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PARTIAL REPLACEMENT OF DIETARY MAIZE WITH FERMENTED CASSAVA PEEL MEAL SUPPLEMENTED WITH ENZYME COMPLEX INFLUENCES GROWING TURKEY PERFORMANCE AND BLOOD PROFILE

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Abstract: **Unsustainable maize supply and high cost necessitate investigation into alternative energy sources in turkey diets. This study examined effects of fermented cassava peel meal (FCPM) as a partial substitute for maize, on growth performance, haematology, and serum biochemistry of growing turkeys fed diets supplemented with an enzyme complex (protease, amylase, xylanase, cellulose, β-glucanase, and lipase). The basal diet contained 49% of maize. Based on it, the maize was replaced with FCPM at 0 (CON), 10% (FCPM10), 20% (FCPM20), 30% (FCPM30), or 40% (FCPM40) respectively. 120 growing British United Turkeys, 8 weeks old mixed sex, averaging 1942.62±16.6 g were randomly assigned to the five dietary treatments, 4 replicates groups of 6 turkeys each in a completely randomized design over a 28-day feeding trial. Data were analyzed using ANOVA and polynomial contrast: linear (L) and quadratic (Q). Results showed that enzyme-supplemented FCPM diets increased final weight (L.Q: P<0.05), feed intake and weight gain (L: P < 0.05). Feed:gain improved (P < 0.05) at 10, 20 and 30 FCPM groups compared with the control. The cost of feed consumed and cost/kg gain reduced (L: P<0.05) with increasing dietary FCPM. Increased levels of white blood cells (L: P < 0.05) and decreased eosinophils (L: P<0.05) were detected with higher inclusion levels (> 20%) of FCPM fed turkeys. Other haematological indices examined revealed no significant difference (L.Q: P > 0.05) across all FCPM groups. There was reduction in total protein and cholesterol (L: P < 0.05) with higher inclusion of FCPM, while other serum indices were not significant (P > 0.05). The replacement of up to 40% dietary maize supplemented with enzyme improved growth performance, reduced cost of production without adverse effect on blood indices of growing turkeys.**

IndexTerms **- fermented cassava peel meal; enzyme complex; growing turkeys; growth; blood indices.**

I. INTRODUCTION

Consumption of poultry meat has increased globally with turkey meat being the second most consumed (Baéza et al., 2022). The meat is a major source of animal protein due to its comparative high protein percentage and low percentage of fat (Marchewka et al., 2013), produced leanest meat and contains minerals such as sodium, potassium and iron (Ferreira et al., 2000). Besides, turkeys have high disease resistant capacity, high market price, low cost of feeding when compared to other poultry birds (Yasmin et al., 2021).

Maize is a major source of energy in the diet of turkey (Baéza, et al., 2022). However, the high cost and scarcity of maize is an impediment to the growth of the poultry sector (Ezeano and Ohaemesi, 2020). The shortfall of maize is due to competition among humans, industries, and livestock for the grain, and the seasonal production. Researchers in Nigeria's poultry industry are facing the challenge of reducing production costs and providing high-quality protein for human consumption by using alternatives feed resources which are cost effective (Atteh and Apata, 2024).

Cassava peel meal (CPM) is a residue left over which is available in large quantities after processing cassava root for human, industrial and export purposes. The peel is low in both energy and protein and also contains higher levels of cyanogenic glucosides (Mukhtar et al. 2023). Due to the peels' poor digestibility, feed intake was low, resulting in poor performance (Agustin et al., 2024). According to Fakir *et al.* (2012), cyanide levels as low as 25 mg/kg negatively affect layer production, egg quality, and hatchability. Similarly, levels as high as 100 mg/kg showed a negative impact on broiler performance.

II. NEED OF THE STUDY.

Several processing methods have been reported and used by different authors to overcome the limitations and improve the utilization of cassava peel for poultry feeding (Dayal *et al*., 2018). According to Ukorebi, (2022) the addition of supplemental feed additives such as enzymes, improved the nutritive value of fibrous plant based ingredients through increased nutrient digestibility in poultry. With other high-fibre feed sources, supplementing enzymes such as Fullyzme helps turkeys to degrade fibre and reduce the harmful effects of the antinutritional components **(**Aguihe et al., 2020). Little is known of the effect of this exogenous enzyme on the use of FCPM as a dietary ingredient, the subject of this study.

