means in the same row bearing different superscripts are significantly different.
*: P ≤ 0.05, NS: not significant. Low dose=200, Medium dose=500 and High dose=700 mg/kg BW.

Table (8): Activity of digestive enzymes in rabbits as affected by bee pollen treatment at the end of experimental period.

BP dose (mg/kg BW)→ Traits ↓	Digestive enzymes					
	Intestinal content (U/g)			Pancreas (U/g)		
	Amylase	Lipase	Protease	Amylase	Lipase	Protease
Control	91.61^{c}	70.26^{c}	61.44^{c}	270.43^{c}	74.05^{c}	83.55^{c}
Low level (200mg/kg BW)	91.23^{c}	69.04^{c}	62.34^{c}	281.16^{b}	120.13^{b}	82.19^{c}
Medium level (500 mg/kg BW)	130.03^{b}	92.11^{b}	90.04^{b}	317.09^{a}	150.26^{a}	141.15^{b}
High level (700 mg/kg BW)	171.11^{a}	111.01^{a}	98.05^{a}	318.12^{a}	151.13^{a}	155.86^{a}
SEM	9.43	6.08	4.33	5.62	4.65	4.30
Sig.	*	**	*	**	*	*

$^{A, b, c}$ Means in the same column bearing different superscripts are significantly different. *: P ≤ 0.05.
Low dose=200, Medium dose=500 and High dose=700 mg/kg BW.

Table (9): Effect of different levels of bee pollen on the relative economical efficiency of growing rabbits.

Items	Control	Levels of bee-pollen (mg/kg BW)		
		Low	Medium	High
Total feed intake /rabbit (kg)	7037	6980	6727	6600
Price bee-pollen in total experimental	-	1.77	4.63	6.52
Price/kg diet (LE)	2.02	2.02	2.02	2.02
Total feed cost/rabbit (LE)	14.21	15.87	18.22	19.85
Average body weight gain (kg)	1.477	1.703	1.947	2.149
Price/kg body weight (LE)	25.00	25.00	25.00	25.00
Selling price (L.E)	36.93	42.58	48.67	53.72
Net revenue (LE)	22.72	26.71	30.45	33.87
Economic efficiency* (EE)	1.60	1.68	1.67	1.71
Relative economical efficiency (%)(REE)	100	105	104	107

*Net revenue/Total cost, Price of kg bee-pollen =100 LE, Low dose=200, Medium dose=500 and High dose=700 mg/kg BW

Total feed cost/rabbit (LE) = Total feed intake /rabbit (kg) x Price/kg diet (LE) + Price bee-pollen in total experimental.

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Bee pollen, rabbits, performance, carcass, digestibility, blood parameters.

الملخص العربي

تأثير اضافة مستويات مختلفة من حبوب اللقاح على الاداء الانتاجى والمناعى فى ذكور الارانب.

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صممت هذه الدراسة لتقييم تأثير إضافة حبوب اللقاح (BP) للأرانب كإضافة غذائية على معدلات الأداء الإنتاجي ، معاملات الهضم ، صفات الذبيحة، والنشاط الميكروبي caecal وبعض مكونات الدم .أستخدم في هذه الدراسة عدد ستون ارنب النيوزيلندى الأبيض فى عمر خمس أسابيع وقسمت الأرانب عشوائيا إلى 4 مجموعات متساوية بكل مجموعة 15 أرنب و كان متوسط وزن الارانب عند خمسة اسابيع(695.75g). غذيت المجموعة الأولى من دون أي اضافة وتعتبر المجموعة المقارنة والمجموعة الثانية و الثالثة والرابعة اعطيت الارانب حبوب اللقاح عن طريق الفم بجرعات تحتوي على 200 (المنخفضة) و500 (المتوسطة) و 700 (العالية) ملج /كجم من وزن الجسم الحى على التوالى يوميا خلال فترة التجربة.

أوضحت النتائج المتحصل عليها من هذه الدراسة أن الأرانب التى تغذت على عليقة مضاف اليها حبوب اللقاح زيادة كبيرة في وزن الجسم ومعدل الزيادة فى الوزن وانخفاض كمية الغذاء المأكول فضلا عن تحسين في التحويل الغذائي بالمقارنة مع مجموعة الكنترول وكان هناك تحسن كبير لمعاملات الهضم والقيم الغذائية للأرانب فى المجموعات المعاملة بالمقارنة بالكنترول. كما وزادت نسبة التصافي ووزن الذبيحة بشكل ملحوظ مع زيادة مستويات حبوب اللقاح.

كما زادت قياسات المناعة (total antioxidants capacity, IgG, IgM and IgA) بشكل ملحوظ مع زيادة حبوب اللقاح. كما زاد نشاط كل من إنزيم الاميليز والليبز والبروتيز فى البنكرياس والامعاءوكانت عالية فى المجموعات التى اعطيت حبوب اللقاح بالمقارنة بالكنترول. افضل كفاءة اقتصادية كانت مع المجموعة التى اعطيت مستوى عالى من حبوب اللقاح بالمقارنة بالمجموعات الأخرى.

التوصية: يمكن أن نخلص إلى أن اضافة مستويات من حبوب اللقاح (المنخفض والمتوسط والعالى) له تاثير إيجابي على الأداء الانتاجى وتنظيم الدهون في الدم نتيجة وجود مضادات الأكسدة.