Effects of Garlic *(Allium Sativum)* Aqueous Extract on Haematological and Serum Biochemical Parameters of Broiler Chickens

[1]Adewole, F. A., [1]Opowoye, I. O., [2]Ayo-Ajasa, O., [2]Ayangbade, O., [2]Okpukpan, W., [2]Egbeyale, L. T. and [2]Sogunle, O. M.

[1]Institute of Food Security, Environmental Resources and Agricultural Research, Federal University of Agriculture, Abeokuta, P.M.B. 2240, Ogun State, Nigeria.

[2]Department of Animal Production and Health, Federal University of Agriculture, Abeokuta, P.M.B. 2240, Ogun State, Nigeria.

Corresponding author: adewolefa@funaab.edu.ng; +2348027170579

Abstract

The use of natural plant extracts to improve animal health and productivity is gaining more attention. An investigation was conducted on the effect of aqueous extract of garlic (Allium sativum) on haematology and serum biochemical parameters of 120 1-day-old Arbor acres broilers at starter (0-4 weeks) and finisher (5-6 weeks) phases. Fresh garlic bulbs were crushed with a mortar and pestle, and mixed at a ratio of 50 g garlic per litre of water. After sieving, the prepared aqueous extract was administered to the respective treatment groups via their drinking water at the designated inclusion levels of 0%, 5%, 10%, and 15% aqueous garlic extract in drinking water. The experiment was laid out in a Completely Randomized Design with the control group received broad-spectrum antibiotics for the first three days and again at four weeks. Feed and water containing test ingredients were offered to the birds ad libitum for 7 weeks. Blood samples were collected from birds' wing webs using sterile syringes and needles. Data were subjected to One Way Analysis of Variance at 5% level of significance. At starter phase, significantly ($p<0.05$) higher values were observed for Packed Cell Volume (PCV), Haemoglobin concentration (Hb) and Red Blood Cell (RBC) of birds on T1 compared to other treatments' values which were similar. White Blood Cell (WBC) significantly ($p<0.05$) decreased in birds on T4 compared to birds on T1. Total protein, Albumin and Alanine aminotransaminase (ALT) showed significantly ($P<0.05$) highest values in T2 and were similar with T1 values. Cholesterol significantly ($P<0.05$) increased in birds on T1 and showed reduction as extract inclusion level increased. For finishers, all haematology indices were not significantly ($p>0.05$) influenced, while Calcium, ALT and Alkaline phosphatase (ALP) significantly ($P<0.05$) increased for birds on treatments with inclusion of extract at different levels compared to T1 showing lowered values. Conclusively, haematology and biochemical parameters in this study suggest broiler chicken's health status at starter or finisher phase was not compromised by aqueous extract of garlic.

Key words: garlic; aqueous extract; haematology; biochemical parameters; broiler chickens

Effets De L'extrait Aqueux D'ail (*Allium Sativum*) Sur Les Paramètres Hématologiques Et Biochimiques Sériques Des Poulets De Chair

Résumé

L'utilisation d'extraits végétaux naturels pour améliorer la santé et la productivité animales suscite un intérêt croissant. Une étude a été menée sur l'effet d'un extrait aqueux d'ail (Allium sativum) sur les paramètres hématologiques et biochimiques sériques de 120 poulets de chair Arbor acres âgés d'un jour, durant les phases de démarrage (0-4 semaines) et de finition (5-6 semaines). Des bulbes d'ail frais ont été broyés au mortier et mélangés à raison de 50 g d'ail par litre d'eau. Après filtration, l'extrait aqueux préparé a été administré aux groupes de traitement respectifs via leur eau de boisson, selon des niveaux d'inclusion de 0 %, 5 %, 10 % et 15 % d'extrait aqueux d'ail. L'expérience a été conçue selon un dispositif complètement randomisé, le groupe témoin recevant des antibiotiques à large spectre durant les trois premiers jours puis à quatre semaines. Les aliments et l'eau contenant les ingrédients testés ont été fournis

ad libitum pendant 7 semaines. Des échantillons sanguins ont été prélevés au niveau des ailes des oiseaux à l'aide de seringues et aiguilles stériles. Les données ont été soumises à une analyse de variance unidirectionnelle au seuil de signification de 5 %. Durant la phase de démarrage, des valeurs significativement plus élevées ($p < 0,05$) ont été observées pour l'hématocrite (PCV), la concentration d'hémoglobine (Hb) et les globules rouges (RBC) chez les oiseaux du groupe T1 par rapport aux autres traitements, dont les valeurs étaient similaires. Les globules blancs (WBC) ont significativement diminué ($p < 0,05$) chez les oiseaux du groupe T4 comparé à ceux du groupe T1. Les valeurs de protéines totales, d'albumine et d'alanine aminotransférase (ALT) étaient significativement plus élevées ($P < 0,05$) dans le groupe T2 et similaires à celles du groupe T1. Le cholestérol a significativement augmenté ($P < 0,05$) chez les oiseaux du groupe T1 puis a diminué avec l'augmentation du niveau d'inclusion de l'extrait. Pour la phase de finition, aucun indice hématologique n'a été significativement influencé ($p > 0,05$), tandis que le calcium, l'ALT et la phosphatase alcaline (ALP) ont significativement augmenté ($P < 0,05$) chez les oiseaux des groupes avec inclusion d'extrait à différents niveaux, comparé au groupe T1 qui présentait des valeurs plus basses. En conclusion, les paramètres hématologiques et biochimiques de cette étude suggèrent que le statut sanitaire des poulets de chair, en phase de démarrage ou de finition, n'a pas été compromis par l'extrait aqueux d'ail.

