

GROWTH PERFORMANCE, HISTOPATHOLOGICAL, HAEMATOLOGICAL AND SERUM BIOCHEMICAL INDICES OF FINISHER BROILER CHICKENS OFFERED NUTRITIONAL SUPPLEMENT OF COMFREY (*Symphytum officinale* L) LEAVES EXTRACT.

Abstract

An experiment was carried out to investigate the effect of nutritional supplement of Comfrey (*Symphytum officinale* L) leaves extract on the performance, haematological and serum biochemical indices of finisher broilers. 200g of Comfrey symphytum leaves were squeezed in 1 liter of H₂O with a juicer to produce comfrey symphytum leaves extract. The leaves extract were further mixed (diluted) in clean drinking water at the rate of 0ml/liter of H₂O, 50ml/liter of H₂O, 100ml/liter of H₂O and 150ml/liter of H₂O designated as T₁, T₃, T₄ and T₅. T₂ was given conventional supplement (Divertamin). Five groups of 30 broiler chicks (28 days old) of Agritech breed were randomly assigned to one of the supplementary Comfrey symphytum leaves extract and the conventional supplement for 28 days in a completely randomized design. Each group was subdivided into three replicates of 10 birds each. The birds were fed normal broiler finisher diet for all the groups. At the end of the 28 days trial, performance indices result showed that average initial weight, average final weight and average daily feed intake were not significantly ($P>0.05$) affected. Average weight changes, Average daily weight gain and feed conversion ratio were significantly ($P<0.05$) affected. T₅ (150ml extract/liter of H₂O) gave best value ($P<0.05$) in terms of performance for feed conversion ratio, average weight changes and average daily weight gain but statistically similar ($P>0.05$) to the control. The indices for economics of production showed that T₅ had high earnings for revenue and gross margin with reduced cost/kg weight gain. The haematological indices (haemoglobin, packed cell volume, red blood cell and white blood cell) and the biochemical indices (urea, creatinine, total protein, cholesterol, serum glutamate oxaloacetate transaminase, serum glutamate pyruvate transaminase) were not affected significantly ($P>0.05$). Histological observations revealed that comfrey leaves extract caused effaced tissue stromal elements, congestion and marked cellularity of the liver. It was concluded that Comfrey symphytum leaves extract could be used as a nutritional supplement to improve performance of broiler finishers at 150ml of extract/liter of H₂O, to reduce cost/kg weight gain and increase revenue and profit margin.

Key words: Comfrey leaves, Extract, Performance, Histology, Haematology, Biochemistry.

INTRODUCTION

The need to improve on the growth and liveability of broilers is becoming very urgent due to the need to produce healthy table meat within a short period of time for the increasing population of mankind (Esiegwu 2019). FAO (2010) reported that out of the 53g of protein per caput per day, Nigeria obtains 10 - 15g per caput per day from animal source as against the recommended 35g per caput per day. This challenge is as a result of limited production of animal products due to high cost of production which is incident on the final consumers in the form of high price of animal and poultry products leading to a drop in consumption. Most poultry feed may not be nutritionally adequate due to lack of essential nutrients necessary for growth and good health. The implication is the purchase of exorbitant synthetic nutrients fortified with vitamins, minerals and some essential amino acids to enhance the nutritional status of the animals which adds to the cost of production and consequently, high cost of broiler meat (Esiegwu, 2019). Protein from plant leaves source is perhaps the most naturally abundant and the cheapest potential source of protein (Fasuyi and Nonyerem, 2007). Leaf vegetables supply minerals, proteins and vitamins which could complement the inadequacies of most feed stuffs (Hon and Basir, 1980). It is necessary to exploit the benefits of most of these vegetables as nutritional supplement in order to reduce the cost of using conventional supplements that may add heavily to the cost of production. Leaves as a nutritional supplement is good but limited due to high fiber content which is a burden to the digestive system. A healthy body with sound digestive system has the capacity to disintegrate about 35% of the ingredients from vegetables and assimilate them where as the body is able to absorb 95% of substances contained in juice (Gala *et al.*, 2003). The use of juice extract as a dietary supplement from plant leaves appear to be more effective and efficient way of deriving the nutrients and enzymes in leaves of plants required in the body for proper metabolism and utilization in the tissues for growth and the other metabolic functions.

