

## Comparative Study on the Yield, Amino Acid Profile and Cowhide (*Ponmo*) Tenderization Efficiency of Papain from Different Parts of Papaya (*Carica papaya*)

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(Received January 7, 2025; Revised February 10, 2025; Accepted February 13, 2025)

**Abstract:** This study evaluated the yield, amino acid profile, and cowhide (*Ponmo*) (tenderization efficiency of papain from different parts of papaya (*Carica papaya*). The fresh leaves and fully grown but unripe pawpaw fruits were harvested from 1<sup>st</sup> to 7<sup>th</sup> August 2024. Papain was extracted from the peels, leaves, and seeds of papaya and purified. *Ponmo* from adult cattle was obtained from commercial producers after slaughtering and subjected to enzyme treatment, evaluation of amino acid profile, and meat tenderness. The highest yield of purified papain was obtained from the papaya seed (50.1%), while the lowest was obtained from the leaves (38.8%). *Ponmo* treated with papain from papaya peel recorded the highest values for all the essential amino acids and non-essential amino acids except for glycine (5.10%) and proline (5.18%), which were high in *Ponmo* treated with papain from papaya seed and control (animal fed with untreated diet) respectively. The shear force values of all the treated samples were significantly ( $p < 0.05$ ) lower than the control sample (5.41 kg/cm<sup>3</sup>). The study has clearly shown that when you treat *ponmo* with papain, more beta-pleated bonds are broken down, the meat becomes more tender and essential amino acids become more available and quantifiable than the control (untreated *ponmo*).

**Keywords:** Ponmo; papain; papaya; amino acid profile; tenderness. © 2025 ACG Publications. All rights reserved.

### 1. Introduction

Meat is regarded as the most valued product from livestock, and it serves as the preferred choice of animal protein by many people. Meat is either consumed as a home-made delicacy or as processed products [1]. The quality of meat is generally considered based on the intended usage where meat and meat products' values are determined based on various factors, which include social, cultural, ethical, symbolic, nutritional, functional, and organoleptic factors [2].

*Ponmo* (cowhide) is consumed as an edible product from animal sources, which is obtained from the skin (hides) of large animals like camels, cattle, and buffaloes [3]. The skin of animals is removed alone or together with meat and is prepared and consumed as a delicacy in Nigeria [4]. In Nigeria,

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## Comparative study on papaya parts' papain yield, amino acids, tenderness

cowhide is generally preferred over other animal skin, and it is commonly referred to as Akpukpo anu by Igbo people, ponmo by Yoruba people, kanda by the Northerners, Ano by Igala people, and Ohian by the people of Edo [5]. *Ponmo* consumption is no longer regarded as a meal for the poor people in Nigeria, where its consumption was predominant amongst the poor and uneducated Yoruba within the southwestern part of Nigeria, but it is now consumed by all classes of people [6]. Processing of *Ponmo* from their raw state to the final stage of consumption involves different processes. These processes include the removal of the animal skin (skinning), cutting, singeing, washing, and preservation [7]. Hides meant for consumption are obtained in larger quantities from abattoirs all over Nigeria, especially in the Northern parts of Nigeria where the majority of these animals are being reared [8].

Cowhide is made up of collagen. According to Lawrie [9], collagen is made up of three (3) polypeptide chains that are joined together to form X-helix-like 3 strands, folded and bonded together by hydrogen bonding. It is a sequential chain of amino acids that are folded and bound to form a very strong and fibrous molecular structure. He also stated that the age of the animal affects the quality of the meat product. As the animal grows older, the cross-linkages in the collagen chains increase, thereby making the meat tougher.

Tenderness has been regarded as the factor of most importance when considering consumers' satisfaction and their views on the meat's taste. The length of the sarcomeres, as well as the integrity of the myofibril structure that influences the actomyosin toughness, is known to affect meat tenderness [10]. Consumers prefer meat that is tender. Therefore, the production of meat with good tenderness is required to maintain consumers' confidence in quality meat, especially over other types of meat that have no toughness problems [11]. Consumers' preference for meat is majorly influenced by the eating quality of the meat as well as the sensory properties, which include flavor, meat color, juiciness, texture, water holding capacity, and cooking losses [12].

