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Evaluation of Adverse Effects of Antibiotics on Broiler Chickens

Berghiche Amine[1,2*], Khenenou Tarek[2] , Bouzid Riad[3] , Rahem Soumia[2], Labied Ibtessem[2] and Boulebda Nadji[1,2]

[1] *Laboratory of Science and Technic of living Mohamed Cherrif Messaadia University, Souk Ahras, Algeria*
[2]*Agro veterinary institute, Taoura, Mohamed Cherrif Messaadia University, Souk Ahras, Algeria*
[3]*Chadli Ben Djedid University El Taref, Algeria*
[*]Corresponding author's Email: amine_berghiche@yahoo.com; ORCID: 0000-0001-6355-3591

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ABSTRACT

To evaluate the impact of uncontrolled use of veterinary drugs on broilers in eastern Algeria, an experimental plan was developed for the evaluation and identification of drug toxicity in 60 chickens (30 treated and 30 non-treated with antibiotics) using analysis of serum biochemical parameters, autopsy, morpho-metric and histopathological analysis of certain internal organs. The results of the serum biochemical analysis revealed that the uric acid and aspartate aminotransferase values in antibiotic-treated chickens were high, while the lesion status showed a dominance of respiratory lesions, followed by digestive lesions, particularly hepatic lesions. The morphometric study of the internal organs (liver, kidney, and intestine) demonstrated that abnormal liver appearance was very important with minor atrophic changes in the kidney, while the histopathological examination of the liver revealed the presence of deposition in the center of the hexagons in the apical area with an apparent homogeneous structure of fibrous connective tissue. Also, there were apparent deep sinus defects in peripheral areas with an overload of fibrin. The histopathological examination of the kidneys revealed proximal tubular atrophy in the renal parenchyma along with loss of distal intratubular consistency to the peripheral zone of homogeneous structure persuading the peripheral edema. It is concluded that the uncontrolled use of antibiotics in the poultry industry leading to a moderate to severe toxicity.

Key words: Adverse effects, Antibiotics, Broiler chicken, Self-medication

INTRODUCTION

According to the Swiss Toxicological Information Centre (Curti et al., 2009), drugs are responsible for more than 20% of animal toxicity cases, with varying degrees of severity. Drug toxicity in broilers, especially due to antibiotics, is an important issue that has severe economic consequences for the poultry industry and negative impacts on human and animal health. Throughout the world, antibiotics are used in the poultry industry for preventive and therapeutic purposes. The massive use of antibiotics in poultry farming is due to their growth promotion effects and the high prevalence of self-medication (Berghiche et al., 2018a). In Algeria, poultry farmers use high doses of antibiotics to fatten chickens and save money without consulting a veterinarian. In addition, poultry farmers do not respect the withdrawal period of antibiotics, thus the poultry products are consumed by humans before the drug residues are removed from the body of the birds (Berghiche et al., 2019). The objective of the present study was to assess the impacts and risks associated with the use of antibiotics in broiler chickens.

MATERIALS AND METHODS

Ethical approval

The experiment was carried out according to the National Regulations on Animal Welfare and Institutional Animal Ethical Committee.

Animals

The experimental study was conducted on 60 broiler chickens (Ross 308) aged 7 to 8-week-old. The chickens were divided into two groups (30 treated and 30 non-treated) raised on the floor, on straw bedding in non-air-conditioned greenhouse buildings.

Treatment plan

Both groups of animals received the treatment including three types of medication administered orally: antibiotic erythromycin (Vigal 2 X, Ceva production, Algeria), anticoccidial sulfaquinoxaline sodium

(Coccidiopon, Avico production, Jordan), and a vitamin and mineral supplement (Amin'Total, Laprovet, France). The chickens were treated according to the following plan:

Day 1 to Day 5: Vitamin supplement (1 gram per 2 liters of water) + Antibiotic (1 gram per 1 liter of water). Day 8 and 9: Vitamin supplement (1 gram per 2 liters of water). Day 10 to 12: Amin'total (1 gram per 1 liter of water). Day 15 and 16: Vitamin supplement (1 gram per 2 liters of water). Day 17 to 19: Antibiotic (1 gram per 1 liter of water). Day 22 to 23: Vitamin supplement (1 gram per 2 liters of water). Day 24 to 28: Antibiotic (1 gram per 1 liter of water)+ Anticoccidial (1 gram per 1 liter of water).