Mots-clés : ail ; extrait aqueux ; hématologie ; paramètres biochimiques ; poulets de chair
Running title: Garlic Extract Effects on Broiler Blood Parameters

Introduction

The contribution of poultry to food security, protein supply, and peoples' livelihood makes them valuable on a global level (Vaarst *et al.*, 2015). In appropriate amounts, animal source foods are valuable sources of complete, high-quality, easily digestible protein including numerous essential micro-nutrients such as iron, zinc, calcium, vitamin A and vitamin B_{12} (Schönfeldt, 2013) and as reported by Dowarah (2013), high quality protein are provided by chicken eggs and meat which are also significant sources of vitamins, minerals and essential fatty acids. Marangoni *et al.* (2015) reported the association between poultry consumption, within a balanced diet, and a reduction in the risk of developing cardiovascular diseases and their risk factors (overweight and insulin resistance and tumors). This health benefit including the provision of high-quality, affordable animal protein, a high chance for investment, job opportunities and a source of income for smallholders worldwide (Attia *et al.*, 2022) has attached much importance to the poultry industry and in the past five decades, the poultry industry has grown through antibiotic use combined with strict biosecurity and hygiene measures that have helped prevent the negative impacts of many avian diseases (Mehdi *et al.*, 2018). Antibiotics are used in the poultry sector primarily for treatment, prophylaxis, and growth promotion. In many parts of the world, food-producing animals are given antibiotics daily to make them grow faster and prevent diseases (Letlhogonolo *et al.*, 2020). Antibiotics have been used for many decades in the poultry industry to enhance production, promote growth performance and protect birds from pathogenic microbes (Ayalew *et al.*, 2022). However, the overuse use of antibiotics has resulted in a substantial increase in the rates of antibiotic excretion and environmental release and also the growth of drug resistant bacterial strains; this represents a global problem and a considerable threat to human health. Resistant bacteria are responsible for spread of infections that are more difficult to treat, requiring the use of drugs that are more toxic and more expensive (Serwecinska, 2020). According to Yang *et al*. (2015) there have arisen a significantly increased number of studies focused on the search for alternatives to antibiotics that possess similar antimicrobial and growth-promoting effects without inducing bacterial resistance and potential side effects to

animals. Lately, some feed additives including phytogenics are being used to replace antibiotic growth promoter and they are reported to enhance performance, feed conversion ratio, carcass meat safety and quality in animals (Upadhaya and Kim, 2017). Phytogenic feed additives are natural, less toxic, residue-free and ideal feed additives for poultry compared with synthetic antibiotics. As natural alternative to antibiotic growth promoters in poultry production, they have attracted an increasing attention and can be included in feeds as dried, solid and ground forms or as extracts (crude, concentrated and purified) (Abdelli *et al.,* 2021). They are plant-origin extracted compounds used in poultry production and they include a broad range of substances like herbs, spices, botanicals, oleoresins and essential oils (Ayalew *et al.,* 2022).

Garlic (*Allium sativum*) is a spice and native medicine that has been in use for many years. It is reported to possess antibacterial, antifungal, antiparasitic, antiviral, antioxidant, anti-cholesteromic, anti-cancerous, and vasodilator characteristics (Karangiya *et al*., 2016). Garlic extract and or components were reported to prevent chemically induced tumours or acute toxic effects of chemicals and the chemo-preventive potential of garlic has been attributed to the presence of several bioactive organosulphur compounds such as diallyl sulfide, diallyl disulfide, and allicin (Fadlalla *et al*., 2010). As a natural feed supplement, broiler performance and decreased mortality rate have been enhanced due to garlic (Makwana *et al.*, 2015; Karangiya *et al.*, 2016; Puvača *et al.*, 2019).

The supplementation of garlic in broiler feeds according to Motasem *et al.* (2018) has been recognized for its strong stimulating effect on the immune system and positive effects on digestion in birds due to its very rich aromatic essential content. In addition, garlic possess active compounds which are capable of substituting the role of synthetically made antibiotics used in chicken production (Taufik and Maruddin, 2019). Previously, garlic was reported as a growth promoter which aided reduced cost of production by improving growth performance and enhancing bird health. Also, Oleforuh-Okoleh *et al.* (2015) stated that the physiological, pathological and nutritional status of poultry is affected by haematological parameters which are greatly impacted upon by garlic. This study was therefore carried out to determine the effects of aqueous extract of garlic *(Allium sativum)* administered through drinking water on blood parameters of broiler chickens.

Materials and Methods

Experimental Site

The study was carried out at the Livelihood Support and Development Center, Sliden Africa, Agbede community near Federal University of Agriculture Abeokuta, Ogun state, Nigeria. The blood haematology and serum biochemical parameters of birds was determined at the Department of Biochemistry, Federal University of Agriculture Abeokuta, Ogun State, Nigeria. The coordinates of the area lie within latitude 7.2437^{o} N and longitude 3.3433^{o} E. It has an annual mean temperature of 34.7^{o}C and average relative humidity is about 82%.

Preparation of aqueous extract of garlic

The garlic was purchased from an open market in Abeokuta. Each bulb of garlic was peeled, washed and crushed manually with the use of local mortar and pestle made of wood. Afterwards, the crushed garlic was weighed using a sensitive scale and mixed with clean water at a ratio of 50 g to 1 litre of water at room temperature in a container. Routinely, the following morning, it was sieved and added to the drinking water of the birds at 5, 10 and 15% inclusion levels. This was measured as 50, 100 and 150mL of garlic extract per litre of drinking water.

