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Effect of an Aqueous Extract of a Plant Used in Traditional Ivorian Veterinary Medicine on Experimentally Induced Colibacillosis in Broiler Chicks: Case of *Vitex doniana* (Sweet)

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Abstract **Original Research Article**

Avian colibacillosis is an infection that leads to high morbidity and mortality, resulting in considerable economic losses in poultry farming. This study aimed to evaluate the effect of an aqueous extract of *Vitex doniana* on experimentally induced colibacillosis in broiler chicks. Eighty (80) one-day-old chicks were divided into 4 groups (n = 20/group) and fed identically for 28 days. Chicks in group 1 received only conventional feed. Chicks in groups 2, 3, and 4 received a single 0.3 ml dose of E. coli culture (1 x 10^9 CFU/ml) orally. After infection, chicks in group 3 received aqueous extract of *V. doniana* (5 mg/kg bw) orally for 14 days, while those in group 4 received leofloxacin (the reference antibiotic) at a dose of 1 mg/10 kg bw orally for 7 days. During the experiment, the behavior, clinical signs, and mortality of the chicks in each group were recorded. On days D0, D7, D14, and D28 of the experiment, blood samples were collected from chicks for hematological and biochemical analysis. The results showed that the aqueous extract of *V. doniana* resulted in an improvement in clinical signs in infected chicks and a reduction in their mortality compared to group 2. Furthermore, the extract of this plant had a more beneficial effect on the hematological and biochemical parameters evaluated compared to leofloxacin. This plant could be used as a veterinary product to control avian colibacillosis.
Keywords : *Vitex doniana,* aqueous extract, *E. coli,* experimentally colibacillosis, chicks.

INTRODUCTION

Poultry production is one of the main sources of animal protein in many developing countries (Kamagaté *et al*., 2017). In Côte d'Ivoire, this production represents a strategic sector for food security and the rural economy (I.P.R.A.V.I., 2015). However, the performance of this sector is frequently compromised by the impact of several infectious diseases. Among these pathologies, avian colibacillosis (Kamagaté *et al*., 2017), caused by Escherichia coli, is paramount. This common disease in broiler chickens leads to high morbidity and mortality, resulting in considerable economic losses (Mellata, 2013; Ewers *et al*., 2022).

To effectively combat colibacillosis, farmers frequently resort to synthetic antibiotics. Faced with the limitations of antibiotic use in treating colibacillosis, due to the emergence of microbial resistance and its repercussions on public health, interest in therapeutic alternatives derived from traditional medicine continues to grow (Cabi, 2007). Indeed, several medicinal plants have been cited in the literature as having antimicrobial and immunomodulatory properties capable of restoring the physiological balance of infected animals. Among them is *Vitex doniana* (Sweet). It is a medicinal plant widely distributed in West Africa (Forcados *et al*., 2021). It is used in traditional medicine for its therapeutic properties. Indeed, traditionally, the leaves of this plant are used to treat diabetes and gastroenteritis (Owolabi *et al*., 2022; Odugbemi, 2008). Decoctions of its roots are used to relieve fevers, diarrhea, and respiratory illnesses (Ajiboye, 2015). The trunk bark, on the other hand, is used against skin infections as well as several other infections (Burkill, 1984). Several scientific studies have reported the antimicrobial and antioxidant activities of this plant (Akinmoladun *et al*., 2021; Mahmoud *et al*., 2024). The work of Abdulrahman *et al*., (2025) demonstrated numerous biological activities of this plant, also describing its richness in various chemical molecules. This study was undertaken with the aim of evaluating the effect of aqueous extract of *Vitex doniana* on experimentally induced colibacillosis in broiler chicks.

MATERIALS AND METHODS

Study plant

The leaves of *Vitex doniana* were collected in Lataha, in the northern region of Côte d'Ivoire, in June 2025. The plant was identified by botanists from Jean Lorougnon Guédé University in Daloa. A reference specimen was also deposited at the same institution.

Study animals

Eighty (80) healthy, one-day-old Cobbs's broiler chicks were obtained from the Ivorian Compound Food Manufacturing Company (SOFACI), a local company certified and recognized by the Ivorian government. Throughout the study period, these chicks were fed identically with the classic broiler chick rearing feed supplied by the same company (SOFACI), which is also recognized in the manufacture of poultry feed.

