O r i g i n a l i n v e s t i g a t i o n s / c o m m e n t a r i e s

Effect of adding Zeolite to the diet in health status of broiler infected with pathogenic *E. coli O157:H7*

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Abstract. This study was conducted at environmentally controlled chicken housed to study the protective effect of Zeolite against bacterial infection induced by orally infected with *E.Coli O157:H7*, by adding Zeolite at concentration 2%. conducted in the poultry farm in the college of veterinary medicine/ university of Baghdad lasted from 4/5/2022 to 20/7/2022. 180chicks (Ross 308) one day old were divided into four equal groups (45/group), with 3 replicates (15/replicate). The results of this study recorded significant ($P \leq 0.05$) differences between groups in all results.The group G4 (2% Zeolite) recorded significant ($P \leq 0.05$) decrease in the log count of *E.Coli O157:H7* in the intestine, compared to groups G2 andG3, group G4 illustrated significant ($P \leq 0.05$) increase in RBCs and Hb value compared to G3 (antibiotic group), while the control group G1 and G4 recorded significant ($P \leq 0.05$) decrease in WBCs and H/L compared with G2 and G3.The results obtained significant ($P \leq 0.05$) increase in the value of ALT and AST of G2 and G3.Histomorphology of Liver and kidney in broiler infected with *E. coli O157:H7* in groups G4 supplemented with 2% Zeolite showed no important pathological change .

Keywords: Zeolite, broiler, E. coli O157:H7

Introduction

Poultry production is an important economic activity in Iraq (Kanaan et al, 2019). Both, the feed industry and the poultry production sector, suffer from huge losses due to the contamination of food with pathogenic bacteria and impacts in the animal. This is lead to highly use of antibiotics at the same time dissemination of antibiotic resistant strains of pathogenic and non-pathogenic organisms (Agyare *et al*., 2018). *Ecoli* especially E. coli O157:H7 strain is one of the mostly responsible for outbreaks of food-borne illnesses in developing nations (Khudhir, 2021), saab Khudhir, 2020) ,(Al-Shedidi, 2017) and (Al-Khyat, 2008). In chicken, Infections with E. coli include egg peritonitis, omphalitis, swollen head syndrome, cellulitis, and colisepticaemia (Azza *et al*., 2018). This prompted several studies to examine the characteristics of natural materials that enhance poultry production without hazard on health of both animals and human and minimize the use of antibiotics (Al-Yasiri & Mohammed, 2019). Zeolite has been one of these natural materials that has been in the spotlight recently by many researchers (Volostnova *et al* .,2022).It may enhance the production of poultry in a number of ways, like decrease in feed intake without losing important nutrients and vitamins (Pavlak *et al* ., 2022). Zeolites have been shown to have bactericidal effects (Sienkiewicz & Członka, 2022). In addition to having a large surface area and good porosity, which allows them to selectively absorb bacteria and produced plaques, shielding the vital tissues from bacterial infection (Zarrintaj *et al*., 2022). Also decrease in broiler chicken mortality and a decrease in viable counts of Salmonella enteritidis and Escherichia coli in the proximal and distal intestines (Hassan *et al*., 2018).

Materials and Methods

Experimental design

180 birds from the Ross broiler source (Belgian) were used in the experiment as day-old chicks, from a local hatchery, At arrival, chicks were weighed and dispersed at random into a floor pen coated in wood shavings, where they were subsequently separated into four equal groups (45 per group) with three replicates (15 each replicate). The first group was give basal diet as control negative group (G1). In the groups (G2, G3 and G4) infected with challenge dose of *E.coli O157:H7* orally at 27 days old. The ciprofloxacin was given to the antibiotic group (G3) only after three days 100ml/200L (each ml contain 100mg ciprofloxacin) for 5 days. In the (G4) the Zeolite 2% was added to the diet throughout the experiment period from the one day old to marketing (the source of Zeolite from Al-Rahimiya located in the western side of the Najaf city (Yasir & Al-Janabi, 2020). The chicks remained under observation until the age of 35 days

Inoculation procedure

The *E. coli* strain used for inoculation, *O157 H7*, was isolated from of liver of broiler chicks. Detected via

MacConkey Sorbitol Agar (Sorbitol Agar), HiCrome™ UTI Agar and Confermed by PCR 16s.

