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Research Article

Effect of *Jatropha curcas* Supplemented Diet on Broilers

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Abstract: Several authors have shown that farmers in Africa feed their poultry with partial or total dietary supplement, especially, diet based on local and cheaper ingredients in order to maximize profit, this however may have opposite effects because some of these supplements are toxic to the birds leading to low productivity or high mortality among the birds. This study was therefore conducted to determine the effect of raw *Jatropha curcas* supplemented diet on broilers' health and growth using some biochemical parameters. A total of forty one-day old Arbor acres broilers were used for this study, they were acclimatize for four weeks during which medication and other managerial practice were administered to the birds as directed by the supplier before they were randomly allocated into four different experimental groups receiving different experimental diets containing 0, 4, 8 and 12% raw *Jatropha curcas* respectively. After four weeks of treatment with different experimental diets, blood samples were collected from the birds for biochemical analysis. The results showed that feeding broilers with raw *Jatropha curcas* supplemented feed resulted in elevation in liver biomarkers (AST, ALT, ALP ,Bilirubin) and kidney biomarkers (Urea, Creatinine). Also there was increase in serum cholesterol with a marked decrease in both serum protein and packed cell volume. From the result, it was obvious that raw *Jatropha curcas* supplemented diet is hepatotoxic, nephrotoxic and can affect blood circulation in the birds even in small quantity and should be detoxified before it can be used as a supplement or substitute in broiler's feed.
Keywords: *Jatropha curcas*, supplement, broilers, toxic, biomarkers

INTRODUCTION

The common protein sources and other ingredients used in poultry feeds have become too expensive because of their high demand this had led to the evaluation of lesser known and underutilized crop seeds for their nutritive values. In this regards, *Jatropha curcas* seeds have been proposed as a cheap supplement to the conventional forage crops [1-3].

Jatropha curcas, a member of the Euphorbiaceae family is a drought resistant multipurpose tree of significant economic importance. The kernel has a protein content of 27–32 % by weight while the residue after oil extraction (fully defatted meal) has a relatively high protein content around 53–58% by weight [4].This relatively high protein content of *Jatropha curcas* can be advantageous since this rich source of protein is not utilized by humans like commonly consumed food crops such as soy and wheat [4,5], however, at present the use of *Jatropha* in feed is limited owing to the presence of toxic and anti nutritional constituents[6].Several studies showed that the *Jatropha curcas* seeds are toxic to humans and animals due to the presence of phorbol esters and certain proteins [4, 6- 9]. The biological effects of these compounds include tumors promotion, wide range of negative biochemical and cellular effects, alteration of cell morphology, induction of platelet aggregation and also serve as lymphocyte mitogens [10].

To maximize profit farmers have been feeding their poultry with partial or total dietary supplement, particularly diet based on local and cheaper ingredients [3, 11-17].This however may have negative effects on the bird because some of these supplements are toxic to the animals leading to low productivity or high mortality among the birds. Therefore, it is necessary to investigate the toxicity of some of these supplements before they are used as supplement.

The purpose of this study was to evaluate the effects of raw *Jatropha curcas* incorporation into broiler's diets on some biochemical parameters in broiler in order to assess its toxicity

EXPERIMENTAL SECTION

Sample Collection And Preparation

Jatropha curcas seeds were collected from Agwada area of Nasarawa state Nigeria and proper identification was carried out at the National Institute for Pharmaceutical Research and Development (NIPRD) located at Idu industrial area – Abuja. The seeds were dried and hand-cracked. The kernels obtained were milled using a mechanical grinder, air dried at room temperature and then packaged until when needed.

Birds Management and duration of the experiment

A total of 40 one- day old Arbor acres broiler chicks strain were purchased from Mararaba, Nasarawa state. They were brooded together for four weeks and fed

with commercial broiler starter obtained from poultry feed vendor. All medications and required managerial practices were applied as at when due. After four weeks, the chicks were divided into their various experimental groups with each group receiving one of the experimental diets and clean drinking water ad-libitum for a period of four weeks.

Commercial broiler diet

The commercial feed used was obtained from a reputable feed company and recompose into the desired experimental diet.

