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Rabbits fed fermented cassava starch residue II: Enzyme supplementation influence on performance and health status

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SUMMARY

The probability of increasing the optimum replacement level of maize with microbially fermented cassava starch residue (MFCSR) with multi-enzyme supplementation in rabbit diets was evaluated in a 2x3 factorial arrangement in eight weeks-trial. While the total weight gain (TWG) and feed conversion ratio (FCR) significantly (P<0.05) improved by 2.37% and 2.04%, respectively, in enzyme supplemented diets, the MFCSR inclusion level (0-100%) led to 18.96-25.24% and 21.76-26.61% decrease in the rabbit TWG and FCR, respectively. The slaughtering weight, skin, head and limb, foreleg, loin and abdominal wall, neck, lung, kidneys heart and bile were significantly (P<0.05) affected by dietary levels of MFCSR. Enzyme supplementation did not significantly (P>0.05) influence the haematological indices measured, but the packed cell volume (PCV), red blood cells (RBC), haemoglobin concentration (HBC), monocytes, granulocytes and platelets were by the levels of MFCSR in the diets. Enzyme supplementation led to decrease in alanine aminotransferase (ALT) while the total serum protein, globulin, cholesterol, bilirubin, aspartate aminotransferase (AST) and glucose though significantly (P<0.05) affected did not follow a particular trend. Using performance indices as response criteria, the addition of multi-enzyme at 0.35g/kg led to the complete replacement of maize in the diets of growing rabbit with MFCSR.

Additional keywords

Cassava sievate.
Health implication.
Agro industrial waste.
Growth promoter.
Monogastric herbivores.

Les lapins nourris avec de l'amidon de manioc fermenté résidus II: enzyme supplementation

RÉSUMÉ

La probabilité d'augmenter le niveau de remplacement optimale du maïs avec de l'amidon de manioc fermenté d'origine microbienne (résidus MFCSR) avec multi-enzyme supplementation en régimes de lapin a été évalué dans un arrangement factoriel 2x3 à huit semaines au procès. Bien que le gain de poids total (GTT) et le ratio de conversion d'alimentation (FCR) a significativement (P<0,05) s'est amélioré de 2,37 % et 2,04 % dans des suppléments alimentaires, l'enzyme MFCSR niveau d'inclusion (0-100 %) conduit à 18.96-25.24 % et 61 %.21.76-26 diminution du GTT de lapin et le FCR respectivement. Le poids d'abattage, de la peau, de la tête et un membre, patte antérieure, la longe et de l'abdomen, du cou, du poumon, des reins et du cœur bile étaient significativement (P<0,05) affectés par les niveaux de MFCSR. La supplémentation enzymatique n'a pas significativement (P> 0,05) a influencé les indices hématologiques mesuré, mais l'hématocrite (PCV), les globules rouges (RBC), du taux d'hémoglobine (HBC), les monocytes, les granulocytes et les plaquettes ont été par les niveaux de MFCSR dans l'alimentation. La supplémentation enzymatique a conduit à diminuer dans l'alanine aminotransférase (ALT) alors que le nombre total de protéines sériques, globuline, cholestérol, bilirubine, aspertate aminotransférase (AST) et le glucose bien que significativement (P< 0,05) n'a pas suivi une tendance particulière. À l'aide d'indices de performance comme critères d'intervention, l'ajout de multi-enzyme à 0,35g/kg a entraîné le remplacement complet du maïs dans l'alimentation des lapins en croissance avec MFCSR.

Mots clés supplémentaires

Sievate le manioc.
Implication de la santé, de l'
Agro les déchets industriels.
Promoteur de croissance.
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INTRODUCTION

Rearing of rabbits has been identified as one of the solutions to the problem of dietary protein intake prevalent in Nigeria and most developing countries (Oloruntola et al., 2015). However, the high cost of rabbits due to the exorbitant price of the conventional feedstuffs for rabbits' feed production makes rabbit's meat unaffordable for the consumers. Therefore, the need for the use of alternatives to the popular conventional feedstuffs in rabbit's feed preparation becomes important (Oloruntola et al., 2015; Adeyeye et al., 2018).

