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Biological Mechanisms of the Aqueous Extract of (*Alcea kurdica*) on the Productive Performance and Microbial Content of the Intestines of Broiler Chickens

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ABSTRACT

The main focus of this study to investigate the biological mechanisms by which a (*Alcea kurdica*) extract impacts chicken productivity and gut microbiome. A total of 225 chicks were randomly allotted to five treatments provided the first as a control (C-1), Levosav as antibiotics (Le-2), with Aqueous Extract **(**AE) of *Alcea kurdica* (AK) at doses 5% (AE-1), 10% (AE-2), and 15% (AE-3) on drinking water, each treatment contained 3 replicates (15 birds each replicates). The results showed that there were significant differences between the experimental treatments. *(p*>0.05*)* changes in performance relative weight between different treatments. In contrast, there was a significant increase in the rate of body weight, body weight gain was observed across all treatments receiving the aqueous extract flowers compared to (C-1), and (Le-2) treatments. For the feed intake and feed conversion the treatments (C-1), and (Le-2) showed significantly higher feed intake compared with the other treatments 5% (AE-1), 10% (AE-2), and 15% (AE-3). AE treatments had significantly feed conversion than the control *(p<0.05)*. Microbial examination data of the broiler intestine showed the positive effect of marshmallow flower extract, as the total bacterial counts and coliform bacteria in the ileum were significantly decreased compared to the control and antibiotic treatment, while the lactic acid bacteria counts were significantly increased in the fifth treatment (15%) compared to the control and the rest of the experimental treatments. As for the microbial content of the jejunum, the total bacterial count decreased significantly in all treatments of the extract and the colonic bacteria decreased significantly in the fifth treatment, while the aqueous extract had no significant effect on the lactic acid bacteria count, as there were no significant differences between the treatments of the aqueous extract and the control and antibiotic treatment.

الآليات البيولوجية للمستخلص المائي (*Alcea kurdic*a) على الأداء الإنتاجي والمحتوى الميكروبي لأمعاء فروج اللحم

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الخلاصة

ينصب التركيز الرئيسي لهذه الدراسة على التحقيق في الآليات البيولوجية التي يؤثر بها مستخلص (*Alcea kurdic*a) على إنتاجية فروج اللحم والمحتوى الميكروبي للأمعاء. تم تخصيص ما مجموعه 225 فرخ بشكل عشوائي لخمس معاملات , الأولى معاملة سيطرة (C-1) ، اضافة 0.5 غم Levosav كمضادات حيوية (Le-2) ، مع مستخلص مائي (*Alcea kurdic*a) 5 ٪ (AE-1) ، 10 ٪ (AE-2) ، و 15 ٪ (AE-3) لكل لتر ماء ، كل معامله تحتوي على 3 مكررات (15 طائرا كل تكرار). أوضحت النتائج ان هناك فروق معنوية بين معاملات التجربة في المقابل، كان هناك زيادة كبيرة في معدل وزن الجسم، ولوحظ زيادة وزن الجسم في جميع المعاملات التي تتلقى المستخلص المائي مقارنة مع(C-1) و (Le-2). بالنسبة لاستهلاك العلف ومعامل التحويل الغذائي أظهرت كل من معاملة السيطرة والمضاد الحيوي استهلاكاً أعلى مقارنة بالمعاملات الأخرى. وسجلت معاملات المستخلص المائي أفضل معدل لكفاءة التحويل الغذائي مقارنة بمجموعة السيطرة. أظهرت بيانات الفحص الميكروبي لأمعاء فروج اللحم التأثير الإيجابي لمستخلص ازهار الخطمي اذ انخفضت معنوياً اعداد البكتيريا الكلي وبكتيريا القولون في اللفائفي مقارنة مع معاملة السيطرة ومعاملة المضاد الحيوي بينما ارفعت معنوياً اعداد بكتيريا حامض اللاكتيك في المعاملة الخامسة (15%) مقارنة مع معاملة السيطرة وبقية معاملات التجربة. اما المحتوى الميكروبي للصائم فقد انخفضت معنوياً العدد الكلي للبكتريا في جميع معاملات المستخلص, وبكتيريا القولون انخفضت معنوياً في المعاملة الخامسة في حين لم يكن للمستخلص المائي تأثير معنوي على اعداد بكتيريا حامض اللاكتيك اذ لم توجد فروق معنوية بين معاملات المستخلص المائي وبين معاملة السيطرة والمضاد الحيوي.

الكلمات المفتاحية: ازهار الخطمي، فروج اللحم، المحتوى الميكروبي.

