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Effect Of Using Water Extract Of Nettle Leaf (Urtica Dioica) On Some Microbial Characteristics Of Broiler Chickens

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Abstract

This study was conducted in the poultry field belonging to the Department of Animal Production at the College of Agriculture at Al-Muthanna University for the period from 10/25/2017 to 12/5/2017 to know the effect of using a82queous extract of nettle leaves on some microbial characteristics of broilers. In this experiment, 240 chicks were used. From broiler strain (Ross308), one day old, with an average weight of (43) gm, it was randomly distributed into four parameters, at 60 chicks per treatment, with three replications for each treatment, 20 chicks/repetition, and was raised in four-story batteries, each floor contains a cage With an area of 1.5 * 1 square meter. The experimental treatments included: the first treatment, T1, the control treatment, without any addition. As for the other treatments (T2, T3, and 4 T), the aqueous extract of nettle leaves was added to it in three concentrations (10, 15, 20) ml/liter of drinking water, respectively. The results of the study showed that there was a significant decrease (P≤0.05) in the logarithmic numbers of both total aerobic and coliform bacteria, with a significant increase (P≤0.05) in the logarithmic numbers of lactobacilli in the duodenum, jejunum, and ileum in favor of the aqueous extract of discoid leaves compared to the control treatment.

keywords: Nettle, Urtica Dioica, , water extract ,Bacterial Count, Chicken Intestine, Broilers.

I. Introduction

Nettle is a herbaceous plant that spreads in different places Of Iraq and some Arab and foreign countries. Nettle is one of the genera of the Urticaceae family, which includes a group of plants with fine thorns, and the genus Nettle consists of several species, there are three types of them in Iraq: Urtica dioica And Urtica Urens, and Urtica Pilulifera, which differ from each other in leaf size and plant height (AL-Rawi and Chakravarty, 1988) The nettle plant is one of the medicinal plants distinguished by its therapeutic efficacy, so it is among the groups in small areas of Africa (Kavalali, 2004, Bhat & Moskovitz, 2009; Roberts; 2012). Some studies point to the importance of using nettle as additives in animal diets, especially poultry (Hughes et al., 1980 (The use of plant extracts for domestic birds can

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grow plant growth and reduce research workers from nutrients), 2002) where Safanah et al. (2012) noted the extract The aqueous of the pineapple leaves, is beautiful in the synthesis of E. coli bacteria by its action of the compounds it contains, flavonoids, phenols, and it can also be used as a food preservative, as it contains the properties of the bacterial synthesis. Farhan (2012) confirmed this that the aqueous extract with a concentration of 0.5 g / l had an effective effect in inhibiting the growth of E. coli when studying the effect of aqueous extracts on different types of bacteria. Abouhosseini et al. (2016) also noted an improvement in increasing the numbers of E. coli. Lactobacillus, with reduced numbers of Escherichia Coli bacteria, when studied, in which nettle root extract was used with 5.0 g / kg of nettle root extract to evaluate its effects on the microbial traits of Ros 308 males for 42 days on 120 birds. In addition to the previous studies and the lack of local studies on the aqueous extract of nettles leaves and its effect on the microbial characteristics of broilers, as well as to educate breeders about the importance of using medicinal plant extract as a safe alternative to the use of chemical drugs that hurt the health of the consumer, the current study came intending to know the effect of adding the extract. Watery nettle leaf to drinking water in some microbial traits of broiler strain (Ross 308).

II. Materials And Methods

The experimental treatments included:

Experimental treatments included: First treatment (T1) Control treatment without any addition of plant extract, the other treatments (T2, T3, and T4) were added to the water extract of nettle leaves with three concentrations (10, 15, 20) ml/l in the drinking water, respectively. Birds were fed on two types of diets, a starter for 1-21 days, containing 23.04% protein and 2951kcal per kg, and a finisher for 22-42 days, containing 20.55% protein and 3098kcal per kg of energy. The pelleted feed was produced by Al-Hafiz company, Karbala Private, Iraq.

Table 1. Ingredients and chemical composition of the starter and finisher diets

Ingredients %	Starter (1-21) days	Finisher (22-42) days
Corn	38	39.3
Wheat	21	26
Soybean meal 44%	38.4	31
Mixture Vitamins And metal	1	1
Plant oil	0.5	1.8
Limestone	0.8	0.6
Day Calcium Phosphite	0.3	0.3

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Total	100	100
Installation Chemist Calculated		
Crude protein %	23.04	20.55
ME kcal/kg	2951	3098
Calcium %	0.93	0.85
Phosphorous Available %	0.48	0.45
Methionine %	0.55	0.5
Lysine%	1.35	1.25
Methionine + Cysteine %	0.91	0.85
Folic acid %	1.2	1.1

Calculated analysis according to NRC (1994).

