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Pigmentation and Meat Quality of Broiler Chickens Fed Maize Replaced with *Panicum maximum* with or Without Roxazyme-G And Ronozyme-P Supplementation

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Abstract: Carcasses and meat from a flock of 240 broiler chickens in which 15% of the dietary maize was replaced with *Panicum maximum* leaf meal with or without Roxazyme-G and Ronozyme-P supplementation at 100 and 200 mg kg^{-1} were assessed pigmentation, taste, water and lipid contents and oxidative stability during refrigerated storage. Shank and skin pigmentation scores significantly ($p<0.05$) increased with increasing levels of either or combination of the enzymes in the *Panicum maximum* containing diets. The taste scores of meat were not affected ($p>0.05$) by diet and interaction of diet and muscle types (breast, drumstick and thigh). Taste scores were significant ($p<0.05$) for muscle types (6.2±2.1, 6.1±2.2 and 6.9±1.7 for thigh, drumstick and breast, respectively). Moisture content of meat was not significantly ($p>0.05$) influenced by diets and lipid contents were 5.5±0.3, 5.0±0.2 and 4.6±0.2 for thigh, drumstick and breast ($p<0.05$). Oxidative stability of meat, though significant ($p<0.05$), showed no trend in relation to diets. Meat susceptibility to oxidation was in order of thigh>drumstick>breast. Partial substitution of maize with *Panicum maximum* with or without enzyme supplementation had no adverse effect on meat quality.

Key words: Broiler chickens, *Panicum maximum* leaf meal, maize, meat quality, roxazyme-G, ronozyme-P

INTRODUCTION

Poultry meat is very popular and is relished by many people all over the world. This is attributable to the fact that little or no social or religious bias is associated with chicken meat (Mourteney, 1975). The meat from young (8-12 weeks old) broiler chickens whether male or female is tender with soft, pliable and smooth textured skin and flexible breast bone cartilage (Singh and Panda, 1988). Broiler production has been observed as a way towards realizing improved animal protein intake because of its inherent potentials as capable of meeting the protein needs of the increasing population of Nigeria.

Poor nutrition of livestock generally causes poor growth, reduced productivity and emaciation which may affect meat quality. Butson and Carter (1985) reported that different skeletal muscle exhibit differential rate of accretion in response to nutritional influences. Mulder (1995) observed that the extent of replacing conventional and unconventional feed raw materials deserves economic consideration to avoid negative effect on consumer's preference. According to Ikeme (1990), poultry meat varies in acceptability to consumers according to the tenderness, juiciness and flavour of the meat when cooked. Broiler meat is rich in unsaturated fatty acids may be very susceptible to lipid deterioration (Onibi, 2000).

Proximate composition of Guinea grass (*Panicum maximum*) harvested at 2 to 4 weeks after emergence of foliage at the Federal College of Agriculture, Akure, Nigeria showed 16% crude protein on dry matter basis (Ogunsola, 2005). In a bid to reduce the quantity of maize needed for broiler production in Nigeria, Ogunsola (2005) evaluated the replacement of 15% of maize in 542 g kg^{-1} maize-based starter and 592 g kg^{-1} maize-based finisher diets with *Panicum maximum* leaf meal with or without Roxazyme-G and Ronozyme-P supplementation. This author reported no significant difference in performance characteristics of broilers fed the enzyme supplemented diets compared with the control. Roxazyme-G and Ronozyme-P are commercial enzymes. Roxazyme-G was developed to complement digestive enzymes needed for breaking down non-starch polysaccharides into simpler molecules which birds can digest and utilise. Ronoxyme-P acts on phytin P to improve the bioavailability of phosphorus for the use of poultry (Alam *et al*., 2003).

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It is however, not enough to produce least-cost rations and cheap animal products without considering consumer's acceptability or perception of the quality and stability of the products to lipid oxidation. Thus, this study was conducted to investigate consumer's preference (as judged by Sensory Panellist) and extent of oxidation (during cold storage) of meat from broiler chickens fed diets containing *Panicum maximum* with or without Roxazyme-G and Ronozyme-P in place of maize.

