



The effect of adding local myrtle leaves powder and industrial antioxidant Butylated Hydroxy Toluene to laying hens feeding on some biochemical characteristics and special enzymes against oxidative stress in laying hens blood

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Abstract

This experiment conducted in the poultry field "Al Sayada" affiliated to the Department of Animal Production at the College of Agriculture in the University of Kirkuk for the period 20/2/2022 to 12/6/2022 for 112 days (16 weeks), 200 laying hens, type "LOHMANN BROWN", at the age of 54 weeks, were randomly distributed to 10 nutritional treatments, with 5 replications for each treatment, and one repetition included 4 birds. The treatments were represented as follows: first treatment: control (without any adds), second treatment: first treatment + 1.0% myrtle leaves powder, third treatment: first treatment + 2.0% myrtle leaves powder, fourth treatment: first treatment + 0.05% Butylated hydroxytoluene, fifth treatment: The second transaction +0.05% Butylated hydroxytoluene, the sixth transaction: the third transaction +0.05% Butylated hydroxytoluene. It is clear from the statistical analysis of the study that there is a significant decrease ($P \leq 0.05$) in the levels of cholesterol, total protein and globulin for the addition treatments compared with the control treatment (without any addition) and an improvement in the levels of glucose, high-density lipoprotein, low-density lipoprotein and albumin, as well as an improvement in the antioxidants Malondialdehyde and Glutathione In blood serum, it can be concluded that the addition of local myrtle leaves powder at rates of 1% and 2% with (BHT) at 0.05% enhanced the improvement of biochemical properties and antioxidant enzymes in serum of laying hens.

Key words: myrtle leaves, BHT, laying hens, biochemical characteristics, oxidative enzymes.

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Introduction

Interest has increased recently in medicinal plants and other sources found in nature, due to the effective compounds they contain in their various parts,[1]and why these compounds play an important role in improving digestion, appetite[2] and immunological characteristics, as well as the blood characteristics of the organism.[3] It does not leave side effects for the consumer. [4]Scientific studies and research have occurred on its use in appropriate ways, such as using the whole plant or parts of its extracts from the active compounds present in its parts. One of the medicinal plants is the myrtle plant, whose scientific name is *M.Communitis*. It is an annual aromatic plant that belongs to the exponential family and contains more than 5,500 species. [5]The leaves, flowers and bark produce an oil known as Angels water, which is characterized by a refreshing aromatic scent. It is considered important in the perfume industry.[6]The plant contains analgesic, antiseptic and anti-inflammatory properties [7]and antioxidant substances, and recent studies have shown that the use of leaf powder showed an antifungal effect against some bacterial microorganisms and pathogenic fungi [8]As well as raising the concentration levels, of natural antioxidants in the tissues Farawn and stimulating the functions of the digestive system by increasing the secretion of digestive enzymes and enhancing the performance of the pancreas and liver as well as the small intestine and thus increasing the utilization of the food intake and filling the body's need for nutritional components and thus improving growth and the food conversion factor and improving system's immunological and hematological traits [9]Therefore, the study aimed to find out the use of different levels of local myrtle leaf powder and compare with an industrial antioxidant (BHT) in the biochemistry and antioxidant enzymes in the blood serum of brown Luhmann laying hens.

Materials and methods

This experiment was carried out in the field of poultry/Al-Sayada, affiliated to the

Department of Animal Production in the College of Agriculture at the University of Kirkuk for the period from 20/2/2022 to 12/6/2022 for a 112 days (16 weeks), randomly distributed 200 laying hens type BROWN LOHMANN, age 54 week to 6 nutritional treatments with 5 repetitions/treatment (one repetition included 4 birds). The aim was to study the use of local myrtle leaf powder and compare it with a type of industrial antioxidant Butylated Hydroxy Toluene (BHT) in laying hens diets and their effect on some biochemical characteristics of chicken blood. whiteness. The nutritional treatments included the following:

T1: control.

T2: T1 + 1.0% myrtle leaves powder.

T3: T1 + 2.0% powder of myrtle leaves.

T4: T1+0.05% BHT.

T5: T1 +0.05% BHT.

T6: T1 +0.05% BHT.

Chemical analysis of the active compounds in the local myrtle leaf powder:

The chemical analysis of the active compounds in the powder of the leaves of the local myrtle plant was carried out in the laboratory of the College of Agriculture - Salahaddin University/Erbil, and the results of the analysis are as shown in the table (1).

