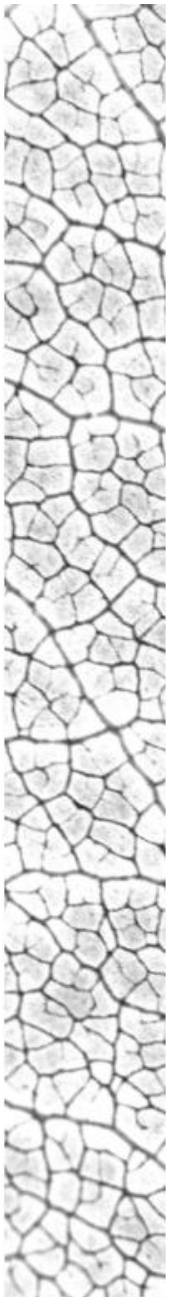

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Effect of adding different levels of crushed laurel leaves (*Laurus nobilis*) to the diet on some blood parameters for broiler chickens

Alaa K. Mohammed [1], Nihad A. Ali [1*], Chassan R. Mousa [2]

[1] Department of Animal Production, College of Agriculture, University of Al-Qasim Green, IRAQ
[2] Al-Furat Al-Awsat Technical University, Al-Mussaib Technical College, IRAQ
*Corresponding author: dr.nihad@agre.uoqasim.edu.iq

Abstract
This experiment was conducted in the field of Poultry birds belonging to Department of Animal Production, College of Agriculture, Al-Qasim Green University in order to study the effect of adding different levels of crushed laurel leaves (*Laurus nobilis*) to the diet of broiler chickens on some physiological traits for blood. A 180 unsexed broiler chickens (ross 380 strain), with one-day age were used, where they were randomly divided into four treatments, with a rate of 45 birds per treatment and each treatment consisted of three replicates (15 birds per replicate). The treatments of experiment were as follows: First treatment (control) without adding crushed laurel leaves to the diet, second treatment: Adding crushed laurel leaves with amount of (1 g / kg feed), third treatment: Adding crushed laurel leaves with amount of (2 g / kg feed) and treatment Fourth: Adding crushed laurel leaves with amount of (3 g / kg feed). The experiment included the following traits: Red blood cell count, white blood cell count, Haematocrit, hemoglobin concentration, White Blood Cell Differential Count, estimating the mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and the mean corpuscular hemoglobin concentration (MCHC). The results indicated that adding laurel leaves with the rate of (2 and 3 g / kg feed) led to significant improvement ($p < 0.05$) in the red and white blood cell counts, hemoglobin concentration, Haematocrit, the value of mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC), a significant decrease ($p < 0.05$) in the Heterophil/lymphocyte ratio (H/L ratio). From the present experiment, it is concluded that adding laurel leaves with the rate of (2, 3 g / kg feed) to the diet can lead to improve some blood parameters for broiler chickens.

Keywords: laurel leaves, blood parameters, broiler chickens

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INTRODUCTION

Broiler chickens is characterized by high body weights when marketing as a result of selection, genetic improvement and development of chicken breeding equipment (Jackie 2003) which reflected negatively on the body's immunity and resistance of birds to various diseases and stresses, where a negative correlation between body weight and immune response was observed in broiler chicks (Qureshi and Havenstein 1994). which prompted the producers of broiler chickens to intensify using of medical drugs during the breeding period to reduce the incidence of infections and reduce the percentage of mortality, which led researchers to seek methods to raise the body's immunity and reduce the chances of bacterial infections in broiler chickens, These include using medicinal plants as feed additives in the diets of broiler chickens (Al-Nadawi 2003). One of these plants is Laurus nobilis, the English name for the Bay tree, which is also called sweet bay tree or Grecian tree. The laurel tree has olive-like fruits that distinguish it with its dark brown color and be with clusters form (Al-Armoush 1999). It is classified as an evergreen herbal medicinal plant belonging to the Lauraceae family, where it is used in treating the gastrointestinal diseases and stomach ulcers (Kivcak and Mert 2002). It is found in Asia Minor and growing in North Africa. It is extracted from the fruits of laurel volatile oil (Eugenol), Its percentage about (1 to 3%) in addition to possessing many active substances such as Monoterpenes, Cinnamaldehyde, Thymol, and Carvacrol. As well as, it has the antioxidant trait because it contains the phenolic compounds and antibacterial trait (Erturk et al. 2006). The laurel leaves are used as fresh or dried aromatic

