

Growth and Haematological Effects of Rabbits Fed *Delonix Regia* Seed Diets

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Abstract

The growth and haematological response of growing rabbits to diets containing *Delonix regia* seeds were studied for a period of 63 days. Forty (40) weaner rabbits with an average weight of 390-590 g obtained from College of Agriculture and Science Mando Kaduna State Nigeria, were assigned to eight dietary treatments containing *Delonix regia* seeds cooked at 100°C for 0, 15, 30, 45, 60, 75 and 90 minutes respectively. The rabbits were assigned in such that their ages, sizes and group weights were balanced as much as possible. The animals were allowed to adjust to the test diets and cages for 7 days before the start of the experiment.

During the period of acclimatization, the rabbits were dewormed with invermectin injection. Each of the treatments contained five (5) rabbits in a Completely Randomised Design (CRD). All cages were supplied feeders and drinkers. The diet were formulated to contain over 18% crude protein and fortified with minerals and Vitamin premix in accordance with NRC nutrient requirements for rabbits. Proximate analysis and haematological parameters were determined. So also average daily feed intake, weight gain, feed efficiency and cost per kg live weight were calculated using the prevailing market price of that season.

Initial and final weights were also observed. Generally, *Delonix regia* seed diets performed better than the control group as the duration of cooking increases up to 60 minutes of cooking. Feed efficiency also increases significantly ($P < 0.05$) as the duration of cooking increases. Numerically, rabbits fed on seeds cooked for 60 minutes had the highest feed consumed and the best feed efficiency. The best Packed Cell Volume (PCV) was obtained from the control diet. The absence of monocyte and eosinophils indicated that *Delonix regia* seed diets could be fed to rabbit from 30-50% inclusion without causing any side effect, thus providing a cheaper source of feeding and feeding conversion ratio.

Keywords: Nutritive value; Growth; Haematological parameters

Introduction

The animal protein for human consumption in Nigeria (from cattle, pigs, poultry, sheep and goats) has not been able to bridge the gap between demand and supply. Despite the numerous advantages associated with the consumption of animal protein, the minimum intake recommended by FAO (1992) has not been met. This is mostly because the price of meat has gone beyond the means of most Nigerians.

The domestic rabbit (*Oryctolagus cuniculus*) is an important non ruminant herbivore for meat production. Rabbit meat is a source of healthful food as it is low in cholesterol but high in protein 22 g/1000 [1]. Rabbit can also utilize the available proteins in cellulose rich plants whereas, it is not economical to feed these to chickens and turkeys, the only animals with higher energy and protein efficiency. Since there is high demand for additional source of food world wide the exploitation of plant seeds of low economic importance would be a step toward better resource utilization Telek et al. [2] which is in line with the strategy to achieve sustainable animal production system by matching them with locally available feed sources [3].

The continued increase in cost of conventional feeding ingredients especially grains, cakes and meals and the effective competitions from both poultry industry and human population are the major constraints for progressive growth of the rabbitary in developing countries. One of the methods that have been utilized for some time now in alleviating this problem is the replacement of percentage grains and cakes in the diet of monogastric animals especially rabbits. However, the present trend in the annual increase in the cost of these products have necessitated the need to use unconventional feeding stuffs for rabbitary that are free of competition from other industries.

Materials and Methods

Seed collection and preparation

The seeds of *Delonix regia* were obtained from College of Agriculture and Science, Mando Kaduna State, Nigeria. The pods were soaked in a pool of water for 3 days for the seeds to split open after sun drying. The seeds collected were divided into six batches; one batch was left raw, while other batches were cooked for various durations, namely, 0, 15, 30, 45, 60, 75 and 90 minutes, respectively. Timing commenced immediately after adding the seeds in boiling water. The boiled seeds were drained off the water and sun dried for 4 days. The raw and boiled seeds were milled in a hammer mill and stored in screw-capped container until required for feed formulation.