INTROUCTION

Recently, researchers have increased interest in medicinal plants and their extracts and their use as an alternative to antibiotics (Mombenin *et al.*, 2017, Kamil *et al*.,2021). They are considered one of the modern trends and strategies for sustainable poultry production as they are natural, non-toxic, and free of chemical residues and are considered safe, cheap, and easy to obtain alternatives (Mnisi et al., 2022). In addition to their effective role in promoting growth and production and improving the quality of animal products because they contain many biologically active, antimicrobial, anti-inflammatory, and antioxidant compounds (Rafeeq *et al*., 2022).

Alcea kurdica (AK), a member of the *Malvaceae* family is a medicinal plant known for its therapeutic properties (Abdulqader *et al.* 2023; Bahadori *et al*. 2022). Research on AK has focused on various aspects including the isolation and identification of bioactive compounds from its flowers and leaves such as flavonoids, phenols, and sterols (Abdulqader *et al.* 2023). In addition, studies have highlighted the potential of the plant as an anti-pest and immune-strengthening agent, showing positive effects on growth rate, immune status, and lipid profile (Jabbar, 2022). It improved the immunity of birds against New Castle disease (ND) and Infectious Bronchitis (IB) (Shokry and Mustafa, 2023). Studies have indicated that dietary supplements and extracts containing marshmallow extract can improve the growth rate, feed conversion ratio, and immune response of broilers.

This experiment aimed to investigate the biological activity of *Alcea kurdica* extract on the productivity and gut microbiome of Ross308 broilers and the possibility of using it instead of antibiotics as a form of sustainability in animal production.

MATERIAL AND METHODS

A total of 225 1-day-old broiler chickens with similar health conditions were randomly allocated into treatments provided the first as a control (C-1), Levosav as antibiotics (Le-2), with Aqueous Extract **(**AE) of (*Alcea kurdic*a) at doses 5% (AE-1), 10% (AE-2), and 15% (AE-3) per on drinking water, each treatment contained 3 replicates (15 birds each replicates). The birds were housed in floor pens as groups in random manner, each pen was enclosed by thick wooden boards to create a confined space, equipped with heating and cooling systems, feeders, separate water supply systems and meters to monitor daily water consumption. The environment was maintained with 24-hour light, proper air ventilation, and under standard management conditions. The experimental diet was formulated according to the two phases: 1-7 days and 8-35 days.

Prior to commencing the experiment, replicates were weighed to determine the initial weight, and the 7th and 35th days of the experiment were recorded as the final weight. Feed consumption for each replicate 7 days before the experiment and the whole period was calculated, and the feed consumption of dead chickens and eliminated chickens was corrected. The average body weight of broilers at 7 days and 35 days, and the average daily gain, average daily feed intake and feed conversion rate (FCR) of broilers weekly.

Microbial sample collection:

Collect samples from different segments of the broiler's intestinal tract, ileum (Apajalahti et al., 2004; Torok et al., 2011), and Microbial enumeration: Perform serial dilutions of the intestinal samples and culture the dilutions on selective media to enumerate the populations of specific microbial groups, such as total bacteria, lactobacilli, and *E. coli* according to (Jozefiak et al., 2010).

Statistical Analysis

Excel 2020 was used for preliminary data sorting, and SAS 9.4 was used for variance analysis and multiple comparison of data among different dietary treatment groups. All test data were expressed as "mean ± standard deviation". P<0.05 was considered as significant difference (Dancun, 1953).

RESULTS AND DISSCUSION

The results of the table (1) show the effect of adding the aqueous extract of AK flower to drinking water on the average live body weight of broilers. It was noted from the results of the experiment that there were no significant differences at the level of ($p \leq 0.05$) between the coefficients of aqueous extract and antibiotic compared with the control group in the rate of live body weight at the first and second weeks. While we note in the third week there was a moral effect of the concentration of the aqueous extract, as the body weights of the birds that were added to their drinking water with the aqueous extract of the *A.kurdica* flower increased morally at a concentration of 10% compared to the concentrations of 15% and 5%. In general, there were no moral differences between the birds of these transactions and the birds of the antibiotic and control treatments. While in the fourth week we observe a gradual increase in the weight of the birds with increasing concentration, as the fifth treatment surpassed 15% over all the experimental and control treatments. As for the fifth week, we find the superiority of all the transactions of the aqueous extract of the *A.kurdica* flower over the antibiotic treatment and the control treatment, and the high concentration of the extract had the obvious effect, as the fifth treatment increased 15% significantly over the fourth and third treatments 10%, 5%, respectively.