Manihot spp. and its agro-industrial by-products like starch resides and peels when incorporated in monogastric diets could be a viable means of reducing feed cost, which could lead to a reduction in animal protein production cost with a concomitant increase in animal protein affordability by the resource-poor in developing countries (Olafadehan, 2011). However, the major drawback to the use of Manihot root harvest is its high content of cyanogenic glucosides which releases hydrocyanic acid on hydrolysis. Also, it has been reported that the starch residue which is a waste product of Manihot processing is very high in crude fibre (Aro et al. 2010) which further limits its use in monogastric animal feeding. The proximate composition (crude protein: 6.86%; lipid: 1.88%; fibre: 0.60%, carbohydrate: 87.53%) of cassava peels was reported by Nwoko et al. (2016).

Previous animal feeding studies revealed that when processed cassava wastes were included above certain levels in diets of monogastrics and pseudo-ruminants, some negative effects on their performance and health status were precipitated (Oluremi and Nwosu, 2002, Osakwe and Nwose, 2008 and Oloruntola et al., 2016). Therefore, processing methods such as sun-drying, soaking, fermentation and enzyme supplementation among others are advocated to enhance the utilization of cassava agro-industrial by-products in monogastric nutrition. Specifically, positive results which varied from improved digestibility, feed conversion ratio to reduced mortality have been reported when Manihot waste such as peels and starch residues supplemented with enzymes were fed to rabbit (Fernandez et al., 1996; Bolis et al., 1996 and Eiben et al., 2004).

Earlier reports by us showed that microbially (rumen liquor) fermented cassava starch residue (MFCSR) could be used to replace maize up to 50% in growing rabbit's diets, without the negative effect on their growth performance and health status (Oloruntola et al., 2018). It is conceivable that this optimum replacement value of MFCSR can be increased when fibre degrading multi-enzyme are supplemented in the diets. A recent report by Ayodele et al. (2016) showed that a multi-enzyme approach could be a viable way

Table I. Gross and analysed composition of experimental diets (g100g^{-1}) (Gross et analysé la composition des régimes expérimentaux (g100g^{-1}))

	Percentage Level of MFCSR Inclusion					
	Diet 1 0+E	Diet 2 0-E	Diet 3 75+E	Diet 4 75-E	Diet 5 100+E	Diet 6 100-E
Ingredients						
Maize	43.00	43.00	10.75	10.75	00.00	00.00
MFCSR	00.00	00.00	32.25	32.25	43.00	43.00
Maize husk	22.40	22.40	22.40	22.40	22.40	22.40
Wheat offal	8.00	8.00	8.00	8.00	8.00	8.00
BDG	10.00	10.00	10.00	10.00	10.00	10.00
Soya bean meal	14.85	14.85	14.85	14.85	14.85	14.85
Bone meal	1.00	1.00	1.00	1.00	1.00	1.00
Methionine	0.15	0.15	0.15	0.15	0.15	0.15
Lysine	0.10	0.10	0.10	0.10	0.10	0.10
Premix	0.25	0.25	0.25	0.25	0.25	0.25
Salt	0.25	0.25	0.25	0.25	0.25	0.25
Total	100.00	100.00	100.00	100.00	100.00	100.00
Calculated analysis						
Lysine (%)	0.76	0.76	0.68	0.68	0.65	0.65
Methionine (%)	0.39	0.39	0.33	0.33	0.31	0.31
Calcium (%)	0.43	0.43	0.42	0.42	0.42	0.42
Avail. Phosphorus (%)	0.38	0.38	0.36	0.36	0.35	0.35
*ME (kcal/kg)	2965.04	2965.04	2961.17	2961.17	2959.88	2959.88
Analyzed composition						
Dry matter	92.78	92.79	94.12	94.13	94.55	94.55
Crude protein	15.48	15.46	16.36	16.39	16.45	16.42
Crude fibre	11.35	11.35	13.03	13.03	13.68	13.68
Ether extract	2.12	2.12	2.04	2.04	2.02	2.02
Ash	6.84	6.84	6.33	6.33	6.13	6.13
Nitrogen-free extract	56.99	57.02	56.36	56.34	56.27	56.3

BDG: Brewer dried grain; MFCSR: microbially fermented cassava starch residue.
*ME: metabolizable energy= (37x%CP) + (81.8x%FAT) + (35.5x%NFE) (Pauzenga, 1985).
E= Enzyme which was supplemented at 0.35 g/kg

to fully capture the nutritive value of feed high in fibre. Achieving this will help to increase the use of cassava starch residue, which constitutes about 10% cassava root harvest waste in rabbit production with a resultant increase in rabbit meat affordability among the resource poor. This is envisaged will enhance food security in regions such as South America, Asia and Africa where hunger and starvation are endemic and *Manihot spp*. waste is largely available but unutilized in animal feeding. The objective of this feeding trial was to study the possibility of increasing the optimum level of maize with MFCSR by multi-enzyme supplementation in rabbit diets.