Management of experimental birds

One hundred and twenty, unsexed, one-day old broiler chicks (Arbor acres) were purchased from a reputable farm in Oyo State, Nigeria and brooded on deep litter for fourteen days. The birds were randomly distributed into four treatments groups from day old: Treatment 1(T1) birds were given no garlic extract at all (control-0%); Treatment 2 (T2), Treatment 3 (T3) and Treatment 4 (T4) birds were given garlic extract in their drinking water at 5, 10 and 15% inclusion levels, respectively. Each treatment contained three replicates with ten birds per replicate. Experimental period was for 6 weeks and prior to arrival of the birds, experimental pens were washed and disinfected properly. Daily routine management practices were observed and vaccination schedule for the farm was duly followed. Clean cool water or water mixed with the appropriate percentage of garlic extract for the different treatments and compounded diet were supplied to the birds *ad libitum*. The diet composition is presented in table 1

.

Table 1: Diet composition (%) for broiler chickens fed garlic aqueous extract

Ingredient	Starter	Finisher
Maize	58.00	53.50
Soya bean meal	29.40	20.20
Fish meal	1.60	0.40
Groundnut cake	6.00	10.00
Wheat offal	0.00	10.80
Bone meal	2.50	3.00
Oyster shell	1.50	1.00
Common salt	0.25	0.25
Methionine	0.25	0.25
Lysine	0.25	0.25
Premix	0.25	0.25
Total	**100.00**	**100.00**
Calculated nutrient levels		
Crude Protein (%)	23.29	22.11
Crude fibre (%)	3.27	4.03
Metabolizable Energy (Kcal/kg)	3176.11	3170.43

Collection and evaluation of blood

Blood samples were collected for haematological and biochemical parameters from birds, through veins in their wing webs using a sterile disposable 5ml syringe and needle at the end of 4^{th} and 6^{th} week from the starter and finishers, respectively. Blood samples were collected from two birds randomly selected from each replicate per treatment. The blood samples for haematological assay were collected into labeled Ethylene Diamine Tetra Acetic acid (EDTA) treated test tubes. Blood samples for serum biochemical analysis were collected into EDTA free test tubes. The following parameters were determined for haematology: Packed Cell Volume (PCV), Haemoglobin concentration (Hb), White Blood Cell count (WBC), Red Blood Cell count (RBC), Neutrophil, Lymphocyte, Basophil, Eosinophil, Monocyte, Mean Corpuscular Haemoglobin Concentration (MCHC). They were determined as previously described by Adeyemo *et al*. (2010) and Madubuike and Ekenyem (2006). The MCV, MCHC and MCH were estimated by calculation using a standard formula (Jaime and Howlett, 2008). For serum biochemistry, parameters determined were: Glucose, Total Protein,

Albumin, Uric acid, Cholesterol, Bilirubin, Aspartate transaminase (AST), Alanine transaminase (ALT) and Alkaline phosphatase (ALP) as previously described by Adeyemo *et al.* (2010). Calcium, phosphorus and other minerals were determined using commercial colorimetric kits (QuimicaClinicaAplicada S A Amposta, Spain).

Statistical Analysis

All data collected were subjected to Analysis of Variance (ANOVA) in a Completely Randomized Design (CRD) using Statistical Analysis Software (SAS, 2012) at 5% level of significance. Significantly ($p<0.05$) different means among variables were separated using Duncan's Multiple Range Test contained in the same SAS (2012) version. Model for the study: $Yij = \mu + Ti + \Sigma ij$ Where: Yij = Individual Observation, μ = Overall Mean, Ti = Treatment Effect (inclusion levels of garlic extract in drinking water), Σij = Experimental error.

The effect of inclusion levels of garlic extract at 0, 5, 10 and 15% on haematological indices of broiler chicks at starter and finisher phases are shown in tables 2 and 3, respectively. The Packed Cell Volume (PCV), Haemoglobin concentration (Hb), White Blood Cell (WBC) and Red Blood Cell (RBC) for broiler chicks at starter phase were significantly ($p<0.05$) influenced across the treatment groups. Significantly ($p<0.05$) highest PCV (31.33%), Hb (10.67g/dl) and RBC (3.70 x 10^6/L) of birds on T1 (control treatment) compared to the values for birds on T2, T3 and T4 which were similar and not significantly different from each other. WBC was reduced significantly ($p<0.05$) in birds on T4 (17.97 x 10^6/L) compared to birds on T1 (25.33 x 10^6/L). The haematological parameters of birds at starter phase including values for all haematological indices of broiler finishers showed no significant ($p>0.05$) differences among the treatment groups with varying levels of inclusion of aqueous garlic extracts.

