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Evaluation of Ethanol Extract of *Lagenaria breviflora* as a Replacement for Antibiotics in Broiler Chicken Production

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A B S T R A C T

An eight week study was conducted to evaluate the effect of oral administration of ethanol extract of *Lagenaria breviflora* (LB) fruit as an additive on the performance, carcass and sensory characteristics of broiler chicken meat avoid the problems of accumulation of antibiotic residue in poultry products and resistance of pathogenic organisms to antibiotics. Two hundred (200) day- old broiler chickens were randomly divided into five groups of forty birds each and assigned to five treatments in a Completely Randomized Design experiment consisting of four replicates per treatment. The treatments were Neoceryl antibiotics (T1), distil water/untreated (T2), 1ml (T3), 2 ml (T4) and 3ml (T5) ethanol extract of LB. Data were collected on production performance, nutrient digestibility, carcass and meat sensory characteristics. Data were analyzed by one way analysis of variance at 5 % probability level. Results showed that average daily gain of the birds that were given 1ml (39.07g), 2ml (41.76g) and 3ml (41.32g) LB were higher ($P<0.05$) than that of the group that was not treated (36.56 g) while the value obtained in the group given Neoceryl (41.82g) was similar to those given LB. Dry matter and crude protein digestibility were higher ($P<0.05$) in birds that were given LB than in those that were not medicated (T2) while those that were treated with Neoceryl had similar values. Feed intake, feed cost/kg weight gain, mortality and FCR were lower ($P<0.05$) and better in birds that were given LB extract than those that were not treated. Primal cuts, organ weights and sensory characteristics were not affected by LB. In conclusion, 2ml and 3ml ethanol extract of LB can be used as an additive in place of commercial antibiotic in broiler chicken production in the interest of public health.

Introduction

The use of antibiotics either for prophylactic or therapeutic effect or as growth promoter is a conventional management practice in livestock industry since 1830s when it was discovered. This practice has revolutionize livestock industry because they boost the productivity of the animals, control subclinical level of

infection in the animal and improve efficiency of feed utilization by the animal (Rushton, 2015). However, the high incident of antibiotic resistant bacteria are becoming more common; a situation that has been attributed to indiscriminate use of antibiotics in livestock industry as growth promoter (Lekshmi *et al.,* 2017). In the recent years, very few new antibiotics are been developed while pathogenic organisms are developing resistance to the old ones at a faster rate. This has led many European countries to place ban on the use of antibiotics as growth promoter and the restriction of some antibiotics from use in animal treatment. Moreover, the premium placed on organic food by many consumers of animal products call for alternatives to the use of antibiotic (Kumar *et al.,* 2014).

The greatest challenges facing livestock producers today is how to sustain the present level of production without the use of antibiotic growth promoters. Researchers have therefore directed their efforts into evaluating various plants products as additives for use in livestock industry. Phyto- additives are plant parts or their extracts that contain phyto- chemicals that are able to destroy or arrest the growth of pathogenic organisms. Such plants contain secondary metabolites in the class of isoprene derivatives, flavonoids and glucosinolate which have been suggested to act as antibiotics or as antioxidants (Huang *et al.,* 2022). Phytogenic plants that have received the attention of researchers include cinnamon, cumin, ginger, garlic, turmeric, black pepper, alligator pepper, thyme, wild colocynth (*Lagenaria breviflora)*.

Lagenaria breviflora (LB) also called wild colocynth is one of the medicinal plant that is native to most tropical environment including Nigeria. The plant belongs to the family of Cucurbitacaea and has been reported to have broad spectrum antibacterial (Tomori *et al.,* 2007) and antiviral activities. This plant contains different phytochemicals which makes it a potential treatment for methicillin-resistant staphylococcus and other known resistant strain of bacteria (Shan *et al.,* 2002). In traditional medicine, LB has been reported to be used for the treatment of small pox, measles, cold and schistosomiasis in man (Ajayi *et al.,* 2002; Oladunmoye and Kehinde, 2011). It has also been reported to possess haematinic and immune-stimulatory activities (Saba *et al.,* 2009; Onasanwo *et al.,* 2011). It's used in Ethinoveterinary medicine for the treatment of Newcastle disease and coccidiosis has been reported (Oridupa *et al.,* 2011). The extract of the fruit of LB was reported to improve the health status of Yaffa brown pullets (Ekunseitan *et al.,* 2017). Addition of aqueous extract of the fruit of LB has also been reported to improve production performance, health status and survivability of broiler chicken (Oladunjoye *et al.,* 2023).

