BIOKEMISTRI 16(1):49-55 (June 2004)

Printed in Nigeria

An international journal published by the

Nigerian Society for Experimental Biology

Assessment of biological value of Terminalia catappa seed meal-based diet in rats

Nasir O. MUHAMMAD* and Oyelola B. OLOYEDE

Department of Biochemistry, University of Ilorin, Ilorin, Nigeria.

Received 23 November 2003

MS/No BKM/2003/052, © 2004 Nigerian Society for Experimental Biology. All rights reserved.

Abstract

The effects of defatted *Terminalia catappa* seed meal on the growth performance and carcass of rats were investigated. Twelve weaning albino rats (*Rattus norvegicus*) with an average weight of 24.0 ± 3.4 g were maintained on diets composed of defatted *T. catappa* seed meal (Tc meal) and soybean meal (control) for six weeks. The weights of the rats were monitored on weekly basis, at the same period of the day and before being served the (weighing) day's feed. The organs and carcasses of the rats were weighed after they were sacrificed and disemboweled, and the chemical compositions of the carcasses were also determined. The body weights of the rats maintained on defatted Tc meal were significantly reduced to one-third (1/3) of the weights of the control animal (p<0.05). The organ to body weight ratio of the Tc meal rats were significantly higher (p<0.05) than that of the control. In the carcass of rats fed Tc meal diet, ether extract, crude protein and ash contents were significantly reduced (p<0.05) when compared with the control. However, the crude fibre and the nitrogen free extract (NFE) were significantly higher in the Tc meal rats than that of the control (p<0.05). It can be deduced that the defatted *Terminalia catappa* seed meal could cause depression in growth rate, enlargement of rat tissues with adverse effects on carcass of rats.

Key words: Terminalia catappa, defatted meal, growth depression, carcasses

^{*}Author to whom all correspondence should be addressed.

E-mail: <u>alphamno2@yahoo.com</u> Tel: 08033931900

INTRODUCTION

Of all the food nutrients, protein requirements are fundamental, without which life would be impossible.¹ Soybean has been found to be superior to other plant protein supplements.² Soybean is however, presently not sufficiently produced to meet the demand of its use as plant protein ingredient in both livestock and human feeds. This limitation coupled with the high cost of animal source of protein (which are of course of better quality than plant sources) poses a challenge for a search for alternative source of protein for human and plant animal consumption. Furthermore, the lack of adequate information on the chemical and biological values of many plant materials of potential value as human and animal food has limited utilization of such materials, particularly protein and oil rich seeds; such should therefore be exploited. Among such alternatives is the seed of Terminalia catappa.

Terminalia catappa is a medium size tree whose branches form layers of canopy. The leaves of this tree have been reported to have medicinal values^{3,4}. The fruit is edible, fleshy, green (unripe) and yellow or red (when ripe). The exocarp is relatively thin and smooth, and the endocarp is hard. When cracked, a kernel is obtained and this can be consumed as well. The kernel of T. catappa has aphrodisiac activity and may be useful in the treatment of certain form of inadequacies such as premature sexual ejaculation.⁵ The seed is very rich in protein (19 -22%) and oil (50 - 52 %).⁶ The amino acid and mineral profiles of this seed has equally been documented.⁷

Information on the biological value of T. catappa seed consumption is still very scanty in literature. This study therefore aimed at investigating its effect on the growth performance and carcass of rats.

MATERIALS AND METHODS

Materials

The ripe fruits of Terminalia catappa were

picked from the premises of the University of Ilorin, Ilorin, Nigeria and authenticated at the Deparment of Plant Biology of the University. The albino rats were inbred in the Department of Biochemistry, University of Ilorin, Ilorin, Nigeria.

Processing of Seed meal

The riped fruits were oven-dried at 60°C and then cracked to remove the seeds, using 125mm Bench vice, FUKUNG Brand made in The People's Republic of China. The seeds were then milled using a manual grinding machine and the resulting meal was subsequently defatted with Soxleht extractor using petroleum ether $(40^{\circ}\text{C} - 60^{\circ}\text{C})$ as the extracting solvent. The extracted meal was autoclaved at 121°C and 15kgcm² for 30 minutes and was subsequently used in compounding rat feed using defatted sovbean meal as control⁸. The formulated diets are shown in Table 1.

