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Effect of Different Methods of Processing Neem (*Azadirachta indica*) Seeds on Performance of Young Rabbits

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Abstract: A feeding trial using sixty (60) weaner rabbits with an average initial live weight of 560 ± 0.80g was conducted to determine the effects of different methods of processing neem seeds on their performance and carcass characteristics. The rabbits were assigned to five dietary treatments based on their initial live weight and sex. There were twelve individually caged rabbits per treatment. The diets were formulated to be isonitrogenous (18% CP). Diet 1 (control) was a Maize-Groundnut cake based diet without neem seed. Diets 2, 3, 4 and 5 had raw milled neem seed (RMNS), hydraulic press neem seed cake (HNSC), Solvent Extracted neem seed cake (SNSC) and Expeller neem seed cake (ENSC) included at 20% level, respectively. Feed and water were offered *ad libitum*. Weekly feed intake and body weight changes were determined and feed efficiency calculated. At the end of the 63-day feeding trial, four rabbits per treatment were selected based on the group average weight and sacrificed for carcass evaluation. Blood samples were also obtained from the animals to evaluate Packcell volume (PCV), Haemoglobim (Hb) and Total protein (TP). The results showed that rabbits fed the processed neem seed cake diets had growth performance that were statistically comparable to those fed the control diet. Animals on processed neem seed cake diets had significantly ($P<0.05$) improved performance over and above those fed the raw neem seed diets. Animals on raw neem seed cake diet had consistently low value for PVC, Hb and TP. The use of solvent extracted neem seed cake in rabbit diet was favoured in this study.

Key words: Neem, solvent extraction, expeller extraction, hydraulic extraction, rabbit

Introduction

Neem *(Azadirachta indica}* seed cake (NSC), a by-product of neem oil industry, is a non-conventional feed ingredients showing great potential for livestock feeding (Nath *et al.,* 1974; Gowda *et al.,* 1998; Bawa *et al.,* 2005). Neem seed cake has been noted as a rich protein source (34-38% CP). Feeding Neem seed cake in raw form to livestock is generally discouraged due to the presence of bitter triterpenoids (Musalia *et al.*, 2000). Elangovan *et al.* (2000) noted that despite its high protein content of 300-400g/kg, NSC could not earn favour as a good livestock feed due to its pungent smell and bitter taste imparted by the presence of toxic triterpenoids; azadirone, nimbin and salanin. However recent reports indicate that it can be fed to rabbits in limited amount without any adverse effect (Fajinimi *et al.,* 1990; Salawu *et al.,* 1994; Bhosale, 1994). A practical processing technology to make neem seed cake wholesome for animal feeding at higher level is required. Such a methodology needs to be tested and perfected for maximum debitterization of neem kernel cake with minimum cost (Gowda and Sastry, 2000). The processing of neem seed to remove the antinutritional factors can be done either by solvent extraction of the neem oil or mechanical extraction of the neem oil. However, much emphasis has not been laid on which of these processes give the best result in terms of improving the growth performance of the animal. This study therefore is aimed at examining the effect of different methods of processing neem seeds on performance of young rabbits.

Materials and Methods

Processing of neem seeds: The neem seed cakes used for this study were residues of oil extraction from neem seeds using different methods of processing. Before oil extraction, neem fruits were soaked in water for 4-5 days and depulped using depulper machine. The seeds with the pulp were then washed and sun-dried for 7 days. The dried seeds were decorticated using a winnowing machine and then crushed after further drying for 3 days. For the expeller neem seed cake, the crushed kernel was steamed and the oil was pressed out using expeller machine. The hydraulic press neem seed cake processing was devoid of heat. The milled kernel was cold-pressed using the hydraulic press machine until the oil content of the residue (cake) was minimal. To reduce the oil content of the cake, it was further defatted using hexane. For the solvent extracted neem seed cake, the oil from the crushed neem kernel was extracted using organic solvent of high polarity mixed with water and filtered. The cakes obtained using the different methods of oil extraction was sun-dried before incorporated into the experimental diets. Raw