MATERIALS AND METHODS

This study was carried out upon approval by the Research Committee of the Department of Animal Production and Health, The Federal University of Technology, Akure, Nigeria.

Experimental animals' arrangement and management

In part I of this study, the optimal replacement level of maize with MFCSR was observed to be 50% (Oloruntola et al., 2018). The possibility to further increase this level with multi-enzyme supplementation was evaluated using 180 healthy, 5-weeks old growing rabbits of crossbreed (New Zealand x Chinchilla) and mixed sexes. The rabbits were randomly allotted to the six dietary treatments (10 replicates/treatment, 3 rabbits/replicate). The experimental design was a completely randomized one in 2x3 factorial arrangement comprising two (2) enzyme levels (0 and 0.35 g/kg) and three (3) MFCSR inclusion levels (0, 75 and 100%).

Test ingredients and experimental diets

a commercial enzyme (Bioenzyme PH) manufactured by Biomix S.A, Carrera 47C, Sabaneta-Colombia was procured from a reputable vendor in Nigeria. The enzyme contains cellulose (700,000U.A), α-amylase (800,000U.A), β-glucanase (300,000U.BG), phytase (1,200F.T.U), protease (800,000 UP), lipase (20, 000 UI) and xylanase (500,000U.X).

Cassava starch residue was collected from cassava starch processing industry into sacks, pressed with the hydraulic press to drain out the water, sun-dried and milled. Layer's droppings were obtained from poultry in the Teaching and Research Farm of the Agricultural Technology Department of the Federal Polytechnic, Ado Ekiti, sundried, autoclaved to destroy any microbial growth, re-sundried and milled. Rumen liquor was collected from freshly slaughtered cattle at government abattoir in Ado Ekiti, Nigeria and immediately squeezed out of the rumen content using a muslin cloth. The ground cassava starch residue was mixed with the autoclaved ground dried layer droppings at the rate of 100 g kg^{-1} of CSR in the black polythene bag, sprayed with rumen liquor at the rate of 250ml and fermented anaerobically for a duration of 7 days. The fermented CSR was sundried for 4 days (Oloruntola et al., 2017. Six experimental diets were formulated and designated as diets 1 (0-E), diet 2(0+E), diet 3(75-E), diet 4(75+E), diet 5(100-E) and diet 6(100+E). Diet 1 and 2 served as negative and positive control respectively. Diets 1, 3 and 5 were not supplemented with the enzyme (E) but had their maize replaced with MFCSR at 0, 75 and 100%; while diets 2, 4 and 6 had their maize replaced with MFCSR at 0, 75 and 100% respectively and supplemented with an enzyme. Thereafter, the diets were pelletized (4mm diameter and 8 mm long). The gross composition and analysed composition of the diets are as presented in **Table I**.

Response criteria

The weekly weight gain, feed consumption, feed conversion ratio, haematological and serum biochemical indices were obtained as described in the paper I (Oloruntola et al., 2018) and by Oloruntola et al. (2016). In brief, the weight gain was calculated as the difference between the initial and final weight while the feed consumption was the difference between the feed given and feed left over. The feed conversion ratio was the ratio of total feed consumed to the total weight gain. Ten rabbits were randomly selected from each treatment group of thirty rabbits at the end of the 8 weeks feeding trial. These rabbits were fasted overnight, slaughtered according to the guidelines of the World Rabbit Science Association (WRSA; Blasco and Ouhayoun, 1996) and skinned. Pelts, rabbit's heads and limbs were removed and weighed. Their internal organs namely; lungs, liver, kidney, heart, pancreas, bile and gastrointestinal tract were removed and weighed. Thereafter, dressed weights were determined and used to calculate the dressed percentage for the rabbits, while the gastrointestinal tract and other internal organs were expressed as a percentage of slaughtered weight as previously described in the paper I of this study.

