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Effects of Biochemically Treated *Jatropha curcas* Seed cake on the Performance Characteristics and Haematological Indices of Growing Rabbits

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Abstract

The experiment was conducted to investigate the effect of Biochemically treated *Jatropha curcas* seed cake on the performance characteristics (Feed intake and digestibility) and haematological indices of growing rabbits (N=16, average initial weight = 805g). The rabbits were divided into four treatments of four replicates each containing four rabbits in a Complete Randomized design model. The animals were fed the experimental diets *ad libitum* with Treatments A (Control with 100% Soyabean cake), B (50% Biochemically Treated Jatropha seed cake), C (75% Biochemically Treated Jatropha seed cake) and D (100% Biochemically Treated Jatropha seed cake) for a 56 day period. The results revealed higher dry matter intake (127.33g/d), crude protein intake (12.25g/d), ether extract intake (17.63g/d) and ash intake (23.52g/d) for Treatment A (Control) compared with the Biochemically treated groups. Contrarily, Treatment B had the highest crude fibre intake among other Treatments. Among the Biochemically Treatments, B had the highest dry matter intake and ash intake while Treatment C had highest crude protein intake and D the highest ether extract intake. It is interesting to note that Treatment D had the highest dry matter and ether extract digestibility. All haematological indices were within the reported normal range for rabbits of that weight. Conclusively, Biochemically treated *Jatropha curcas* seed cake could be included as a protein source in the dietary treatment at a level of 50% for growing rabbits without any deleterious effect.

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Key words: Biochemically treated *Jatropha curcas* seed cake, *Aspergillus niger*, feed intake, digestibility coefficient, haematological indices

Introduction

As the population increases in Nigeria so also is the competition between man and livestock for the conventional feed stuffs (maize, soyabean cake and groundnut cake, etc.). This leads to increase in the prices of conventional feed-stuffs and ultimately increase in the cost of production. Therefore, there is the need to search for alternative unconventional feedstuffs without compromising the quality and quantity.

Jatropha curcas is one of the lesser known multipurpose shrub trees which has significant economic values due to its several industrial and medicinal potentials. The seed of the plant could be used as cheap source of protein for livestock if properly processed. Jatropha seed cake (oil extraction) contains very good nutrient profile compared to Soyabean cake. The crude protein content is between 58 and 64% , (90% of which is the true protein) higher than Soyabean cake. The cake is currently being used as organic fertilizer but its utilization as livestock feed should be encouraged (if properly processed) because of the high level of the essential amino acids. (Makkar and Becker, 1997). However, both the seed and the oil are found to be toxic (Heller, 1996, Aderibigbe *et al*., 1997, Belewu *et al.*, 2013) containing numerous antinutrients (saponin, tannin, phorbolester, Lectin).

Various methods (mechanical, physical and chemical) detoxification of Jatropha seed cake are well documented in literature (Makkar and Becker, 1997). All the aforementioned methods are not very effective. However, the Biochemically method reported herein seems most promising. Apart from its efficacy there is less threat of residual chemical effect on the health of both livestock and man.

Belewu *et al*. (2011a, b) reported the use of the fungus- *Aspergillus niger* to detoxify Jatropha seed cake with encouraging results. This study therefore, aimed at evaluating the dietary potential of Biochemically treated Jatropha seed cake on the performance characteristics (feed intake and digestibility coefficient) and haematological indices of growing rabbits.

Materials and Methods

Experimental site

The experiment was conducted at the Teaching and Research farm of the University of Ilorin, Kwara State, Nigeria. University of Ilorin is located in the Central part of Nigeria with 8° 29' 47.90"N Latitude and Longitude of 4° 32' 31.70" E and a temperature of between 18^0C and 35^0C. The rainfall varies from 94mm to 125mm and the humidity of 30%.