MATERIALS AND METHODS

Experimental layout: Three hundred broiler chicks were electrically group brooded at the Teaching and Research Farm of the Federal University of Technology, Akure, Nigeria. During the first week of brooding, the chicks were fed a commercial broiler starter mash (23% crude protein). Thereafter, 240 chicks were selected and fed the experimental broiler starter and finisher diets for 3 and 4 weeks, respectively in metabolism cages (Table 1). Experimental design was completely randomised with 10 birds per replicate and 4 replicates per each of the 6 experimental diets. Diet 1 was the control diet with 54.15 and 59.15% maize for starter and finisher rations, respectively. Diets 2-6 for both starter and finisher rations had 15% of the maize replaced with *Panicum maximum*. Diet 2 had no enzyme inclusion; diets 3 and 4 had Roxazyme-G supplementation at 100 and 200 mg kg^{-1}, respectively; diet 5 contained 100 mg kg^{-1} Roxazyme-G and 100 mg kg^{-1} Ronoxyme-P and diet 6 had 200 mg kg^{-1} of each of Roxazyme-G and Ronoxyme-P. Crude protein content of experimental starter and finisher diets were 23 and 20%, respectively while both diets had 13 MJ kg^{-1} metabolisable energy. Feed and water were provided *ad libitum*. At the end of the feeding trial, 4 male chickens from each replicate were slaughtered, dressed, eviscerated and chilled at 4°C for 4 h before assessment of shank and skin for pigmentation and dissection into parts.

Table 1: Basal composition of experimental diets (%)

	Starter diets		Finisher diets	
Ingredients	1(Control)	2-6	1(Control)	2-6
Maize	54.15	47.03	59.15	50.78
Panicum maximum[1]	-	8.12	-	8.87
Soyabean meal	32.00	31.00	29.00	28.50
Fish meal	4.00	4.00	2.00	2.00
Others[2]	9.85	9.85	9.85	9.85
Total	100.00	100.00	100.00	100.00
Calculated analysis				
Crude Protein (%)	23.12	23.04	20.11	20.10
Metabolisable energy (MJ/kg)	13.22	13.21	13.30	13.29

[1]*Panicum maximum* at 15% replacement of maize; [2]Others: 5% brewers dried grain, 1% palm oil, 2.5% bone meal, 0.5% oyster shell, 0.15% DL-methionine, 0.2% premix and 0.5% salt; Enzyme supplementation: Diets 1 and 2 contained no enzyme; Diet 3 contained 100 mg kg^{-1} Roxazyme-G; Diet 4 contained 200 mg kg^{-1} Roxazyme-G; Diet 5 contained 100 mg kg^{-1} Roxazyme-G and 100 mg kg^{-1} Ronoxyme-P; Diet 6 contained 200 mg kg^{-1} Roxazyme-G and 200 mg kg^{-1} Ronoxyme-P

Shank and skin pigmentation: The carcasses were randomly coded with 3 digit numbers unknown to any of the 10 panellists to prevent bias judgment. The carcasses were thereafter displayed on white tiled table and the shanks and skin were visually evaluated for extent of pigmentation on a 6-point hedonic rating scale where 1 = not pigmented and 6 = highly pigmented.

Preparation of meat samples for sensory evaluation and oxidative studies: After dissections, the breast, thighs and drumsticks were wrapped in labelled cellophane bags. They were stored in a deep freezer for one week before the sensory evaluation tests were carried out. Meat samples for oxidative studies were refrigerated at 4°C for 1, 3 and 6 days before frozen storage at -18°C.

At the time of the sensory evaluation test, the thigh, drumsticks and breasts were thawed at room temperature for one hour. The meat samples (400-450 g) were packed into aluminium cooking pots (according to dietary treatments and muscle types) containing 150 mL of 1% salt solution and cooked for 20 min. After cooking, the meat samples were coded with random numbers, arranged on white enamel plates and presented to each of the 10-member panel to evaluate for appearance, taste and overall acceptability using 9-point hedonic ranking system (Larmond, 1982) (1 = dislike extremely and 9 = like extremely). Members of the panel were told to refrain from eating chewing gum or drinking for at least 3 h before the commencement of test. No information about the objective for the test was given to members of the panel and they were asked to rinse their mouths with water after each sample and to allow an interval of not less than 2 min in-between samples. The assessment was carried out at the Sensory Evaluation Laboratory of the Department of Food Science and Technology of the Federal University of Technology, Akure.

