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Effect of unconventional marinades on beef quality

[1]*Obajuluwa, O. V., [1]Sanwo, K. A., [1]Akinola, O. S., [2]Sobukola, O. P., [1]Adegoke, A.V. and [2]Faloye, O. R.

[1]*Department of Animal Production and Health, College of Animal Science and Livestock Production, University of Agriculture, Abeokuta, Nigeria.*

[2]*Department of Food Science and Technology, College of Food Science and Human Ecology, University of Agriculture, Abeokuta, Nigeria*.

***Corresponding author:** olusolaobajuluwa@gmail.com; 07068500907

Abstract

There is consumer's preference for tender meat. The use of tenderizers to soften meat helps to reduce nutrient losses due to prolonged cooking. This study was conducted to compare the effects of four unconventional marinades (Pawpaw leaves extract, Lime-juice, and Carbonated drinks and distilled water) on the quality of beef. One thousand five hundred grams (1500 g) of beef excised from the thigh muscle was cut into twelve whole pieces of similar sizes and were randomly distributed into four experimental groups in a Complete Randomized Design (CRD). Treatment groups were randomly allotted to experimental marinades measuring 300mL with each marinade containing 100mL of tenderizers. Marination spanned for a period of 24 hours at a temperature of 4 ^{0}C. Data collected were: beef proximate composition, weights with pH of beef and marinades, cook and refrigeration losses and sensory scores. Results showed significantly (P<0.05) higher fat and Fe content in beef treated with Lime-juice and Pawpaw leaves extract-based marinades respectively. Marination did not affect (P>0.05) crude protein, ash, moisture content, and calcium. Weights and pH from beef and marinades varied significantly (P<0.05) after marination. Pawpaw leaves extract-based marinades produced more (P<0.05) tender beef; while beef marinated with carbonated drink marinade had significantly (P<0.05) better flavour and overall acceptability. It is therefore concluded that carbonated drink based marinades produced beef with the best quality.

Keywords: Pawpaw leaves, lime juice, carbonated drink, beef, marinades, meat quality

Introduction

Nigeria is one of the largest meat consumers and producers in West Africa (Osho and Asghar, 2005). The household consumption of various meat products in order of consumption rates are: beef (77%) followed by fish (68%), chicken (22%), and mutton and chevon (15%) while pork and mini-livestock were (1% and 4%) respectively (Ezedinma *et al.,* 2006). Mutton, Beef, Chevon, Veal, Pork, Chicken, Bacon, etc are meat from farm animals which help supply protein to man. The projected meat consumption across countries and regions in developing countries in Africa has been estimated to increase at the rate of 36.7 Kilogram (Kg) of meat per year between 2000 and 2030 (FAO 2003).

In spite of the relatively high consumption of beef compared to other meat products, beef produced in Nigeria is relatively tough and thus, increasing the cooking time needed to tenderize it before consumption by consumers, leading to reduction in its nutritive quality. In other to soften beef during processing, some methods are employed which can be grouped into chemical, mechanical and manual methods. These methods involve quick freezing and aging before rigor mortis, marination, pressure cooking, disruption of muscles by blades or hammer, and muscle stretching

(Alvanado and Sams, 2003). Amongst the chemical approach is the use of refined natural proteolytic enzymes like papain, ficin, and bromelin (Cunningham, 1998). Also, natural acids like acetic acid or vinegar, lactic acid, and citric acids are being used for softening beef (Macrae *et al.,*1993, Warriss, 2000). The proteolytic enzymes and acids soften meat by denaturing protein and breaking down the collagen, muscle fibres and tissues that connect them.

In order to improve palatability qualities of meat (juiciness and flavour) alongside with tenderness, seasoning, spices, and oil are included in the tenderizing solution to form marinades. Marination is the process of soaking foods in a seasoned, often acidic liquid before cooking (Corriher *et al.,* 2012). Marination is often used as flavour in foods and to tenderize meat. (Filippone, 2012). Bille and Taapopi, (2008) used two commercial meat tenderizers (acidic and enzymatic) on different cuts parts of goat meat (namely, the back, the ribs, and the hind limbs). Also, in search for alternative tenderizers, which are cheaper and safer, different researches have been conducted which include the use of proteolytic enzymes such as: Papain, Bromelin, and Ficin (from plant) and Bacillus protease, Aspartic protease (micro-organism origin) by Chris (2007) in the softening of beef. Acids like: Citrus juice (Kahraman *et al.,* 2012), citric acid, lemon juice, (Shuling *et al.,* 2002), and Salts like NaCl, $CaCl_2$, tripolyphosphate, and ammonium hydroxide has been used singly and in combination to enhance beef tenderness (Shuling *et al.*, 2002; Smith, *et al.,* 2008). All researches reported a significant improvement in beef quality. Although several researchers have reported on the use of commercial and local tenderizers in various localities across the globe (Shuling *et al.,* 2002; Bille and Taapopi, 2008; Maiti and Ahlawat, 2011; Kahraman *et al.,* 2012), however, there is a knowledge gap in terms of common tenderizers available for meat processing in Nigeria. Meat processors use plant part and extract that has tenderizing properties for the softening of beef. Some of such are Pawpaw leaf extract, Lime extract, and carbonated drink. Therefore, this research will be evaluating the effects of these locally utilized tenderizers in marinade solutions on beef.

