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Effects of rigor state and muscle type on physicochemical properties, processing and storage stability of two different pig skeletal muscles

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***Target audience:** meat processors, consumers, researchers*

Abstract

Twelve (12) growing male Large White x Landrace crossbred pigs, aged 12 weeks and weighing between 25 and 30 kg were used to assess the impact of rigor state on pork quality characteristics with respect to muscle position. Animals were fed on grower's ration (crude protein = 18.06 %) for 10 weeks. Six pigs per treatment (pre- and post-rigor) were slaughtered at the age of 6 months to evaluate pH, shear force tenderness, water-holding capacity (WHC), cooking and storage losses. Data were subjected to General Linear Model in a 2 x 2 factorial arrangement. pH was higher ($p<0.05$) at pre-rigor state than at post-rigor; while those of the back and thigh muscles did not differ significantly. Rigor state did not influence WHC. Higher shear force ($p=0.014$) of 2.08 kg were observed at pre-rigor state while a lower value (0.16 kg) was obtained post-rigor. Cooking losses were influenced ($p=0.000$) by rigor state. Thigh muscle had higher refrigeration and freezing thaw losses. It was concluded that pig meat in post-rigor state could yield better quality product in terms of pH value, shear force, tenderness while minimal cooking and storage losses could be achieved at pre-rigor state.

***Key words:** pig meat; rigor state; muscle position; shear force; cooking loss; storage stability.*

Description of Problem

Over recent decades, there has been a transformation in the nature and objectives of swine production, most especially the establishment of a large scale integrated agricultural industry, producing large number of high quality meat products due to increasing demand for meat. As a matter of fact, global pork production has increased up to four times over the last fifty years and is expected to continue to grow over the next three decades (1). Hence, swine production is necessarily concerned with the quality of the pork produced and the efficiency of its production (2).

For many centuries, man has used animal tissues for food without paying much attention either to their living functions, or to the changes that might take place in them before consumption. After slaughter, the circulatory system ceases to function and Adenosine triphosphate (ATP) levels in the cells decrease. This is gradually followed by post-mortem muscle contraction (3). During this period, there is shortening and stiffness of the muscles causing the joints to be immobile (4). The process of *rigor mortis* is initiated as soon as ATP reservoir becomes used up and is unable to resynthesize from glycogen and creatine phosphate. Thus, factors that influence their content at slaughter indirectly affect *rigor mortis* onset and intensity (5).

Among other factors such as animal age (6), gender (7), storage temperature (8); muscle type could be an important determinant of meat tenderness. Variation in tenderness occurs among different muscle types differing in location and functions in an animal body. These differences are generally attributed to the collagen content or differences in contraction or stretching during *rigor mortis* (9). Tenderness is a complex trait which is influenced by two primary structural features of muscle namely: integrity of myofibrils (termed actomyosin effect) and the connective tissue contribution (termed background effect).

To the consumer, appearance is the major criterion for purchase, selection and initial evaluation of meat quality. The satisfaction derived from a product is determined by consumers' perception of colour, texture, juiciness and flavor (10). Technological qualities which are a reflection of the ability of a product to undergo processing also determine meat quality. They are particularly connected to a reduced water-holding capacity during cooking and cold storage or the damage which results from the subjection to external pressure such as cutting, gravity, heating, centrifugation and pressing (10). Yet, the impact of rigor state on pork quality characteristics with respect to muscle position requires further studies. Therefore, the objective of this study is to determine the effects of rigor state and muscle position on pH, water-holding capacity, cooking and storage stability, and shear force tenderness of pre- and post-rigor pig meat.

Materials and Methods

Experimental Site

The first phase of the experiment was carried out at the Piggery Unit of Directorate of Farms, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria while the second phase took place at the Meat Processing Laboratory, Animal Production and Health Department of the University.

