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Comparative Effects of Moist and Dry Heat on Nutrient Potentials of *Tetracarpidium conophorum* Nut in Rats

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Abstract: The nutritional potential of moist and dry heat treated *Tetracarpidium conophorum* nut was evaluated using male albino rats (45-55 g). Twenty-four (18) weanling rats were divided into three groups of six rats each on the basis of body weight. These rats were fed for thirty-five (35) days. Caesin (CAS), Boiled *T. conophorum* (BTC) and Roasted *T. conophorum* (RTC) were fed these three (3) groups of rats respectively. All chemicals and statistical analysis were based on modern techniques. *In vivo* bioassay revealed that growth food and nitrogen intakes and Protein Efficiency Ratio (PER) were higher in rats fed the RTC diet than in rats fed the BTC diet. However, rats fed the BTC diet gained more weight than those fed the RTC diet. The rats fed the RTC diet had higher food and nitrogen intakes, faecal digested and retained nitrogen, Biological Value (BV) and Net Protein Utilization (NPU) than rats fed the BTC diet. Rats fed the test diets-RTC and BTC- had more than 95% N intake, digested and retained nitrogen. The results indicate that dry-heat treatment improved Nitrogen availability in *T. conophorum* than moist-heat.

Key words: Biological value, protein efficiency ratio, net protein utilization, *Tetracarpidium conophorum*, digested nitrogen, retained nitrogen

INTRODUCTION

In developing countries, animal food products which are better sources of dietary proteins and energy have become very costly and almost out of reach due to economic recession, political instability and civil wars. This necessitates the need to explore other plant sources as alternatives to protein requirements of individuals in these countries.

Generally, nuts, legumes and seeds contain antinutrients which inhibit important pathways in metabolic processes. They reduce utilization of nutrients in the body such as protein, vitamin and mineral (Theil, *et al.*, 1997). Some of these antinutrients occur naturally such as phytates in cereals, oil seeds; trypsin inhibitors, haemagglutinins and tannins in legumes and cereals. However, others such as maillard compounds, oxidized forms of sulphur amino acids, D-amino acids and Lysinoalanine (LAL) are formed during heat or alkaline processing of protein products (Gilani *et al.*, 2005). The presence of some of these antinutrients causes substantial reduction in protein and amino acids digestibility and others bind to vitamins, minerals and enzymes making them unavailable to the body (Gilani *et al.*, 2005). Processing however, significantly reduces the adverse effects of these antinutrients.

T. conophorum nut (conophor nut) commonly called *ukpa* in the Igbo speaking tribes and *awusa* or *asala* in the Yoruba tribes of Nigeria is basically consumed as snacks after boiling or roasting. The nut extracts has been reported to have antimicrobial and antifungal activities (Ajaiyeoba and Fadare, 2006). This work was undertaken to compare the effect of both processing methods on the nutritive value of conophor nut.

MATERIALS AND METHODS

All chemicals used were purchased from Sigma Chemicals, St Louis USA, as analytical grade. The ingredients for diet formulation were bought from Harlan Teklad Inc, Madison, Wisconsin USA. Kits for evaluation of liver and renal functions were purchased from Randox Laboratory Ltd, Ardmore, Diamond Road, Crumin Co, Antrim UK. Kits for electrolyte estimation were purchased from Teco Diagnostics 1268 N Lakeview Ave, Anaheim. CA 92807, USA.

Collection and processing of *Tetracarpidium conophorum*: Fresh and mature conophor nuts were purchased from *Ibeagwa* market, a small market near University of Nigeria, Nsukka. The nuts were mechanically dehusked.

Boiled *Tetracarpidium conophorum* (BTC): The dehusked nuts were boiled for 2 h in a boiling water bath, macerated and dried in a hot air oven at 60°C for 48 h. The dried samples were ground and stored with freezer bags at 4°C until use.

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Roasted *Tetracarpidium conophorum* (RTC): The fresh dehusked nuts were roasted in hot sand (100°C) for 40 min with constant stirring. The nuts were macerated and dried at 60°C after which they were ground and stored with freezer bags at 4°C until use.

Diet formulation: The diets were formulated using AIN 93G method for growth, pregnancy and lactating phase in laboratory rats (Reeves *et al*., 1993).
The ingredients were weighed into a bowl starting with those with larger weights and quantities. Mixing was done manually until a homogenous state was attained. The diets were pelleted manually and the pellets dried in hot air oven at 60°C.

