

Beef shank Pineapple vinegar marinade optimization: Relationship between textural and physical characteristics

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ABSTRACT

Shank meat has a tough texture that limited its culinary applications due to its high concentration of connective tissue. The purpose of this study was to determine how marinating beef shank with pineapple vinegar affected its physical characteristics (pH and water-holding capacity (WHC)), texture profile (hardness, springiness, and cohesiveness), and the relationship between both. A completely randomized design with six replications was employed. The treatments consisted of a control, not marinated, and marination in pineapple Vinegar solutions at concentrations of 8% and 16%. Water-holding capacity and hardness of the meat were significantly ($P < 0.01$) reduced with the increase in the concentration of pineapple vinegar. Contrastively, springiness and cohesiveness were not affected ($P > 0.05$). Soaking beef shank with pineapple vinegar has enhanced the quality and tenderness of the meat, and the most favorable concentration is found to be 16%. The association between WHC and meat texture, however, is closer than between WHC and pH. It thus provides useful insights into the utilization of natural meat tenderizers to increase the quality of tougher meat cuts and perspectives for industrial and domestic applications.

Introduction

Each cut of the carcass has unique biochemical properties and compositions (Hwang *et al.*, 2010). The shank is cut from the chuck and back leg of female and male cattle and comprises an estimated 3.3% of the entire carcass weight (Grand View Research, 2019). Extracellular matrix of beef shank contains a rich composition of connective tissues including collagen, elastin, reticulum and mucopolysaccharides, which gives it a tough texture. Collagen is especially required for meat textural characteristics and strongly influences consumer acceptance of shank cuts (Wu *et al.*, 2021; Weston *et al.*, 2002). Moreover, the shank is a low fat cut of meat because it is an active muscle of ambulation which makes the meat tough and coarse (Weston *et al.*, 2002; Gambaro *et al.*, 2023). Such features reduce the culinary use of the shank and make it an undesirable cut of meat, as it is tough and coarse (Gambaro *et al.*, 2023). Usually, beef shanks are ground and got into meat products like tendon meatballs in Indonesia.

The ability of marinating meat to increase its tenderness after death has been widely acknowledged. Marinating tough meat cuts can significantly alter their textural characteristics by promoting proteolysis, breaking down myofibrillar proteins, and increasing their water-holding capacity (Bekhit *et al.*, 2013). Particularly, marinades based on acid have demonstrated promise in tenderizing meat by allowing acidic solutions to enter muscle fibers through osmosis, which denaturates proteins, improves flavor, and increases moisture retention (Lampe, 2015; Septinova *et al.*, 2023). Numerous studies have examined the impact of vinegar, a popular acidic marinade, on the quality of meat (Cumbay, 2008; Gomez-Salaza *et al.*, 2018; Septinova *et al.*, 2023). According to Unal *et al.* (2023) Depending on the vinegar's chemical composition (particularly polyphenol and organic acid content) the potency of the marinating changes.

Texture is one of the most important factors in meat quality, which influences consumers' purchasing decisions and satisfaction while eating (Dar and Light, 2014). So, making tough cuts like that beef shank tender is of the utmost importance. One of the natural tenderizers proposed is pineapple vinegar, which is obtained by pineapple fermentation. Boon-

daeng *et al.* (2022) showed that the acetous fermentation of pineapple sludge produces 7.35 % acetic acid, 3.93-4.10 pH, a total phenolic acid content of 245.31 μg GAE/ml, as well as a significant antioxidant activity of 189.52 μg TE/g. It also has volatile constituents such as ethanol, acetic acid ethyl ester and 2,3-butanediol, which can contribute to its ability to tenderize.

Pineapple vinegar has some good potential but there is few research regarding the use of pineapple vinegar in the quality and tenderness of beef shanks. This study aimed to fill this void by investigating the effect of pineapple vinegar as a natural tenderizer in beef shank.