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plants (as a spice) in cooking to take advantage of its distinctive aroma and flavor (Nazia and Perween 2006). Karaalp and Genc (2013) observed when using laurel leaves with quail diet intended for egg production led to lower the concentration of malondialdehyde (MDA) in serum compared to the control treatment. Musa et al., (2011) improved some yield traits for quail when using laurel leaves added to the diet, where improved egg production and egg weight and lowering the concentrations of cholesterol and triglyceride compared to the control treatment. In view of the above and the lack of research in the use of this plant in poultry diets, this study aims to determine the effect of leaves of laurel plant added to the diet on some blood parameters for broiler chickens (Ross 308) and to know the best percentages that can be used in the diet.

MATERIALS AND METHODS

This study was conducted in the field of Poultry birds belonging to the Department of Animal Production, College of Agriculture, Al-Qasim Green University for the period from 21/3/2019 to 25/4/2019, which continued to 35 days. A 180 unsexed broiler chickens (ross 380 strain), with the average weight amounted to (40 g / chick), were used. The chicks were reared in-ground cages, and the chicks were randomly distributed on 4 treatments, each consists of 3 replicates where each replicates contains 15 chicks. Feeds were provided to the birds freely, where two diets were fed, initiator diet from (1 to 21 days) and final diet from 22 to 35 days. The crushed laurel leaves were added to the diet (manual mixing) from the age of one day as follows: First treatment (control) without adding crushed laurel leaves to the diet, second treatment: Adding crushed laurel leaves with amount of (1 g / kg feed), third treatment: Adding crushed laurel leaves with amount of (2 g / kg feed) and treatment Fourth: Adding crushed laurel leaves with amount of (3 g / kg feed). The effect of the treatments on red blood cell count, white blood cell count, Haematocrit, hemoglobin concentration, White Blood Cell Differential Count, estimating the mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and the mean corpuscular hemoglobin concentration (MCHC), were studied, where blood was collected at the fifth week of the end of the experiment from 6 birds of each treatment (2 birds for each replicate) randomly from the jugular vein where tubes containing anticoagulant (potassium EDTA) were used to prevent blood clotting. The red and white blood cell count was estimated according to the method indicated by (Natt and Herrick 1952). The Haematocrit was calculated using special capillary tubes containing an anticoagulant according to the method indicated by (Archer 1965). The hemoglobin concentration was estimated by converting it into a complex compound Cyanomethemoglobin using the Drabkins reagent

Table 1. Percentage of Feed materials included in the formation of the Initial Diet and Final diet Used in the Experiment with the Calculated Chemical Composition for Both Diets

Feed material	Initial diets (1-21 days) %	Final diets (22-35 days) %
yellow corn	30	30
Local wheat	27.7	35.5
Soybean meal (44% protein)	28	20
The concentrated Protein*	10	10
Vegetable oil	3	3
limestone	1	1.2
Food salt	0.3	0.3
Total	100%	100%
The Calculated Chemical Analysis **		
Metabolic Energy (kg / kg)	3078	3125.2
crude protein (%)	22.74	20.16
Energy/protein ratio	135.35	155.07
Lysine (%)	1.02	0.95
Methionine + Cysteine (%)	0.83	0.75
Calcium (%)	0.97	1.0
phosphorus availability (%)	0.41	0.48

* The concentrated protein from Al Hayat Company (Jordanian origin) contains 44% protein, 2800 kcal/kg metabolized energy, 12% fat, 25% ash, 5% calcium, 2.9% phosphorus, 2.55% methionine + Cysteine, 2.8% lysine.
** The chemical composition was calculated according to the analysis of feed materials mentioned by (NRC, 1994).

