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IN-VIVO QUALITY ASSESSMENT OF *Dambu-nama* TREATED WITH SOME HURDLES

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----------ABSTRACT----------
A study was conducted to assess the protein quality of beef based Dambu-nama "A shredded dried Nigeria meat product" by IN-VIVO studies using Swiss Strain Albino rats. Four hurdles were used in the preparation of three samples namely; Citric acid at 0.1 (B), 0.2 (D) and 0.3 (G) % levels of concentration, Salt at 2.0%, Sugar at 4.0% and All spices (ginger, garlic, onions, hot pepper, sweet pepper and magi cube) was added at 2.0% and 4.0% respectively and a control (AB) which was not treated with any hurdles. Result showed that the proximate composition of the test diets B, D, G, AB, BD and CD (Casein diet) , varied significantly at (P≤0.05). All diets recorded positive weight gain except BD (Basal diet) that was negative. The Protein Efficiency Ratio (PER), Protein Utilization (PU), Net Protein Ratio (NPR) and Apparent digestibility (AD) values varied significantly (P≤0.05) in all diets. Faecal nitrogen, liver nitrogen and carcasses nitrogen also varied significantly in all at P≤0.05. Rats fed basal diet (BD) recorded a negative PER (-2.26) while those fed on 0.3% citric acid (G) have a value of 2.32and those fed on 0.2% 0.1% citric acid (D and B) have a PER value of 2.34 and 2.40 respectively and the control (AB) which was untreated have a lower PER value of 2.18. This implies that the use of citric acid significantly increase the protein quality of dambu-nama and may as well increase the shelf-stability due to the reduction of the activity of micro-organisms.

***KEYWORDS**: Dambu-nama, In-vivo, Albino Rats, Hurdles, Protein Efficiency Ratio (PER)*

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I. INTRODUCTION

The dearth of animal protein in the diets of persons living in developing countries has been an issue of concern to Government and Individuals over the years. This is because what is obtained from the major sources of meat supply can no longer sustain the growing demand due to increase in human population (1). In order to bridge the gap between demand and supply of animal protein, preservation of meat is therefore necessary.
The microbial safety and stability as well as the sensory and nutritional quality of most foods is based on an application of combined preservative factors called hurdles (2). From an understanding of the hurdle effect, the hurdle technology has been derived (3), which means that hurdles are deliberately combined to improve the microbial stability and the sensory quality of foods as well as their nutritional and economic properties. Thus, hurdle technology aims to improve the total quality of foods by application of an intelligent mix of hurdles.
According to (2), the most important hurdles used in food preservation are temperature (high or low), water activity (a_w), acidity (pH), redox potential (Eh), preservatives (e.g. nitrite, sorbate, sulfite), and competitive micro-organisms (e.g. lactic acid bacteria).

Some hurdles (e.g. mallard reaction products) will influence the safety and the quality of foods, because they have antimicrobial properties and at the same time improve the flavour of the products. The same hurdles could have a positive or a negative effect on foods, depending on its intensity. In developing countries the application of hurdle technology for foods that remain stable, safe and tasty if stored without refrigeration is of paramount importance. The inclusion of animal products in the diet of an average Nigerian has been diminishing from year to year mainly due to poverty, high cost of animal feeds and political and economic instability coupled with poor infrastructural development (4).The use of citric acid, salt, sugar and all spices hurdle combination for this study significantly improved the sensory, microbial stability and nutritional quality of the *dambu-nama*. Citric acid, a food acidulant is not only often used in meat marination to improve the water-holding capacity and tenderness of beef muscle but is also commonly used as a chelator to control the activity of pro oxidants metals (5, 6). The salt, sugar and spices also play preservative roles as well as improving the sensory and nutritional quality of dambu-nama.

The objective of this research is to investigate the effect of citric acid, salt, sugar and spices (hurdles) on the nutritional and sensory qualities of *dambu-nama* using albino rats as the vehicle.

