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## Evaluation of Fish Meal Replaced with Leaf Protein Concentrate from *Glyricidia* in Diets for Broiler - Chicks: Effect on Performance, Muscle Growth, Haematology and Serum Metabolites

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**Abstract:** The equi-protein replacement of fishmeal (FM) protein with *Glyricidia* leaf protein concentrate (GLPC) was evaluated in diets for broiler chicks. Prior to the feeding trial, the GLPC was analyzed for the amino acids and any phytin and tannin contents. Thereafter, five diets were formulated. The control (diet 1) contained 5% FM whose protein was gradually replaced at 25, 50, 75 or 100% with GLPC in diets 2 to 5. Thus the inclusion level of GLPC was 1.81, 3.62, 5.43 or 7.24 respectively for diets 2 to 5. A batch of 300 starter-chicks was randomly assigned in triplicate to these dietary treatments (60 chicks/treatment). The final weight and average weight gain of the chicks fed 1.81% GLPC - based diet was similar ( $P = 0.05$ ) to those fed the control diet; both being significantly ( $P = 0.05$ ) higher than those fed 3.62, 5.43 or 7.24% GLPC - based diets. The weight gain, average feed consumption as well as feed efficiency declined as the level of GLPC in the diets increased at the expense of FM protein. Nitrogen retention (NR) of chicks fed 1.81, 3.62 or 5.43% GLPC - based diets was similar to those fed the control diet. At the end of the feeding trial, the chicks were sacrificed for carcass characteristics, relative organ and muscle measurements as well as blood and serum analyses. Relative weights of the heart and belly fat were significantly ( $P \leq 0.05$ ) influenced by dietary treatments. Among the muscles weighed, only the relative weight of *Pectorialis thoracicus* was significantly ( $P \leq 0.05$ ) influenced. The weight of *P. thoracicus* was highest in chicks fed 1.81% GLPC - based diet among the test diets and was similar to those fed the control diet ( $P = 0.05$ ). Relative lengths of *Supra coracoideus* and *P. thoracicus* averaged 17.6 and 20.7  $\text{cmkg}^{-1}$  body weight, respectively while the relative breadth averaged 3.9 and 8.4  $\text{cmkg}^{-1}$  body weight, respectively. These values did not vary widely as indicated by the low coefficient of variations. Among the haematological indices, only the Erythrocyte Sedimentation Rate (ESR) significantly ( $P = 0.05$ ) increased as the level of GLPC increased in the diets while the serum constituents showed no consistent trends related to the dietary treatments. It was concluded that FM protein in the diet for broiler - chicks can be replaced by 25% with GLPC protein without adverse effect on performance, carcass characteristics, muscle development or haematological variables in broiler - chicks.

**Key words:** Fishmeal protein, *Glyricidia* leaf protein concentrate, *Pectorialis thoracicus*

### Introduction

The realization that feeding alone currently accounts for over 75% of intensive non-ruminant (poultry and swine) production in the third world countries, including Nigeria, has stimulated research interest aiming at exploiting different locally available alternative feeding resources. A notable feed resource quite often used in feed formulation, which requires urgent research attention for an alternative, is fishmeal due to its high cost. The high cost of fishmeal derives largely from the competition between man and livestock for fish and fish products. Consequently, nutritionists have in recent times attempt to replace it with different locally available protein resources. For instance, maggot meal (Atteh and Ologbenla, 1993; Akpodiete *et al.*, 1997; Akpodiete and Inoni, 2000) and cassava leaf protein concentrate (Fasuyi, 2000) have been used either to replace fishmeal wholly or partly in broiler diets with remarkable

success especially in relation to some growth indices. An ingredient with high nutrient density deserving evaluation in this regard is the leaf protein concentrate from *Glyricidia sepium*. The leaves of *G. sepium* abound in Nigeria and are rich in protein (30-32%), minerals (Aletor and Omodara, 1994) and vitamin A. The leaf protein concentrate contains high crude protein (48.9%) and crude fat (14.5%). Apart from the relatively high mineral and protein contents, it has an amino acid profile which in most cases compares with those of fishmeal. *Glyricidia* leaf protein concentrate have been reported to enhance growth in rats when it replaced soybean flour in infant weaning food (Agbede, 2000). Conceivably, a successful use of *Glyricidia* leaf protein as alternative protein source to fishmeal in broiler diet should lower substantially high cost of fishmeal and eventually the acute shortage of animal protein supply in developing countries, including Nigeria. In spite of the

nutritional potential of GLPC, there remains a dearth of information on its use in broiler - starter diets. Consequently, this study was designated to investigate the effect of protein-for-protein replacement of fish meal with GLPC in the diets for broiler - starter chicks on (1) performance, nutrient utilization, carcass characteristics and relative organ measurement; (2) the muscle growth with respect to *Pectoralis thoracicus*, *Supra coracoideus* and *Gastrocnemius* and (3) some haematological and serum components.