The chickens were fed on diets supplemented by 0.1% of antibiotic Vigal 2 X only for sick chicken and healthy chickens were fed on a basal diet without any supplementation.

Biochemical analysis

To biochemical analysis**,** 1 mL blood of 60 broilers was taken from the cutaneous ulnar vein using a syringe and disposable needle. The blood samples were collected in dry and heparinized tubes. The serum was separated and used for biochemical analysis. The biochemical parameters including creatinine, uric acid, alanine aminotransferase (ALAT), and aspartate aminotransferase (ASAT) were measured in a human medicine laboratory.

Morphometric, macroscopic, histological and histopathological studies

Sixty animals were euthanized and immediately dissected to maintain the integrity of the organs and tissues. The intestines carefully removed to avoid the risk of crushing and then placed in vials filled with fixative (10% formaldehyde) (Khenenou et al., 2019). The necropsy examination was carried out according to the classical technique (Khenenou et al., 2019; Berghiche et al., 2018c). The morphometric study consisted of measuring the fresh weight of the liver and kidney using a precision balance, also, the length and width of the liver and kidney using a caliper.

Statistical Analysis

The statistical analysis was performed using PAST software (Palaeontologia Electronica, Norway, Version 6.0, Free edition). The results were considered statistically significant at a significance level of $p<0.5$.

RESULTS AND DISCUSSION

Biochemical and biological modifications in broilers

Serum biochemical analysis revealed a significant increase ($p<0.01$) in uric acid and ASAT values in treated sick chickens compared to the healthy group (Table 1). Whereas the treated chickens did not show any significant change in serum creatinine and ALAT concentrations compared to the healthy group.

Macroscopic and morphometric aspects of tissues in different systems

Necropsy analysis

In the autopsy of 194 dead chickens collected from the study farm, the respiratory lesions represented a 30.15% incidence rate, followed by the digestive lesions (25.59%), particularly the hepatic lesions (14.95%). The locomotor and splenic lesions had a rate of 24.48% and 14.69%, respectively. The heart lesions were observed in nine cases (5.08 %) (Table 2).

Morphometric study

Our analysis showed very significant morpho-histological changes in the liver with minor changes in the kidney (Table 3).

Histopathological analysis

Microscopic changes of the liver

Microscopic examination of the broilers' liver revealed lesions in the peripheral parts and lobules due to the action of antibiotics. The presence of deposition in the center of the hexagonal surface in the apical zone, an apparent homogeneous structure of fibrous connective tissue, and apparent degeneration in the peripheral area with clear edema were observed (Figures 1 and 2).

Microscopic changes of the intestine

The congestion related to stasis was demonstrated in different parts of the intestine. lesions consisted of degeneration of enterocytes, the presence of significant edema, hydropic degeneration with functional repercussions (Figure 3).

Microscopic changes of kidney

Active congestion related to hyperemia was observed that is the indicator of chronic tubulointerstitial nephritis with the toxic origin. Microscopic examination of the renal cortex demonstrated hydropic degeneration of the tubular system resulting from drug intoxication caused by self-medication. The microscopic examination of proximal convoluted tubule showed areas of degeneration with necrosis points; caryolysis and picnosis (Figure 4).

Critical points for the use of broiler chicken as an experimental model in toxicology

Broiler chicken is not a suitable experimental model to determine drug toxicity by antibiotics because the harmful effects of the antibiotics are usually chronic, while the production cycle in broilers does not exceed two months. it is noted that there is no exact time for the appearance of an injury or adverse effect, according to Haber's rules the dose is important in the expression of effects, acute toxicity of a substance includes all the specific phenomena that occur shortly after administration of a toxic substance after a single dose, chronic toxicity is less normalized and usually involves several non-fatal doses at administration (Paris et al., 2006)