The frozen breast, drumstick and thigh for oxidative studies were chopped into small pieces after removing the bones, skin, visible fats and connective tissues. Each sample was homogenized, filtered and assayed in accordance with the method of Pikul *et al.* (1989). Extent of oxidation was expressed in mg malonaldehyde (MDA)/kg muscle. Moisture and lipid contents of the uncooked meat were determined according to AOAC (1995) method. Score data for sensory evaluation were square root transformed and all data were subjected to One-way Analysis Of Variance (ANOVA) and factorial analysis as appropriate, using the Minitab Statistical Package (v.10.2, Minitab, Inc. USA).

RESULTS

Organoleptic characteristics of meat: Results of the shank and skin pigmentation are presented in Table 2. The skin pigmentation was significantly ($p<0.05$) influenced by the dietary treatments. Diet 1 (feed without *Panicum maximum* and no enzyme) had 2.5±0.53 while diet 2 (15% *Panicum maximum* without enzyme) had a score of 1.3±0.05. However, skin pigmentation scores increased from diet 3 to diet 6. Diet 3 (feed with 15% *Panicum maximum* with 0.01% Roxazyme-G) had a value of 3.0±0.47 while diet 6 (with 15% *Panicum maximum* with 0.02% Roxazyme-G + 0.02% Ronozyme-P) had a value score of 6.0±0.00) indicating higher pigmentation at higher level of inclusion of the 2 enzymes. Shank pigmentation followed the same pattern with that of skin pigmentation. Diet 6 had a value of 5.2±0.79 while the smallest value was recorded for diet 2. Shank pigmentation increased with increasing level of enzymes in the diet.

Appearance and taste scores of the cooked meat sample are presented in Table 3. The appearance of the cooked meat samples was not significantly ($p>0.05$) influenced by the dietary treatments, muscle type and the interaction of these 2 factors. Appearance scores of the cooked meat was numerically highest for muscles from birds fed diet 6 (0.02% Roxazyme G + 0.02% Ronozyme P + 15% *Panicun maximum*) being 7.0±2.1. The breast muscle had the highest score (6.8±1.9) while the lowest score (6.3±2.2) was that of thigh. Taste score was significantly influenced ($p<0.05$) by muscle type but not by diet or interaction between diet and muscle type. The taste score of the cooked meat was numerically highest for muscles from birds fed diets 3 (15% *Panicum maximum* + 0.01% Roxazyme G) being 7.0±1.3 while the lowest score was from the muscle from birds fed diet 5 (0.01% Roxazyme G + 0.01% Ronozyme P + 15% *Panicum maximum)* being 5.9±2.3. Breast muscle recorded the highest significant ($p<0.05$) taste score of 6.9±1.7, thigh had 6.2±2.1 and the lowest taste score of 6.1±2.2 was for drumstick.

Tabl 2: Shank and skin pigmentation of broiler chickens Fed *Panicum maximum* with Roxazyme-G and Ronozyme-P in Place of Maize

Diet	Skin pigmentation	Shank pigmentation
1	2.5±0.53[f]	3.2±0.41[b]
2	1.3±0.50[e]	2.4±0.84[c]
3	3.0±0.47[d]	3.3±0.82[b]
4	4.0±0.81[c]	3.7±0.82[b]
5	4.9±0.74[b]	4.7±0.95[a]
6	6.0±0.30[a]	5.2±0.79[a]

[a]Mean±SD; n = 10 (representing 10-member panel); Means with different superscript within the same column are significant ($p<0.05$)

Table 3: Appearance and Taste of Meat from Broiler Chickens Fed *Panicum maximum* with Roxazyme-G and Ronozyme-P in Place of Maize