## Materials and Methods

**Leaf Protein Production:** The Glyricidia leaf protein concentrate (GLPC) was produced using a village-scale fractionation method (Fellows, 1987). Hot whey was siphoned using rubber hose while the protein coagulum was separated from other fractions by filtering through a muslin bag; followed by pressing with screw-press (Aletor, 1993). The GLPC was rinsed, pulverized, sun-dried and milled. Thereafter, the GLPC was used to formulate diets along with other ingredients purchased from Egun Olu Farm, Km 9 Ikirun Road, Osogbo, Nigeria.

**Experimental Diets:** Five *iso*-nitrogenous and *iso*-caloric diets were formulated with the basal as well as the proximate composition as shown in Table 1 and 2, respectively. Diet 1 was the control with 5% fish meal (FM). The fishmeal protein was substituted at 25, 50, 75 or 100% with GLPC in diets 2, 3, 4 and 5, respectively. The five diets were analyzed for their amino acid contents and presented as part of Table 2.

**Management of chicks and Experimental layout:** Three hundred day-old broiler chicks (150 males + 150 females) used for the experiment were electrically brooded at the Teaching and Research Farm of the Federal University of Technology, Akure, Nigeria for the first week. They were brooded on the floor for the first 4 days and sexed as described by Laseinde and Oluyemi (1997), and later transferred to a metabolism cage for 3 days to allow the chicks acclimate to the experimental conditions. During this period they were offered a 23g kg<sup>-1</sup> crude protein commercial broiler-starter mash (Guinea Feed) and water *ad libitum*. Completely randomized design (CRD) was adopted for the trial with a total of 15 experimental units. At the end of the acclimation period, the chicks were weighed and 30 males and 30 females (total = 60) were assigned to each of the five triplicate dietary treatments (10 males + 10 females/replicate), such that the group meal weights per diet were identical (106.3 ± 1.4g). The chicks were fed their respective experimental diets *ad libitum* for 21 days during which the daily feed consumption and group weight changes were measured. Faeces voided during the last five days were collected, weighed, dried at 55-60°C in an air-circulating oven for 72 hrs, and preserved

while the corresponding feed consumed was recorded.

**Blood Collection for Analysis:** At the end of the feeding trial, the chicks were starved overnight so as to empty the crop and the chicks weighed and sacrificed first by stunning followed by severing the jugular vein. The blood was then allowed to flow freely into labeled bijoux bottles one of which contained a speck of EDTA while the others were without EDTA. The blood in the EDTA - containing bijoux bottles was processed for haematology while those in bottles without EDTA were processed for serum. The serum was kept deep frozen prior to analysis.

**Carcass Characteristics and Organs Measurements:** After slaughtering and bleeding the chicks, the carcasses were scalded at 65 °C in water both for 30 seconds before defeathering. The dressed chicks were later eviscerated. Ten eviscerated carcasses per replicate were used for the measurement of the carcass characteristics viz: dressed weight%, eviscerated weight%, thigh, drumstick, shank, chest, back, neck, wing, belly fat and head and organ measurements. The organs were dissected out and blotted with filter paper. The organs measured were the liver, kidneys, lungs, pancreas, heart, spleen, bursa of fibricus and gizzard. All the carcass characteristics and organs measured were expressed in gkg<sup>-1</sup> body weight except the dressed and eviscerated weights, which were expressed in percentages of the live weight.

**Muscle measurement:** The remaining ten eviscerated carcasses per replicate were weighed and cooled under the fan for 3 min. before dissection. The dressed and eviscerated weights were taken before dissecting out the chest as well as thigh muscles. The inner chest muscle (*Supra coracoideus*), outer chest muscle (*Pectoralis thoracicus*) and thigh (*Gastrocnemius*) were carefully dissected out from their points of origin and insertion. Measurements of the fresh weights, length and breadth of these muscles were taken. The muscle weights were expressed in gkg<sup>-1</sup> body weight, while the length and breadth were expressed in cmkg<sup>-1</sup> body weight.