Dose prepared and counted before (18-24) hours from challenging control and treated broilers. A 250 ml of boosting doubled enriched TSBYE broths were inoculated by ten purified seed colonies of *E. coli O157 H7* then mixed and homogenized by vortex. Incubation at 37 C overnight was followed, then counting regime via modified built-in roll-pour plate decanting technique for approximately adjusting microbial log titer to McFarland ten logs (10^8 CFU.ml^{-1}) for each ml (Elutade *et al*., 2019).

Then the suspension was diluted 1:100 with sterile broth to obtain a cell number of approximately 106 CFU/ml (Azar Daryany *et al*., 2008).

Blood collection

Blood samples were collected from 2 birds from each pen from the wing vein in a test tube without anticoagulant for serological parameters and with coagulant for complete blood count according (Parasuraman, 2010). The serum was separated by centrifugation for 10 minutes at 3000 rpm and stored in a deep freeze (-20) until analysis.

Cloacal swabs

Using sterile cotton swabs, cloacal swabs were taken from 24 randomly selected birds to ensure that they are free from *E coli O157H7* before induced infection and put the samples in sterile (Universal bottle) contain 20-25 ml from Tryptone Soya Yeast Extract Broth (TSB-YE) then incubated in the incubator for 24 hours at 37°C. After the induced infection, the procedure was done in the same way and then a bacterial culture and count was performed (Barrow et al., 1986).

Letter samples collection:

Using a clean and sterilized spatula, 24 samples of poultry litter were randomly collected to ensure that they are free from *E coli O157H7* before induced infection and put (0.5-1g) in sterile (Universal bottle) contain 20-25 ml from Tryptone Soya Yeast Extract Broth (TSB-YE) then incubated in the incubator for 24 hours at 37°C. After the induced infection, the procedure was done in the same way and then a bacterial culture and count was performed (Barrow et al., 1986).

The Statistical Analysis

Statistical analysis

Statistical analysis of data was performed using SAS (Statistical Analysis System - version 9.1). One-way ANOVA and Least significant differences (LSD) post hoc test were performedto assess significant differences among means. P < 0.05 is considered statistically significant.
Means with a different small letter in the same column are significantly different (P<0.05)
Means with a different capital letter in the same row are significantly different (P<0.05)

Results

Escherichia coliO157:H7 count after experimental challenge

In the table (1) the results of *Escherichia coli O157:H7* serotype count from cloacae and letter showed high significant (P ≤ 0.05) decreased at G4 (Zeolite group) compared with G2 and G3.

Table 1. Estimated count of *Escherichia coliO157:H7* serotype from cloacae and letter (log 10 CFU/g).

Groups	Cloacae	Letter
G2	B7.37±0.31a	A8.30±0.25a
G3	B6.34±0.09b	A7.28±0.14b
G4	B4.09±0.04c	A6.44±0.28c

LSD= 0.62, N= 24.

The effected of addition 2% Zeolite on ALT and AST of broiler infected with *E. coli O157:H7.*

The effect of supplant diet 2% Zeolite on ALT and AST enzymes of broiler infected with *E. coli O157:H7* in table (2). The results illustrated significant differences (P ≤0.05) among groups in the value ALT and AST. The mean value of ALT recorded significant (P ≤0.05) increase in group G2 compared to the control and other groups. No significant (P ≤0.05) differences among G1, G3 and G4. The mean value of AST illustrated significant (P ≤0.05) decrease difference in group G1 and G4 compared to G2 and G3. The group G2 recorded high significant (P ≤0.05) increase in the AST compared with G1, G3 and G4.

Table 2. The effected of addition 2% Zeolite on ALT and AST (U/L) of broiler infected with *E. coli O157:H7.*

parameters	RBC (x106/ul)	HB (g/dl)	PCV (%)
G1	2.99±0.56ab	13.40±0.85a	30.52±3.45a
G2	2.62±0.49ab	8.48±0.59b	28.36±2.65a
G3	2.27±0.59b	8.70±0.69b	25.92±2.95a

G4	3.07±0.15a	11.94±0.49a	30.04±2.66a
LSD	1.48	2.01	8.83

LSD ALT=10.76, AST=12.09. n=24
RI (UL) of ALT =7-55 AST=70-220 (Makama *et al*., 2021).