III. Preparation of aqueous extract and method of use

After obtaining the dried nettle leaves from the local market, they were thoroughly cleaned of impurities, and then milled by an electric grinder into a fine powder, then prepared according to the modified method of Hernandez et al. (1994); By mixing an amount of dry powder with a quantity of distilled water at a ratio of 1 g: 20 ml of distilled water, and mixing both the solvent (distilled water) and the solute (nettle leaf powder) using an electric mixer. To obtain a homogeneous solution, then transfer this solution to the water bath at a temperature of 55 ° C for one hour, To ensure complete solubility and homogeneity, and at the end of the extraction process, the solution was left for 24 hours at room temperature, after which it was filtered with gauze. To capture the precipitate particles and take the leachate that was produced from the extraction process to be ready for use in the experiment.

Table 2. Results of the qualitative and quantitative chemical detection of the nettle leaf infusion.

Active compounds	Ml / µg
flavonoids	0.2993
phenols	3.826
Saponins	0.816
Tannins	0.773
Steroids	0.5415
Terpenes	0.7891

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Materials	Weight (g)

The analysis was done in the modern science laboratories in Diwaniyah / Hay Al-Jazayer.

IV. Preparation of peptone and culture media

Prepare the peptone water solution

Prepare by dissolving 1 g of peptone in 1000 ml of distilled water, then distribute to decimal dilution tubes and sterilize the Autoclave at 121 °C and pressure 1.5 atmospheres for 15 minutes (Harrigan and McCance, 1976).

V. Agricultural media

The following culture media were used that were sterilized in the Autoclave at a temperature of o121 m and a pressure of 1.5 atmospheres for 15 minutes.

VI. Preparation of Nutrient Agar

Prepare according to the instructions of the Indian prepared company (Himedia), by dissolving 28 g of the nutrients in 1000 ml of distilled water, then heat the mixture to a boil for one minute and use it to estimate the total number of bacteria.

VII. Preparation of MacConKey Agar

Prepare according to the instructions of Oxiod, an English prepared company, by dissolving 51 grams of MacConKey solid in 1000 ml of distilled water, then heat the mixture to a boil for one minute and use it to estimate the number of coliform bacteria.

VIII. Preparation of MRS Agar

It was used for the total count of Lactobacillus acidophilus cells prepared as reported by Harrigan and McCance (1976) by dissolving the following components in one liter of distilled water. Shown in Table (3).

Table 3. the most important components of MRS Agar.

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IX. Microbial examinations

Peptone	10
Meat Extract	10
Sodium acetate	5
Yeast Extract	5
Triammonium Citrate	2
K3HPO4	2
Tween 80	0.2
MgSo4	0.2
MnSo4.4H2o	0.05
Glucose	20
Agar	15

The microbial examinations were conducted in the postgraduate laboratory at the College of Agriculture at Al-Muthanna University, as 1g of the contents of the small intestine (duodenum, jejunum, and ileum) was taken from (3 birds/treatment), and 9 ml of the previously prepared peptone water solution was added to be the first dilution of 10-1, and save the solution In the refrigerator at a temperature of 4 ° C until the microbial count is performed.

X. Estimate the total number of bacteria

Four glass tubes containing 9 ml of Pepton water solution were prepared, and 1 ml of the initial dilution solution 10-1 was taken to the first tube to be the dilution 10-2 and 1 ml of it was taken to the second tube, and so on to the fourth tube so that the ratio of dilution was 10-5, then transferred 1 ml of Each decimal dilution by sterile pipette into two empty sterile Petri dishes (Duplicate), and directly to each dish, 15 ml of sterile Nutrient Agar are added to estimate the total number of aerobic bacteria according to the Pour Plate Count method mentioned in (1978) APHA.

XI. Estimate the total number of coliform bacteria

Three glass tubes containing 9 ml of Pepton water solution were prepared, and 1 ml of the initial dilution solution 10-1 was taken to the first tube to be the 10-2 dilution, and 1 ml of it was taken to the second tube, and so on to the third tube so that the dilution ratio was 10-4, and it was also used. The Pour Plate Count method mentioned in (1978) APHA to estimate the total number of total coliform bacteria, by transferring 1 ml of each decimal diluent using a sterile pipette into two empty sterile Petri dishes (Duplicate), and directly add 15 ml of sterile culture medium to each plate. The pre-prepared MacConKey Agar feeder, then the colony rate is taken and then multiplied by the dilution reciprocal to obtain the number of germs colonies / g of the gut sample (colony /g).

XII. Estimate the total number of Lactobacilli bacteria

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I prepared six glass tubes containing 9 ml of Pepton's water solution, and 1 ml of the initial dilution solution 10-1 was taken to the first tube to make the dilution 10-2, and 1 ml of it was taken to the second tube and so on to the sixth tube so that the dilution ratio was 10-7, and I used the Pour method The Plate Count reported by Speak (1984) using solid MRS to estimate the total number of L. acidophilus bacteria.

XIII. statistical analysis

A complete random design (CRD) was used to study the effect of different parameters on microbial traits. The significant differences between the averages were compared with the Duncan (1955) polynomial test under the 0.05 significance level, and the program SPSS (2018) was used in the statistical analysis.