Proximate analysis

Moisture content : Five grams of the sample was weighed into pre-weighed aluminum drying dish. The sample was dried to a constant weight in an oven at 105°C for 4 h [4]. The moisture content was then determined as follows:

$$\text{Moisture content} = \frac{m_1 - m_2 \chi 100}{m_1 - m_0 \chi 1}$$

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m_0 = Weight of aluminum dish

m_1 = Weight of fresh sample + dish

m_2 = Weight of dried sample + dish

Ash content: Five grams of the sample was weighed into a porcelain crucible, previously ignited and weighed. Organic matter was charred by igniting the material on a hot plate in the fume cupboard. The crucible was placed in the muffle furnace and maintained at 60°C for 6 h. It was then cooled in a desiccator and weighed out immediately. The percentage ash was determined using the following formula:

$$\text{Ash} = \frac{(\text{Weight of crucible+ash}) - (\text{Weight of empty crucible}) \times 100}{\text{weight of sample}} \times 100$$

Crude fat: Five grams of the sample was put in the thimbles and plugged with cotton wool. The thimbles were dried and inserted into a Soxhlet system HT2. The extraction cups were dried and weighed, 25 ml of petroleum ether was added to each cup. The cup was inserted in the soxhtec, and the sample was extracted for 15 min in a boiling position and 45 min in a rising position. The percentage fat in the sample was calculated as follows:

$$\text{Fat} = \frac{W_3 - W_2 \times 100}{W_1 \times 1}$$

W_1 = weight of sample

W_2 = weight of empty cup

W_3 = weight of cup with extracted oil

Crude fibre: The trichloroacetic acid method of Joslyn [5] and Adeniji et al. [6] was used. The sample was defatted with petroleum ether. Five grams of the defatted sample was weighed into a 600 ml beaker and 100 ml trichloroacetic acid was added. The sample was boiled and refluxed for 4 min. The cooled sample was filtered with a filter paper, Whatman No. 4.

The residue was washed with distilled water and methylated spirit. The filter paper together with the sample were transferred to porcelain crucible and dried in an oven overnight at 100°C. The sample was cooled in a desiccator and weighed. It was then ashed in a muffle furnace at 600°C for 6 h and weighed again after cooling. The loss in weight during incineration was equivalent to the amount of crude fiber.

Sample weight:

Weight A = weight of sample after drying

Weight B = weight of sample after ashing

Crude protein : For determination of crude protein, a Kjeldahl nitrogen method was used. One gram of the sample was introduced into the 800 ml digestion flask. Kjeldahl catalyst (5 Selenium tablets) was added to the sample. 20 ml of concentrated sulphuric acid was added to each sample and fixed to the digestion flask until a clear solution was obtained. The cooling digest was transferred into 100 ml volumetric flask and made up to the mark with distilled water.

The distillation apparatus was set up and rinsed for 10 min after boiling; 20 ml of 4% Boric acid was pipetted into conical flasks, 5 drops of Methyl red was added to each flask as indicator and the digest was diluted with 75 ml distilled water. 10 ml of the digest was made alkaline with 20 ml of 20% NaOH and distilled. The steam exit of the distillator was closed and the change in colour of boric acid solution to green was

noted. The mixture was distilled for 15 min [4]. The boric acid along with distillate was then titrated against 0.1N hydrochloric acid and, consequently, the total nitrogen was calculated as indicated below:

Total nitrogen = Normality = 0.1 N

% Crude protein — % Total Nitrogen x 6.25

Carbohydrate: This was obtained by subtracting the sum or the percentage moisture, ash, crude fiber, fats and protein from 100.

Blood Collection and Clinical Examination

Blood samples were collected at the 8th week from the jugular veins of the rabbits into a set of Bijou bottles, containing Ethylene diamine tetra acetic (EDTA) [7,8] immediately after collection. The blood samples were taken to the laboratory for analysis.

Packed Cell Volume Determination

The Packed Cell Volume (PCV) values of the blood were determined according to the micro-haematocrit method of Benjamin (1978) with slight modification Oladele [9] in timing of the centrifuge. Blood containing EDTA was aspirated into a set of plain capillary tubes. The blood sample was centrifuged at 9,500rpm for 15 minutes and PCV read directly from the graphic reader in percentage.

Haemoglobin Estimation

The values of haemoglobin for the rabbits were estimated by dividing the PCV value by 3 Maxwell et al. [10], Makinde and Olowookurun [11].

Total Protein of Determination

The total proteins in the blood of rabbits were determined using hand refractometric method of Benjamin [7]. Data collected were subjected to analysis of variance (ANOVA) Duncan's Multiple Ranges was used to separate the significant means both contained in SSPSS for Windows 2003

Results and Discussion

The result of the proximate composition of *Delonix regia* seed diets are presented in Table 1. Dry matter ranges from 90.48-93.35g while protein is from 18.29-19.02%. Dry matter intake in animal nutrition is very important as it ensures the intake of large quantity of nutrients when compared to high moisture feeds. Significant difference ($P>0.05$) exist in the dry matter, protein, ether extract, ash and free nitrogen extract when the seeds were cooked at different durations. However, there was no significant difference ($P>0.05$) in the fibre contents. Crude fibre is useful for maintaining bulk mortality and increase intestinal peristalsis by surface extension of the food in the intestinal track [12].