Table 1: Biological Mechanisms of the Aqueous Extract of (*Alcea kurdic*a) on body weight (g)

Age	C-1	Le-2	AE-1 (5%)	AE-2 (10%)	AE-3 (15%)
7 d	175.22±1.73 a	176.99±2.91 a	175.77±1.06 a	176.22±197 a	177.33±1.01 a
14 d	476.22±4.62 a	471.55±3.28 a	461.89±2.77 a	470.22±15.93 a	471.55±1.46 a
21 d	966.10±2.80 ab	976.51±26.14 b	939.80±21.92 b	1035.41±32.73 a	961.10±2.22 b
28 d	1611.11±8.23 c	1639.86±9.49 b	1598.14±4.18 c	1658.35±9.35 b	1686.66±6.94 a
35 d	2364.42±12.57 c	2400.51±3.51 c	2460±21.18 b	2461.54±20.91 b	2520.19±5.60 a

Values are expressed as Mean ± SE. Means bearing different superscripts in the same row differ significantly (P < 0.05).

The results of the table (2) show the effectiveness of adding the aqueous extract of *A.kurdica* flowers to drinking water in the rate of weight gain of broilers, as it did not have a significant effect in all the parameters of the experiment per week(1-2) . At the age of (2-3) weeks, the fourth treatment was morally superior to the third treatment and showed no significant differences between the second and fifth treatments compared to the control group, while the third treatment recorded the lowest level of this trait. While at (3-4) weeks, the fourth treatment recorded a significant decrease in the rate of weight gain compared to the birds of the fifth treatment, which recorded the highest rate, while there were no significant differences between them and the rest of the transactions. At the age of (4-5) weeks, the third and fifth treatments were morally superior to the control treatment and the second treatment, and there were no moral differences between them and the fourth treatment. We note from the rate of total weight gain from the age of (1-5) weeks a moral superiority of the aqueous extract of *A.kurdica* flower over the control and antibiotic treatments, and the fifth treatment gave the best rate, as it morally surpassed all the transactions.

Table 2: Biological Mechanisms of the Aqueous Extract of (*Alcea kurdic*a) on body weight gain (g)

Age	C-1	Le-2	AE-1 (5%)	AE-2 (10%)	AE-3 (15%)
7-14 d	301.0±5.73 a	294.55±0.58 a	286.12±3.30 a	294.0±14.10 a	294.23±2.40 a
15-21 d	489.88±6.95 ab	504.±23.70 Ab	477.91±20.20 b	565.19±40.73 a	489.55±2.42 ab
22-28 d	645.0±9.51 b	663.35±34.37 ab	658.33±22.49 ab	622.93±32.25 b	725.55±5.55 a
29-35 d	753.31±16.55 b	760.65±11.79 b	862.71±20.52 a	803.19±29.80 ab	833.52±7.70 a
7-35 d	2189.20±11.47 c	2223.52±2.92 C	2285.08±22.24 b	2285.32±18.95 b	2342.86±4.87 a

Values are expressed as Mean ± SE. Means bearing different superscripts in the same row differ significantly ($P < 0.05$).

The results of the table (3) show the effectiveness of adding water extract of *A.kurdica* flower to the drinking water of broilers in the rate of feed consumption, we note at the age of (1-2) weeks the antibiotic treatment was morally superior to the experimental coefficients and there were no moral differences between them and the control treatment and the fifth treatment was morally superior to the third and fourth treatments and the latter did not show any moral differences between them. At week (2-3), the antibiotic treatment recorded the highest rate of feed consumption and the fourth treatment the lowest rate compared to the control treatment and the rest of the trial transactions. The antibiotic treatment continued to outperform it, registering the highest rate at the age of (3-4) weeks and thus morally outperformed ($p<0.05$) the control treatment and the rest of the transactions, while the fourth and fifth transactions decreased morally compared to the control treatment and the third treatment, the latter did not show any significant differences with the control treatment. At the age of (4-5) weeks, the second treatment was morally superior to the third treatment and there were no moral differences between it and the control treatment and the rest of the transactions. From the observation of the total duration of the experiment (1-5) weeks, we find a significant decrease in the rate of feed consumption in the aqueous extract of *A.kurdica* flowers compared with the control treatment and antibiotic treatment and a significant decrease in the fourth treatment compared with the third and fifth treatments.