Materials and methods

The experiment was conducted, to compare the effects of four unconventional marinades made of different locally available tenderizers at the meat processing laboratory of the Department of Animal Production and Health, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria. Muscle from the thigh of a freshly slaughtered cattle was harvested and conveyed in a cold container to the laboratory. Also, each marinade was constituted as follows: 100mL of seasoned water, 100mL canola oil and 100mL of tenderizers. Tenderizers in respective treatments are as follows: treatment 1, 100mL of distilled water, treatment 2 contained 100mL of carbonated drink, treatment 3 contained 100mL of lime juice, and treatment 4 contained 100mL of pawpaw leaf extract. Collected data includes the Proximate compositions of fresh beef and marinated beef, weights of fresh beef and marinated beef, pH of fresh and marinated beef, percentage loss from beef after refrigeration and cooking, and sensory scores of beef after cooking.

Marination of beef

A chunk of meat weighing 1500 g was excised from the bulk of muscle by cutting along the fat marbling. The excised beef was cut into twelve (12) pieces of the same size (10 X 6 cm) and thickness (1.5 – 2.0 cm) weighing approximately 125 g. Each of

the pieces of meat were placed in a polypropylene box. The boxes with the beef were randomly divided into four groups with each treatment group being in triplicate. A 300 ml marinade was poured into each of the boxes such that the pieces of meat were completely submerged. The type of marinade for each box will be determined by the treatment group it belongs. Thereafter, the marinade mixture was agitated using a stirring rod ensuring even distribution of the solute. All boxes were over-wrapped with a polyethylene cover and held at 4°C for 24 hrs inside a refrigerator. After 12 hrs, the meat samples were turned over, to ensure uniform marination. Thereafter, the meat samples were removed from the boxes and excess liquid allowed to drain off for 5 min at 4°C.

Experimental procedures

Proximate composition of fresh and marinated beef samples were determined according to the methods outlined by the Association of Official Analytical Chemists (AOAC, 2010). Also, pH meter was first calibrated before use. It was switched on and allowed to stabilize for a period of five minutes. The pH meter was standardized with buffer solutions with known pH values of pH4, pH7, pH9 to ensure sensitivity and accuracy of the meter. The pH of the samples was taken by the insertion of the Xerolyte electrode (JENWAT 3015 pH meter) into the incision to take readings. The electrode was immersed in distilled water to prevent the carry-over effect. Also, the pH of marinades from each treatment was determined using the same procedure. Furthermore, the marinated meat samples from each replicate across the experimental treatments were labelled and weighed on a sensitive scale. Samples from each treatment was put in plastic bags and cooked in a Uniscope laboratory water bath at 70 °C for 20 mins. The cooked weight of samples was taken after allowing samples to cool at room temperature for 30 min. Cooking loss percentage was determined as the difference between pre-cooked and post-cooked weights and divided by pre-cooked weights of meat multiplied by 100 (Sanwo *et al.*, 2011). Also, marinated meat samples across the experimental treatments were labelled and weighed. The weights were taken using a sensitive scale before and after refrigeration. Refrigeration temperatures were at 4°C for 24 h (Sanwo *et al*., 2011). Furthermore, a sensory evaluation was conducted immediately after cooking. All the cooked samples (12 in number) were evaluated in one session. Ten untrained panellists were used in the assessment procedure. Each panellist was seated at his/her table with a bottle of clean water and twelve samples of the experimental beef were served in turns. They were instructed to chew a small slice of the experimental beef samples and score it for colour, flavour, texture, juiciness, and tenderness. They rinsed their mouth with the bottled water after scoring each sample to remove carryover effects. The panellists scored each sample for the mentioned qualities (colour, flavour, texture, juiciness, and tenderness) on a nine-point hedonic scale (9 = like extremely, 8 = like very much, 7 = like moderately, 6 = like slightly, 5 = neither like nor dislike, 4 = dislike slightly, 3 = dislike moderately, 2 = dislike very much and 1 = dislike extremely). Averages of each panellist scores for each beef quality were subjected to statistical analysis (Sanwo *et al*., 2011).

