TROPICAL VEGETABLE (AMARANTHUS CRUENTUS) LEAF MEAL AS ALTERNATIVE PROTEIN SUPPLEMENT IN BROILER STARTER DIETS: BIONUTRITIONAL EVALUATION

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ABSTRACT

Amaranthus cruentus is a tropical leaf vegetable grown in most tropical regions of the world for its vegetable protein. The fresh matured leaves of the plant were harvested and sun dried until a moisture content of between 12-13% was obtained. The sun dried leaves (Amaranthus cruentus leaf meal, ACLM) were milled and analysed for their proximate composition. Crude protein was $23.0\%\pm0.55$; crude fat, $5.4\%\pm0.01$; crude fibre, $8.8\%\pm0.02$; ash, $19.3\%\pm0.01$ and gross energy, 3.3 ± 0.01 kcal/g all on dry matter basis. Methionine and to a lesser extent, lysine, arginine, leucine and aspartate were high. The ACLM was incorporated into five formulated broiler starter diets at varying inclusion levels. The control diet 1 had no ACLM inclusion. All the six diets including control diet 1 were formulated isocaloric and isonitrogenous and fed to the experimental chicks (n = 540). Birds kept on diet 2 (5% ACLM inclusion level) had the best average weight gain (WG) of 372.9 ± 29.94 g/chick. The feed efficiency (FE) value and the protein efficiency ratio (PER) for birds on diet 2 were similar (P > 0.05) to values obtained for diet 2 were highest at 1.48 ± 0.24 gN/chick/day and $63.12\%\pm10.28$, respectively. Except for dressed weight and the back of chicken all the organs weights taken were similar (P > 0.05). Haematological examinations were similar (P > 0.05). Results generally indicated that ACLM could be a useful dietary protein source for broiler starter chicks at 5% inclusion level.

Key words: Amaranthus cruentus leaf meal, antinutrients, nitrogen utilization, broiler chicken



INTRODUCTION

Amaranthus plant is a popularly grown leaf vegetable in tropical regions of the world including Africa, India, Bangladesh, Sri Lanka and the Caribbean. It is also grown as leaf vegetable through South-East Asia and Latin America. Grain amaranth is produced commercially in the United States in wet and dry areas. The economic and nutritional advantage of the amaranth as a leaf vegetable is accentuated by its agronomic superiority over many plant protein sources. For instance, harvesting is done 20-30days after transplanting and then every 2-3weeks for a period of one to two months [11]. Another potential advantage of the amaranthus is the chemical composition which is highly in favour of the plant leaves as a veritable source of plant protein [2, 11] and its rich source of vitamins and minerals. The world shortage of animal protein particularly in developing countries in Africa has necessitated investigations of several novel nutritional materials for possible incorporation into animal feeds (particularly poultry) as replacements for the expensive conventional sources such as fish meal, groundnut cake and soyabeans. The acute shortage of protein has been attributed to the phenomenal rise in the prices of animal feeds which account for about 75-85% of the recurrent production inputs in intensive monogastric animal production [8]. A growing interest in the use of unconventional sources of protein and energy in poultry feed has gained prominence [7, 18, 23]. This study therefore investigated the chemical, amino acids and antinutritional constituents in the processed Amaranthus cruentus leaf meal (ACLM) as a prelude to incorporation into broiler starter diets. The performance characteristics, nitrogen utilization, carcass characteristics, relative organs weights, muscle development, haematological indices, serum and liver metabolites of experimental birds were thereafter investigated as a measure of acceptability of ACLM in poultry feed.

MATERIALS AND METHODS

Collection and preparation of Amaranthus cruentus leaf meal (ACLM)

Amaranthus cruentus plants were harvested fresh from maturing stems at about 20-30days after transplanting to the field from the nursery. The fresh leaves were immediately subjected to sundrying in an open cleaned concrete floor space until moisture content became constant at 13%. The sundried leaves were later milled using a commercial feedmilling machine (Artec, model 20). The proximate analysis, amino acid profile and mineral content were determined to chemically evaluate the nutritional potentials of the ACLM. Thereafter, the ACLM was used to formulate diets along with other ingredients purchased locally.

Proximate composition, gross energy, amino acids and mineral content determination

Proximate composition of the ACLM was determined by AOAC (1995) method while the amino acids were determined using amino acid analyzer model 80-2107-07 Auto Loader. The sodium and potassium contents were determined by flame photometry while phosphorus was determined by the Vanado-molybdate method (AOAC, 1995). The other mineral elements were determined after wet digestion with a mixture of nitric, sulphuric and hydrochloric acid using Atomic Absorption Spectrophotometer (AAS model SP9). Gross energy of the ACLM sample and the 6 formulated diets were determined against thermocouple grade benzoic acid using a Gallenkamp ballistic bomb calorimeter (Model CBB-330-0104L). The results showing the above determinations are presented in Tables 1 and 2.

Determination of Phytin and Oxalate

The extraction and precipitation of phytin in the sundried ACLM were done by the described method [26] while iron in the precipitate was determined as described [8]. Phytin was determined by using a 4:6 Fe/P ratio to calculate phytin phosphorus and multiplying the phytin phosphorus by 3.55 as suggested [27]. Oxalate content was determined by the titrimetric method [14] as modified [17]. Where extracts were intensely coloured, they were decolourised with activated charcoal [4].