Results

Table 2: Effect of inclusion levels of garlic extract on haematological indices of broilers at starter phase

Parameters	T1(0)	T2 (5%)	T3 (10%)	T4(15%)	SEM
PCV (%)	31.33[a]	26.17[b]	23.83[b]	25.33[b]	1.00
Haemoglobin (g/dL)	10.67[a]	8.90[b]	8.12[b]	8.68[b]	0.34
RBC (x10^6/L)	3.70[a]	3.08[b]	2.81[b]	2.94[b]	0.12
WBC(x10^6/L)	25.33[a]	19.52[ab]	20.70[ab]	17.97[b]	1.09
Neutrophilis (%)	21.00	20.33	21.33	14.67	1.42
Lymphocytes (%)	73.67	76.33	74.00	80.17	1.60
Basophils (%)	0.33	0.33	0.50	0.17	0.14
Eosinophils (%)	2.33	1.50	1.17	2.83	0.32
Monocytes (%)	2.67	3.17	3.00	2.17	0.31
MCHC (g/100mL)	34.02	34.07	34.07	34.27	0.09

[a,b;] Means bearing different superscript in a row differ significantly (P<0.05); PCV = Packed Cell Volume, WBC = White Blood Cell, RBC = Red Blood Cell, MCHC = Mean Cell Haemoglobin Concentration, SEM = Standard Error of Mean

Table 3: Effect of inclusion levels of garlic extract on haematological indices of broilers at finisher phase

Parameters	T1(0%)	T2(5%)	T3(10%)	T4(15%)	SEM
PCV (%)	32.00	36.00	33.33	37.67	1.12
Haemoglobin (g/dL)	10.83	12.17	11.25	12.75	0.38
RBC ($x10^6$/L)	3.79	4.30	3.97	4.50	0.14
WBC($x10^6$/L)	13..77	11.87	16.62	18.15	1.14
Neutrophilis (%)	24.83	27.00	25.50	27.83	2.16
Lymphocytes (%)	71.16	69.83	69.50	66.33	2.21
Basophils (%)	0.00	0.50	0.17	0.67	0.13
Eosinophils (%)	2.33	1.17	3.17	2.50	0.33
Monocytes (%)	1.67	1.50	1.67	2.67	0.39
MCHC (g/100mL)	33.83	33.80	33.73	33.81	0.06

PCV = Packed Cell Volume, WBC = White Blood Cell, RBC = Red Blood Cell, MCHC = Mean Cell Haemoglobin Concentration, SEM = Standard Error of Mean

Tables 3 and 4 show the effect of the inclusion levels of garlic extract at 0, 5, 10 and 15% on serum biochemical indices of broiler chicks at starter phase and finisher phase respectively. The serum biochemical parameters differed significantly ($P<0.05$) at starter phase (Total Protein, Albumin, Cholesterol and ALT) and finisher phase (Calcium, ALT and ALP). In the broiler starter phase, the significantly ($P<0.05$) influenced values obtained for serum total protein, albumin and ALT were highest at T2 and lowest at T4. However, the significantly ($P<0.05$) high values in birds on T2 were similar with values for birds on T1. Cholesterol values were significantly ($P<0.05$) highest in birds on T1 and decreased gradually across the treatments with increased inclusion level of garlic extract. The finisher birds showed significantly ($P<0.05$) increased values for birds on treatmentsT4 (9.53mg/dL), T2 (8.80mg/dL) and T3 (8.36mg/dL), with inclusion of garlic extract at different levels compared to those on T1 (7.21mg/dL) which recorded the lowest values for Calcium. ALT and ALP followed similar trend.

Table 4: Effect of inclusion levels of garlic extract on serum biochemical indices of broilers at starter phase

Parameters	T1(0%)	T2(5%)	T3(10%)	T4(15 %)	SEM
Glucose(mg/dL)	124.23	122.03	121.03	113.33	5.17
Total Protein (g/L)	64.50^{ab}	68.20^{a}	61.47^{b}	60.52^{b}	1.09
Albumin (g/l)	35.98^{ab}	38.22^{a}	35.43^{ab}	33.45^{b}	0.58
Uric acid (mg/dL)	8.69	7.50	9.43	7.79	0.42
Cholesterol (mg/dL)	161.33^{a}	129.62^{ab}	116.60^{b}	99.50^{b}	7.78
Calcium (mg/dL)	8.24	7.99	8.07	8.25	0.08
Phosphorus (mg/dL)	6.29	5.29	5.69	5.68	0.26
Bilirubin (mg/dL)	0.55	0.68	0.63	0.62	0.03
AST (µ/L)	92.88	107.33	107.37	90.92	4.16
ALT (µ/L)	25.50^{a}	26.12^{a}	16.40^{b}	24.87^{ab}	1.60
ALP (µ/L)	280.52	231.27	235.32	232.13	15.67

$^{a,b:}$ Means within the same row having different superscripts differ significantly (P<0.05); AST = Aspartate transaminase, ALT = Alanine transaminase, ALP = Alkaline phosphatase, SEM = Standard Error of Mean

Table 5: Effect of inclusion levels of garlic extract on serum biochemical indices of broilers at finisher phase

$^{a,b:}$ Means within the same row having different superscripts differ significantly (P<0.05); AST = Aspartate

Parameters	T1(0%)	T2(5%)	T3(10%)	T4(15%)	SEM
Glucose(mg/dL)	136.3	126.00	147.30	143.38	4.69
Total Protein (g/L)	72.90	68.00	73.68	73.17	1.13
Albumin (g/L)	42.30	40.78	43.00	44.10	0.71
Uric acid (mg/dL)	5.20	5.30	6.14	4.97	0.32
Cholesterol (mg/dL)	107.92	101.95	95.47	103.38	2.61
Calcium (mg/dL)	7.21^{c}	8.80^{ab}	8.36^{b}	9.53^{a}	0.25
Phosphorus (mg/dL)	4.75	7.41	6.22	5.69	0.49
Bilirubin (mg/dL)	0.58	0.75	2.27	0.75	0.37
AST (µ/L)	94.47	91.13	84.35	74.48	3.49
ALT (µ/L)	14.85^{b}	16.53^{ab}	21.60^{a}	16.20^{ab}	1.02
ALP (µ/L)	160.12^{b}	259.43^{a}	196.37^{b}	185.70^{b}	9.72

transaminase, ALT = Alanine transaminase, ALP = Alkaline phosphatase, SEM = Standard Error of Mean