The quality of meat can be improved by chemical and/or physical treatment or treatment with proteolytic enzymes, which are one of the most recognized methods for meat tenderization. Proteolytic enzymes like papain obtained from plant sources have been used mostly as meat tenderizers worldwide, and they are used in many industries for commercial purposes [13]. United States federal agencies generally regard papain, bromelain, *Aspergillus oryzae* protease, ficin, and *Bacillus subtilis* protease as safe for meat tenderization [10].

Meat tenderness is an important quality trait critical to consumer acceptance, and determines satisfaction, repeat purchase and willingness-to-pay premium prices (Robyn *et al.*, 2021). Tenderness depends on the structural integrity of the myofibrils and of connective tissue which surrounds the muscle fibers and on their properties. The contribution of connective tissue to the secondary toughness of meat is dependent on the quantity, type and intermolecular cross-links of collagen which is the main component of connective tissue. By formation of the cross-links between the collagen molecules, the meat of old animals becomes harder. Cowhides (*Ponmo*), a rich source of protein, are composed of a complex structure consisting of both alpha-helical and beta-pleated arrangements. The alpha-helical structure, characterized by a spiral conformation, provides elasticity and flexibility to the hide. In contrast, the beta-pleated structure, comprising a sheet-like arrangement, contributes to the hide's strength and toughness. However, this tough structural complexity renders *Ponmo* resistant to breakdown, hindering the release and bioavailability of essential amino acids. As a result, the nutritional potentials of ponmo remains underexploited, and their consumption may not provide the expected health benefits. Moreso, due to the toughness of the meat, it takes a longer time to be cooked properly. The use of papain in treating ponmo is a strategic approach to break down robust beta-pleated structures that impede the bioavailability of amino acids. Papain, a cysteine protease, has been widely recognized for its ability to hydrolyze proteins and break down complex structures. By treating *Ponmo* with papain, the beta-pleated structures can be effectively degraded, releasing amino acids and making them more bioavailable for utilization.

## 2. Materials and Methods

### 2.1 Materials

*Ponmo* from adult cattle was obtained from commercial producers after slaughtering at a Mami-approved abattoir in the Abakpa military barracks in Enugu State, Nigeria. The fresh leaves and fully grown but unripe pawpaw fruits of a known variety (Obukpa) used for this experiment were harvested from 1<sup>st</sup> to 7<sup>th</sup> August 2024. The fresh leaves and unripe mature pawpaw fruits used in this study were sourced from local farms in Enugu State. The botanical identification of the plant materials was carried out at the Enugu State Ministry of Agriculture, where a team of experienced botanists and agricultural experts verified the authenticity of the samples. The identification process involved a thorough examination of the morphological characteristics of the leaves and fruits, including their shape, size, color, and texture. Based on this identification process, the fresh leaves and unripe but fully matured pawpaw fruits were confirmed to be authentic samples of *Carica papaya*. The samples were sorted and washed properly with clean water to remove surface dirt. The papaya peels were manually removed with a kitchen knife, and the fruits were longitudinally cut to remove the seeds (see Figure 1-6).

### 2.2 Extraction and Purification of Papain

The method of Mahmood *et al.* [14] was employed for the extraction of papain. The fresh leaves, seeds, and peels were dried at 55°C in an oven (model: KZ 760 4SS China) to a final moisture content of 5%. The samples were grounded using a mechanical grinder {model: BLG-595(MK2) China}. Five (5) g of ground papaya leaf, seed, and peel powder were dissolved in 20 mL of distilled water, and the extract was filtered through filter paper.