Diet	Muscle type	Appearance	Taste
1	Thigh	6.7±1.95	5.7±2.6
	Drumstick	6.4±1.60	6.9±1.7
	Breast	6.9±2.3	8.1±0.9
2	Thigh	5.4±3.0	6.6±1.4
	Drumstick	5.4±1.5	5.7±1.8
	Breast	6.4±1.4	6.7±1.1
3	Thigh	5.6±2.3	6.9±1.3
	Drumstick	5.8±1.1	7.0±1.6
	Breast	6.9±2.4	7.0±0.9
4	Thigh	6.4±2.2	6.0±2.1
	Drumstick	5.6±2.4	5.6±3.1
	Breast	6.7±1.1	71±1.7
5	Thigh	6.8±0.9	6.1±2.3
	Drumstick	5.9±2.6	5.0±2.4
	Breast	6.1±2.8	5.8±2.2
6	Thigh	6.6±2.3	5.8±2.4
	Drumstick	6.6±2.6	6.4±2.1
	Breast	7.8±1.0	5.9±2.2
Statistical significance			
Diet		NS	NS
Muscle type		NS	*
Diet x Muscle type		NS	NS
Mean comparison			
Dietary effect			
1		6.8±1.9	6.9±2.1
2		6.4±2.2	6.3±1.5
3		6.4±2.0	7.0±1.3
4		6.2±1.9	6.2±2.4
5		6.3±2.2	5.9±2.3
6		7.0±2.1	6.0±2.2
Muscle type effect			
Thigh		6.3±2.2	6.2±2.1[b]
Drumstick		6.5±2.1	6.1±2.2[b]
Breast		6.8±1.9	6.9±1.7

[a]Mean±SD; NS = Not significant ($p>0.05$); * = ($p<0.05$) n = 10 (representing 10-member panel); Means with different superscripts within the same column and for the same parameter are significant ($p<0.05$)

Moisture and lipid contents of meat: Moisture and lipid contents of meat from broiler chickens fed *Panicum maximum* leaf meal with or without Roxazyme-G and Ronozyme-P are presented in Table 4. There was no significant ($p>0.05$) difference in the moisture content due to the dietary treatments and the interactive effect of the diets and the muscle type. However, muscle type affected ($p<0.05$) moisture content. The mean moisture content ranged between. 71.1±1.9 and 72.3±1.6% in all the muscle types. Thigh and drumstick had moisture contents that were significantly higher ($p<0.05$) than that of breast.

The lipid content of the meat was significantly influenced by dietary treatments ($p<0.001$), muscle type ($p<0.001$) and the interaction of these factors ($p<0.01$). The lipid content was highest in thigh muscle averaging 5.5±0.3% with a range of 4.9±0.2 to 6.2±0.2%. The lipid content in drumstick on average was 5.0±0.2%. The least lipid contents was observed in breast muscle with an average of 4.6±0.2% ($p<0.05$). The lipid content of the muscle from birds fed diet 4 was the highest being 5.3±0.6% on the average while birds on diet 2 had the least lipid content of 4.6±0.4% ($p<0.05$).

Table 4: Moisture and Lipid Contents (%) of Meat from Broiler Chickens Fed *Panicum maximum* with Roxazyme-G and Ronozyme-P in Place of Maize

Diet	Muscle type	Muscle type	Lipid
1	Thigh	73.5±0.2	4.9±0.2
	Drumstick	72.0±1.5	4.8±0.3
	Breast	70.4±0.5	4.6±0.1
2	Thigh	72.3±1.2	5.1±0.4
	Drumstick	73.5±0.6	4.7±0.2
	Breast	71.1±0.9	4.3±0.1
3	Thigh	69.7±2.6	5.6±0.5
	Drumstick	72.4±1.3	5.1±0.1
	Breast	71.2±4.7	4.8±0.1
4	Thigh	71.1±2.4	6.2±0.2
	Drumstick	73.1±0.3	5.1±0.1
	Breast	69.6±1.1	4.8±0.1
5	Thigh	72.4±0.3	5.2±0.2
	Drumstick	71.8±0.5	5.1±0.1
	Breast	71.0±1.5	4.8±0.1
6	Thigh	71.4±0.8	5.6±0.2
	Drumstick	71.8±0.2	5.2±0.1
	Breast	69.8±1.3	4.8±0.2
Statistical significance			
Diet		NS	***
Muscle type	***	***	
Diet x Muscle type		NS	**
Mean comparison			
Dietary effect			
1		72.3±1.6	4.8±0.2^{ab}
2		72.3±1.3	4.6±0.4^{a}
3		71.1±1.9	5.2±0.5^{bc}
4		71.3±2.0	5.3±0.6^{c}
5		71.7±1.0	5.0±0.2^{ac}
6		71.0±1.2	5.2±0.4^{bc}
Muscle type effect			
Thigh		71.7±0.8^{b}	5.5±0.3^{c}
Drumstick		72.6±1.0^{b}	5.0±0.2^{b}
Breast		70.5±1.3^{a}	4.6±0.2