Discussion

The inclusion of garlic aqueous extract at 5, 10, 15% levels in broiler drinking water in this study showed these levels were adequate for the birds and while there were variations in blood PCV, Hb, RBC and WBC, there was absence of toxicity, anaemic conditions and dehydration in the birds. Evidence of significantly influenced haematological parameters in broilers by garlic supplementation have been reported previously (Rehman and Munir, 2015; Khan *et al.,* 2017; Bondona *et al.,* 2019; Kairalla *et al.,* 2022). Significant variations in red blood cells, packed cell volume haemoglobin and white blood cells were also reported by Onunkwo *et al.* (2019) and this was attributed to garlic powder having an effect on normal erythropoiesis and conveying oxygen, chemicals and nutrients essential for life. While the result of this study showed increased PCV for starter birds in the control group (T1) compared to those in the garlic treated groups, the findings of Elagib *et al.* (2013) and Bondona *et al.* (2019) reported higher PCV values in garlic treated groups compared to birds in the control. The significant effect of garlic treatments on Hb in this study could be attributed to the presence of several haemolytic bioactive constituents, diallyl sulfide, diallyl disulfide, allicin and/or their metabolites in garlic (Elagib *et al.,* 2013). The significant differences observed in WBC for starter birds in this study agrees with Fadlalla *et al.* (2010) who reported significantly increased WBC in garlic treated groups compared to the control group. Onunkwo *et al.* (2019) reported there was significant difference in WBC with higher values in garlic treated groups for both broiler starter and finisher phases and this indicated garlic's immuno-stimulant properties (Oluwole, 2001). Diet supplementation with garlic according to Hanieh *et al*. (2010) resulted in increased WBC, lymphocytes, and immunoglobulin G in broilers.

The range of values for PCV (23.83 - 31.33%), Hb (8.12 – 10.67g/dL), RBC (2.81-3.70x10^6/L) and WBC (17.97-25.33x10^6/L) in this study were within the normal range for broilers as reported by Mitruka (1977) and Banerjee (2009) and this indicates well-being of birds all through the experimental period as observed by the absence of disease in the birds (Olafedehan *et al.,* 2010). It also shows the absence of challenge to cellular respiration and metabolic reactions due to the treatments (Ekine *et al.,* 2017). In addition, the normal range of PCV values in this study indicate that the quantity of blood within the broiler body system was enough for their normal body functions to be carried out. This further emphasizes the good status of bird health in the current study since low PCV shows anaemia depicting a condition where RBC is few or deficient in haemoglobin thus resulting in poor health (Bot *et al.,* 2022). George-Gay and Parker (2003) demonstrated that WBC decrease was due to reduced bone marrow production or destruction caused by viral infection or toxic reaction but, there were no reduced WBC or signs of infection in the birds for this current study. The WBC counts in were within the normal range hence they possess capability of generating antibodies through phagocytosis with high degree of disease resistance (Soetan *et al*., 2013) and improved adaptability to disease conditions that are locally prevalent (Kabir *et al*., 2011). In contrast with this study, Ao *et al.* (2011) reported the lack of garlic effect on broiler leukocyte numbers.

The non-significant influence observed in Neutrophilis, Lymphocytes, Basophils, Eosinophils, Monocytes and MCHC for broiler starters and all haematological parameters for broiler finishers considered in this study agrees with the reports of Yang *et al.* (2007) and Elagib *et al.* (2013). Ismail

et al. (2022) also reported that garlic powder had no significant influence on PCV, WBC and Monocytes of broilers. Noman *et al.* (2015) documented that aqueous extract of garlic administered with drinking water had no significant influence on the blood haematology of broilers. Furthermore, RBC and MCHC according to Ekine *et al*. (2017) were not significantly influenced in a study on the effect of various management strategies and garlic granules on blood parameters.

Serum biochemistry is a reliable biochemical system in an organism which could reflect its condition and the changes happening to it under the influence of internal and external factors (Toghyani *et al.,* 2010). The variations observed in the serum biochemistry indices at both phases in this study are similar to the report of Bondona *et al.* (2019) that ALT and total serum cholesterol differed significantly among different experimental groups of broilers fed garlic powder. Moreover, Onunkwo *et al.* (2019) described that there were significant variations observed in all serum biochemical indices except for creatinine of starter and finisher broilers fed graded levels of garlic powder. This contrasts with the observation of Khan *et al.* (2012) that there were no differences in serum biochemical parameters (total protein, albumin, globulin, cholesterol, triglycerides, ALT, and AST) of broilers fed diet enriched with garlic.

Total protein (60.52 – 68.20g/L) in this study was within the normal range of 50 g/l – 70 g/L as reported by Barnejee (2009). Total protein and albumin values were highest at 5% inclusion level. Oleforuh – Okoleh *et al*. (2015) reported a significant increase in albumin of ginger or garlic treated birds when compared to the control group. Important nutrients are carried and delivered to body cells by albumin. For tissue growth and healing, albumin is essential as it supplies appropriate amino acids required for tissue proteins synthesis during quick somatic bird growth (Piotrowska *et al*. 2011), and also binds to toxins and free radicals hereby minimizing oxidation process and preventing cell damage.