Site Preparation

The poultry house was thoroughly disinfected, fumigated with 1 part of Potassium Permanganate pellets to 3 parts of formalin. Thereafter, the house was rested for 2 weeks before the arrival of the experimental broiler starter chicks.

Experimental rations formulation

The feed ingredients used in ration formulation were purchased locally. The ACLM was sourced as earlier discussed. The results of the proximate compositions earlier determined were used as guides in the manual ration formulation of the six experimental diets. The experimental diets were prepared and adequately mixed in the mixer. All diets were compounded to contain identical crude protein content (isonitrogenous) and gross energy (isocaloric). Diet 1 was the control diet and was formulated without the inclusion of ACLM. Diets 2, 3, 4, 5 and 6 were formulated such that ACLM was incorporated at 5%, 10%, 15%, 20% and 25% respectively.

Other notable protein sources in all diets were fish meal at 2% inclusion levels, palm kernel cake at 10% inclusion

Composition (g/100g)	ACLM
Dry matter	88.6 <u>+</u> 0.01
Crude protein	23.0 <u>+</u> 0.55
Ether extracts	5.4 <u>+</u> 0.01
Crude fibre	8.8 <u>+</u> 0.01
Ash	19.3 <u>+</u> 0.01
Nitrogen free extract	43.5 <u>+</u> 0.52
Gross energy (kcal/g)	3.25 <u>+</u> 0.01
Amino acids	
Alanine	1.24
Aspartic acid	1.78
Arginine	2.11
Glycine	0.63
Glutamic acid	0.12
Histidine	0.61
Isoleucine	1.02
Lysine	2.01
Methionine	3.52
Cystine	0.81
Meth. + Cys.	4.33
Leucine	1.85
Serine	0.81
Threonine	0.52
Phenylalanine	1.51
Valine	1.04
Tyrosine	0.94
Tryptophan	0.64

Table 1: Proximate composition (g/100g), gross energy(kcal/g) and amino acid content (%) of Amaranthuscruentus leaf meal (ACLM) (means, n = 2)

Table 2: Mineral composition of *Amaranthus cruentus* leaf meal (ACLM) (means, n = 2)

ACLM	Ca	Р	K	Na	Mg	Fe	Mn	Cu	Zn
-			g/100g				(pp	om)	
	2.4	1.8	5.8	7.2	3.1	1175	198	36	890

Table 3: Phytic acid, phytin-P and oxalic acid content of Amaranthus cruentus
leaf meal (ACLM) (means, $n = 2$)

				$O_{1} + (100)$
ACLM	Phytic acid	Phytin-P	Phytin-P	Oxalate (mg/100g)
	Mg/100g	(mg/100g)	As % of total P	
	680	160	12.2	620

level and groundnut cake at 33%, 32%, 29%, 27%, 26% and 24% in diets 1, 2, 3, 4, 5, and 6, respectively. All diets were also supplemented with feed grade methionine and lysine.

Broiler birds husbandry and experimental design

A total of 540 day-old broiler chicks of the anak heavy strain were purchased from Zartech hatchery, a division of Zartech Farms, Ibadan, Oyo-State (a reputable hatchery in Nigeria). All chicks were electrically brooded at the Gabof Research Farms, Aule Government Residential Area, Akure. They were fed a 24% crude protein broiler starter commercial ration ad libitum for the first 3 days after arrival from the hatchery prior to the commencement of the experiment. The chicks were also sexed on the second day of brooding as described [10]. Water was also provided ad libitum with appropriate antibiotics and antistress particularly after arrival. Routine medications and vaccinations were administered.

The experimental design was the completely randomized type with a total of 18 experimental units/replicates. After the uniform brooding of 3 days, the sexed chicks (15 males and 15 females) were randomly distributed into 18 experimental units. The chicks were assigned at

the rate of 90chicks/diet in 3 replications of 30chicks/ replicate such that the mean group weights were similar at the beginning of the experiment. The chicks were fed the experimental diet ad libitum for 21 days during which records on daily feed consumption and 3 days periodic weight changes were recorded.

Estimation of nitrogen retention, nitrogen digestibility and protein efficiency ratio

Total faeces voided during the last 5 days were collected, weighed, dried at 65-70°C in an air circulating oven for 72hrs and preserved while the corresponding feed consumed was also recorded for nitrogen studies. The nitrogen contents of the samples were determined by the method of AOAC (1995). Nitrogen retained was calculated as the algebraic difference between feed nitrogen and feacal nitrogen (on dry matter basis) for the period. Nitrogen digestibility was computed by expressing the nitrogen retained as a fraction of the nitrogen intake multiplied by 100. The protein efficiency ratio was calculated as the ratio of weight gain to total protein consumed.

