Effects of Hexane Extract of Garlic on Hematological, Biochemical and Histological Parameters in F1 Crossbred Chicks Non-Infected and Infected with *Salmonella typhimurium*

Tchoupou-Tchoupou EC¹*, Ndofor-Foleng HM¹, Nwenya JM¹, Okenyi N.J¹, Ikeh Nnana E¹, Ngwu NR¹, Nwakpu PE², Ossei J³ and Onyimoniy EA¹

¹University of Nigeria, Nsukka, Faculty of Agriculture, Department of Animal Science
²University of Abakaliki, Nigeria; Department of Animal Science
³University of Lome-Togo, Department of Food Science and Technology-Agro-food Industries

*Corresponding author: tchoupouedith@gmail.com; edith.tchoupou.pg03689@unn.edu.ng

Article History: 21-429 Received: 26-Oct-21 Revised: 15-Nov-21 Accepted: 18-Nov-21

**ABSTRACT**

An experiment was undertaken to evaluate the effects of garlic extract on hematological, biochemical, and histopathological profiles in F1 crossbred chicks (White Leghorn c× Nigerian Heavy Ecotype chickens) non-infected and infected with *Salmonella typhimurium*. This experiment was carried out to contribute to the genetic improvement of local Nigerian chickens while controlling salmonellosis and combating antibiotic resistance. Two hundred and forty (240) chicks at two weeks old were randomly divided into six treatments: T1 (No Salmonella and no garlic), T2 (garlic only), T3 (Ciprofloxacin only), T4 (Salmonella + Ciprofloxacin), T5 (Salmonella + garlic) and T6 (Salmonella without garlic or ciprofloxacin) and reared for six days. On day seven, birds were sacrificed, and blood samples were collected for hematometry and biochemistry analysis, while liver and small intestines were also collected for histopathology. Data obtained were subjected to analysis of variance (ANOVA), and were means were found to be significant. Duncan’s New Multiple Range Test was used to separate the means at a 5% probability level. The results revealed that white blood cell counts, neutrophils percentage, liver enzymes, total prot, and histological damage were also observed. However, feeding birds with garlic counteracted the effect of ST on those parameters. It can be concluded that hexane extract of garlic can alleviate the harmful effect of ST in birds’ systems and, thus, used in Poultry Farms against antibiotic resistance.

**Key words:** *Allium sativum*, Birds, Blood parameters, Histopathology and *S. typhimurium*.

**INTRODUCTION**

Animal Breeders in Nigeria have over the past years researched on improving the performance and disease resistance of the indigenous native chickens. The target is to make available safe and cheap animal protein to a greater proportion of the citizens, thus achieving the Sustainable Development Goals (SDGs) 1 and 2 (Morton et al. 2017). The Nigeria native chickens are preferred by the population for their good source of protein, the flavour of their meat and eggs compared to exotic chickens. They are also characterised by their capacities to hatch their eggs, hardy and resistant to diseases (Iyasere et al. 2019). However, the productivity rate of these local chickens remains very low due to inadequate nutrition, poor disease management and unimproved genetic potential in terms of their body weight (Munisi et al. 2015). To increase the productivity (body weight and eggs laying rate) of the Nigeria indigenous heavy ecotype chickens, they can be crossed with an improved breed of chicken such as the White Leghorn which are known for their higher egg and meat production. This productivity can only be maximized if poultry related diseases such as salmonellosis are also controlled.

*Salmonella* is a gram-negative bacterium that colonizes the gastrointestinal tract of chicken (Lee et al. 2020). The Centres for Disease Control and Prevention has reported that, *Salmonella* is one of the bacteria that causes foodborne disease (Froebel et al. 2020). Several *Salmonella serovars* have been identified by scientists and, among them *Salmonella enterica* serovar *typhimurium*.

