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Nutritional implications of Cassoya diet supplemented with protease enzyme on growth performance, nutrient digestibility, haematology and serum of broiler finisher chickens

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***Target Audience:** Animal Scientists, Researchers, Farmers and Policy Makers*

Abstract

This study evaluated the implications of cassoya-based nutrition (mixture of cassava root meal and full-fat soybean in ratio 40:60) supplemented with protease enzyme on growth performance and nutrient digestibility of broiler finisher chickens. Three hundred three weeks old ROSS 308 broiler starter chicks were placed on 6 dietary treatments for 3 weeks; in 5 replicates of 10 birds each. The first to third treatments had cassoya inclusion levels of 0, 50 and 100 % without enzyme while treatments 4 to 6 had protease enzyme supplementation at 10 g/tonne. Data collected were analyzed using Analysis of Variance in a Completely Randomized form. The diets supplemented with protease enzyme significantly improved (P<0.05) all the measured parameters except protein efficiency ratio. Treatments had significant (p<0.05) effects on all haematological parameters except packed cell volume and mean corpuscular haemoglobin concentration. Serum metabolites were significantly influenced by cassoya supplemented with enzyme. Birds efficiently utilized the feeds with 50% cassoya maize replacement inclusion without any detrimental effect and at a reduced cost of production. Cassoya diet at 50% level of inclusion with enzyme supplementation in broiler finisher diet is recommended for improvement of growth performance and nutrient digestibility.

Key words: Diet; feed cost; haemoglobin; processing; proximate; soybean; treatment

Description of Problem

Poultry require nutrients for maintenance and growth. The diet must contain essential materials for maintenance, production and reproduction (1). Increase in feed price is one of the major constraints of poultry production. The total cost of production of feed is 70% (2) which means efforts to increase poultry industry productivity should be directed towards improved energy feedstuff that could replace maize during period of scarcity to ensure optimum performance of the birds. Maize (*Zea mays*) has been the most commonly used conventional energy source of plant origin in feed formulation for poultry. It is sometimes highly expensive, limited or even scarce. Apart from this, maize and other cereal grains are highly competed for by other animals and humans as food. Hence, a non-conventional energy source needs to be considered and studied for better output in poultry farming (3). Exploiting the potentials of other energy sources such as cassava root as alternative for maize in poultry diets is a good initiative (4).

Cassava (*Manihot esculenta*), is a common tropical root crop extensively grown in the tropics as a major source of energy food for both man and animals (5). In 2020, global production of cassava root was 304 million tonnes which is expected to reach 350.8 million tonnes by 2027. Nigeria is the world's largest producer of cassava with a production capacity of 60,000,000 metric tonnes (6). Different processing methods including peeling, chipping, grating, fermenting, drying and ensiling with different materials such as rumen filtrate, rumen digesta and soybeans to improve the protein content has being employed (7). In view of the above, full-fat soya can serve as a good source of protein and help to reduce the dustiness in cassava. It improves texture and palatability of the feed, decreases dust production and reduces wastage in mash feeds and may improve the digestibility of remaining nutrients of the diet. The use of *cassoya* (mixture of cassava root meal and full-fat soybean in ratio 40:60) as an alternative to conventional energy feedstuff like maize could help to reduce feed costs and increase meat production. According to (1), introduction of feed enzymes in the feed will exert beneficial effect on nutrient utilization. (8) stated that enzyme supplementation increases final live weight of birds thereby increases feed availability for digestion and absorption. Thus, alternative cheaper sources of energy feedstuff such as cassoya for poultry feed must be exploited.

This study therefore evaluated the growth performance, nutrient digestibility and blood parameters of *ROSS 308* broiler birds fed with *cassoya* diet supplemented with protease enzyme.

Materials and Methods

Experimental Site

The studies were carried out at the Poultry Unit of the Directorate of University Farms (DUFARMS), College of Animal Science and Livestock Production, Federal University of Agriculture, Abeokuta, Ogun-State, Nigeria. The farm is located on latitude 7° 13' 35.48"N and Longitude 3° 26' 12.38"E with an elevation of 415 feet and eye altitude of 700 feet (9). This area lies in the rainforest vegetation zone which has humid climatic condition with an average rainfall of 1,037 mm, a mean ambient temperature of about 34.7ºC and yearly average relative humidity of 83%.

Experimental Materials

Cassava tubers were purchased from a private farm in Odeda Local Government while full-fat soybean grains and other feed ingredients were purchased from a reputable feed mill in Abeokuta. Cassava tubers were washed in clean water, grated, screw pressed and dried with solar drier. Cassava and full-fat soya were mixed manually in ratio 40:60 respectively. Enzyme was sourced from

Nutrivitas limited.

The singled enzyme used was Ronoxyme(R)proACT enzyme.

Proximate Analysis of Cassava and Full-fat Soya

The proximate analysis of cassava and full-fat soya used in *cassoya* diet formulation as seen in Table 1 shows that they contained 94.0% and 84.50% dry matter, 1.75% and 34.74% crude protein, 5.0% and 16.5% ether extract, 13% and 8% crude fibre and 1.5% and 6.5% ash respectively. Neutral detergent fibre, acid detergent fibre and acid detergent lignin for cassava were 26%, 7% and 0.59% while that of full-fat soya were 44%, 29% and 3.0% respectively.

Parameter	Cassava	Full-fat soybeans
Moisture (%)	6.00	15.50
Ash (%)	1.50	6.50
Ether Extract (%)	5.00	16.50
Crude fibre (%)	13.00	8.00
Dry matter (%)	94.00	84.50
NFE (%)	78.75	34.26
Crude protein (%)	1.75	34.74
ADL (%)	0.50	3.00
NDF (%)	26.00	44.00
ADF (%)	7.00	29.00
Calculated		
Metabolisable energy (kcal/kg)	3265.88	3839.76

NFE: Nitrogen free extract, ME: metabolisable energy, NDF: neutral detergent fibre, ADF: acid detergent fibre, ADL: acid detergent lignin.