Blood samples were collected into a blue top tube containing potassium ethylene diamine tetraacetic acid (K-EDTA) and into plain purple top tubes. The blood in EDTA containing bottles were analysed for packed cell volume, red blood cells, white blood cells, haemoglobin concentration, mean cell volume, mean cell haemoglobin, mean cell haemoglobin concentration, platelets, lymphocytes, monocytes and granulocytes using Shenzhen Mind ray Auto Haematology Analyzer, Model Bc-3200 (Shenzhen Mind ray Biomedical Electronics Co. Hamburg 20537, Germany). The sera which were separated from plain purpled top bottle blood samples were analysed for cholesterol, urea, creatinine, bilirubin, aspartate amino transaminase (AST), alanine aminotransaminase (ALT), alkaline phosphatase (ALP), amylase, glucose, total protein, albumin and globulin with a Reflectron ® Plus 8C79 (Roche Diagnostic, GmbH Mannheim, Germany), using commercial kits.

Statistical Analysis

Proximate composition of MFCSR, maize, wheat offals, maize husk, brewers dried grain (BDG) and soybean meal and experimental diets were determined as described by AOAC methods (AOAC, 1995). The data collected were analysed using General Linear Model procedure of SPSS version 20 (SPSS 2011) for complete randomized design with 2x3 factorial arrangements. The data were tested for the main effects (Enzyme and MFCSR inclusion) and two-way interactions. The significance was assessed at P<0.05.

RESULTS

Table II shows that the weight changes and the feed conversion ratio (FCR) of the rabbits significantly (P<0.05) increased by the multi-enzyme supplementation in the diets but decreased (P<0.05) with increased inclusion level of MFCSR from 0 through 75 to 100%. The enzyme x MFCSR interaction was not significant (P>0.05).

Table III reveals that the effect of the multi-enzyme supplementation was not significant (P>0.05) for the carcass traits measured. Slaughter weight (SW) of rabbits on the control (1372.00g/rabbit) and those on 75% MFCSR-based diet (1385.75g/rabbit) were similar but significantly (P<0.05) higher than those fed on 100% MFCSR-based diet (1190.25g/rabbit). Also, the relative weights of the skin, head and limb, foreleg and loin and abdominal wall of rabbits fed on the control and 75% MFCSR-based diets were similar but consistently significantly (P<0.05) lower than those fed 100% MFCSR-based diets. The relative weight of the neck of rabbits fed the control diet (3.00%) was significantly (P<0.05) higher than those fed 75 and 100% MFCSR-based diets (2.13 and 2.28 % respectively). **Table IV** shows that the internal organs (% slaughter weight) of the rabbits were only significantly (P<0.01, P<0.05) affected by the levels of MFCSR in the diets. The relative weight of lung, kidney, heart, pancreas and bile of rabbits fed the control and 75% MFCSR-based diets were similar, but significantly (P<0.01, P<0.05, respectively) lower than those fed 100% MFCSR-based diets.

Of the haematological indices measured, only the packed cell volume (PCV), red blood cells (RBC), haemoglobin concentration (HBC), platelets, monocytes and granulocytes were significantly (P<0.05) affected by the levels of MFCSR in the diets with rabbits fed on 75% MFCSR-based diets in most cases having the lowest values of these indices **(Table V)**. The interaction between enzyme and MFCSR inclusion level was only significant (P<0.05) for mean cell haemoglobin (MCH) and platelets. **Table VI** shows that while only the ALT was significantly (P<0.05) affected by the enzyme supplementation, the total protein (TP), globulin (GLO), cholesterol (CHO), bilirubin (BIL), alanine aminotransaminase (ALT) and glucose (GLU) were significantly (P<0.01, P<0.05) affected with the level of replacement of maize with MFCSR in the diets. Of these, rabbits fed 75% MFCSR-based diets consistently had the highest values of TP, GLO, CHO and ALT.