**Haematological and Serum analyses:** The packed cell volume (PCV), was estimated by spinning about 75 µl of each blood sample in heparinized capillary tubes in a haematocrit micro centrifuge for 5 minutes while the total Red Blood Cell count (RBC) was determined using normal saline as the diluting fluid. The Haemoglobin concentration (HBC) was estimated using cyanomethaemoglobin method while the Mean Corpuscular Haemoglobin Concentration (MCHC), Mean Corpuscular Haemoglobin (MCH) and the Mean Corpuscular Volume (MCV) were calculated as

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Table 1: Composition of Experimental Diet (g/100g)

Ingredients	Diets				
	1	2	3	4	5
	% of Fish meal replaced with GLPC protein				
	0	25	50	75	100
Maize (11%)	56.10	55.54	54.98	54.42	53.86
Groundnut cake (42%)	28.00	28.00	28.00	28.00	28.00
Fish Meal FM (68%)	5.00	3.75	2.50	1.25	-
GLPC (47%)	-	1.81	3.62	5.43	7.24
Brewer Dried Grain (29%)	5.00	5.00	5.00	5.00	5.00
Oil	2.00	2.00	2.00	2.00	2.00
Bone Meal	2.50	2.50	2.50	2.50	2.50
Oyster Shell	0.50	0.50	0.50	0.50	0.50
DL Methionine	0.15	0.15	0.15	0.15	0.15
Premix *	0.25	0.25	0.25	0.25	0.25
Salt	0.50	0.50	0.50	0.50	0.50
TOTAL	100.0	100.0	100.0	100.0	100.0
Crude Protein (g/100g)	23.0	23.0	23.0	23.0	23.0
ME (MJ/kg)	13.0	13.0	13.0	13.0	13.0

\*contained vitamins A (4,000,000iu); D (800,000iu); E (14000iu); K (760mg); B<sub>12</sub> (7.6mg); Riboflavin (2800mg); Pyridoxine (1520mg); Thiamine (880mg); D Pantothenic acid (4400mg); nicotinic acid (18,000mg); Folic acid (560mg); Biotin (45.2mg); and Trace elements as Cu (3200mg); Mn (25600mg); Zn (16,000mg); Fe (12800mg); Se (64mg); I<sub>2</sub> (320mg) and other items as Co (160mg); Choline (190,000mg); Methionine (20,000mg); BHT (2,000mg) and Spiramycin (2,000mg) per 1.0kg. GLPC; Glyricidia leaf protein concentrate

Table 2: Proximate Composition (g/100g) and Amino Acid Content (%) of Experimental Diets

Composition	Diets				
	1	2	3	4	5
	% of Fish meal replaced with GLPC protein				
	0	25	50	75	100
Dry matter	90.4	90.1	90.8	90.3	90.4
Crude protein	22.6	22.4	22.1	22.1	22.0
Ether extracts	9.4	9.4	9.7	10.3	10.8
Crude fibre	7.4	8.2	8.6	8.9	10.3
Ash	4.0	4.3	4.6	6.1	6.6
Nitrogen free extract	56.6	55.7	55.0	52.6	50.3
Amino acids					
Arginine	2.11	2.11	2.11	2.11	2.11
Histidine	0.57	0.57	0.57	0.57	0.57
Isoleucine	1.08	1.08	1.08	1.08	1.08
Leucine	1.95	1.96	1.98	1.99	2.00
Lysine	1.31	1.26	1.23	1.19	1.16
Methionine	0.45	0.45	0.44	0.43	0.42
Cystine	0.16	0.15	0.15	0.14	0.14
Methionine + cystine	0.61	0.60	0.59	0.57	0.56
Phenylalanine	1.18	1.20	1.22	1.24	1.26
Threonine	0.86	0.87	0.88	0.88	0.90
Tyrosine	0.94	0.94	0.95	0.98	0.99
Valine	1.26	1.26	1.28	1.29	1.31
Glycine	0.34	0.33	0.32	0.30	0.29
Tryptophan	0.72	0.72	0.72	0.72	0.72

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described by Lamb (1981). The Erythrocyte Sedimentation Rate (ESR) of the blood as well as the total protein, albumin and globulin of serum were determined as described (Lamb, 1981).

**Chemical and Statistical analysis:** The proximate composition of the ingredients, diets and faecal samples were determined by the method of AOAC (1990). Nitrogen retained was calculated as the algebraic difference between feed nitrogen and faecal nitrogen (on dry matter basis) for the period. The amino acid contents of the GLPC and the diets were analyzed by hydrolyzing the LPCs (50-75mg) for 24 hours in a heating block previously heated at  $110 \pm 1$  °C. The hydrolysate was cooled and quantitatively transferred to a -50ml flask and diluted to volume with water. After filtration, a 10ml aliquot of the filtrate was heated in a rotary evaporator (40 °C) to remove excess acid before analysis using high-performance liquid chromatography (HPLC) with a Varian HPLC system (Palo Alto, CA) and a Shimadzu RF-535 Fluorescence detector (Tokyo, Japan) set at an excitation wavelength of 325nm and an emission wavelength of 465nm. Separation was achieved in adsorbosphere OPA-HR (150 x 4.6 mm) column (Alltech, Carnforth, UK). The mobile phase was 1,4-dioxan and 2-propanol (HPLC grade). Methionine was determined as methionine sulfone and cysteine as cysteic acid after performic acid oxidation while tryptophan was determined as described by Miller (1967). To correct for slight fluctuations in amino acid peaks, DL-Amino-n-butyric acid was used as internal standard. The determination of some anti-nutritional factors typified by phytin as described (Makkar and Goodchild, 1996) and tannin by the methods (Young and Greaves, 1940; Makower, 1970; Wheeler and Ferrel, 1971) were done. Data collected on the performance indices; haematology and total serum protein were subjected to analysis of variance (Steel and Torrie, 1980). Where significant differences were found, the means were compared using the Duncan's Multiple Range Test (DMRT) (Duncan, 1955). Data on muscle growth were subjected to coefficient of variation analysis (Snedecor and Cochran, 1973).