Birds can be infected with *Salmonella enterica* serovar typhimurium (*S. typhimurium*) after ingesting contaminated feed or water. Salmonellosis symptoms in poultry farms include diarrhea, ruffled feathers, wings drooping, reluctant to move and egg laying disorder (Bandyopadhyay 2017). It has been reported that, *S. typhimurium* is a zoonosis transmitted to humans through contaminated chicken eggs and meat. It causes considerable mortality and morbidity in animals and humans (EFSA BIOHAZ et al. 2019). Antibiotics such as ciprofloxacin have been used during the past century to fight against this bacterium. However, the misuse of these products has resulted in the emergence of a resistant microbial population (Aldeyab et al. 2020). For this reason, it is essential to control salmonellosis by increasing the natural resistance of chickens, thereby reducing the socioeconomic damages worldwide in the one hand and fight against antibiotic resistance on the other hand. Several alternatives, such as probiotics, prebiotics, phytobiotics and feed additive have been used worldwide to combat the spread of salmonellosis infection (Rebollada-Merino et al. 2020; Sugiharto et al. 2020).

Garlic (*Allium sativum* L.) plant of the Alliaceae family has long been used as a seasoning in kitchen, as well as an ethno-medical plant to prevent and treat infectious diseases. The medical characteristics of garlic depends on many compounds including diallyl thiosulfonate (allicin), diallyl sulfide, S-allyl-cysteine sulfoxide (alliin), amino acids and vitamins (Kodera et al. 2017; Mansingh et al. 2018; Shang et al. 2019). Used in humans, animals and plants, garlic and its formulations have various biological properties such as antiviral, antimicrobial, antibacterial, anti-inflammatory and antioxidant (Rouf et al. 2020; Chitra 2020). Due to such properties, garlic can therefore be used in poultry farms as a potential alternative to antibiotics in order to improve bird health and production performance.

The current work focused on evaluating the effects of hexane extract of garlic on haematological, biochemical and histopathological profile in F₁ crossbred chicks (White Leghorn cocks × Nigerian Heavy Ecotype hens) non-infected and infected with *S. typhimurium*.

**MATERIALS AND METHODS**

**Location of the Study Area**

This research work was carried out in the Poultry Unit of the Department of Animal Science Teaching and Research Farm (DASTRF), University of Nigeria, Nsukka (UNN). The farm is situated within the equatorial rainforest belt of the tropics and falls specifically within the derived savannah vegetation zone. Nsukka is positioned on latitude 5°22' North, longitude of 07°24' East and altitude of 447°25m above sea level; with two main seasons, dry season (November to March) and rainy season (April to October) (Ndofor-Foleng et al. 2015). The study was carried out after having consulted the institutional guideline on the use of animals for scientific research and obtaining ethical clearance.

**Experimental Preparation of *Allium sativum* Hexane Extract**

Fresh pinkish garlic (*Allium sativum*) was purchased from Orie Orba Market, Nigeria and the hexane extract of garlic prepared according to Tchoupou-Tchoupou et al. (2021) was used.

**Experimental Animals/Breeding Protocol**

The base population for the exotic chickens (White Leghorn) were acquired from the local market (Orie Orba Market, Nigeria) and quarantined before being introduced into the experimental unit. The population of the local birds (female) were the heavy ecotype chickens which had undergone 7th generation of index selection at the DASTRF, UNN.

The White Leghorn males were crossed with the heavy ecotype female using Artificial Insemination. Eggs were collected and hatched to have a population of 240 F₁ chicks. On hatching, the chicks were weighed and distributed according to treatments (Fig. 1): T1 (Negative Control: No Salmonella and no extract), T2 (Extract only), T3 (Ciprofloxacin only), T4 (Salmonella + Ciprofloxacin), T5 (Salmonella + Extract) and T6 (Positive Control: Salmonella without extract or ciprofloxacin). Each treatment had 40 birds, replicated 4 times with 10 birds per replicate. The birds were raised on conventional deep litter. Pens and all routine management practices were carried out.

A standard strain of *S. typhimurium* was received from the National Veterinary Research Institute in Vom Jos. The Chicks received a challenge of 10⁸ organisms/ml NaCl solution of *S. typhimurium* by oral gavage (0.5mL) at two weeks of age and the faecal swab samples were used to determine the presence or absence of salmonellosa infection. The samples were cultured in tetrazionate broth (TBB) then, incubated for 18h at 37°C. The growth of TTB broth was streaked on Brilliant Green Agar (BGA) and MacConkey agar. These culture plates were incubated for 48h at 37°C. The plates were inspected for typical colonies of Salmonella. On MacConkey agar, the colour of the colonies is pale why on BGA the colour is pink (Dar et al. 2018).