DISCUSSION

This study was conducted to confirm whether a multi-enzyme supplementation would help to increase the earlier reported 50% optimum replacement value of MFCSR for maize in the diets of growing rabbit to a higher level. In this study, while the weight gain of the rabbits decreased by 18.96-25.24%, the FCR decreased by 21.76-26.61% as the MFCSR without enzyme supplementation in the diets increased from 0% to 75% and 100%, thus further confirming that beyond 50% replacement level of maize with MFCSR the weight gain vis-à-vis feed utilization by the rabbit will decline. The plausible reason for this observed decline in the weight changes and FCR could be as a result of the cumulative effect of the residual HCN in MFCSR as earlier reported (Oloruntola et al., 2016). In cyanide poisoning, cya-

Table II. Performance of weaner rabbits fed graded levels of microbially fermented cassava starch residue supplemented with multi-enzyme (Performances des porcelets sevrés lapins recevant des niveaux gradués de résidus d'amidon de manioc fermenté par voie microbienne complétées avec multi-enzyme)

	ILW	FLW	TWG	DWG	TFI	DFI	FCR
Enzyme (g)							
0.00	549.39	1532.03^{b}	985.89^{b}	17.61^{b}	3803.13	67.91	3.92^{b}
0.35	548.03	1557.81^{a}	1009.78^{a}	18.03^{a}	3815.99	68.14	3.84^{a}
SEM	6.35	6.36	1.46	0.03	27.89	0.50	0.03
P value	0.876	0.008	0.003	0.002	0.749	0.748	0.038
MFCSR (%)							
0	554.49	1724.74^{a}	1170.25^{a}	20.89^{a}	3744.07	66.86	3.20^{a}
75	554.91	1493.24^{b}	948.33^{b}	16.93^{b}	3874.46	69.19	4.09^{b}
100	546.73	1421.65^{c}	874.92^{c}	15.62^{c}	3810.01	68.04	4.36^{c}
SEM	7.77	7.79	1.79	0.03	34.16	0.61	0.03
P value	0.657	0.002	0.002	0.002	0.381	0.381	0.002
Enzyme x MFCSR							
SEM	10.99	11.02	2.53	0.05	48.30	0.86	0.05
P value	0.736	0.865	0.321	0.321	0.768	0.768	0.601

Means with different superscripts in the same row are significant (P<0.05).
MFCSR: microbially fermented cassava starch residue; ILW: Initial live weight (g/rabbit); FLW: Final live weight (g/rabbit); TWG: Total weight gain (g/rabbit); DWG: Daily weight gain (g/rabbit/day); TFI: Total feed intake (g); DFI: Daily feed intake (g/rabbit/day); FCR: Feed conversion ratio.

Table III. Carcass of weaner rabbits fed graded levels of microbially fermented cassava starch residue supplemented with enzyme (Carcasse d'sevré lapins recevant des niveaux gradués de résidus d'amidon de manioc fermenté par voie microbienne complétée avec l'enzyme).

Enzyme (g)	Slaughtering weight(g)	Dressing %	Skin, head and limb (%)	Hind leg (%)	Foreleg (%)	Breast and rib cage (%)	Loin and abdominal wall (%)	Neck (%)
0.00	1316.83	48.01	20.97	15.43	6.47	9.43	13.30	2.49
0.35	1315.17	48.05	20.97	15.39	6.44	9.38	13.24	2.46
SEM	22.96	0.92	0.32	0.46	0.11	0.32	0.32	0.16
P value	0.955	0.966	0.991	0.953	0.832	0.912	0.881	0.903
MFCSR (%)								
0	1372.00 ^{a}	47.44	21.71 ^{b}	15.59	6.24 ^{b}	9.71	13.00 ^{b}	3.00 ^{a}
75	1385.75 ^{a}	44.95	19.44 ^{b}	15.28	5.99 ^{b}	9.21	12.33 ^{b}	2.13 ^{b}
100	1190.25 ^{b}	48.12	21.74 ^{a}	15.37	7.13 ^{a}	9.28	14.47 ^{a}	2.28 ^{b}
SEM	28.12	1.12	0.39	0.56	0.13	0.39	0.35	0.35
P value	0.001	0.723	0.022	0.923	0.002	0.633	0.002	0.021
Enzyme x MFCSR								
SEM	39.76	1.59	0.56	0.79	0.19	0.56	0.49	0.29
P value	0.993	0.991	1.000	0.991	0.991	0.992	0.994	1.000

Means with different superscripts in the same row are significant (P<0.05).
MFCSR: microbially fermented cassava starch residue.