This study was conducted to evaluate the effect of addition of ethanolic extract of *Lagenarian breviflora* to drinking water of broiler chicken on the production performance, blood, organs, meat, and carcass characteristics of broiler chicken.

Materials and Methods

Experimental Site

The study was carried out at the Poultry Brooder Unit, Teaching and Research Farm, Ladoke Akintola University of Technology, Ogbomoso, Oyo State, Nigeria. The site is in the derived savanna zone of South Western part of Nigeria and lies on latitude $8^0$8'31.7974 North of the equator and Longitude $4^0$14'426696" East of the Greenwich Meridian. The altitude is between 300m and 600m above the sea level while the mean temperature and annual rainfall are 27^0C and 1247mm respectively (Google Earth Map, 2021).

Preparation of Ethanolic extract of Lagenaria breviflora

Mature fruits of wild colocynth (*Lagenaria breviflora* were sourced from fallow land within the university community. They were cut into pieces and air dried under shade until it was about 10% moisture content. The dried product was milled to powdery form and was taken to laboratory for extraction. Fifty (50) grams of the meal was mixed with two hundred and fifty milliliters of ethanol in a conical flask and stirred together properly. The mixture was tightly covered, kept for 48 hours and then sieved with muslin cloth to collect the filtrate. The filtrate was then poured into a cleaned conical flask and placed on a settled electric heating mantle to allow ethanol to be recovered from the filtrate (90%) into a separate flask. The extract was then stored in a plastic container and stored in the refrigerator at 4^0C until it was used.

Feed Preparation

Birds were fed conventional diets at starter and finisher phases. The composition of the diets is shown in Table 1.

Birds and Management

Two hundred day-old broiler chicks of Abor Acre strain (average 42± 3.5g) were used for the study. The birds were divided into five groups of forty birds each and the groups randomly assigned to five medication treatments in a Completely Randomized Design (CRD) experiment consisting of four replicates of 10 birds in each replicate. The treatments consists of positive control experiment in which the birds were given Neoceryl antibiotic (T1), negative control experiment (T2) in which the birds were not medicated but ordinary water was added to their drinking water, the group that was medicated with 1ml of ethanol extract of LB in 10 ml of water (T3), the group that was medicated with 2ml of ethanol extract (T4) and the one that was medicated with 3ml of the extract (T5). The birds were medication at week 3, week 5 and week 7 of the study. The conduct of the experiment followed the ethical rule governing animal experiments in the university. Birds were housed in deep litter pens measuring 1.4m × 1.2m and were offered feed and water *ad libitum* throughout the eight weeks duration of the study.

Data Collection

Data were collected on feed intake, weight gain, while feed to gain ratio was determined from feed intake and weight gain.

Feed intake: A known quantity of feed was weighed and offered to the birds in each replicate at 8.00am daily while left over was measured and recorded. Feed consumed was then estimated as the difference between feed offered and the left over.

Feed Intake per bird = Feed offered –Feed leftover/Number of birds housed

Weight Gain: Birds were weighed at the beginning of the experiment and at the end of the experiment using digital weighing scale. Weight of the birds per day was then determined from the two as shown below:

Weight Gain (bird/day) = Final weight – Initial weight /Number of the birds weighed*56 days

Feed Conversion Ratio: This was calculated as the total feed consumed divided by total weight gain

Feed: Gain ratio = Total feed intake/Total weight gain

Mortality: The number of birds lost throughout the period of the experiment in a replicate was recorded and expressed as the percentage of the birds housed at the beginning of the experiment.