Table 1: Composition of Experimental Diets

Diet	1	2	3	4	5
Ingredients	Control	RNSMD	HNSCD	SNSCD	ENSCD
Maize	64.20	52.29	52.29	52.29	52.29
GNC	18.80	10.71	10.71	10.71	10.71
NSC	-	20.00	20.00	20.00	20.00
Blood meal	4.00	1.00	4.00	1.00	4.00
Bone meal	2.00	2.00	2.00	2.00	2.00
Limestone	0.50	0.50	0.50	0.50	0.50
Salt	0.30	0.30	0.30	0.30	0.30
[1]Vitamin premix	0.20	0.20	0.20	0.20	0.20
Total	100.00	100.00	100.00	100.00	100.00
Cost per kg of diet	24.28	21.49	18.49	18.49	18.49
Calculated analysis					
Crude protein (%)	18.00	18.00	18.00	10.00	18.00
Metabolizable Energy (Kcal/kg)	2792.78	2433.31	2433.31	2433.31	2433.31

[1]Bio organics starter vitamin-premix contributed the following per kg of diet:
Vitamin A, 12,000 I.U; Vitamin D_3, 1,200 I.U; Vitamin E, 3.6 I.U; Vitamin K, 1.8mg;
Vitamin B_2, 3.6mg; Nicotinate, 18mg; Calcium-d - Pantothenate, 9.6mg; Biotin, 0.3mg Vitamin B_{12}, 0.012mg; Choline chloride, 120mg; Chlortetracycline, 4.8mg; manganese 24mg; Iron, 48mg; Zinc, 96mg; Copper, 60mg; Iodine, 1.8mg; Cobalt, 48mg.

Table 2: Proximate composition of diets based on groundnut cake, Raw neem Seed meal or neem seed cake obtained from different methods of processing fed to young rabbits

Treatment	1	2	3	4	5
Parameters	Control	RNSMD	HNSCD	SNSCD	ENSCD
Dry matter (%)	96.69	96.80	96.65	96.25	96.50
Crude protein (%)	24.31	24.63	23.88	24.31	24.13
Ether extract (%)	8.15	8.16	7.54	7.00	7.63
Ash (%)	6.17	8.36	6.69	6.52	6.40
Crude fibre (%)	6.15	5.17	5.44	5.44	5.24
NFE (%)	55.22	53.68	43.55	56.73	56.60

RNSM - Raw Neem Seed Meal. HNSCD - Hydraulic press neem Seed cake diet. SNSCD - Solvent extracted Neem Seed cake diet ENSCD - Expeller Neem Seed cake diet.

Table 3: Proximate composition of Raw neem Seed meal and neem seed cake from different processing methods

Parameters	RNSM	HNSC	SNSC	ENSC
Dry matter (%)	89.87	86.29	88.96	91.35
Crude protein (%)	23.19	22.69	23.06	22.50
Crude fibre (%)	9.35	4.14	7.80	12.60
Oil (%)	38.61	21.02	33.44	22.22
Ash (%)	9.33	7.16	8.26	8.95

RNSM - Raw Neem Seed Meal. HNSC- Hydraulic press neem Seed cake. SNSC - Solvent extracted Neem Seed cake.
ENSC - Expeller Neem Seed cake.

neem kernel was also ground and used directly without oil extraction in this experiment.

Experimental diet: The experimental diets (Table 1) were formulated to be iso-nitrogenous (18% crude protein). The experimental diets consisted of five treatments viz: control diet, Raw Neem seed meal diet (RNSMD), expeller neem seed cake diet (ENSCD), solvent extracted neem seed cake diet (SNSCD) and hydraulic press Neem seed cake diet (HNSC). The diets were further fortified with adequate minerals and vitamins in accordance with the NRC (1988) nutrient requirement for rabbits. The proximate analysis of the experimental diets and the major ingredients were carried out at the Biochemistry Laboratory unit of Animal Science Department, Ahumadu Bello University, Zaria according to AOAC (1990) procedure. The proximate composition of the experimental diets and the neem seed cakes are presented in Tables 2 and 3, respectively.