## Results

**Amino acid profile, Phytin and Tannin contents of GLPC:** The amino acid profile of GLPC showed that it contained lysine: 6.60, histidine: 2.51, arginine: 6.30, aspartic acid: 9.81, threonine: 5.08, serine: 5.01, glutamic acid: 11.66, proline: 4.98, glycine: 5.78, alanine: 6.25, cystine: 1.64, methionine: 2.05, valine: 6.18, isoleucine: 5.14, leucine: 9.33, tyrosine: 4.79, phenylalanine: 6.22 and tryptophan: 1.92 (g/16N). Also GLPC contained 10.0mg/100mg Phytin and 0.3g/100g DM poly phenols as tannic acid equivalent.

**General observation:** The chicks used were all alert during the experimental period. The chicks fed the GLPC-based diets had yellow pigmentation on the skin, beaks and shanks. Pigmentation was also obvious in their faeces and it increased with increasing dietary level of GLPC.

**Performance of chicks and Nitrogen utilization :** Table 3 shows that average weight gain, feed consumption and feed efficiency were significantly ( $P \leq 0.05$ ) influenced by the dietary treatments. Weight gain of chicks fed the control diet was similar to those fed on 1.81% GLPC - based diet, but both were significantly ( $P \leq 0.05$ ) higher than those fed diets containing 3.62-7.24% GLPC. The average weight gain decreased progressively with increased FM protein substitution with GLPC. However, the least average weight gain ( $12.1 \pm 0.7$ g/chick/day) was observed for chicks fed 7.24% GLPC - based diet (diet 5). Feed consumption (FC) by chicks fed the control diet and those diets containing 1.81, 3.62 or 5.43% GLPC were similar while those fed on 3.62, 5.43 or 7.24% GLPC - based diets were not significantly ( $P = 0.05$ ) different from each other. Nevertheless, FC decreased with increased substitution of FM protein with GLPC protein in the diets. Like the FC, the feed efficiency decreased as the FM protein substitution with GLPC protein increased in the diets.

Nitrogen retention (NR) of the chicks fed the control diet and those fed on diets containing 1.81, 3.62 or 5.43% GLPC were significantly ( $P \leq 0.05$ ) higher than those fed 7.24% GLPC - based diet (Table 3). However, the nitrogen retained by the chicks correlated negatively ( $r = -0.99$ ;  $P \leq 0.05$ ) with the level of fishmeal replacement with GLPC.

**Carcass characteristics and Relative Organ Measurement:** All the carcass characteristics measured (Table 4), except the belly fat and neck were not significantly ( $P \geq 0.05$ ) influenced by the dietary treatments. The belly fat in the chicks fed on diets with 5.43 or 7.24% GLPC were similar, but those fed 7.24% GLPC - based diet were significantly ( $P \leq 0.05$ ) higher than those fed 0, 1.81 or 3.62% GLPC - based diets. The relative weights of the neck were least in chicks fed the control diet ( $36.3 \pm 6.3$ gkg<sup>-1</sup> body weight) and highest in chicks fed 7.24% GLPC - based diet ( $60.7 \pm 12.8$ gkg<sup>-1</sup> body weight).

Table 5 shows that, with the exception of the relative weight of the heart, all other organs measured were not significantly ( $P \geq 0.05$ ) influenced by the dietary treatments. Relative weight of the heart was higher in the chicks fed GLPC - based diets than those fed the control. However, the highest relative weight of the heart was observed for chicks fed 7.24% GLPC - based diet ( $8.2 \pm 0.5$ gkg<sup>-1</sup> body weight) and least in chicks fed the control diet ( $5.0 \pm 0.9$ gkg<sup>-1</sup> body weight). Generally,

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Table 3: Performance and nitrogen utilization of broiler-starter fed experimental diets