nide ions bind to and inhibit ferric (Fe^{3+}) heme moiety form of mitochondrial cytochrome C oxidase. This blocks the fourth step in the mitochondrial electron transport chain (reduction of O_2 to H_2O), which could result in the aerobic metabolism, poor activity and death from histolytic anoxia. Tissues such as the heart and brain that heavily depend on aerobic metabolism are particularly susceptible to this effect (Olowoyeye, 2016) and this might account for the decline in the weight changes and FCR observed in the current study. This further suggests that increasing the level of replacement of maize with MFCSR in growing rabbit diet to 75% or 100% without enzyme supplementation might not bring a visible weight gain vis-à-vis economic gain. Contrarily, the multi-enzyme supplementation of the diet led to a significant respective improvement of 2.37% and 2.04% for total weight gain and FCR of the rabbits respectively, suggesting that with the multi-enzyme supplementation, the replacement level of MFCSR for maize could be increased to 100%. This is consistent with the earlier report of Ogunsipe (2014), who recorded higher significant weight in enzyme supplemented diets over the non- enzyme supplemented diets in rabbits. The improved weight gain due to

Table IV. Internal organs (% slaughter weight) of weaner rabbits fed graded levels of rumen liquor fermented cassava starch residue supplemented with multi-enzyme (Les organes internes (% poids) de l'abattage des lapins de porcelets sevrés nourris niveaux gradués du rumen de fécule de manioc fermenté alcool complétée de résidus avec multi-enzyme).

Enzyme (g)	Lung	Liver	Kidney	Heart	Pancrease	Bile	Gastrointestinal tract
0.00	0.61	2.50	0.75	0.23	0.05	0.88	17.76
0.35	0.71	2.52	0.79	0.24	0.05	0.80	17.74
SEM	0.06	0.15	0.02	0.01	0.00	0.00	0.53
P value	0.273	0.904	0.232	0.402	0.357	0.307	0.983
MFCSR (%)							
0	0.60 ^{b}	2.64	0.67 ^{b}	0.22 ^{b}	0.05 ^{b}	0.07 ^{b}	17.46
75	0.50 ^{b}	2.26	0.59 ^{b}	0.21 ^{b}	0.04 ^{b}	0.08 ^{ab}	17.25
100	0.87 ^{a}	2.63	1.04 ^{a}	0.26 ^{a}	0.06 ^{a}	0.09 ^{a}	18.55
SEM	0.07	0.18	0.03	0.01	0.01	0.01	0.65
P value	0.012	0.271	0.001	0.023	0.013	0.038	0.349
Enzyme x MFCSR							
SEM	0.10	0.26	0.04	0.02	0.04	0.01	0.92
P value	0.981	0.992	0.173	0.976	0.946	0.121	0.989

Means with different superscripts in the same row are significantly different (P<0.05).
MFCSR: microbially fermented cassava starch residue; LG: Lung, LV: Liver, KY: Kidney, HT: Heart, PC: Pancrease, BL: Bile, GT: Gastrointestinal tract.

Table V. Haematology of weaner rabbits fed graded levels of microbially fermented cassava starch residue supplemented with enzyme (L'hématologie de sevré lapins recevant des niveaux gradués de résidus d'amidon de manioc fermenté par voie microbienne complétée avec l'enzyme)

Enzyme (g)	PCV	RBC	WBC	HBC	MCV	MCH	MCHC	PLA	LYM	MON	GRA
0.00	32.15	5.31	5.48	11.02	65.15	21.90	33.40	406.00	2.08	0.57	2.63
0.35	31.95	5.11	5.65	11.13	65.65	22.45	34.07	409.33	2.23	0.62	2.62
SEM	1.30	0.23	0.29	0.48	0.63	0.21	9.68	9.68	0.20	0.04	0.07
P value	0.924	0.565	0.689	0.872	0.583	0.094	0.262	0.813	0.614	0.352	0.874
MFCSR (%)											
0	33.80^{a}	5.80^{a}	6.25	12.30^{a}	65.80	22.50	34.10	377.50^{b}	1.85	0.70^{a}	3.45^{a}
75	28.00^{b}	4.96^{b}	5.10	9.62^{b}	64.65	22.35	34.25	510.00^{a}	2.23	0.58^{ab}	1.95^{c}
100	34.35^{a}	5.32^{ab}	5.35	11.30^{a}	65.75	21.68	32.85	335.50^{c}	2.40	0.50^{b}	2.48^{b}
SEM	1.59	0.93	0.35	0.59	0.77	0.26	0.48	11.86	0.25	0.05	0.88
P value	0.032	0.031	0.094	0.023	0.514	0.092	0.122	0.001	0.322	0.028	0.001
Enzyme x MFCSR											
SEM	2.25	0.42	0.50	0.84	1.09	0.36	0.68	16.78	1.85	0.06	0.12
P value	0.876	0.275	0.151	0.912	0.493	0.014	0.921	0.043	0.832	0.156	0.458