Purification of papain was done as described by Sarote *et al.* [15]. The filtered samples were mixed with 40 mM cysteine at a ratio of 3:1(w/v), and the pH of the suspension was adjusted to 5.6 using 6 M HCl. The mixture was stirred for 15 min at 4 °C and filtered. The pH of the filtrate was adjusted to 9.0 using 6 M NaOH. The filtrate was centrifuged at 10000 rpm for 30 min at 4 °C, and the supernatant was precipitated with ammonium sulphate at 45 % saturation. The salt-enriched solution was incubated at 4°C for 30 min. The precipitation was centrifuged at 10000 rpm for 30 min and dissolved using 20 mM Cysteine. Sodium chloride (10% w/v) was added to the solution and kept at 4 °C. The mixture was stirred slowly for 30 min and centrifuged at 10000 rpm for 30 min. The enzyme papain was dissolved in water and stored at 4 °C.



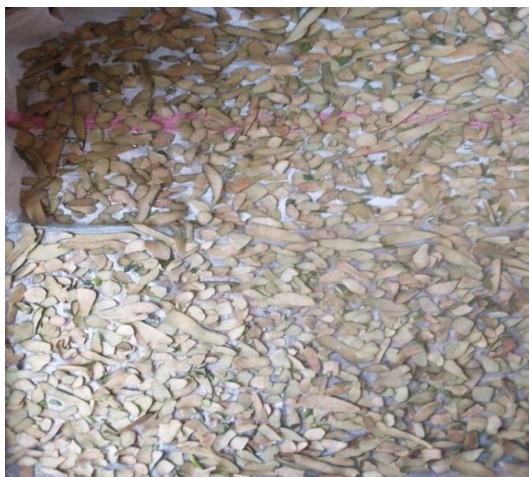
**Figure 1.** Obukpa papaya leaves



**Figure 2.** Obukpa papaya seeds



## Comparative study on papaya parts' papain yield, amino acids, tenderness



**Figure 3.** Obukpa papaya peels



**Figure 4.** Obukpa papaya fruits variety



**Figure 5.** Extracted papain from Obukpa papaya variety



**Figure 6.** Kpmo meat from adult cattle

### 2.3 Kpmo Preparation

The method described by Ionescu *et al.* [16] was used. The kpmo meat (animal skin), 24 hours after slaughter, was divided into equal pieces, having the approximate weight of 100 g. The pieces of meat were divided into 4 groups; each group, having at least 5 pieces, was treated in a different way:

Sample A - the blank sample served as the control, free from enzyme treatment

Sample B – the meat was injected with 10 mg of papain extracted from the leaves of *Carica papaya*

Sample C – the meat was injected with 10 mg of papain extracted from the seed of *Carica papaya*

Sample D – the meat was injected with 10 mg of papain extracted from the peels of *Carica papaya*

Sample E – casein will be used to feed the rats

The injection of 10 mg of papain was manually made with a single needle to each group of the meat. After the injection of papain, they were packed and stored at a temperature of 4°C for 24 hours. After the ageing, each of the group samples were taken to a closed experimental tube and boiled in a water bath until the samples attained a temperature of 70°C. At this temperature, the samples were allowed to boil for 10 minutes. After boiling, the samples were cooled and then taken out from the experimental tubes for further analysis.

### 2.4 Determination of Amino Acid Profile

Determination of amino acid profile was done using ion-exchange chromatography (IEC) as described by Ibegbulem and Belonwu [17]. The samples were first defatted before carrying out acid digestion. After digestion, the samples were taken to the amino acid analyzer. The samples were defatted using the standard methods of AOAC [18]. Six grams (6 g) of the minced samples were weighted and taken into an extraction thimble. Lipid soluble matter extraction was done using chloroform/methanol