Blood collection for analysis

At the end of the feeding trial, a male chick per replicate

				(8 8)		
]	Diets		
	1	2	3	4	5	6
Ingredients		C	% inclusion	levels of A	CLM	
	0	5	10	15	20	25
Maize (11.0% CP)	50.70	46.70	44.70	41.70	37.70	34.70
Groundnut cake (45.0% CP)	33.00	32.00	29.00	27.00	26.00	24.00
Palm kernel cake (18.8% CP)	10.00	10.00	10.00	10.00	10.00	10.00
Fish meal (68.0% CP)	2.00	2.00	2.00	2.00	2.00	2.00
ACLM* (23.0% CP)	-	2.00	10.00	15.00	20.00	25.00
Bone meal	2.50	2.50	2.50	2.50	2.50	2.50
Oyster shell	0.50	0.50	0.50	0.50	0.50	0.50
Nacl	0.50	0.50	0.50	0.50	0.50	0.50
DL-methionine	0.15	0.15	0.15	0.15	0.15	0.15
DL-Lysine	0.15	0.15	0.15	0.15	0.15	0.15
Premix**	0.50	0.50	0.50	0.50	0.50	0.50
Total	100.00	100.00	100.00	100.00	100.00	100.00
Analysed composition						
Crude protein (%)	23.42	23.41	23.39	23.41	23.40	23.29
Crude fibre (%)	4.31	4.42	4.71	4.70	4.85	5.34
Ether extract (%)	7.21	6.41	6.40	6.71	6.51	6.48
GE*** (kcal/100g)	462.4	462.3	462.1	461.5	461.7	462.1

Table 4: Composition of experimental diets (g/100g)

*ACLM, Amarathus cruentus leaf meal

**contained vitamins A (10,000,000iu); D(2,000,000 iu); E (35000 iu); K (1900mg); B12 (19mg); Riboflavin (7,000mg); Pyridoxine (3800mg); Thiamine (2,200mg); D Pantothenic acid (11,000mg); Nicotinic acid (45,000mg); Folic acid (1400mg); Biotin (113mg); and Trace elements as Cu (8000mg); Mn (64,000mg); Zn (40,000mg); Fe (32,000mg) Se (160mg); I₂ (800mg) and other items as Co (400mg); Choline (475,000mg); Methionine (50,000mg); BHT (5,000mg) and Spiramycin (5,000mg) per 2.5kg.

GE*** (kcal/100g) calculated based on 5.7kcal/g protein; 9.5kcal/g lipid; 4.0kcal/g carbohydrate. (Ng and Wee, 1989)