Effect of dietary supplementation of the 2% Zeolite on hematological parameters of broiler infected with *E. coli O157:H7*:

The effect of supplant diet 2% Zeolite on haematological parameters of broiler infected with *E. coli O157:H7* in table (3) .The results illustrated significant differences (P ≤0.05) among groups in the RBC, HB and PCV. Significant increase (P ≤0.05) were obtained in RBC value at G1, G2 and G4, There was superiority of group G4 (Zeolite) which showed a highest value compared to other groups (3.07±0.15).A high significant (P ≤0.05) increase in the HB was observed in G1 and G4 compared with G2and G3. The PCV illustrated no significant (P ≤0.05) difference among groups.

Table 3. Effect of dietary supplementation of the 2% Zeolite on RBC, HB and PCV of broiler infected with *E. coli O157:H7*.

Groups	ALT	AST
G1	55.60±2.99b	216.20±1.96b
G2	82.40±5.68a	231.40±5.40a
G3	60.60±2.51b	227.20±5.51ab
G4	54.60±2.03b	216.20±0.97b
LSD	10.76	12.09

LSD RBC=1.48, HB=2, 01, PCV=8.83, n =24. Means with a different small letter in the same column are significantly different (P ≤0.05)
*RI: RBC (x106/ul) =2.50-3.90, HB (g/dl), PCV (%) =30.00-49.00
*(Al-Nedawi, 2018).

Effect of dietary supplementation of the 2% Zeolite on Leukocytes counts of broiler infected with *E. coli O157:H7*:

The effect of supplant diet 2% Zeolite on Leukocytes counts of broiler infected with *E. coli O157:H7* in table (4). The results illustrated significant differences (P ≤0.05) among groups in the lymphocyte, Monocyte and HeterophilThe result recorded significant decrease (P ≤0.05) in WBC at G1 and G4 compared with G2and G3. A high significant (P ≤0.05) increase of the lymphocyte were observed in G1 and G4 compared with G3 and G2. Significant decrease (P ≤0.05) in Monocyte and Heterophil were obtained at G1 and G4 compared with G2and G3.The ratio of Heterophil / lymphocyte illustrated low significant difference in group G1, G2and G4 compared to G3, a group G4 decrease recorded lowest significant (P ≤0.05) value.

Table 4. Effect of dietary supplementation of the 2% Zeolite on WBC= 1.01, LYM, MON, HETER, H\L of broiler infected with *E. coli O157:H7*.

Groups	WBC	LYM%	MONO%	HETRO%	H/L Ratio
G1	5.84±0.36b	94.20±1.50a	5.12±1.41b	0.68±0.19b	0.007±0.002bc
G2	7.97±0.23a	88.40±2.00b	10.24±1.63a	1.36±0.37a	0.016±0.004ab
G3	7.01±0.20a	86.48±1.78b	11.78±1.30a	1.74±0.48a	0.021±0.006a
G4	5.58±0.47b	97.76±0.17a	2.10±0.17b	0.14±0.02b	0.001±0.0002c

LSD WBC= 1.01, LYM= 4.61, MON= 3.78, HETER=0.56, H\L= 0.011, N=24
RI of WBC (x10^6/ul) =1.90 –9.50 *(Al-Nedawi, 2018).

Effect of dietary supplementation of the 2% Zeolite on Liver and kidney of broiler infected with *E. coli O157:H7:*

Histopathological of liver (G1) shows vascular degeneration of hepatocytes and sinusoidal congestion (fig. 1 and 2). While the kidney (fig.3) showed normal appearance of glomerulus and renal tubules. Liver histomorphology of broiler infected with E. coli O157:H7 in group control positive (G2) showed kuffer cells infiltration , vascular degentration hepatocytes and sinusoids congested (fig.4).The kidney showeddilated bumman and hemorrhage with congestion (fig.5).Also multifocal area of mononuclear cells infiltration, MNC infiltration in interstitial area (fig.6).Liver histomorphology of broiler infected with E. coli O157:H7 in group infected with antibiotic (G3) showed mild dilated sinusoids , acute cellular swelling hepatocytes , mild lymphocytic infiltration (fig.4.7). also infiltration of mononuclear cells in hepatocytes ,necrotic with narrowing sinusoids , fatty change hemorrhage and congestion blood vessels (fig.8).The figure of the kidney showed congestion of central vein and normal renal tubules (fig. 9). Also multi focal area of lymphocytic granuloma, hypercelluratiy of tubules nuclei and intertubular hemorrhage (fig. 10).