Statistical analyses

All laboratory data obtained were arranged in a Completely Randomized Design and analyzed using analysis of variance (ANOVA) with the general linear model of (SPSS 1999). While sensory scores were arranged in a Randomized Complete Block Design and analyzed using the Linear

Mixed Models (LMM) so as to remove the panellist effect. Significant means were separated using Duncan multiple range test of SPSS software. Probability level of $P<0.05$ was considered for significance in all the mean comparisons.

Experimental model

Completely Randomized Design

$Y_{ij} = \mu + M_i + E_{ij}$

Where:

Y_{ij} = Observed value of beef quality

μ = Population mean

M_i = Effect of the i^{th} marinade (i.e. Control, Pawpaw leaf, Lime juice, or carbonated drink)

E_{ij} = Random error

Randomized Complete Block Design

$Y_{ijk} = \mu + M_i + B_j + E_{ijk}$

Where:

Y_{ijk} = Observed sensory score

μ = Population mean

M_i = Effect of the i^{th} marinade (i.e. Control, Pawpaw leaf, Lime juice, or carbonated drink)

B_j = Randomized effect of the j^{th} panellist

E_{ijk} = Random error

Results and discussion

Chemical composition of fresh beef before marination

Table 1 shows the chemical composition of fresh beef before marination. Beef used for the experiment were very lean as percentage fat content was between 2.88% to 3.05% which was lower numerically when compared with values of 5.95% reported by Daniela *et al.,* (2011) who marinated beef using the biceps femoris muscle. This could be due to differences in nutrition and excised animal parts used from the respective cattle. However, moisture content and protein content ranges from 76.767% - 77.870% and 17.637% - 18.530% respectively, which were within range 76.1% and 17.5% respectively reported by Daniela *et al*. (2011).

Table 1: Chemical composition of fresh beef before marination

Parameters	Control (Water)	Carbonated drink	Lime-juice	Pawpaw leaves extract	SEM
Moisture (%)	78.19±0.12	77.57±0.67	78.35±0.25	77.96±0.64	0.226
Fat (%)	2.92±0.01	3.05±0.05	2.88±0.04	2.97±0.08	0.031
Ash (%)	1.09±0.03	1.19±0.02	1.01±0.04	1.13±0.04	0.019
Crude Protein (%)	17.81±0.08	18.53±0.32	17.68±0.17	18.26±0.65	0.192
Iron (%)	0.37±0.01	0.34±0.00	0.36±0.00	0.55±0.18	0.046
Calcium (%)	0.12±0.00	0.12±0.00	0.13±0.00	0.12±0.00	0.000

SEM = Standard Error of Mean

Chemical composition of marinated beef

Table 2 shows the chemical composition of marinated beef. There was significant ($P<0.05$) rise in the percentage fat content of beef after marination across all experimental samples. This might be a result of the absorption of oil from the marinades; which contributes to the increase in fat content in all beef samples. Samples soaked in lime juice and carbonated drink absorbs the most of oil compared to other marinades. This is due to the acidic media provided by both marinades. Similar phenomenon was observed by Raichlen (1992) who soaked fish in marinade containing citrus juice to enhance oil absorption.

Table 2: Chemical composition of marinated beef

Parameters	Control (Water)	Carbonated drink	Lime-juice	Pawpaw leaves extract	SEM
Moisture (%)	76.85±1.77	69.82±3.95	70.64±0.65	75.88±1.43	1.297
Fat (%)	2.99±0.12^{b}	3.98±0.50^{a}	3.87±0.07^{ab}	3.13±0.11^{ab}	0.174
Ash (%)	1.20±0.06	1.50±0.17	1.46±0.03	1.18±0.06	0.062
Crude Protein (%)	18.92±0.80	24.72±3.14	24.01±0.56	19.80±0.65	1.065
Iron (%)	0.31±0.01^{ab}	0.33±0.00^{a}	0.31±0.00^{ab}	0.30±0.00^{b}	0.003
Calcium (%)	0.11±0.01	0.11±0.00	0.11±0.00	0.11±0.00	0.002