Management of Experimental Animal

A total of three hundred three-weeks old ROSS 308 broiler starter chicks were used for this study. The poultry pen and the equipment were thoroughly washed and disinfected before the arrival of the chicks. The birds were raised on battery cage system for 3 weeks. Routine management and medication (drug and vaccine schedules were observed. Feed and water were supplied to the birds *ad libitum.*

Experimental Design

The three hundred three-weeks old broiler chicks were randomly allotted to six (6) dietary treatments. Each treatment group was further divided into five (5) replicate groups of ten birds each in a 3x2 factorial arrangement, using complete randomized design (CRD).

Experimental Diets

The experimental diets consist of six

(6) dietary *cassoya* inclusion levels replacing 0%, 50%, and 100% maize with and without protease enzyme supplementation at 10 kg/tonne. Tables 2 shows the six (6) experimental diets formulated.

Table 2: Percentage composition of experimental diets during the finisher phase (4–6 weeks)

	-Enzyme (%)			+Enzyme (%)		
	0	50	100	0	50	100
Ingredients	**D1**	**D2**	**D3**	**D4**	**D5**	**D6**
Maize	48.0	25.96		48.0	25.96	
FFSB	36	18		36	18	
Cassoya		42.9	85.8		42.9	85.8
Wheat offal	12	12	11	12	12	11
Bone meal	0.7	0.7	0.7	0.7	1.0	0.7
Oyster shell	2.0	2.0	2.0	2.0	2.0	2.0
Lysine	0.5	0.1	0.5	0.5	0.1	0.5
Methionine	0.2	0.3	0.2	0.2	0.3	0.2
Premix	0.3	0.3	0.3	0.3	0.3	0.3
Salt	0.3	0.3	0.3	0.3	0.3	0.3
Total	100.00	100.00	100.00	100.00	100.00	100.00
Calculated Analysis						
Crude protein (%)	20.34	20.01	19.57	20.34	20.01	19.57
Fat (%)	9.00	8.51	8.50	9.00	8.51	8.50
Fibre (%)	6.50	6.71	7.54	6.50	6.71	7.54
Ash (%)	8.60	8.60	10.00	8.60	8.60	10.00
NFE (%)	56.83	55.08	53.62	56.83	55.08	53.62
Lysine (%)	1.13	1.10	0.98	1.13	1.10	0.98
Methionine (%)	0.47	0.46	0.44	0.47	0.46	0.44

FFSB - full-fat soya bean. Premix composition per 2.5kg feed the following; Vitamin A (I.U.) 12,000,000, Vit. D_3 (I.U.) 2,500,000, Vit. E (mg) 40,000, Vit. K (mg) 2,000, Vit. B_1 (mg) 3,000, Vit. B_2 (mg) 4,000, Biotin (mg) 75, Folic Acid (mg) 1,000, Niacin (mg) 50,000, D Caal. Pn (mp) 11,000, Co. (mg) 300, Cu (mg) 8,000, I (mg) 1,500 Se (mg) 120, Zn (mg) 60,000, Fe (mg) 40, 000, Mn (mg) 100,000, Cl (mg) 300,000. NFE - Nitrogen free extract.

Data collection

Performance characteristics

The initial weights of the birds were taken. The live weights of the birds as well as the feed consumption of each replicate were measured weekly. Feed conversion ratio for each replicate was calculated by dividing the feed intake by the weight gain.

Digestibility Study

At the 3rd weeks of the study, two birds per replicate were randomly selected and transferred into separate metabolic cages. Known quantity of feed was supplied and excreta were collected using total tract collection method. Three days of acclimatization period were allowed prior to the collection of droppings. The wet excreta samples collected were weighed and dried at 65°C. The dried excreta samples were milled, allowed to pass through 0.5 mm sieve and used for proximate analysis to determine

(6) dietary *cassoya* inclusion levels replacing 0%, 50%, and 100% maize with and without protease enzyme supplementation at 10 kg/tonne. Tables 2 shows the six (6) experimental diets formulated.

$$\text{Dry matter digestibility} = \frac{\text{Feed intake (gDM)} - \text{Excreta output (gDM)}}{\text{Feed intake (gDM)}} \times 100$$

$$\text{Protein Digestibility} = \frac{\text{Protein in feed} - \text{Protein in Excreta}}{\text{Protein in feed}} \times 100$$

$$\text{Crude fibre Digestibility} = \frac{\text{CF in feed} - \text{CF in excreta}}{\text{CF in feed}} \times 100$$

$$\text{Fat digestibility} = \frac{\text{Fat in feed} - \text{Fat in excreta}}{\text{fat in feed}} \times 100$$

$$\text{Ash digestibility} = \frac{\text{Ash in feed} - \text{Ash in excreta}}{\text{Ash in feed}} \times 100$$

Haematological and Serum Parameters

At the end of third week of the study, 2.5 mL of blood was collected from two birds per replicate through the jugular vein into bottles containing ethylene diamine tetra acetate (EDTA) for haematological parameters (packed cell volume, haemoglobin concentrate concentration, white blood cell and red blood cell counts). Another 2.5 mL was collected in a hypodermic syringe to determine serum metabolites (total protein, globulin, albumin, uric acid and creatinine).