Table 2. Preventive Health Program for Chicks of the experiments

Age (day)	Type of vaccine
1	Giving oily vaccine of (Newcastle + bird flu) and IB vaccine dropping in the eye.
10	Newcastle vaccine type (1B) in drinking water.
14	Camboro vaccine (Lockhart) in drinking water.

according to the method indicated by (Varley et al. 1980). The heterophil/lymphocyte ratio was estimated using glass slides where a drop of blood was placed on the glass slide and spread very carefully with another glass slide placed above the blood drop and pulled over the first slide at a 45 angle without being pressed firmly and left to dry at a rate of 10 min. Thereafter, the slices are stained with a mixture of Wright Gimsa dyes according to (Shen and Patteron 1983). The counting is done using an optical microscope under a magnification power (1000) by placing an oil drop on the slide according to the method of (Burton and Guion 1968). The Completely Randomized Design was used to study the effect of different treatments on the studied traits, the significant differences between the averages were compared using Duncan's Multiple Range Test (Duncan 1955) and the SAS (SAS 2010) was used to analyze the data.

Preventive Health Program

The preventive health program was used as shown in **Table 2**, where the prevalent and approved vaccinations were conducted for broiler chicks against the two diseases of Newcastle and Camboro with no antibiotics were given during the breeding period, but the vitamins mixture was added after each vaccination process and for three days by adding it to drinking water according to the instructions of the manufacturer.

Table 3. Effect of adding different levels of crushed laurel leaves (Laurus nobilis L.) to diet on the percentage of red and white blood cell counts, the percentage of Haematocrit, hemoglobin concentration and H / L ratio for broiler chicks at the age of 5 weeks (statistical average ± standard error)

Traits \ Treatments	First treatment (control)	Second treatment (1 g/kg)	Third treatment (2 g/kg)	Fourth treatment (3 g/kg)
Red blood cell count (× 10^6.mm^{-3} blood)	0.41 ± 1.95 b	0.17 ± 2.36ab	0.10 ± 2.71a	0.08 ± 2.93a
White blood cell count (× 10^3.mm^{-3} blood)	27.6 6 ± 1801.23 b	32.33 ± 557.77 a	33.66 ± 760.11 a	33.33 ± 760.16 a
Haematocrit (%)	1.35 ± 29.16b	29.34 ± 1.30b	31.15 ± 1.29ab	33.25 ± 1.40a
Hemoglobin concentration (g/100 blood)	9.72 ± 0.26b	9.78 ± 0.12b	0.08 ± 10.38ab	11.08 ± 0.31a
H/L ratio	0.33 ± 0.03 a	0.31 ± 0.08 a	0.28 ± 0.02 b	0.29 ± 0.03 b

* Different letters within the row indicate significant differences between the treatments at the significant level (p<0.05).