(2:1; v/v) mixture in a Soxhlet extraction apparatus. After extraction, 4 g of the samples were transferred into a glass ampoule and 8 mL of 6 N HCl was added it. Oxygen was removed from the sample/acid mixture by passing gaseous nitrogen into the glass ampoule. After that, the ampoule was sealed over a Bunsen burner flame before transferring into an oven and allowed to stand for 22 h at 105°C. After that, the samples were allowed to cool and filtered using Whatman No 52-filter paper. The filtrate was taken to hot air oven and evaporated to dryness. The residue was dissolved in 5 mL acetate buffer (pH = 2.0) and stored in a plastic tube at a temperature of -4°C until used for analyses. Ten (10  $\mu$ L) volume of the digest was displayed into the cartridge of High-Performance Liquid Chromatography (HPLC) (Column: ninhydrin column, mobile phase: Water with a gradient of sodium acetate, flow rate: 1.0-1.5 mL/min, injection volume: 10-50  $\mu$ L, column temperature: 30-40°C. Calibration Standards: amino acid standard mixture concentration range: 0.1-1.0 Mm) and amino acids determined by the HPLC based on the color reaction of amino acids with the oxidation agent ninhydrin.

### 2.5 Determination of Shear Force

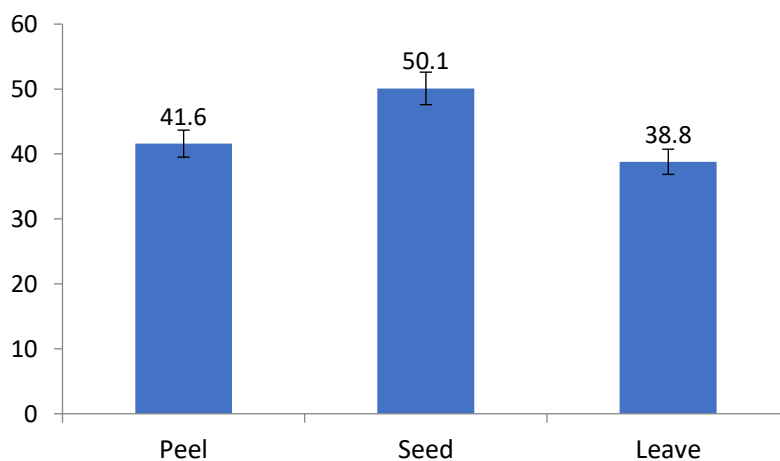
Warner–Bratzler shear force device (capacity 25 kg X 50 GMS, Salter Brecknell, Fairmont, Minnesota, USA) was used to measure cooked *Ponmo* tenderness according to the method of Nicola *et al.* [19]. A piece was cut off from the outside of the meat so that the direction the muscle fibers are lying in the meat can be identified. Three meat samples were taken from each group sample and cut exactly to specification; 10 mm x 10 mm cross section, and a length, parallel to the fiber axis, of at least 25 mm and placed into the ‘V’ shaped blade. The samples were perpendicularly sheared to the muscle fiber at three places, and the force at which the samples were sheared was recorded and averaged.

### 2.6 Data Analysis

Analysis of variance was used for the determination of significant differences ( $p < 0.05$ ) among treatment means, and separation of means was carried out using the SPSS version 20.0. Separation of means would be carried out by Duncan's Multiple range test, and values would be reported as means and standard deviation.

## 3. Results and Discussion

The yields of the purified papain from different parts of *papaya* are shown in Figure 7. The percentage crude yield ranged from 15.76 – 21.20%. The highest yield of crude papain was obtained from the seed (21.20%), while the lowest yield was obtained from the peel (15.76%). The variation in the crude yield may be due to the differences in the concentration of papain from the different parts of the *papaya*. The crude yield varies from the ranges of 2.92 - 24.87% for the local variety and 1.60 - 14.67% for the California variety, as reported by Diah *et al.* [20] for papain enzyme from papaya fruits of the California variant and the Indonesian local variant.