E. coli strain

A pure culture of a pathogenic strain of avian *E. coli* was obtained from the microbiology and molecular biology laboratory of the Pasteur Institute of Côte d'Ivoire. It was preserved on MacConkey agar and Eosin Methylene Blue agar throughout the handling period. This *E. coli* culture was intended to infect chicks orally during the experimental trial.

Preparation of the aqueous extract

The leaves of *Vitex doniana* were pre-washed with distilled water and dried in the laboratory for four weeks. They were then on the ground using an IKAMAG type grinder. The aqueous extract of *Vitex doniana* was obtained by macerating 100 g of sample pulp in 1 L of distilled water for 24 h. The homogenate obtained was successively filtered twice through hydrophilic cotton and once through Whatman n°2 paper. The filtrate obtained was concentrated in a Med Center Venticell oven at 50 °C until a dry extract was obtained. The dry extract was then stored in a refrigerator for further study.

Preparation of *E. coli* inoculum

To prepare the bacterial inoculum, the standard *E. coli* culture was subcultured onto a nutrient broth and incubated at 37°C for 24 hours. After this incubation, the number of bacteria was adjusted to 1 x 10^9 CFU/ml by referring to the turbidity with the McFarland tube.

Experimental plan

After a week of acclimatization in the laboratory, the chicks were weighed individually and randomly divided into four equal groups (n= 20/group) on the eighth day, named D0. All chicks in the four groups received the provided feed and had free access to water for the entire duration of the experiment (28 days). The chicks in group 1 received only the provided feed (control group) for 28 days. On Day 0, unlike group 1, the chicks in groups 2, 3 and 4 each received a single dose of 0.3 ml of *E. coli* culture (1 x 10^9 CFU/ml) orally for 28 days. After being infected, the chicks in group 3 received aqueous extract of *V. doniana* (5 mg/kg bw) orally for 14 days. In contrast, the chicks in group 4, after contamination, received Leofloxacin (reference antibiotic) at a dose of 1 mg/10 kg body weight orally for 7 days. Throughout the experiment, the behaviors, clinical signs and mortality of the chicks in each group were noted. Furthermore, on Day D0; D7; D14 and D28 of the experiment, blood samples were taken from the wing vein of the chicks and packaged in different tubes for hematological and biochemical examinations using automatic analyzers and biochemical analysis kits according to the manufacturers' recommendations.

RESULTS AND DISCUSSION

Clinical signs in different groups of chicks

During the experiment, the chicks in group 1 (uncontaminated and untreated control) showed no abnormal clinical signs. They remained healthy throughout the experimental trial. However, those in group 2 (chicks infected with the *E. coli* strain and untreated) showed clinical signs 24 hours after infection. For the chicks in this group 2, various clinical signs were observed, including depression, lethargy, shortness of breath, and respiratory distress. As the days passed, these chicks grew physically weak due to their loss of appetite. They gradually developed whitish, pasty, watery, and mottled diarrhea, which turned reddish in later stages. They huddled together in a corner with ruffled feathers. As the weeks went by, these clinical signs intensified until the end of the second week. In group 3 (chicks infected with *E. coli* and treated with aqueous extract of *V. doniana*), the clinical signs observed were generalized stress from the first days, lethargy, occasional shortness of breath with mild diarrhea which appeared at the end of the first week. After the first week, the chicks' droppings began to become less solid, and the chicks began to become lively again until Day D14. From D14 onwards, the chicks in group 3 were more active and had solid feces. The clinical signs observed were like those of normal group 1.

The chicks in group 4 (chicks infected with *E. coli* and treated with the reference antibiotic) showed signs like those in group 3. However, the birds in group 3 were livelier and more active than those in group 4. The various observations indicate a successful *E. coli* infection in chicks, resulting in behavioral changes and the appearance of various clinical signs. These observations also demonstrate the effects of the aqueous extract of *Vitex doniana* and the reference antibiotic (leoflaxacin) against the avian *E. coli* strain used to infect the chicks. Similar studies and results have already been reported by several other authors with other plant species (Patil *et al*., 2018).