Table (1) Chemical analysis of the active compounds in the local myrtle leaf powder

compound	%
alpha.-Pinene	18.47
Eucalyptol	14.10
1,6-Octadien-3-ol, 3,7-	2.35
dimethyl-, 2- aminobenzoate	
Silane,	3.95
dimethyloctyloxytridecyloxy	
1-(+)-Ascorbic acid 2,6-	3.25
dihexadecanoate	
2,2-Dimethyl-4-octenal	4.01
5.alpha.,6.alpha.-Epoxy-	5.44
17-oxo-6.beta.- pentyl-4-	
nor-3,5secoandrostan-3-	
oicacid	
2-Phenanthreneacetic acid,	3.15
tetradecahydro-7-hydroxy-	
1,4b,8,8-tetramethyl-10-	

oxo-	
6-Octadecenoic acid, (Z)-	1.25
1,2,3-Benzenetriol	10.25
1,6-Octadien-3-ol, 3,7-dimethyl-	3.28
4-Chlorobenzoic acid, pentadecyl ester	5.01
Germacr-4-en-12-oic acid, 6.alpha.-hydroxy-,gamma.-lactone,(11S)-	4.12
4a,6a-Dimethyl-2-oxo-Durohydroquinone	3.66
3.beta.-Myristoylolean-12-en-28-ol	1.55
Oxalic acid, hexadecyl 1-menthyl ester	8.64
3-Cyclohexene-1-methanol, .alpha.,.alpha.,4-trimethyl-, (S)-	5.87
	1.66

Table (2) The proportions of feed materials with the calculated chemical composition of the diets of the experimental treatments.

Ingredients %	T1	T2	T3	T4	T5	T6
Wheat	65.8	64.85	59.37	65.84	64.80	59.32
Barley	2.65	1.83	5.01	2.65	1.83	5.01
myrtle leaf powder	-	1.0	2.0	-	1.0	2.0
BHT	-	-	-	0.05	0.05	0.05
Soybean %47 meal	16.0	16.41	17.19	16.00	16.41	17.19
oil (sunflower seed)	3.30	3.75	4.27	3.30	3.75	4.27
Limestone	9.26	9.26	9.26	9.26	9.26	9.26
Dicalcium phosphate	2.0	2.0	2.0	2.0	2.0	2.0
Salt	0.20	0.20	0.20	0.20	0.20	0.20
Vitamin-mineral (a)	0.10	0.10	0.10	0.10	0.10	0.10
L-Lysine	0.15	0.15	0.15	0.15	0.15	0.15
D-methionine	0.20	0.20	0.20	0.20	0.20	0.20
choline chloride 60%	0.25	0.25	0.25	0.25	0.25	0.25
Total	100	100	100	100	100	100

Chemical composition (b)						
Metabolizable energy (MJ/kg)	2792	2789	284	2792	2825	2841
Crude protein %	16.5	16.50	17.7	16.56	17.12	17.73
Calcium %	3		2			
Phosphorus %	4.00	4.00	4.00	4.00	4.00	4.00
Methionine %	0.36	0.36	0.36	0.36	0.36	0.36
Lysine %	0.44	0.441	0.46	0.442	0.441	0.468
	2		7			
	0.83	0.838	0.87	0.835	0.838	0.874
	4		2			

(a) 1 kg of the mixture of vitamins and minerals prepares: Vitamin A (8,000,000 IU), Vitamin D3 (1,500,000 IU), Vitamin E (1,000 IU), K3 (2,000 mg), B1 (500 mg), B2 (500 mg), B6 (200 mg), B12 (8 mg), Folic Acid (50 mg), Niacin (8000 mg), Calcium (4000 mg), Magnesium (400 mg), Zinc (150 mg), Iron (53 mg) , copper (43 mg)and choline (40 mg)

(b) According to the chemical composition of the feed materials according to what was stated in US National Research Council [10].

Measurement of glucose concentrations, cholesterol, albumin, globulin, low-density cholesterol and high-density cholesterol

These tests were conducted using a test kit (Kit) produced by a Spanish company (Klabkit). The readings of the samples were measured using a spectrophotometer at different wavelengths according to each measurement and according to the method of work attached by the company.

Estimation of antioxidant status in blood serum:

Determination of malondialdehyde (MDA) concentration in blood serum:

The thiobarbituric acid (TBA) reaction method used by the researchers [11]was used to measure the concentration of mannodialdehyde, which is one of the main products of the lipid peroxidation process, and its level is an indicator of it as it depends on the principle of interaction between lipid peroxidation and TBA.

Estimation of glutathione (GSH) concentration in blood serum:

The concentration of glutathione in blood serum was measured using the modified Ellman reagent method. The reagent was prepared in the laboratory by dissolving 0.004 g of 5-5-dithio bis- 2nitro benzoic acid in 100 ml of phosphate buffer solution [11].