Experimental design and management of animals: Sixty (60) weaner rabbits with an average initial weight of 560±80g obtained from Animal Science Departmental rabbitary were assigned to five dietary treatments in such a way that age, sex and group weights were balanced as much as possible. The animals were allowed 7 days adjustment period before the start of the experiment. Each of the treatments contained 12 rabbits in a completely randomized design. All the cages were equipped with feeders and drinkers.

The animals were fed the treatment diets and fresh clean water *ad libitum* at 8.00am throughout the 63-day trial period. The rabbits were weighed at the beginning of the trial and also at weekly intervals. At the end of the experiment, average daily feed intake, average daily live weight gain, feed conversion efficiency and feed cost per kg live weight gain were computed. At the end of the trial, 6 rabbits from each treatment group were selected based on the group average weight and used for carcass analysis at the animal science meat laboratory.

Before slaughtering, the live weight of each rabbit was taken. External cleaning of the slaughtered rabbits was done using the flaying method. The internal organs were then removed and weighed. The carcass weight of each rabbit was taken and used to determine the dressing percentages of the rabbits. The weight of the major organs and retail cuts were taken and expressed as percentages of carcass weight. The blood samples were collected and analyzed for packed-cell volume (PVC), total protein (TP) and haemoglobin (Hb).

Statistical analysis: The data obtained from this study were subjected to Analysis of Variance and where statistical significance were observed, the means were compared using the Duncan's Multiple Range Test according to SAS (1995).

Results and Discussion

The result of the performance characteristics of young rabbits fed raw neem seed meal or differently processed neem seedcake diets are presented in Table 2. Final live weight showed a significant (P<0.05) difference across dietary treatments. Animal on control diet had the highest final live weight. Rabbits on solvent extracted neem seed cake diet and expeller neem seed cake were significantly different (P<0.05) from those fed raw neem seed meal, hydraulic press neem seed cake and control diets.

Feed intake of the rabbits fed the experimental diets had significant (P<0.05) difference across dietary treatments. Rabbits fed treatment 2 and 3 had the least feed intake compared to those fed diets 1 (control), 4 and 5. This is in line with the reports of Vasanthakumar *et al*. (1999), and Gowda and Sastry (2000). Vasanthakumar *et al.* (1999) observed lowered feed intake, nutrient digestibility and growth in rabbits fed 20% raw neem seed meal. Gowda and Sastry (2000) reported that neem seed cake is toxic and bitter to taste due to triterpenoids, which restricts its safe inclusion in livestock diets. The depressed feed intake observed in this study could be due to the bitter principles inherent in the raw neem seed. Debitterization through solvent (hexane, ether) extraction, water washing, alkali soaking and urea-ammunition had improved the palatability and nutritive value of the cake (Jana, 1997; Katiyar *et al*., 1991; Katiyar *et al.*, 1993). These authors also stated that several feeding trials with raw cake have revealed poor palatability and adverse performance among different categories of livestock and poultry with ruminants tolerating reasonably higher levels of the cake than the monogastrics.

Daily weight gains were significantly (P<0.05) influenced by dietary treatments. Rabbits fed raw neem seed meal diet had a significantly (P<0.05) depressed weight gain compared to those on the control diet. The depressed weight gain observed in rabbits fed raw neem seed cake could be attributed to the high level of bitter principles inherent in that diet. Rabbits fed treatment 4 (Solvent extracted neem seed cake) had a non significantly (P>0.05) higher daily weight gain compared to the rabbits on other neem seed cake diets. The similarity in daily weight gain of rabbits fed processed neem seed cake compared to control diet in this study is in line with Khan (1994) who observed comparable growth performance of rabbits fed alkali treated or water-washed neem kernel meal when compared to rabbits fed soybean meal diets.