Liver histomorphology of broiler infected with E. coli O157: H7 in group Zeolite (G4) showed mild diffused vacuolar degeneration of hepatocytes and congested sinusoids blood vessel **((**fig. 4.11). The kidney showed normal appearance of glomerulus and renal tubules (fig.4.12). But founded mild diffused hemorrhages (fig.4.13).

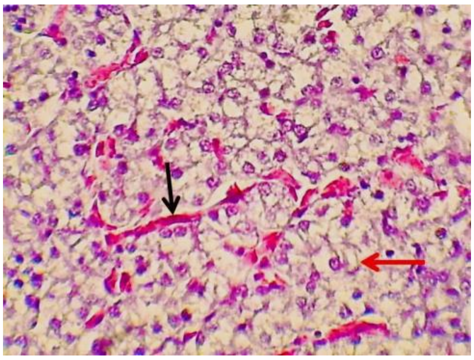

Figure 1. Histopathological of liver (G1) shows vascular degeneration of hepatocytes (Red arrow) & sinusoidal congestion (Black arrow) H&E stain.400x.

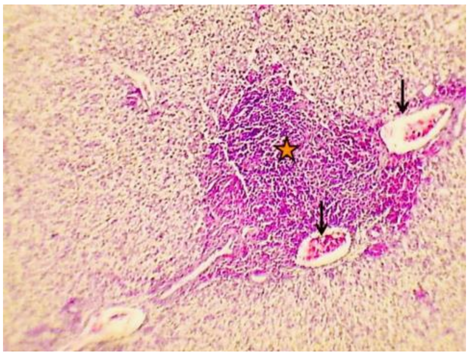

Figure 2. Histopathological of liver (G1) shows mild vascular congestion (Arrows) & peri lymphocytic cuffing of lymphocytes. H&E stain.100x.

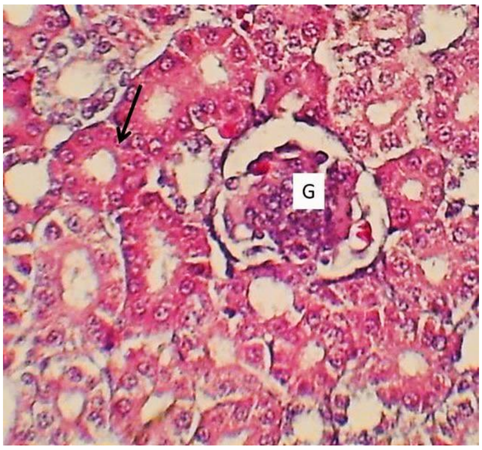

Figure 3. Histopathological of kidney (G1) normal appearance of glomerulus (G) & renal tubules (Arrow). H&E stain.400x

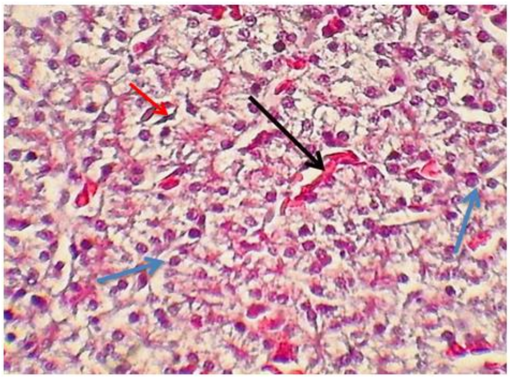

Figure 4. Histopathological of liver (G2) shows: a- kuffer cells infiltration (read arrow)**,** b- vasculadegentration hepatocytes (blue arrow), c- sinusoids congested (black arrow). H&E stain.400x.