Comfrey plant leaves appear to have the potential to supply needed proteins, vitamins and minerals and could serve as a nutritional supplement to enhance growth. Comfrey (*Symphytum officinale* L) is native to Europe, Central Asia and prevalent in the South Eastern part of Nigeria, where it is used in bone healing, treating anemia and as a blood booster. Greeks and Romans used Comfrey to stop heavy bleeding, treat bronchial problems, heal wounds and broken bones; poultices were made for external wounds and tea was consumed for internal ailments (Teynor *et al.*, 1992). Comfrey leaves is used in making salad as a rich source of protein and vitamin B₁₂. Comfrey like most green vegetables is deficient in methionine and low in phenylalanine (Teynor *et al.*, 1992). Comfrey leaves is very high in crude protein (between 21 to 31% crude protein) and is a forage of high digestibility (between 37 to 77%) (Robert, 1983). The fresh leaves of comfrey are eaten by sheep, pigs and poultry but are often not palatable for cattle and rabbits (Robert, 1983). Cattle and rabbits enjoy leaves when the forage is wilted or ensiled. The forage is also fed

to horses, goats, chinchillas and caged birds. Robert (1983) also reported that comfrey contains 18 amino acids – Glutamic acid, (12.49%); Leucine, (10.04%); Aspartic acid, (9.34%); Valine, (7.02%); Arginine, (6.37%); Phenylalanine, (6.34%); Alanine, (5.79%); Proline, (5.45%); Glycine, (5.37%); Isoleucine, (5.31%); Lysine, (5.21%); Cystine, (4.67%); Threonine, (4.51%); Tyrosine, (3.74%); Serine, (2.96%); Methionine, (2.46%); Histidine, (2.02%); Tryptophan, (0.91%). The comfrey is particularly high in K and is higher than many other forage crops in Ca, P, Fe and Cu (Robert, 1983). It is also rich in minerals such as Potassium, (5.86%); Nitrogen, (3.53%); Calcium, (1.44%); Phosphorus, (0.50%); Magnesium, (0.30%); Aluminum, (385parts/million); Iron, (364parts/million); Manganese, (116parts/million); Sodium, (70 parts/million); Zinc, (45parts/million); Nickel, (2parts/million); Chromium, (1part/million); Cadmium, (<1part/million). Despite the nutritional benefits of comfrey, it has toxic or anti-nutritional factors such as pyrrolizidine alkaloids (which is carcinogenic) and allantoin. Huxtable *et al.* (1986) cited cases of hepatic veno-occlusive disease that were produced by using pyrrolizidine alkaloid capsules in USA. Comfrey has been reported to cause liver damage and cancerous tumours in rats (Teynor *et al.*, 1992). Alkaloids extracted from Quaker comfrey leaves and injected in rats at dosage of 9-71mg of alkaloid/kg body weight three times per week for several weeks caused liver damage or death (Culvenor *et al.* 1980). Allantoin chemical stimulates cell production and promotes wound healing. The allantoin (C₄H₆N₄O₃) concentration in comfrey particularly the root, has led to its extensive use in natural and folk medicine for promoting healings of wounds, ulcers, broken bones, swellings and burns (Hills, 1976). In Nigeria, families and herbal doctors are still harnessing the benefits of comfrey leaf for treatment of various ailments, and as a rich source of vegetable and blood tonic. There is dearth of information on the nutritional and health benefit of comfrey leaves by researchers and its potential toxic risks in Nigeria. This research is basically to explore the rich nutritional value of comfrey leaves extract on broiler finisher chickens performance and its effect on some blood indices.

Therefore, the aim of this research is to investigate the effect of comfrey symphytum leaves extract on the growth performance, Histopathology, haematological and serum biochemical indices of finisher broiler chickens.

MATERIALS AND METHOD

Experimental site

This experiment was carried out at the poultry unit of teaching and research farm, Imo State University, Owerri, which is located within the Southern-Eastern agro ecological zone of Nigeria. Owerri lies between latitude 5^o29'North and longitude 7^o20'East. It is about 91m above sea level with annual rainfall, temperature and humidity ranging from 1,500mm to 2,200mm, 20.0 - 27.5^oc and 75 - 90% respectively (Accuweather, 2015).