Statistical analysis:

A complete random design (CRD) was used to study the effect of the studied coefficients on the different traits, and the ready-made statistical program [12] was used in the statistical analysis, and the averages were compared using Duncan's multinomial test [12] according to the following mathematical model: $Y_{ij} = M + T_i + E_{ij}$

Results and discussion

It is clear from the results of the statistical analysis in Table (3) that there are no significant differences ($P \leq 0.05$) between the transactional birds for the level of glucose in the blood serum of the transactional birds, but it has decreased by an amount (5.36, 8.43, 5.76, 19.54 and 11.11%), respectively, compared with control treatment birds, as the local myrtle plant has a significant effect on lowering the level of sugar in the blood, Myrtle acts as an anti-diabetic by inhibiting the A-glucosidase enzyme present in the small intestine, which reduces the absorption of glucose, as well as increasing the rate of glycolysis by increasing the activity of the enzyme (glucokinase). Also, because of the presence of Myricetin in the composition of the myrtle plant, and this substance is one of the compounds included in the class of flavonoid antioxidants, on the other hand, the reason is that it has a hypoglycemic effect by increasing the activity of waxy cells, and these cells work to absorb glucose in the blood. On the other hand, insulin present in the blood stimulates further reduction of glucose levels in the blood (12,13,7) While there were

significant ($P \leq 0.05$) differences between the blood plasma of the birds of the treatment for the level of cholesterol, compared with the blood plasma of the birds of the first treatment (free of any addition), and it decreased significantly ($P \leq 0.05$) the level of cholesterol in the blood birds for the second, third, fourth, fifth and sixth by (5.30, 5.09, 8.41, 6.85 and 5.76%), respectively. Compared with the first treatment, the reason for this is due to the fact that the local myrtle plant contains effective compounds such as saponins, which work to reduce cholesterol in the blood serum by controlling the production and secretion of enzymes responsible for fat and cholesterol metabolism [2] As for the level of high-density protein lipids (HDL), the second treatment containing 1% of the local myrtle leaf powder recorded the highest rate for this characteristic, with an amount of 4.38%, and in a significant way ($P \leq 0.05$) The rate of blood plasma of the first treatment control birds While the rest of the experimental treatments did not record any significant differences with the first treatment, while low-density lipids (LDL) did not record any significant differences between the experimental treatments. As for protein, it is one of the most important main components in the blood serum of animals in general, as well as fats, carbohydrates, vitamins, minerals and enzymes. Protein has a very important and vital role in bringing about the natural balance of the body. It is a storehouse of amino acids and a carrier of food compounds that cannot be transferred without being linked, such as fats that are transmitted in the form of lipoproteins, as well as some carbohydrates that are transmitted in the form of glycoproteins. Therefore, the rate of total protein, albumin, and globulin in the blood plasma of the comparison treatment birds (the first) in Table (3) was significantly higher ($P \leq 0.05$) than the rest of the blood plasma of other treatment birds, and it amounted to (4.33, 4.06, 4.09, 3.93

and 3.85) for total protein. and 4.05 mg/100), respectively, and for the level of albumin concentration it was (2.05, 2.05, 2.02, 1.91, 1.96 and 2.02 mg/100 ml) and for the level of globulin it was (2.61, 2.01, 2.24, 2.01, 1.89 and 2.03 mg/100) where The liver in poultry birds is the main factory for the manufacture of proteins of all kinds

Table (3): The effect of using powder from local myrtle leaves and a synthetic antioxidant (BHT) on glucose, cholesterol, high-density and low-density lipids, total protein, albumin and globulin in the blood serum of Luhmann laying hens (mean ± standard error).

Treatments	Glucose mg/100ml	Cholesterol mg/100ml	HDL mg/100ml	LDL mg/100ml	T-protein mg/100ml	Albumin mg/100ml	Globulin mg/100ml
T1	0.13±5.22	0.57±151.0 a	0.33±44.96 b	0.34±15.03	0.08±4.33 a	0.02±2.05	0.30±2.61 a
T2	0.53±4.94	1.52±143.0 b	0.09±47.18 a	0.28±15.26	0.06±4.06 b	0.05±2.05	0.01±2.01 b
T3	0.48±4.78	1.45±143.3 b	0.64±45.56 ab	0.33±14.60	0.02±4.09 b	0.19±2.02	0.017±2.24 ab
T4	0.90±4.90	4.17±138.3 b	0.70±45.95 ab	0.31±14.70	0.04±3.93 bc	0.08±1.91	0.06±2.01 b
T5	0.52±4.20	1.45±140.66 b	0.43±46.53 ab	0.43±14.96	0.08±3.85 c	0.06±1.96	0.05±1.89 b
T6	0.29±4.67	1.85±142.3 b	0.52±46.32 ab	0.48±15.04	0.01±4.05 b	0.05±2.02	0.02±2.03 b
incorporeal	NS	*	*	NS	*	NS	*

NS - no significant differences within the column at the probability level (P≤0.05).

* - There are significant differences within the column at the probability level (P≤0.05).