Collection and Preparation of Seed Cake

Dried matured fruits of *Jatropha curcas* were collected from the University of Ilorin *Jatropha curcas* Plantation which was established in 2009. The husk of the fruits were removed and the seeds dried and later milled and the oil was extracted using hydraulic press. Remaining oil in the cake was extracted using petroleum ether. The cake attained was autoclaved at 121^0C 15psi for 30 minutes to get rid of any microbes that could be present in the cake.

Collection of *Aspergillus niger*, inoculation and incubation

The fungus used was *Aspergillus niger* which was obtained from the Department of Crop Protection, University of Ilorin, Nigeria. A bijou bottle of pure cultured of *Aspergillus niger* was used to inoculate petri dishes containing sterilized Potato dextrose agar (PDA) medium using a wire loop, sterilized in Bunsen burner flame till it was red hot and later cool.

The inoculated petri dish was incubated for 7days at room temperature during which the fungus enveloped the medium and the fungus growth was terminated by oven drying the cake at $70^{O}C$ for 24 hours. The dried sample was used in formulating diets for the rabbits (Table 1).

Experimental Diets

The cooled, oven dried *Jatropha curcas* seed cake was used in the formulation of diets in replacement of Soyabean cake at varying inclusion levels of 50% (Diet B), 75% (Diet C) , and 100% (Diet D) while Diet A was the (Control). Other ingredients were of fixed proportions (Table 1).

Management of the Experimental Animals

Sixteen growing rabbits were randomized against the experimental diets with four animals per treatment in a Completely Randomized design model. The rabbits were housed in individual cages and managed intensively. Feed and water were given *ad-libitum* to the animals for 8 weeks and the following parameters feed intake, digestibility coefficient and haematological indices were evaluated.

Table 1: Composition of the Experimental Diets

Ingredients	Diet A (Control)	Diet B (50%)	Diet C (75%)	Diet D (100%)
Cassava waste meal	30.00	30.00	30.00	30.00
Rice bran	23.50	23.50	23.50	23.50
Soya bean cake	10.00	5.00	2.50	-
Fungus treated Jatropha Seed cake	-	5.00	7.50	10.00
Palm kernel cake	26.50	26.50	26.50	26.50
Bone meal	2.50	2.50	2.50	2.50
Vitamin – Mineral premix	0.50	0.50	0.50	0.50
Salt	0.50	0.50	0.50	0.50
Total	100.00	100.00	100.00	100.00

Chemical analysis

Samples of the Treated and Untreated Jatropha cake, the experimental diets and faeces were subjected to proximate analysis using the methods of AOAC (1990) at the Department of Animal Production Laboratory, University of Ilorin, Nigeria.

Statistical analysis

All data collected were subjected to Analysis of Variance (ANOVA) of a Completely Randomised designed model and significant differences were separated using Duncan (1955) multiple range test.

Results and Discussion

The percentages dry matter (92.25%), crude protein (15.89%), ether extract (49.27%) and ash (11.17%) of the fungus treated *Jatropha curcas* seed cake were significantly higher than that of the untreated cake (Table 2) due probably to the action of the fungus on the substrate. Contrarily, the crude fibre of the Fungal treated *Jatropha curcas* seed cake was significantly lower that the untreated cake due probably to the action of the fungus which could have utilized the fibre for their own growth (Table 2). The observation was in agreement with the results of Jacqueline and Visser (1996).

The increased Crude protein content after treatment could be attributed to the addition of microbial protein during the fermentation process. This corroborated the submissions of Jacqueline and Visser (1996) and Belewu *et al.* (2011a).

The improvement of the ether extract of the Fungal treated sample agreed with the report of Belewu et *al.* (2011a,b) and Belewu *et al.* (2013).The increased ether extract could be due to the lipolytic action of *Aspergillus niger*. This corroborated the report of Souza *et al.* (2014) that *Aspergillus niger* has lipolytic ability.