Table 1: Composition of experimental diets (g)

	Diet			
Ingredient	CAS	RTC	BTC	NFD
Casein	114.400	-	-	-
RTC	-	415.220	-	-
BTC	-	-	368.240	-
Cornstarch	572.069	331.500	368.997	663.589
Sucrose	143.017	82.880	92.249	165.897
Fibre	50.000	50.000	50.000	50.000
Soy bean oil	70.000	70.000	70.000	70.000
Mineral mix+	35.000	35.000	35.000	35.000
Vitamin mix+	10.000	10.000	10.000	10.000
L-cysteine	3.000	3.000	3.000	3.000
Choline bitartrate	2.500	2.500	2.500	2.500
BHT	0.014	0.014	0.014	0.014
Total	1000.000	1000.000	1000.000	1000.000

BTC-Boiled *Tetracarpidium conophorum*, CAS-Caesin, BHT-Butylated-hydroxytoluene. AIN-93G diet recommendations (Reeves *et al*., 1993). NFD-Nitrogen Free Diet. +Purchased from Harlan Tekland Inc. Wisconsin USA

Housing and feeding experiment: Twenty-four (24) weanling male albino rats (45-55 g) supplied by the Veterinary Department of the University of Nigeria Nsukka were divided into four groups of six rats each on the basis of body weight. The diets were; Casein (CAS), Boiled *Tetracarpidium conophorum* (BTC), Roasted *Tetracarpidium conophorum* (RTC) and Nitrogen Free Diets (NFD). The rats were housed in individual metabolic cages equipped to separate faeces and urine. After a six day adjustment period, the rats were weighed prior to access to the test diets. The groups of rats were fed 15 g each of BTC and RTC diets as well as casein diet for 28 days. During the growth study, food intake was on daily basis and body weight was recorded on weekly intervals. The NFD group was fed with normal rat chow for 28 days. On the 28th day, 2 mg of carmine red was added to the diets to indicate the beginning of nitrogen balance period. The coloured faeces that appeared on day 29 and subsequent black faeces were retained until another carmine red was added to the diets on day 35. On day 36, only black faeces were collected. The coloured faeces were discarded to mark the end of N balance period. The urine was measured, treated with few drops of 0.01 N HCl and stored with air tight containers in deep freezer. The faeces were dried at room temperature, weighed, ground and stored in freezer bags until analyzed. The respective faecal and urinary samples were pooled for various analyses. The rats were sacrificed on day 36, blood samples were collected for some biochemical analysis using the ocular method. The crude protein content of *T. conophorum*, the diets, faeces and urine was estimated by the AOAC (1995) procedures. The data collected were analyzed using Statistical Package for Social Sciences (SPSS version 12.0). One way Analysis of Variance (ANOVA) and Fisher's Least Significant Difference (F-LSD) were used to separate the means and compare.

RESULTS AND DISCUSSION

The percent crude protein content of the BTC and RTC diets was 27.15% and 24.09% respectively.

Table 2: Growth food and nitrogen intakes, weight gain and protein efficiency ratio of rats fed differently processed *T. conophorum* diets

	Diets		
	CAS	BTC	RTC
Food intake, g	236.50±14.29	166.75±12.84	172.5±32.03
Weight gain, g	54.00±10.74	11.25±5.12	10.25±7.63
N intake, mg	19.87±1.2	7.25±0.56	13.28±2.47
PER	2.72±0.4	1.54±0.67	0.74±0.47

Values are written as means ± SD (6 rats). PER = Protein Efficiency Ratio

Table 2 presents the food and Nitrogen (N) intakes, weight gain and protein efficiency ratio of rats fed three diets. Food intake in the present study was influenced by source of nitrogen. Ene-Obong and Obizoba (1995) observed the same phenomenon in rats. The food intake for the rats fed CAS diet was much higher than those fed test (BTC and RTC) diets. The higher intake might be attributed to improved palatability due to desirable essential amino acid profile of casein. On the other hand, the similar food intake of rats fed BTC and RTC diets (166.75 g and 172.5 g) showed that both diets had the same quality ($p>0.05$); however, the RTC group of rats had an edge over the BTC rats (172.5 g vs 166.75 g). The weight gain of rats fed CAS diet was much more than those of other rats fed BTC and RTC diets (54.0 g, 11.25 g and 10.25 g respectively). The reason for the higher weight gain for rats fed casein diets was because these rats consumed more and retained more for tissue synthesis. Obizoba (1985) observed that weight gain of rats fed different diets might partly be influenced by food intake. The rats fed RTC diet had higher food intake but lower weight gain than those fed BTC diet. Sugiura *et al*. (1998) had earlier noted that moderate thermal treatment in humid conditions (such as boiling) enhances digestibility due to partial protein unfolding and destruction of trypsin inhibitors. It appears that boiling enhanced digestibility of *T. conophorum* protein much more than roasting to increase availability for body protein anabolism.

The higher Nitrogen (N) intake of rats fed CAS diet ($p<0.05$) over those of rats fed test diets could be due to high food intake and better pattern of essential amino acid content of casein. This explanation could be equally true for rats fed the RTC diet that had higher food and N intake than those rats fed the BTC diet.