Experimental Animals and Management

Twelve growing male Large White x Landrace crossbred pigs, aged 12 weeks and weighing between 25 and 30 kg were used for the study. The pigs were fed on grower ration (CP – 18.06%, M.E – 2906kcal) for ten (10) weeks. Feed and water intake were monitored and good hygiene practices were observed. Animals were housed in individual pen and dewormed using Kepromec ® oral. The composition (%) of the experimental diet is presented in Table 1:

Table 1: Percentage Composition of Experimental Diet

Ingredients	%
Maize	45.00
Groundnut cake	20.00
Wheat offal	20.00
Palm kernel cake	12.00
Bone meal	2.00
*Grower premix	0.45
Salt	0.50
Methionine	0.05
Total	100.00
Calculated analysis	
Crude protein (%)	18.06
Crude fibre (%)	5.84
Calcium (%)	0.72
Phosphorus (%)	0.34
Metabolisable energy (kcal)	2906.00

*****Contained the following per kg diet: Vit. A 12,600IU; Vit. D 2800IU; Vit. E 49IU; Vit K_3 2.8 mg; Vit. B_1 1.4 mg; Vit B_2 5.6 mg; Vit B_6 1.4 mg; Vit B_{12} 0.014 mcg; Niacin 21 mg; Pantothenic acid 14 mg; Folic acid 1.4 mg; Biotin 0.028 mcg; Choline chloride 70 mg; Mn 70 mg; Zn 140 mg; Fe 140 mg; Cu 140 mg; Iodine 1.4 mg; Selenium 0.28 mg; Co 0.7 mg; Antioxidant 168mg.

Slaughtering of animals

At the age of five months, six pigs (one per replicate) were stunned and slaughtered via the jugular vein followed by bleeding. Carcasses for treatment 1 (pre-rigor) were used immediately for data collection before the onset of *rigor mortis.* Carcasses for treatment 2 (post-rigor) were stored in a cold room at 4ºC for 24 hours to allow for the process of *rigor mortis* to be completed.

Data Collection

Data were collected on the pH, cooking losses, refrigeration and thaw losses, and shear force tenderness both at the pre- and post-rigor stages from the back and thigh muscle.

pH Determination

The pH of the back muscle (*longissimus dorsi*) were taken thrice between the 4th and 10th rib of the thoracic vertebrae and on the left thigh muscles (*biceps femoris*) using Jenwayº 3015 model glass electrode pH meter by inserting the probes to a depth of 4-5 cm into the intact muscles postmortem at pre- and post-rigor stages.

Shear force Determination

Warner-Bratzler shear force was determined according to (11) guidelines. The meat samples were cooked in a water bath to an internal temperature of 80ºC for 20 minutes. After cooling to room temperature for 2-3 hours, about 1.3 cores were taken in triplicates from each sample parallel to the muscle fibres using a driller. The cores were sheared perpendicular to the orientation of the muscles fibres. Shear force was determined using a Warner-Bratzler shear force device.

Water-holding Capacity

This was measured using the Filter Paper Press Method (FPPM) as modified by (12). 1g of each meat sample was weighed and then placed between two filter papers (Whatman No. 1); the filter-papers with meat sample were pressed between two plexi-glass plates at approximately 35.2kg/cm^2 for 1 minute. The pressed samples were oven dried at 70ºC for 24 hours in order to determine the moisture contents. The amount of water released from the sample was measured indirectly by measuring the areas of the filter paper wetted relative to the areas of the pressed sample using a tracing paper and a graph sheet.

$$WHC = 100 - \frac{(Ar - Am)x\ 9.47}{Wm\ x\ Mo}$$

where Ar = Area of water released from meat (cm^2); Am = area of meat sample (cm^2); Wm = weight of meat sample (mg); Mo = moisture content of the meat (%) and 9.47 is constant.

Cooking Loss

This was determined according to (13). 50g each of fresh pork samples were cooked in a water bath to an internal temperature of 70ºC for 20 minutes. The cooking loss was obtained as the difference between pre-cooking weight and post- cooking weight.

Cooking loss % = [(pre-cooking weight – post-cooking weight)/pre- cooking weight] × 100

Refrigeration loss

50g each of uncooked pork samples were weighed separately before refrigeration. They were reweighed after 24 hours of refrigeration at 4ºC. Refrigeration loss was the difference between the pre- and post- refrigeration weight of the samples.

Refrigeration loss % = [(pre-refrigeration weight – post-refrigeration weight)/pre- refrigeration weight] × 100

Freezing Thaw Loss

This was determined according to the procedures described by (14). The pork samples were frozen for 24 hours at -10ºC after which they were removed and allowed to thaw for 2 hours. Freezing thaw loss was the difference between the pre- and post-freezing weight of the samples.