Source and processing of comfrey symphytum leaves extract

The comfrey plant was planted in Imo State University teaching and research farm Owerri. 200g of the fresh leaves were squeezed in 1 liter of water with a juicer to produce comfrey symphytum leaves extract. The comfrey leaves extract were mixed in drinking water at the rate of 0ml/liter of H₂O, 50ml of extract/liter of H₂O, 100ml of extract/liter of H₂O and 150ml of extract/liter of H₂O. Initially, the comfrey leaves meal were dried and sent to the laboratory for proximate, phytochemical and mineral analysis according to AOAC (2010).

Experimental diets

The birds were fed normal broiler finisher diet containing 19% crude protein and 2900kcal/kg of energy as shown in Tables 1. T₁ was the control and contained no comfrey leaves extract. T₂ contained conventional supplement (Divertamin) and used at the factory recommended level (4g to 4litres of H₂O). T₃, T₄ and T₅ contained 50ml comfrey leaves extract/liter of H₂O, 100ml of comfrey leaves extract/liter of H₂O and 150ml of comfrey leaves extract/liter of water, respectively. The Divertamin contained multivitamine, electrolyte and amino acids and was used at the factory recommended level. The Divertamin used contained Vitamin A, 375000 IU; Vitamin D₃, 110000 IU; Vitamin E, 1000 IU; Vitamin K₃, 4350 mg; Vitamin B₂, 4350 mg; Vitamin B₆, 2350 mg; Vitamin B₁₂, 11350 mg; Vitamin C, 10000 mg; Nicotinic acid, 16700 mg; DL-Methionine, 10000 mg; L-lysine, 15000 mg; Potassium chloride, 87000 mg; Manganese sulphate, 12000 mg; Sodium sulphate, 212000 mg; Sodium Chloride, 50000 mg; Copper sulphate, 12000 mg; Zinc sulphate, 12000 mg. It is used in poultry to achieve optimum growth and production in the management of stress related to high temperature etc. It is indicated in the treatment of vitamin and mineral deficiency and in the management of stressful condition.

Table 1: Ingredient and calculated nutrient composition of the experimental diet

Ingredient	Quantity
Maize	60.00
Soya bean	20.00
Fish meal	2.00
Blood meal	1.00
Palm kernel cake	4.25
Palm oil	1.0
Wheat offal	8.00
Bone meal	3.00
Salt	0.25
Vitamin premix	0.25
Lysine	0.15
Methionine	0.10
Calculated nutrient composition	
Crude protein (%)	19.00
ME kcal/g (%)	2926.33
Ether extract (%)	13.95

Ash (%)	3.08
Ca (%)	1.28
P (%)	0.97
Lysine (%)	1.08
Methionine (%)	0.70

Experimental birds and design

One fifty (150) day old Agritech broiler chicks were purchased from a certified poultry vendor in Owerri. The chicks were brooded for four weeks. Thereafter, the birds were divided into five groups of 30 birds each in a completely randomized design. Each group was further divided into three replicates of 10 birds each. Each of the groups was assigned to one of the comfrey leaves extract. All the treatment groups were fed same normal broiler finisher diet. The initial live weights of the birds were taken and weekly thereafter. The birds were fed *ad libitum*. The birds in each treatment were served six (6) litres of the comfrey leaves extract mixture per day for the comfrey groups, six (6) litres of the Divertamin mixture per day for the Divertamin group and normal drinking water for the control. Clean fresh drinking water was supplied to the birds after the consumption of the comfrey leaves extract and the Divertamin water. The trial lasted for four weeks.

Data collection

Feed intake was recorded daily and the birds were weighed weekly after the initial body weights were taken. Feed intake was determined by weighing the feed offered and the left over the next morning. The difference between the two values was the feed consumed. Feed conversion ratio was determined by dividing average daily feed intake by average daily body weight gain.

Economics of production

Economic parameters determined were; Average weight changes; Average daily weight gain, Average daily feed intake, cost/kg weight gain, cost of total feed consumed

Cost of production (₦) = cost/kg weight gain multiplied by average weight changes.