T1: control. T2: T1 + 1.0% myrtle leaves powder. T3: T1 + 2.0% powder of myrtle leaves. T4: T1+0.05% BHT. T5: T1 +0.05% BHT. T6: T1 +0.05% BHT.

As for the results of the statistical analysis of the characteristics of antioxidants in the blood, there were no significant differences (P≤0.05) between the birds of the addition treatments when compared with the control treatment of the concentration of malondehyde (MDA) in the blood. It is evident from these

results that the local myrtle leaf powder and BHT improved the antioxidant status by decreasing the level of MDA and increasing the level of GSH in the blood serum of the addition treatments in the experiment. The ability of the active substances present in the powder of local myrtle leaves and BHT to raise the level of GSH and reduce the level of MDA. This decrease is attributed to the extent of the effect of antioxidants on the secretion of corticosterone hormone from the adrenal cortex, which works to raise the concentration of FSH and LH. This was shown by a decrease

in the level of MDA and a rise in GSH [14][15].indicated a positive correlation between the concentration of LH and GSH, and a negative correlation with MDA that may have the greatest effect on raising the concentration of LH in serum the blood [13].

Table (4): The effect of using powder from local myrtle leaves and a synthetic antioxidant (BHT) on the level of antioxidants in the blood serum of Luhmann laying hens (mean ± standard error)

Treatments	MDA	GSH
	micromol/mol	micromol/mol
T1	0.29±2.19	1.630.02 c
T2	0.04±2.10	0.03±1.77 b
T3	0.07±2.11	0.005±1.81 b
T4	0.03±2.05	0.06±1.85 b
T5	0.06±1.93	0.07±1.92 a
T6	0.06±2.06	0.07±1.87 a
Incorporeal	NS	*

NS - no significant differences within the column at the probability level ($P \leq 0.05$).

* - There are significant differences within the column at the probability level ($P \leq 0.05$).

T1: control. T2: T1 + 1.0% myrtle leaves powder. T3: T1 + 2.0% powder of myrtle leaves. T4: T1+0.05% BHT. T5: T1 +0.05% BHT. T6: T1 +0.05% BHT.

Conclusion

1. Addition of 1% and 2% of local myrtle leaf powder and 0.05% of BHT reduced glucose level, cholesterol level, total protein level and raised HDL level in the blood serum of birds.
2. The addition of 1% and 2% of local myrtle leaves powder and 0.05% of BHT improved the antioxidant status in the blood serum of birds by decreasing the level of MDA and raising the level of GSH.

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تأثير اضافة مسحوق اوراق نبات الآس المحلي ومضاد الاكسدة الصناعي Butylated Hydroxy Toluene في تغذية الدجاج البياض على بعض الصفات كيميوية والانزيمات الخاصة ضد الاكسدة لدم الدجاج البياض

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• البحث مسئل من اطروحة الدكتوراه للباحث الاول.

المستخلص

تمت هذه التجربة في حقل الطيور الداجنة / الصيادة والتابع لقسم الانتاج الحيواني في كلية الزراعة داخل جامعة كركوك للفترة من 2022/2/20 ولغاية 2022/6/12 ولمدة 112 يوماً (16 اسبوع)، وزعت عشوائيا 200 دجاجة بياضة نوع BROWN LOHMANN بعمر 54 اسبوعاً الى 6 معاملات تغذوية وواقع 5 مكررات/معاملة ، مثلت المعاملات على النحو الآتي: المعاملة الأولى: السيطرة ، المعاملة الثانية: المعاملة الاولى + 1.0 % مسحوق اوراق نبات الآس ، المعاملة الثالثة: المعاملة الاولى + 2.0 % مسحوق اوراق نبات الآس ، المعاملة الرابعة: المعاملة الاولى +0.05% BHT ، المعاملة الخامسة: المعاملة الثانية +0.05% BHT ، المعاملة السادسة: المعاملة الثالثة +0.05% BHT . ويتضح من التحليل الاحصائي للدراسة عن وجود انخفاض معنوي ($P \leq 0.05$) في مستويات الكولسترول والبروتين الكلي والكلوبيولين لمعاملات الاضافة مقارنة مع معاملة السيطرة (الخالي من أي اضافة) وتحسن في مستويات الكلوكرز و HDL و LDL والالبومين وكذلك تحسن في مضادات للاكسدة ال MDA و GSH في مصل الدم ، يمكن الاستنتاج ان اضافة مسحوق اوراق نبات الآس المحلي بنسب 1% و 2% مع (BHT) بنسبة 0.05% قد عززت من تحسن صفات كيميوية والانزيمات المضادة للأكسدة في مصل الدم لدجاج البياض .

الكلمات المفتاحية: أوراق الآس ، BHT ، الدجاج البياض ، الصفات كيميوية ، الإنزيمات الاكسدة.