III. **RESEARCH METHODOLOGY**

3.1 Experimental site

The research was carried out at the poultry unit of the Teaching and Research Farm, Kwara State University, Malete, Nigeria, which is located in the Southern Guinea Savanna Vegetation Ecological zone on longitude 11^0 71''N and latitude 40^0'44''E 671m above sea level.

3.2 Preparation of fermented cassava peel meal

Fresh cassava peels were collected from cassava *(garri)* processing factory in Malete. The peels were washed thoroughly with clean water. The peels were dried in a hot air oven at 55℃, milled, and subjected to solid state fermentation (SSF) process. The fungus used for fermentation was *Aspergillus. niger* MCB-003 and the stock were obtained from the Microbiology Laboratory, Kwara State University, Malete.

In preparing suspension, *A. niger* MCB-003 was cultivated on Potato-Dextrose Agar (PDA) at 28℃ for 7 days, after which it was sub-cultured to obtain a pure suspension containing 5×10^4 spore ml^{-1} following the procedure outlined by Hesseltine (1983). A batch (1 kg) of the cassava peel meal (CPM) was autoclaved at 121^0C for 15 min. Autoclaved meal was cooled to room temperature for 1 h, hydrated with distilled water at a ratio of 1:1.5 (w/v), and inoculated with 20ml of spore suspension of *A. niger* per kg of wet meal. The meal was thoroughly mixed and fermented in an air-tight plastic bucket for 7 days. At the end of the fermentation period, *A. niger* fermented CPM was covered by a thin layer of black mycelia which knitted the substrate into a cake. The fermented cassava peel meal (FCPM) was then re-autoclaved at 121^0C for 15min to destroy *A. niger* and oven dried at 60 ^{0}C for 4 h. The FCPM dried product was kept in bags and stored at ambient temperature until required for feeding trial and chemical analysis.

3.3 Turkeys, diets and management

One hundred and twenty (120) 8- week old grower turkeys of average weight of 1942.62±16.6 g were obtained from a local commercial farm. The basal diet contained 49% of maize. Based on it, the maize was replaced with FCPM at 0%, 10%, 20%, 30%, or 40%, which were referred to as CONT, FCPM10, FCPM20, FCPM30 or FCPM40, respectively, to formulate 5 experimental diets (Table 1).

Diets (FCPM10, FCPM20, FCPM30 and FCPM40) were supplemented with Fullzyme® at 0.05%. The enzyme preparation derived from yeast culture and *Bacillus subtilis*, contain amylase, protease, cellulase, lipase, pectinase, xylanase, β-mannanase, β-glucanase, and phytase manufactured by Biofeed® Technology Inc., Brossard QC, Canada. The level of enzyme supplementation was chosen according to the manufacturer's recommendations. The diets were fed *ad libitum* throughout the 28-day period of study. Routine management was implemented. During the period of study, all the turkeys in each pen were weighed weekly, and the results were divided by the total number of birds. Weekly records were used to calculate the cumulative mean value of feed intake. The feed to gain ratio was computed by dividing the amount of feed consumed per bird by the total weight gain during the period of the experiment. Cost–benefit analysis was based on the prevailing market prices of feed ingredients. Feed: gain x cost of feed per/kg gave the cost/kg gain.

3.4 Analysis of fermented cassava peel meal

The proximate composition of CPM and FCPM samples were determined by AOAC (2016) methods. Cyanide contents of the samples were determined according to the method of Surleva et *al*., (2013).