Table 1: Compositions of the commercial Feed

Constituents	Percentage composition (%)
Cereals/grains	25
Vegetable protein	13
Premix (vitamins/minerals)	9
Essential Amino acids	20
Salt	5
Antioxidants	9
Anti-toxins	9
Prebiotic	5
Enzymes	5
Nutritional composition	
Crude protein	20
Crude fat	10
Crude fibre	9
Calcium	1
Available phosphorous	0.45
Metabolizable Energy	2800kcal/kg

SOURCE: Label on the feed as stated by the feed company

Experimental design

After four weeks of acclimatization, the birds were randomly allocated into four different dietary groups as shown below.

Group 1: Fed with normal commercial diet without *Jatropha curcas.*

Group 2: Fed with experimental diet containing 4% *Jatropha* curcas by composition.

Group 3: Fed with experimental diet containing 8% *Jatropha* curcas by composition.

Group 4*:* Fed with experimental diet containing 12% *Jatropha curcas* by composition.

Table 2: Experimental diet composition

Composition	Group 1	Group 2	Group 3	Group 4
Commercial diet (%)	100	96	92	88
Raw *Jatropha curcas* (%)	0	4	8	12
Total (%)	100	100	100	100

Physical and Clinical observations

The birds were keenly observed for any discharge, evidenced allergic reaction, behavioural changes, change in weight, mortality and any appearance of pathological condition

Analysis of raw *Jatropha curcas*

The moisture, protein, fat, ash, fiber and nitrogen free extract were analysed by the methods of AOAC[18]. Phytate was determined using the method descibed by Mohamed et al.[19] While Saponin and total oxalate were determined according to the methods of Pearson [20] and Adeniyi et al., 2012 respectively.

Collection of blood samples

5ml of blood samples were collected in duplicate from each bird. The first one was collected into an anticoagulant bottle while the second one was collected into plain bottle without anticoagulant.

Haematological study

The blood samples in the anticoagulant (heparinized) bottles were used to determine the packed cell volume (PCV).

Serum biochemistry study

The blood sample in the plain bottles was allowed to clot for about two hour. The clotted blood was centrifuged at 3,500rpm for 30mins to recover the serum from the clotted blood. Serum was separated with sterile syringes and needles and stored frozen until when needed. The following biochemical analysis was performed on the sera samples: Serum aspartate amino transferase (AST) and alanine amino transferase (ALT)

activities were estimated with the Randox reagent kit using 2, 4-dinitrophenylhydrazine as substrate according to the method described by Reitman and Frankel [22]. Urea, creatinine and total bilirubin concentrations were determined by the methods of Patton and Crouch [23]; Henry *et al*.[24] and Pearlman and Lee [25], respectively. Total cholesterol was measured by the procedure described by Allain *et al*.,[26] . Protein content was determined by the method of Lowry *et al*.[27] while Alkaline phosphatase was assessed as described by Principato et al.,[28]. All the assays were carried out at the Department of Biochemistry, Bingham University Karu Nasarawa state, Nigeria.

Statistical analysis

The data are expressed as mean ± SD. Statistical analysis was performed using SPSS (Version 17). A level of $p < 0.05$ was considered to be significant results

RESULTS AND DISCUSSION

The nutritional compositions of raw (undefatted) *Jatropha curcas* seeds are shown in tables 1. The moisture contents is lower than 10% moisture content limit recommended for storage stability of flours[29] which suggest that the seed flour will have a long shelf life. The ash content (table1) of the *J. curcas* seed flour is an indication that it may have a reasonable quantity of mineral elements for building healthy body and proper function of the body tissues while the moderate fiber content means it will enhance easy movement of the bolus in the large intestine. The average crude protein obtained (33.07 ± 0.09%) is higher than the value reported by Abou- Arab and Abu-Salem[30] and that of the seed of Jatropha gossipifolia (13.40) reported by Ogbobe and Akano[31].This high crude protein content makes it a good source of protein and substitute or supplement for soybean and other protein rich legumes.

As expected high value was observed for the crude fat (34.01%) (Table1) which has been shown to contain most of the antinutritional factors especially phorbol. Relatively high carbohydrate level (20.29%) was also obtained. Crude fibre content (2.69%) (Table 1) is however lower than that reported by Abou- Arab and Abu-Salem [30] for raw *Jatropha curcas* but higher than 0.2% reported for soybean [32]. Crude fiber in diet consists mostly of plant polysaccharides that cannot be digested, it therefore increases stool bulk and decreases the time that waste materials spend in the gastrointestinal tract. [33, 34].