Means with different superscripts in the same row are significantly different (P<0.05).
MFCSR: microbially fermented cassava starch residue; WBC: White blood cells (x10^9/l); LYM: Lymphocytes (x10^9/l); MON: Monocytes (x10^9/l); GRA: Granulocytes (x10^9/l); RBC: Red blood cells (x10^{12}/l); HBC: Haemoglobin conc. (g/dl); PCV: Packed cell volume (%),MCV: Mean cell volume (fl); MCH: Mean cell haemoglobin (pg); MCHC: Mean cell haemoglobin concentration (g/dl); PLA: Platelets (10^9/l).

enzyme supplementation in the rabbits may be due to the active enzymes in the multi-enzyme used in this study which might have help in breaking down long-chain polysaccharides into utilizable form, increased passage rate of digesta and decreased viscosity of intestinal content which possibly resulted in improved performance of the rabbits (Ogunsipe, 2014).

The cutting of carcass into primal parts gives room for possible comparison among variously defined areas of the animal carcass. In this study while multi-enzyme supplementation did not have significant effect on the cut parts, the MFCSR inclusion in the diets exerted profound variation in the slaughter weight, skin, head and limb, fore leg, loin and abdominal wall and neck with rabbits fed the control diet and those with 100% MFCSR maize in most cases consistently having higher values than those fed 75% MFCSR+25% maize-based diet, suggesting that higher inclusion level of MFCSR might not be detrimental to the development of the cut parts, especially that these parts are also a function of the final live weight. This is in contrast to our earlier report but consistent with the earlier report by Idowu et al. (2006).

Table VI. Serum biochemical profiles of weaner rabbits fed graded levels of microbially fermented cassava starch residue supplemented with enzyme (Les profils biochimiques sériques sevré de lapins recevant des niveaux gradués de résidus d'amidon de manioc fermenté par voie microbienne complétée avec l'enzyme)

Enzyme (g)	TP	ALB	GLO	CHO	URE	CRE	BIL	AST	ALT	ALP	AMY	GLU
0.00	59.33	43.26	16.06	113.00	37.23	1.31	1.22	134.45	91.32^{a}	189.83	656.00	59.78
0.35	58.50	42.83	15.66	105.77	35.10	1.32	1.27	159.83	80.10^{b}	185.50	705.83	58.43
SEM	1.29	0.76	0.26	2.86	2.04	0.08	0.15	19.96	3.55	10.02	34.60	1.01
P value	0.658	0.765	0.291	0.093	0.473	0.926	0.814	0.393	0.046	0.773	0.331	0.364
MFCSR (%)												
0	60.75^{a}	46.74	14.00^{c}	104.50^{b}	39.50	1.27	1.44^{a}	164.50	65.63^{b}	187.50	718.75	41.30^{b}
75	61.00^{a}	43.3	17.60^{a}	122.25^{a}	36.85	1.36	0.72^{b}	113.90	119.25^{a}	195.75	639.75	61.90^{a}
100	55.00^{b}	39	16.00^{b}	101.40^{b}	32.15	1.31	1.57^{a}	163.03	72.25^{b}	179.74	684.25	66.13^{a}
SEM	1.58	0.92	0.32	3.51	2.50	0.09	0.18	24.45	4.35	12.27	42.38	1.23
P value	0.033	0.154	0.002	0.001	0.152	0.774	0.012	0.293	0.001	0.664	0.438	0.003
Enzyme x MFCSR												
SEM	2.23	1.31	0.45	4.96	3.54	0.13	0.26	34.58	6.15	17.36	59.94	1.75
P value	0.783	0.741	0.114	0.847	0.658	0.271	0.883	0.804	0.163	0.776	0.751	0.513

Means with different superscripts in the same row are significantly different (P<0.05).
MFCSR: microbially fermented cassava starch residue; TP: Total protein (g/l); AL: Albumin (g/l); GLO: Globulin (g/l); CHO: Cholesterol (mg/dl); URE: Urea (mg/dl); CRE: Creatinine (mg/dl); BIL: Bilirubin (µ/l); AST: Aspertate amino transaminase (µ/l); ALT: Alanine Transaminase (µ/l); ALP: Alkaline phosphate (µ/l); AMY: Amylase (µ/l), GLU: Glucose (mg/dl).