Price/kg meat (₦) = price of selling one kg of meat

Revenue (₦) = price/kg meat multiplied by average weight change

Gross margin (gain) = Revenue minus cost of production

Haematological and serum indices studies

At the last day of the feeding trial, three birds per treatment were randomly selected to determine their haematological and serum indices. 5ml blood samples were collected from the wing vein of the birds using syring and needle and placed in the specimen bottles with EDTA (Ethylene Diamine Tetra Acetate) for haematological studies. Blood was analysed within three hours of collection for haemoglobin(HB), packed cell volume(PCV), red blood cell(RBC), mean cell volume(MCV), mean cell haemoglobin(MCH), mean cell haemoglobin concentration(MCHC), and white blood cell(WBC) as outlined by Ochie and Kolhatkar (2000). Another 5ml of blood samples were collected and placed in the specimen bottles without EDTA for serum biochemical analysis. Blood samples placed in the specimen bottles without EDTA were used to analyse the serum biochemical parameters such as urea, total protein, creatinine, cholesterol, serum electrolytes and liver enzymes as outlined by Ochie and Kolhatkar (2000).

Histological Studies

Histological studies were carried out according to Baker *et al.* (1989). Excised organs were fixed in 10% formal saline for 24 hours and dehydrated in ascending grades of alcohol (70, 80 and 90% absolute) and de-alcoholized in xylene for 30 minutes. The tissues were impregnated in molten paraffin wax and subsequently embedded using disposable plastic embedding moulds. The embedded tissues were sectioned with haematoxylin and eosin (H/E) staining procedure. The sections were examined under microscope (DM 500 leica binocular microscope) and observations noted. The examined slides were photo-graphed with leica DM 500 binocular microscope with photomicrographic accessories.

Statistical analysis

Data collected were subjected to analysis of variance using the SPSS software (2012). Where analysis of variance indicated significant treatment effects, means were compared using Duncan's New Multiple Range Test (DNMRT) (SPSS, 2012)

Results and Discussion

Proximate, mineral and phytochemical composition of comfrey leaf meal.

Proximate, mineral and phytochemical composition of comfrey leaf meal is presented in Table 2. The values are similar to the values reported by Esiegwu and Obi (2021) in a similar experiment with broiler starter chicks.

Performance of finisher broilers offered comfrey leaves extract

Data on the performance of finisher broilers offered comfrey leaves extract are presented in Tables (3). There were no significant treatment effect ($P>0.05$) on the average initial weight, average final weight and average daily feed intake. Significant treatment effect ($P<0.05$) were recorded on average weight changes, average daily weight gain and feed conversion ratio. The average daily feed intake which showed no significant treatment effect ($P>0.05$) implied that the anti nutritional factors (ANF) in comfrey leaves extract did not depress the appetite of the birds. It could suggest that the ANF present in the juice extract were within tolerable level. The average weight changes, average daily weight gain and feed conversion ratio showed significant differences ($P<0.05$). However, T_5 was

significantly increased ($P < 0.05$) compared to T_3 but similar statistically ($P > 0.05$) to the rest treatments. The comfrey leaves extract at T_5 (150ml extract/liter of H_2O) gave better feed conversion ratio but statistically similar to the control (T_1) and the Divertamin group (T_2). The feed conversion ratio was lower than the value 3.33 to 3.70 for finisher broilers offered *Moringa oleifera* leaf water (Esiegwu, 2019) but within the range 2.07 to 2.56 for broiler chickens fed varying levels of *Seasum indicum* leaves (Adedeji, 2019) and higher than the reference value 1.7 to 2 for broilers (Ghosh, 2015). The performance of T_5 (150ml comfrey extract/liter of H_2O) for Average weight changes (1876.67g), average daily weight gain (65.719) and feed conversion ratio (2.08) was best across treatments indicating effective metabolism and utilization of amino acids in the comfrey leaves extract. The heavier body weight gain of broilers offered comfrey leaves extract could be attributed to a likely higher intake of amino acids (Tijani *et al.*, 2016). This shows that the inclusion or use of comfrey leaves extract improves performance considerable. Comfrey leaves extract enhanced performance better than conventional supplement which could be due to improved digestibility and nutrient utilization.

