



The Potentiality of Processed Mango Kernel in Animal Feed

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors designed the study. Author AO performed the statistical analysis and wrote the protocol. We both managed the analyses of the study as well as the literature searches. Both authors read and approved the final manuscript.

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ABSTRACT

Biological quality of processed mango (*Mangifera indica*) kernel (PMK) in a 50% substituted commercial feed was evaluated as soluble protein equivalent (spe) in a feeding experiment with albino rats. The methodology employed was novel and involved determination of protein retention using whole uncentrifuged blood sample. The rats of the 'test' group showed similar protein efficient ratio (PERspe) with those of 'control', despite the difference in total crude protein contents of their diets (11 and 16% respectively). However, higher values of BVspe and NPUspe were observed with the test diet, implying that available essential amino acids there-in supported growth and maintenance better than with the control diet.

Keywords: *Mangifera indica*; processed mango kernel; soluble protein equivalent; biological quality.

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1. INTRODUCTION

Mango (*Mangifera indica*) plant of the member of *Anacardiaceae* family yields one of the most celebrated fruits in tropical countries. The tasty fleshy mesocarp of the ripe fruits are eaten raw, and manufactured into juice and jellies [1]. For several decades, the stony endocarp which comprises the shell, testa, and kernel have been discarded as waste in Africa whereas in Asia, interest has mainly been on the lipid component of the kernel because of its potential application in the confectionery industry as a source of cocoa-butter substitute [2].

The usefulness of the whole mango kernel, the defatted kernel flour and the oil, in comparison with mango juice has not yet been harnessed in both cottage and large-scale industries in Africa, and particularly Nigeria. Consequently, our laboratory embarked on waste conversion programme of the abundant but discarded Nigerian mango kernel.

Our knowledge of a local (Ikanekpo) mango variety handled on several occasions shows that the fresh kernel constitutes 7% of the weight of the fruit, and 48% of the weight of stony endocarp (seed) [3]. Arogba further reported the level of unsaturated fatty acids as twice those of the saturated; and in comparison with standard proteins, each of six essential amino acids available was greater than 70% [3].

Further studies successfully showed that the kernel was convertible to edible state by processing. These studies bordered on composition, toxicology, and functionality of the kernel before and after processing into flour [3], the browning activity of its polyphenol oxidase [4], characterization of the tannic constituents [5], employing the *processed* flour in biscuit-formulation [6], characterization of the biscuit with respect to effect of temperature on moisture adsorption behavior [7], quality/shelf-characteristics of the biscuit [8], and phenolics as well as cytotoxicity of the processed flour [9].

The present study reports on the nutritional contribution of the processed mango kernel (PMK) to a diet of growing albino rats. The analytical design of the experiment employed soluble protein fractions of the feed and blood of fed rats to compute and express PER, NPU, NPR, apparent BV, and nitrogen digestibility (AND) as their soluble protein equivalent (spe).

This methodology is novel and the justification is given in the Discussion section.

2. MATERIALS AND METHODS

2.1 Materials Collection and Handling

Freshly discarded mango seeds of Ikanekpo variety collected in Anyigba town (Kogi State, Nigeria) and its environ during the ripening season of March to May 2004.

The methodology of Arogba, [3] was replicated to obtain PMK for use in a test feed in this study. The PMK was hand-milled into granules of a physical state similar to that of a commercial feed to be used.

The commercial feed, branded as VITAL, was purchased from a local market at Anyigba. The company, Grand Cereals and Oil Mills Co. Ltd., located at Bukuru, Jos, Nigeria declared crude protein content of 16% for the batch #,k051756.

Twenty albino rats comprising ten males and ten females were purchased from a local breeder in the locality. Weights ranged between 84 and 270 g. Rats of 84 to 175 g constituted the test group while those of 200 to 270 g were for control. Nevertheless, each group was subdivided into cages on the basis of gender to avoid mating. They were housed in our research laboratory with an ambient temperature range of 27°C, 50 to 60% relative humidity, and 12 hour cycle of light and dark.

2.2 Experimental Design

Five rats were taken from each group and of the same gender, on weekly basis for four weeks thus (Table 1).

2.3 Feeding Experiment

Experiment diets comprised the unsupplemented commercial feed which served as control, and the feed supplemented at 50% weight by proportion with the PMK granules served as test diet. Formulation of the latter provided the nearest isonitrogenous diet of 11% protein (in comparison with the expected 10%). Water was provided *ad libitum*.

Weights of whole rats were taken on zero day. Similar measurements were taken on weekly basis and also for their respective kidneys and liver after anaesthesia and dissection in order to assess weight gain and growth rate.

Table 1. Experimental design. Five rats were taken from each group and of the same gender, on weekly basis for four weeks thus

Control	Week 1	Week 2	Week 3	Week 4
Male (5+)	+	-	+	-
Female (5+)	-	+	-	+
Test				
Male (5+)	+	-	+	-
Female (5+)	-	+	-	+

To avoid excessive spillage and contamination, 30 g of feed were introduced daily into plastic bottles previously drilled on the near-bottom side to a diameter of 3 cm and each firmly placed in the cages.