RESULTS AND DISCUSSIONS

Table 3 shows the effect of the adding different levels of the crushed laurel leaves to the diet on the percentage of red and white blood cell counts, the percentage of Haematocrit, hemoglobin concentration and H / L ratio for broiler chicks at the age of 5 weeks, where it is observed excelling the third treatment (adding 2 g / kg feed of crushed bay leaves) and the fourth treatment (adding 3 g / kg feed of crushed bay leaves) significantly (P <0.05) on the first treatment (control), which they recorded the following values: (2.71, 2.93 million cells / mm^3 blood), respectively, While the first treatment (control) recorded the following value (1.95 million cells / mm^3 blood) and without significant difference from the second treatment, which recorded (2.36 million cells / mm^3 blood). This increase in the red blood cell counts may be attributed to the body's need to meet the new requirements for transporting nutrients and oxygen to the cells due to increased metabolic rate for birds that have eaten crushed laurel leaves which have been added to the diet. The high number of red blood cells in laurel leaves treatments may be caused by an effective antioxidant that helps to protect blood cells from damage that may occur as a result of oxidation (Craig 1999). It also observed that the treatments of laurel leaves (second, third, fourth) were significantly excelled (P <0.05) in the white blood cells counts compared to the first treatment (control), which recorded the following numbers: (32.33, 33.66, 33.33 × 103 / mm3 blood), respectively. While the first treatment recorded (27.66 × 103 / mm3 blood). As for the trait of Haematocrit (%) and the concentration of hemoglobin (gm /100 ml), we note that the fourth treatment was significantly (P <0.05) excelled on both the first treatment (control) and the second treatment where the highest trait for the Haematocrit and hemoglobin amounted to (33.25%, 11.08 g/100 ml), respectively. While the first and second treatments recorded values amounted to (29.16, 29.34%), respectively, for the trait of Haematocrit and (9.72, 9.78 g / 100 ml), respectively for hemoglobin, While the third treatment recorded the values amounted to (31.15%, 10.38 g / 100 ml), respectively, without significant difference with all treatments. The values of the Haematocrit took a somewhat similar trend to the changes in the red blood cells counts in the fifth week, and the Haematocrit scale is related to the red blood cells counts where the greater the red blood cells count lead to an increase in the Haematocrit scale (Sturkie 1986). The concentration of laurel leaves had a significant effect on the Haematocrit scale. From **Table 3**, the higher the concentration of laurel leaves in the diet, the values of Haematocrit scale increased, and the increase in the concentration of hemoglobin is similar to that of red blood cell counts, where the concentration of hemoglobin is directly related to the red blood cells counts (Al-Hassani 2000). Therefore, the nature of this increase in hemoglobin concentration was strongly correlated with increasing the concentration of laurel leaves in the feed. The same **Table 3** shows a significant decrease (P <0.05) in the Heterophil/lymphocyte ratio, where the lowest level for this ratio recorded in the third and fourth treatment which amounted to (0.28, 0.29), respectively, Then this ratio increased in the second treatment significantly (P> 0.05) compared to the previous treatments (third and fourth), which recorded (0.31), Then, the level of the Heterophil/lymphocyte ratio reached the highest level in the first treatment (control) where it amounted to (0.33) and with significant difference (P> 0.05) for the third and fourth treatments. The reducing H/L ratio gives a good impression about the health of the bird. Al-Darraji, (1995) mentioned that the H / L ratio is the best measure for detecting the general bird condition and the level of stress to which it is exposed and the rise of this ratio than the general average indicates that birds are under extreme stress. The reason for the decrease in the third and fourth treatment may be due to the fact that laurel leaves are considered a natural antioxidant, which plays important biological roles in improving health, reducing the risk of infecting with disease and acting as an anti-inflammatory and fungus (Ojala et al. 2000). Hence, the increase in concentration has been shown to play an important role in increasing the immunity represented by rising lymphocytes (Adeyeye et al. 2016).

Table 4. Effect of adding different levels of crushed laurel leaves (Laurus nobilis L.) to diet on the mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and the mean corpuscular hemoglobin concentration (MCHC) for broiler chicks at the age of 5 weeks (statistical average ± standard error)

Traits \ Treatments	First treatment (control)	Second treatment (1 g/kg)	Third treatment (2 g/kg)	Fourth treatment (3 g/kg)
Mean corpuscular volume (MCV) (μ^3)	149.53 ± 5.09a	4.28 ± 124.32ab	114.94 ± 4.87 b	113.48 ± 5.94 b
Mean corpuscular hemoglobin (MCH) (pg/cell)	49.84 ± 1.51 a	41.44 ± 1.69 a	38.30 ± 1.80 ab	37.81± 1.98 b
Mean corpuscular hemoglobin concentration (MCHC) (%)	33.333 ± 0.004 a	33.333 ± 0.003 a	33.322±0. 003 a	33.323 ± 0.003 a

* Different letters within the row indicate significant differences between the treatments at the significant level (p<0.05).

Table 4 shows the effect of adding different levels of crushed laurel leaves (Laurus nobilis L.) to diet on mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and the mean corpuscular hemoglobin concentration (MCHC) for broiler chicks at the age of 5 weeks. Where the third and fourth treatment recorded the lowest value for the mean corpuscular volume MCV (μ^3) and with a significant difference (P <0.05) compared to the first treatment which amounted to (114.94, 113.48 μ^3) followed by the second treatment which recorded the value of MCV amounted to (124.32 μ^3). The first treatment recorded the highest value of MCV amounted to (149.53 μ^3) without significant differences from the second treatment. As for the mean corpuscular hemoglobin (MCH) (pg/ cell), where the first and second treatment recorded the highest value of MCH and without significant difference from the third treatment, While the fourth treatment recorded the lowest value of MCH (pg/cell) and a significant difference (P <0.05) than the first and second treatments, As for the mean corpuscular hemoglobin concentration (MCHC), it is noticed that there were no significant differences between the treatments (Rezapour-Nasrabad 2018).

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