Materials and methods

Materials

The study utilized 1.8 kg of beef shank cut from Ongole Crossbred cattle, obtained from the Way Laga Slaughterhouse (RPH) in Bandar Lampung, Indonesia. The meat was cut along the muscle grain into pieces weighing approximately 100 g each, with dimensions of 6×4×4 cm. Before marination, the beef shank was refrigerated at a temperature of approximately 4°C for 24 hours. The marination solution consisted of commercially available pineapple vinegar (marketed under the brand "Vinegar") and water.

Method

The experiment was conducted using a completely randomized design (CRD). Shank meat samples were randomly divided into three groups based on the concentration of pineapple vinegar used for marination: 0% (T0 or control no marination), 8% (T1), and 16% (T2). Each treatment consisted of six replications.

Meat marinating procedure

Shank meat pieces were immersed in marination solution according to designed treatment. The weight to volume ratio of shank meat (g) to

marination solution (ml) was maintained at 1: 2. The samples were packed in airtight jars and stored in a refrigerator at a controlled temperature of approximately 4°C for 27 hours.

pH measurement

To measure the pH, 5 g of shank meat samples were weighed and homogenized with 45 mL of distilled water (aquadest). The homogenized mixture was placed in a container, and the electrode of a calibrated pH meter was immersed in the solution. The pH value was recorded directly from the digital display of the pH meter (Septinova et al., 2023). The tool used is the AMT20 Benchtop digital pH meter.

Water holding capacity (WHC) Measurement

The water holding capacity (WHC) was measured following a modification of a method described by Kissel et al. (2009). Meat samples weighing between 0.28-0.32 g were used for analysis. Each sample was placed on a filter paper measuring 5 x 5 cm and sandwiched between two glass plates (25x25 cm). A 10 kg weight was applied to the top glass plate, and the setup was left undisturbed for 5 minutes. Afterward, the sample was reweighed, and the WHC was calculated using the following formula:

$$\text{WHC (\%)} = 100\% - (W_0 - W_1)/W_0 \times 100\%$$

where:

W₀: Initial weight of the sample (g)

W₁: Final weight of the sample (g)

Texture profile analysis

Texture profile analysis was performed to evaluate hardness, springiness, and cohesiveness using a Brookfield Ametek CT3 texture analyzer. The measurement procedure was conducted according to the method described by Brookfield Ametek (2023).

Data analysis

All data were analyzed using analysis of variance (ANOVA) to evaluate the effects of treatments on the measured parameters. Further tests used Duncan's test at the 5% and 1% level. The relationship between pH, water holding capacity (WHC), and texture parameters (hardness, elasticity, and cohesiveness) was assessed using Pearson correlation analysis. Statistical analysis was performed using Microsoft Excel 2016

Results and Discussion

pH value

Table 1 shows that marinating using pineapple vinegar (T1 and T2) has a very significant effect ($P < 0.01$) on the pH of the shank meat. The pH value of the meat at T1 and T2 is lower than at T0 (6.19). The higher the concentration of pineapple vinegar, the lower the pH of the beef shank meat ($P < 0.01$).

The significantly different pH values between treatments indicated that extrinsic factors, such as marinating with pineapple vinegar, are effective in changing the chemical environment of the shank beef pieces. Pineapple vinegar contains several organic acids. The higher the concentration of pineapple vinegar, the more acid ions enter the meat, so that the pH of the beef shank meat is lower or more acidic. The results of this study are similar to Łepecka et al. (2023). Marinating meat with 16% pineapple vinegar causes the pH of beef shank meat to drop below the isoelectric pH. These results are similar to Jones et al. (2019), who found that giving vinegar can reduce the pH of beef from 5.64 to 4.91.