Experimental model

$$Y_{ijkl} = \mu + S_i + P_j + (SP)_{ij} + \sum_{ijk}$$

Where

Y_{ijk} = Trait of interest

μ = Population mean

S_i = Fixed effect due to ith cassoya level

P_j = Fixed effect due to jth enzyme supplementation

Results and Discussion

Main effect of *cassoya* inclusion and enzyme supplementation on performance characteristics and cost benefit analysis of finishing broiler chickens (4-6 weeks)

Table 3 shows the main effect of *cassoya* inclusion level and enzyme supplementation on the finishing broiler chickens. The result showed that final live weight, total weight gain, daily weight gain, daily feed intake, feed conversion ratio, protein efficiency ratio, cost of feed per kg and cost of feed per kg weight gain were significantly (P<0.05) influenced by *cassoya* inclusion levels. There was a linear decrease in value of final live weight, total weight gain, daily weight gain, daily feed intake, protein efficiency ratio and cost of feed per kg as the inclusion level increased. In poultry nutrition, one of the factors for improving growth is feed intake. Decreased feed intake resulted to decreased final weight and weight gain. Decreased feed intake as observed in this study may be attributed to reduced acceptability as a result the high fibre level and viscous nature of the cassava products. It might have created a gut feeling effect thereby reducing the appetite, due to high proportions of cassava in the diets compared to those on 0% and 50% *cassoya* based diets. This also agrees with the result of (11) who reported that acceptability is the major attribute of feed intake. According to (12) in an experiment, decrease in weight gain with diets containing higher proportion of cassava may be due to high fibrous level. Birds fed 0% and 50% *cassoya* had similar and higher means for final live weight (1666.40 g, 1642.30 g), total weight gain (1003.20 g, 981.40 g), daily weight gain (47.77 g, 46.73 g), daily feed intake (113.69 g, 108.73 g), feed conversion ratio (2.38, 2.34) and protein efficiency ratio (2.39, 2.36) while those on 100% *cassoya* recorded the least values. The improvement observed could be attached to the addition of full-fat soya which enhanced the protein and amino acid profiles. According to (13), broiler diets based on cassava meal were improved nutritionally by the addition of protein. Similar statistical values obtained for birds fed 0% and 50% cassoya in this study suggested that maize could be replaced by cassoya up to 50% without any impairment on the growth response The observation in this study supported the findings of (14) who reported that body weight was the highest when cassava was replaced with 50% maize. (15) opined that there was no reduction in growth performance when cassava peels meal substituted maize in broiler diet. This justification is further supported by the findings of (16) in his assertion that broiler birds can also perform better if maize is replaced by cassava. Improved processes methods have yielded satisfactory results and increased the use of cassava in livestock feeds (17).

Feed cost per kg varied significantly ($p<0.05$), from N122.62 to N154.50. Feed cost per kg weight gain ranged from N320.14 to N366.93, birds fed 0% and 100% *cassoya* recorded higher mean values while the least was recorded for 50% *cassoya* inclusion. Enzyme supplementation had significant ($p<0.05$) effect on final live weight, feed conversion ratio, protein efficiency ratio and cost of feed per kg. However, the trend observed was that birds fed enzyme supplemented diets had higher weight gain (666.47 g), final live weight (1605.13 g), protein efficiency ratio (2.32 g), and cost of feed per kg (N151.32), compared to those fed diet without enzyme. Feed conversion ratio was least in birds fed diets with enzyme supplementation. The improved values recorded for FCR might be attributed to the potential role of enzymes in breaking down the substrate, and as a result making more nutrient available to the birds resulting in improved growth. (18) opined that there was an increase in growth rate, feed intake and feed conversion when broiler diet was supplemented with exogenous enzyme. Bedford and Partridge (19) also reported that birds fed protease enzyme supplemented diets showed improved weights and increase metabolic activities.

Interaction effect of *cassoya* inclusion and enzyme supplementation on performance characteristics and cost benefit analysis of finishing broiler chickens (4-6 weeks)

The interaction effect (Table 4) of *cassoya* inclusion and enzyme supplementation revealed that diets significantly ($P<0.05$) influenced all the parameters measured, with the exception of protein efficiency ratio. Mean values for final live weight and initial weight ranged from 11120.20 g in birds fed with 100% *cassoya* without enzyme to 1738.60 g and 528 g to 734.40 g respectively, birds fed 0% and 50% *cassoya* with enzyme supplemented diets recorded similar and higher ($P<0.05$) values, while the least values were recorded in birds fed diet containing 100% *cassoya* without enzyme supplementation. This showed the advantage of incorporating enzymes in diets of poultry birds. According to (8) enzyme supplementation increases final live weight of birds through improved gut health and increases feed availability for digestion and absorption. Protein efficiency ratio was better for birds on enzyme supplemented diets. This indicates that feed nutrients were adequately digested. However, these results were not in agreement with the findings of (20) that dietary enzyme supplementation had no effect on body weight gain, feed intake and feed conversion ratio of broilers. Birds fed 0% and 50% *cassoya* without enzyme gave similar values for all the parameters measured with the exception of feed cost per kg and daily feed intake. Total weight gain and daily weight gain values ranged from 584.20 g to 1031.20 g and 27.82 g to 49.10 g respectively, thus birds fed 0% and 50% *cassoya* with enzyme supplementation had higher numerical but similar ($p>0.05$) values than those on 0% and 50% *cassoya* without enzyme supplementation, while those birds on 100% *cassoya* without enzyme had the least ($p<0.05$) values. The similarities observed indicated that *cassoya* diets were favorably compared to the control diet. This observation agrees with the report of (21) that nutritional fortification of cassava flour with fresh soybean meal has shown promising results in the poultry industry. Varying degrees of success have been documented with the use of cassava as replacement for maize in poultry diets (16). (22) observed that feeding concentrates supplemented with 50% cassava root meal showed advantages in broiler chickens compared to the use of conventional feedstuff. (19) opined that there was an increase in growth rate, feed intake and feed conversion when broiler diet was supplemented with exogenous enzyme. Feed intake values ranged from 87.06 g to 144.44 g, birds fed 50% *cassoya* without enzyme had the highest feed intake while those on 100% *cassoya* without enzyme recorded the least value. Those on 0% and 50% *cassoya* with enzyme and 0% *cassoya* without enzyme had similar feed intake values. Improved value observed in feed intake could be due to enzymatic effect and the oil from full-fat soya which might have improved the palatability and reduced the dustiness of the feed. Lowest feed conversion ratio of 2.10 was obtained from birds fed 50% *cassoya* with enzyme supplementation while those on 100% cassoya without enzyme had the highest mean value of 3.14 g. This was an indication of better feed utilization of *cassoya* diets with enzyme supplementation in meat type chickens which has earlier been reported by (23). Feed cost per kg varied significantly ($p<0.05$) and ranged from N109.73 to N151.10 across the experimental diets; highest values were recorded in birds fed 0% *cassoya* with enzyme while the least was observed in birds fed 100% *cassoya* without enzyme supplementation. Feed cost per kg weight gain ranged from N317.01 in birds fed 50% *cassoya* with enzyme to N388.30 obtained in birds on 0% *cassoya* with enzyme. The economic implication of cassava in broiler production was revealed by the decrease in feed cost as the inclusion level increased which reduced the cost of production and maximized profit. Similar report was revealed by (24) that the production cost could be reduced when cassava replaced maize in feed industry. Proact enzyme is used to reduce feed cost and does so successfully at 50% level of *cassoya* inclusion. Higher feed cost per kg was obtained in birds fed enzyme supplemented diets compared to those without enzyme; hence the benefit of enzyme inclusion was revealed with higher weight gain in birds fed enzyme supplemented diet and insignificant cost per kg weight gain among the dietary treatments. Feed cost per kg varied with enzyme supplementation. This was as a result of disparity in cost of maize and cassava. Despite the significant difference in weight gain, cost per kg weight of those on enzyme supplementation was reduced due to improved feed to gain ratio, this showed the beneficial effects of enzyme supplementation on *cassoya* based diet for broiler chickens.