This is also necessary for food digestion. The result shows that the crude fibre contents of the seeds are within the ranges recommended by Anugwa in 1998 and therefore suitable as materials for monogastric animals.

Table 2 is the effect of cooking *Delonix regia* seeds at 100°C for various durations. Significant difference exists between seeds cooked at different duration of time. The best cooking time for *Delonix regia* seed was 60 minutes. At 60 minutes cooking, it had the best feed efficiency, the highest feed consumed and there was no mortality reported. The higher ether extract value indicates a higher fat content.

The crude protein values were well above NRC recommended values for monogastric animals [13]. Studies Aletor and Aladetimi

Ingredients	Control	2	3	4	5	6	7	8
Maize	56.55	46.82	46.82	46.82	46.82	46.82	46.82	46.82
Groundnut cake	25.05	14.78	14.78	14.78	14.78	14.78	14.78	14.78
<i>Delonix regia</i>	-	20.00	20.00	20.00	20.00	20.00	20.00	20.00
Maize offal	15.0	15.0	15.0	15.0	15.0	15.0	15.0	15.0
Bone meal	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Limestone	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Lysine	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Methionine	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Salt	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Vitamin Premix	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Total	100	100	100	100	100	100	100	100

Calculated Analysis of Diets

Metabolizable energy (kcal/kg)	2952.88	2672.45	2672.48	2672.48	2672.48	2672.48	2672.48	2672.48
Crude Protein (%)	18.50	16.89	16.89	16.89	16.89	16.89	16.89	16.89
Crude Fibre (%)	4.01	7.15	7.15	7.15	7.15	7.15	7.15	7.15
Ether Extract (%)	4.09	13.62	13.62	13.62	13.62	13.62	13.62	13.62
Ash (%)	2.23	3.22	3.22	3.22	3.22	3.22	3.22	3.22
Calcium (%)	0.32	0.46	0.46	0.46	0.46	0.46	0.46	0.46
Phosphorus (%)	0.40	0.32	0.32	0.32	0.32	0.32	0.32	0.32
Lysine (%)	0.80	4.38	4.38	4.38	4.38	4.38	4.38	4.38
Methionine (%)	0.43	0.60	0.60	0.60	0.60	0.60	0.60	0.60
Cost per kg (₦)	70.18 ^a	42.37 ^c	60.23 ^{ba}	44.74 ^{bc}	44.41 ^{bc}	59.68 ^{ba}	51.99 ^{bc}	51.23 ^{bc}

A vitamin-mineral premix provides per kg of diet: Vit. A, 13.340. i.u; Vit D₃, 2680 i.u; Vit. K, 2.68 i.u; Calcium pantothenate, 10.68 mg; Vit B₁₂, 0.022 mg; Folic acid, 0.668 mg; Choline chloride 400 mg; Chlorotetracycline, 26-28 mg; Manganese, 133.34 mg; Iron, 66.68 mg; Zinc, 53.34 mg; Copper, 3.2 mg; Iodine, 1.86 mg; Cobalt, 0.268 mg; Selenium, 0.108 mg.

Table 1: Diets Compositions and Calculated Analysis of *Delonix regia* Seed Meals Diets.

Parameters	0 min	15 min	30 min	45min	60 min	75 min	90 min	SEM	LOS
Crude Protein (%)	92.05 ^e	90.48 ^a	93.35 ^a	92.60 ^e	93.14 ^b	93.13 ^c	92.77 ^d	0.00	*
Crude Fibre (%)	18.75 ^b	18.38 ^b	19.41 ^b	18.36 ^b	19.02 ^b	18.56 ^b	18.29 ^b	0.208	*
Ether Extract (%)	6.80 ^{ab}	6.57 ^b	7.60 ^a	6.31 ^b	6.67 ^{ab}	6.07 ^b	6.21 ^b	0.164	*
Ash (%)	8.52 ^{bc}	6.73 ^e	8.38 ^{bcd}	8.97 ^b	9.94 ^a	8.09 ^{cd}	8.20 ^{cd}	0.117	*
Nitrogen Free Extract (%)	8.34 ^f	9.16 ^a	8.64 ^c	8.37 ^e	8.64 ^c	8.86 ^b	8.37 ^e	0.00	*
	50.00 ^f	59.76 ^a	59.14 ^d	57.78 ^c	51.1 ^c	58.59 ^b	58.46 ^b	0.036	*

Figures followed by the same letter(s) in each row are not significantly different (P< 0.05) using DMRT

SEM: Standard error of means

* LOS: level of significance

*: Significant (P < 0.05)

NS: Non-significant difference

Table 2: Proximate Composition of Experimental Diets of *Delonix regia* Seeds Meal Cooked at 100°C for Varying Duration (in minutes).