II. MATERIALS AND METHODS

Sample Procurement and Preparation

Pre-rigored beef from the hind quarters was purchased from Makurdi International Market abattoir. All spices (onions, ginger, garlic, sweet pepper, hot pepper and maggi), salt, sugar and citric acid (food grade) were also purchased from Makurdi International Market. Grand cereal vegetable oil used for frying was purchased from a supermarket in Makurdi. Four kilogram (4kg) of beef was used for each treatment (four) totalling 16kg. The beef was trimmed of fats and connective tissue and cut into nearly equal sizeable sizes and then cured in 0.02g of meta-bisulphate for 30 minutes (7). 500ml of clean tap water was poured into each treatment containing 4kg of beef, 0.1, 0.2 and 0.3% of citric acid, 2.0% of salt and sugar and 4.0% of all spices (processed into dry powder), mixed properly and cooked for 90 minutes. The cooked tender beef was then pounded into a soft matrix using pestle and mortar. The matrix which is fluffy in nature was then separated manually. The shredded fluffy beef was dried briefly for 30 minutes at 60^oC to reduce the moisture content before frying in a hot air oven . The beef was then fried mildly for 10 minutes and re-dried in a hot air oven at 60^oC for 3 – 4 hrs. The shredded dried meat (*Dambu-nama*) is then cooled and packaged in a high density polyethylene (HDPE) from were samples were drown for subsequent analysis.

Feeding Trials with Rats

Swiss strain of albino rats aged between 10-12 weeks were purchased from NITROM (National Institute of Trypanosomiasis Research, Vom, Plateau State) and were used for the feeding experiment following a modification of the method described by (8) as reported by (9). Thirty rats were randomly distributed to six stainless cages with 5 (five) rats per cage. The rats were given food and water *ad Libitum*. Each of the animal groups was fed one of the test diets (B, D, G, AB, CD and BD) respectively. Weight gained by the rats and food consumed were measured every day for 21 days. Faeces were collected daily during the first 7 days and last 7 days and stored in a freezer. The faeces were later pooled together, thawed, air (sun) dried and ground into powder, packaged in airtight bottles until when required for analysis. At the end of the twenty one (21) days of the experiment, the rats in each cage were scarified following the procedure described by (9). The rats were anaesthetised using chloroform; the dead rats were incised vertically through the stomach, the neck and the head. Individual organ were removed, weighed separately and dried in a hot air oven at 60^oC. Subsequently ground into powder and packaged in an airtight plastic container from where samples were drawn for Nitrogen analysis.

Assessment of Protein Quality

Protein quality of *Dambu-nama* – a shredded dried Nigerian meat product which were fed to weaning albino rats was evaluated following method described by (8) were used to calculate PER, FCE, NPR, PU, AD and TPD respectively.

Statistical Analysis

Data collected were subjected to statistical analysis of variance (ANOVA) and significant differences between means were separated using Duncan's Multiple Range test using SPSS 17.0 package.

III. RESULTS AND DISCUSSION

The results of the proximate composition of the test diets fed to the albino rats is as presented in Table 1. The crude protein value showed some level of significance ($P \leq 0.05$) and ranged 16.45% to 28.25% respectively. The basal diet (BD) has the lowest while diet treated with 0.2% citric acid (D) has the highest value. Table 2 shows the result of nitrogen content of diets, carcasses, liver and faeces. Significant difference exist at ($P \leq 0.05$) for the nitrogen content in liver, faeces and diets but no significant difference for nitrogen in carcasses. According to (10) higher liver nitrogen is a function of protein quality, nitrogen intake and digested nitrogen because nitrogen is used in the synthesis of liver. The result of the body weight changes, feed intake and feed conversion efficiency of albino rats fed with test diets (*dambu-nama)* is as presented in Table 3. The mean weight gain (MWG) for the rats fed diets which was not treated with any hurdles (AB-control) was lower (46.45g) and the group fed basal diet (BD) have a weight loss (-2.68g). The nitrogen intake and protein intake showed some level of significance at ($P \leq 0.05$) for rats fed the test diets. This observation is in agreement with the reports of (9) who fed their albino rats with protein enriched *mumu.* Similarly, (11) who fed rats with extruded African bread fruits based diets observed similar trends. Table 4 presents the result of the protein quality indices of the test diets. The protein efficiency ratio (PER = weight gain of test group/protein consumed by test group) is the official method in