In some cases some modifications on biochemical parameters and slight changes on the microscopic aspect of the internal organs were found and here the degree of severity is classified as benign (biochemical modification) and moderate (modification of volume and weight) (Joint FAO and WHO Expert Committee on Food Additives, 2009; Berghiche et al., 2018b). These modifications can be explained by the poor preparation and conservation of treatments by unqualified persons and the lack of specific diagnostic methods in poultry farming in the study region (Done, 1964). In the frame of treatment, the antibiotic is generally used at a defined dose and for a limited time scale, generally has low toxicity in the treated animal and the overdose in poultry farming is accidental and rare (Gustafson and Bowen, 1997; Sarmah et al., 2006). Nevertheless, some antibiotics have a high toxicity that limits their use in many animal species. Any antibiotic therapy must be performed by the practitioner, to avoid the existence of direct effects in the case of organic toxicities that are specific to each antibiotic; there are also two types of adverse effects that are indirect, a disturbance of the digestive flora and development of resistance, which is due by the therapeutic failures (Berghiche et al., 2018d).

Table 1. Comparison of serum biochemical parameters of chickens fed on diets supplemented with 0.1% Vigal 2 X antibiotic (treatment) with those of group fed on a basal diet (control)

Parameter	Groups	
	control	treatment
Creatinine	0,41 ± 0,03	0,49 ± 0,03
Uric acid	2,45 ± 0,09	2,93 ± 0,11*
ALAT	19,33 ±0,29	21,17 ± 0,82
ASAT	78,15 ± 4,83	85,33 ±2,57*

*p<0.01

Table 2. The incidence of gross lesions in the different systems of autopsied chickens

Lesions in different systems		Number	Frequency (%)
Digestive	Hepatic	29	14,95
	Others	21	10,64
Respiratory		59	30,15
Cardiac		9	5,08
Locomotor		48	24,48
Lymphoid system (Splenic)		28	14,69

Table 3. Comparison between dimensions and weight of internal organs of chickens fed on diets supplemented with 0.1% Vigal 2 X antibiotic (treatment group) with those of chickens fed on a basal diet (control group).

Organs	Liver		Kidney	
Parameters	control	treatment	control	treatment
Weight (g)	59.80 ± 4.1	67.33 ± 0.5*	14.98 ± 2.33	12.45 ± 1.67*
Length (mm)	34 ± 11.05	50 ± 15.67**	16 ± 3.66	16 ± 5.05
Width (mm)	10 ± 0.87	15 ± 4.87*	13 ± 1.55	16 ± 3.33*
Height (mm)	72 ± 9.33	79 ± 6.33*	21 ± 2.87	24 ± 1.87*

Age of broiler: 7^{th}-8^{th} weeks. Data are expressed as mean ± standard deviation. (*p < 0.05; **p < 0.01).

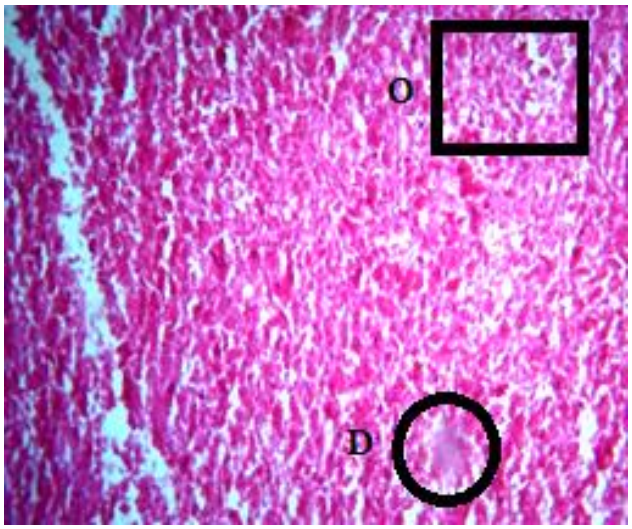

Figure 1. Histological appearance of liver of antibiotic-treated broiler chickens. Hepatic *degeneration* (D); edema (O)