The Protein Efficiency Ratio (PER) of rats fed CAS diet was higher than those fed test diets. This probably indicates the superiority of CAS diet over those of test diets.

Table 3: Growth performance and nitrogen utilization of rats fed differently processed *T. conophorum* based diets

	Diets		
	CAS	BTC	RTC
Food intake, g	36.25±10.04	39.25±3.59	36.25±6.39
Weight gain, g	2.75±0.26	4.75±0.95	3.50±0.90
N intake, mg[a]	3.05±0.84	1.71±0.15	2.79±0.49
Faecal N, mg[1]	0.27±0.03	0.27±0.02	0.34±0.04
Digested N, mg[1]	2.78±0.81	1.44±0.13	2.45±0.45
Urinary N, mg[1]	0.04±0.01	0.13±0.02	0.05±0.02
Retained N, mg[1]	2.74±0.80	1.31±0.11	2.40±0.43
Biological value	98.56±0.98	90.97±0.84	97.96±0.96
Net protein utilization	89.84±0.95	76.61±0.73	86.02±0.88
True digestibility	91.14±0.96	84.21±0.86	87.81±0.91

Values are written as means±SD (6 rats).
[1]Corrected for endogenous N losses.
[a]7-day N intake

Table 3 presents food and nitrogen intake, weight gain, digested N, faecal and urinary N, retained N, biological value and net protein utilization of groups of rats fed three different diets. The food intake of the groups of rats differed. The range was from 36.25 g to 39.25 g. However, the groups of rats fed the Casein (CAS) and the RTC diets had comparable food intake (36.25 g). The group of rats fed the BTC diet had 3.0 g food intake more than those of the rats fed the CAS and the RTC diets. The slightly higher food intake of the rats fed the BTC diet (39.25 g) might be due to the sources of protein and their essential amino acid profile. It is known that rats consume more food if the essential amino acid profile of the protein is desirable to produce palatability. The weight gain of the groups of rats was a function of food intake. The BTC groups of rats had the highest weight gain when compared with the other two groups of rats-CAS and RTC (4.75 g, 2.75 g and 3.50 g, respectively). This might be that the BTC diet had better pattern of essential amino acids which the rats utilized to synthesize more body protein than others. Ezeokonkwo and Dodson (2004) reported that quality of a given protein influences body weight of animals (rats). The Nitrogen (N) intake of the groups of rats varied, it ranged from 1.71 mg to 3.05 mg. The group of rats fed the CAS diet had the highest N intake (3.05 mg) followed by that of the group fed the RTC diet (2.79 mg). The group of rats fed the BTC diet had the least (1.71 mg). The higher N intake for the rats fed the CAS diet might be attributed to (a) its better essential amino acid profile and (b) its high palatability. These two factors are known to influence food and N intake in rats.

The faecal N output of the groups of rats differed. The CAS and the BTC groups of rats had comparable faecal N (0.27 mg). The RTC group of rats had slightly higher facal N output (0.34 mg).

The urinary N output of the groups of rats varied. The variation ranged from 0.04 mg to 0.13 mg. The group of rats that consumed less N had the highest urinary N output (0.13 mg). On the other hand, the CAS and the RTC groups of rats had similar urinary N output (0.04 mg and 0.05 mg). The retained N for the groups of rats differed. It ranged from 1.31 mg to 2.74 mg. The CAS group had the highest N retention (2.74 mg) followed by the RTC group (2.40 mg). However, the BTC group of rats had the least value (1.31 mg). The higher N retention of the group of rats fed the CAS diet was due to its low faecal and urinary N output. The BTC diet had the least N retention because of its high faecal and urinary N output.

The Biological value for the three groups of rats was influenced by protein quality and intake as well as metabolic output (faeces and urine). The Biological Value (BV) of the groups of rats fed CAS and the RTC diets had the highest and comparable values as against that of the BTC group of rats (98.56% and 97.96% vs 90.97%). The lower BV for the BTC group of rats (90.97%) was due to high faecal and urinary N excretions as well as lower N intake. The net protein utilization of the CAS group of rats was much higher than those of the BTC and RTC groups of rats (89.84%, 76.61% and 86.02%, respectively). The lower net protein utilization for the BTC group of rats demonstrated its poor protein utilization. The poor N utilization might be attributed to low N intake and higher faecal and urinary N excretion.

Conclusion: The results discussed so far strongly suggests that *T. conophorum* nut contains high quality protein capable of supporting growth and maintenance of tissues in rats. However, dry heat treatment seem to enhance the protein qualities as seen in the BV, NPU and TD results which favoured the maintenance of body tissues, although the wet heat treatment improved the protein qualities which favoured growth of the weanling rats over dry heat treatment.

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