XIV. Results and discussion

It is evident from the tables below (4,5,6) that the aqueous extract treatments contributed to obtaining good results through increasing the numbers of Lactobacilli bacteria and reducing the numbers of total aerobic and coliform bacteria in the parts of the digestive system (duodenum, jejunum, and ileum) according to the level of addition compared With a control treatment.

The reason may be attributed to the efficacy of the aqueous extract of nettle leaves due to the potent compounds it contains represented by tannins and flavonoids, which inhibit the action of harmful bacteria in the gastrointestinal tract (Vijayanand and Hemapriya, 2011), so that the active compounds have their role in promoting and supporting the microbial balance within the gut and ultimately reflects on the general health of birds (Vasudha et al 2011). This result is in agreement with the findings of Safanah et al. (2012) that the aqueous and alcoholic extracts of nettle leaves have complete effectiveness in inhibiting bacteria, due to the effective compounds they contain (glycosides, tannins, flavonoids, phenols), and it is consistent with what was reported by Abouhosseini and others. (2016) on the efficacy of nettle root in increasing the numbers of Lactobacillus bacteria, and reducing the numbers of coliform bacteria in the gut of broilers compared with the control treatment. Cram-positive bacteria such as Coli are sensitive to plant extracts by interfering with the cell's cell membrane and changing its permeability by exchanging positive ions such as k + and H +, which contribute to cell breakdown (Ultee et al. 2002) and phenolic compounds have an active role in this (Lambert et al., 2001). (Unlike the Cram-negative bacteria, which are equipped with an outer layer surrounding the cell membrane and act as a permeability barrier that prevents the hydrophobic compounds from reaching hydrophobic compounds, and since most plant extracts compounds are hydrophobic, the Gram-negative bacteria such as Lactobacillus exhibit greater resistance than the positive for the Cram stain) (Orndorff et al. It can be said that medicinal plants have a positive effect

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in multiple ways, such as stimulating the digestive process, as well as possessing anti-microbiological properties.

T1: a control. T2: adding an aqueous extract of nettle leaves at a level of 10 ml/liter of water. T3: adding the aqueous extract of nettle leaves at a level of 15 ml/liter of water. T4: add aqueous extract of nettle leaves at a level of 20 ml/liter of water. * The different letters within one column indicate that there are significant differences between the groups at a probability level of 0.05.

Table 6. Effect of nettle leaf water extract on the Ileum of broilers (mean ± standard error)

Treatments	Total aerobic bacteria	Coliform bacteria	Lactobacilli
T1	8.78±0.005 a	5.13±0.005 a	3.86±0.005 d
T2	7.12±0.005 b	4.23±0.008 b	5.12±0.008 c

Table 4. Effect of nettle leaf water extract on the Duodenum of broilers (mean ± standard error)

Treatments	Total aerobic bacteria	Coliform bacteria	Lactobacilli
T1	10.76±0.005 a	6.42±0.005 a	2.13±0.005 d
T2	9.84±0.005 b	5.91±0.003 b	3.06±0.014 c
T3	9.06±0.011 c	5.66±0.005 c	3.43±0.005 b
T4	8.73±0.005 d	5.22±0.005 d	3.88±0.005 a
significance	*	*	*

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T3	6.72±0.008 bc	3.91±0.003 c	5.41±0.005 b
T4	5.35±1.000 c	3.71±0.012 d	5.68±0.005 a
significance	*	*	*

Table 5. Effect of nettle leaf water extract on the Jejunum of broilers (mean ± standard error)

Treatments	Total aerobic bacteria	Coliform bacteria	Lactobacilli
T1	9.74±0.005 a	5.86±0.005 a	3.44±0.006 d
T2	8.53±0.005 b	4.62±0.005 b	4.68±0.008 c
T3	8.28±0.008 c	4.36±0.005 c	4.94±0.008 b
T4	8.02±0.008 d	3.95±0.015 d	5.34±0.011 a
significance	*	*	*

T1: a control. T2: adding an aqueous extract of nettle leaves at a level of 10 ml/liter of water. T3: adding the aqueous extract of nettle leaves at a level of 15 ml/liter of water. T4: add aqueous extract of nettle leaves at a level of 20 ml/liter of water. * The different letters within one column indicate that there are significant differences between the groups at a probability level of 0.05.

T1: a control. T2: adding an aqueous extract of nettle leaves at a level of 10 ml/liter of water. T3: adding the aqueous extract of nettle leaves at a level of 15 ml/liter of water. T4: add aqueous extract of nettle leaves at a level of 20 ml/liter of water. * The different letters within one column indicate that there are significant differences between the groups at a probability level of 0.05.

XV. Conclusion

The addition of nettle leaf (Urtica Dioica) aqueous extract to drinking water contributed to the improvement of the microbial characteristics of broilers by increasing the beneficial bacteria in the gastrointestinal tract and inhibiting harmful bacteria in comparison with the control treatment.

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