**Figure 7.** Extraction yield of pure papain from the 500 g sample

## Comparative study on papaya parts' papain yield, amino acids, tenderness

The concentrations of essential and non-essential amino acids are shown in Table 1. Histidine ranged from 2.04 – 2.68%, isoleucine ranged from 2.80 – 3.69%, leucine ranged from 5.35 – 6.79%, lysine ranged from 5.02 – 5.68%, methionine ranged from 2.16 – 2.58%, phenylalanine ranged from 5.49 – 6.27%, tryptophan ranged from 0.19 – 0.32%, valine ranged from 2.58 – 5.24%, threonine ranged from 3.80 – 5.03% and arginine ranged from 3.84 – 5.85%. *Ponmo* treated with papain extracted from the papaya peel had the highest values for all the essential amino acids. This could be due to the specificity of papain from *papaya* peel. Papain from *papaya* peel might have a higher specificity for breaking down proteins into amino acids, resulting in higher amino acid values. This is followed by *Ponmo* treated with papain extracted from the papaya seeds. The lowest percentage of essential amino acid was obtained from tryptophan for all the samples. There were significant ( $p < 0.05$ ) differences among all the samples for the essential amino acids except for methionine and threonine where no significant ( $p > 0.05$ ) differences existed between samples treated with papain from papaya leaf and the control; and samples treated with papain from papaya seed and the control respectively. These values were higher than the findings (0.88% for arginine), (0.41% for histidine), (0.61% for isoleucine), (1.00% for leucine), and all the essential amino acids reported by Hannah *et al.* [21] for cooked beef but similar to the findings reported by Jorfi *et al.* [22] for all the essential amino acids tested on beef, mutton, chevon, chicken and pork and Islam *et al.* [23] for essential amino acid profile of *Longissimus dorsi* muscle of indigenous cattle.

The results of the non-essential amino acids showed that alanine ranged from 5.57 – 8.90%, aspartic acid ranged from 4.63 – 6.34%, glycine ranged from 3.68 – 5.10%, cysteine ranged from 5.16 – 6.09%, ornithine ranged from 0.08 – 0.14%, serine ranged from 2.44 – 1.68%, tyrosine ranged from 0.19 – 0.32%, glutamic acid ranged from 11.35 – 17.67%, pyrolysine ranged from 0.69 – 0.99% and proline ranged from 4.23 – 5.18%. *Ponmo* treated with papain extracted from the papaya peel had the highest values for all the non-essential amino acids except for glycine obtained from *Ponmo* treated with papain extracted from the papaya seed and proline obtained from the control. The lowest percentage of non-essential amino acid was obtained from ornithine for all the samples. No significant ( $p > 0.05$ ) differences existed between samples treated with papain from papaya leaf and the control for ornithine and glutamic acid and samples treated with papain from papaya seed and peels for proline. The results from these findings showed that *Ponmo* is very rich in glutamic acid with a range between 11.35 - 17.67%. The values obtained from this study vary from (5.70% for alanine), (5.40% for glycine), (3.64% for serine), (4.23% for proline), (8.58% for aspartic acid) and (14.3% for glutamic acid) reported by Emmanuel *et al.* [24] for amino acid composition of kilishi - Nigerian (beef jerky) meat and Josef *et al.* [25] for amino acid levels in muscle tissue of eight meat cattle breeds but much higher than the findings reported by Hannah *et al.* [21] for cooked beef and Kamal *et al.* [26] for amino acid profile of sheep meats reared extensively in Morocco. Differences in the values of the essential and non-essential amino acids could be a result of differences in animal age, feeding pattern, genotype, species, meat part, and sex. According to Koutsidis *et al.* [27], a major role is played by the animal sex when checking for both essential and non-essential amino acids. Hollo *et al.* [28] discovered that continuous feeding of the animal with grass/grass silage and concentrate containing linseed supplements caused variations in the levels of some amino acids in beef meat. According to Kamal *et al.* [26], genotype also has a major influence on the value of the meat protein.