Mortality in different groups of chicks

No mortality was observed in the chicks of group 1 (control) throughout the experiment. Furthermore, they were healthy and lively throughout the study period. However, in group 2 (chicks infected with *E. coli* and not treated), 7 out of 20 chicks died (32%) during the experimental study period. Indeed, two (02) chicks died after infection with *E. coli* between days D0-D7. Three (03) chicks also died between D7- D14 followed by two (02) other chicks between days D14 and D28. In group 3 (chicks infected with *E. coli* and treated with *V. doniana*) and group 4 (chicks infected with *E. coli* and treated with the reference antibiotic), two and three chicks died respectively out of a total of 20 chicks. Mortality in birds of group 3 was observed during the first week (01 chick) and the second week (01 chick) giving a mortality rate of 10%. In the case of the birds in group 4, mortality was observed during the first week (1 chick), the second week (1 chick) and between the third and fourth week (1 chick) giving for this group a mortality rate of 15%. These various observations demonstrate that the aqueous extract of *Vitex doniana* leaves has an inhibitory effect on the *E. coli* strain used to infect infected chicks while reducing the mortality rate.

Analysis of hematological parameters by group

Figure 1: shows the concentrations of hematological parameters evaluated during experimentation in the different groups of chicks.

In the control group 1 (uncontaminated and untreated), the mean hemoglobin concentration values in the chicks remained almost stable, varying from 9.88 ± 0.3 g/dL to 10.25 ± 0.29 g/dL. This result undoubtedly shows a normal hematological status, without infectious disturbance in the chicks. However, in group 2 of the contaminated and untreated chicks, after infection, a sharp decrease in hemoglobin concentration was observed, falling from 9.73 ± 0.35 g/dL (D0) to 7.58 ± 0.39 g/dL (D7). The latter indicates a successful infection. Indeed, this decrease in hemoglobin would indicate anemia linked to colibacillosis. Several studies have reported these same observations (Gross, 1994; Panth, 2019; Umar *et al*., 2017). From day 7, this concentration began to increase in group 2 until reaching 7.85 ± 0.39 g/dL on day 14 and 9.81 ± 0.1 on day 28, which would be due to a resumption of immunoprotective activity. The hemoglobin levels in group 3 gradually increased. These values remained higher than the other groups, with levels of 10.19 ± 0.42 g/dL (Day 7) and 10.33 ± 0.17 g/dL (Day 28). This suggests an immunoprotective effect of *Vitex doniana*, probably via its antioxidant and anti-inflammatory actions (Abdulrahman *et al*, 2025) and especially its prevention in the destruction of lymphocytes, and the stimulation of the adaptive response (Akinmoladun *et al*., 2019; Olorunnisola *et al*., 2011). Unlike group 3, after infection, the hemoglobin levels of the chicks treated with leofloxacin remained almost constant until day 7 (9.95 ± 0.1 g/dL) and fell from day 7 to day 14. From day 14, these levels increased and reached an Hb level of 9.78 ± 0.24 g/dL on day 28. This result probably indicates partial protection against immunosuppression, with an intermediate efficacy between group 2 and group 3. After infection, lymphocyte levels decreased very significantly in infected and untreated chicks (group 2) until day 7 (48.15 ± 0.09%). This phenomenon reflects not only a successful infection but, more importantly, post-infection immunosuppression, which may be linked to lymphocyte lysis by the infectious agent or to a systemic inflammatory response (Umar *et al*., 2017; Gross, 1994). From day 7 onward, an increase in these levels was observed until day 14 (53.47 ± 0.12%) and day 28 (61.42 ± 0.26%), likely to indicate spontaneous immune recovery. However, these rates remain significantly lower than those of other groups of chicks. Furthermore, in these other groups of chicks (groups 1 ;3; 4), this same evolution of lymphocyte levels was observed. However, group 4 (chicks infected and treated with the reference antibiotic) had the lowest lymphocyte counts. This was followed by group 3 (chicks infected and treated with the aqueous extract of the plant) and group 1 (a group of normal control chicks). Furthermore, lymphocyte counts in groups 1 and 3 were like those in groups 2 and 4. PCV levels decreased very significantly in group 2 chicks from day D0 (27.11 ± 1.28%) to day D7 (24.06 ± 0.33). This drop would reflect marked anemia probably associated with the destruction of red blood cells by infection and systemic inflammation (Gross, 1994; Umar *et al*., 2017). These