Diet	Initial weight (g)	Final weight (g)	Average weight gain (g/chick/day)	Average feed consumption (g/chick/day)	Feed efficiency	N-retention (g/chick/day)
1	106.9±4.0	586.2±14.6 <sup>a</sup>	22.0±0.7 <sup>a</sup>	43.7±1.7 <sup>a</sup>	2.0±0.1 <sup>c</sup>	0.8±0.3 <sup>a</sup>
2	108.3±4.2	545.3±32.7 <sup>a</sup>	20.8±1.6 <sup>a</sup>	46.8±5.3 <sup>a</sup>	2.3±0.2 <sup>b</sup>	0.7±0.3 <sup>a</sup>
3	104.5±0.2	451.9±33.4 <sup>b</sup>	16.5±1.6 <sup>b</sup>	38.9±4.7 <sup>ab</sup>	2.4±0.1 <sup>b</sup>	0.8±0.4 <sup>a</sup>
4	106.2±4.2	408.4±5.8 <sup>bc</sup>	14.4±1.2 <sup>bc</sup>	38.1±1.4 <sup>ab</sup>	2.6±0.1 <sup>ab</sup>	0.8±0.2 <sup>a</sup>
5	105.8±3.4	359.5±15.2 <sup>c</sup>	12.1±0.7 <sup>c</sup>	34.1±1.9 <sup>b</sup>	2.8±0.2 <sup>a</sup>	0.6±0.3 <sup>b</sup>

Means with differing superscripts in the same column are significantly different (P=0.05)

Table 4: Carcass traits of broiler-chicks fed GLPC - Based Diets from age 7-28 days

Traits	Diets				
	1	2	3	4	5
Dressed weight %	87.1±5.3	90.1±1.2	90.1±0.4	89.6±1.6	91.1±1.3
Eviscerated weight %	79.1±4.6	83.3±0.7	82.1±0.3	83.8±18.0	82.6±5.1
Thigh (gkg <sup>-1</sup> body wt)	49.3±3.3	49.1±5.4	44.8±7.9	44.9±16.0	41.2±1.7
Drumstick (gkg <sup>-1</sup> body wt)	99.8±10.1	102.7±10.2	94.7±3.3	96.8±20.3	97.6±9.9
Shank (gkg <sup>-1</sup> body wt)	31.3±5.1	32.5±3.1	35.4±6.3	30.5±9.0	29.5±2.2
Wing (gkg <sup>-1</sup> body wt)	36.8±2.1	42.0±3.8	40.5±0.5	38.5±9.5	40.7±7.6
Chest (gkg <sup>-1</sup> body wt)	142.4±16.1	137.3±18.7	123.5±12.8	127.2±38.0	109.6±9.1
Back (gkg <sup>-1</sup> body wt)	67.8±11.6	72.4±4.6	66.8±4.2	71.3±22.4	65.1±0.6
Head (gkg <sup>-1</sup> body wt)	46.2±2.2	46.6±1.6	46.6±9.7	48.7±8.6	45.4±4.8
Belly fat (gkg <sup>-1</sup> body wt)	2.52±1.2 <sup>b</sup>	7.0±1.7 <sup>b</sup>	2.0±3.5 <sup>b</sup>	8.6±5.1 <sup>ab</sup>	16.3±4.5 <sup>a</sup>
Neck (gkg <sup>-1</sup> body wt)	36.3±6.3 <sup>b</sup>	53.4±7.8 <sup>ab</sup>	45.8±3.6 <sup>ab</sup>	45.6±10.9 <sup>ab</sup>	60.7±12.8 <sup>a</sup>

\*Means are for 60 chicks/diet (mean ± SD), \* Means with differing superscripts in the same column are significantly different (P= 0.05)

chicks fed GLPC - based diets had higher relative weight of the heart than the control diet.

**Relative Weight, Length and Breath of Muscles:** Table 6 shows the responses of muscle weights in chicks fed varying levels of fishmeal substitution with GLPC. There were no wide variations in the weights of *S. coracoideus* and *Gastrocnemius* of chicks fed the control or other diets as indicated by their CV values of 15.4 and 12.4%, respectively. The relative weight of *P. thoracicus* of chicks fed the control diet (39.3±3.0gkg<sup>-1</sup> body weight) were similar to those fed 1.81% GLPC - based diet (31.7±6.4gkg<sup>-1</sup> body weight) but significantly (P=0.05) different from those chicks fed on 3.62 to 7.24% GLPC - based diets. Table 6 also showed that the relative length of *S. coracoideus* and *P. thoracicus* did not vary widely as shown by their CV values of 7.4 and 9.7%, respectively. Similarly, the relative breadth of *S. coracoideus* and *P. thoracicus* did not vary widely as shown by the low CV values of 10.3 and 8.3%, respectively.