Table 3: Biological Mechanisms of the Aqueous Extract of (*Alcea kurdic*a) on feed intake (g)

Age	C-1	Le-2	AE-1 (5%)	AE-2 (10%)	AE-3 (15%)
7-14 d	365.88±1.55 a	371.32±4.70 A	363.22±1.54 c	332.33±1.07 c	355.66±0.57 b
15-21 d	623.33±3.34 b	647.74±0.56 A	622.65±3.20 b	601.09±0.62 c	625.66±1.38 b
22-28 d	866.66±5.03 b	904.06±2.46 A	871.42±4.17 b	840.71±4.01 c	851.39±0.71 c
29-35 d	1063.5±33.21 ab	1078.29±2.23 A	1016.80±1.72 b	1038.01±1.25 ab	1061.85±0.89 ab
7-35 d	2919.45±9.10 b	3001.42±4.83 A	2847.10±4.86 d	2815.15±5.88 e	2894.57±2.14 c

Values are expressed as Mean ± SE. Means bearing different superscripts in the same row differ significantly (P < 0.05).

The table (4) shows the effectiveness of the aqueous extract of *A.kurdica* flowers on the nutritional conversion coefficient, The results indicate that there were moral differences in the first week, as the fourth treatment with a concentration of 10% gave the best rate of this trait compared to the antibiotic treatment, while there were no moral differences between it and the two concentrations of 5%, 15% and the control treatment, as well as in the second week, the fourth treatment also recorded the best rate of the food conversion factor, as it decreased morally compared to the control treatment and the rest of the trial transactions, while in the third week the fifth treatment was the best, as it decreased morally compared to the antibiotic treatment and there were no moral differences between it and the control treatment and the rest of the trial transactions. In the fourth and fifth weeks, we note the moral effect of the aqueous extract of *A.kurdica* flower and all concentrations on the rate of food conversion coefficient, as it decreased morally in all transactions of the aqueous extract compared with the control and antibiotic transactions.

Table 4: Biological Mechanisms of the Aqueous Extract of (*Alcea kurdica*) on feed conversion

Age	C-1	Le-2	AE-1 (5%)	AE-2 (10%)	AE-3 (15%)
7-14 d	1.216±0.02 ab	1.260±0.01 A	1.175±0.01 ab	1.135±0.05 b	1.208±0.01 ab
15-21 d	1.273±0.02 a	1.288±0.05 A	1.307±0.05 a	1.074±0.07 b	1.278±0.01 a
22-28 d	1.344±0.01 ab	1.370±0.07 A	1.327±0.05 ab	1.357±0.07 ab	1.173±0.01 b
29-35 d	1.412± 0.04 a	1.418±0.02 a	1.179±0.02 c	1.295±0.04 b	1.274±0.01 bc
7-35 d	1.311±0.01 a	1334±0.002 a	1.247±0.01 b	1.215±0.01 c	1.233±0.003 bc

Values are expressed as Mean ± SE. Means bearing different superscripts in the same row differ significantly (P < 0.05).

Table 5: Biological Mechanisms of the Aqueous Extract of (*Alcea kurdica*) on gut flora

Type	C-1	Le-2	AE-1 (5%)	AE-2 (10%)	AE-3 (15%)
			ileum		
Total bacterial	13.50±0.34a	11.37±0.19b	12.47±0.25a	10.67±0.57bc	10.17±0.12c
E coli	13.12±0.62a	12.57±0.08a	11.15±0.56b	10.10±0.04b	11.15±0.12b
Lactobacillus	10.70±0.78b	10.94±0.70b	10.65±0.48b	8.30±0.04c	12.87±0.04a
			Jejunum		
Total bacterial	11.00±0.69 b	11.63±1.08a	8.45±0.27d	9.55±0.29 c	9.27±0.04 c
E coli	12.60±0.75a	11.35±0.58ab	11.37±0.27ab	10.00±0.15bc	9.55±0.13c
Lactobacillus	10.12±0.69b	12.50±0.38a	11.45±0.57ab	10.17±0.14b	11.30±0.04ab

Values are expressed as Mean ± SE. Means bearing different superscripts in the same row differ significantly (P < 0.05).

We note from the results of the statistical analysis shown in the table (5) the effect of different concentrations of *A.kurdica* flower aqueous extract on the microbial content of broiler intestines throughout the 5 weeks of the experiment, noting a significant decrease in the total number of bacteria in the ileum with an increase in concentration as the fifth treatment (15%) recorded the lowest value of this attribute and reached (10.17) compared with the control treatment and antibiotic treatment (11.37,13.50) respectively , This was followed by the fourth treatment, which decreased from significantly compared with the control treatment while between it and the antibiotic treatment there were no significant differences .While the aqueous extract of *A.kurdica* flower for all concentrations had a pronounced effect on the numbers of total bacteria, it was significantly reduced in all transactions compared with the control and antibiotic treatments . While the high concentration played a role in increasing the numbers of beneficial bacteria for the ileum , the numbers of *Lactobacillus* increased significantly in the intestines of the birds of the fifth treatment (15%) compared with the birds of the control treatment and the rest of the experimental transactions, while their numbers decreased significantly in the birds of the fourth treatment compared with the birds of the control and antibiotic treatments and the third treatment, which did not have a significant effect in this capacity. From the results of the same table, we note the significant effect of *A.kurdica* flower aqueous extract on the total number of Jejunum bacteria, as their numbers decreased significantly in all transactions compared with the control and antibiotic treatments, as for colon bacteria, they were affected by high concentrations of *A.kurdica* flower extract, they decreased significant in the fifth treatment (15%) and mathematically in the fourth treatment (10%) compared with the control and antibiotic treatments, reaching their numbers (9.55, 10) and(11.35, 12.60) respectively .