levels remained almost stable until day 14, after which they began to increase again until day D28 (29.82 ± 0.35%). This improvement on day D28 can be explained by a gradual return of normal immunity. However, in this group 2, PCV levels remained significantly lower than in the other chick groups. In the other chick groups (groups 1, 3, and 4), an increase in PCV levels was observed up to day D28, with maximum levels of 31.24 ± 0.12% and 30.02 ± 0.13% recorded by groups 1 and 3, respectively, on day D14. Similar results were reported by Gross (1994) and Umar *et al*. (2017). The different eosinophil levels generally evolved in the same way in the chick groups. However, a non-significant difference between these levels was observed on Day D14, even though group 3 (contaminated chicks treated with the aqueous extract) showed the lowest values during the experiment. According to Chauhan *et al.* (2020), eosinophils are involved in the allergic and parasitic response, which would indicate good physiological stability. The results obtained are like those of Maxwell and Robertson (1998). The observed fluctuations could be explained by unregulated, fluctuating immune responses sometimes seen during incomplete treatments. As for groups 3 and 4, they underwent practically the same type of evolution of total protein levels until day D14 where they recorded respective protein levels of 4.28 ± 0.09 g/dL and 4.24 ± 0.04 g/dL. However, the total protein levels of group 3 were closer to those of the control group until day D14. This latter result indicates a protective effect of the aqueous extract linked to its antioxidant and hepatoprotective properties reported by numerous studies in the literature (Omoregie and Osagie, 2012). However, the total protein levels of group 3 were closer to those of the control group until day D14. This latter result indicates a protective effect of the aqueous extract linked to its antioxidant and hepatoprotective properties reported by numerous studies in the literature (Omoregie and Osagie, 2012). These high elevations would indicate renal damage induced by the infection. On day 14, these values decreased very significantly, with these groups reaching concentrations of 0.88 ± 0.05 mg/dL (group 4) and 0.73 ± 0.09 mg/dL (group 2). These results can be explained by partial renal recovery. Groups 1 and 3 showed very low variation in concentration in chicks with respective values of 0.78 ± 0.01 mg/dl and 0.81 ± 0.03 mg/dl on day D28. This suggested that group 1 exhibited normal renal function without pathological stress. As for group 3, the results obtained were linked to a protective role of *Vitex doniana*, whose phenolic compounds limit oxidative stress and lipid peroxidation in the kidneys (Olorunnisola *et al*., 2011). Furthermore, the nephroprotective effect of *Vitex doniana* extract could also be associated with its antioxidant properties which limit oxidative stress and renal inflammation (Akinmoladun *et al*., 2019; Olorunnisola *et al*., 2011). AST values remained almost constant from day 0 to day D7 in groups 1, 3, and 4 (108.3 ± 0.3 IU/L - 109.2 ± 0.98 IU/L). These results indicate a normal profile, without major liver damage. On Day D14, AST values fell significantly for group 3 (103.65 ± 0.48 IU/l) and group 4 (103.73 ± 0.28 IU/l) while they increased in the chicks of group 1 (113.34 ± 0.91 IU/l). These results would justify the hepatoprotective roles of the different treatments. Group 2 (contaminated and untreated chicks) showed the highest AST values on days D7 and D14 before reaching a low value (115.15 ± 3.02 IU/l) like that of the other groups on day D28. These observations suggest acute liver damage likely related to *E. coli* infection. Following significant stress, the progressive decrease in AST levels appears to be linked to spontaneous physiological recovery. Similar studies have been obtained by several other researchers (Suvarna *et al*., 2017). ALT activities in chicks from groups 1, 3 and 4 varied between 11.95 ± 0.2 IU/l (D1) and 12.12 ± 0.31 IU/l (D28) thus showing a slight variation (± 0.17 IU/l). However, in chicks in group 2 (contaminated and untreated chicks), ALT activity increased significantly to reach a value of 17.93 ± 0.19 IU/l on day D7. From day D7 onwards, this activity gradually decreased to reach values of 18.83 ± 0.34 IU/l and 11.81 ± 0.17 IU/l respectively on days D14 and D28. The results obtained with the ALT values suggest that contamination of the chicks with the E. coli strain caused liver dysfunction. Furthermore, the aqueous extract studied, as well as the reference antibiotic, attempted to stabilize the resulting disruption in liver function.