Reduced cholesterol with increased inclusion level of garlic extract in this study was in agreement with Issa and Omar (2012) who established that addition of garlic powder significantly reduced triglyceride, cholesterol and LDL concentration in Cobb broilers and increased their HDL levels. The findings reported by Mansoub (2011) also reported a reduction in total cholesterol for broilers supplemented with 1g/kg of garlic. Ismail *et al.* (2021) reported that supplementing diets with garlic powder and phenyl acetic acid decreased blood total cholesterol and LDL and increased total protein, globulin and HDL in broilers. The mechanism involved in the potential effect of garlic decreasing cholesterol, triglyceride and LDL according to Yeh and Liu (2001) is reduced activities of hepatic lipogenic and cholesterogenic enzymes such as fatty acid synthase, malic enzyme, 3-hydroxy-3-methyl glutaryl-CoA (HMG CoA) reductase and glucose-6 phosphate dehydrogenase. Stanacev *et al.* (2011) opined that hypocholesterolemic effect of garlic in chicken manifested because the most important enzymes that participate in cholesterol and lipid synthesis are being inhibited.

The ALT is a liver enzyme that catalyzes the transfer of amino groups from L-alanine to α–ketoglutarate in order to produce hepatic metabolite oxaloacetate (Kim *et al*., 2008). ALT is more specific to the liver and can be an indicator to detecting liver injury. The ALT and ALP in both starter and finisher birds in this study did not follow any particular trend. They increased with higher inclusion level of garlic extract in drinking water for birds and values were

lowest for birds on control. This is in contrast with the findings of Oluwafemi *et al*. (2021) who reported that ALT, AST and ALP decreased as the level of ginger and garlic oil mixture inclusion increased in the diet. In another study (Kairalla *et al*., 2022), broiler diets supplemented with garlic powder were reported to have decreased values for ALT, total cholesterol, triglycerides, and low density lipoprotein (LDL) compared to values for birds in the control group.

Conclusion

In this study, the range of values for haematological and biochemical parameters were within the normal range and cholesterol in serum decreased with increased inclusion level of garlic extract. Thus, haematological and biochemical indices in this study indicate that broiler chicken's health status at either starter or finisher phase were not compromised by the aqueous extract of garlic.

References

Abdelli, N., Solà-Oriol, D. and Pérez, J.F. (2021) Phytogenic feed additives in poultry: Achievements, prospective and challenges. *Animals* 11(12):3471. https://doi.org/10.3390/ani11123471

Adeyemo, G.O., Ologhobo, A.D. and Adebiyi, O.A. (2010) The effect of graded levels of dietary methionine on the haematology and serum biochemistry of broilers. *International Journal of Poultry Science* 9(2): 158- 161.

Ao, X., Yoo, J.S., Zhou, T.X., Wang, J.P., Meng, Q.W., Yan, L., Cho, J.H. and Kim, I.H. (2011). Effects of fermented garlic powder supplementation on growth performance, blood profiles and breast meat quality in broilers. *Livestock Science*, 141:85-89.

Attia, Y. A., Rahman, M. T., Hossain, M. J., Basiouni, S., Khafaga, A. F., Shehata, A. A. and Hafez, H. M. (2022). Poultry production and sustainability in developing countries under the COVID-19 crisis: Lessons learned. *Animals: An Open Access Journal from MDPI*, *12*(5). https://doi.org/10.3390/ani12050644

Ayalew, H., Zhang, H., Wang, J., Wu, S., Qiu, K., Qi, G., Tekeste, A., Wassie, T. and Chanie, D. (2022). Potential feed additives as antibiotic alternatives in broiler production. *Frontiers in Veterinary Science*, *9*. https://doi.org/10.3389/fvets.2022.916473

Banerjee. G.C. (2009). A textbook of animal husbandry. 8th Edition. Oxford & IBH Publishing Co. PVT. Ltd., New Delhi. P132-139

Bondona, B., Joga, D. M., Deben, S., Bornalee, H. and Rafiqul, I. (2019). Effect of feeding garlic (*Allium sativum*) on haematological, serum biochemical profile and carcass characteristics in broiler chicken. *International Journal of Current Microbiology and Applied Science* 8(10):492-500. https://doi.org/10.20546/ijcmas.2019.810.054

Bot, M.H., Udom, I.E., Brengshak, S.B., Aliyu, I., Akure, C.O., Kpanja, E., Davou, G.C., Imoisi, O. and Duru, S. (2022). Haematological and serum biochemical indices of broiler chickens fed diets containing red finger millet (*Eleucine coracana*) with and without phytase enzyme. *Nigerian J. Anim. Sci*. 24 (3): 183-191

Dowarah, R. (2013). The role of poultry meat and eggs in human nutrition.

Poultry Line, June, 31-33. https://www.researchgate.net/publication/321254565

Ekine, O.A., Akinola, L.A.F. and Idakwo, E. (2017). Effect of various management strategies and garlic granules on haematological status of broilers in the tropics *Nigerian Journal of Animal Science*. (1):166 - 172.

Elagib, H.A.A., El-Amin, W.I.A., Elamin, K.M. and Malik, H.E.E. (2013). Effect of dietary garlic (*Allium sativum*) supplementation as feed additive on broiler performance and blood profile. *J. Anim. Sci. Adv*. 3(2): 58-64.

Fadlalla, I.M.T., Mohammed, B.H. and Bakhiet, A.O. (2010). Effect of feeding garlic on the performance and immunity of broilers. *Asian Journal of Poultry Science*, 4:182-189.