Table 3: Main effect of *cassoya* inclusion and enzyme supplementation on performance and cost benefit analysis of finishing broiler chickens (4-6 weeks).

	Cassoya inclusion (%)				Enzyme supplementation		
Parameters	0	50	100	SEM	-enzyme	+enzyme	SEM
Initial weight (g/bird)	663.10^{a}	660.90^{a}	542.80^{b}	12.93	578.13^{b}	666.47^{a}	10.55
Final live weight (g/bird)	1666.40^{a}	1642.30^{a}	1226.10^{b}	19.28	1418.07^{b}	1605.13^{a}	43.21
Total weight gain (g/bird)	1003.20^{a}	981.40^{a}	683.30^{b}	24.62	839.93	938.67	32.30
Daily weight gain (g/bird)	47.77^{a}	46.73^{a}	32.54^{b}	1.17	39.10	44.70	1.52
Daily feed intake (g/bird)	113.69^{a}	108.73^{a}	91.65^{b}	2.66	106.06	103.32	2.90
Feed conversion ratio	2.38^{a}	2.34^{a}	2.89^{b}	0.07	2.72^{a}	2.35^{b}	0.70
Protein efficiency ratio (g/bird)	2.39^{a}	2.36^{a}	1.66^{b}	0.08	1.96^{b}	2.32^{a}	0.13
Feed cost per kg (₦)	154.50^{a}	138.20^{b}	122.62^{c}	0.01	125.55^{b}	151.32^{a}	3.90
Feed cost per kg gain (₦)	366.93^{a}	320.14^{b}	351.00^{a}	9.60	354.25	337.79	7.15

^{ab} Means on the same row having different superscript are significantly different (P<0.05)

Table 4: Interaction effect of *cassoya* inclusion and enzyme supplementation on performance characteristics and cost benefit analysis of finishing broiler chickens (4-6 week)

		-Enzyme (%)			+Enzyme (%)		
Parameters	0	50	100	0	50	100	SEM
Initial weight (g/bird)	592.00^{bc}	614.40^{b}	528.00^{d}	734.40^{a}	707.40^{a}	557.60^{cd}	18.31
Final live weight (g/bird)	1596.00^{b}	1546.00^{b}	1112.20^{d}	1736.80^{a}	1738.60^{a}	1340.00^{c}	27.27
Total weight gain (g/bird)	1004.00^{a}	931.60^{a}	584.20^{c}	1002.40^{a}	1031.20^{a}	782.40^{b}	34.82
Daily weight gain (g/bird)	47.81^{a}	44.36^{a}	27.82^{c}	47.73^{a}	49.10^{a}	37.26^{b}	1.65
Daily feed intake (g/bird)	116.68^{b}	144.44^{a}	87.06^{d}	110.70^{b}	103.01^{bc}	96.23$_{cd}$	3.76
Feed conversion ratio	2.44$_{bc}$	2.58$_{bc}$	3.14^{a}	2.32$_{cd}$	2.10^{d}	2.64^{b}	1.00
Protein efficiency ratio (g/bird)	2.31	2.14	1.42	2.46	2.58	1.90	0.13
Feed cost per kg (₦)	141.62^{c}	125.30^{e}	109.73^{f}	167.37^{a}	151.10^{b}	135.50^{d}	0.01
Feed cost per kg gain (₦)	345.55^{b}	323.28^{b}	344.55^{b}	388.30^{a}	317.01^{b}	357.45^{ab}	13.58

^{abcdef} Means on the same row having different superscript are significantly different (P<0.05)

Main effect of *cassoya* inclusion and enzyme supplementation on nutrient digestibility of finishing broiler chickens (4 - 6weeks)

