Proximate Biological Value (BV) and Protein Efficiency Ratio (PER) of Processed Bush Mango (*I. gabonensis*) Kernel (PBMK).

Arogba, Sunday S.¹ & Matanmisi, Julius A.²

^{1,2}Biochemistry Department, Kogi State University, PMB 1008, Anyigba. Nigeria.

Abstract: A novel proximate technique of determining soluble protein equivalents (spe) of Protein Efficiency Ratio (PER) and Biological Value (BV) using Biuret method, is described. The protein quality of processed Bush mango (I. gabonensis) kernel (PBMK) in five feed formulations of Morinda lucida leaf-PBMK was studied. Young goats weighing 5.4 ± 0.5 kg were fed for seven weeks with 25, 50, 75, and 100% PBMK substitutions in the feeds. Results showed that an optimum substitution of 50% promoted liver growth, overall body weight gain, and improved biological protein status of the animals employed.

Keywords: protein quality, proximate indices, soluble protein equivalent, BVspe, PERspe.

I. Introduction

Protein nutrition serves to provide the required amino acids for body protein retention, and maintenance. Albumin and other plasma proteins aid the maintenance of blood viscosity, osmolarity, and transportation of lipids and other solutes. Issues of protein quality are of concern especially in third world countries where there is poor quality protein-intake as well as insufficient total caloric intake, which are associated with restricted dietary patterns. Supplementation with other foods becomes a necessity. In contrast to high quality proteins, low quality proteins are deficient in one or more essential amino acid(s) which limit the rate of protein synthesis [1]. The quality of protein is measured by the relative proportion of essential amino acids in a food with that required for good nutrition, using egg or milk protein as standards in most instances [2,3]. Since excess amino acids are not stored for future use, they are rapidly degraded and the nitrogen is excreted as urea and other products through urine, faeces, and desquamating skin.

Popular and measurable indices of protein quality include BV, and PER. Others are Net Protein Utilization (NPU), Chemical Score (CS), and most recently, Protein Digestibility Corrected Amino Acid Score (PDCAAS). The United States of America Food and Nutrition Board estimated from nitrogen balance studies that an average maintenance requirement for an adult was 0.47g per kg body weight per day allowing for excretory losses, 30% variability in protein metabolism and 25% variations in biological value of dietary proteins.

Orthodox standard methodologies for protein quality determination have limited relevance when results are interpreted, for instance, BV and CS do not account for digestion and actual absorption of protein unlike PDCAAS and NPU; PER is always measured using young rats and argued to be of little relevance to human physiology; PDCAAS accepts only a highest possible score of 1.0, regardless of the apparent protein quality when a protein contains essential amino acids in excess of human requirements. Furthermore, these orthodox methodologies are encumbered by duration to accomplish experimentation, sensitivity to digestibility factor, overall energy consideration, availability of equipment, and cost. For these reasons, there has been continuous modifications of biological methods for protein evaluation to make them simpler and vet maintaining accuracy. For example, NPU experiments changed from use of whole carcass, through hind limb, to liver [4,5,6]. In similar vein, we have in the past five years developed in our laboratory speedy, economical and reproducible proximate technique to evaluate 'soluble protein equivalent's (spe) of BV and PER. Samples are drawn from the bloodstream for analysis to account for digestion, absorption, energy consideration, and possible protein losses in urine and faeces. Results obtained are independent of experimental animals employed. The experimental feeds provide sufficient protein and energy required for maintenance of tissue, and growth of the animal. The present study, therefore, reports this application on the comparative biological qualities of PBMK and a conventional animal feed of Morinda lucida leaves fed to goats for seven (7) weeks.

II. Materials And Methods

2.1. Procurement and handling of raw materials

Bush mango (*Irvingia gabonensis*) seeds, and (female) goats were purchased at Anyigba (Kogi State, Nigeria) central market while *Morinda lucida* leaves were plucked from shrubs within Anyigba and its environ.

The methodology of Arogba[7] was adopted to obtain processed BM kernels. Testa was retained. The dried kernels were ground manually into powdery form (grits). Similarly, the leaves of *M. lucida* were ovendried at 65° C until constant weight was achieved, cooled, and ground.

2.2. Experimental design

The dried ground leaves of *M. lucida* were substituted at ordinal intervals of 25% with PBMK grits and fed to five young she-goats weighing 4.8 to 5.9 kg, in fabricated cages, for 7 weeks. Water was provided adlibitum. The sixth animal which served as a reference, was sacrificed on the first day of the experiment (zero day). Over the period, physical weights of animals, liver, kidney, feeds, and faeces were taken. Soluble protein concentration equivalents (spe) in the blood, feeds, and faeces were determined by biuret method [8]. The blood samples (2.5ml each) were freshly syringed on sacrificing the animals, and made up to 100ml with normal saline. Varied volumes (0.2 - 1.0ml) were withdrawn for use in the biuret test. Each determination was replicated twice. At the end of 7th week, parameters measured were respectively 'normalised' with respect to the weight of the reference animal.