Residual feeds and faeces were collected separately, weight on weekly basis, and amounts of feeds consumed were calculated accordingly. The care and use of the animals as well as the experimental protocol of this research were in accordance with the Experimental Animal Care and Use regulation of Kogi State University, Nigeria, which were also in agreement with the internationally accepted principles for laboratory animal use (EEC Directive of 1986; 86/609/EEC).

2.4 Statistical Analysis

All the data were analysed using GraphPad prism 6. Results were expressed as mean data \pm the standard error of mean (Mean \pm SEM) of n independent experiments. n numbers are represented in figure legends. Student's t -test was used to determine the level of significance between the two groups. $P < 0.05$ was used as criteria for statistical significance.

2.5 Chemical Analysis

Blood samples were withdrawn from the heart using plastic disposable syringes within two minutes of anesthesia and dissection of a rat. Following some trial experiments to avoid coagulation, known weight of 1.0 ml blood sample was immediately mixed with distilled water and made up to 50 ml-mark of a measuring cylinder on each occasion.

Two (2) ml of the diluted blood sample was then used for total soluble (serum + plasma) protein content determination, employing the Biuret method [10]. Corresponding concentration for samples analyzed on weekly basis were derived by extrapolation from linear regression curves prepared similarly and using a standard commercial albumin as reference protein.

The soluble protein contents of the feeds, and faeces were analyzed in similar manner described above.

In all instances, measurements were replicated in triplicate, and results expressed per 100 g-body wt. of rat to ease comparison.

2.6 Dimensional Analysis

The contribution of soluble protein equivalent (**spe**) of the diets to biological quality was evaluated using the following parameters:

Protein intake as

(Average gram feed consumed x Soluble protein factor).

Protein efficiency ratio (PER) as

$PER_{spe} = (\text{Av. Wt. gain on diet} \times \text{Soluble protein factor}) / \text{Soluble protein intake}$

Nitrogen retained as equivalent of

(Av. Protein concentration in blood).

Nitrogen absorbed as equivalent of

(Protein intake – Faecal protein).

Biological value (BV) as apparent

$BV_{spe} = \text{Soluble protein retained} / \text{Soluble protein absorbed}$

Net protein utilization (NPU) as apparent

$NPU_{spe} = \text{Soluble protein retained} / \text{Soluble protein absorbed}$

Net protein retention (NPR) as

$NPR_{spe} = NPU_{spe} / 0.16$

Apparent nitrogen digestibility (AND) (Lape & Treche, 1994) as

$$\text{AND}_{\text{spe}} = (\text{Soluble protein retained} / \text{Soluble protein absorbed}) = \text{NPU}_{\text{spe}} / \text{BV}_{\text{spe}}$$

Urinary nitrogen as equivalent of urinary protein (U_p) where

$$U_p = \text{protein intake} - [\text{Digestibility factor} \times \text{Absorbed protein} \times \text{protein intake} \times (1 - \text{Digestibility factor})] / [\text{Absorbed protein} - (\text{Digestibility factor} \times \text{protein intake})].$$

body weight basis, consumed more of the test diet daily by about 23% (Test: 22.23 ± 0.7 versus Control: 17.33 ± 1.06 ; $P=0.04$; Table 5).

Nutritive value of diet partly depends on degradability of the nitrogen fraction of the diet [12]. Degradability, in turn, is principally affected by the amino acid sequence within the protein. Consequently, growth and biological maintenance must be satisfied through the quality of available protein [13]. This fact explains the lower digestibility index (AND) of the test diet observed in this study (Test: 64.8 ± 1.66 versus Control: 77.6 ± 1.39 ; $P=0.05$; Table 5), despite the difference in their crude protein contents.

3. RESULTS AND DISCUSSION

Incorporation of similar processed mango kernel (PMK) flour in model human food system (biscuit) at 50% wheat flour supplementation was reported by Arogba [6,8]. For this study, PMK was reproduced by similar methodology and used to supplement rat diet at the same level of substitution. The test diet had pH of 5.7 while the control had (6.1). The presence of residual tannin of about 2.3% by weight [3] could justify the lower pH value of the test diet. The latter has nutritional advantage over the control diet in this respect for the known efficacy of tannin as a cholesterol lowering agent in rats [11]. The added advantage with the formulation was that palatability of the diet improved as rats, on 100 g

On the contrary, soluble protein intake and weight gained there-of were maximal and commensurate with the 11 and 16% crude protein contents of the test and control diets indicated earlier in this report. It could be implied therefore that the composition of the test diet had no adverse nutritional effect on the metabolism of the rats. Rather, that both groups of animals exhibited similar protein efficiency ratio (PER) suggested that the test diet adequately met the needed maintenance requirement as the control diet. Similarly, the protein retention values also tended to support the inference. The chemical protein quality of PMK applied in this formulation had been reported by Arogba [3].