3.5 Blood samples collection and analysis

At the end of the experimental period, two turkeys were randomly selected from each replicate and slaughtered by decapitation. Blood samples were collected into bottles containing ethylene diamine and tetraacetic acid (EDTA) anticoagulant for haematology and clean dry tubes for serum constituent analysis. Serum samples were separated by centrifugation (1600 x g for 15 min) and stored at -4 ^{0}C. The blood was analyzed for hemoglobin (Hb) concentration, red blood cells (RBC), pack cell volume (PCV), platelet (PLT), white blood cell count (WBC), white cell differential count, the mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) using a DYMIND AUTO Haematology Analyser (Shenzhen Dymind Biotechnology Co., Ltd., China) in accordance with the manufacturer's instructions. Serum total protein (TP), albumen (Alb), globulin (Glo), cholesterol (CHO), triglyceride (TRIG), high density lipoprotein (LDL), low density lipoprotein (LDL), aspartate aminotransferase (AST, EC 2.6.1.1), alanine aminotransferase (ALT, EC 2.6.1.2) and alkaline phosphatase (ALP, EC 3.1.3.1) activities were analysed using an automated BIOELAB AS-160 (Sancare Biomedical Ltd, Hong Kong, China) according to the manufacturer's instructions.

3.6 Statistical Analysis

Data were subjected to one-way analysis of variance (ANOVA), multiple comparisons among means were made by Duncan's multiple range test, and polynomial contrast (linear, quadratic ratio) was applied to determine the effect of FCPM with enzyme supplementation on the parameters determined using SPSS program (version 21) (IBM). Other data were compared using Student's t-test. All statement of differences were based on significance at P <0.05.

IV. RESULTS AND DISCUSSION

4.1 Results

Table 4.1: Formulation (%) of the experimental diets

	Diets				
	CON	**FCPM10**	**FCPM20**	**FCPM30**	**FCPM40**
Ingredients					
Maize	49.00	44.10	39.20	34.30	29.40
Fermented cassava peel meal	0	4.90	9.80	14.70	19.60
Wheat offal	11.00	10.75	10.45	9.97	9.85
Soybean meal	8.20	8.10	8.10	8.20	8.40
Palm kernel cake	12.80	12.60	12.20	11.90	11.05
Groundnut cake	10.30	10.50	10.90	10.90	11.10
Fish meal	2.20	2.40	2.70	2.90	3.10
Soya bean oil	1.50	1.60	1.80	1.90	2.45
Bone meal	4.00	4.00	4.00	4.00	4.00
Methionine	0.25	0.25	0.25	0.25	0.25
Lysine	0.25	0.25	0.25	0.25	0.25
Premix[a]	0.25	0.25	0.25	0.25	0.25
Fullyzme®	0	0.05	0.05	0.05	0.05
Salt	0.25	0.25	0.25	0.25	0.25
Nutrient content (%)[b]					
Crude protein	18.40	17.99	18.00	17.96	17.92
Crude fibre	4.53	5.15	5.75	6.38	6.94
Metabolizable energy (MJ/kg)	13.58	13.38	13.23	12.96	12.74

[a]: Provided: vitamin A (all trans-retinol acetate) – 15 000 IU, vitamin D3 (cholecalciferol) – 5 000 IU, vitamin E (all-rac-α-tocopheryl acetate) – 100 mg, vitamin K3 – 4 mg, vitamin B1 – 5 mg, vitamin B2 – 15 mg, vitamin B6 – 6 mg, niacin – 100 mg, biotin – 0.35 mg, pantothenic acid – 32 mg, nicotinic acid – 100 mg, folic acid – 4 mg, choline chloride – 700 mg, Mn – 100 mg, Zn – 80 mg, Fe – 60 mg, Cu – 20 mg, I – 1.5 mg, Se – 0.3 mg, Ca – 1.07 g. [b]: Determined values except metabolizable energy, which was calculated from the published NRC (2024) compositions of the ingredients used

Table 4.2: Proximate composition (%) and cyanide component (mg/kg) of cassava peel meal (CPM) and fermented cassava peel meal (FCPM)

	CPM	**FCPM**
Proximate composition (%)		
Dry matter	92.8	90.16
Crude protein	5.40[b]	7.53[a]
Crude fibre	15.01[a]	7.48[b]
Ether extract	2.10	1.84
Ash	5.92	6.68
Nitrogen free extractives	64.37[b]	66.63[a]
Antinutrient		
Cyanide (mg/kg)	0.71[a]	0.48[b]

[a,b]Means in the same row with different letters are significantly different at $P < 0.05$

Table 4.3: Partial replacement of maize with fermented cassava peel meal (FCPM) on growth performance and cost benefit of growing turkeys.