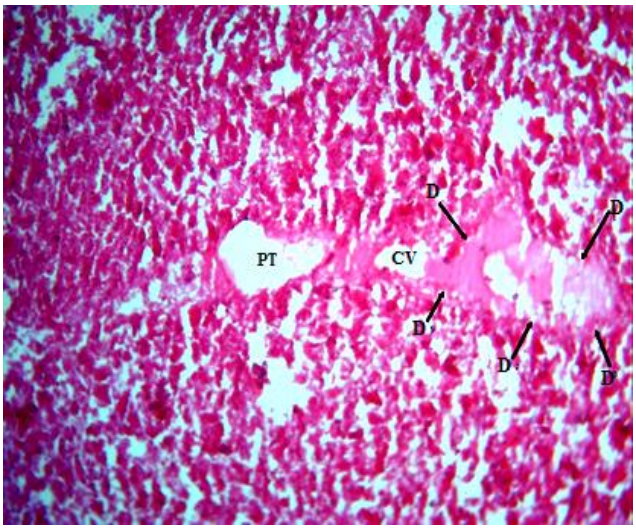

Figure 2. Microscopic lesion of the liver of broiler chickens fed on diets supplemented with 0.1% Vigal 2 X antibiotic (x100); CV: central vein; D: tissue degeneration; PT: portal triads

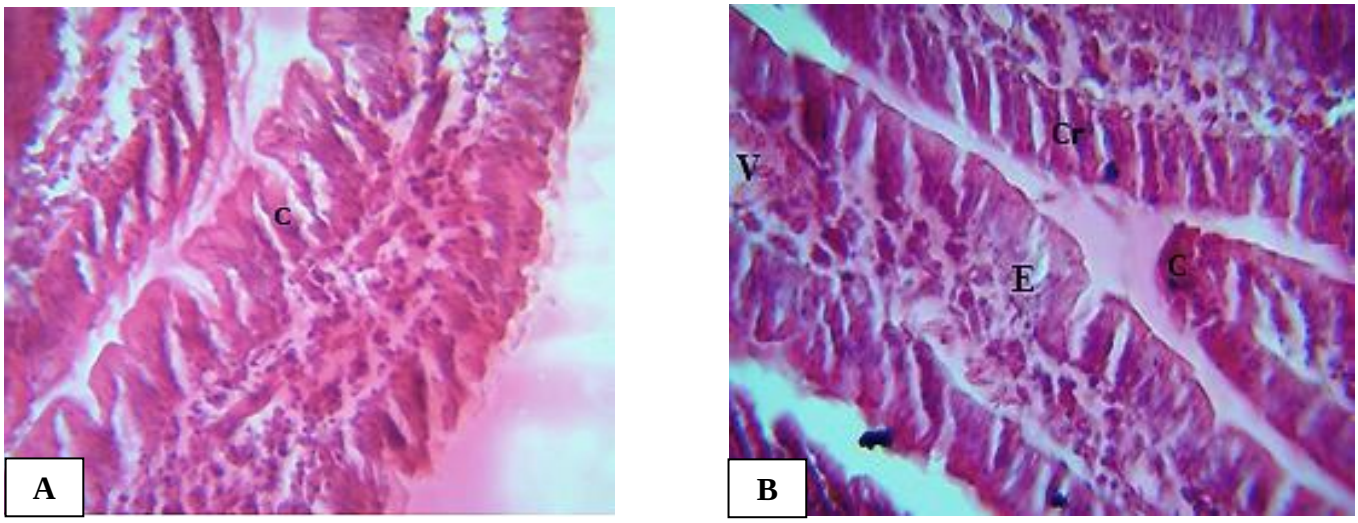

Figure 3. A): Microscopic aspect of the intestine of broiler chickens fed on a basal diet (normal appearance). **B):** Microscopic lesions of intestine of broiler chickens fed on diets supplemented with 0.1% Vigal 2 X antibiotic. (x100); V: Vessel; Cr: Crypt; C: Congestion; E: Edema