Table 2. Soluble protein concentration in blood (g/wk/100 g rat)

Animals	Week 1	Week 2	Week 3	Week 4
Test	$0.182 \pm 0.03^*$	0.191 ± 0.08	0.202 ± 0.06	0.202 ± 0.07
Control	0.102 ± 0.01	0.163 ± 0.03	0.233 ± 0.02	0.263 ± 0.04

All data are presented as mean \pm SEM; $n=5$ in each group; $*P<0.05$, Student's *t*-test was used to compare the two groups tested; test versus control

Table 3. Soluble protein concentration (or factor) of feed and faeces (g/100 g sample)

Animals	Feed	Faeces
Test	$2.22 \pm 0.01^*$	5.00 ± 0.08
Control	4.18 ± 0.02	5.26 ± 0.04

All data are presented as mean \pm SEM; $*P<0.05$, Student's *t*-test was used to compare the two groups tested; test versus control. This represents three independent experiments

Table 4. Growth rate of kidneys and liver (g/wk/100 g rat)

Organs	Week 1	Week 2	Week 3	Week 4	Average
Kidney (test)	0.673 ± 0.11	0.444 ± 0.08	0.654 ± 0.01	0.574 ± 0.02	0.586 ± 0.04
Kidney (control)	0.673 ± 0.03	0.716 ± 0.05	0.679 ± 0.09	0.666 ± 0.06	0.683 ± 0.04
Liver (test)	4.088 ± 0.03	2.942 ± 0.09	4.035 ± 0.08	2.934 ± 0.02	3.500 ± 0.05
Liver (control)	3.328 ± 0.07	3.002 ± 0.03	3.383 ± 0.09	3.202 ± 0.00	3.229 ± 0.11

All data are presented as mean \pm SEM; $n=5$ in each group; Student's *t*-test was used to compare the two groups tested; test versus control

Table 5. Other nutritional parameters

Parameter (Av.grm/wk/100 g rat)	Test	Control
Weight gain (diet)	22.23±0.7*	17.33±1.06
Weight gain (protein)	0.489±0.01*	0.728±0.02
Protein intake	2.018±0.02*	3.258±0.06
Faecal protein	0.710±0.01	0.731±0.01
Protein retained	0.202±0.02	0.231±0.03
Protein absorbed	1.308±0.04	2.527±0.05
PER _{spe}	0.242±0.01	0.223±0.02
BV _{spe} %	15.44±0.52*	9.140±0.31
NPU _{spe} %	10.01±0.18	7.091±0.92
NPR _{spe} %	62.56±0.12*	44.31±0.26
AND _{spe} %	64.8±1.66*	77.6±1.39
UP (computed)	-0.138±0.08	+1.235±0.01

All data are presented as mean ± SEM; n=5 in each group; *P<0.05, Student's t-test was used to compare the two groups tested; test versus control

Table 4 showed similar average growth rate of the liver samples of both group of animals. Even the firmer organs, such as the kidneys, did not show any appreciable differences in growth rate (P=ns). Further nutritional advantages of the formulated diet over the control were observed in the higher BV and NPU data (Table 5).

It is known that as the amount of dietary protein increases beyond 'NPU_{standardized}', more is catabolized and proportionally less is retained in body protein [12]. The fact is further buttressed by previous studies of Nti and plahar [13] where lower BV, TND, and NPU were recorded with cowpea supplemented diet of 81% protein score than that with maize of 42%. Indeed, the mathematical relationship between these three parameters could explain the positive correlation associated with the computed UP of the control animals in contrast to the negative value obtained using the test diet (Test: -0.138±0.08 versus Control: +1.235±0.01; Table 5).

Since the test diet did not seem to have altered the metabolic pattern of the test animals, it was not surprising that both groups of animals experienced maximum protein intake, as well as similar excretion rate of faecal protein (Table 3). It implied maximum soluble protein absorption resulting from optimum peptidase activity had occurred in their intestinal tracts.

Comparing the computed retained protein (Table 5) with the weekly protein concentrations in the blood (Table 2) of both groups of rats, it appeared the animals had acclimatized to the diets provided by the end of third week. The observation could partly justify the following

claims of the methodology adopted for this experimentation:

- (i) Nitrogen intake minus that excreted in faeces and urine was equated to nitrogen retention and was determined by analysis of *total* soluble protein content. Total blood protein content was a better index than serum or plasma protein alone ad serum: plasma ratio was found to be (1.0): (5.9).
- (ii) Protein content is a product of nitrogen content and a factor. Lack of knowledge of the factor for evaluating nutritional parameters does not affect computation.
- (iii) For routine purpose, it is simpler to conduct the colorimetric analysis than use the kjeldahl method.
- (iv) The technique is valuable for comparing nutritional qualities of two or more animal diets.
- (v) In assessing apparent NPU, the carcass analysis is avoided by direct analysis of the blood sample.
- (vi) In the absence of standard metabolic cages, it is easier to collect faeces alone for faecal protein determination in contrast to urine.
- (vii) Apparent nitrogen digestibility (AND) can be derived easily from the ratio of the protein absorbed to protein intake without the labour involved by experimental technique.

4. CONCLUSION

The study has shown that formulated animal feed containing 50% *processed* mango kernel would support growth and maintenance of albino rats, and much so if the diet is NPU_{standardized}.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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