Mortality (%) = Number of the birds lost/Number of the birds housed*100

Feed cost per kilogram and Feed cost per kilogram live weight gain: Feed cost per kilogram was estimated from the cost of each ingredient used in the preparation of the feed while feed cost per kilogram live weight gain was estimated from feed cost per kilogram and feed: gain ratio.

Metabolic Trial

Metabolic trial was conducted using 2 birds per replicate (8 birds per treatment). The birds were housed in metabolic cages equipped with feeder, drinkers and faecal trays. Birds were served known quantity of feed at 7.30 am in the morning and the leftover weighed the following morning at the same time to allow for the estimation of feed intake. Birds were allowed 3 days pre-collection period to acclimatize to the cage environment. Faecal collection was carried out for three days. The faecal samples collected were weighed using digital weighing scale and then oven-dried at 70^0Cfor 48 hours. Samples collected from the same treatment were bulked, mixed together and representative samples stored in sealed bottles until it was analyzed.

Apparent nutrient digestibility was calculated using the formula:

$$\text{Apparent Nutrient Digestibility (\%)} = \frac{\text{Nutrient in the Feed (gDM)} - \text{Nutrient in the Faeces (gDM)}}{\text{Nutrient in the Feed (gDM)}} * 100$$

Carcass Evaluation

Two birds per replicate (8 bird per treatment) were selected for carcass evaluation at the end of the study. The birds were fasted for 24 hours, tagged for easy identification, weighed, bled, cleaned of the feather, eviscerated and dressed. Live weight, bled weight, defeathered weights and dressed weight were taken using digital weighing scale which were expressed as the percentage of the live weight of the birds. Internal

organs: gizzard, proventriculus, kidneys, liver, pancreas and caeca were carefully excised, weighed and expressed as the percentage of the live weight. Carcasses were also cut into primal cuts (Neck, breast, back, thigh, drum stick, wings, shanks, head and abdominal fat pad), weighed and the weights expressed as percentage of live weight.

Sensory Evaluation

Twelve trained individuals were used for sensory evaluation. The panelists consisted of male (N =6) and females (N= 6) with their age ranged between 18 and 25 years. Samples of breast muscles collected from birds selected from each treatment were washed in clean water, tagged properly and packed in transparent polythene bags for proper identification. They were thereafter boiled in water bath for 20 minutes at 80^0C. The meat samples were allowed to cool and then served to the panelists for sensory evaluation using the nine-point hedonic scale according to the procedure of AMSA (1995). Meat were graded from point 1 to 9, where 1 refers to extremely liked and 9 refers to extremely dislike. Each panelist was allowed to masticate one sample per treatment and rate his/her preference for the followings: Colour, texture, flavour, juiciness, tenderness and overall acceptability.

Laboratory Analysis

Feed and faecal samples from each treatment were analyzed for proximate composition according to the methods of AOAC (1990).

Data Analysis

Data were analyzed by One-way Analysis of Variance using SAS statistical software package (SAS, 1990) at 5% probability level and where significance were indicated, Duncan's option of the same statistical software was used to separate the means.

Results and Discussion

The performance of the broiler chicken medicated with ethanol extract of wild colocynth (LB) is shown in table 2. Final weight, Total weight gain and Average daily gain were similar in birds that were medicated with either Neoceryl or ethanol extract of *Lagenaria breviflora* (LB). Birds that were medicated (T, T3, T4 and T5) had higher ($p<0.05$) values in these parameters than those that were not medicated (T2). Feed intake was higher ($P<0.05$) in birds that were not medicated than in those that were medicated either with Neoceryl or ethanol extract of *LB*. Feed conversion ratio was better and lower ($P<0.05$) in the birds that were medicated with Neoceryl, 1ml extract, 2ml extract and 3ml extract of LB compared with those that were not medicated. Mortality was lower ($P<0.05$) in the birds that were medicated either with Neoceryl antibiotic or ethanol extract of LB. Feed cost per kilogram weight gain was also lower ($P<0.05$) in birds medicated with ethanol extract or Neoceryl compared with the group that was not medicated. Apparent nutrient digestibility of broiler chicken given ethanol extract of wild colocynth as additive is shown in table 3. No significant effect ($P<0.05$) of the extract was observed in the digestibility of crude fibre, ether extract, ash and nitrogen free extract.