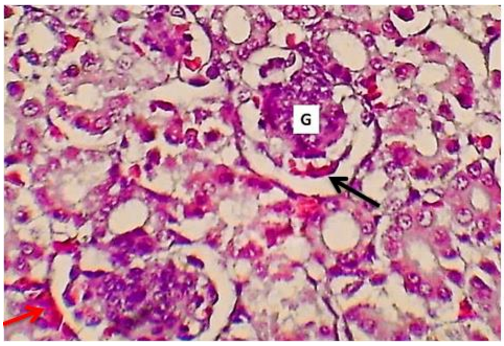

Figure 5. Histopathological of kidney/cortex layer (G2): a- dilated bumman space (black arrow) b- hemorrhage & congestion (read arrow). H&E stain.400x.

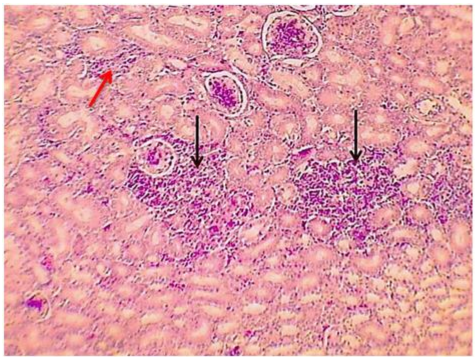

Figure 6. Histopathological of kidney (G2) shows: multifocal area of mononuclear cells infiltration. MNC infiltration in interstitial area (Red arrow) H&E stain.100x

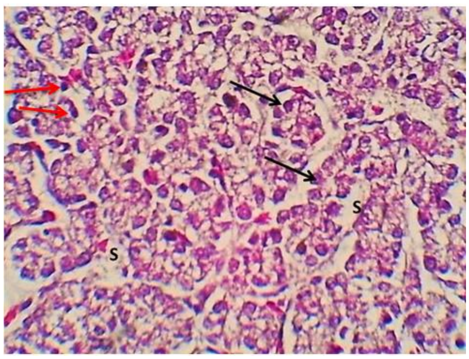

Figure 7. Histopathological liver group (G3) show: a- mild dilated sinusoids (s), b- acute cellular swelling hepatocytes (black arrow), c- mild lymphocytic infiltration .100x

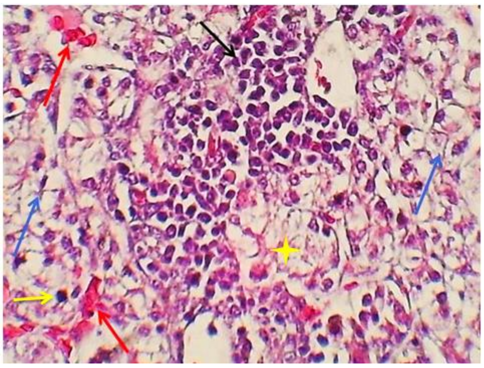

Figure 8. Histopathological liver group (G3) show: a- infiltration of mononuclear cells in hepatocytes (black arrow), b- necrotic hepatocytes (star) with narrowing sinusoids (blue arrow), d- fatty change (yellow arrow), e- hemorrhage and congestion blood vessels (read arrow) 400x.

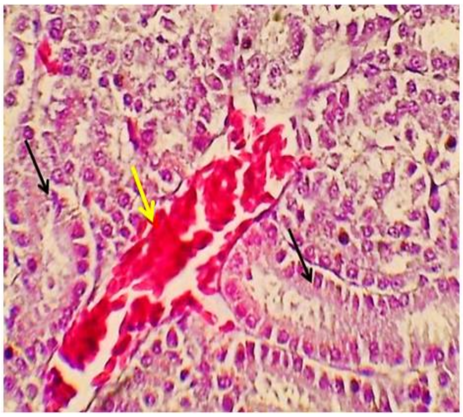

Figure 9. Histopathological kidney (G3) shows: congestion of central vein (yellow arrow) and normal renal tubules (Black arrow). 400x