$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	ters $\frac{1}{0} = \frac{2}{5} = \frac{3}{96} \frac{4}{10 \text{ lnclusion levels of ACLM}} = \frac{5}{15}$ $\frac{0}{10} = \frac{15}{15} = \frac{20}{20}$ $\frac{114.7 \pm 3.80}{116.7 \pm 6.35} = \frac{10}{116.7 \pm 6.35} = \frac{20}{20}$ $\frac{114.7 \pm 5.03}{116.7 \pm 6.35} = \frac{10}{116.7 \pm 6.35} = \frac{20}{20}$ $\frac{114.7 \pm 5.03}{116.7 \pm 6.35} = \frac{20}{116.7 \pm 6.35}$ $\frac{484.0 \pm 31.80}{39.42^{\pm} \pm 2.92} = \frac{470.0 \pm 72.04}{33.16^{45} \pm 3.10} = \frac{358.1^{44} \pm 65.84}{47.90^{15} \pm 0.30}$ $\frac{366.0^{4} \pm 38.19}{39.42^{\pm} \pm 2.92} = \frac{372.9^{4} \pm 1.03}{43.16^{45} \pm 3.10} = \frac{358.1^{4} \pm 65.84}{45.96^{15} \pm 0.30}$ $\frac{39.42^{\pm} \pm 2.92}{1.58^{4} \pm 0.19} = \frac{332.2^{4} \pm 1.08}{1.66^{45} \pm 0.16} = \frac{358.1^{4} \pm 65.84}{1.69^{15} \pm 0.25} = \frac{359.1^{4} \pm 65.84}{1.60}$ $\frac{2.25^{4} \pm 0.05}{1.58^{4} \pm 0.19} = \frac{2.94^{5} \pm 1.04}{1.66^{45} \pm 0.16} = \frac{2.81^{5} \pm 0.51}{1.84^{45} \pm 0.25} = \frac{356^{5} \pm 0.47}{1.61^{5} \pm 0.25}$ $\frac{616t}{1.89^{4} \pm 0.20} = \frac{2.16^{45} \pm 0.16}{1.66^{45} \pm 0.16} = \frac{2.81^{5} \pm 0.51}{1.84^{45} \pm 0.25} = \frac{2.5^{5} \pm 0.47}{1.61^{5} \pm 0.25}$ $\frac{7}{1.61^{5} \pm 0.25} = \frac{2.1^{41} \pm 0.19}{1.56^{45} \pm 0.16} = \frac{2.81^{5} \pm 0.51}{1.61^{5} \pm 0.25} = \frac{2.5^{5} \pm 0.47}{1.61^{5} \pm 0.25} = \frac{2.5^{5} \pm 0.61}{1.56^{45} \pm 0.25} = \frac{2.5^{4} \pm 0.05}{1.66^{45} \pm 0.16} = \frac{2.81^{5} \pm 0.25}{1.61^{5} \pm 0.25} = \frac{2.5^{5} \pm 0.47}{1.61^{5} \pm 0.25} = \frac{2.5^{5} \pm 0.47}{1.61$					Diets		
ters $\begin{array}{c c c c c c c c c c c c c c c c c c c $	% Inclusion levels of ACLM 5 10 15 20 80 117.3±8.62 119.3±7.51 114.7±5.03 116.7±6.35 80 496.2±32.88 470.0±72.04 505.6±20.53 511.9±32.99 80 496.2±32.88 470.0±72.04 505.6±20.53 511.9±32.99 19 372.9 ⁴ ±29.94 332.2 ⁴ ±10.81 358.1 ⁴ ±65.84 359.1 ⁴ ±57.61 2 42.14 ^{ab±} ±1.32 43.16 ^{ab±} ±3.10 46.96 ^{b±} ±1.01 47.90 ^{b±} ±0.30 2.31 ^{av±} ±0.19 2.94 ^{b±} ±1.04 2.81 ^{b±} ±0.51 2.85 ^{b±} 0.47 1.61 ^{b±} ±0.25 1.75 ^{ab±} ±0.19 1.66 ^{ab±} ±0.16 1.84 ^{ab±} ±0.25 1.61 ^{b±} ±0.25 1.61 ^{b±} ±0.25 tow are significantly different (P<0.05).			2	ε	4	5	9
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$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	80 496.2 ± 32.88 470.0 ± 72.04 505.6 ± 20.53 511.9 ± 32.99 19 $372.9^{a}\pm29.94$ $332.2^{a}\pm108.1$ $358.1^{a}\pm65.84$ $359.1^{a}\pm57.61$ 2 $42.14^{ab}\pm1.32$ $43.16^{ab}\pm3.10$ $46.96^{bc}\pm1.01$ $47.90^{bc}\pm0.30$ 2 $2.31^{ac}\pm0.10$ $2.94^{b}\pm1.04$ $2.81^{b}\pm0.51$ $2.85^{b}\pm0.47$ 1.75^{ab}\pm0.19 $1.66^{ab}\pm0.16$ $1.84^{ab}\pm0.25$ $1.61^{b}\pm0.25$ row are significantly different (P<0.5).	Initial weight	114.7 ± 13.80	117.3 ± 8.62	119.3 ± 7.51	114.7±5.03	116.7±6.35	117.3 ± 1.90
$\begin{array}{llllllllllllllllllllllllllllllllllll$	19 $372.9^{a}+29.94$ $332.2^{a}+108.1$ $358.1^{a}+65.84$ $359.1^{a}+57.61$ 2 $42.14^{ab}-1.32$ $43.16^{ab}-3.10$ $46.96^{bc}-1.01$ $47.90^{bc}-0.30$ 2 $32.1^{ac}+0.10$ $2.94^{b}+1.04$ $2.81^{b}+0.51$ $2.85^{b}+0.47$ 1.75^{ab}-0.19 $2.94^{b}+1.04$ $2.81^{b}-0.51$ $2.85^{b}-0.47$ 1.75^{ab}-0.19 $2.94^{b}+1.04$ $2.81^{b}-0.51$ $2.85^{b}-0.47$ 1.75^{ab}-0.19 $2.94^{b}+1.04$ $2.81^{b}-0.55$ $1.61^{b}-0.25$ row are significantly different (P<0.05).	(g/cinck) Final Weight	484.0 ± 31.80	496.2 ± 32.88	470.0±72.04	505.6 ± 20.53	511.9 ± 32.99	506.2 ± 43.38
chick/day) $2.25^{a}\pm0.05$ $2.31^{ac}\pm0.10$ $2.94^{b}\pm1.04$ $2.81^{b}\pm0.51$ $2.85^{b}\pm0.47$ $1.89^{a}\pm0.29$ $1.75^{ab}\pm0.19$ $1.66^{ab}\pm0.16$ $1.84^{ab}\pm0.25$ $1.61^{b}\pm0.25$	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	(g/cmck) Average Weight Gain Average Feed	$366.0^{a} \pm 38.19$ $39.42^{a} \pm 2.92$	$372.9^{a}+29.94$ $42.14^{ab}+1.32$	$332.2^{a}+108.1$ $43.16^{ab}+3.10$	$358.1^{a} \pm 65.84$ $46.96^{bc} \pm 1.01$	$359.1^{a}+57.61$ $47.90^{bc}+0.30$	$218.9^{b}+67.42$ $51.63^{c}+3.38$
	row are significantly different (P<0.05). ole 6: Nitrogen utilization of broiler chicks fed ACLM-based diets DIETS	Consumption (g/chick/day) Feed Efficiency Protein	$2.25^{a}\pm0.05$ 1 89 ^a +0 29	$2.31^{\rm ac} \pm 0.10$ 1.75 ^{ab} + 0.19	$2.94^{b}+1.04$ 1 66 ^{ab} +0 16	$2.81^{b} \pm 0.51$ 1.84 ^{ab} \pm 0.25	$2.85^{b}\pm0.47$ 1.61^{b}\pm0.25	$4.45^{d}\pm0.47$ 1 $44^{b}\pm0.25$
	row are significantly different (P<0.05). ole 6: Nitrogen utilization of broiler chicks fed ACLM-based diets DIETS	Efficiency (PER)		1	1	1	1	1
		1			, ,		1	Ň

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 $46.77^{c} \pm 6.78$

 $56.73^{b} \pm 6.55$

 $48.43^{bc} \pm 6.29$

 $59.09^{ab} \pm 15.90$

 $63.12^{a} \pm 10.28$

 $59.71^{ab} \pm 12.45$

Digestibility

Means with different superscripts in the same horizontal row are significantly different (P < 0.05); ACLM – Amaranthus cruentus leaf meal.