George-Gay, B. and Parker, K. (2003). Understanding the complete blood count with differential. *Journal of Perianesthesia. Nursing.* 18:96-114. doi:10.1053/j.pan.2003.50013

Hanieh, H., Narabara, K., Piao, M., Gerile, C., Abe, A. and Kondo, Y. (2010) Modulatory effects of two levels of dietary *Alliums* on immune response and certain immunological variables following immunization, in White Leghorn chickens. *Animal Science Journal*; 81:673-80. https://doi.org/10.1111/j.1740-0929.2010.00798.x

Ismail, I.E., Alagawany, M., Taha, A.E., Puvača, N., Laudadio, V. and Tufarelli V. (2021) Effect of dietary supplementation of garlic powder and phenyl acetic acid on productive performance, blood haematology, immunity and antioxidant status of broiler chickens. *Animal Bioscience.* 34(3): 363–370. doi: 10.5713/ajas.20.0140

Issa, K.J. and Omar, J.M.A. (2012) Effect of garlic powder on performance and lipid profile of broilers. *Open Journa of Animal Science.* 2012;2:62-8. https://doi.org/10.4236/ojas.2012.22010

Jaime, S. and Howlett, J. C. (2008) Avian Medicine. 2nd Edition, Mosby Elevier pp. 46

Kabir, M., Akpu, G.N., Nwagu, B.I., Adeyinka, I.A and Bello, U.I. (2011). Sexual dimorphism, breed and age characteristics of rabbits in Zaria, Nigeria. Proceedings of the 16th Annual Conference of Animal Science Association of Nigeria, 133-137.

Kairalla, M.A., Alshelmani, M.I. and Aburas, A.A. (2022). Effect of diet supplemented with graded levels of garlic (*Allium sativum* L.) powder on growth performance, carcass characteristics, blood hematology and biochemistry of broilers. *Open Veterinary Journal*, Vol. 12(5): 595–601. doi: 10.5455/OVJ. 2022.v12.i5.1

Karangiya, V. K., Savsani, H. H., Patil, S. S., Garg, D. D., Murthy, K. S., Ribadiya, N. K. and Vekariya, S. J. (2016). Effect of dietary supplementation of garlic, ginger and their combination on feed intake, growth performance and economics in commercial broilers. *Veterinary World*, *9*(3), 245-250. https://doi.org/10.14202/vetworld.2016.245-250

Khan, R.U., Nikousefat, Z., Tufarelli, V., Naz, S., Javdani, M. and Laudadio, V. (2012). Garlic (*Allium sativum*) supplementation

in poultry diets: Effect on production and physiology. *Worlds Poultry Science Journal*. 68:417-24. https://doi.org/10.1017/S0043933912000530

Khan, M. S. I., Prodhan, M. S., Islam, M. S., Hasan, M. N. and Islam, M. S. (2017) Effect of garlic extract on growth performances and hematological parameters of broilers. *Asian Journal of Medical and Biological Research*, 3:317-322.

Letlhogonolo, A.S., Zahra, M.H., Tlou, G.M. and Monnye, M. (2020). The current status of the alternative use to antibiotics in poultry production: An African perspective. *Antibiotics (Basel)*. 9(9):594 pp 1-18. doi: 10.3390/antibiotics9090594. PMID: 32932751; PMCID: PMC7559726.

Madubuike F. N and Ekenyem B.U (2006) Haematology and serum biochemistry characteristics of broiler chicks fed varying dietary levels of *Ipomea asarifolia* leaf meal. *International Journal of Poultry Science*, 5(1), 9-12.

Makwana, R.B., Raval, A.P., Chauhan, H.D., Kulkarni, R.C., Srivastava, A.K., Bhagwat, S.R. and Rajgor, B.B. (2015). Effects of garlic (*Allium sativum*) supplementation on growth performance, carcass characteristics and economics of broilers. *Journal of Animal Research.* 5(4), 843–848.

Mansoub, H.N. and Nezhad, M.A.M. (2011). The effects of using thyme, garlic and nettle on performance, carcass quality and blood parameters. *Annals of Biological Research*, 2: 315-320.

Marangoni, F., Corsello, G., Cricelli, C., Ferrara, N., Ghiselli, A., Lucchin, L. and Poli, A. (2015). Role of poultry meat in a balanced diet aimed at maintaining health and wellbeing: An Italian consensus document. *Food Nutrition Research*. 59:27606. doi: 10.3402/fnr.v59.27606.

Mehdi, Y., Létourneau-Montminy, M-P., Gaucher, M-L., Chorfi, Y., Suresh, G., Rouissi, T., Brar, S.K., Côté, C., Ramirez, A.A. and Godbout, S. (2018). Use of antibiotics in broiler production: Global impacts and alternatives. *Animal Nutrition.* 4(2): 170-178. ISSN 2405-6545, https://doi.org/10.1016/j.aninu.2018.03.002.

Mitruka, B. M., and Rawnsley, H. M. (1977). Clinical biochemical and hematological reference values in normal experimental animals. pp.134-135. USA: Masson Publishing Inc.

Motasem, A. M., Ramamneh, D.A., Sharafat, A.A. and Hussain, N. (2018). Effect of using garlic on the economical and physiological characteristics of broiler chickens. *International Journal Environment Science Nature Research*. 10(2): 555783 pp 054-058. doi: 10.19080/IJESNR.2018.10.555783

Noman, Z., Hasan, M., Talukder, S., Sarker, Y., Paul, T. and Sikder, M. (2015). Effects of garlic extract on growth, carcass characteristics and haematological parameters in broilers. *Bangladesh Veterinarian*, 32:1-6.