Amino acid profile is used as the major factor for the biological value of products that contain protein [29]. A balanced amino acid profile is mainly used to determine the values of meat as a product rich in protein. [30]. Evaluation of the biological value of meat is solely dependent on the amino acid profile of a protein [31]. According to FAO/WHO [32], leucine/isoleucine is most ideal at 2.36%. The values obtained from this present study were higher than 2.36%. Therefore, we can suggest that *kpomo* meat is a rich source of these essential amino acids when consumed as a source of protein in food. At a high level of leucine imbalance in the diet, niacin metabolism is impaired, leading to niacin deficiency amongst sorghum consumers [33]. Mechanistic target of rapamycin can be activated by high levels of leucine in beef meat, signaling a pathway to stimulate the synthesis of protein and ameliorate sarcopenia [34]. Amino acids like alanine, glutamic acid, and glycine, which are important nutrients in protein-rich meals, are responsible for the meaty flavor of meat products [35]. High glutamic acid contents in beef

play an important role as protein synthesis stimulators, main metabolic fuels, immune response modulators, and epithelial integrity and small intestine function regulators [36-37].

**Table 1.** Amino acid profile of the ponmo treated with papain from different *papaya* parts

Amino acid profile	Seed (%)	Peel (%)	Leaf (%)	Control (%)	FAO/WHO/UNU standards (%)
<b>Essential</b>					
Histidine	2.59 <sup>b</sup> ±0.01	2.68 <sup>a</sup> ±0.02	2.14 <sup>c</sup> ±0.01	2.04 <sup>d</sup> ±0.02	1.1
Isoleucine	3.38 <sup>b</sup> ±0.04	3.69 <sup>a</sup> ±0.01	3.08 <sup>c</sup> ±0.05	2.80 <sup>d</sup> ±0.14	2.8
Leucine	6.06 <sup>b</sup> ±0.01	6.79 <sup>a</sup> ±0.06	5.44 <sup>c</sup> ±0.03	5.35 <sup>d</sup> ±0.02	6.6
Lysine	5.57 <sup>b</sup> ±0.02	5.68 <sup>a</sup> ±0.04	5.02 <sup>d</sup> ±0.02	5.22 <sup>c</sup> ±0.03	5.8
Methionine	2.40 <sup>b</sup> ±0.01	2.58 <sup>a</sup> ±0.04	2.23 <sup>c</sup> ±0.01	2.16 <sup>c</sup> ±0.04	2.5
Phenylalanine	6.13 <sup>b</sup> ±0.01	6.27 <sup>a</sup> ±0.02	5.79 <sup>c</sup> ±0.03	5.49 <sup>d</sup> ±0.01	6.3
Tryptophan	0.26 <sup>b</sup> ±0.01	0.32 <sup>a</sup> ±0.01	0.19 <sup>d</sup> ±0.01	0.23 <sup>c</sup> ±0.01	0.5
Valine	4.14 <sup>b</sup> ±0.01	5.24 <sup>a</sup> ±0.01	3.29 <sup>c</sup> ±0.04	2.58 <sup>d</sup> ±0.02	3.5
Threonine	4.49 <sup>b</sup> ±0.02	5.03 <sup>a</sup> ±0.11	3.80 <sup>c</sup> ±0.14	4.36 <sup>b</sup> ±0.06	3.4
Arginine	5.28 <sup>b</sup> ±0.04	5.85 <sup>a</sup> ±0.07	3.84 <sup>d</sup> ±0.08	4.01 <sup>c</sup> ±0.01	CE
Total	40.3	44.3	34.82	34.24	
<b>Non essential</b>					
Alanine	7.83 <sup>b</sup> ±0.01	8.90 <sup>a</sup> ±0.01	6.07 <sup>c</sup> ±0.01	5.57 <sup>d</sup> ±0.01	NE
Aspartic acid	5.53 <sup>b</sup> ±0.11	6.34 <sup>a</sup> ±0.01	5.09 <sup>c</sup> ±0.01	4.63 <sup>d</sup> ±0.15	NE
Glycine	5.10 <sup>a</sup> ±0.01	5.04 <sup>b</sup> ±0.01	4.07 <sup>c</sup> ±0.01	3.68 <sup>d</sup> ±0.01	NE
Cysteine	1.04 <sup>b</sup> ±0.01	1.12 <sup>a</sup> ±0.02	0.95 <sup>c</sup> ±0.02	0.78 <sup>d</sup> ±0.01	2.5
Ornithine	0.11 <sup>b</sup> ±0.01	0.14 <sup>a</sup> ±0.01	0.08 <sup>c</sup> ±0.01	0.08 <sup>c</sup> ±0.01	NE
Serine	2.57 <sup>b</sup> ±0.01	2.68 <sup>a</sup> ±0.01	2.48 <sup>c</sup> ±0.01	2.44 <sup>d</sup> ±0.02	NE
Tyrosine	5.80 <sup>b</sup> ±0.04	6.09 <sup>a</sup> ±0.02	5.24 <sup>c</sup> ±0.02	5.16 <sup>d</sup> ±0.01	6.3
Glutamic acid	16.48 <sup>b</sup> ±0.01	17.67 <sup>a</sup> ±0.76	12.00 <sup>c</sup> ±0.14	11.35 <sup>c</sup> ±0.07	NE
Proline	5.08 <sup>b</sup> ±0.01	5.06 <sup>b</sup> ±0.03	4.23 <sup>c</sup> ±0.03	5.18 <sup>a</sup> ±0.01	NE
Total	49.54	53.04	40.21	38.87	