Apparent digestibility of dry matter and crude protein were however significantly ($P<0.05$) affected by the extract. Apparent digestibility of dry matter and crude protein were significantly ($P<0.05$) higher in the birds that were given ethanol extract of L.B (T3, T4 and T5) compared to those that were given distil water (T2). The birds that had ethanol extract of LB however had similar dry matter and crude protein digestibility with those given Neoceryl antibiotic as additive.

The carcass characteristics of broiler chicken given ethanol extract of wild colocynth is shown in table 4. Addition of ethanol extract of wild colocynth had no significant effect ($P>0.05$) on pre-slaughter weight, carcass weight, carcass yield and the relative weights of neck, breast, back, thighs, drumsticks and wings. Relative weights of the head and shanks of the birds that were not given additive (T2) were however higher ($P<0.05$) than those given ethanol extract of L.B and Neoceryl.

The relative weight of internal organs of broiler chicken given ethanol extract of L.B is shown in table 5. No significant effect ($P>0.05$) of the additive was observed in all the parameters measured: Kidneys, liver, lungs, heart gizzard, spleen, pancreas and proventriculus.

The sensory evaluation of meat derived from broiler chicken given ethanol extract of LB is shown in table 6. Addition of ethanol extract of LB had no significant ($P>0.05$) effect on the meat colour, flavor, juiciness, tenderness and meat general acceptability.

Table.1 Gross Composition of Starter and Finisher Diet

Ingredient (%)	Starter	Finisher
Maize	45.00	47.00
Soy bean meal	18.00	16.00
Wheat offal	5.50	12.80
Corn bran	5.00	3.00
Palm kernel meal	6.0	6.75
Fish meal	3.00	2.00
Groundnut cake	13.00	7.00
Dicalcium phosphate	1.50	2.00
Oyster shell	1.95	2.50
Common salt	0.25	0.25
Lysine	0.25	0.20
Methionine	0.25	0.20
***Premix**	0.30	0.30
Total	100.00	100.00
Calculated Analysis		
****ME (Kcal/kg)**	2900	2800
Crude Protein	23.00	21.78
Crude fibre	4.20	4.0

*premix composition: Vitamin A, 200,000,00IU, Vit, D_3, 40,000,00 IU, Vitamin E (Mg) 460, Vitamin K_3(kg) 40, Vitamin B_1 (Mg) 60, Vitamin B_2 (Mg) 120, Niacin (Mg) 1,000, Calcium pantothenate (Mg) 200, Vitamin B_6 (Mg) 100, Vitamin B_{12} (Mg) 05, Folic acid (Mg), 20, Biotin (Mg) 1, Chlorine chloride (Mg) 8,000, Manganese (Mg) 2,400, Iron (Mg) 2,000, Zinc (Mg) 1,600 Copper (Mg) 170, Iodine (Mg) 30, Cobalt (Mg) 6, Selenium (Mg) 24, Anti-oxidant (Mg) 2,400.
** Calculated

Table.2 Performance of Broiler Chicken given Ethanol Extract of *Lagenaria breviflora* as an Additive

Parameter	+ve Control Antibiotics (T1)	-Control/ Untreated T2	1m Extract (T3)	2ml Extract (T4(	3ml Extract (T5)	P value
Initial Wt (g)	37.21	37.41	37.32	37.52	37.41	-
Final WT(Kg)	2.38^{a}	2.09^{c}	2.23^{b}	2.38^{a}	2.35^{a}	0.04
TWG (Kg)	2.34^{a}	2.05^{c}	2.19^{b}	2.34^{a}	2.31^{a}	0.04
ADG (g)	41.82^{a}	36.56^{c}	39.07^{b}	41.76^{a}	41.32^{a}	0.03
TFI (Kg)	4.22^{c}	4.71^{a}	4.38^{b}	4.21^{c}	4.17^{d}	0.04
ADFI (g)	75.27^{c}	84.09^{a}	78.14^{b}	75.17^{c}	74.38^{c}	0.05
FCR	1.8^{c}	2.3^{a}	2.0^{b}	1.8^{c}	1.8^{c}	0.03
Mortality (%)	5.30^{b}	16.2^{a}	6.20^{b}	6.80^{b}	6.40^{b}	0.02
Feed cost (N)	970	970	970	970	970	0.30
Feed cost/kg WT gain (N)	1746^{c}	2231^{a}	1940^{b}	1736^{c}	1746^{c}	0.30