Variable	Levels of FCPM replacement for maize						Orthogonal contrast	
	CON	FCPM10	FCPM20	FCPM30	FCPM40	SEM	Linear	Quadratic
Initial weight (g per turkey)	1,940.54	1946.12	1952.10	1938.90	1935.50	2.23	NS	NS
Final weight (g per turkey)	4,521.40^{b}	4767.68^{a}	4710.66^{a}	4761.86^{a}	4825.66^{a}	27.23	0.001	0.035
Feed intake (g per turkey day-^{1})	224.18^{c}	231.37^{bc}	225.07^{c}	236.62^{ab}	245.95^{a}	2.32	0.001	NS
Weight gain (g per turkey day-^{1})	92.17^{b}	100.77^{a}	98.52 ^{a}	100.82^{a}	103.22^{a}	1.05	<0.001	NS
Feed : gain (g/g)	2.42^{a}	2.29^{b}	2.28^{b}	2.35^{b}	2.38^{a}	0.01	NS	NS
Cost of feed (₦/kg)	589.48^{a}	557.54^{b}	530.75 ^{c}	499.32^{d}	484.26 ^{e}	8.85	<0.001	0.022
Cost of feed consumed (₦)	3612.75^{a}	3344.78^{b}	3310.49^{b}	3336.41^{b}	3335.92^{b}	25.96	<0.001	NS
Cost/kg gain (₦/bird)	1426.54 ^{a}	1276.99 ^{b}	1210.11^{c}	1173.40^{d}	1137.96^{e}	23.36	<0.001	NS

Values are means of four replicates per treatment. Means in the same row with different superscripts are significantly ($P < 0.05$) different; SEM = Standard Error of Means, NS = Not Significant.
₦1,200 = $1

Table 4.4: Effect of fermented cassava peel meal on haematological values of growing turkey

Variable	Levels of FCPM replacement for maize						Orthogonal constrast	
	CON	FCPM10	FCPM20	FCPM30	FCPM40	SEM	Linear	Quadratic
Packed cell volume (%)	30.01	31.80	32.40	32.47	32.80	0.52	NS	NS
Haemoglobin (gdl)	9.72	9.96	9.41	9.60	10.01	0.49	NS	NS
Red blood cells (x10^6µl)	2.53	2.56	2.64	2.58	2.72	0.023	NS	NS
White blood cells ($x10^6$µl)	15.11^{b}	15.70^{b}	15.02^{b}	19.05^{a}	19.72^{a}	0.66	<0.004	NS
Platelet ($x10^3$µl)	121.32	122.57	115.30	117.24	119.12	1.45	NS	NS
Lymphocytes (%)	65.39	65.26	68.69	70.20	69.41	1.47	NS	NS
Heterophils (%)	25.54	26.42	25.67	28.20	28.83	1.05	NS	NS
Monocytes (%)	2.88	2.98	2.82	3.20	3.01	0.07	NS	NS
Eosinophils (%)	3.71^{a}	3.67^{a}	3.41^{a}	2.33^{b}	0.21^{b}	0.21	0.001	NS
Basophils (%)	0.29	0.28	0.31	0.33	0.34	0.01	NS	NS
Mean corpuscular volume (fl)	120.39	124.02	122.96	120.58	122.68	0.63	NS	NS
Mean corpuscular haemoglobin concentration (%)	31.33	31.36	29.67	29.51	30.59	0.45	NS	NS
Mean haemoglobin concentration (pg)	38.38	38.88	35.52	37.12	37.21	0.65	NS	NS

Values are means of four replicates per treatment. Means in the same row with different superscripts are significantly (P<0.05) different; SEM = Standard Error of Means, NS = Not Significant.