^{a,b} mean values within the same rows with the same superscript are not significantly different at 5% level
SEM = Standard Error of Mean

pH and Weights of marinades and beef

Table 3 and 4 shows the initial and final values of weights and pH from beef and marinades respectively. The Final beef pH was significantly (P<0.05) least in lime-juice based marinade 4.32 compared to other marinated beef whose pH falls between 5.32 and 5.60. This is due to the citric acid composition of lime (Ganguly, 2013). Also, during marination, there was a significant (P<0.05) reduction in beef pH. This is because beef pH has not yet attained the optimum pH values (5.3-5.7) at the beginning of the experiment as the initial pH for control marinade was 6.40 while final pH was 5.34. This is similar to the findings of Kaharman *et al*. (2012) who soaked beef in citrus juice marinade and reported a pH of 5.5 but different from the findings of Laima *et al*. (2013) who soaked beef and venison in red wine marinades and reported a slight pH rise from 5.1 to 5.2 in the course of marination. This variation might be due to the difference in pH of the marinade used. There were significant (P<0.05) variations in the pH of respective marinades before and after marination. This may be due to the differences in the pH of the tenderizers in the marinades. There were significant differences (P<0.05) in the marinades' weights. This is due to the dissimilarity in the weights of the tenderizers used in the preparation of the respective marinade. After marination, significant differences (P<0.05) in marinades' final weights were due to the absorption of marinades through osmotic process and diffusion of ions out of the beef. Furthermore, Beef soaked in slightly acidic marinades (control and Pawpaw leaves extract) absorbed significantly (P<0.05) more marinades compared to other marinades which were more acidic (Lime and carbonated drink). This is because acidic media induce protein denaturation (Shuling *et al*, 2002), resulting in decreased water binding ability of myosin, actin and other myofibrillar components of meat thus, reducing the absorption of marinades. This low marinade absorption by beef in acidic marinades agrees to results obtained by Kaharman *et al*. (2012) who soaked beef in citrus juice marinade with a starting pH of 5.6 and final pH of 4.4.

Table 3: Weights and pH of beef before and after marination

Parameters	Control (Water)	Carbonated drink	Lime-juice	Pawpaw leaves extract	SEM
Initial Weight (g)	124.33±1.75	127.77±1.47	125.17±2.45	125.40±0.49	0.819
Final Weight (g)	135.17±8.15^{a}	119.03±1.34^{ab}	115.23±4.79^{b}	135.07±1.77^{a}	3.437
Initial pH	6.40±0.00	6.20±0.00	6.20±0.00	6.50±0.00	0.392
Final pH	5.34±0.04^{a}	5.32±0.14^{a}	4.32±0.06^{b}	5.60±0.16^{a}	0.155

^{a,b} mean values within the same rows with the same superscript are not significantly different at 5% level
SEM = Standard Error of Mean

Table 4: Weights and pH of marinades before and after marination

Parameters	Control (Water)	Carbonated drink	Lime-juice	Pawpaw leaves extract	SEM
Initial Weight (g)	299.10±0.32^{bc}	302.97±4.21^{b}	329.80±1.93^{a}	291.70±2.26^{c}	4.475
Final Weight (g)	287.50±6.25^{c}	310.63±6.07^{b}	338.10±4.60^{a}	281.20±4.14^{c}	7.179
Initial pH	6.10±0.00^{b}	4.00±0.05^{c}	3.00±0.00^{d}	6.43±0.06^{a}	0.432
Final pH	5.92±0.03^{b}	5.33±0.06^{c}	4.51±0.02^{d}	6.06±0.03^{a}	0.186

^{a,b,c,d} mean values within the same rows with the same superscript are not significantly different at 5% level
SEM = Standard Error of Mean

Cooking and refrigeration loss

Tables 5 and 6 show respectively the cook and refrigeration loss of marinated beef. There were no significant differences (P>0.05) in the cooking losses, refrigeration losses, cooking loss percentages and refrigeration percentage losses of all experimental marinades. This is in agreement with the findings of Barbantia and Pasquini (2005) who reported no significant effect of marinades on cook losses of chicken breast. These insignificant losses among marinated beef might mean that the tenderizer used in the marinades did not adversely disrupt the structure of the cells making up each muscle fibre; hence, causing no loss in cell content.