Table 1: Composition of Diet	

Concentration		
(g/kg)		
Control*	Tc Meal	
516	416	
250^{\dagger}		
	350^{\dagger}	
40	40	
100	100	
40	40	
4	4	
10	10	
40	40	
	(g/) Control* 516 250 [†] 40 100 40 4 10	

* Soybean meal-based diet

[†] To provide 20% protein level

^a Vitamin mix (per Kg of diet): Thiamin hydrochloride, 6mg; Pyridoxine hydrochloride, 7mg; nicotinic acid, 30mg; folic acid, 2mg; calcium pantothenate, 16mg; biotin, 0.2mg; cyanocobalamin, 0.01mg; retinol palmitate, 4000 IU; cholecalciferol, 1000 IU; α -tocopherol acetate 50 IU; menadione 0.05mg; chlorine chloride, 2g.

Mineral mix (g/Kg diet): $CoCl_2.6H_2O$ (0.001); CuSO₄.5H₂O (0.07); MnSO₄ (0.178); KI (0.032); KH_2PO_4 (15.559); $CaSO_4$ (15.25); NaCl (5.573); ZnCO₃ (1.6); FeSO₄ .7H₂O (1.078); MgSO₄.7H₂O (2.292).

Proximate Analysis of the Diet

The proximate analysis of the composed diet was carried out using the method described by AOAC.⁹

Animals and Diets

Twelve male, 3-weeks old weaning albino rats of average weight $24.0 \pm 3.4g$ were divided into two groups of six (6) animals each. The rats were maintained on normal rat chow, allowed to acclimatized for a week, fasted for 24 hours, and then placed on the different diets.

The first group of rats (the control) was placed on the defatted soybean meal-based diet while the second group of rats (Tc meal) was placed on defatted *Terminalia catappa* seed meal-based diet.

The rats were housed in $33 \text{cm} \times 20.5 \text{cm} \times 19 \text{cm}$ plastic metabolic cages and were fed with their respective diets and water *ad libitum* for six weeks at $25^{\circ}\text{C} \pm 1^{\circ}\text{C}$, $50\% \pm 5\%$ humidity and, 12 hours light and 12 hours dark conditions. They were weighed weekly and at the end of the experimental feeding period, were sacrificed by anaesthetizing with (cotton wool soaked in) chloroform. They were then quickly dissected to excise the brain, liver, kidney, stomach and small intestine. The kidney was decapsulated and the stomach and intestine cleaned free of food. The organs and the carcasses were then weighed. The carcasses were dried in an oven at 80°C , and then analysed using the method of AOAC⁹. Statistical analysis was carried out using the students' t-test.¹⁰

RESULTS

Table 2 shows the proximate analysis of the formulated diets (test and control) on which the rats were maintained. There was no significant difference (p>0.05) in the various components of the diets. Rats maintained on the Tc mealbased diet had growth depression as shown by the body weights (Table 3) and growth pattern (Figure 1). The weights of rats fed with Tc meal were significantly reduced (p<0.05) to about one-third (1/3) of the control. The carcass weights of the experimental animals are shown in Table 3. The weights of the various organs and organ:body weight ratio of the rats are shown in Table 4. The weights of the organs of rats maintained on the Tc meal were significantly lower (p<0.05) than those of the control animals, however, there were significant increases (p<0.05) in the organ:body weight ratio of rats reared on the Tc meal diet compared to the control.

The proximate composition of the carcasses of rats in both group are shown in Table 5. When compared with the control, the crude protein, ether extract and ash contents of the carcass of rats fed with the Tc meal diet were significantly reduced (p<0.05) while there were significant increases (p<0.05) in the crude fibre and nitrogen free extract (NFE) of the Tc meal rats.

 Table 2: Proximate Composition (%) of the Formulated Diets

Group	Dry Matter	Crude Protein	Ether Extract	Crude Fibre	Ash	Carbohydrate (by diff.)
Control	96.20 ± 3.1	19.90 ± 2.4	15.45 ± 2.2	4.96 ± 1.4	0.023±0.002	59.45 ± 2.1
Tc meal	94.80 ± 2.4	20.56 ± 1.6	16.12 ± 3.3	5.22 ± 2.5	0.027 ± 0.004	58.22 ± 1.5

Values are expressed as mean of three determinations \pm SEM. Values are not significantly different (p>0.05)

Table 3: Body and Carcass weights (g) of rats
maintained on soybean meal-based- and T.
catappa seed meal-based diets for six weeks

	Body weight				
Group	Initial	Final	Carcass		
Control	23.83 ±	151.3 ±	54.4 ± 2.1		
	4.79	6.11			
Тс	24.0 ±	61.7 ±	38.80 ±		
meal	2.28	4.73*	1.2*		

Values are expressed as mean of six determinations \pm SEM.