 $0.82^{b+0.03}$

 $1.29^{ab}\pm0.29$

 $1.05^{c}\pm0.03$

 $1.13^{b}\pm0.17$

 $1.48^{a} \pm 0.24$

 $1.24^{ab} \pm 0.24$

Nitrogen Retention

gN/chick/day Nitrogen D (%)

Nitrogen Intake gN/chick/day

 $\frac{25}{1.73^{b}+0.36}$

 $\frac{20}{2.25^{a}+0.26}$

 $2.19^{a} \pm 0.22$

10 1.98 ^{ab}±0.44

 $\frac{5}{2.34 \ ^{a}\pm 0.06}$

 $\frac{0}{2.08^{a}\pm0.10}$

Parameters

15

% Inclusion levels of ACLM

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was randomly selected, weighed and scarified by severing the jugular vein and blood allowed to flow freely into labeled bottles one of which contained a speck of EDTA while the other without EDTA was processed for serum. The serum was kept deep frozen prior to analysis. The packed cell volume (PCV%) was estimated by spinning about 75:1 of each blood sample in heparinized capillary tubes in an haematocrit micro centrifuge for 5 minutes while the total red blood cell (RBC) count 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.

Carcass, muscle and organ measurements

After slaughtering, the carcasses were scalded at 75°C in a water bath for about 30 seconds before defeathering. The dressed chicks were later eviscerated. The measurement of the carcass traits (dressed weight %, eviscerated weight %, thigh, drumstick, shank, chest, back, neck, wing, bellyfat and head) were taken before dissecting out the organs. The organs measured were the liver, kidneys, lungs, pancreas, heart, spleen, bursa of fabricus and gizzard. The following muscles: 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 weight, length and breadth of these muscles were taken. All the carcass traits, except the dressed and eviscerated weights, were expressed as percentages of the live weight while the organs and muscles were expressed in gkg⁻¹ body weight, while the length and breadth of the muscles were expressed in cmkg-1 body weight.

Statistical analysis

Data were analysed using the ANOVA (SPSS 11.0 for Windows) (SPSS Inc., Chicago IL, USA).

RESULTS AND DISCUSSION

Proximate composition, gross energy, amino acids, mineral content and antinutritional factors

The results of proximate composition, gross energy and amino acids content are presented in Table 1 while the mineral composition is presented in Table 2. The Amaranthus cruentus leaf meal (ACLM) was relatively high in crude protein at 23.0%+0.55; fat at 5.4%+0.01 and sugar + starch (NFE) at 43.5%+0.52. The ACLM was remarkably rich in mineral elements such as Ca, K, Na, Mg, Fe and Zn compared to reported levels of these mineral elements in most plant protein sources. The

Table 7: Haematological indices of broiler chicks fed ACLM-based diets	Diets	1 2 3 4 5 6	ters % Inclusion levels of ACLM	0 5 10 15 20 25		but $2.1^{a}\pm0.2$ $2.0^{a}\pm0.1$ $2.3^{a}\pm1.0$ $2.0^{a}\pm0.3$ $2.1^{a}\pm0.3$ $2.1^{a}\pm0.3$ $2.1^{a}\pm0.3$	mm^3)		(%) $7.4^{a}\pm0.9$ $7.2^{a}\pm0.4$ $7.1^{a}\pm0.7$ $7.0^{a}\pm0.2$ $7.1^{a}\pm0.4$ $7.0^{a}\pm0.5$	$9.5^{a} \pm 0.3$ $9.3^{a} \pm 0.4$ $9.4^{a} \pm 0.7$ $9.3^{a} \pm 0.8$	$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	in) $4.3^{a} \pm 0.6$ $4.3^{a} \pm 0.5$ $4.1^{a} \pm 0.4$ $4.2^{a} \pm 0.3$ $4.2^{a} \pm 0.5$ $4.1^{a} \pm 0.5$	= Amaranthus cruentus Leaf Meal; PCV = Packed Cell Vol	MCHC = Maan Cell Haemonlohin Concentration: MCH = Mean Cell Haemonlohin: MCV = Mean Cell Volume: FSR = Fruthrowthe Sedimentation Rate
			Parameters		PCV (%)	RBC count	$(x10^{6}/mm^{3})$	Hbc $(g/100ml)$	MCHC (%)	MCH (pg)	MCV (μm^3)	ESR (mm)	ACLM = $Amar$	MCHC = Mean Call

TROPICAL VEGETABLE (AMARANTHUS CRUENTUS) LEAF MEAL AS ALTERNATIVE PROTEIN SUPPLEMENT IN BROILER STARTER DIETS: BIONUTRITIONAL EVALUATION