The garlic hexane extract (800mg/kg) was diluted in drinking water and administered within a period of 5 days after infection to T2 and T5 while ciprofloxacin (antibiotic) used as a standard product was administered to T3 and T4. Throughout the experiment, birds were equally observed daily for clinical signs and mortality.

**Hematological and Biochemical Parameters**

After five days of treatment, blood samples were collected from three birds per replicate to determine the hematological indices such as white blood cell counts (WBC), packed cell volume (PCV), red blood cell counts (RBC), hemoglobin (HB), neutrophile, lymphocyte, monocyte, basophile and eosinophile. The blood was collected in sample bottles containing ethylene diamine tetra acetic acid as anticoagulant. Blood samples were also collected for serum biochemistry analysis such as alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) and total protein (TP).

**Histopathology**

At the end of the experiment, liver and small intestine (ileum) were collected and fixed in 10% neutral buffered formalin for 48 hours before being dehydrated in 4 grades (70, 80, 90 and 100%) of ethyl alcohol. The tissues samples were then prepared according to the standard procedure described by Luna (1968). The slides were mounted in
coverslips with distyrene, plasticizer and xylene, and examined with a Motic™ compound light microscope using x4, x10 and x40 objective lenses. The photomicrographs were taken using a Motic™ 5.0 megapixels microscope camera at x160 and x200 magnification.

Statistical Analysis
Data generated on the haematological and biochemical parameters were subjected to analysis of variance (ANOVA) using IBM statistic version 20 (IBM Corp, Armonk, NY, USA). Significantly different means were compared by using Duncan’s New Multiple Range Test procedure accepted at 5% probability test.

RESULTS
The result of the effects of feeding garlic hexane extract to uninfected and infected birds with S. typhimurium on haematological parameters is presented in Table 1. White blood cells (WBC), PCV, lymphocytes, RBC and HB were highly significant affected by treatment with the p-value of (P<0.0001). Moreover, neutrophil and lymphocyte were also affected by treatments, while monocyte, basophile and eosinophile were not.

The average values of WBC on T4 (13.83×10³/L), T5 (13.32×10³/L) and T6 (13.60×10³/L) were significantly (P<0.05) higher than the values of 8.98×10³/L, 8.93×10³/L and 8.95×10³/L recorded for birds in the control groups T1, T2 and T3 respectively. However, the average values of RBC and HB counts were significantly decrease (P<0.05) in the birds infested with S. typhimurium on T4, T5 and T6 compared to groups T1, T2 and T3 non-infested. The highest value was obtained on T1, while the lowest was recorded in positive control. Packed Cell Volume was significant increase (P<0.05) in the non-infected groups compared to the infected groups. The average values in birds fed with garlic significantly increased from 26.35 to 31.25% when compared to the positive control (21.75%). The lymphocytes count showed the highest values in the non-infected groups compared to the infected groups. However, the neutrophils in the infected birds had higher values than the values in the non-infected birds.

The result of serum biochemistry analysis presented in Table 2 showed that birds in the positive control group had higher AST, ALT, ALP and TP than groups T1, T2, T3, T4 and T5. Nevertheless, the concentrations of these parameters were reduced in groups fed garlic (T2 and T5) and ciprofloxacin (T3 and T4).

The weights of the liver and small intestine (Table 3) were significantly (P<0.05) affected by treatments. The value recorded on T6 was higher compared to the values obtained in other treatments. Feeding bird with garlic or ciprofloxacin reduce the weight of the organs.

Table 1: Least square mean±SEM of hematological parameters.