While the aqueous extract did not affect the number of Lactobacillus, as there were no significant differences between the third, fourth and fifth transactions and the control and antibiotic treatment, except for the fourth treatment, which decreased significantly compared to the antibiotic treatment. Although there were no significant differences between the number of Lactobacillus of the Jejunum bacteria in birds treated with the aqueous extract and the control and antibiotic treatments, they maintained the microbial balance of the intestine, which was positively reflected in the productive performance of the birds.

The poultry industry is currently facing great challenges as a result of intensive breeding, improvement, and rearing operations to meet the market requirements of poultry products, which negatively affected their immune system, making them more sensitive to diseases, especially after the international ban on the use of antibiotics in the animal production sector (Tufan *et al*., 2015; Abdulwahid *et al*.,2022). Therefore, researchers have turned to the use of medicinal plants and their extracts as feed additives and as alternative strategies to antibiotics (Jamil *et al.,* 2022 ; Shaker *et al.,*2023, Saed *et al*.,2025) , as Hussein *et al.*,2021 and Beghout *et al.*,2022) indicated the role of medicinal plants in improving the productive performance of broilers and increasing the utilization of feed by enhancing immunity and antioxidant systems and improving the secretions of digestive enzymes such as amylase, protease, and trypsin, thus utilizing maximize the nutrients in the feed and increase their absorption by increasing the permeability of the intestinal mucosa and preserving it from oxidative stress caused by pathological microbes or their toxins and scavenging free radicals, which reflects positively on the health status of the broilers. There is a link between gut health and bird health and performance, as the gut ecosystem plays an important role in removing infectious agents and toxins from the gut (Diaz Carrasco *et al*., 2019). This was observed from the results of our experiment, as the application of the aqueous extract of *A. kurdica* increased body weight, improved metabolic conversion factor, maintained gut microflora, and inhibited the growth of pathogenic microbes in the intestine. Alshawsh *et al* (2012) reported the ability of *A. kurdica* to inhibit the growth of Gram-negative and Gram-positive bacteria, yeasts, and filamentous fungi. The same result was obtained by Mahbuba et al. (2022), as the aqueous extract of *A.kurdica* showed an antibacterial effect against E.coli and enterococci. This is because it contains many active compounds such as flavonoids and phenols (Gallic acid, Syrinigic acid, Quercetin, and Chrologenic acid) that are characterized by their antioxidant and antimicrobial activity (Mohammed *et al*. 2022). Phenolic acids modify mucosal cell membranes (Jabbar, 2022) by increasing their hydrophobicity and thus affect the adhesion of bacteria to the intestinal mucosal walls (Mohiti-Asli and Ghanaatparast-Rashti, 2018). Phenols are characterized by their lipophilic nature, enabling them to penetrate the microbial cell membrane and accumulate in the lipid layer of the membrane and mitochondria (Christaki *et al.*, 2020). They interfere with the electron transport chain and cellular respiration and reduce ATP production, leading to disruption of bacterial cell function (Kubo *et al.*, 2003). There are many studies that emphasize the role of Gallic acid, one of the most abundant phenolic acids in *A. kurdica*, and its antioxidant and antimicrobial activity by inhibiting DNA gyrase, which is involved in the synthesis of bacterial DNA and RNA, leading to programmed cell death (Kahkeshani *et al*., 2019).

CONCLUSION

The use of *A. kurdica* flower aqueous extract improved the production performance of broilers and increased the utilization of nutrients in the feed by maintaining intestinal health from oxidative stress caused by pathological microbes, as the phenolic acids contained in the extract inhibited the growth of *E.coil* and total bacteria and maintained the microbial balance in the intestine, so *A.kurdica* is an effective alternative that competes with antibiotics.

CONFLICT OF INTEREST

The authors declare no conflicts of interest associated with this manuscript.

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