[14], Sobayo et al. [15] have shown that high protein in diets of rabbits could produce some undesirable physiological and biological alterations which may manifest in the haematological characteristics of animal when fed such diets. The test diets shows protein content of *Delonix regia* seeds. Table 3 shows the haematological parameters of rabbits fed *Delonix regia* seed diets. It is well established that changes in haematological and biochemical parameters reflect the physiological status of animal [16]. Packed Cell Volume (PCV) values obtained in this study were not significantly (P>0.05) influenced by dietary treatments.

The Packed Cell Volume (PCV) obtained fell within the range of 31.0-48.6% reported by Mitraka and Rawnsley [17], 33-50% ASAN (2006) and 33-50% reported by Medi Rabbit [18]. The PCV is the fraction or portion occupied by the red blood cell Bush (1991). Haemoglobin levels were within the reported range of 9.9-19.3% reported by Turfey (1995). This normal range of haemoglobin suggests an efficient oxygen transport system. The higher PCV and Hb of rabbits in all the treatments were apparently, due to better nourishment of these diets on the rabbits. This result is in agreement with the

findings of Hackbarth et al. [19], who demonstrated that PCV and Hb concentrations indicate the nutritional status of the diets.

The White Blood Cell count was also observed to be within values of 2.0 – 15.0x10⁶/ml reported by Medirabbit [18]. This is an indication that defensive system of the rabbits was not affected. The non-significant (P>0.05) higher values of neutrophils content in all the treatments indicate similar environmental conditions of stress on the rabbits. This shows that their resistance to infection was stable. The rare occurrence of monocytes suggests that *Delonix regia* seeds did not cause inflammatory reactions in the blood system of the rabbits. So also, the absence of eosinophils shows that there was no allergic condition when *Delonix regia* seeds are consumed. The values obtained compared favourably with those obtained Hewlett et al. [20], RAR [21] and Amata [13]. Thus, *Delonix regia* seeds may be included in the diet of rabbits without compromising the health status of the rabbits.

Conclusion

The results from the feeding trials indicate normal haemoglobin

	Control	0 min.	15 min.	30 min.	45 min.	60 min.	75 min.	90 min.	SEM	LOS
Packed cell volume (%)	35.25 ^b	41.65 ^{ab}	46.50 ^a	33.75 ^b	35.00 ^b	35.00 ^b	38.00 ^{ab}	38.75 ^{ab}	1.11	NS
Total protein	6.4 ^a	7.75 ^a	6.00 ^a	6.65 ^a	6.27 ^a	6.46 ^a	6.20 ^a	6.10 ^a	0.10	NS
Haemoglobin	11.73 ^b	12.20 ^b	15.55 ^a	11.23 ^b	11.63 ^b	12.62 ^b	11.90 ^b	12.88 ^b	0.32	NS
Leucocytes	10.84 ^a	10.65 ^a	8.75 ^a	9.00 ^a	7.57 ^a	15.06 ^a	11.70 ^a	10.11 ^a	0.90	NS
Neutrophils	24.00 ^a	30.25 ^a	31.50 ^a	32.00 ^a	31.00 ^a	28.40 ^a	32.60 ^a	31.50 ^a	2.39	NS
Lymphocytes	75.50 ^a	68.75 ^a	68.25 ^a	67.25 ^a	75.50 ^a	71.00 ^a	67.00 ^a	68.25 ^a	2.43	NS
Monocytes	1	3	1	3	3	2	1	1		
Eosinophils	1	0	1	0	0	0	0	0		

Figures followed by the same letter(s) in each row are not significantly different ($P < 0.05$) using DMRT

SEM: Standard error of means

* LOS: Level of significance

*: Significant ($P < 0.05$)

NS: Non-significant difference

Table 3: Haematological Parameters of Rabbits Fed Delonix regia Seed Cooked at 100°C for Varying Duration.

and higher PCV values of rabbits in all treatments, thus showing the nutritional status of the seeds. The rare occurrence of monocytes and the absence of eosinophils shows that the seeds did not cause any inflammatory and allergic reactions respectively. It can therefore be included on rabbit diets without compromising the health status of the rabbits.

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