Main effect of *cassoya* inclusion is shown in Table 5. *Cassoya* inclusion significantly ($p<0.05$) influence all the parameters measured with the exception of ash digestibility. Birds fed 0% and 50% *cassoya* inclusion had significantly higher means for crude protein, ether extract and crude fiber digestibility while the least was obtained in those on 100% *cassoya* inclusion, their mean values ranged from 55.42% to 68.47%, 56.13% to 68.67% and 57.56% to 67.98% respectively. The decreased nutrient digestibility observed in birds fed 100% *cassoya* diet can be attributed to shorter transit time of the high fibrous diets in the intestine of the birds. This is in accordance with an earlier study by (25) that digestibility indices decrease with increasing level of inclusion of ensiled cassava leaves in the diet. (26) also opined that high fibre increases the bulkiness of the diet, exact effect on gut transient time and reduce nutrient digestibility. Dry matter digestibility was highest ($p<0.05$) in birds fed 0% *cassoya* inclusion (77.24%) while those on 50% and 100% *cassoya* had lower values. In spite of high digestibility value of dry matter observed in 100 %, *cassoya*, crude protein digestibility was reduced compared to those on 50% *Cassoya.* This is supported by the report of Arigbede *et al.* (27) that the level of dietary protein in the ration did not affect dry matter digestibility. Enzyme supplementation significantly ($p<0.05$) increased crude protein, ether extract and crude fibre digestibility while those birds on diets without enzyme had lower values. The significant increase in digestibility of crude protein and ether extract with enzyme supplementation might be due to the fact that the enzyme had a complementary influence on digestive enzyme of gastrol intestinal tract that led to the increase in digestibility of birds fed the experiment diets (28)

Interaction effect of *cassoya* inclusion and enzyme supplementation on nutrient digestibility of finishing broilers chicken (4-6 weeks)

Interaction effect of *cassoya* inclusion and enzyme supplementation on nutrient digestibility of finishing broilers chicken as shown in Table 6, significantly ($p<0.05$) affected all the parameters measured. Birds offered 0% and 50% *cassoya* with enzyme supplementation had similar ($p>0.05$) and higher significant means for crude protein 70.00%, 72.16% and 72.19%, 72.54% for ether extract digestibility while those on 100% *cassoya* without enzyme recorded the least values. This is an indication that enzyme had increased the feeding value of raw material which allow digestion to occur more rapidly and completely (29). Crude fibre digestibility ranged from 58.56% in birds fed 100% *cassoya* with enzyme to 69.97% in birds fed 50% *cassoya* with enzyme. For ash digestibility, lowest mean value was obtained in birds that received 100% *cassoya* with enzyme while 50% *cassoya* with enzyme had the highest significant mean of 69.11%. Least values obtained in 100% *cassoya* without enzymes attest to the observed depressed performance. This reduction in feed digestibility might be attributed to decrease in nutrient available for growth, which could lead to low performance. According to (30), nutrient digestibility increases with age and differs between poultry species and feed ingredients. Dry matter digestibility varied significantly ($p<0.05$) with the highest in birds fed 0% *cassoya* without enzyme and the least was obtained in those on 50% *cassoya* with enzyme which is quite possible for dry matter digestibility.

Table 5: Main effect of *cassoya* inclusion and enzyme supplementation on nutrient digestibility of finishing broiler chickens (4–6 weeks)

	Cassoya inclusion (%)				Enzyme supple mentation		
Parameters (%)	0	50	100	SEM	-Enzyme	+Enzyme	SEM
Crude protein	68.01^a	68.47^a	55.42^b	1.46	61.05^b	67.21^a	1.86
Ether extract	66.75^a	68.67^a	56.13^b	1.37	60.30^b	67.40^a	1.77
Crude fibre	66.78^a	67.98^a	57.56^b	1.09	62.72^b	65.06^a	1.49
Ash	67.04	69.05	61.58	0.91	65.97	65.53	1.31
Dry matter	77.24^a	76.30^b	76.37^b	0.12	76.99	76.30	052

^{ab} Means on the same row having different superscript are significantly different ($P<0.05$)

Table 6: Interaction effect of *cassoya* inclusion and enzyme supplementation on nutrient digestibility of finishing broiler chickens (4-6 weeks)

	-Enzyme (10 kg/tonne)			+Enzyme (10 kg/tonne)			
Parameters (%)	0	50	100	0	50	100	SEM
Crude protein	65.79^b	65.98^b	51.38^d	70.00^a	72.16^a	59.47^c	2.36
Ether extract	61.30^b	64.81^b	54.78^d	72.19^a	72.54^a	57.47^{cd}	2.27
Crude fibre	65.08^a	64.60^b	58.48^c	66.65^a	69.97^a	58.56^c	1.99
Ash	67.49^{ab}	68.20^a	62.22^{bc}	66.59^{ab}	69.11^a	60.89^c	1.81
Dry matter	77.74^a	76.84^b	76.38^c	76.76^b	75.76^d	76.36^c	1.02

^{abcd} Means on the same row having different superscript are significantly different ($P<0.05$)

Main effect of *cassoya* inclusion and enzyme supplementation on haematological indices of finishing broiler chickens (4-6 weeks)