2.3. Formulae

 $PER_{(spe)}$ Weight gain x soluble protein factor / soluble protein intake Where: Weight gain \equiv grams of gain /normalised body weight of animal Protein factor \equiv gram protein / gram feed Protein intake \equiv feed consumed x protein factor $BV_{(spe)}$ Soluble protein retained / soluble protein absorbed Where: Soluble protein absorbed \equiv soluble protein intake – soluble protein in faces

III. Results And Discussion

Early researchers had experienced low food intake by animals in short periods of investigation and so, advocated long experimental period of between 4 and 8 weeks to allow for more consistent food consumption [9]. Indeed, previous experimentation with bucks fed for four weeks in our laboratory yielded no significant increase in weight gain. However, the seven weeks duration employed on this occasion has shown overall body weight gain in all animals of $5.1 \pm 0.4\%$, but the order of % gain in organ weights as reflected in kidney / liver was 68/56, 11/57, 9/19 at 0%, 50%, 100% feed substitution respectively. It implied that weight gain by kidney decreased with increased PBMK level in feed. By its excretory function, vital nutrients such as soluble proteins and minerals could have been bio-unavailable through chelation with some phytochemicals. The earlier reports of Arogba et al [10] and Arogba[7,11,12,13] identified the presence of polyphenolic substances, particularly tannin, in the conventional mango (*Mangifera indica*) and *I. gabonensis* kernels (PBMK). Combined soaking and heat treatments during processing was further stated to appreciably reduce the anti-nutritional effects of such phytochemicals.

The residual concentrations of the phenolics in the processed kernels is implicated in the growth performance of experimental animals through their antioxidant activity [14], and could explain why liver growth was not adversely affected up to 50% PBMK inclusion. The observation was further supported by results in Tables 1 and 2. There was progressive increment of soluble protein concentration in the blood as substitution with PBMK increased. The central role of the liver in integrated metabolism identified with the overall weight gain observed and the BV*spe* and PER*spe*, for which statistical separation of mean scores of the five feed formulations are shown.

The BV*spe* and PER*spe* values increased proportionally with the percentage PBMK in feed compared with the use of M. lucida leaves alone. The correlation coefficient, r, was calculated to be 0.9871. Similar correlation has been reported between NPU and PER in a single experiment [9]. While the observations from this work and literature might not be surprising based on the relationship in definition between BV and NPU, the results obtained in this experiment validate the reliability of the technique employed. From the experimental data on BV*spe* and PER*spe*, the 50% substituted PBMK feed described an optimum level, of statistical significance (p < 0.05) in the array. It was therefore, recommended as the best animal feed in the formulations for physiological and economic reasons.

IV. Conclusion

The study has described a sensitive and reproducible proximate technique for biological protein quality assays. A 50% PBMK substitution in feed formulation promoted liver growth, overall body weight gain, and improved protein quality status of livestock.

TABLE 1: PROTEIN EFFICIENCY RATIO (PERspe) of M. lucida leaf-PBMK formulations fed to goats for 7 weeks

	Feed formula						
Animal	1	2	3	4	5		
% PBMK in feed	0	25	50	75	100		
Soluble protein factor in feed (g/g))A)	0.275	0.301	0.325	0.373	0.399		
Total feed consumed (g)	1758	1664	1564	1521	1505		
Total feed consumed per 'Normalized body Weight' (g/g) (B)	0.285	0.269	0.252	0.245	0.242		
Soluble protein intake for 7 weeks (g/g) (C)	0.078	0.081	0.082	0.091	0.097		
Weight gained (g)	270	280	300	300	320		
Weight Gained Per Normalized Body Weight (g/g) (D)	0.044	0.045	0.048	0.048	0.051		
Weight gained $_{(spe)}$ (g/g) (E)	0.012	0.014	0.016	0.018	0.021		
$PER_{(spe)}$ (se=0.001) (F)	0.154 ^d	0.168 ^c	0.191 ^b	0.198 ^b	0.211 ^a		
(Means with different letters across a row differed significantly at $p = 0.05$)							

Where:

spe = soluble protein equivalent

PER= protein Efficiency ratio $C = A \times B$ $E = A \times D$

 $E = A \times D$ F = E / C

se = Standard error

TABLE 2: BIOLOGICAL VALUES (BVspe) of M. lucida leaf-PBMK formulations fed to goats for 7 weeks

	Feed formula						
Animal	1	2	3	4	5		
% PBMK in feed	0	25	50	75	100		
Soluble protein in faeces g/normalized body wt (A)	0.050	0.120	0.131	0.216	0.215		
Total weight of faeces for 7 weeks (gram)	634	636	628	640	632		
Total weight of faeces in 7 weeks per normalized body weight (g/g)	0.103	0.103	0.102	0.103	0.102		
(B)							
Total soluble protein in faeces g/g normalized body weight (C)	0.0052	0.0124	0.0132	0.0222	0.0219		
Total soluble protein intake from feed (g/g) (D)	0.078	0.081	0.082	0.091	0.097		
Soluble protein absorbed (g/g) (E)	0.073	0.0686	0.0688	0.0688	0.0751		
Soluble protein absorbed (mg/g) (F)	73.00	68.60	68.80	68.80	75.10		
Soluble protein retained in the blood (mg/g) (G)	4.67	4.75	5.53	5.65	6.34		
BV (spe) (se=0.4) (H)	6.4 ^c	6.9 ^b	8.0 ^a	8.2 ^a	8.4 ^a		

(Means with different letters across a row differed significantly at p=0.05)

BV=Biological value

spe = soluble protein equivalent

C = A x B, E = D - C, H = G x 100/F

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