Freezing thaw loss % = [(pre-thawing weight – post-thawing weight)/pre- thawing weight] × 100

Statistical Analysis

Data were evaluated by ANOVA (SPSS Inc., 2010) (15) using General Linear Model in a 2 x 2 factorial design that included rigor state and muscle type as factors.

Results

Table 2 shows the effects of rigor state and muscle type on the pH, water-holding capacity (WHC) and shear force tenderness of two different pig skeletal muscles. Rigor state influenced (p=0.000) the muscle pH. Muscles in pre-rigor state recorded a higher pH value (7.02) than those of post-rigor (6.15). However, pH was not affected (p=0.414) by the muscle type. Meat samples from the back muscles (6.53) recorded a numerically lower pH compared to the thigh (6.63) muscles. There were no significant interactions between rigor state and muscle type.

State of rigor and muscle type did not affect WHC. Meat samples obtained from back muscles recorded WHC value of 59.30 %, while those from the thigh muscles had a value of 37.46 %. The interaction between rigor state and muscle type revealed no significant differences between the two factors.

Rigor state showed higher (p=0.014) shear force value of 2.08 kg in cooked meat samples pre-*rigor mortis* while a lower value of 0.16 kg was observed post-rigor. Although, shear force was not influenced (p=0.576) by muscle type, it was observed that cooked meat samples from back muscles had a higher shear force value (1.30 kg) compared to those obtained for thigh muscles (0.94 kg).

The main effects of rigor state and muscle type on percentage cooking, refrigeration and freezing thaw losses of two different pig skeletal muscles are shown in Table 3. Percentage cooking losses of meat samples were influenced (p=0.000) by rigor state, with those at pre-rigor state recording minimal losses (12.63%) than those at post-rigor (36.02 %) state. Meat samples obtained from back (23.55 %) and thigh (25.10 %) muscles did not exhibit significant differences between the two factors. No interaction was observed between rigor state and muscle type for percentage cooking losses.

Rigor state did not (p=0.05) influence refrigeration losses of meat samples. Meat samples at pre-rigor state recorded a refrigeration loss of 8.25 % while post-rigor samples had a lower loss of 6.50 %. Samples from back muscles had a more reduced (p=0.00) percentage refrigeration loss of 4.29% while those of thigh muscles had a significantly higher loss (10.46 %). Significant interaction (P = 0.000) was noted for refrigeration loss between the two factors studied (pre-rigor back = 3.16 %, pre-rigor thigh = 13.34 %, post-rigor back = 5.42 %, post-rigor thigh = 7.58 %).

Pre-rigor (10.24 %) and post-rigor (7.21 %) states had no significant impact on freezing thaw loss of pig meat samples. However, significant (p<0.05) differences were observed in the percentage freezing thaw loss of meat samples obtained from back (6.03 %) and thigh (11.42 %) muscles.

Table 2: Main effects of rigor state and muscle type on pH, water-holding capacity and shear force of two different pig skeletal muscles

Parameters	Rigor state (R)		Muscle (M)		SEM	P-value		
	Pre-rigor	Post-rigor	341	Thigh		R	M	R x M
pH	7.02	6.15		6.63	0.06	0.000	0.414	0.989
WHC (%)	41.72	55.03	59.30	37.46	0.486	0.239	0.070	0.558
Shear force (kg)	2.08	0.16	1.30	0.94	0.971	0.014	0.576	0.493

SEM: Standard Error Mean

Table 3: Main effects of rigor state and muscle type on cooking, refrigeration and freezing thaw losses of two different pig skeletal muscles

Parameters	Rigor state (R)		Muscle type (M)		SEM	P-value		
	Pre-rigor	Post-rigor	Back	Thigh		R	M	R x M
Cooking loss (%)	12.63	36.02	23.55	25.10	0.665	0.000	0.255	0.995
Refrigeration loss (%)	8.25	6.50	4.29	10.46	0.486	0.081	0.000	0.000
Freezing thaw loss (%)	10.24	7.21	6.03	11.42	0.971	0.129	0.009	0.170