Table 2: **Chemical Composition of the Fungal Treated and Untreated *Jatropha curcas* Seed Cake***

Parameters (%)	Untreated Seed cake (T1)	Treated Seed cake (T2)	Significances (P<0.05)
Dry matter	90.60	92.25	T1 vs T2
Crude Protein	12.79	15.89	T1vs T2*
Crude fibre	34.61	16.78	T1 vs T2*
Ether extract	35.15	49.27	T1 vs T2*
Ash	8.72	11.17	T1 vs T2*

***Means of Six Determinations**

Table 3: Feed Intake of the Experimental Animals (g/d)

Parameters	Treatment A (Control)	Treatment B	Treatment C	Treatment D	±SEM
Dry matter intake	127.33[a]	104.48[ab]	95.49[b]	92.42[b]	4.85
Crude Protein intake	12.25[a]	8.83[b]	9.02[b]	8.54[b]	0.45
Crude fibre intake	14.86	20.09	17.94	16.56	0.90
Ether extract intake	17.63[a]	12.56[b]	12.74[b]	13.05[b]	0.65
Ash intake	23.52[a]	16.48[b]	15.49[b]	13.97[b]	0.77

Means on the same row with similar superscripts are not significantly different (p>0.05)

With the exception of the highest Crude fibre intake recorded for animals on fungal treated diets compared to the Control (Table 3) every other nutrient (Dry matter intake, Crude Protein intake, Ether extract intake and Ash intake) was highest for the Control Treatment. The increasing Crude fibre intake may be due to the facts that Rabbits are pseudoruminant and may need to satisfy their fibre requirement (NseAbasi *et al*. (2014a). The higher Crude fibre intake by the rabbits could also be due to the predigestion of the fibre by the inoculated fungus (*Aspergillus niger)*. Allied with the aforementioned is the efficient ability of rabbits to convert 30% of Crude fibre to meat as against 10% of most poultry species (Makinde *et al*., 2015).

The Crude Protein, Dry matter, Ash intake and Ether extract intake met the nutrients need for rabbits of this weight as recommended by NRC (1977). The values also fell within the ranges reported by Omole *et al*. (2005).

Table 4: **Apparent Digestibility Coefficient of Growing Rabbits Fed Biochemically Treated *Jatropha curcas* Seed cakes**

Parameters	Treatment A (Control)	Treatment B	Treatment C	Treatment D	±SEM
Dry matter digestibility	92.13[ab]	92.51[ab]	89.18[b]	94.58[a]	0.69
Crude Protein digestibility	88.37[a]	89.83[a]	80.40[b]	87.91[a]	0.87
Crude fibre digestibility	57.13[b]	67.42[ab]	70.75[ab]	83.80[a]	3.06
Ether extract digestibility	89.75[a]	88.53[a]	79.64[b]	90.55[a]	0.94

Means on the same row with similar superscripts are not significantly different (p>0.05)

There was significant differences in the Dry matter digestibility (DMD) among the Treatments (Table 4). The highest DMD was noted for animals on Diet D and lowest for animals on Diet C. The DMD reported herein was higher than the value noted by Ramchurn *et al*. (2000). The difference in the results could probably due to variation in the diets. Contrarily, the Crude Protein digestibility (CPD) was highest for Treatment B followed by Treatments A and D which were similar (P>0.0%) but significantly different from Treatment C (80.40%). The CPD noted in this study was higher than the reported value of Omole *et al*. (2005). The variation could be due probably to the variation in the composition of the experimental diets in the two studies. Conversely, the Crude fibre digestibility (CFD) was highest for Treatment D followed by Treatments C, B and A in that order. It is interesting to note that CFD of Treatments C and D was highest compared to the values reported by Omole *et al*. (2005). The poor CFD of Treatment A (Control) might be due to the low Crude fibre content of this Treatment. The Ether extract digestibility (EED) reported herein fell within the values reported by Ranchurn *et al* (2000) for rabbits.

The higher digestibility values of nutrients reported in this study might be due to the higher nutritive values of the treated test feedstuff. This corroborated the report of Ajmalkhan *et al*. (2008) that the digestibility of any feed is influenced by its nutritive value.