**Haematological variables:** Table 7 presents the haematological variables of broiler-chicks fed the various diets. The ESR was significantly (P=0.05) influenced by diet treatments. The PCV was highest in chicks fed 5.43% GLPC - based diet (31.7±1.2%) and

lowest in those fed the control diet (29.3±1.5%) while RBC was highest in chicks fed on 1.81% GLPC - based diet (2.7±0.6 x 10<sup>6</sup>/mm<sup>3</sup>) and lowest in those fed the control diet (2.0±0.0 x 10<sup>6</sup>/mm<sup>3</sup>). HBC was lowest in the chicks fed the control diet (1.7±0.2g/100ml) and highest in those fed 1.81% GLPC - based diet (2.7±1.2g/100ml). The MCHC% varied between 5.3±0.5% in chicks fed the control diet to 8.9±6.1% in chicks fed on 3.62% GLPC - based diet. MCH (pg) of chicks fed 3.62% GLPC - based diet was the highest (11.5±7.1) while the MCV (um<sup>3</sup>) was highest in chicks fed on 5.43% GPLC-based diet (153.9±15.1). The ESR (mm/hr) of chicks significantly (P=0.05) decreased with increased level of FM protein substitution with GLPC protein but there was no significant (P=0.05) difference between the ESR of chicks fed the control and those fed 1.81, 3.62 or 5.43% GLPC-based diets.

**Total Serum Protein (TSP):** Table 8 shows the response of serum metabolites to the dietary treatments. TSP, albumin and globulin of the chicks fed the control diet were not significantly (P=0.05) influenced by the diet treatments. Albumin/Globulin ratios of chicks fed 3.62 or 5.43% GLPC - based diets were similar but those fed 3.62% GLPC - based diet were significantly

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Table 5: Relative organ weight (g/kg<sup>-1</sup> body wt) of broiler chicks fed GLPC - based diets from age 7 - 28 days

Organs	Diets				
	1	2	3	4	5
Heart	5.0±0.9 <sup>b</sup>	7.1±1.2 <sup>ab</sup>	6.3±1.2 <sup>ab</sup>	7.3±0.9 <sup>ab</sup>	8.2±0.5 <sup>a</sup>
Lung	5.6±0.2	7.1±1.6	7.2±1.2	6.5±1.5	6.9±1.5
Liver	20.6±3.0	20.3±4.6	21.1±2.2	21.9±1.4	23.9±2.1
Spleen	1.9±0.1	1.92±0.1	2.2±0.5	2.2±0.3	2.1±0.1
Pancreas	1.9±0.1	3.2±1.1	4.4±0.9	2.9±1.1	4.1±1.9
Gizzard	35.7±6.0	42.7±3.7	41.9±6.1	46.0±5.9	45.3±11.7
Kidney	8.1±1.4	8.3±0.5	9.1±2.0	10.4±3.7	8.2±0.5
Bursa	3.7±0.1	3.2±1.1	2.2±0.5	2.2±0.3	2.8±1.2

\*Means are for 60 chicks/diet (Mean ± SD). \* Means with differing superscripts in the same column are significantly different (P= 0.05)

Table 6: Effect of varying levels of dietary GLPC on relative weight (gkg<sup>-1</sup> body weight), length and breadth (cmkg<sup>-1</sup> body weight) of Some muscles in broiler-starter

Diets	Weight			Length		Breadth	
	<i>Supra coracoideus</i>	Pectoralis thoracicus	Gastrocnemius	<i>Supra coracoideus</i>	Pectoralis thoracicus	<i>Supra coracoideus</i>	Pectoralis thoracicus
1	11.2±1.5	39.3±3.0 <sup>s</sup>	35.7±4.6	16.2±1.0	19.4±2.0	3.7±0.1	9.0±1.0
2	8.2±2.3	31.7±6.4 <sup>ab</sup>	34.5±1.2	17.9±3.1	20.6±3.4	3.9±0.3	8.6±0.4
3	9.1±0.9	27.4±1.0 <sup>b</sup>	31.9±5.8	19.6±6.6	21.5±1.6	4.4±0.9	7.2±1.3
4	9.5±1.2	29.2±1.0 <sup>b</sup>	229.1±5.5	17.6±1.4	23.7±1.3	4.0±0.2	8.5±1.5
5	7.6±1.6	25.4±2.0 <sup>b</sup>	26.1±1.7	16.7±1.7	18.5±2.5	3.4±0.4	8.9±0.7
MEAN	9.1	30.6	31.5	17.6	20.7	3.9	8.4
STD	1.4	5.4	3.9	1.31	2.0	0.4	0.7
CV%	15.4	17.4	12.4	7.4	9.7	10.3	8.3

CV; Coefficient of Variation, Means with differing superscript along the same column are significantly (P=0.05) different

(P=0.05) higher than those fed the control, 1.81 or 5.43% GLPC - based diets.