<table>
<thead>
<tr>
<th>PARAMETERS</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCV (%)</td>
<td>33.25±1.37a</td>
<td>31.25±1.10a</td>
<td>32.00±0.81a</td>
<td>26.25±1.31b</td>
<td>26.35±0.92b</td>
<td>21.75±0.5c</td>
</tr>
<tr>
<td>RBC *10³/L</td>
<td>2.13±0.04a</td>
<td>2.08±0.06a</td>
<td>1.98±0.03a</td>
<td>1.46±0.06b</td>
<td>1.57±0.10b</td>
<td>1.57±0.04b</td>
</tr>
<tr>
<td>WBC *10³/L</td>
<td>8.98±0.39b</td>
<td>8.93±0.28b</td>
<td>8.95±0.20b</td>
<td>13.83±0.10a</td>
<td>13.32±0.99a</td>
<td>13.60±0.55a</td>
</tr>
<tr>
<td>HB (g/dl)</td>
<td>10.32±0.14a</td>
<td>10.15±0.29a</td>
<td>10.17±0.21a</td>
<td>7.10±0.27b</td>
<td>7.27±0.47b</td>
<td>6.77±0.22a</td>
</tr>
<tr>
<td>Neutrophil (%)</td>
<td>15.75±2.17a</td>
<td>21.00±1.91b</td>
<td>22.25±3.01bc</td>
<td>36.00±7.25ab</td>
<td>40.25±10.61a</td>
<td>34.25±2.83b</td>
</tr>
<tr>
<td>Lymphocyte (%)</td>
<td>80.50±3.01a</td>
<td>76.75±1.37a</td>
<td>76.00±2.82a</td>
<td>62.00±7.29ab</td>
<td>49.50±10.97b</td>
<td>63.50±3.42d</td>
</tr>
<tr>
<td>Monocyte (%)</td>
<td>2.00±1.08</td>
<td>1.50±0.28</td>
<td>1.25±0.47</td>
<td>1.00±0.40</td>
<td>1.07±0.41</td>
<td>1.50±0.64ab</td>
</tr>
<tr>
<td>Basophile (%)</td>
<td>1.00±0.70</td>
<td>0.50±0.28</td>
<td>0.50±0.28</td>
<td>0.50±0.28</td>
<td>0.37±0.23</td>
<td>0.50±0.28</td>
</tr>
<tr>
<td>Eosinophil (%)</td>
<td>0.75±0.47</td>
<td>0.25±0.25</td>
<td>0.00±0.00</td>
<td>0.50±0.50</td>
<td>0.07±0.07</td>
<td>0.25±0.25</td>
</tr>
</tbody>
</table>

Table 2: Least square mean±SEM of some serum biochemical parameters in different treatments

<table>
<thead>
<tr>
<th>PARAMETERS</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
</tr>
</thead>
<tbody>
<tr>
<td>AST (U/L)</td>
<td>209.30±17.64ab</td>
<td>208.95±18.67ab</td>
<td>181.15±7.66a</td>
<td>187.55±7.70ab</td>
<td>209.02±9.93ab</td>
<td>222.95±2.77a</td>
</tr>
<tr>
<td>ALT (U/L)</td>
<td>16.58±1.90b</td>
<td>15.40±2.77b</td>
<td>11.87±7.16a</td>
<td>29.50±5.91b</td>
<td>16.31±1.59b</td>
<td>30.19±7.10a</td>
</tr>
<tr>
<td>ALP (U/L)</td>
<td>4860±640a</td>
<td>4309±723b</td>
<td>4635±423b</td>
<td>7650±490ab</td>
<td>5291±863b</td>
<td>8794±910b</td>
</tr>
<tr>
<td>TP (g/dL)</td>
<td>3.02±0.13ab</td>
<td>3.10±0.40ab</td>
<td>3.10±0.17ab</td>
<td>3.20±0.15ab</td>
<td>2.90±0.58ab</td>
<td>3.65±0.13a</td>
</tr>
</tbody>
</table>

Data shown are Mean±SEM. a,b,c: Row means with different superscripts are either significant at P<0.05 or highly significant at P<0.01. T1=control; T2=Garlic only; T3=Cipro only; T4=Salmonella+Cipro; T5=Salmonella+Garlic; T6=Salmonella only; WBC=White Blood Cell; PCV=Packed cell volume; RBC=Red Blood Cell; HB=Hemoglobin concentration.
Fig. 1: Treatment protocol.