Olafedehan, C.O., Obun, A.M., Yusuf, M.K., Adewumi, O.O., Olafedehan, A.O., Awofolaji, A.O. and Adeniji, A.A. (2010). Cassava peel meals on haematological and biochemical indices of growing rabbits.

Proceedings of 35th Annual conference of Nigerian Society for Animal Production. (P.212).

Oleforuh-Okoleh, V.U., Ndofor-Foleng, H.M., Olorunleke, S.O. and Uguru, J.O. (2015). Evaluation of growth performance, haematological and serum biochemical response of broiler chickens to aqueous extract of ginger and garlic. *Journal of Agricultural Science*, 7: 167–174.

Oluwafemi, R.A., Daniel, S.E. and Alagbe, J. O. (2021). Haematological and serum biochemical indices of broiler chicks fed different inclusion levels of ginger *(Zingiber officinale)* and garlic *(Allium sativum)* oil mixture. *International Journal of Discoveries and Innovations in Applied Sciences.* 1:4. e-ISSN: 2792-3983 www.openaccessjournals.eu | Volume: 1 Issue: 4

Oluwole, F.S. (2001). Effects of garlic on some haemotological and biochemical parameters. *African Journal Biomed Research*, 4:139-141.

Onunkwo, D.N., Ezike, J.C., Wisdom, A., Daniel-Igwe, A. (2020). Effect of garlic rhizome (Allium sativum) powder supplemented diets on the haematology and serum biochemistry of broiler chickens at starter and finisher phases. *Nigerian Journal of Animal Production* 46(1):149-156 DOI: 10.51791/njap.v46i1.1312

Piotrowska, A., Burlikowska, K. and Szymeczko, R. (2011). Changes in blood chemistry in broiler chickens during the fattening period. *Folia Biologica (Kraków)*, 59:183–187. doi:10.3409/fb59_3-4.183-187.

Puvača, N., Ljubojević Pelić, D., Čabarkapa, I., Popović, S., Tomičić, Z., Nikolova, N. and Lević, J. (2019). Quality of broiler chickens carcass fed dietary addition of garlic, black pepper and hot red pepper. *Technology Engineering. Management.* 2, 218–227.

Rehman, Z.U. and Munir, M.T. (2015). Effect of garlic on the health and performance of broilers. *Veterinaria*, 3(1):32-39.

SAS. 2012. Statistical Analysis System (2012). SAS Users Guide Statistics. SAS Institute Inc., Cary, NC.

Schönfeldt, H.C, Pretorius, B. and Hall, N. (2013). The impact of animal source food products on human nutrition and health. *South African Journal of Animal Science*, 41(3) 394-412. ISSN 0375-1589 (print), ISSN 2221-4062 (online) http://dx.doi.org/10.431/sajas.v43i3.11

Serwecińska L. (2020) Antimicrobials and Antibiotic-Resistant Bacteria: A Risk to the Environment and to Public Health. *Water*. 12(12):3313. https://doi.org/10.3390/w12123313

Soetan, K. O., Akinrinde, A. S. and Ajibade, T. O. (2013). Preliminary studies on the haematological parameters of cockerels fed raw and processed guinea corn (*Sorghum bicolor*) Proceedings of 38th Annual Conference of Nigerian Society for Animal Production. pp. 49-52.

Stanacev, V., Glamoeic, D., Milosevic, N., Reri, L., Puvaca, N., Milic, D. and Plavsa, N. (2012). Influence of garlic (*Allium sativum*) and copper as phytoadditives in the feed on the content of cholesterol

in the tissues of chickens. *Journal of medical plants Research*. 6: 2816 - 2819.

Taufik, M. and Maruddin, F. (2019). The effect of garlic solution supplementation on performance, carcass weight and abdominal fat of broiler chickens. *IOP Conference series: Earth and environmental science*. pp 1-6. Bristol, UK: IOP Publishing.

Toghyani, M., Toghyani, M.A., Gheisari, G. and Mohammedrezaei, M. (2010). Growth performance, serum biochemistry and blood haematology of broiler chicks fed different levels of black seed (*Nigella sativa*) and peppermint (*Mentha piperita*). *Livestock Science*. 129:173 - 178.

Upadhaya, S.D. and Kim, I.H. (2017). Efficacy of phytogenic feed additive on performance, production and health status of monogastric animals–A review. *Annals of Animal Science*, 17(4): 929-948. doi: https://www.doi.org/10.1515/aoas-2016-0079

Vaarst, M., Steenfeldt, S., and Horsted, K. (2015). Sustainable development perspectives of poultry production. *World's Poultry Science Journal*, 71(4), 609-620. doi:10.1017/S0043933915002433

Yang, W.Z., Benchaar, C., Ametaj, B.N., Chaves, A.V., He, M.L. and McAllister, T.A. (2007). Effects of garlic and juniper berry essential oils on ruminal fermentation and on site and extent of digestion in lactating cows. *Journal of Dairy Science*. 9: 5671-5681.

Yang, C., Chowdhury, A. K., Hou, Y., and Gong, J. (2015). Phytogenic compounds as alternatives to in-feed antibiotics: Potentials and challenges in application. *Pathogens,4*(1)137-156. https://doi.org/10.3390/pathogens4010137

Yeh, Y-Y and Liu, L. (2001). Cholesterol lowering effect of garlic extracts and organosulfur compounds: Human and animal studies. *Journal of Nutrition.* 131: 989-993.

Date received: 24th January, 2025
Date accepted: 11th July, 2025