[a]Mean±SD; NS = Not significant (p>0.05); n** = p<0.01; *** = p<0.001; n = 4; Means with different superscripts within the same column and for the same parameter are significant (p<0.05)

Oxidative stability of meat: Results of extent of oxidation, measured as the concentration of malonaldehyde (MDA) in meat during refrigerated storage are presented in Table 5. There was no significant (p>0.05) difference due to diets on MDA concentration at day 1 of storage. On day 3 of storage, diet had significant (p<0.01) influence on MDA concentration. Muscle type influenced the MDA concentration at days 1, 3 and 6 of storage (p<0.001). Interaction effects of diet and muscle types were not significant (p>0.05).

MDA concentrations in meat significantly (p<0.01) increased with increasing storage length irrespective of diet and muscle type. The MDA concentrations in meat did not follow any trend in relation to dietary treatments on days 1, 3 and 6 of refrigerated storage. Thigh muscle had the highest value of 0.51±0.06 mg MDA kg^{-1} meat, followed by drumstick with 0.45±0.05 mg MDA kg^{-1} meat while the least was recorded for breast (0.43±0.04 mg MDA kg^{-1} meat) (p<0.05). The MDA concentrations for days 3 and 6 followed the same pattern with that of day 1.

Table 5: Oxidative Stability (mg MDA/kg meat) of Refrigerated Meat from Broiler Chickens Fed *Panicum maximum* with Roxazyme-G and Ronozyme-P in Place of Maize

		Storage Length at 4°C (Days)			
Diet	Muscle Type	1	3	6	Statistical Significance
1	Thigh	0.52±0.05	0.77±0.02	0.87±0.07	
	Drumstick	0.46±0.05	0.71±0.03	0.88±0.04	**
	Breast	0.44±0.04	0.67±0.01	0.78±0.03	***
2	Thigh	0.54±0.04	0.71±0.04		***
	Drumstick	0.46±0.03	0.61±0.03	0.90±0.02	***
	Breast	0.46±0.01	0.69±0.04	0.74±0.04	***
3	Thigh	0.54±0.05	0.65±0.07	0.78±0.04	***
	Drumstick	0.48±0.01	0.60±0.05		**
	Breast	0.41±0.07	0.53±0.09	0.83±0.05	***
4	Thigh	0.48±0.04	0.66±0.03	0.79±0.01	***
	Drumstick	0.42±0.10	0.53±0.09	0.73±0.01	**
	Breast	0.41±0.12	0.62±0.05		***
5	Thigh	0.52±0.01	0.54±0.05	0.95±0.02	***
	Drumstick	0.43±0.04	0.51±0.05	0.92±0.04	***
	Breast	0.41±0.02	0.52±0.08	0.77±0.02	***
6	Thigh	0.47±0.03	0.62±0.01		**
	Drumstick	0.43±0.02	0.63±0.04	0.91±0.01	***
	Breast	0.44±0.02	0.49±0.05	0.86±0.03	***
Statistical significance					
Diet	NS	**	**		
Muscle type		***	***	***	
Diet x Muscle type		NS	NS	NS	
Mean comparison					
Dietary effect					
1		0.47±0.05	0.68±0.10^{b}	0.84±0.10^{b}	
2		0.49±0.05	0.64±0.07^{ab}	0.81±0.09^{ab}	
3		0.48±0.07	0.60±0.07^{a}	0.78±0.07^{a}	
4		0.44±0.09	0.63±0.10^{ab}	0.88±0.09^{ab}	
5		0.45±0.06	0.56±0.08^{a}	0.83±0.10^{ab}	
6		0.44±0.03	0.58±0.08^{a}	0.80±0.11^{ab}	
Muscle type effect					
Thigh		0.51±0.06^{b}	0.68±0.07^{c}	0.90±0.07^{c}	
Drumstick		0.45±0.05^{a}	0.63±0.06^{b}	0.83±0.08^{b}	
Breast		0.43±0.04^{a}	0.54±0.06^{a}	0.74±0.05^{a}	