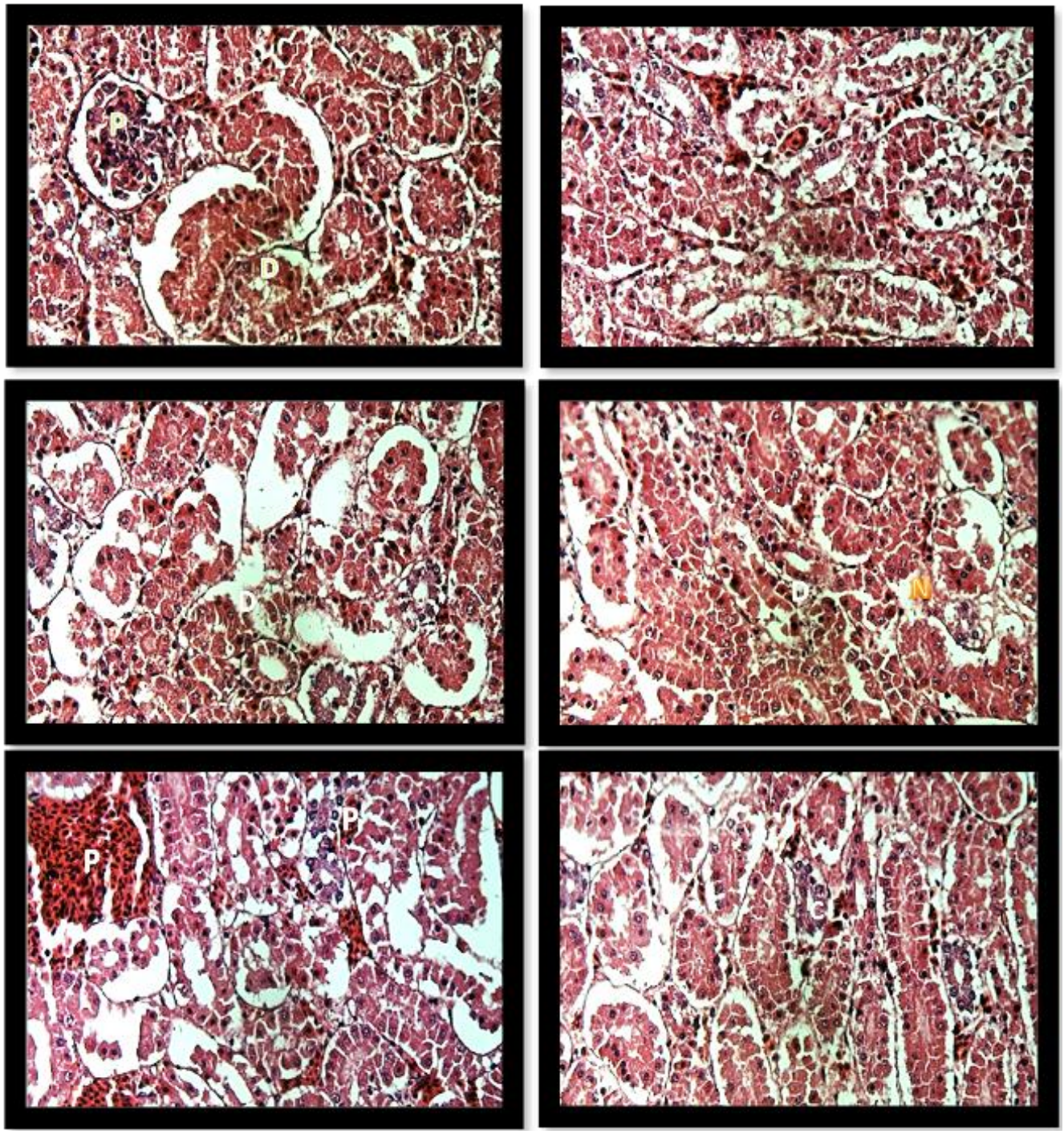

Figure 4. Microscopic lesions observed in the kidneys of broiler chickens fed on diets supplemented with 0.1% Vigal 2 X antibiotic. degeneration (D);necrosis points (N); caryolysis (C) and picnosis (P) (H&E. x100).

CONCLUSION

The results showed that antibiotic treatment in chickens had effects on serum biochemical parameters and demonstrated an abnormal appearance on the liver and minor atrophic changes in the kidney. Histopathological examination of the liver, kidney, and intestine revealed the presence of remarkable changes in their histological structures. The self-medication in poultry farming impacts the animal and human health and the attention of

veterinarians should be focused on the fight against self-medication in poultry farming, in particular antibiotics.

DECLARATIONS

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Competing interests

The authors have no competing interests to declare.

Authors' contributions

Berghiche created the idea and designed the study, performed the statistical analysis, and draft the manuscript. Rahem, Labied, and Berghiche collected data. Khenenou, Boulebda, Bouzid, and Berghiche read and approved the final manuscript.

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