			Γ	Diets						
	1	2	3	4	5	6				
Parameters	% Inclusion levels of ACLM									
	0	5	10	15	20	25				
Total Serum Protein	$9.8^{a} \pm 0.4$	9.6 ^a <u>+</u> 1.4	9.7 ^a ±1.2	9.3 ^a ±1.3	$9.4^{a} \pm 1.2$	9.5 ^a +1.3				
(g/100g)										
Albumin (g/100g)	$0.6^{a} \pm 0.1$	$0.7^{a}+0.2$	$0.6^{a} \pm 0.2$	$0.7^{a}\pm0.3$	$0.7^{a}\pm0.4$	$0.6^{a} \pm 0.5$				
Globulin (g/100g)	9.1ª <u>+</u> 0.5	$9.2^{a} \pm 0.4$	$9.0^{a} \pm 0.2$	9.1 ^a ±0.2	9.3 ^a <u>+</u> 0.3	$9.2^{a}+0.4$				
Albumin/Globulin Ratio	$0.1^{a} \pm 0.2$	$0.1^{a}\pm0.2$	$0.1^{a}\pm0.1$	$0.1^{a}\pm0.1$	$0.1^{a}\pm0.1$	$0.1^{a}+0.1$				

Table 8: Serum Metabolites of broiler chicks fed ACLM-based diets

Means with different superscripts in the same horizontal row are significantly different (P<0.05)

]	Diets		
	1	2	3	4	5	6
Parameters			% Inclusion	levels of ACLN	1	
	0	5	10	15	20	25
Total Liver Protein	$10.1^{a} \pm 0.4$	9.9^{a} <u>+</u> 0.1	$10.0^{a} \pm 0.4$	$10.1^{a}\pm0.2$	$9.9^{a}+0.1$	$10.0^{a} \pm 0.2$
(g/100g)						
Albumin (g/100g)	$2.6^{a} \pm 0.1$	$2.7^{a}\pm0.2$	$2.6^{a} \pm 0.3$	$2.6^{a}\pm0.1$	$2.5^{a}\pm0.1$	$2.6^{a}\pm0.1$
Globulin (g/100g)	7.4^{a} <u>+</u> 0.4	7.3 ^a <u>+</u> 0.3	7.5 ^a <u>+</u> 0.3	7.5 ^a <u>+</u> 0.2	$7.4^{a}\pm0.1$	$7.3^{a}\pm0.2$
Albumin/Globulin Ratio	0.3 ^a ±0.1	0.3 ^a ±0.2	0.4 ^a ±0.1	0.3 ^a <u>+</u> 0.2	0.3ª0.1	$0.4^{a}\pm0.1$

Means with different superscripts in the same horizontal row are significantly different (P<0.05).

phytic acid and oxalate levels (Table 3) were relatively higher than most other plant protein origins at 680mg/ 100g and 620mg/100g, respectively. The phytin-P was also high at 160mg/100g. The protein level and amino acids composition of ACLM clearly give it a rating in the category of other conventional protein sources especially of plant origins [15] at 23.0%+0.55 CP and the well balanced amino acid profile particularly its rich source of methionine and lysine [1]. About 75% of the total nitrogen in most vegetables is protein-nitrogen although this proportion varied with vegetable species [22]. The ash (mineral) content was remarkably high and a further investigation revealed that ACLM was a rich source of Ca, Mg and Fe and to a lesser extent K, Na and Zn [2]. The notable antinutritional factors (ANFs) found in ACLM are phytins and oxalates [2, 11].

Broiler performance characteristics

The performance characteristics data are presented in Table 5. The average weight gain (WG) value of birds on diet 2 (5% ACLM inclusion) was consistently higher at $372.9\pm29.9g$ /chick than for some other diets. The average feed consumption (FC) increased geometrically across the diets from diet 1 to diet 6. However, the FC values of birds on diets 1, 2 and 3 were similar (P>0.05). The feed efficiency (FE) values of birds on the control (basal) diet 1 and diet 2 were similar (P > 0.05). The protein

efficiency ration (PER) of birds on diet 2 was the best but statistically similar (P > 0.05) to PER values obtained for birds on diets 1, 3, 4, 5 and even 6. The average weight gain (WG) of birds on diet 2 (5% ACLM inclusion) was consistently higher than for other diets. The feed consumption (FC) of birds increased from diet 1 to diet 6 where the FC value was highest. The feed efficiency (FE) and protein efficiency ratio (PER) revealed birds on diet 2 as having a relatively good feed conversion into body weight gain and protein efficiency parameter. The highest WG value obtained for birds on diet 2 with 5% ACLM inclusion level may not be unconnected with the recognized well balanced amino acid profile in ACLM [1] and the growth factors earlier reported in certain amaranth plants [22]. The increased feed consumption experienced by birds on diet 2 to diet 6 may be as a result of the increased fibre level inadvertently introduced by the increased levels of inclusion of ACLM from diet 2 to diet 6.

It has earlier been recognized that the major drawbacks to the use of vegetable materials as major sources of nutrients by monogastrics (including man) are their high fibre and bulkiness which call for large quantities to be consumed to provide adequate levels of nutrients [2]. Another possible drawback is the content of phytin. Phytic acid can bind with proteins to form phytate-protein

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complexes [21]. This complex can adversely affect the digestibility of protein [19] by inhibiting a number of digestive enzymes in the gastro-intestinal tract such as pepsin [6], trypsin [5] and chymotrypsin [24] thereby reducing the digestibility of proteins and amino acids.