Table 5: Cooking loss and cooking loss percentages of marinated beef

Parameters	Control (Water)	Carbonated drink	Lime-juice	Pawpaw leaves extract	SEM
Initial Weight (g)	25.00±0.46	25.87±2.69	22.43±2.34	25.30±2.14	0.979
Final Weight (g)	16.93±1.73	18.03±2.06	14.30±0.90	17.53±1.60	0.820
Cook loss (g)	8.07±2.15	7.83±0.71	8.13±1.45	7.77±0.60	0.602
Cook Loss (%)	32.00±8.18	30.45±1.36	35.73±2.51	30.69±1.97	2.001

SEM = Standard Error of Mean

Table 6: Refrigeration loss and refrigeration loss percentages of marinated beef

Parameters	Control (Water)	Carbonated drink	Lime juice	Pawpaw leaves extract	SEM
Initial Weight (g)	15.80±2.74	14.00±1.31	17.77±5.17	20.13±1.13	1.473
Final Weight (g)	15.57±2.61	13.87±1.29	17.43±5.10	19.80±1.10	1.439
Refrigeration Loss (g)	0.23±0.13	0.13±0.03	0.33±0.06	0.33±0.03	0.041
Refrigeration loss (%)	1.30±0.53	0.95±0.20	2.03±0.27	1.65±0.06	0.181

SEM = Standard Error of Mean

Sensory scores

Table 7 shows the sensory scores of marinate and cooked beef. Colour score of beef soaked in pawpaw leaves extract marinade was significantly (P<0.05) lower 4.37 than the other marinades. Pawpaw scored low because of the chlorophyll from leaves which made the beef unattractive. This contrast with the findings of Islam and Molinar-Toribio (2013) who used pawpaw peel at 30% and 60% Kg weight for beef softening. The difference might be due to the presence of green pigment on the beef after marination. However, beef soaked in a carbonated drink had highest (P<0.05) score for colour. This is because a

carbonated drink used is colourless; hence, does not alter the colour of beef. Beef soaked in lime juice-based marinades showed intermediate colour score. This is due to its pale look as a result of its high pH after marination. Furthermore, beef soaked in lime juice-based marinade was less (P<0.05) tender and juicy, compared to other marinades. This is contrary to the report of Burke *et al*. (2003) who soaked shin beef into 31% lemon juice and found an increase in tenderness and juiciness of the marinated beef. The difference may be as a result of the lower percentage of lime used in this experiment which results to lesser proteolysis in the marinated samples thus, decreasing water binding ability and solubility of myosin, actin, and all other myofibrillar components in the beef (Shuling *et al*., 2002). However, there were no significant changes (P>0.05) in the meaty flavour and saltiness of beef after marination. From the result, pawpaw leaves marinade significantly (P<0.05) produced the most tender beef. This is because of the aggressiveness of the enzyme, papain which causes significant degradation of both myofibrillar and collagen protein in meat (Ashie *et al*. 2002). Also, carbonated drink based marinades significantly (P<0.05) produced beef with the best flavour among the experimental marinades while pawpaw leaves extract-based marinade produced beef with the poorest flavour. This agrees with the work of Islam and Molinar-Toribio (2013) who reported pawpaw treated beef having poor flavour and bitter taste after marination. However, beef soaked in carbonated drink based marinade was significantly (P<0.05) the most acceptable among the beef from other marinades.

Table 7: Sensory scores of marinated and cooked beef

Parameters	Control (Water)	Carbonated drink	Lime-juice	Pawpaw leaves extract	SEM
Colour	6.148±0.305^{a}	6.385±0.283^{a}	5.808±0.272^{a}	4.778±0.382^{b}	0.167
Juiciness	6.185± 0.302^{a}	6.115±0.413^{a}	4.538±0.465^{b}	6.148± 0.353^{a}	0.202
Meaty Flavour	6.630±0.325	6.385± 0.368	5.731± 0.390	5.462±0.408	0.190
Tenderness	6.889±0.258^{b}	4.692±0.350^{c}	4.115±0.424^{d}	7.630±0.257^{a}	0.215
Saltiness	2.852±0.260	3.769± 0.279	3.846±0.302	3.074±0.392	0.160
Overall Flavour	5.519±0.363^{ab}	6.192±0.364^{a}	5.192±0.314^{b}	3.963±0.445^{c}	0.201
Overall Acceptability	5.889 ± 0.393^{a}	6.720±0.329^{a}	5.500±0.369^{a}	3.593±0.466^{b}	0.225

^{a,b,c,d} mean values within the same rows with the same superscript are not significantly different at 5% level
SEM = Standard Error of Mean

Conclusion

The study showed that pawpaw leave extract produced the tenderest beef. However, it is scored badly on colour, flavour and overall acceptability. Therefore, it could be concluded that the best beef quality can be obtained by soaking it in carbonated drink-based marinades as it produced beef with the least refrigeration and cook loss, and the best overall acceptability concerning sensory test after marination compared with other marinade treatments.

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