Table 3: Proximate composition of *Jatropha curcas*

Composition	Percentage (%)
Moisture	5.97 ± 0.014
Ash	6.76 ± 1.380
Crude protein	33.07 ± 0.090
Crude fat	34.01 ± 0.790
Carbohydrates	20.29 ± 0.160
Crude fiber	2.69 ± 0.860
Nitrogen free extract	5.20 ± 0.140

All values are mean of triplicate determination ± standard deviation.

Evaluation of antinutional content of raw *Jatropha curcas* showed that the saponin content (Table 4) is lower than 4700mg/100g reported by Makkar et al., [29] for Soybean meal. Saponin is linked with some negative effects on animals including reduction of palatability and intake of nutrient [35, 36]. The phytate content (Table 4) in the present study agrees with that of soybeans reported by Reddy and Pierson [37]. Phytates have been implicated in decreasing protein digestibility by forming complexes and also by interacting with enzymes such as trypsin and pepsin[37].It also form complexes with divalent minerals thereby decreasing the bioavailability of these elements for absorption[38].The high concentration of oxalate observed can affect calcium absorption.

Table 4: Antinutritional factors in raw *Jatropha curcas*

Anti-nutritional factors (mg/100g)	Content(mg/100g)
Phytate	48 ± 0.57
Saponin	2500 ± 141.42
Oxalate	16.5 ± 0.85

All values are mean of triplicate determination ± standard deviation

Measurement of the activities of various enzymes in body fluids plays a significant role in disease investigation, diagnosis and detection of tissue damage. From the results (Table 5) there was an increase in the liver biomarker enzymes examined, this increase was dose dependent, that is, increase in the

quantity of *Jatropha curcas* in the feed resulted in increase in the level of these enzymes in the serum. The increase in serum liver enzymes (ALT, AST, and ALP) levels is an indicator of liver damage and cytotoxic effect of raw J. *curcas* seed on the liver cells leading to leakage of these enzymes from damaged hepatocytes cytosols into bloodstream[39,40,41]. ALT an enzyme produced in cytosol of hepatocytes of the liver is the most sensitive marker for liver than the remaining ones and it's serum level is an indication of hepatocellular damage that can therefore provide quantitative assessment of the degree of damage sustained by the liver [42].The trend of ALP observed gave an indication that the hepatic capacity of the liver is grossly affected by *Jatropha curcas*[43] .Total bilirubin was also elevated in the serum of the broilers administered with *Jatropha curcas*; bilirubin is a conventional indicator of liver diseases and its elevation in the serum has been associated with hepatocellular damage and hepatic biliary tract obstruction. These results agree with previous reports by El Badwi et al.[44] and Samia et al[45] who reported the negative effect of *Jatropha curcas* inclusion in feed. The toxic effects were ascribed to the toxic substances in the oil of raw *Jatropha curcas* [44, 45].

Table 5: Effect of different graded level of raw *Jatropha curcas* in broilers feeds on hepatic function biomarkers

GROUPS	AST(U/I)	ALT(U/I)	ALP(U/I)	BILIRUBIN (mg/dl))
Group 1	72.55 ±5.98[a]	64.26±5.67[a]	33±11.00[a]	0.53 ± 0.08[a]
Group 2	84.00 ± 20.65[a]	83.08 ± 7.52[a]	46.20 ± 18.70[a]	0.73 ± 0.17[a]
Group 3	106.00 ±9.58[b]	242.90 ± 8.90[b]	70.40 ± 27.60[b]	1.40 ± 0.03[b]
Group 4	106.17 ± 9.64[b]	283.8±11.00[b]	79.2±14.34[b]	1.61 ± 0.06[b]

The values are mean ± standard deviation of five observations. Values with different superscript in the same column are significantly different at $p< 0.05$.

The effect of *Jatropha curcas* on the kidney functions was assessed by the levels of serum creatinine and urea, as the levels of serum urea and creatinines are often regarded as reliable markers of renal function [46]. The significant elevation of creatinine and urea (Table 6) is a pointer to renal dysfunction in chickens given *Jatropha curcas*[47]. Creatinine is a break-down product of creatine. It is usually produced at a fairly constant rate by the body and filtered out of the blood by the kidneys. If the filtering capacity of the kidney is deficient, creatinine blood level rises [48, 49]. Urea is the major end product of protein catabolism in animals and is the primary vehicle for removal of toxic ammonia from the body. It is primarily produced in the liver and excreted by the kidneys. In general, increased urea levels are associated with compromise in kidney function [46,50].Therefore, from the result the toxicants or antinutrional factors in raw *Jatropha curcas* can cause a damage to the kidney there by distorting renal function.