Economics of production

Data on economics of production are shown in Tables (4). Cost of feed (₦220.11) was increased at T_2 (Divertamin group). This was attributed to the high cost of conventional supplements. Cost per kg weight gain was also highest at T_2 (₦515.06) compared to other treatments as a result of high cost of feed and poor feed conversion ratio. Cost of production rose very high above others at T_2 due to high cost per kg weight gain and relatively low average weight changes. T_5 (150ml extract/liter of H_2O) revealed a higher revenue earnings (₦2439.67) and better gross margin (profit) (₦1756.56). This was attributed to better performance of the comfrey leaves extract in producing heavier average weight changes and high feed conversion ratio (low value for feed conversion ratio). In financial terms, T_5 revealed a lower cost per kg weight gain, higher revenue (₦2439.67) and higher gross margin (₦1756.56) comparatively. In the market every money counts. The factor responsible for the financial merit of T_5 (150ml extract/liter of H_2O) were better value of feed conversion ratio due to the efficacy of the non conventional supplement (comfrey leaves extract), reduced cost of comfrey leaves extract and heavier body weight change. It could be concluded from economics of production that comfrey leaves extract gave heavier body weight gain, low value for feed conversion ratio leading to lower cost per kg weight gain, higher revenue and gross margin (profit) earning. Therefore, it is more economical to use some natural sources of nutrient supplement such as comfrey leaves extract to improve performance of broilers and have higher returns on investment due to cost effectiveness.

Haematological and serum biochemical indices

Data on the haematological and serum biochemical indices of broiler finishers offered comfrey leaves extract are presented in Tables 5 and 6. All the haematological indices analysed did not show any significant difference ($P > 0.05$) across treatments. Haemoglobin (Hb) did not show any significant treatment effect ($P > 0.05$), however the values were within normal reference

Table 2: Proximate, mineral and phytochemical composition of comfrey leaf meal.

PARAMETERS	QUANTITY (%Dm)
Moisture	9.90
ash	6.90
Protein	18.37
Ether extract	5.25
Fiber	47.12
Nitrogen free extract	11.87
MINERALS(Mg/Kg)	
Cu	47.85mg/kg
Zn	374.2 mg/kg
Mn	41.85 mg/kg
Fe	349.65 mg/kg
Ca	3242.2 mg/kg
Mg	262.8 mg/kg
Na	2433.4 mg/kg
K	2568.5 mg/kg
PHYTOCHEMICALS	
Alkaloid (% DM)	5.40
Flavonoid (% DM)	18.36
Saponin (% DM)	14.95
Tannin (% DM)	0.543

Table 3. Performance characteristics of broiler finisher chickens Offered comfrey leaves extract.

PARAMETER	T ₁	T ₂	T ₃	T ₄	T ₅	SEM
Average Initial weight (g)	479.33	470.00	478.00	479.00	476.67	17.45
Average final weight (g)	2286.67	2126.67	2038.67	2299.33	2353.33	94.62
Average weight changes (g)	1807.34 ^{ab}	1656.67 ^{ab}	1560.67 ^b	1820.00 ^{ab}	1876.67 ^a	86.79
Average daily weight gain (g)	64.55 ^{ab}	59.17 ^{ab}	55.74 ^b	65.01 ^{ab}	67.02 ^a	3.02
Average daily feed intake (g)	135.23	133.19	139.07	133.13	134.22	3.35
Feed conversion ratio	2.09 ^{ab}	2.25 ^{ab}	2.49 ^a	2.05 ^b	2.00 ^b	0.14

Ab means within the same row with different superscripts are significantly different (P<0.05).

T₁ = Control (Normal water, 0ml/liter of H₂O); T₂ = conventional supplement (Divertamin)

T₃=50ml extract/liter of H₂O; T₄=100ml extract/liter of H₂O; T₅=150ml extract/liter of H₂O

Table 4: Economics of production of broiler finishers offered comfrey leaves extract.