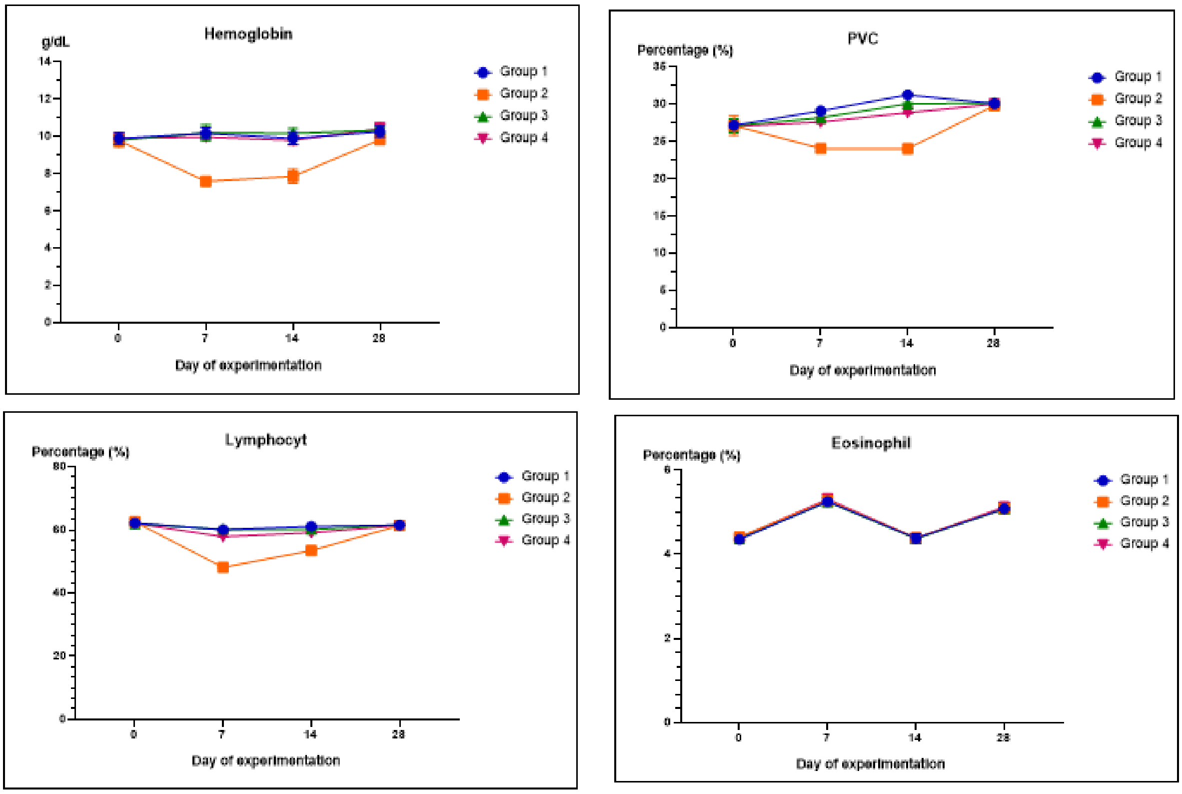
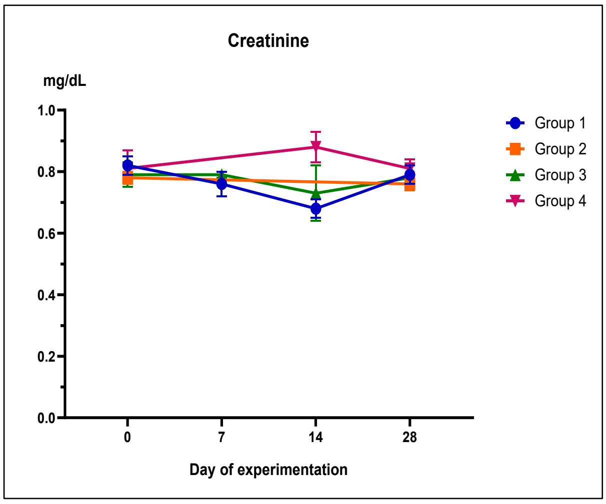