Canada and the United State for assessing protein quality of foods. PER values less than 2.0 are attributed to low quality protein foods while PER values of more than 2.0 are for normal quality protein foods (12,13). The result showed PER values for diets with 0.3% and 0.2% citric acid (G) and (D) have a PER values of 2.34 and 2.32 respectively while that treated with 0.1% citric acid (B) have 2.40 , the untreated sample (AB – control) have a PER value of 2.18. The reference standard Casein (CD) has a value of 2.58. The PER as an index of protein quality shows the relationship between weight gain by the test animals and the corresponding protein as N- intake (11). According to (14) the addition of citric acid at higher intensity may affect the sensory attributes. Similarly, the microstructure of beef muscle changes dramatically upon citric acid acidification but returns to a surprising normal structure upon readjustment back to the normal post mortem pH of raw beef muscle. PER (protein efficiency ratio) has been heavily criticized and NPR (Net protein ratio) has been the recommended method by the AOAC and the Protein Advisory Group of the United Nations (15,16). The NPR (weight gain/loss by 0.0% protein group/weight of protein consumed) credits the protein used for both growth and maintenance (12).

IV. CONCLUSION

The result of the PER demonstrate that the value are above 2.0 as recommended by the protein Advisory Group of the United Nations except the group feed basal diets that recorded a negative value. The mean weight gain (MWG) also showed significant increase with increase in hurdle concentration.

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Table 1: Proximate Composition of Test Diets (%)

Parameters	Diets						
	B	D	G	AB	BD	CD	LSD
Crude protein	26.30^{c}	28.25^{a}	27.55^{b}	21.35^{d}	16.45^{f}	18.55^{e}	0.16
Crude fat	7.15^{b}	7.35^{a}	7.20^{b}	6.30^{c}	3.20^{e}	3.40^{d}	0.10
Ash	5.30^{c}	5.75^{b}	6.15^{a}	4.70^{d}	3.80^{f}	4.05^{e}	0.12
Crude Fibre	1.20^{a}	1.21^{a}	1.19^{a}	1.13^{b}	0.80^{d}	0.93^{c}	0.02
Moisture	2.20^{d}	2.60^{a}	2.65^{a}	2.40^{b}	2.20^{d}	2.30^{c}	0.07
Carbohydrate	59.05^{d}	56.05^{f}	56.50^{e}	65.25^{c}	74.35^{a}	71.75^{b}	0.16

Sample mean values not followed by the same superscripts in the same row are significantly different ($P \leq 0.05$)

Key:

B = Citric Acid (0.1%), Salt (2.0%), Sugar (4.0%), All Spices (2.0%)
D = Citric Acid (0.2%), Salt (2.0%), Sugar (4.0%), All Spices (2.0%)
G = Citric Acid (0.3%), Salt (2.0%), Sugar (4.0%), All Spices (4.0%)
AB = Control (Hurdleless)
BD = Basal diet
CD = Casein Diet
LSD = Least Significant Difference

Table 2: Nitrogen Content of Diet, Carcasses, Liver and Faeces

Parameters	Diets						
	B	D	G	AB	BD	CD	LSD
N-Diet	4.20^{c}	4.50^{a}	4.35^{b}	3.45^{d}	2.60^{f}	2.95^{e}	0.12
N-Carcasses	5.05^{c}	5.30^{a}	4.90^{d}	5.20^{b}	5.30^{a}	5.10^{c}	0.07
N-Liver	4.50^{d}	4.70^{b}	4.80^{a}	4.65^{b}	4.30^{e}	4.60^{c}	0.07
N-Faeces	0.35^{d}	0.36^{c}	0.34^{e}	0.38^{b}	0.38^{b}	0.41^{a}	0.01

Sample mean values not followed by the same superscripts in the same row are significantly different ($P \leq 0.05$)

Key:

B = Citric Acid (0.1%), Salt (2.0%), Sugar (4.0%), All Spices (2.0%)
D = Citric Acid (0.2%), Salt (2.0%), Sugar (4.0%), All Spices (2.0%)
G = Citric Acid (0.3%), Salt (2.0%), Sugar (4.0%), All Spices (4.0%)
AB = Control (Hurdleless)
BD = Basal diet
CD = Casein Diet
LSD = Least Significant Difference
N-Diet = Nitrogen in Diet
N-Carcasses = Nitrogen in Carcasses
N-Liver = Nitrogen in liver
N-Faeces = Nitrogen in Faeces

Table 3: Body Weight Changes, Feed Intake and Feed Conversion Efficiency of Rats Fed *Dambu-nama* Treated with some Hurdles