Fig. 2: Photomicrograph of liver sections from experimental birds of treatments 1, 2, 3, 4, 5 and 6, H and E ×400: Hepatic artery (HA), Portal vein (PV), Bile ductile (BD), Centrilobular venule (CV); (T1, T2 and T3): normal histology; (T4): aggregates of lymphoid cells (arrow); (T5): normal hepatic histomorphology; (T6): multifocal areas of necrosis (arrow) and multifocal aggregates of lymphoid cells.

Table 3: Organ’s weight

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Treatments</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
</tr>
</thead>
<tbody>
<tr>
<td>Liver</td>
<td></td>
<td>10.23±0.42b</td>
<td>8.74±0.99b</td>
<td>9.81±0.56b</td>
<td>10.19±0.41b</td>
<td>8.97±0.76b</td>
<td>10.88±0.48a</td>
</tr>
<tr>
<td>Small Intestine</td>
<td></td>
<td>22.47±1.37b</td>
<td>21.13±2.22b</td>
<td>28.45±1.81a</td>
<td>21.01±0.97b</td>
<td>23.75±1.10b</td>
<td>28.52±0.78a</td>
</tr>
</tbody>
</table>

Data shown are Mean±SE. abc - Row means with different superscripts are either significant at P<0.05 or highly significant at P<0.01. T1=control; T2=Garlic only; T3=Cipro only; T4=Salmonella+Cipro; T5=Salmonella+Garlic; T6=Salmonella only.

Photomicrographs of liver and ileum sections from experimental birds on T1, T2, T3, T4, T5 and T6 are shown in Fig. 2 and 3 respectively. Fig. 2 shows normal hepatocytes with normal hepatic sinusoids containing RBCs on T1, T2, T3 and T5. The sections on T4 presented random multifocal variably sized and mostly well circumscribed aggregates of lymphoid cells (arrow). These lymphoid aggregates were mostly organized adjacent to the centrilobular vein or the portal tracts. On T6, it was observed multifocal areas of necrosis (arrow), multifocal aggregates of lymphoid cells and dissociated hepatocytes. Fig. 3 revealed normal intestinal villi (white arrow) and crypts (black arrow) lined by normal enterocytes with normal lamina propria of the mucosa on T1, T2 and T3. The sections on T4 showed a relatively normal intestinal histomorphology. However, a few areas presented intense
cellular infiltration of the lamina propria of the mucosa (black arrow) with formation of variably sized and often well circumscribed lymphoid nodules (white arrow). On T5, necrosis and sloughing of the enterocytes at the villus tips (black arrow) with mild to moderate leucocytic infiltration of the lamina propria were observed (white arrow). The sections observed on T6 revealed marked necrosis of the enterocytes at the villus tips with marked leucocytic infiltration of the lamina propria (white arrow) as well as necrosis of the crypt lining cells (black arrow).

DISCUSSION

The main objective of this study was to evaluate the effects of garlic on F₁ chicks (White Leghorn cocks × Nigerian Heavy Ecotype hens) uninfected and infected with S. typhimurium. It is well known that haematological profile (Table 1) is an essential indicator of the physiological or pathophysiological status of the animals’ well-being (Onasanya et al. 2015). The present findings revealed that, the range of WBC in the infected groups was higher than the normal ranged from 8.85-10.22×10³/L for bird reported by Abdi-Hachesoo et al. (2011). Moreover, it was also observed the increase in the number of neutrophils in those groups. This may probably be because of the present of S. typhimurium in the bird systems, which is responsible for infection and inflammation (Khan and Chousalkar 2020). It has been reported that, during the early stage of an infection a large number of neutrophils migrate from the peripheral blood to the site of inflammation where the microorganism is eliminated by both phagocytic and non-phagocytic mechanisms (Mortaz et al. 2018). The observed increase in the number of neutrophils in the treatments fed garlic (T2 and T5) compared to the negative control could prove the capacity of garlic to stimulate these leucocytes, which are the most dominant and efficient cells during both inflammation and phagocytosis (Rosales 2018). These results may be attributed to the diallyl sulfide, a sulfur compound found in garlic with the potential to increase immunity by stimulating several cells including macrophages, neutrophils and lymphocytes (Subramanian et al. 2020). In agreement with the present results, Ademola et al. (2004) stated that birds fed garlic powder have an increase value of neutrophils count than their counterparts without garlic inclusion. This observation against immuno-stimulatory effects of garlic also corroborates with the report of Zeryehun et al. (2017).