Haematological parameters have been used as indices of dietary effect on animals to meet their metabolic, physiological and pathological statuses. Table 7, shows the main effect of *cassoya* inclusion and enzyme supplementation on haematological indices of finishing broiler chickens. The result indicated that haemoglobin, red blood cell, white blood cell and monocytes were significantly ($p<0.05$) influenced by *cassoya* inclusion while mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration, heterophil, lymphocytes, eosinophil and basophil were not significantly affected ($p>0.05$). Haemoglobin mean values ranged from 8.51 to 10.03 g/dl, the highest mean was obtained in birds fed 50% *cassoya* inclusion followed by those on 100% *cassoya* inclusion while the least was obtained in birds fed 0%

cassoya inclusion. The values obtained for haemoglobin (8.51-10.03 g/dl) in this study correlated with the haemoglobin value of 7.66 - 10.00 g/dl obtained by (31). Birds on 50% cassoya had the highest mean haemoglobin value of 10.03 g/dl which implies greater potential and a better state of health. This was the reflection of the performance characteristics and an indication of adequate nutrition (32). Similar trend was observed in red blood cell, white blood cell and monocytes, their mean values ranged from (1.85 to 2.15) x 10^{12}/L, (5.99 to 7.06) x 10^{13}/L and (0.8 to 0.9%)respectively. Red blood cell values obtained in *cassoya* diets were higher than that of the control diet. This is in harmony with the findings of (33) that broilers fed diets containing cassava products exhibited improved health status compared to those fed maize-based diets. Results of the main effect of enzyme supplementation on haematology indices of broiler chickens showed that mean corpuscular haemoglobin, heterophil and lymphocytes recorded higher significant ($p<0.05$) mean values in birds fed enzyme supplemented diets. This shows the efficient activities of enzyme on the diet which may have directly resulted in efficient erythropoiesis. This agrees with the result of (34) that the variations in haematological indices values measured tend to confirm diet effect on blood profile of chicken. It also conforms with the report of (35) that nutrient quality affects blood values of animals. Although, red blood cell 1.8-2.28x10^{12}/L reported in this study is not in line with the report of (3.0x10^{6}/L) (36) for chicken. The observed variation could be as a result of variations in nutrient and age of birds. The observed increase in white blood cells and monocytes could be attributed to better response of experimental birds' defense mechanism to handle anti-nutritional factors and infections. Consistently, cassava is known to contain arginine which prevents bacterial and viral diseases and enhances the immune system (37).

Interaction effect of *cassoya* inclusion and enzyme supplementation on haematological indices of finishing broiler chickens (4-6 weeks)

Interactive effect of *cassoya* inclusion and enzyme supplementation on haematological indices of broiler finisher phase is shown in Table 8. Significant ($p<0.05$) differences were found on haemoglobin, red blood cell, mean corpuscular haemoglobin, mean corpuscular volume, white blood cell, heterophil, lymphocytes, eosinophil and basophil. Heamoglobin mean values ranged between 8.30 and 10.54 g/dl, highest value was obtained in birds fed 50% *cassoya* with enzyme while the least similar statistical values were obtained in other dietary treatments Birds fed enzyme supplemented diets had higher values of mean corpuscular haemoglobin; this is an indication of better oxygen carrying ability of the red blood cells and efficient respiratory function as observed by (38). Similar trend was observed for red blood cell; the values ranged between 1.80 and 2.28x10^{12}/L. Mean corpuscular haemoglobin values ranged ($p<0.05$) between 41.56 pg and 47.50 pg, higher but similar values were obtained in birds fed 100% *cassoya* inclusion with enzyme and 50% *cassoya* without enzyme, while the least value was obtained in birds fed 100% *cassoya* without enzyme. White blood cell values ranged between 5.50 and 7.32x10^{3}/L, similar statistical means were obtained in birds fed 50% *cassoya* with enzyme and 50% *cassoya* without enzyme supplementation. Those fed 0% and 100% *cassoya* with enzyme and those fed 100% *cassoya* without enzyme had similar values while least value was obtained in 0% *cassoya* without enzyme. For mean corpuscular volume, its value ranges from 113.24 fl and 114.49 fl, highest mean was recorded in birds fed 0% *cassoya* with enzyme while the least value was obtained in birds fed 100% *cassoya* without enzyme. Other dietary treatments recorded similar values ($p>0.05$). Heterophil ranged from 26.20 % to 40.80%, highest mean 40.80% was obtained in birds fed 0% *cassoya* with enzyme while the least 26.20% was recorded in birds fed 0% *cassoya* without enzyme supplementation. However, birds fed 50% *cassoya* with enzyme and 100% *cassoya* without enzyme had similar values 31.40% and 31.20%. Lymphocytes ranged ($p<0.05$) from56.40% recorded in birds fed 0% *cassoya* without enzyme to 68.60% obtained in birds fed 0% *cassoya* with enzyme. Heterophil and lymphocyte mean ranges fall within 20 – 50% and 40 – 75 % respectively as reported by (39). Eosinophil mean values ranged from 2.40 to 4.20%, highest similar values were obtained in birds fed 0% and 100% *cassoya* with enzyme and 50% *cassoya* without enzyme while the least value was obtained in birds feds 0% *cassoya* without enzyme supplementation. For monocytes, highest significant ($p<0.05$) mean of 1.20% was obtained in birds fed 0% *cassoya* with enzyme supplementation while birds fed 100% *cassoya* without enzyme and 100% *cassoya* with enzyme had the lowest mean values. White blood cell and all its differentials were significantly affected by *cassoya* and enzyme supplementation, monocytes values ranged from 0.2 to 1.2% and the values are within the normal range 0–3% reported by (39).

This implies that enzyme supplementation enables the birds to utilize the nutrients properly.

Table 7: Main effect of *cassoya* inclusion and enzyme supplementation on haematological parameters of finishing broiler chickens (4–6 weeks).