Parameters	Diets						
	B	D	G	AB	BD	CD	LSD
Initial Wt. (g)	87.60^{c}	84.75^{e}	92.15^{b}	85.95^{d}	93.15^{a}	83.90^{f}	0.37
Final Wt. (g)	150.85^{b}	150.45^{c}	156.45^{a}	132.25^{d}	90.43^{f}	131.75^{e}	0.12
MWG/L	63.28^{c}	65.75^{a}	64.31^{b}	46.45^{e}	-2.68^{f}	47.90^{d}	0.09
DFI	38.18^{c}	38.25^{c}	35.90^{e}	38.80^{b}	37.58^{d}	39.80^{a}	0.09
FCE	2.25^{d}	6.70^{b}	9.96^{a}	1.76^{e}	-289.00^{f}	34.01^{c}	0.11
NI	4.20^{c}	4.48^{a}	4.35^{b}	3.40^{d}	2.55^{f}	2.99^{e}	0.11
PI	26.25^{c}	28.28^{a}	27.61^{b}	21.40^{d}	16.48^{f}	18.55^{e}	0.11

Sample mean values not followed by the same superscripts in the same row are significantly different ($P \leq 0.05$)

Key:

B = Citric Acid (0.1%), Salt (2.0%), Sugar (4.0%), All Spices (2.0%)
D = Citric Acid (0.2%), Salt (2.0%), Sugar (4.0%), All Spices (2.0%)
G = Citric Acid (0.3%), Salt (2.0%), Sugar (4.0%), All Spices (4.0%)
AB = Control (Hurdleless)
BD = Basal diet
CD = Casein Diet
LSD = Least Significant Difference
Wt. = Weight
MWG/L = Mean Weight Gain/Loss
DFI = Daily Feed Intake
FCE = Feed Conversion Efficiency
NI = Nitrogen Intake
PI = Protein Intake

Table 4: Nutritional Quality Indices of Hurdled *Dambu-nama* Diet Fed to Albino Rats

Parameters	Diets						
	B	D	G	AB	BD	CD	LSD
FER	1.67^{c}	1.72^{b}	1.79^{a}	1.21^{d}	-0.07^{e}	1.21^{d}	0.02
FCE	1.28^{b}	1.23^{c}	1.18^{d}	1.76^{a}	0.00^{e}	1.76^{a}	0.02
PER	2.40^{b}	2.32^{d}	2.34^{c}	2.18^{e}	-0.16^{f}	2.58^{a}	0.01
R-PER	4.56^{b}	4.38^{d}	4.44^{c}	4.12^{e}	-0.30^{f}	4.89^{a}	0.02
NPR	2.30^{b}	2.22^{c}	2.23^{c}	2.05^{d}	0.33^{e}	2.44^{a}	0.02
R-NPR	3.80^{c}	3.66^{e}	3.69^{d}	6.38^{a}	0.54^{f}	4.03^{b}	0.02
PU	8.74^{d}	9.13^{b}	9.03^{c}	9.73^{a}	-165.00^{f}	8.17^{e}	0.04
AD (%)	91.50^{a}	92.00^{a}	92.00^{a}	88.50^{b}	85.50^{d}	87.00^{c}	1.22
TPD	1.71^{b}	1.44^{c}	1.92^{a}	1.00^{d}	1.00^{d}	0.33^{e}	0.01

Sample mean values not followed by the same superscripts in the same row are significantly different ($P \leq 0.05$)

Key:

B = Citric Acid (0.1%), Salt (2.0%), Sugar (4.0%), All Spices (2.0%)
D = Citric Acid (0.2%), Salt (2.0%), Sugar (4.0%), All Spices (2.0%)
G = Citric Acid (0.3%), Salt (2.0%), Sugar (4.0%), All Spices (4.0%)
AB = Control (Hurdleless)
BD = Basal diet
CD = Casein Diet
LSD = Least Significant Difference
FER = Feed Efficiency Ratio
FCE = Feed Conversion Efficiency
PER = Protein Efficiency Ratio
R-PER = Relative Protein Efficiency Ratio
NPR = Net protein Ratio
R-NPR = Relative Net protein Ratio
PU = Protein Utilization
AD = Apparent Digestibility
TPD = Total Protein Digestibility