The significant decrease (P<0.05) of the RBC and HB counts in groups challenged with Salmonella indicates that, the bacteria negatively affect the erythrocyte precursor hormone (erythropoietin) responsible for the synthesis of RBC. Zivot et al. (2018) reported that, the production of the RBC is stimulated by erythropoietin (EPO), which is a humoral cytokine produced in the kidney and released into the systemic circulation with the primary role to regulate oxygen transport to organs. However, the infection of this organ can decrease the quantity of RBC into the bloodstream leading to the dyserythropoietic diseases including leukaemia and anaemia. Consequently, inefficient erythropoiesis results in the increase in quantity of immature RBC precursors and leukocytes in the general circulation (Rosche et al. 2015; Moras et al. 2017). The reduction of the oxygen carrying capacity of these cells therefore affect the well-being of birds. Feeding bird with garlic without S. typhimurium infection did not show any negative effect. This could mean that the consumption of garlic for 6 days did not cause anaemia and hypoxia to birds. This could be attributed to the capacity of garlic to stimulate EPO trough erythropoiesis and thus preventing tissue hypoxia (Brines and Cerami 2012). In contrast, Al-
Jowari (2017) reported that, the administration of garlic powder at 1% and 5% compared to the control group in male rabbits for 21 days cause a significant (P<0.05) increase in the RBCs count. This difference may be due to the garlic preparation, the duration of the experiment or/and the species variation.

One of the tests giving more information about anaemia and the rate of dehydration is PCV. The normal PCV for chickens ranged from 22-43% as reported by Santosso et al. (2015). The authors observed on this study that, the percentage of PCV on T6 was lower than the ranges earlier reported. This may be due to the harmful effect of S. typhimurium inducing liver and kidney inflammation, folic acid and serum iron deficiency (Shubham et al. 2020). In addition, Chandra et al. (2014) stated that, Salmonella can also destroy the haematopoietic precursors in the organs such as liver. In the same vein, Dar et al. (2018) also reported that RBC, PCV and HB decreased significantly (P<0.05) in bird system after the S. typhimurium infection. However, feeding bird with garlic did not cause any common conditions. This is because allicin, the main phytochemical in garlic has the potential to inhibit the growth of S. typhimurium and maintain normal the level of RBC in the organism (Salehi et al. 2019). In addition, Vitamin C or ascorbic acid an efficient water-soluble antioxidant found in garlic can also improves the absorption of iron by capturing non-heme iron and storing it in a form that can be absorbed by the organism. Iron plays a key role in the production of hemoglobin, which is responsible for oxygen transportation (He et al. 2018; Timoshnikov et al. 2020). This result agrees with the findings of Zeryehun et al. (2017) who noted that the administration of garlic extract at 1% significantly (P<0.01) improved the PCV value of White Leghorn chicken.

The values recorded for lymphocytes count were within the normal ranged from 24-84% for chickens as reported by Santosso et al. (2015). The non-significant values obtained in the infected groups could be attributed to the natural resistance of the local chicken used as parent stock.

Serum biochemistry profiling is a major tool used in the diagnosis and treatment of livestock diseases through the investigation of many parameters influencing serum bio-chemical indices (Onasanya et al. 2015). Aspartate aminotransaminase (AST), ALT, ALP and TP are often used to detect liver damage and dysfunction. This is because during pathological infection, ALT and AST are released by the liver in the blood to reflect the status of the animal and, TP to express the parenchymal damages (Kalra et al. 2021). The increased quantity of ALP in the blood, is mostly due to liver diseases, bile duct obstruction, gallbladder diseases, or bone disorders (Babu et al. 2020). The normal ranges of the concentration of liver enzymes and total protein are: 70-220U/L (AST), 568-8831U/L (ALP) (Odunitan-Wayas et al. 2018) and 2.8-3.32g/dL (TP) (Tóthová et al. 2019). In the present study, AST and TP on T6 were out of the normal ranges earlier reported. Alkaline phosphatase was withing the normal ranges for all the treatment. The significant (P<0.05) decreased results of the liver enzymes and TP in chicks fed garlic could be due to the protective effect of the biochemical components it contains such as saccharides and phenolic compounds (Mhyson 2017). The plant extract may also be non-toxic to the liver. These results agree to the findings of Omer et al. (2019) who also made similar observation on the capacity of garlic to improve bird health.