Feed conversion ratio of rabbits fed the raw neem seed meal showed significant (P<0.05) difference compared to those fed the control diet, Jana (1997) reported lowered body weight gain and feed conversion efficiency when the broiler chicks were fed raw neem seed meal up to 150g/kg level in diet from day old to six weeks of age. Rabbits on diets 3, 4 and 5 efficiently utilized their feed better than those on the raw neem seed meal diet although the difference was not significant.

Cost per kg gain in weight ranged from N67.67 – N87.67. The highest feed cost per kg weight gain was obtained on rabbits fed raw neem seed meal diet (T_2) and this could be attributed to poor rate of feed conversion which may be due to the level of bitter principles in the diet. However hydraulic press neem seed cake diets had the least feed cost per kg gain among dietary treatments. There was no mortality due to dietary treatments. This is in consonance with Fajinmi *et al.*, 1990; Salawu *et al.,* 1994 and Gowda *et al.*, 1998 who did not observe mortality in rabbits fed processed neeem seed cakes.

The carcass characteristics of rabbits fed differently processed neem seed cakes are presented in Table 4. The difference among dietary treatments was non-significant (P>05) for the dressing percentage, lungs, liver and head (% yield) and length of small intestine (cm). However significant (P<0.05) difference was observed across dietary treatments for pre-slaughter weight, carcass weight, heart, kidney, stomach, intestine, skin, feet, tail, shoulder, loin and thigh (% yield), and length of large intestine (cm).

The significant (P<0.05) difference in pre-slaughter weight may be a reflection of feed intake, which was lower for rabbits fed treatments 2 and 3. The pre-slaughter weight of rabbits fed treatments 4 and 5 were statistically similar compared to the pre-slaughter weights of the rabbits on the control diet. The kidney (%) of the animals fed control and solvent extracted neem seed cake diets showed a significant (P<0.05) difference compared to the raw neem meal, hydraulic press neem seed cake and expeller neem seed cake diets. The kidney (%) was higher for animals on treatments 2, 3 and 5, which was in line with Musalia *et al.* (2000). Musalia *et al.* (2000) observed that kidneys of lambs fed urea-treated neem seed kernel cake were

Table 4: Performance of rabbits fed experimental diets

Parameters	Treatment 1	2	3	4	5	SEM	Level of Significance
	Control	RNSMD	HNSCD	SNSCD	ENSCD		
Initial live weight (g)	543.75	600.00	550.00	566.67	543.75	52.29	NS
Final live weight (g)	1642.67^{a}	975.00^{C}	1125.00^{c}	1325.00^{b}	1300.00^{b}	50.60	*
Daily feed intake (g)	34.62^{a}	23.07^{b}	28.53^{b}	31.38^{ab}	31.07^{ab}	5.05	*
Daily weight gain (g)	11.31^{a}	5.76^{b}	7.99^{ab}	8.46^{ab}	8.43^{ab}	1.20	*
Feed efficiency ratio	2.97^{a}	4.08^{b}	3.66^{ab}	3.93^{ab}	3.80^{ab}	0.36	*
Protein efficiency ratio	1.96^{a}	1.37^{b}	1.57^{ab}	1.53^{ab}	1.53^{ab}	0.16	*
Cost per kg gain (N)	72.11	87.67	67.67	72.67	70.26	8.07	

a,b,c: Means value on the same row with different superscripts differ significantly (P<0.05). *: Significant (P<0.05) difference
NS: Non-significant different SEM: Standard Error of Mean RNSM: Raw Neem Seed Meal
SNSC: Solvent extracted Neem Seed cake HNSC: Hydraulic press Neem Seed cake ENSC: Expeller Neem seed cake

Table 5: Carcass characteristics of rabbit fed raw and processed neem seed cake diets