Table6: Effect of different graded level of raw *Jatropha curcas* in broilers feeds on renal function biomarker

GROUP	UREA(mg/dl))	CREATININE(mg/dl))
Group 1	30.83 ± 4.52	0.45 ± 0.07
Group 2	42.08 ± 4.01[a]	0.51 ± 0.06
Group 3	58.33 ± 466[a]	1.580 ± 0.227[a]
Group 4	70.00 ± 4.79[a]	2.882 ± 0.611[a]

The values are mean ± standard deviation of five observations. a = significant difference at $p<0.05$ compare with the Control.

The administration of *Jatropha curcas* caused an increase in serum cholesterol and marked decrease in serum protein concentration (hypoproteinemia)[Table 7].The hypoproteinemia observed may be attributed to the direct toxic effect of phorbol esters leading to degeneration and necrosis of hepatocytes[39,51] which are considered to be the main site of protein synthesis[52]. The interaction of phorbol ester with protein kinase C (PKC) affects activities of several enzymes, including the enzymes involved in biosynthesis of protein, DNA, polyamines, cell differentiation processes, and gene expression[39,53].This may be responsible for the low levels of protein in the serum of the chicken fed with graded levels of *Jatropha curcas.* Reddy and Salunkhe[54], also attribute the decreases in total protein to inhiition of protein utilization in the broilers. Increase in serum cholesterol in broilers fed with graded level of *Jatropha curcas*, may be as a result of stimulation of lipolytic hormones action on the fat depots due to inhibition of insulin[55,56,57].This elevated blood cholesterol levels have been reported by

Omage *et al*.[58] as the most important risk factor in heart disease which result result in the death of the chicken.

Table 7: Effect of different graded level of raw *Jatropha curcas* in broilers feeds on their serum protein and cholesterol

GROUP	TOTAL PROTIEN (mg/dl)	CHOLESTEROL(mg/dl)
Group 1	8211.20 ± 226.10	55.23 ± 17.96
Group 2	6326.40 ±440.40	72.56 ± 12.94
Group 3	5120 ± 33.94	119.37 ± 24.87
Group4	4524 ± 369.20	143.14 ± 11.91

The values are mean ± standard deviation of five observations.

Table 8 shows the results of the packed cell volume of the broilers progressive decrease in packed cell volume was observed as the level of raw *Jatropha curcas* supplement increases. The decline in % PCV with an increase in raw *Jatropha curcas* in the diet indicates blood loss and destruction of erythrocytes, decrease in % PCV with an increase in raw *Jatropha curcas* all point to the development of anaemia in the broilers. Chivandi et al.[59] reported that the anaemia could have been haemorrhagic as a result of blood loss through the gastrointestinal tract (GIT). Furthermore damage to the GIT was also reported to have resulted in maldigestion and malabsorption of nutrients required for erythropoiesis[59].This was also in line with our observations that broilers fed with higher concentration of raw *Jatropha curcas* have stunted growth and looked very weak

Table 8: Effect of different graded level of raw *Jatropha curcas* in broilers feeds on their packed cell volume.

GROUP	PACKED CELL VOLUME(PCV) %
Group 1	28.60 + 2.07
Group 2	24.60 + 2.41
Group 3	19.60 + 1.14
Group 4	19.00 + 1.58

The values are mean ± standard deviation of five observations in percentage

FURTHER OBSERVATIONS

During the administration of the experimental diets the broilers fed with higher concentration of raw *Jatropha curcas*(8% and 12%) have stunted growth, looked very weak and their feathers sheds often. Also, mortality rate increased according to the percentage of raw *Jatropha curcas* meal ingested but no death was recorded in the control group.

CONCLUSION

From the results and observations, it was evident that raw *Jatropha curcas* supplemented feed is hepatotoxic, nephrotoxic and can affect blood circulation in the birds even in small quantity and should therefore be detoxified before it can be used as a supplement or substitute in animal feed.

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