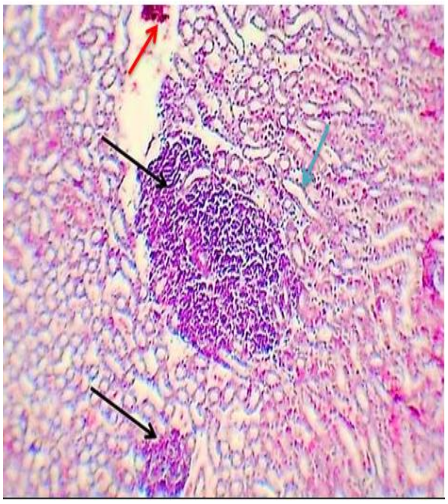

Figure 10. Histopathological kidney group (G3) show: a- Multi focal area of lymphocytic granuloma (black arrow), b- hypercelluratiy of tubules nuclei (blue arrow), c- intertubular hemorrhage (read arrow). 100x

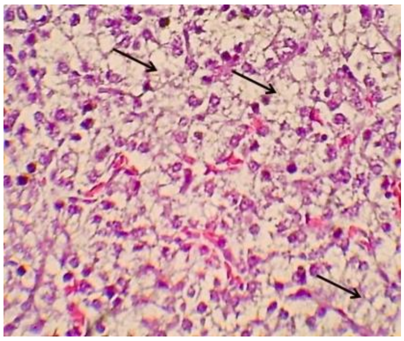

Figure 11. Histopathological liver group (G4) show: a-mild diffused vacuolar degeneration of hepatocytes (Arrows) b-congested sinusoids blood vessel (read arrow) .H&E stain.400x.

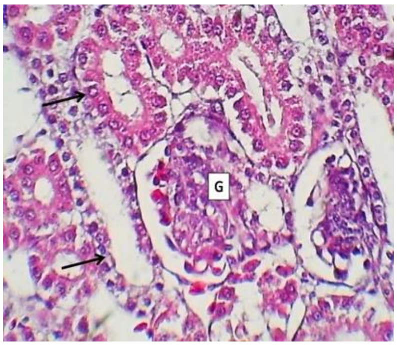

Figure 12. Histopathological kidney group (G4) normal appearance of glomerulus (G) & renal tubules (Arrows). H&E stain.400x

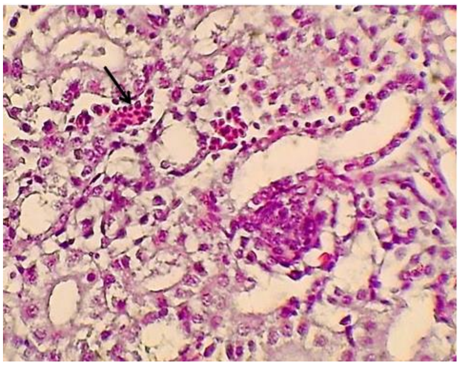

Figure 13. Histopathological kidney group (G4) show: mild diffused hemorrages (nucleated RBCs). H&E stain.400x

Discussion

Escherichia coliO157:H7 count

The results after induce infection at table (1) was noticed that the Zeolite 2% can reduced of bacterial count in cloacae and increase the shedding of bacteria in litter (washing out) so lead to increase bacterial log at litter. This result agree with (Al-Nasser et al., 2011) reported that adding Zeolite in the broiler feed is beneficiary in reducing Salmonella contamination in ceca. (Prasai et ql., 2017) suggested that the low (1%) and medium dosage (2%) of zeolite in feed have the ability to reduce poultry pathogens including levels of Escherichia-like and low abundant

Salmonella reduced by zeolite without disturbing beneficial bacteria. (Mallek et al., 2012) reported that addition of Natural zeolites (0.5 or 1% w/w) in a broiler diet produced a significant (P < 0.05) reduction in total culturable microbial levels, as compared to the control group. The effect of zeolite particles against bacteria (bactericidal or bacteriostatic effect phylotypes) like (washing out) and reducing abundance in the gut. (Prasai et al., 2016) reported that zeolite can selectively remove pathogens without reducing microbial richness and diversity in the poultry gut. This also explains the higher percentage of bacteria in the letter than in the cloaca. Zeolite also exhibits antibacterial effects, including activity against Salmonella spp. and Escherichia coli (Tang et al., 2014).