Parameters	Treatment 1	2	3	4	5	SEM	Level of Significance
	Control	RNSMD	HNSCD	SNSCD	ENSCD		
Pre slaughter weight (g)	1642.67^{a}	975.00^{c}	1125.00^{c}	1325.00^{b}	1300.00^{b}	5060	*
Carcass weight (g)	820.00^{a}	424.50^{c}	487.50^{ba}	625.00^{b}	624.67^{b}	48.50	*
Dressing percentage (%)	49.92	42.25	43.11	47.20	48.15	2.72	NS
Heat (% of carcass)	0.61^{b}	0.99^{ab}	0.61^{b}	0.73^{b}	1.29^{a}	0.15	*
Lung "	1.55	2.25	2.11	1.60	2.26	0.26	NS
Liver "	7.17	11.43	10.30	8.79	8.84	1.35	NS
Kidney "	1.83^{b}	2.81^{a}	3.02^{a}	1.99^{b}	2.84^{a}	0.21	*
Stomach "	2.44^{b}	3.80^{a}	3.61^{a}	3.03^{ab}	3.05^{ab}	0.32	*
Intestine "	12.97^{b}	23.44^{a}	18.28^{ab}	14.18^{ab}	15.86^{ab}	2.88	*
Head "	18.30	22.47	20.97	17.97	16.03	2.05	NS
Skin "	19.50^{a}	14.35^{c}	13.50^{c}	16.84^{b}	16.03^{b}	0.51	*
Feet "	5.73^{c}	9.57^{a}	8.15^{ab}	6.64^{bc}	6.66^{bc}	0.50	*
Tail "	1.30^{b}	1.37^{b}	1.65^{a}	1.36^{b}	1.55^{ab}	0.08	*
Shoulder "	36.33^{b}	39.75^{ab}	43.53^{a}	38.99^{ab}	38.41^{ab}	1.58	*
Loin "	29.58^{a}	20.12^{b}	20.48^{b}	27.84^{a}	21.26^{b}	1.02	*
Thigh "	33.98^{ab}	32.28^{b}	35.62^{ab}	34.24^{ab}	38.86^{a}	1.48	*
Length of small intestine (cm)	265.67	260.00	276.50	267.50	267.00	5.61	NS
Length of large intestine (cm)	152.67	152.50^{b}	156.59^{ab}	143.00^{c}	164.67^{a}	2.72	*

a,b,c Means value on the same row with different superscripts differ significantly (P<0.05)
*: Significant (P<0.05) difference. NS: Non-significant different. SEM: Standard Error of Mean

Table 6: Blood profile of rabbits fed experimental diets

Parameter	1	2	3	4	5	SEM	Level of Significance
	Control	RNSMD	HNSCD	SNSCD	ENSCD		
Pack cell volume	42.00^{a}	27.50^{b}	38.00^{ab}	45.00^{a}	43.50^{a}	3.51	*
Total Protein	7.00^{ab}	5.75^{C}	7.75^{a}	6.75^{b}	7.10^{ab}	0.23	*
Haemoglobin	13.95	9.15^{b}	12.60^{ab}	13.45^{a}	14.45^{a}	1.18	*

a, b, c Means values on the same row with different superscripts differ. * Significantly (P<0.05)

larger (P<0.05) than the groundnut cake fed animals.

Table 5 shows the blood profile of rabbits fed experimental diet. It revealed a significant (P<0.05) difference in all the parameters. Rabbits fed the raw neem seed cake had a significant (P<0.05) difference compared to those fed control and processed neem seed cake diets for the Packed-Cell Volume (PCV), Total Protein (TP) and Haemoglobin (HB) which is an indication of the negative effect of the bitter principles of neem on the animals. The findings of this experiment favour the use of solvent extracted neem seedcake in the diet of young rabbits. It is concluded that solvent extraction, as a method of processing should be employed in the processing of neem seeds for inclusion in rabbit diets.

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