^{abc} Means bearing different superscripts along the same row are significantly different($P<0.05$).
+ = Positive; - = Negative; NFE = Nitrogen free extract:

Table.3 Apparent Nutrient Digestibility of the Broiler Chicken given Ethanol extract of *Lagenaria breviflora* as an Additive

Parameter (%)	+ Control Antibiotics (T1)	-Control/ Untreated T2	1ml (T3)	2ml (T4(	3ml (T5)	P value
Dry matter	88.30[a]	84.10[c]	86.02[b]	88.20[a]	88.72[a]	0.03
Crude protein	83.56[a]	78.87[c]	80.20[b]	83.46[a]	83.78[a]	0.04
Crude fibre	76.32	76.43	77.01	77.33	76.92	0.12
Ether extract	88.22	88.72	88.64	88.05	88.57	0.13
Ash	91.01	92.5	90.73	91.8	90.4	0.21
NFE	74.82	76.3	75.3	74.8	76.4	0.33

[ab] Means bearing different superscripts along the same row are significantly different(P<0.05).
+ = Positive; - = Negative; NFE = Nitrogen free extract.

Table.4 Carcass Characteristics of Broiler Chicken given Ethanol Extract of L*agenaria breviflora* as an Additive

Parameter	+ Control Antibiotics (T1)	-Control/ Untreated (T2)	1ml Extract (T3)	2ml Extract (T4)	3ml Extract (T5)	P value
Pre-slaughter WT (Kg)	2.32	2.43	2.16	2.30	2.34	-
Carcass WT (Kg)	1.86	1.62	1.73	1.84	1.87	0.18
Carcass yield (%)	70.17	69.8	70.00	70.00	69.91	0.13
Head (%)	2.24[b]	3.31[a]	3.26[a]	2.25[b]	2.22[b]	0.04
Neck (%)	3.81	3.88	3.44	3.82	3.56	0.13
Shank (%)	3.84[b]	5.86[a]	4.75[a]	3.62[b]	3.82[b]	0.03
Breast (%)	23.22	23.41	24.05	23.92	23.81	0.32
Back (%)	11.05	11.32	11.21	11.15	11.32	0.32
Thigh (%)	12.14	12.06	12.01	12.25	12.18	0.20
Drumstick (%)	9.87	9.72	9.80	9.40	9.15	0.13
Wings (%)	7.32	7.48	7.60	7.80	7.50	0.32
Abdominal fat	1.37	1.34	1.35	1.36	1.32	0.10

[ab] Means bearing different superscripts along the same row are significantly different(P<0.05).
+ = Positive; - = Negative; NFE = Nitrogen free extract.

Table.5 Internal Organ Characteristics of Broiler chicken given Ethanol Extract of *Lagenaria breviflora* as an Additive

Parameter (%)	+ Control Antibiotics (T1)	-Control/ Untreated (T2)	1ml (T3)	2ml (T4)	3ml (T5)	P value
Kidneys	0.38	0.35	0.37	0.36	0.34	0.13
Liver	1.65	1.67	1.63	1.64	1.66	0.10
Heart	0.38	0.37	0.36	0.35	0.39	0.21
Lungs	0.61	0.63	0.62	0.59	0.60	0.30
Gizzard	1.58	1.62	1.61	1.57	1.59	0.22
Spleen	0.04	0.03	0.05	0.04	0.03	0.10
Pancreas	0.21	0.23	0.22	0.24	0.25	0.20
Proventriculus	0.34	0.35	0.36	0.37	0.33	0.20

[abc] Means bearing different superscripts along the same row are significantly different(P<0.05).
+ = Positive; - = Negative; NFE = Nitrogen free extract.