Nitrogen utililization

The nitrogen utilization records are presented in Table 6. The nitrogen intake, nitrogen retention and nitrogen digestibility values for birds on diet 2 were superior to the values obtained for birds on other diets although statistically similar (P > 0.05) to some other values obtained. There was a clear superiority in the nitrogen utilization indices of birds on diet 2 over the other diets particularly the nitrogen retention (NR) and nitrogen digestibility (ND). The consumption of ACLM in measured quantities may not have a negative nitrogen digestibility index and if properly consumed with other veritable protein sources as supplements may actually produce a comparable nitrogen utilization with the conventional nitrogen sources of acceptable standards [13, 15].

Haematology

The haematological serum and liver metabolites indices are shown in Table 7, 8 and 9 respectively. All haematological parameters investigated showed no significant differences (P > 0.05) in their mean values. This was also true for serum and liver metabolites which showed similar values (P > 0.05) for all treatment means. The haematological indices, serum and liver metabolites investigated in the present study revealed similar values with no significant difference. The values obtained for most of these haematological indices were similar and generally agreed with standard values obtained in previous studies [1, 3]. The blood variables most often affected by dietary influences were identified as PCV, plasma protein, glucose and clotting time [1]. These values in the present study were consistently higher than most values earlier reported and comparable with the report for chicks fed soya bean in place of fish meal [3]. On a similar note, the MCHC, MCH and Hbc were not significantly affected by the dietary treatments suggesting similar haemoglobin contents. The ESR of the birds on the ACLM based diets were similar to the control diet indicating that the test diets did not predispose the birds to any known general infection or malformation of any kind. It has been reported that ESR are increased in cases of acute general infection, malignant tumors and pregnancy [9]. There was also no mortality throughout the experimental period. The total serum protein (TSP), albumin, globulin and albumin/globulin ratio had similar (P > 0.05) values. This was also true for total liver protein (TLP), albumin,

490.0+13.5 46.1 ± 1.6^{a} 102.2±1.9 t3.0+6.8^a 52.0+0.8^a 91.3 ± 0.7 82.1+0.7 25 9 50.3 + 4.440.4 + 1.22.6+1.0 30.4+1.7 43.1 ± 7.4^{a} 20 485.0+14. $91.4\pm0.8^{\circ}$ 81.5±0.4^a 01.8 ± 1.8 Ś 51.9 ± 0.4 46.5+1.7 2.5±1.4^a 1.0+1.9 50.6 ± 0.1 40.0 ± 1.7 % Inclusion levels of ACLM 5 46.1 ± 1.3^{a} 102.0 ± 1.8^{a} 4 t90.1<u>+</u>13. Means with different superscripts in the same horizontal row are significantly different (P<0.05); ACLM, Amaranthus cruentus leaf mea 90.0+1.2^{bc} 32.0+0.5^a 30.8+1.8^a 52.9+7.0 33.8+0.3 t2.9+6.1⁸ 2.6+1.1^a 39.9+5.4 Diets 10 187.0 ± 15.2 $39.5+1.0^{ab}$ 30.3+1.8 39.7+4.3 53.1+3.5 81.5+0.7 46.3+2.7 101.3 + 2.13.0+6.3 78.9+0.3 2.3 ± 0.3 485.2 ± 12.0 01.5+1.8 90.7 ± 1.3^{ab} 81.9+0.5^a 46.1+2.8^a 2 $3.1+6.0^{a}$ $63.0+3.4^{a}$ 30.1+2.1 87.3+0.9 40.1 ± 3.5 $2.5+0.1^{a}$ 480.0+13. 82.0+0.8^a $89.7+0.9^{a}$ $46.8+3.5^{a}$ 42.9+5.9^a $63.4+2.6^{a}$ C 29.9+3.7 81.2+0.5 39.1 ± 3.3 02.1+2. 2.7±0.7° body weight) body weight) body weight) Thigh (gkg⁻¹ body weight) body weight) body weight) body weight) Back (gkg⁻¹ body weight) Eviscerated weight (%) Parameters Dressed weight (%) Drumstick (gkg⁻ Live weight (g) Backfat (gkg⁻ Shank (gkg⁻ Neck (gkg⁻¹ Wing (gkg⁻ Head (gkg⁻