Parameters	T ₁	T ₂	T ₃	T ₄	T ₅
Cost/kg of feed (₦)	170.20	220.11	173.01	174.03	175.00
Cost of feed consumed (₦)	670.62	853.28	707.43	674.64	683.11
Cost/kg weight gain (₦)	371.06	515.06	453.29	370.68	36.40
Cost of production (₦)	670.62	853.28	707.43	674.64	683.11
Revenue (₦)	2349.54	2153.67	2020.87	2366.00	2439.67
Gross margin (₦)	1678.95	1300.39	1313.43	1691.36	1756.56

Note: Selling Price per kg meat is ₦1300

Range, 7-13g/100ml (Banergee, 1998 and Jain, 1989) and 6.5-9g/100ml (Swenson, 1977). The packed cell volume (PCV) were not significantly different (P>0.05) across treatments but fall within the normal reference value 25-45% (Banergee 1998); 22-35% (Jain 1989) and 30-33% or 35-40% for male chickens (Swenson, 1977). The red blood cells (RBC) were also within normal reference value 7-1.2 X 10¹²/l (Jain, 1989). Awodi *et al.* (2005) and Chineke *et al.* (2006) reported that packed cell volume, haemoglobin and mean cell haemoglobin are major indices for evaluating circulating avian erythrocytes and are very useful in the diagnosis of anaemia and also serve as a useful indices of the bone marrow capacity to produce red blood cells as in mammals. The values of the PCV, Hb, and RBC showed that the birds were not anaemic and the bone marrow was intact. The comfrey leaves extract supplied enough Fe and proteins for the haemoglobin to circulate adequate erythrocytes in the blood. The comfrey leaves extract did not indicate any deleterious effect on the haematological indices of the birds as the values did not fall

below the normal reference range. Kepeme *et al.* (2011) reported that when the PCV values are below the normal range, the chickens were anaemic. The mean cell volume (MCV), mean cell haemoglobin (MCH) and mean cell haemoglobin concentration (MCHC) were not significantly different ($P>0.05$). The MCHC fall within normal range (Jain, 1989). This non significant treatment ($P>0.05$) effect on these indices implies that the production of red blood cells by the bone marrow was adequate across treatment. White blood cell fall within normal range $9-31 \times 10^3/\text{mm}^3$ (Banergee, 1998). The white blood cell differentials were statistically similar $P>0.05$. This suggests that there was no toxin or infection arising from the ingestion of the comfrey leaves extract. It has been reported that leukocyte counts as well as heterophil and lymphocytes ratio were used as indicators of stress responses and sensitive bio markers that are crucial to immune functions (Shaniko, 2003). The white blood cells and the differentials are good indicators of infection or presence of toxin in the blood.

Table 5: Haematological indices of broiler finishers offered comfrey leaves extract

PARAMETER	T ₁	T ₂	T ₃	T ₄	T ₅	SEM
Haemoglobin(g/dl)	12.77	13.00	12.90	12.63	12.60	0.24
Packed cell volume (%)	39.67	41.33	40.00	38.00	37.67	1.98
White blood cell(t)	11.60	11.93	11.33	11.60	11.40	0.21
Neutrophil%	54.67	56.33	53.33	55.00	53.33	1.83
Eosinophil%	1.33 ^{ab}	1.00 ^b	1.33 ^{ab}	1.00 ^b	2.00 ^a	0.21
Basophil%	0.00	0.00	0.00	0.00	0.00	0.00
Lymphocytes%	42.67	41.33	43.33	42.00	43.00	1.71
Monocytes%	1.33	1.33	2.00	2.00	1.67	0.26
Red blood cell ($\times 10^{12}/\text{L}$)	12.90	13.13	13.13	12.63	12.77	0.24
ESR($\text{MM}^3/1^{\text{st}}\text{hour}$)	26.67	23.33	26.67	33.33	36.67	6.15
MCV(PI)	30.73	31.47	30.43	30.03	29.43	0.99
MCH(Pg)	9.87 ^b	9.90 ^{ab}	9.83 ^b	10.00 ^a	9.83 ^b	0.04
MCH(g/dl)	32.27	31.57	32.27	33.33	33.67	1.07

Ab means within the same row with different superscripts are significantly different ($P<0.05$).