Additionally, Table 1 demonstrates that the beef shank meat's pH at T0 is a relatively high 6.19. Haq et al. (2015) stated that beef has a pH

between 5.47 and 6.24. Additionally, according to Villarroel et al. (2003), premium meat has a pH between 5.5 and 5.6. Because shank meat is a muscle used for movement, it has little glycogen left, which contributes to its high pH value. As a result, the pH of the meat rises, and there is less buildup of lactic acid during the 24-hour aging process when glycogen is converted to lactic acid.

Table 1. Physical characteristics of beef shank.

Parameters	Pineapple vinegar		
	0% (T0)	8% (T1)	16% (T2)
pH	6.19 ± 0.29c	5.40 ± 0.22b	4.84 ± 0.24a
DIA (%)	57.16 ± 3.13b	46.96 ± 1.21a	49.03 ± 5.00a

Different superscripts in the same row indicate a highly significant difference ($P < 0.01$).

Water holding capacity

Marinating beef shank meat in pineapple vinegar solution at concentrations of 8% (T1) and 16% (T2) resulted in WHC that was significantly ($P < 0.01$) lower than the control (T0). However, the WHC values for T1 (46.96 ± 1.21) and T2 (49.03 ± 5.00) did not show any significant difference ($P > 0.05$) (Table 1).

Pineapple vinegar contains organic acids such as citric acid and acetic acid (Boondaeng et al., 2022), which can denature meat proteins by breaking the peptide chains (Zhang et al., 2020). This denaturation, especially in myosin protein, reduces the ability of meat to retain water (Sarker et al., 2021). According to Soeparno (2005), WHC meat reaches its lowest point at isoelectric pH because the number of positive and negative charges in the protein is balanced. In this condition, the space between the myofilaments is very close so that there is less space to retain water.

Marinating shank meat with pineapple vinegar causes the pH of the meat to decrease, which is closer to the isoelectric pH. According to Septinova et al. (2018), when the pH of the meat approaches the isoelectric pH (5.4-5.6), the imbalance between positive and negative electrostatic charges decreases so that the gap between the myofilaments becomes closer and the ability of the meat protein to bind water decreases. The results of this study are similar to Bulgaru et al. (2022), but different from Sarker et al. (2021), which stated that the pH value is opposite to WHC. The lower the pH, the higher the WHC value.

Hardness

Beef shank meat soaked in pineapple vinegar concentrations of 8% (T1) and 16% (T2) significantly ($P < 0.01$) had lower hardness values than the control (T0) (Table 2). Furthermore, Table 3 also shows that the hardness value of the shank meat will decrease with increasing pineapple vinegar concentration ($P < 0.01$). The findings of this study are in line with the research of Sengun et al. (2021), which found that soaking coarse-textured beef in fruit vinegar reduces its hardness.

Meat hardness was decreased using marination with a solution that contains organic acids due to the fiber thickness and diameter reduction (Unal et al., 2023). In this study, myofibrillar proteins and connective tissue in shank meat were broken down by organic acids from pineapple vinegar. This breaks the chain of meat protein and makes the meat softer and easier to cut.

Meat hardness is influenced by the structure of the myofibril, contracted or relaxed, connective tissue content, the amount of cross-linked fibers, and the ability of the proteins to retain water and meat juice (Rahman et al., 2023). Based on this research pointed out, organic acid contained in pineapple vinegar decrease pH on meat, which have an impact toward protein denaturation. This denaturation affects in weakening these bounds until the hardness of the meat are reduced.

It is the protein content that affects the hardness of the meat. Although soluble protein content is a factor that most affects the hardness of meat, myofibrillar protein content has the least effect (Bulgaru et al.,

2022). The hardness two at both T1 and T2 also led to the decrease in hardness. WHC is decreasing (as more free water leaves the muscle tissue), which subsequently makes the meat structure looser and therefore easier to bite.

Springiness

Springiness indicates the elasticity of the material or meat. Springiness is the ability of meat to return to its original condition after the pressure is removed. The smaller the springiness value of the meat, the more chewy the meat is (Paredes *et al.*, 2022). Table 2 indicates that marination with pineapple vinegar had no significant effect ($P > 0.05$) on the springiness of beef shank. But the springiness values were lower in the marinated samples than the control.