Values show the mean of duplicate analysis and ± standard deviation. Figures with different superscript down the row are significantly different ( $p < 0.05$ ). CE: conditionally essential, NE: not essential

From this study, it was observed that *Ponmo* meat is a very good source of glutamic acid. The Institute of Medicine recommends a daily dietary allowance intake of methionine plus cysteine by 70 kg adult humans from meat, wheat flour, and white rice as 39, 390, and 700 g DM, respectively [38]. The presence of a high amount of carbohydrates in the body caused by consumption of wheat flour or white rice can be converted into fat, therefore contributing to the development of dyslipidemia, obesity, and other metabolic disorders [39]. Intake of *Ponmo*, which contains less fat on a DM basis, plays a major role in supporting the synthesis of protein, improvement of insulin sensitivity, sustaining skeletal-muscle mass, relieving aging-related sarcopenia, and reduction in white fat accretion [34]. All the *Ponmo* samples contain both essential and non-essential amino acids at varying levels of concentration. This will be needed to meet the physiological and nutritional needs of both children and adults.

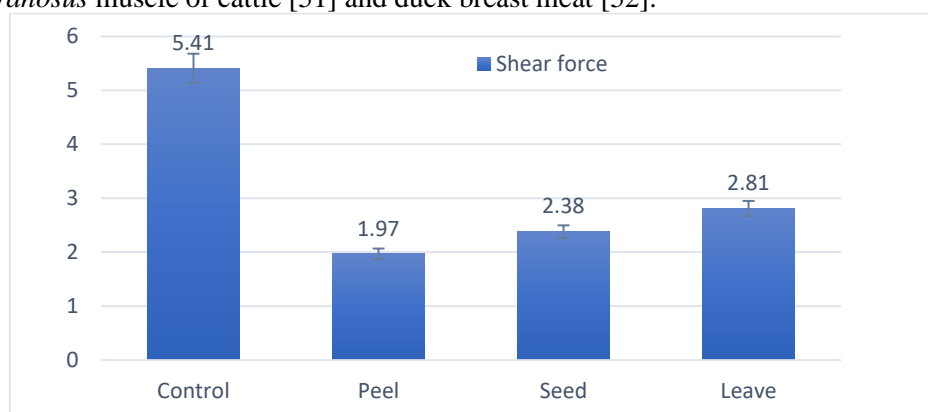
### Comparative study on papaya parts' papain yield, amino acids, tenderness

According to Wu [40], the consumption of food rich in animal protein plays a vital role in optimal human health. More so, syntheses of low-molecular-weight metabolites such as vasodilators, neurotransmitters, signaling molecules, and immune response mediators from the high supply of all amino acids play an important role in the maintenance of physiological homeostasis in humans [39].