The response of organ weights of animals to any test feed resource could be an indicator of whether the feed resource will precipitate toxicity to the organs or not. Thus, any observed change in organ weight could be as a result of overweight and/or an indicator of toxicity as organ will be affected by the suppression of body weight (Lu, 1996). In this study, while the effect of enzyme supplementation was not significant on the major vital internal organs, hypertrophies of internal organs such as the lung, kidney, heart, pancreas and bile in rabbits fed with 100% MFCSR-based diet were observed, suggesting the possible toxicity of the cumulated HCN in the MFCSR, which the processing method adopted in this study could not have removed. Thus, the observed enlargement of these internal organs could be attributed to their responses to the detoxification mechanisms to the hydrogen cyanide toxicity (Dutta et al., 1986, Olowoyeye 2016) in the diet having 100% MFCSR inclusion level. However, the observed values in this study are similar to those reported by Ogunsipe et al., (2014) when high sorghum offal was included in rabbit diets. The present study also showed that the effect of enzyme supplementation in the utilization of MFCSR for haematopoetic activity was not significant and that the nutritive potential of the higher replacement value of MFCSR for maize was significant for PCV, RBC, HBC, PLA, MON and GRA. The PCV, HBC, MON and GRA values were higher in rabbit fed on the control and in most cases followed by those fed on 100% MFCSR-based diets with the only interaction of the two main factors significant for MCH and PLA. These observations were in contrast to our earlier report in part I of this study, where in most cases, the blood parameters improved with the increase in MFCSR inclusion levels. However, in both instances, the blood parameters are within the normal range reported by Flecknell (2000).

In this study, all the serum metabolites measured were not statistically influenced by the multi-enzyme supplementation except ALT, while the MFCSR inclusion level significantly affected the TP, GLO, BIL, ALT and GLU. Cholesterol, a high molecular weight sterol is used in the body as raw material for healing process (Enig, 2000); useful in the normal function of the brain and it is an important component of the cell membrane including organelle inside the cell. In this study, the effect of the MFCSR inclusion level on the cholesterol level, though falling within the normal range for healthy rabbits, did not follow a well-defined pattern. Bilirubin is an end product of haemoglobin breakdown. An increase in serum bilirubin produces jaundice. This signals acute haemolytic disorder or liver diseases, while the decreased value is known for decreased red blood cell production. The occurrence of possible liver problem due to MFCSR inclusion might not be visible because the values of bilirubin were within the normal range of 0-12 mmol/l (Latimer et al., 2003). This further supports the suitability of MFCSR as an ingredient for rabbit production. However, the present observation is not in consonance with the earlier report, where the inclusion levels of MFCSR did not affect the bilirubin levels of the rabbits. The alanine aminotransferase (ALT) is important in the diagnosis of heart and liver disease. In this study, this enzyme (ALT) was affected by the multi-enzyme supplementation and level of MFCSR inclusion in the diets. The exo-enzyme supplemented diets gave a decreased ALT values while the levels of MFCSR did not produce a defined pattern of effect on the enzyme level. This is not consistent with the result of our earlier study, where the MFCSR inclusion did not precipitate any effect on ALT. However, the ALT values in both studies fell within the normal range (55-260 µ/l) reported by Flecknell (2000). This finding supports the safety of MFCSR for rabbit production as clinical/ health hazard was not observed by the rabbit particularly when the diets were supplemented with the exogenous enzyme. The MFCSR level at 75% produced a rise in glucose levels. Increased glucose value is associated with lack of insulin, severe stress and endocrine disorder (Peter and Susan, 1991; Dennis, 2005). Although, none of these situations could have existed in this study because the glucose level of the rabbits at the various MFCSR inclusion levels falls within the normal range (75-155 mg/dl) as reported by Latimer et al. (2003).

CONCLUSIONS

Addition of multi-enzyme such as used in this study at 0.35 g/kg level led to 2.37% and 2.04% improvement in weight gain and feed conversion ratio, respectively, and by extension increased in the level of maize replacement in the diets of growing rabbits to 100% MFCSR. Contrarily, without multi-enzyme supplementation, only 50% of the maize component of growing rabbit diet can be replaced with MFCSR.

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