### Discussion

The yellow pigmentation observed on the skin, beaks, ear lobes and shanks of the birds fed the test diets could be attributed to the high level of beta - carotene (precursor of vitamin A) that have been reported to be present in LPCs (Eggum, 1970; Fasuyi, 2000). In the present study, an increase in GLPC in the diet beyond 1.81% led to an almost linear depression in growth performance, feed consumption and feed efficiency. The identical performance of chicks fed the control and 1.81% GLPC - based diets might be due to the similarity of their dietary amino acids profiles (Table 2), except for cystine, lysine and glycine which decreased by 6.25, 3.82 and 2.94%, respectively. However, there was a decrease in the concentration of some of the amino acids in the diets as the levels of GLPC substitution increased. This was more obvious in methionine, lysine, cystine and glycine, which decreased with 6.67, 11.45, 12.5 and 14.71%, respectively. Thus, the observed decline in the performance of the chicks on the test diets may be attributed in part to the adverse effect of amino imbalances. This corroborated an earlier report of

Agbede and Aletor (1997) that amino acids imbalance adversely impair appetite and feed intake with attendant reduction in performance in chicks. The observed growth depression, reduced feed intake and feed efficiency of chicks on the test diets may also in part be attributed to the level of dietary fibre (Table 2) which increased with increasing fish meal protein substitution with GLPC as high dietary fibre may cause poor performance by impaction of the intestinal tract. Similarly, the higher the level of GLPC in the diet, the higher will be the level of the residual anti-nutrient, particularly phytin (Agbede, 2000) with the attendant adverse cumulative effect on the performance. In fact, the levels of phytin in the test diets varied from 0.81% in diet 2 to 0.72% in diet 5. Therefore, the decrease in the performance of chicks fed especially on 2.62 - 7.24% GLPC - based diets may be attributed to a variety of factors including, amino acids imbalances, increased dietary fibre and possible adverse effect of residual anti-nutrients (phytin) in GLPC.

The similar nitrogen retained by birds fed the control diet and those fed 1.81 - 5.43% GLPC - based diets could be attributed to similar dietary protein contents (*iso*-nitrogenous) in the diets. The NR of chicks fed on 7.24% GLPC - based diet was the lowest and this might be caused by high dietary crude fibre with low contents of

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Table 7: Haematological indices of broiler - starter fed varying dietary levels of GLPC

Diets	PCV (%)	RBC ( $\times 10^6/\text{mm}^3$ )	HBC (g/100ml)	MCHC (%)	MCH (pg)	MCV ( $\mu\text{m}^3$ )	ESR (mm/hr)
1	29.3 $\pm$ 1.5	2.0 $\pm$ 0.0	1.7 $\pm$ 0.2	5.3 $\pm$ 0.5	7.7 $\pm$ 1.2	145.5 $\pm$ 9.2	5.0 $\pm$ 0.0 <sup>b</sup>
2	30.7 $\pm$ 2.3	2.7 $\pm$ 0.6	2.7 $\pm$ 1.2	8.6 $\pm$ 3.4	9.6 $\pm$ 2.2	116.0 $\pm$ 17.9	5.0 $\pm$ 0.0 <sup>b</sup>
3	30.3 $\pm$ 3.2	2.3 $\pm$ 0.2	2.6 $\pm$ 1.6	8.9 $\pm$ 6.1	11.5 $\pm$ 7.1	135.6 $\pm$ 25.7	5.7 $\pm$ 0.4 <sup>ab</sup>
4	31.7 $\pm$ 1.2	2.1 $\pm$ 0.2	2.0 $\pm$ 0.2	6.3 $\pm$ 0.9	9.7 $\pm$ 1.2	153.9 $\pm$ 15.1	5.7 $\pm$ 0.6 <sup>ab</sup>
5	31.0 $\pm$ 2.7	2.3 $\pm$ 0.4	2.1 $\pm$ 0.5	6.6 $\pm$ 1.1	8.9 $\pm$ 0.7	135.4 $\pm$ 10.6	6.0 $\pm$ 0.0 <sup>a</sup>

Means with differing superscript in the same column are significantly different (P=0.05). PCV = Packed Cell Volume; RBC = Red Blood Cell; WBC = White Blood Cell; HBC = Haemoglobin Concentration; MCHC = Mean Cell Haemoglobin Concentration; MCH = Mean Cell Haemoglobin; MCV = Mean Cell Volume; ESR = Erythrocyte Sedimentation Rate.