E coli O157 H7 in current study show high resistant to ciprofloxacin and this is agree with (Anyanwu et al., 2022) reported that when compared to isolates from cattle and pigs, chicken isolates *of E coli O157 H7* have much greater rates of ciprofloxacin resistance. According to (Akinduti et al., 2022) evidence of potential resistance gene transmission from fecal shedding or feed into rivers, streams, and wells is the high antibiotic resistance of water and animal Escherichia coli O157 to ciprofloxacin, gentamycin, and of loxacin, which are first-line drugs. Reduced sensitivity of *E. coli* to ciprofloxacin was confirmed in study of (Kocúreková et al., 2021) namely, in 60.78% of broilers. Also (Hess et al., 2022) reported that multidrug resistance (MDR) was found in the majority of isolates (200/209) for Escherichia coli isolated from poultry and seventy-seven isolates proved extensively drug-resistant (XDR). The results didn't agree with (Ahmad et al., 2022 and Halder et al., 2022) reported that E. coli isolated from proiler was highly sensitive against ciprofloxacin.

ALT and AST enzymes:

when ALT increased this could be an indication of tissue damage especially when it was accompanied by the increased activity of other serum enzymes like AST (Safaeikatouli *et al.*, 2011) and this is clear accrued with groups of Bacterial infected and Bacterial infected with antibiotic. Our results agree with (Emam *et al.*, 2019) founded that not increased of ALT and AST for hens drank tap water and fed diet containing 2 % Zeolite. Our results disagree with (El-Ghany& Ismail, 2014) reported that ciprofloxacin displayed the lowest significant (P<0.05) levels of liver enzyme during infection poultry with E coli. And near with (Youssef *et al*., 2019) infected with *E coli* and treated broiler with levofloxacin are revealed a significant increase in (AST & ALT). Elevated levels of liver enzymes in serum indicate the destruction of hepatocytes and dysfunction of the liver. And these findings indicate that the 2% Zeolite has a potential to limit the tissue injuries induced by pathogenic agents.

Hematological parameters:

The results at table (3) showed the Zeolite 2% can help to enhance the hematological parameters and this agree with (Emam *et al.*, 2019) noticed enhance in the blood hematological parameters of hens fed with Zeolite 2 and 4 % that may be due to the good effect of the additional the Zeolite. Also, zeolites play important role in ion-exchangers, participate in certain biochemical transformations, normalize the homeostasis of animals and increase the nutrient conversion, so it lead to reversible in increase of RBC's and Hb values (Andronikashvili *et al.*, 2009). But (El-Hady& Mohamed, 2020) observed that values of haematological parameters of RBCs, Hb, PCV, ratio didn't differ between natural Zeolite and control group.

The current study recorded that the ciprofloxacin caused a decrease in red blood cells, and this was documented in a previous study by (Prisnyi *et al.*, 2022) founded some decrease in RBC count in chickens after administration of Ciprofloxacin during infection with gram negative bacteria . It is a well-known fact that use of fluoroquinolones may cause changes in some blood parameters like development of anemia alsoincrease in erythrocyte sedimentation rate. Also, the drug may have a short-term and reversible effect on haematopoiesis. For instance, it has been described that Temafloxacin triggers haemolytic reactions in the hematopoietic system. Also our results near to (Anaruzzaman *et al*., 2021) who suggested that use of ciprofloxacin for a specific period had some effects on hematological parameters in infected poultry like Hb, and PCV were the highest in control group but no statistical significance in difference of meansamong discriminate and indiscriminate groups, the antibiotics have effects on hematopoiesis and hemolysis but the effects are not significant in small duration. This might be significant if used for longer duration. Our results did not agree with (Noman *et al.*, 2015) who reported not significant of Hb and RBC and of birds in treated group with ciprofloxacin compared with control. In infection group showed no significant difference in RBC and PCV compared with control negative group but decrease in HB that is May due to the *E coli: O157 H 7* causes little stress in poultry due to increase the load of bacterial count in intestine.