Mean±SD; NS = Not significant (p>0.05); ** = p<0.01; *** = p<0.001; N = 4; Means with different superscripts within the same column and for the same parameter are significant (p<0.05)

DISCUSSION

Skin pigmentation of birds fed diet 2 was the least. This may be due to the fact that birds were not able to efficiently utilize *Panicum maximum* without synthetic enzymes since they are simple stomach animals (Ajaja, 2002). Skin pigmentation increased with increasing level of Roxazyme-G and Ronozyme-P. This could also be attributed to increasing utilization of dietary fibre with increasing enzymes levels in the diets thereby making more nutrients, especially beta carotene (precursor of vitamin A) which have been reported to be present in leaves (Agbede and Aletor, 2003) available for pigmentation. Shank pigmentation followed similar trend to that of skin pigmentation. The pigmentation of these 2 parts is in line with the report of Agbede *et al*. (2002), that the more the concentration of enzymes in the diet, the more degradation of the feed components and the more available, all the nutrients are for birds utilization.

The non-significant influence of the diets, muscle type and their interaction on the appearance of the cooked meat showed that muscle types from the different diets have similar appearance. The taste preference for the breast muscle compared with thigh and drumstick by members of the taste panel may be attributed to reduced fatty nature of the breast.

The moisture contents of muscles obtained in this study averaging 71.7±0.8, 72.6±1.0 and 70.5±1.3% for thigh, drumstick and breast, respectively were within range of 65 and 76% reported for poultry by Adrian *et al.* (1982) and similar to those reported for breast (73.8-75.1%), drumstick (73.9-75.8%) and thigh (73.7-75.6%) by Onibi (2006).

The lipid content for thigh, drumstick and breast, respectively fall within the range of 4 and 12% reported by Adrian *et al.* (1982) for poultry. The significant effect of diet, muscle type and interactive effect of these factors on lipid contents of the meat agree with the report of Ikeme (1990) and Onibi (2000) that fat content of meat varies with diets and the different muscles in the same bird. There was a trend of increasing lipid contents of meat with increasing level of supplementation of Roxazyme-G and Ronozyme-P attributable to these enzymes making more nutrients available from the *Panicum maximum* leaf meal. Meat from birds fed diet 2 (*Panicum maximum* leaf meal without Roxazyme-G or Ronozyme-P) had the lowest lipid content. The fat contents obtained in this study for thigh muscle were higher than those of drumstick and breast. This agrees with Ikeme (1990) report that thigh muscles have numerous fat globules than the breast muscles. Also, Olomu (1995), Aduku and Olukosi (2000) reported that lipid content is the most variable components of meat and there are variations in the amount of lipid deposited at different parts of poultry being higher in thigh muscles than drumstick and lowest for breast.

Malonaldehyde concentration increased with increasing length of storage irrespective of muscle type. This agrees with earlier reports of Pikul *et al.* (1989), Monahan *et al.* (1992) and Onibi (2000) reported that deterioration due to oxidation continue to occur during refrigerated storage of meat. The diets did not show any consistent trend in relation to oxidative stability of the meat. The MDA concentrations of breast muscle were the lowest for all the storage length, followed by drumstick and highest for thigh. This showed that the thigh muscle which had highest fat content than drumstick and breast was most susceptible to oxidation and hence the higher MDA concentration. This agrees with earlier studies (Lin *et al.*, 1989; Pfalzgraf *et al.*, 1995), that the higher the quantity of lipid in meat, the higher the susceptibility of the meat to lipid oxidation.

CONCLUSION

Shank and skin pigmentation of the chickens was improved by supplementation of diets containing *Panicum maximum* leaf meal with Roxazyme-G and Ronozyme-P. Overall acceptability of broiler chicken meat was not adversely affected by the dietary treatments. Thus, the meat quality of broilers fed *Panicum maximum* leaf meal would be acceptable to consumers without any undesirable effect. Thus, *Panicum maximum* leaf meal with Roxazyme-G and Ronozyme-P at the levels studied may be considered as a good and low cost replacement for highly expensive maize in broiler chicken diets.

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