Table 8: Serum metabolites of broiler-starter fed varying dietary levels of GLPC

Diets	Total Serum Protein (g/100ml)	Albumin (g/100ml)	Globulin (g/100ml)	Albumin/ Globulin Ratio
1	3.4 $\pm$ 0.9	1.1 $\pm$ 0.1	2.3 $\pm$ 0.9	0.5 $\pm$ 0.2 <sup>b</sup>
2	2.7 $\pm$ 0.3	0.9 $\pm$ 0.1	1.9 $\pm$ 0.3	0.5 $\pm$ 0.1 <sup>b</sup>
3	2.6 $\pm$ 0.4	1.6 $\pm$ 0.5	1.0 $\pm$ 0.5	1.9 $\pm$ 1.0 <sup>a</sup>
4	3.0 $\pm$ 0.4	1.3 $\pm$ 0.2	1.7 $\pm$ 0.2	0.8 $\pm$ 0.1 <sup>ab</sup>
5	3.3 $\pm$ 0.5	0.9 $\pm$ 0.3	2.4 $\pm$ 0.6	0.4 $\pm$ 0.2 <sup>b</sup>

Mean with different superscript in the same column are significantly different (P=0.05)

some limiting amino acids (Table 2) which have been classified among those factors that hinder nitrogen utilization by chicks (Nwokolo *et al.*, 1985; Agbede and Aletor, 1997).

It is evident that neither the 5.0% FM - based diet nor the GLPC - based diets had any significant influence (except the weight of the belly fat and neck) on the carcass 'fast food' cuts, suggesting that identical carcass characteristics are attainable by feeding either FM or GLPC - based diets. The deposition of belly fat in this present study appears to be enhanced by the increased inclusion of GLPC (diets 4 and 5). It is suggested that this higher development of adipose tissue by chicks fed the GLPC - based diets may be attributed to higher dietary fat which increased as the level of GLPC increased in the diets (Table 2). The significantly higher relative weights of the neck in chicks fed all the test diets over the control diet might be attributed to the dietary effect since all the birds used for the study were decapitated neatly at the base of the skull and disengaged from the carcass at the point of insertion. Also, the relative weights of organ (except the heart) were identical in all the treatment groups. The relative weight of the heart tended to increase with increased level of GLPC in the diets. This may be attributed to high fat deposition around the pericardium of the heart as a result of GLPC and that may have implication for the diastolic and systolic systems of the heart of the chicks fed on the test diets. Despite this, the absolute weight gain of the chicks fed the test diets were lower than

those fed the control diets thus further confirming the superiority of FM as good sources of protein and other growth promoting factors than GLPC. Rosochacki *et al.* (1986); Aletor *et al.* (1989) and Schreurs (2000) demonstrated that nutrition influences muscular growth while malnutrition causes an increase in protein degradation in chicken. From this study, the relative weights, lengths and breadths of *S. coracoideus*, *P. thoracicus* and *Gastrocnemius* of chicks fed the control and the test diets were similar as evident in their relatively low CV values. This thus suggests that the control diet as well as the test diets promoted similar chest and hind muscle growth except the relative weight of *P. thoracicus* which was higher in the chicks fed the control diet than those fed the test diets.

Blood represents a means of assessing clinical and nutritional health status of animals in feeding trial and the haematological parameters most commonly used in nutritional studies include PCV, RBC, HBC, MCHC, MCV and clotting time (Aletor and Egberongbe, 1992; Olorede and Longe, 2000; Adeyemi *et al.*, 2000). The results of haematological variables in this study suggest that the test diets did not precipitate any severe effects on the health status of the experimental chicks. However, the values obtained for PCV and RBC were similar to those reported for chicks fed winged bean and full fat jatropha seeds (Igene, 1999; Adeyemi *et al.*, 2000) but the HBC, MCHC, MCH and MCV were lower than the values reported by Aletor and Egberongbe (1992). With regard to the blood physical properties the ESR was significantly influenced by the dietary treatments. It is believed that the frictional resistance of the surrounding plasma, which hold the cells in suspension, and the gravitational pull on the erythrocyte, mostly determines the ESR. Though the ESR increased with the levels of GLPC inclusion in the diets the values obtained in this study may not be a source of worry since they are very low. This also implies that the test diets did not give rise to acute general infection as high values of sedimentation rates could precipitate acute general infections and malignant tumors (Frandsen, 1986). Total serum protein, albumin and globulin syntheses were generally similar for all the dietary treatments in the



present study. Although TSP may be used as an indirect measurement of dietary protein quality (Tewe, 1985), the value observed for the serum generally appears comparable with that reported by Igene (1999) and Adeyemi *et al.* (2000) for chicks of the same age.

**Conclusion:** The study showed that GLPC could replace up to 25% FM protein in broiler-chick diets without adverse effects on the weight gain, carcass characteristics, chest and hind muscles and the blood variables. Consequently, the use of GLPC in the diets for broilers in the third world countries where fish meal usually attracted high cost, hold tremendous promise for making started chicks available for farmers at affordable cost. This is expected to boost broiler production for the consumption of the low-paid populace.

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