Leukocytes counts

The table (4) showed the Zeolite maintained a low level of WBC during infection the WBC count was significant decrease (P ≤0.05) in both groups of Control negative and Zeolite compared with G2 and G3 groups. As it is known white blood cells are directly involved in the body's immune responses. Their main function is to recognize foreign substances, phagocytosis and the recording of information about microorganisms so if increase WBC number that is indicate to tolerance of broiler chickens to chronic stress (Attia *et al*., 2017). results agree with (Oleforuh-Okoleh *et al*., 2014) who suggested that a decrease in white blood cell count is a result of an increase in immune status in poultry by using the natural feed additive ,(Golovacheva *et al*., 2020) reported that use Zeolite decrease in white blood cell count is a result of an increase in immune status in rabbit. The Zeolite action as selection and remove the pathogen by washing out, so the Zeolite may be help the immunity and didn’t need to increase the WBC count by decrees the tolerance of chickens. By using of Zeolite the lymphocytes in both experiments were significant increase ($P \leq 0.05$), birds having the greater density of lymphocytes have stronger immune status (Haque., 2010).

The current study noted that the Heutrophils: lymphocytes (H/L) ratio may significantly enhance in the treated group with 2% Zeolite , agree with (Skomorucha & Sosnówka-Czajka, 2021) suggested that increase in the percentage H/L ratio that mean this group under stressed environment, therefore, the H/L ratio is used as a reliable indicator of stress in birds. And agree with (Prisnyi *et al*., 2022) founded some increase in white blood cell count in infected group with gram negative bacteria and resaved Ciprofloxacin

Liver and kidney histomorphology

Liver as the biggest gland in the body has several roles. In addition to digestive and metabolic activities, Liver has immunological and detoxification activity (Latif, 2011) and (Al-Khalidi& Hamood, 2022). Liver in the body as a detoxifying organ absorbs a significant part of toxins produced from harmful microbes and external particles into the blood (Tavakoli *et al*., 2020).Also The excretory system in the birds consist of paired kidneys and the function of the kidneys are: maintain the balance of both electrolyte and water in the body , eliminate metabolic wastes and particularly nitrogen products of metabolism (except carbon dioxide), (Ghaj, 2021) and (Al-Hiyali, 2005).The health of the tissues of the liver and kidneys in poultry is a great indicator of the health of the animal, as well as the functioning of food additives such as Zeolite and its effect. The result of current study showed that the liver and kidney at group of infected bacteria have pathological change .The infection with pathogenic *E. coli* can cause liver and kidney damage (Mohammad & Al-Mahmood, 2022). (Azza *et al*., 2018) founded that *E. coli O157:H7* caused with severe leucocyte infiltration, severe degenerative changes of hepatocytes and congestion in poultry.

The liver performs several intricate processes, one of which is the detoxification of medicines and their metabolites, including antibiotics. While certain antibiotics can directly harm the liver, others can trigger adverse reactions. The result of current study showed that use of antibiotic at normal dose of ciprofloxacin for 5 days can cause of liver and kidney damage. The results agree with (Siddig *et al*., 2014) reported that the poultry received normal dose of ciprofloxacin for 7 days can cause of liver damage and the histopathology showed: Short Arrows indicate positions of hepatocellular degeneration and centrilobular hepatocellular. While long arrow indicates pressure atrophy and dilated congested central vein. Additionally, if the kidneys are already weak, some antibiotics can harm them further (Khalili *et al*., 2013). The current study founded the use of Zeolite able to maintain the health of the tissues of the liver and kidney of infected poultry with *E. coli O157:H7*. Zeolite has been shown to be an effective uremic toxin adsorbent. They are expected to exhibit selective adsorption with potentially large adsorption capacities, be non-toxic, stable, and have channel systems corresponding to the size of tiny uremic toxin molecules (Koubaissy *et al*., 2017). In current study founded the treatment with Zeolite can reduce the kidney and liver damage during the infection of *E. coli O157:H7* compared with the use of ciprofloxacin. And this is may be due to the *E. coli O157:H7* being resistant to the ciprofloxacin (Hameed *et al*., 2023).

Conclusion

We can conclude that Zeolite 2% in broiler feed can be used as feed additive in broiler to reduce the log count of Escherichia Coli O157:H7 in intestine and improve some hematological parameters (RBC, Hb, WBC and H/L) and biochemical parameters ALT and AST enzymes. And maintain the health statues of liver and kidney. Zeolite is not expensive and is a good option to reduce the pathogen causes in broiler farms. However, it needs more studies to validate the effect of zeolite, because there are few studies on the effects on broiler.

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