Table 10: Carcass traits of broiler chicks fed ACLM-based diets

			D	Diets		
		2	ε	4	5	9
Parameters			% Inclusion le	% Inclusion levels of ACLM		
	0	5	10	15	20	25
Liver	19.2 ± 1.6^{a}	20.0 ± 0.3^{a}	19.7 ± 0.4^{a}	19.3 ± 1.5^{a}	19.6 ± 1.8^{a}	19.9 ± 1.4^{a}
Kidney	6.8 ± 0.1^{a}	$6.5+0.7^{a}$	6.6 ± 0.4^{a}	6.5 ± 1.2^{a}	6.7 ± 1.5^{a}	6.5 ± 1.8^{a}
Heart	7.2 ± 0.3^{a}	7.1 ± 1.8^{a}	8.0 ± 0.4^{ab}	8.5 ± 1.3^{bc}	$8.9\pm1.5^{\circ}$	$8.9\pm 2.1^{\circ}$
Spleen	1.1 ± 0.2^{a}	1.2 ± 0.3^{a}	1.1 ± 0.3^{a}	1.2 ± 0.1^{a}	1.4 ± 0.5^{a}	1.1 ± 0.4^{a}
Pancreas	2.9 ± 0.3^{a}	2.9 ± 0.5^{a}	3.1 ± 0.3^{a}	3.0 ± 0.7^{a}	2.9 ± 0.5^{a}	3.1 ± 0.4^{a}
Bursa	2.7 ± 0.7^{a}	$2.8+0.4^{a}$	$2.8+0.4^{a}$	$2.8+0.9^{a}$	2.7 ± 0.7^{a}	2.7 ± 0.5^{a}
Gizzard	$36.8+3.8^{a}$	37.1 ± 0.5^{a}	37.4 ± 1.8^{a}	$37.0+1.9^{a}$	36.9 ± 0.9^{a}	37.2 ± 1.8^{a}
Lung	6.7 ± 1.0^{a}	6.7 ± 1.8^{a}	6.7 ± 2.1^{a}	$6.6+2.5^{a}$	6.7 ± 1.2^{a}	6.7 ± 1.3^{a}

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				a		Sa		S ^a		
	9		25	8.5 ± 1.9^{a}		23.8 ± 1.8^{a}		31.2 ± 1.8^{a}		
	5		20	8.6 ± 0.8^{a}		23.5 ± 1.8^{a}		31.8 ± 1.8^{a}		<i>ruentus</i> leaf meal
Diets	4	% Inclusion levels of ACLM	15	8.6 ± 0.6^{a}		23.8 ± 1.2^{a}		31.3 ± 0.9^{a}		ACLM, Amaranthus c
	3	% Inclusion	10	8.5 ± 0.9^{a}		$24.0+1.9^{a}$		$31.5+2.0^{a}$		ly different (P<0.05); /
	2		5	8.6 ± 0.2^{a}		23.7 ± 3.2^{a}		31.4 ± 1.9^{a}		ntal row are significant
	1		0	8.6 ± 0.7^{a}		23.4 ± 2.8^{a}		31.2 ± 1.8^{a}		ts in the same horizor
		Muscle		Inner chest muscle	(Supra coracoideus)	Outer chest muscle	(Pectoralis thoracicus)	Thigh muscle	(Gastrocnemius)	Means with different superscripts in the same horizontal row are significantly different (P<0.05); ACLM, Amaranthus cruentus leaf meal

				Diets		
	1	2	ω	4	5	9
Muscle			% Inclusion	% Inclusion levels of ACLM		
	0	5	10	15	20	25
Length of inner chest muscle (Sunra coracoideus)	$19.1_{-4.1^{a}}$	19.3 ± 5.2^{a}	19.4 ± 6.3^{a}	19.1 ± 0.5^{a}	19.2 ± 5.6^{a}	19.3 ± 6.1^{a}
Length of outer chest muscle	21.2 ± 5.7^{a}	22.0 ± 6.1^{a}	21.7 ± 7.2^{a}	21.8 ± 8.1^{a}	21.7 ± 6.2^{a}	21.5 ± 6.8^{a}
(<i>Pectoralis thoracicus</i>) Breadth of inner chest muscle	3.4 ± 0.2^{a}	3.5 ± 0.3^{a}	3.6 ± 0.4^{a}	3.6 ± 1.2^{a}	3.6 ± 0.8^{a}	3.5 ± 1.2^{a}
(Supra coracoideus)						
Breadth of outer chest muscle (<i>Pectoralis thoracicus</i>)	6.9 ± 0.8^{a}	7.1 ± 0.5^{a}	7.1 ± 0.8^{a}	7.1 ± 0.6^{a}	7.0 ± 1.2^{a}	7.1 ± 0.5^{a}

Carcass characteristics

Records on carcass traits are shown in Table 10. All carcass traits measured except the dressed weights and backs of experimental chicks were similar (P > 0.05). The records on the relative organs weight are presented in Table 11. Except for the heart, all other organs measured were statistically similar (P > 0.05). The relative weight and relative length and breadth of some muscles are presented in Tables 12 and 13. The inner chest muscle (Supra coracoideus), outer chest muscle (Pectoralis thoracicus) and thigh muscles (Gastrocnemius) were all similar (P >0.05) in their treatment means for all chicks investigated. The adipose deposition around the heart may have been facilitated by the increase inclusion levels of ACLM. A uniform growth pattern and muscle development in all birds on the ACLM based diets compared with the control diet and also with many previous standard growth patterns and muscle development of birds of the same age and strain [16, 20].

CONCLUSIONS

The results of the study showed that the proximate composition, gross energy, amino acids content and mineral composition all revealed that ACLM is a potentially rich source of nutrients in monogastric feed formulation. The processing effect of sundrying appreciably reduced the antinutritional factors (ANFs) to innocuous levels that enhanced higher tolerant levels in the test animals. Inclusion level of 5% of ACLM in broiler diets was found to be most suitable in facilitating better performance characteristics. The nitrogen utilization, muscle development and haematological indices were all in favour of an inclusion level of 5% in broiler starter diets.

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