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Table 5: Effect of fermented cassava peel meal on serum biochemical constituents of growing turkeys

Variable	Levels of CPM replacement for maize						Orthogonal constrast	
	CON	FCPM10	FCPM20	FCPM30	FCPM40	SEM	Linear	Quadratic
Total protein (g/dl)	5.45[a]	5.22[abc]	5.07[ab]	4.34[c]	4.56[bc]	0.14	0.003	NS
Albumin (g/dl)	2.00	2.14	2.08	1.64	1.97	0.08	NS	NS
Globulin (g/dl)	2.99	3.08	2.99	2.70	2.59	0.10	NS	NS
Aspartate aminotransferase (µl)	255.84	258.17	264.19	265.79	263.79	1.58	NS	NS
Alanine aminotransferase (µl)	26.95	27.06	29.69	30.44	31.22	0.91	NS	NS
Alkaline phosphatase (µl)	358.15	360.55	363.39	367.68	366.75	7.63	NS	NS
Glucose (mg/dl)	367.97	366.55	370.43	363.36	359.95	1.54	NS	NS
Cholesterol (mg/dl)	206.81[a]	200.67[ab]	201.42[ab]	190.24[bc]	187.25[c]	2.90	0.003	NS
Triglyceride (mg/dl)	171.33	171.03	171.35	168.98	167.98	1.75	NS	NS
High density lipoprotein (mg/dl)	144.46	146.53	145.19	137.24	139.37	1.90	NS	NS
Low density lipoprotein (g/dl)	60.09	60.37	56.35	50.99	48.31	2.09	0.033	NS

Values are means of four replicates per treatment. Means in the same row with different superscripts are significantly ($P < 0.05$) different; SEM = Standard Error of Means, NS = Not Significant.

4.1.1. Analytical composition of fermented cassava peel meal

The proximate composition and cyanide component of cassava peel meal (CPM) and fermented cassava peel meal (FCPM) are shown in Table 2. Following 7 days of fermentation, crude protein and nitrogen-free extractives increased ($P < 0.05$) by 39.39% and 3.51%, respectively, while crude fibre and cyanide decreased ($P < 0.05$) by 50.16% and 32.39%, respectively.

4.1.2 Growth Performance

The growth performance and cost benefit of growing turkeys were affected by FCPM-based diets supplemented with the exogenous enzyme at the end of the feeding trial (Table 3). The final weight increased while the cost of feed per kilogram decreased with increase dietary FCPM (L.Q: $P < 0.05$). Average daily feed intake, average daily weight gain increased while cost of feed consumed per bird and cost/kg gain reduced (L: $P < 0.05$) with increasing FCPM in diets. There was significant ($p < 0.05$) improvement in feed:gain with 10, 20, and 30% FCPM diets compared with the control.

4.1.3. Haematology

The haematological values of growing turkeys fed FCPM based-diets are presented in Table 4. For all treatments, packed cell volume, haemoglogbin, red blood cells, platelet, lymphocytes, heterophils, monocytes, basophils, mean corpuscular volume haemoglobin concentration and mean haemoglogbin concentration measured were not affected ($P > 0.05$) by inclusion of FCPM. However, white blood cells increased while eosinophils decreased (L: $P < 0.05$) with higher inclusion levels (> 20%) of FCPM.

4.1.4 Serum Biochemistry

The serum biochemical values of growing turkeys fed FCPM-based diets are presented in Table 5. The albumin, globulin, alanine aminotransferase, alkaline phosphatase, aspartate aminotransferase, glucose, triglyceride and high density lipoprotein were not affected ($P > 0.05$) by inclusion of FCPM in diets. However, total protein, cholesterol and Low density lipoprotein reduced (L: $P < 0.05$) at higher levels of FCPM inclusion (> 20%).