	Cassoya inclusion (%)				Enzyme supplementation		
Parameters	0	50	100	SEM	-Enzyme	+Enzyme	SEM
Packed cell volume (%)	26.60	29.20	30.11	0.74	28.27	28.87	1.14
Haemoglobin (g/dl)	8.51^{b}	10.03^{a}	9.62^{ab}	0.48	9.03	9.74	0.39
Red blood cell ($x10^{12}$/l)	1.85^{b}	2.15^{a}	2.16^{a}	0.09	2.01	2.08	0.76
Mean corpuscular heamoglobin (pg)	45.68	46.67	44.51	1.08	44.16^{b}	47.09^{a}	1.88
Mean corpuscular haemoglobin concentration (g/dl)	31.88	33.30	32.45	0.69	32.08	33.03	0.56
Mean corpuscular volume (fl)	114.36	113.99	113.72	0.24	113.81	114.24	0.11
White blood cell ($x10^{3}$/l)	5.99^{b}	7.06^{a}	6.37^{ab}	0.26	6.18	6.72	0.21
Heterophil (%)	33.50	32.80	34.20	1.45	30.53^{b}	36.47^{a}	1.19
Lymphocytes (%)	62.50	62.70	61.90	1.46	59.73^{b}	65.00^{a}	1.11
Eosinophil (%)	3.20	3.70	3.70	0.27	3.30	3.76	0.22
Monocyte (%)	0.80^{a}	0.90^{a}	0.20^{b}	0.16	0.46	0.80	0.13
Basophil (%)							

[abc] Means on the same row having different superscript are significantly ($P<0.05$) different

Table 8: Interaction effect of *cassoya* diets and enzyme supplementation on haematological parameters of finishing broiler chickens (4 - 6 weeks)

	-Enzyme (%)			+ Enzyme (%)			
Parameters	0	50	100	0	50	100	SEM
Packed Cell Volume (%)	27.00	29.80	28.00	26.20	28.60	31.80	0.68
Haemoglobin (g/dl)	8.30^{b}	9.53^{ab}	9.28^{ab}	8.72^{ab}	10.54^{a}	9.96^{ab}	0.13
Red blood Cell (x10^{12}/l)	1.80^{b}	2.02^{ab}	2.22^{ab}	1.86^{ba}	2.28^{a}	2.10^{ab}	1.52
Mean Corpuscular Haemoglobin (pg)	44.67^{b}	46.23^{a}	41.56^{c}	46.68^{a}	47.11^{a}	47.50^{a}	0.98
Mean Corpuscular haemoglobin Concentration (g/dl)	31.50	33.10	31.60	32.20	33.50	33.35	0.34
Mean Corpuscular Volume (fl)	114.23^{ab}	113.95^{ab}	113.24^{b}	114.49^{a}	114.04^{ab}	114.02^{ab}	0.36
White Blood Cell (10^{3}/l)	5.50^{b}	6.80^{a}	6.26^{ab}	6.48^{ab}	7.32^{a}	6.48^{ab}	2.05
Heterophil (%)	26.20^{c}	34.20^{b}	31.20^{bc}	40.80^{c}	31.40^{bc}	37.20^{ab}	2.07
Lympocyte (%)	56.40^{c}	63.60_{ab}	59.20_{bc}	68.60^{a}	61.80_{bc}	64.6^{a}_{b}	0.39
Eosinophil (%)	2.40^{b}	4.20^{a}	3.40^{ba}	4.09^{a}	3.20^{ba}	4.00^{a}	0.23
Monocytes (%)	0.40^{bc}	0.80^{abc}	0.20^{c}	1.20^{a}	1.00^{ab}	0.20^{c}	0.00
Basinophil (%)							

[abcdef] Means on the same row having different superscripts are significantly ($P<0.05$) differen

Main effect of *cassoya* inclusion and enzyme supplementation on serum metabolites of finishing broiler chickens (4-6 weeks)

Table 9 shows the main effects of *cassoya* inclusion and enzyme supplementation on blood serum parameters of the finishing broiler chickens. The result shows that alanine amino transferase was significantly affected ($p<0.05$) by *cassoya* inclusion; it ranged between 4.30 ui/L and 7.40 ui/L, highest mean value was obtained in 0% *cassoya* inclusion and least similar insignificant values were obtained in 50% *and* 100% *cassoya* inclusion levels. The least values obtained for alanine amino transferase in birds fed 50% *cassoya* were closer to the values (6.5 – 263 u/i) reported by (40). Variation could be linked to age of birds and environment (40). Globulin, albumin, aspartate amino transferase, cholesterol, uric acid and creatinine were not significantly affected ($p>0.05$) by *cassoya* inclusion. Aspartate amino transferase and alanine amino transferase were higher in 0% *cassoya* without enzyme while others were insignificantly reduced, indicating that there was no toxic indication within liver tissues of the birds. This study therefore revealed that the birds' livers were in good condition for nutrient metabolism, production of blood, detoxification process and processing of waste product. The effect of enzyme supplementation on serum metabolite of broiler fed diets containing *cassoya* shows that total protein, alanine amino transferase, uric acid, glucose and creatinine were significantly affected ($p<0.05$) by enzyme supplementation, birds fed diets without enzyme supplementation recorded higher ($p<0.05$) values for alanine amino transferase (7.00 ui/L), uric acid (1.51 mg/dl) and creatinine (0.41 mg/dl), while those fed diet with enzyme had higher total protein mean value of 6.27 g/dl. Cassava as a good source of arginine could be used as an efficient feed additive for enhancing the physiological performance of birds. Enzyme supplemented diet shows that birds are able to utilize the diet without any residual effect on normal muscular break down (41)

Interaction effect of *cassoya* inclusion and enzyme supplementation on serum metabolites of finishing broiler chickens (4-6 weeks)