The number of the WBC increases in acute infection or toxicity. Eosinophil increases in allergy and in parasitic infection (Banergee, 1998). The white blood cells showed that the comfrey leaves extract had no deleterious effect on the blood of the birds. The biochemical indices, urea, creatinine, cholesterol, total proteins and the liver enzymes (alkaline phosphate (ALK), serum glutamic oxaloacetic transaminase (SGOT) and serum glutamic pyruvic transaminase (SGPT)), showed no significant difference ($P>0.05$) across treatments. Serum urea was statistically similar ($P>0.05$) across treatments. Serum urea is a strong indices used in monitoring protein quality and the extent of amino acid balance. It has been reported that high level of serum urea was an indication of low protein quality as a result of imbalance of amino acids (Nworgu, *et al.*, 2007). The non-significant difference ($P>0.05$) of the serum urea was a pointer to normal serum protein quality which means that the comfrey leaves extract did not import any contamination or exact any negative effect to protein-amino acid metabolism. Total protein, albumin and globulin were not affected by treatment significantly ($P>0.05$). The non-significance difference ($P>0.05$) of the

protein is a pointer to the balance of nutrients, consumption and metabolism of adequate protein and amino acids by the birds as seen in the performance indices. The protein needs of the birds across treatments were met. The non-significant difference of the globulin and albumin across treatments was a pointer to intact immune system and healthy birds.

Table 6: Biochemical Indices of broiler finishers offered comfrey leaves extract

PARAMETERS	T ₁	T ₂	T ₃	T ₄	T ₅	SEM
Urea (mg/100ml)	6.83	7.20	7.10	6.67	6.77	0.34
Creatinine (mg/100ml)	62.67	65.67	63.67	59.33	60.67	3.34
Cholesterol (mmol/L)	8.27	8.63	8.47	7.93	8.13	0.37
Total protein (mg/100ml)	60.33	63.00	61.33	58.33	59.33	2.86
Albumin	23.00	23.67	22.33	21.33	22.00	1.53
Globulin	37.33	39.33	39.00	37.00	37.33	1.95
Sodium Na ⁺ (mmol/L)	41.33	42.00	42.33	40.33	41.33	1.08
Potassium K ⁺ (mmol/L)	1.27	1.27	1.23	1.23	1.23	0.04
HCO ₃ ⁻ (mmol/L)	11.27	11.33	11.10	11.17	11.13	0.14
Chloride Cl ⁻	55.67 ^a	22.67 ^a	23.00 ^a	22.00 ^a	22.33 ^a	26.31
ALK (iu/L)	1.23 ^{ab}	1.37 ^a	1.23 ^{ab}	1.20 ^b	1.30 ^{ab}	0.04
SGOT (iu/L)	11.23	11.40	11.30	11.20	11.23	0.14
SGPT (iu/L)	6.47	6.67	6.73	6.73	6.60	0.16

Ab means within the same row with different superscripts are significantly different (P<0.05).

Sanchez Monge *et al.* (2004) reported that increased globulins are seen in chronic infections, liver damage and kidney dysfunction. Serum albumin is a strong predictor of health; a low albumin concentration is a sign of poor health (Kastow, 2009). Serum urea was similar statistically (P>0.05) across treatments. Serum urea is an indices used to measure or assess protein quality. It has been reported that high serum urea was sign of protein insufficiency (Esiegwu, 2019). The non significant (P>0.05) values of the urea were an indication of good quality protein and non interference on the amino acid metabolism. Aletor and Egberongbe (1992) reported that the lower the value of creatinine the higher the protein quality. Ross *et al.* (1978) also reported that abnormal high blood creatinine would indicate muscle wastage and imply that the animal was surviving at the expense of body reserves which also results in weight loss. The non-significant value (P>0.05) of the creatinine was a sign of nutritional adequacy of protein in both quality and quantity. The cholesterol values were not significantly different (P>0.05). This was an indication of proper fat metabolism and utilization. Fat was adequately mobilized to the tissues and utilized for all the treatments. In order words, there was no interference on fat metabolism by any anti-nutritional factor. The SGOT and SGPT were not significantly different. Alk had a significant difference. T₂ (Divertamine group) was significantly increased compared to T₄ (100ml extract/liter of H₂O) but statistically similar to the control. T₁ (control), T₃ (50ml extract/liter of H₂O) and T₅ (150ml extract/liter of H₂O) were similar (P>0.05) statistically. Ukpabi *et al.* (2015) reported that serum enzyme activities are used for checking toxicity as well as monitoring protein quality and damages done to the liver. The