*Significantly different from control (p<0.05)

DISCUSSION

The values obtained from proximate analysis of the formulated diets (Table 2) compared favourably with each other, indicating that the rats in both groups were placed on the same amount of nutrients quantitatively and therefore whatever differences that are noticed might be due to the differences in the quality of the nutrients in these diets or their bioavailability when consumed.

The growth rate depression (Fig. 1) and significant body weight reduction (Table 3) in the rats placed on the Tc meal diet is an

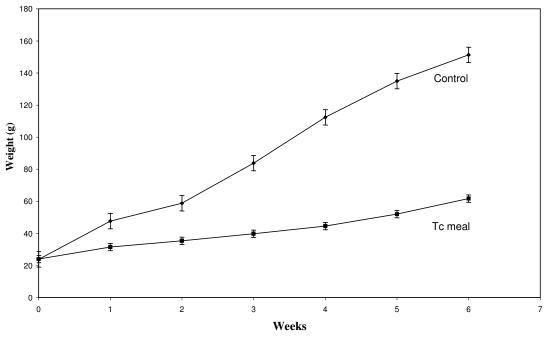


Figure 1: Growth curve of rats fed with Soybean meal- and T. catappa meal-based diets for 6 weeks

Table 4: Weight of organs and Organ:Body weight ratio	of rats	maintained	on soybean	meal-
based- and T. catappa seed meal-based diets for six weeks.				

	Organ Weight		Organ:Body weight ratio		
Organ	Control	Tc meal	Control	Tc meal	
Brain	1.56 ± 0.03	$1.33 \pm 0.04*$	0.011 ± 0.001	$0.024 \pm 0.002*$	
Liver	5.17 ± 0.40	$3.22 \pm 0.30^{*}$	0.034 ± 0.003	$0.057 \pm 0.001*$	
Kidney	1.05 ± 0.32	$0.518 \pm 0.04*$	0.0073 ± 0.000	$0.0096 \pm 0.001*$	
Stomach	0.281 ± 0.03	$0.295 \pm 0.03*$	0.0058 ± 0.001	$0.0121 \pm 0.002*$	
Small intestine	2.52 ± 0.13	$1.45 \pm 0.16^*$	0.056 ± 0.001	$0.060 \pm 0.001*$	

Values are expressed as mean of six determinations \pm SEM. *Significantly different from control (p<0.05)

Table 5: Proximate Composition (% based on dry weight) of Carcass of rats maintained on soybean mealbased and *T. catappa* seed meal-based diets for six weeks.

Group	Crude Protein	Ether Extract	Crude Fibre	Ash	N.F.E ¹
Control	3.76 ± 0.22	37.30 ± 0.72	1.75 ± 0.53	6.60 ± 0.41	50.59 ± 3.12
Tc meal	$2.02 \pm 0.02 *$	23.05 ±1.59*	$4.30 \pm 0.38*$	$2.61 \pm 0.05*$	$69.38 \pm 2.61*$

Values are expressed as mean of six determinations \pm SEM.

*Significantly different from control (p < 0.05)

¹ NFE : Nitrogen-free extract

indication that the defatted Tc meal does not support the growth of the animals. This is probably due to the high phytate, oxalate and tannin contents of the seed, which might have rendered unavailable the protein and mineral content of the seed.⁷ Phytate, an antinutrient, is a strong chelator of many divalent minerals such as Cu, Ca, Mg, Zn and Fe. The ability of phytate to chelate these essential minerals has been recognised as a potential threat in animal and human nutrition.^{11,12} Oxalate has also been reported to chelate with Ca²⁺ forming insoluble calcium oxalate.⁷ One of the nutritional effects attributed to tannins is interference with the absorption of calcium (Ca^{2+}) .¹³ Animals deficient in some minerals like Mg^{2+} , Se^{2+} and Zn²⁺ have been documented to have impaired nucleic acid and protein synthesis.^{14,15}