Figure 8 shows the tenderness of *Ponmo* treated with papain obtained from different parts of *C. papaya*. One of the main quality attributes of meat is toughness, which is caused by the length of the sarcomere, the presence of intramuscular connective tissue, and intramuscular fat [41]. Meat tenderness can be determined by the use of an objective tool that measures the force needed to shear through a piece of meat (Shear force), with values of lower shear force signifying higher tenderness. Meat tenderness has an important role to play when considering meat-eating qualities such as mouth feel, texture, juiciness, flavor, appearance, and palatability, and it is also among the major important properties of prepared meat [42-43]. According to Ashie *et al.* [44], meat tenderness can be enhanced using the enzyme papain and microbial enzymes.

The shear force values of all the treated samples were significantly ( $p < 0.05$ ) lower than the control sample. The lowest shear force value ( $1.97 \text{ kg/cm}^3$ ) was recorded in samples treated with papain extracted from papaya peels, signifying great tenderness. This is followed by samples treated with papain from the seeds ( $2.38 \text{ kg/cm}^3$ ) and then papain from the leaves ( $2.81 \text{ kg/cm}^3$ ). The higher shear force value ( $5.41 \text{ kg/cm}^3$ ) of the control sample is attributed to the high amount of connective tissue in *Ponmo*. According to Miller *et al.* [45], beef with Warner-Bratzler shear force values recorded above 5.7 is classified as being very tough, while above 3.0 to 4.9 is intermediate and below 3.0 as tender. It could be seen that the control sample falls under the category of tough meat. This could be a result of not being treated with the papain, unlike the other treated samples. The meat tenderness varies from the findings of 0.29 - 3.28 m/L reported by Farouq and Sung [46] for tenderness-related traits and quality properties of spent hen meat affected by adenosine 5' -monophosphate during cold storage. This result agrees with the findings of Akpan and Omojola [47] who reported a reduction in the shear value of beef treated with papain and Abdel-Naeem and Mohamed [48] who also reported a reduction in shear value of camel meat burger patties using ginger extract and papain. A significant reduction in shear force values of buffalo meat treated with papain powder was reported by Neveena *et al.* [49]. Meat tenderness is identified as a vital factor that determines eating satisfaction along with nutritional content and taste [50].

This study revealed that papain improved the tenderness of cowhide (*Ponmo*), overcoming its toughness. This might be due to papain's ability to break up the cross-linkage bonds between actin and myosin, thereby releasing free actin. These major findings agree with previous research on the *semimembranosus* muscle of cattle [51] and duck breast meat [52].



**Figure 8.** Tenderness of *Ponmo* treated with papain from different parts of *C. papaya*

## 4. Conclusions

This study provides a detailed procedure for the extraction and purification of papain from the leaf of pawpaw and unripe but fully matured pawpaw seeds and peels. The highest yield of purified papain was obtained from the papaya seed (50.1%). The study has clearly shown that when you treat *Ponmo* with papain, more beta-pleated bonds are broken down, the meat becomes more tender and



essential amino acids become more available and quantifiable. The treatment made the essential amino acids comparable with the international standards. *Ponmo* treated with papain from pawpaw peel recorded highest values for all the essential amino acids determined and as well as non essential amino acids except for glycine and proline which was high in *ponmo* treated with papain from papaya seed and control respectively. The shear force values of all the treated samples were significantly ( $p < 0.05$ ) lower than the control sample. The higher shear force value ( $5.41 \text{ kg/cm}^3$ ) of the control sample is attributed to the high amount of connective tissue in *Ponmo* meat. This study has shown that *Ponmo* contains both essential and nonessential amino acids at varying levels of concentration.

### Acknowledgments

The authors thank all those who played different roles in seeing that this research work was a success.

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## Comparative study on papaya parts' papain yield, amino acids, tenderness

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