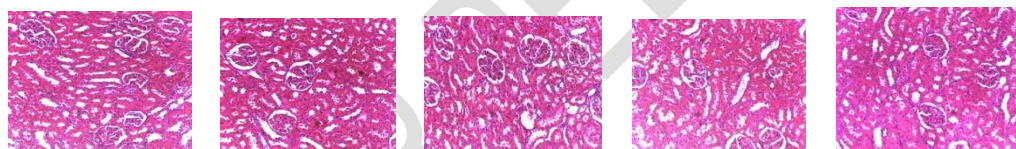
enzymes (ALK, SGPT, SGOT), were statistically similar to the control. It was a sign that there was no toxic effect on the blood and liver of the birds. That means, the comfrey leaves extract did not impact any negative or deleterious effect on the blood of the birds.

Histopathological observations on some organs

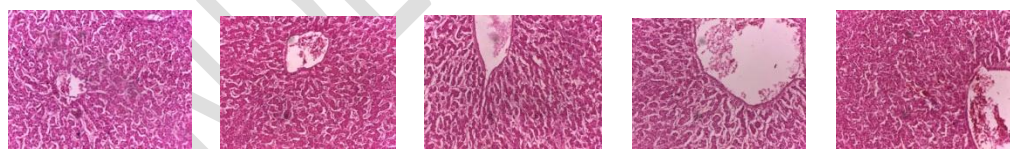
The histopathological observations on the organs are shown on plates 1 to 5 of each organ. The histology section of the heart from Plate (P) 1:T₁ to Plate (P) 5: T₅ showed cardiac muscle with unremarkable myocyte nuclei, muscle fibre and myocardium. There was no observable histologic changes in the heart across treatments. The heart was normal indicating that the comfrey leaves extract was not toxic to the organ. The kidney tissue did not show any histologic change in architecture. Histology section showed kidney tissue with unremarkable glomeuli, bowmans capsule, tubules tufts and stroma. This implies that the comfrey leaves extract had no deleterious effect on the kidney cells within the dosage level used.



P 1: Heart T₁ P 2: Heart T₂ P 3: Heart T₃ P 4: Heart T₄ P 5: Heart T₅



P 1: Kidney T₁ P 2: Kidney T₂ P 3: Kidney T₃ P 4: Kidney T₄ P 5: Kidney T₅



P 1: Liver T₁ P 2: Liver T₂ P 3: Liver T₃ P 4: Liver T₄ P 5: Liver T₅

Note: P means Plate

Histology section of the liver tissue for T₁ and T₂ showed hepatic tissue with unremarkable central vein, sinuoids and lamina of hepatocytes. The hepatocytes and other stroma elements appear normal. T₃ and T₄ (Plate 3 and 4) revealed hepatic tissue with unremarkable central vein

and effaced tissue stromal elements. T₅ (plate 5) showed effaced tissue stromal elements with congestion and marked cellularity. The effaced tissue stromal elements, congestion and marked cellularity observed in T₃, T₄ and T₅ were signs of an onset of toxicity due to the anti nutrients contained in the comfrey leaves extract especially alkaloids. The findings support the report of Culvenor *et al.* (1980) that alkaloids extracted from Quaker comfrey leaves and injected in rats at dosage of 9-71mg of alkaloid/kg body weight three times per week for several weeks caused liver damage or death and Teynor *et al.* (1992) reported that comfrey caused liver damage and cancerous tumours in rats.

CONCLUSION

This study showed that the use of comfrey leaves extract as a nutritional supplement in broiler finisher was beneficial in improving performance at 150ml of comfrey leaves extract/litre of H₂O. It also reduced the cost/kg weight gain, increased the revenue earnings and gross margin (profit). The study also showed that there was no side effect, toxin or contamination of the blood as revealed on the haematological and serum biochemical indices of the bird. Histologically, the study revealed that comfrey leaves extract caused effaced tissue stromal elements, congestion and marked cellularity of the liver. It was therefore recommended that 150ml of comfrey leaves extract per liter of H₂O should be used for oral supplementation on broiler finisher diet.

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