The deficiency or unavailability of these minerals would therefore lead to impairment in growth and development of the animals as observed in the present study. Additionally, phytates react directly with charged groups of proteins mediated by a positively charged mineral ion such as Ca²⁺, due to their ionic nature, thus the resultant phytate-protein and phytate-mineral-protein complexes may also adversely influence protein digestion and bioavailability.¹⁶ Tannins have also been reported to decrease protein quality by decreasing digestibility and palatability.¹³ It is possible that these antinutritional factors inhibit trypsin and pepsin with their protein complexing ability.¹⁶

Trypsin inhibitors have been known to interfere with the physiological process of digestion through interference with the normal functioning of the pancreatic proteolytic enzymes in nonruminants,¹⁷ leading to severe growth depression. It is possible that these antinutrients have interacted directly with the proteolytic enzymes leading to a corresponding reduction in the digestibility of the proteins in the diet.^{18, 19}

Two of the animals on the Tc meal-based diet died before the termination of the experiment and this might be due to the possible toxicity of the Tc meal on the internal organs of the rats as shown by the significant increase in the organ to body weight ratio on the rats maintained on the meal (Table 4). It has been reported that diets with high oxalate content could increase the risk of renal calcium absorption.^{20, 21} Tannins have also been reported to cause damage to the intestinal tract, toxic when absorbed from the gut and exhibit a possible carcinogenic effect.¹³

The results from the carcass evaluation of the rats on the Tc meal diet (Table 5) further corroborated the evidence that the high protein quality, inherent in the seed of Terminalia catappa had not been utilised to form body muscle as a result of possible chelation by the antinutrients. The significant reduction in ether extract of the carcass of rats placed on Tc meal diet (Table 5) further indicated that the lipid metabolism in these animals might have been adversely affected. This is because alteration in protein and lipid metabolism during deficiency of some minerals like Zn^{2+} and Mg^{2+} have been reported in literature.^{14, 15, 22, 23, 24} It may therefore be argued that reduction in the bioavailability of the mineral and protein contents of the Tc meal (due to its antinutritional

factors), has led to the observed significant decrease in weight and depression in growth rate of the animals fed with the Tc meal-based diet.

Furthermore, some of these antinutrients, especially phytates insoluble oxalates and condensed tannins, are heat stable, thus could withstand heat, harsh field conditions, seed/grain transportation, and storage environment, coupled with the fact that food processing alone may not prevent their potential adverse effects as some of them typically remained in the processed food.^{25, 26}

Perhaps, the use of exogenous enzymes for phytate reduction in monogastric nutrition which is recently suggested for the development of thermostable phytases ²⁷ or the use of fungi treatment to reduce antinutrients in foods, ²⁸ would assist in the usage of this protein-rich seed in animal and human nutrition.

REFERENCES

1. Brian, A. F. and Allen, G. C. (1977) Food science: a chemical approach. Pbs. Hodder & Stoughton. Pp. 305 – 332.

2. Galatet, A.G., Bike, D.H. and Norton, H.M. (1972) Protein-containing foods: Soyabean. *Soyabean Digest* **27:** 50–53.

3. Lin, C.C., Hsu, Y.F. and Lin, T.C. (1999) Effects of punicalagin and punicalin on carrageenan-induced inflammation in rats. *Am. J. Chin. Med.* **27**: 371-376.

4. Chen, P.S., Li, J.H., Liu, T.Y. and Lin, T.C. (**2000**). *Terminalia catappa* and its major tannin component, punicalagin, are effective against bleomycin-induced genotoxicity in Chinese hamster ovary cells. *Cancer Lett.* **152**:115-122.

5. Ratnasooriya, W.D. and Dharmasiri, M.G. (2000) Effect of *Terminalia catappa* seeds on sexual behaviour and fertility of male rats. *Asian J. Androl.* **2**: 213 – 219.

6. Adewole, A. and Olowookere, J.O. (1986) Nutritional potentialities of *Irvingia gabonensis* and *Terminalia catappa* fruits. *Nutr. Repts. Intern* **2**:10–13.

7. Osagie, A.U. (1998) Antinutritional factors. In: *Nutritional Quality of Plant Foods.* (A.U. Osagie and O.U. Eka eds). Published by Post Harvest Research Unit, University of Benin, Nigeria. Pp. 221–224.

8. Oloyede, O.B. and Muhammad, N.O. (2001) Effect of magnesium and essential fatty acids deficiency on alkaline phophatase and lactate dehydrogenase activities in the liver and kidney of rats. *NISEB Journal.* 1:169-174.