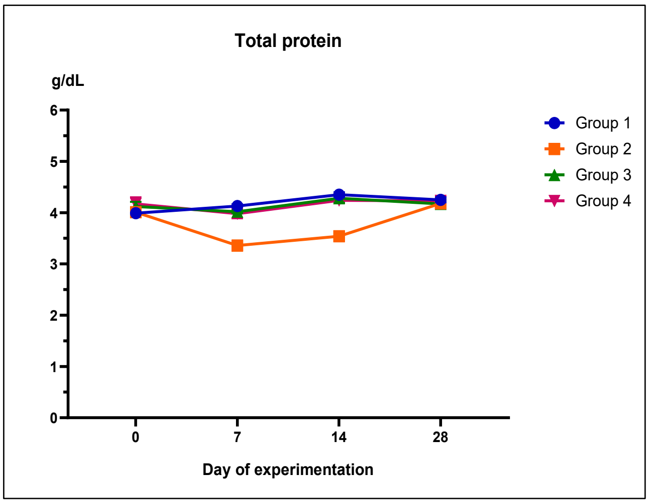
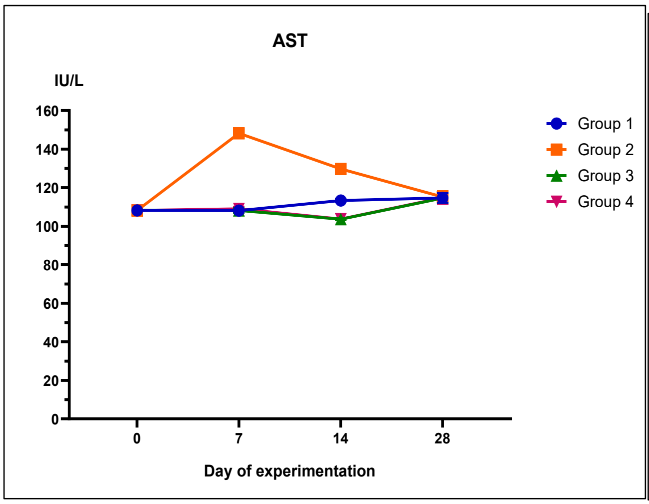