SEM: Standard Error Mean

Discussion

pH which is a degree of acidity or alkalinity of a solution is an important factor which determines meat quality and ranges from about 5.2 to 7.0. According to (16), the highest quality products usually tend to fall within a pH range of 5.7 to 6.0. The lower pH values obtained in post-rigor state could be as a result of postmortem muscle energy metabolism which led to a pH decline due to the accumulation of H^+ (17). Energy reserves depletion as *rigor mortis* develops within the muscle tissues culminate into the lowering effect of the muscle temperature and pH resulting from cessation of blood supply and the chilling process (18). Anaerobic glycolysis that occur within the muscle generates lactate that accumulates, lowering the intracellular pH of meat, so that by 24 hours post- mortem the pH has fallen to an ultimate pH of about 5.4-5.7. The rate at which meat pH declines during rigor development may be one of the most important post-mortem factors which influences quality characteristics of meat with respect to colour, water-holding capacity (WHC) and tenderness (19,20,21,22). Lowered WHC reduces meat yield during processing.

(23) stated that very little drip loss occurs in pre-rigor meat but later after rigor onset, drip losses tend to increase while (24) revealed that meat with a higher pH had greater water-holding capacity than that with a low pH. As the pH of the muscle lowers as it converts to meat, the complex latticework of the myofibrils in the muscle cell shrinks, reducing the space within the myofibrils where water can reside, thereby reducing the WHC of post-rigor meat. Although the mechanism involved is not very clear, however, the reason for lower WHC obtained in meat from thigh muscles compared to higher value in meat samples from the back muscles at pre-rigor states may be position dependent. According to (16), this result could possibly be based on the structure of the muscle cells itself. The structure of the thigh muscle cells (which are particularly used

for locomotion) differ from those from the back muscles, and may have played a vital role in the lower values obtained.

The slightly higher shear force values of meat samples from back muscles may be an indication that pig meat in post-rigor state is more tender than pre-rigor meat due to the low shear force value of the former. Shear force has been used as an objective measurement of meat tenderness (25) and tenderness remains a vital element for the evaluation of eating quality. Connective tissues are responsible for the variation observed in background toughness (26).

The toughening of meat at the onset of *rigor mortis* caused by sarcomere shortening could be responsible for the higher shear force value obtained for the pre-rigor meat (27; 28) while the resolution phase of *rigor mortis*, which is characterized by tenderizing of meat postmortem could be responsible for the lower shear force experienced in the post-rigor meat samples. The connective tissue, sarcomere length and myofibrilar breakdown are the major sources of meat tenderness variation (29). (29) stated that tenderization of meat is impacted by enzymatic breakdown. Proteolysis of the myofibrils forms the basis of the tenderizing phase (26; 30). According to (26) and (31), the major proteolytic enzyme system involved in tenderization of aged pork post mortem is the calpain system, involving two calcium requiring enzymes, µ-calpain and m-calpain, and calpastatin acting as an inhibitor. (32) stated that because of savings in time and money and the difficulty to obtain a well-trained sensory panel, tenderness of cooked samples can be assessed more easily using Warner-Bratzler shear force than trained sensory panel analysis.

An increase in cooking loss (as obtained in post-rigor meat) has a significant financial impact in pork industry and reduces the nutritional quality of meat. Meat and meat products contain substantial amounts of high protein quality and are good sources of a number of essential minerals, which include iron, zinc and vitamins B. The increased loss of such nutrient degrades pork nutritional quality and lowers its purchase (33) which implies that pre-rigor meat could yield a better nutritional quality product.

The non- interaction between rigor state and muscle type for freezing thaw loss is in agreement with (34) who reported that no differences ($p>0.05$) were detected in drip loss of hams or loins or in thaw loss of loin chops; but is in contrast with the findings of (35) who observed an increase in thawing loss from the post-rigor pork samples. Thawing loss is the loss of fluid in meat as a result of exudate formation after freezing and thawing. Water is lost from the meat in the form of purge during storage and in thawing of frozen meat, and during cooking through evaporation and drip loss (stock) (36, 37).

Conclusion and Applications

1. The present study reveals that superior quality meat products could be obtained in pre-rigor pig meat harvested from back muscles as a result of minimal nutritional losses from cooking and refrigeration storage.
2. Pork carcasses suspended while subjected to chilling prior *rigor mortis* onset could produce a more desirable ultimate pH.
3. Due to its low shear force value, post-rigor pork could yield desirable quality for meat processors and consumers requesting for tender meat.

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