In this study, the increase weights of the liver and small intestine recorded in the positive control may be a result of the competition of the bacteria with the microbiota for nutrients in order to overcomes colonization resistance and disseminate in the liver (Gart et al. 2016). Salmonella typhimurium is able to alter the nutritional environment by metabolizing several compounds, such as ethanolamine produces in the intestine, as a source of carbon and nitrogen. The used of ethanolamine is to produced necessary energy to enhance its growth (Báumlmer and Sperandio 2016). Feeding bird with garlic or ciprofloxacin reduce the weight of the organs and thus the mortality rate (result not shown). The findings of this study compare favorably with the results of Adibmoradi et al. (2006) who reported that garlic is able to activate the absorptive process as an antibiotic alternative in chicken. The beneficial effect of garlic observed is probably due to the potency of the bioactive compounds it contains, which are responsible for the antibacterial activity of the garlic hexane extract (El-Saber Battha et al. 2020; Tchoupou-Tchoupou et al. 2021).

According to the previous results, S. typhimurium is responsible to several injuries in the liver (Fig. 2) and ileum (Fig. 3) tissues, even though no gross lesions were observed. The harmful effects of the bacteria recorded on T6 had also been reported by Muna et al. (2016) during S. typhimurium outbreak in Sudan. These authors stated that natural infestation of chicks with Salmonella damaged organs such as liver, intestine, kidney and heart after postmortem examinations. In addition, Kumari et al. (2013) also stated a desquamation of mucosal epithelium, villi and lumen necrosis, infiltration of granulated and mononuclear leukocytes, pathologic hyperplasia, and focal fibroblastic tumors when broiler chickens were infected with S. gallinarum. These observations testify the severity of the infiltration of Salmonella in the bird’s system. However, feeding bird with garlic significantly reduced the tissues lesions. It could be supposed that the mechanisms by which garlic alleviated the hepatocytes lesions in chicks infected with S. typhimurium may be attributed to its antioxidant compounds including allicin. Allicin reacts with thiol-possessing enzymes to inhibit nitric oxide, hydroxyl and superoxide radical accumulation responsible to induce oxidative stress and tissues damages (Salehi et al. 2019). This study also showed that garlic did not causes abnormalities in the bird intestine. It can then be supposed that garlic can be used to balance the microbiota and overcome dysbiosis in order to enhanced diversity of commensal microbes in the gastrointestinal tract.

**Conclusion**

This study has emphasized on the effects of garlic in improving the health of F1 chicks (White Leghorn cocks x Nigerian Heavy Ecotype hens) uninfected and infected with S. typhimurium. It was observed that garlic could be used in poultry industry as an alternative to antibiotics (such as ciprofloxacin) to combat salmonellosis and fight antibiotic resistance. The administration of this probiotic prevents the destruction of the liver parenchyma and cell dysfunction. Garlic also has immuno-stimulatory effects of
neutrophils whose role is known in the destruction of the pathogen in a cell/organism. Histopathological analysis showed that garlic hexane extract has a hepatoprotective power. This demonstrates that garlic hexane extract can be used in poultry farms to fight against microbial resistance induce by antibiotics and then prevent humans from zoonosis.

Acknowledgements

We thank all the technical and research staffs of the Departments of Animal Science and Veterinary Medicine, Faculty of Agriculture, UNN who contributed to the data collection.

Funding

I would like to thank the German Academic Exchange Service (DAAD) for the generous financial support. It is because of you that I have been able to do this work. I am extremely grateful!

REFERENCES


Veterinary Medicine Sciences 16: 16-23. https://doi.org/10.29079/vol16iss1art1411