Figure 1: Hematological parameters evaluated during the experiment in the different groups of chicks.

Figure 2, meanwhile, shows the concentrations of the biochemical parameters evaluated during experimentation in the different groups of chicks. Total protein levels in group 2 fell very significantly until day 7 (3.36 ± 0.06 g/dL). This hypoproteinemia reflects liver damage and/or a mystical inflammatory response that would increase protein catabolism and losses. These observations were made by Coles (1986). From day D7, these values increased until reaching levels of 3.54 ± 0.03 g/dL and 4.18 ± 0.07 g/dL respectively on day D14 and day D28. These results show a rise and partial recovery of immunity. In control group 1, these levels increased progressively until day D14 before undergoing a slight decrease. As for groups 3 and 4, they underwent practically the same type of evolution of total protein levels until day D14 where they recorded respective protein levels of 4.28 ± 0.09 g/dL and 4.24 ± 0.04 g/dL. However, the total protein levels of group 3 were closer to those of the control group until day D14. This latter result indicates a protective effect of the aqueous extract linked to its antioxidant and hepatoprotective properties reported by numerous studies in the literature (Omoregie and Osagie, 2012). On day 7, high creatinine concentrations (1.82 ± 0.05 mg/dL) were obtained in group 4 (contaminated chicks treated with the reference antibiotic). On the same day, group 4 was followed by group 2 (contaminated but untreated chicks), where an increasing trend was observed (1.68 ± 0.03). These significant increases suggest renal damage induced by the infection. On day D14, these values decreased very significantly, with these groups reaching concentrations of 0.88 ± 0.05 mg/dL (group 4) and 0.73 ± 0.09 mg/dL (group 2). These results can be explained by partial renal recovery. Groups 1 and 3 showed very low variation in concentration in chicks with respective values of 0.78 ± 0.01 mg/dl and 0.81 ± 0.03 mg/dl on day D28. This suggested in group 1 the existence of normal renal function without pathological stress. Regarding group 3, the results obtained appear to be linked to a protective role of *Vitex doniana*, whose phenolic compounds limit oxidative stress and lipid peroxidation in the kidneys (Olorunnisola *et al*., 2011). Furthermore, the nephroprotective effect of *Vitex doniana* extract could also be associated with its antioxidant properties, which limit oxidative stress and renal inflammation (Akinmoladun *et al*., 2019; Olorunnisola *et al*., 2011). AST values remained almost constant from day D0 to day D7 in groups 1, 3, and 4 (108.3 ± 0.3 IU/L - 109.2 ± 0.98 IU/L). These results indicate a normal profile, without major liver damage. On Day D14, AST values decreased significantly in group 3 (103.65 ± 0.48 IU/L) and group 4 (103.73 ± 0.28 IU/L), while they increased in chicks in group 1 (113.34 ± 0.91 IU/L). These results support the hepatoprotective effects of the different treatments. Group 2 (contaminated and untreated chicks) showed the highest AST values on days D7 and D14 before reaching a low value (115.15 ± 3.02 IU/l) like that of the other groups on day D28. These observations reflect acute liver damage which would be linked to *E. coli* infection. Following significant stress, the progressive decrease in AST values appears to be linked to spontaneous physiological recovery. Similar studies have been conducted by several other researchers (Suvarna *et al*., 2017). ALT activities in chicks from groups 1, 3 and 4 varied between 11.95 ± 0.2 IU/l (D1) and 12.12 ± 0.31 IU/l (D28) thus showing a slight variation (± 0.17 IU/l). However, in chicks in group 2 (contaminated and untreated chicks), ALT activity increased significantly to reach a value of 17.93 ± 0.19 IU/l on day D7. From day D7 onwards, this activity gradually decreased to reach values of 18.83 ± 0.34 IU/l and 11.81 ± 0.17 IU/l respectively on days D14 and D28. The results obtained with the ALT values suggest that contamination of the chicks with the *E. coli* strain caused liver dysfunction. Furthermore, the aqueous extract studied, as well as the reference antibiotic, attempted to stabilize the resulting disruption in liver function.







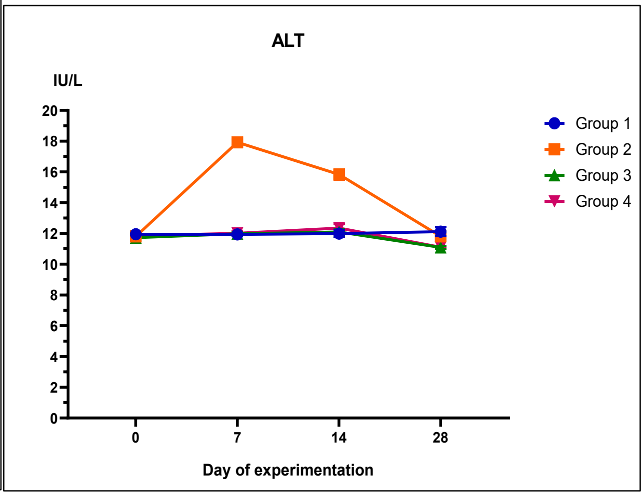


Figure 2: Biochemical parameters evaluated during the experiment in the different groups of chicks.

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CONCLUSION

The results of this study showed that the aqueous extract of *Vitex doniana* best improves clinical signs after infection of chicks by colibacillosis. The aqueous extract studied also had a beneficial effect on most of the hematological and biochemical parameters evaluated. Therefore, this plant could be used as a veterinary product to control avian colibacillosis. However, thorough toxicity studies and the search for possible lesions to vital organs in chicks will need to be considered before formulating an improved traditional medicine usable in poultry farming.

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