9. AOAC (1990) Association of Official Analytical Chemists. *Official Methods of Analysis.* 15th Edition. Washington D.C.

10. Adamu, S.O. and Johnson, T.L. (1997) Statistics for beginners, Book 1. SAAL Publication, Ibadan, Nigeria. Pp. 184 – 199.

11. Reddy, N.R., Pierson, M.D., Sathe, S.K. and Salunkhe, D.K. (1989) Phytates in cereals and legumes. CRC Press, Boca Raton, Florida.

12. Krebs, N.F. (2000) Overview of Zn absorption and excretion in human gastrointestinal tract. *J. Nutr.* **130**:2–12

13. Butler, L.G. (1989) Effects of condensed tannin on animal nutrition. In: "*Chemistry and significance of condensed tannins*" R.W. Hemingway and J.J. Karchesy Eds. Plennum Press, New York. Pp. 391 – 402.

14. Odutuga, A.A. (1982) Effects of low zinc status and EFA deficiency on growth lipid composition of rat brain. *Clin. Exptal. Pharmacol. and Physiol.* **9**:213–221.

15. Muhammad, N.O., Ogunleye, A.J. and Oloyede, O.B. (2001) Effects of selenium and zinc deficiency on the growth and lipid composition of developing rat brain. *Nig. J. Biochem. & Mol. Biol.* 16:169-172.

16. Reddy, N.R. and Pierson, M.D. (1994) Reduction in antinutritional and toxic components in plant foods by fermentation. *Food Res. Int.* **27**:281–290.

17. White, C.E., Campbell, D.R. and Combs, G.E. (1989) Effect of moisture and processing temperature on activities of trypsin inhibitor and urease in soybeans fed to swine. In: J. Huisman, T.F.B. van der Poel and I.E. Liener (Editors). *Recent Advances in Antinutritional Factors in Legume Seeds.* Puduc, Wageningen, Pp. 230 – 234.

18. Nwakolo, E.N. and Bragg, D.B. (1977) Influence of phytic acid and crude fibre on the availability of minerals from four protein supplements in growing chicks. *Can. J. Anim. Sci.* **57**: 475 – 477.

19. Hajos, G., Geleneser, E., Pusztai, A., Grant, G., Sakhri, M. and Bardoez, S. (1995) Biological effects and survival of trypsin inhibitors and agglutinin from soybean in the small intestine of the rat. *J. Agric. Food Chem.* **43**:165–170.

20. Oke, O.L. (1969) Oxalic acid in plants and in nutrition. *World Rev. Nutri. and Dietetics* **10**: 262 – 303.

21. Libert, B. and Franceschi, V.R. (1987) Oxalate in crop plants. J. Agric. Food Chem. **35**: 926-938.

22. Jurgen, V., Theoodor, G., Vera, H. and Klaus, S. (1995) Effects of various degrees and duration on lipid peroxidation and mineral metabolism in rats. *J. Nutr. Biochem.* **6**:681–688.

23. Oloyede, O.B., Muhammad, N.O. and Oladiji, A.T. (1998) Lipid composition of rat liver and kidney in response to dietary magnesium deficiency. *Nig. J. Pure and Applied Sciences* **13**:599-602.

24. Muhammad, N.O. and Oloyede, O.B. (2001) Effects of dietary magnesium and fatty acids deficiency on the protein and magnesium content of rat liver. *Biosci. Res. Commun.* 13:633-637.

25. Aderibigbe, A.O., Johnson, C.O.L.E., Makkar, H.P.S., Becker, K. and Foidl, N. (1997) Chemical composition and effect of heat on organic matter- and nitrogen-degradability and some antinutritional components of Jatropha meal. *Animal Feed Science Technology* **67**:223–243.

26. Reddy, N.R. and Sathe, S.K. (2002) Food phytates CRC Press, Boca Raton, Florida.

27. Bedford, M.R. (2000) Exogenous enzymes in monogastric nutrition – their current value and future benefits. *Anim. feed Sci. Tech.* 86:2-26.

28. Muhammad, N.O., Adeyina, A.O. and Peters, O.M. (2000) Nutritional evaluation of fungi treated cocoa bean shell. *Nig. J. Pure & Appl. Sci.* **15**:1059–1064.