4. 2 Discussion

The fermentation of cassava peel meal resulted into increase in crude protein and decrease in crude fibre. According to Okhonlaye and Foluke (2016), Apata and Atteh, (2016), *Aspergillus niger* produced extra cellular enzymes into the fermenting medium with the growth and proliferation of the fungi in the form of single cell protein which could be responsible for the observed increase in crude protein content of FCPM. Similar findings were reported by Ikpesu *et al*. (2015) that an increase in protein content of cassava peel fermented with *A. niger* resulted in enrichment of cassava products through fermentation. The decreased in crude fibre content observed in this experiment might be due to the ability of *A. niger* to degrade the crude fibre by converting them to volatile fatty acids, methane, and produce microbial biomass (Ikpesu *et al.*, 2015). Similarly, *A. niger* MCB-003 fermentation of CPM decreased cyanide content by 32.4% cyanide. This is in agreement with Adegoke (2018) who reported significant reduction in cyanide level through solid state fermentation of cassava products. In this study, *A. niger* degradation of cyanogenic glycosides in the CPM could be ascribed to the hydrolysis by linamarase and evaporation of cyanide during drying (Olutosin and Kayode, 2021).

The increase in daily feed intake and daily weight gain with higher levels of FCPM fed to growing turkeys is probably related to the addition of enzyme which improved the feed intake and absorption of nutrients. This is in agreement with the reports of Anigbogu *et al.* (2024) and Unigwe *et al.*, (2020) in which supplementation with exogenous enzymes in the diets improved performance of turkeys and piglets, respectively. Similar observation was reported by Oguntoye *et al.*, (2018) with enhanced growth performance in broiler chickens when yam peel based-diets was supplemented with cocktail enzyme. The observed increase in feed intake at higher dietary inclusion level of FCPM could be consequent upon the fact that poultry eat to satisfy their energy requirements (NRC, 2024).

The improvement in feed:gain ratio observed at FCPM inclusion up to 30% replacement of maize could be contingent upon improved digestibility of dietary fibre and reduction in cyanide effect of CPM used in turkey feed. Since cost of feed in poultry accounts for 60 to 70% of the total cost of production (Banson *et al.*, 2015) the reduced cost of feed as FCPM inclusion level increases would lead to higher profitability in turkey production. By implication, farmers could save cost on every kg of feed prepared with FCPM and reduction in cost per kilogram gain of growing turkeys. This finding is similar to the results of Anigbogu *et al*. (2024) who revealed that up to 15% saw dust and pam kernel cake mixture supplemented with exogenous enzymes in turkeys reduced cost of feed production.

Hematological values are good indicators of the physiological status of animals, effect of environmental, nutritional and/or pathological stresses (Marchewka *et al*, 2013; Daniel-Igwe. and Okwara, 2017). In this study, increased white blood cells (WBC) at higher inclusion levels of FCPM could partly be ascribed to physiological adjustment to the residual effect of cyanide. According to Oduguwa, (2006), higher levels of white blood cells have been produced in farm animals in a bid to fight against the foreign bodies. The observed WBC in this study was however within the normal reference values for healthy turkey ($9 – 31 \times 10^3/\mu l$) as reported by Adeyeye *et al.* (2022). Other hematological values are also within the normal range of values (Mitruka and Rawnsley, 1977).

The values of the serum biochemical indices in turkeys fed FCPM demonstrated reduction in cholesterol, low density lipoprotein and total protein at higher levels of inclusion above 20% replacement of maize. This could be of significance to consumers as reduction in LDL and cholesterol leads to consumption of healthy foods. The total protein in this study is similar to Bounous *et al* (2000) who reported the range of 3.6 -5.5g/dl in healthy turkey. This suggest that the protein in the diet was efficiently utilized by the growing turkeys. The values of other serum indices were within the normal reference ranges for turkeys (Mitruka and Rawnsley, 1977), reflecting a normal metabolism in the growing turkeys fed up to 40% FCPM diets. This observation could also suggest that FCPM can spare dietary maize at these substitution levels.

V. CONCLUSION

In conclusion, the present study showed that the solid state fermentation of CPM using *A. niger* MCB – 003 could improve nutritional characteristics of cassava peel meal and decrease the inherent cyanide. The FCPM has a high potential as feedstuff for turkey and could replace up to 40% maize in diets supplemented with enzyme which improved growth performance and reduced cost of production without adverse effect on blood parameters of growing turkeys.

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