The interaction effect of *cassoya* inclusion and enzyme supplementation on serum metabolite is presented in Table 10. *Cassoya* inclusion and enzyme supplementation significantly ($p<0.05$) affected total protein, alanine amino transferase, aspartate amino transferase, cholesterol, uric acid and creatinine. The total protein value ranged from 5.64 g/dl to 6.72 g/dl, highest value was recorded in 50% *cassoya* with enzyme supplementation. Birds fed 50% *cassoya* inclusion and enzyme supplementation had total protein value of 6.75 g/dl and it falls within the range (6.50 - 6.77 g/dl) reported by (31). The significant increase in blood plasma protein, albumin and globulin concentrations in groups fed enzyme supplemented cassoya diets might be attributed to the increase in broilers' body weights, which is in line with (42) who reported that blood plasma or serum electrophoresis showed that albumin is the major part of the total protein in the avian plasma or serum. Alanine amino transferase and aspartate amino transferase values ranged from 4.20 ui/L to 10.40 ui/L and 22.60 ui/L to 32.00 ui/L respectively, highest values were recorded for birds on 0% *cassoya* without enzyme while the least similar values were recorded in other dietary treatments. Cholesterol means ranged ($p<0.05$) between 74.80 mg/dl and 101.64 mg/dl, highest ($p<0.05$) values were obtained in birds fed 0% and 50% *cassoya* without enzyme, similar statistical values were obtained in other dietary treatments. Cholesterol values obtained 74.80101.04 mg/dl were lower to values reported by (43) 100.30-108.2 mg/dl and (31) (93.33-116.67 mg/dl) which are higher. Variation in cholesterol could be attributed to breed of chicken and nutritional pattern. Higher cholesterol value indicates which might result in heart problem while low level causes depression (44). For uric acid, mean value ranged ($p<0.05$) from 1.0 mg/dl in birds fed 0% *cassoya* with enzyme supplementation to 1.66 mg/d, 1.54 mg/dl and 1.54 mg/dl in birds fed 100 and 0% *cassoya* without enzyme and 50% *cassoya* with enzyme. Thus, uric acid values depend on protein quantity and quality in the diet. Creatinine mean values ranged ($p<0.05$) between 0.46 mg/dl and 0.34 mg/dl, highest 0.46 mg/dl and 0.42 mg/dl were obtained in birds fed 0% and 100% *cassoya* without enzyme supplementation. *Cassoya* with enzyme supplementation shows reduced significant values for uric acid and creatinine. This observation reveals reduced toxin content in the diet due to enzyme supplementation as reported by (45). Blood glucose levels obtained ranged from 215 to 268 mg/dl which agrees with the normal range of 200 to500 mg/dl reported by (46).

Conclusions and Applications

Broiler finisher chickens efficiently utilized diets with 50% cassoya replacement of maize without any detrimental effect.

Enzyme supplementation resulted in improved growth performance and consequently low cost/kg gain in broiler chickens fed cassoya as a replacement for maize

Cassoya inclusion up *to 50%* supplementated with enzyme significantly influenced serum biochemical indices.

Cassoya diet at 50% level of inclusion with enzyme supplementation in

Table 9: Main effect of *cassoya* inclusion and enzyme supplementation on serum metabolites of finishing broiler chickens (4-6 weeks)

	Cassoya inclusion (%)				Enzyme Supplementat ion		
Parameters	0	50	100	SEM	-Enzyme	+Enzyme	SEM
Total protein (g/dl)	5.75	6.22	5.71	0.10	5.68^{b}	6.27^{a}	0.05
Globulin (g/dl)	2.55	2.71	2.69	0.11	2.54	2.76	0.93
Albumin (g/dl)	3.20	3.51	3.12	0.09	3.13	3.51	0.73
ALT (u/l)	7.40^{a}	4.30^{b}	5.40^{ab}	0.63	7.00^{a}	4.53^{b}	0.51
AST (u/l)	27.90	25.60	25.40	1.64	27.00	26.80	1.34
Cholesterol (mg/dl)	90.58	87.11	77.88	4.39	90.18	80.20	3.59
Glucose(mg/dl)	274.60	263.20	267.20	4.26	268.07	268.60	3.26
Uric acid (mg/dl)	1.27	1.49	1.56	0.12	1.54^{a}	1.33^{b}	0.97
Creatinine (mg/dl)	0.38	0.35	0.40	0.02	0.41^{a}	0.35^{b}	0.19

^{ab} Means on the same row having different superscript are significantly different (P<0.05). ALT=Alanine amino transferase. AST=Aspartate amino transferase.

Table 10: Interaction effect of *cassoya* inclusion and enzyme supplementation on serum metabolites of finishing broiler chickens

	-Enzyme (%)			+Enzyme (%)			
Parameters	0	50	100	0	50	100	SEM
Total protein (g/dl)	5.64^{b}	5.71^{b}	5.68^{b}	5.86^{b}	6.72^{a}	6.23^{b}	0.13
Globulin (g/dl)	2.42	2.65	2.56	2.68	2.78	2.83	0.16
Albumin (g/dl)	3.22	3.06	3.12	3.18	3.94	3.40	0.13
ALT (u/l)	10.40^{a}	4.20^{b}	6.40^{b}	4.80^{b}	4.40^{b}	4.40^{b}	0.09
AST (u/l)	32.00^{a}	22.60^{b}	26.40^{ab}	27.4^{ab}	28.60^{ab}	24.40^{b}	2.32
Cholesterol (mg/dl)	101.64^{a}	88.02^{a}	80.88^{b}	79.52^{b}	86.20^{ab}	74.80^{b}	6.21
Glucose (g/dl)	275.80	264.80	263.60	273.40	261.60	270.80	4.96
Uric acid (mg/dl)	1.54^{a}	1.44^{ab}	1.66^{a}	1.00^{b}	1.54^{a}	1.10^{ba}	0.17
Creatinine (mg/dl)	0.42^{ab}	0.34^{b}	0.46^{a}	0.34^{b}	0.36^{b}	0.34^{b}	0.03

^{ab} Means on the same row having different superscript are significantly different (P<0.05). ALT = Alanine amino transferase. AST = Aspartate amino transferase.

broiler finisher diets is recommended for improved growth performance, nutrient digestibility and economic production.

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