Table.6 Sensory evaluation of Broiler Chicken given ethanol extract of *Lagenaria breviflora* as an Additive

Parameter	+ Control Antibiotics (T1)	-Control/ Untreated (T2)	1ml (T3)	2ml (T4)	3ml (T5)	P value
Colour	6.80	7.0	6.60	6.90	6.50	0.03
Flavour	7.10	6.90	7.00	6.80	6.95	0.10
Juiciness	6.60	6.40	6.70	6.50	6.80	0.1
Tenderness	7.50	7.30	7.40	6.90	7.20	0.20
General Acceptability	8.10	7.90	8.00	8.20		0.30

+ = Positive; - = Negative; NFE = Nitrogen free extract

This study revealed that live weight, total weight gain and average daily gain were higher in birds that were treated with ethanol extract of LB and those treated with Neoceryl antibiotic compared to those that were not treated. This is in line with the report of Oladunjoye *et al.,* (2023) who earlier reported a higher weight gain in broiler chicken medicated with aqueous extract of LB than those that were not treated.

Ekunseitan (2025) also reported slightly higher curve in the hen-day production of Yaffa layers given 300g LB in 4 liters of drinking water daily towards 15 weeks in lay compared to the control group. Bozkurt *et al.,* (2009) also reported positive effect of herbal essential oil on the productive and reproductive performance of broiler breeder.

Feed conversion ratio was revealed to be better in birds that were medicated with ethanol extract of L breviflora than those that were not medicated. Egbeyale *et al.,* (2021), earlier reported a better feed conversion in broiler chicken that were medicated with hot water extracted LB fruits at finisher phase.

Mortality was revealed to be lower in birds that were medicated with ethanol extract of *L. breviflora* than those that were not medicated in this study. Tomori *et al.,* (2007) had earlier reported broad spectrum antibacterial effects of ethanolic extract of LB on different species of bacteria which could have controlled the pathogenic strain of bacteria in birds that were medicated. The fruit of LB has also been reported to possess haematinic, immune-stimulatory and anti-oxidant activities (Saba *et al.,* 2009; Onasanwo *et al.,* 2011) that could help birds that were medicated to resist diseases better than those that were not treated. Apparent digestibility of dry matter and crude protein was higher in birds that were medicated with extract of LB and antibiotics than the group that were not medicated. This could be due to the alteration of the gut environment by LB such that the microbial population favour the digestion of these nutrients. The lower feed cost/kg live weight observed in birds that were medicated with extract of ethanol in this study is a direct effect of better feed conversion observed in the birds that were medicated.

The carcass yield and primal cuts of the birds were not affected in this study. This implies that the use of LB in broiler production will not affect the market value of the birds. The use of this extract in place of commercial antibiotic will also promote the production of organic poultry meat which is gaining wider acceptability in the recent years.

Also the internal organs of the birds were not affected by the administration of ethanolic extract of LB which indicates that the extract does not contain anti-nutritional factors at a level that can adversely affect the health status of the birds.

Sensory characteristics of the birds given ethanol extract of LB in this study was not affected. This agree with the work of Ekunseitan *et al.,* (2023) who reported that administration of aqueous extract of LB had no effect on the sensory characteristics of broiler breast meat.

In conclusion, It was revealed in this study that birds that were given ethanol extract of *L. breviflora* had higher growth rate, better feed conversion, lower mortality and feed cost per kilogram weight gain than the group that was not medicated. Performance of the birds that were given the extract was also similar to the group given given Neoceryl antibiotic as additive with respect to these parameters. Therefore Ethanol extract of *L Breviflora* can be used to replace commercial antibiotic

as additive in broiler chicken production to enhance organic chicken production.

Author Contributions

I. O. Oladunjoye: Investigation, formal analysis, writing—original draft. A. O. Adeniran: Validation, methodology, writing—reviewing.

Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethical Approval Not applicable.

Consent to Participate Not applicable.

Consent to Publish Not applicable.

Conflict of Interest The authors declare no competing interests.

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