Table 5: Effects of Biochemically Treated Jatropha curcas Seed cake on the Haematological Indices of Growing Rabbits

Parameters	Treatment A (Control)	Treatment B	Treatment C	Treatment D	±SEM
PCV (%)	25.50^{b}	27.00^{a}	23.00^{c}	20.00^{d}	0.13
WBC x 10^9/l	7.40^{a}	7.30^{a}	6.70^{b}	5.40^{c}	0.05
RBC x10^{12}/l	1.80^{b}	2.00^{a}	1.30^{c}	1.30^{c}	0.03
Haemoglobin (g/l)	6.15^{b}	7.10^{a}	5.40^{c}	5.30^{c}	0.04
Lymphocytes (%)	58.50^{d}	70.00^{a}	65.00^{b}	64.00^{c}	0.13
Neutrophils (%)	36.50	30.00	33.00	34.00	0.88
Monocytes (%)	1.50^{a}	0.00^{b}	0.00^{b}	0.00^{b}	0.13
Eosinophils (%)	3.50	2.00	2.00	2.00	0.88
Glucose (mg/dl)	156.00	112.00	74.00	89.00	13.96

Means on the same row with similar superscripts are not significantly different (p>0.05)

The evaluation of the haematological indices (Table 5) was in-line with the report of WHO (1963) that blood examination is a good of assessment of the health status of animals as it plays a vital role in the physiological, nutritional and pathological status of animals. The Packed cell volume (PVC) measures the ratio of the volume occupied by Red blood cell to the volume of whole blood in a sample of capillary/venous/arterial. The PVC value reported herein agreed with the report of Olabanji *et al*. (2007) and it fell within the normal range for rabbits (Mitruka and Rawnsley, 1977).

White Blood Cell (WBC) count fell within the normal range of 2.5 – 12.5 $X10^9$/l reported by PGCVS (1990) and the range of 5 – $13x10^9$/l (Burke, 1994). This shows that the animals were healthy because decrease in number of WBC below the normal level is an indication of allergic conditions, anaphylactic shock and certain parasitism (Reilly, 1993). Red Blood Cell (RBC) and Haemoglobin were lower than the values reported by Mitruka and Rawnsley (1977). Low haematological value is an indication of anaemia (Mitruka and Rawnsley, 1977; Radostits *et al.* (1994)). Additionally, low haematological value could be due to the destruction of red blood cell (NseAbasi *et al*., 2014b)

The Lymphocyte count of Treatments A, C and D was in agreement with the report of Kronfield and Mediway (1975) and Mitruka and Rawnsley (1977). However, the value of Treatment B exceeded the value reported by Kronfield and Mediway (1975) and Mitruka and Rawnsley (1977). This could be a good sign because Campbel and Lasley (1975) postulated that significantly lower lymphocyte count was an indication of a reduction in the ability of experimental rabbits to produce and release antibodies when infections occurs. Neutrophil count of all the Treatments fell within the normal range of between 30.0 and 43.2% (kronfield and Mediway, 1975; Mitruka and Rawnsley1977; Olabanji *et al*., 2007). The result of the Monocyte count agreed with 0.67 -1.5% reported by Olabanji *et al*. (2007).

The Eosinophil count of Treatment B was lower than the value reported by Mitruka and Rawnsley (1977) while Eosinophil count of Treatment A was higher than the value reported by Mitruka and Rawnsley (1977). The higher blood glucose levels recorded for Treatments A and B was in agreement with the previous observation noted by Onifade and Tewe (1993).

Conclusion and Implication

It could be concluded from this study that inclusion of fungal treated *Jatropha curcas* seed cake in the diet of growing rabbits at 50% was not detrimental to the health of the animals, this is an indication that fungus treated *Jatropha curcas* seed cake can successfully replace Soyabean cake in the diet of growing rabbits at 50%. It was noted from this study that dietary inclusion of 50% fungal treated *Jatropha curcas* seed cake is feasible in the diet of growing rabbits.

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