From the marination treatments, the average springiness values indicate that beef shank marinated in 30% pineapple vinegar solution was lower compared to the control. This is a small increase in chewiness of beef shank following marination. This is similar to the findings of Unal *et al.* (2023), an acidic solution showed no significant difference in springiness ($p > 0.05$) but a gradual trend toward lowering values with increasing pH.

The reduced springiness effect in this study may have been due to hydrolysis of collagen, elastin, and other connective tissue proteins by organic acids (Skierka and Sadowska, 2007) from pineapple vinegar. These modifications influence the integrity of the connective tissue, causing it to lose its spring when pressure is placed. In addition to the circumstantial evidence of the reduced operating concentration of pineapple vinegar, its acidity (pH 5.4) is capable of influencing muscle myofibrillar proteins that are possibly condition by myosin and actin interactions. Soaking in a vinegar solution (pineapple) acidic solution acts as a guide to the bonds of actomyosin, themselves the main structure of muscle contraction. This leads to a decrease in elastance of the muscle tissue, therefore, yielding lower springiness values (Lucarini *et al.*, 2020).

The absence of a meaningful difference in springiness notwithstanding, the trend towards elasticity apparently means that marination is probably going to matter to meat texture. This is an important factor to consider in real applications, since that elasticity is one of the most distinguishing features of meat products.

Table 2. The texture of beef shank.

Parameter	Pineapple vinegar		
	0% (T0)	8% (T1)	16% (T2)
Hardness	70.95 ± 3.99a	61.05 ± 9.31b	51.30 ± 7.62c
Springiness	1.63 ± 0.15	1.58 ± 0.08	1.50 ± 0.21
Cohesiveness	0.74 ± 0.03	0.72 ± 0.03	0.71 ± 0.05

Different superscripts in the same row indicate a highly significant difference ($P < 0.01$).

Cohesiveness

Marinade treatment of beef shank with pineapple vinegar at a concentration 8% and 16%, respectively, had no significant effect on cohesiveness values ($P > 0.05$). The cohesiveness of beef shank for all treatments (including the control) was relatively consistent and close to 1 (Table 3). These results are consistent with the data reported by Unal *et al.* (2023), who also found that acidic solutions will not have a major impact on meat cohesiveness.

Cohesiveness is defined as the proportion of the pressure area in the second compression and the pressure area of the first compression, which reflects the cohesiveness of the material (Paredes *et al.*, 2022). Cohesiveness values near 1 indicate that the material holds well together in compression and does not fall apart. Furthermore, the almost uniform cohesiveness values for all treatments indicate that the marination step with pineapple vinegar was not enough to disintegrate muscle structure or connective tissues in the meat (beef shank).

The stability in cohesiveness found here corresponds to the relatively constant springiness scores obtained for different treatments. These findings imply that pineapple vinegar as marination solution had no marked changes in basic structural properties of beef shank muscle tissue. Beef shank has a very high cohesiveness, which is likely due to large characteristics of the cut. Introducing cut The shank is the muscle located at the upper portion of the legs. The shank is a weight-bearing section that has a lot of connective tissues, particularly collagen, which makes it very chewy, therefore slightly hard to bite. While pineapple vinegar does contain organic acids that can assist in softening connective tissues, this treatment would be insufficient on its own to completely denature the massive connective tissues in this cut.

Softening connective tissues in beef shank involves probably other interventions, such as heat treatment, also capable of effectively denaturing those proteins: collagen and elastin. In control treatments no additional treatments were made and the shank connective tissues remain intact consistently resulting in high cohesiveness values for all treatments.

Relationship between pH, WHC, and beef shank texture

Table 3 presents the Pearson correlation analysis between pH, WHC (Water Holding Capacity), and the textural attributes of beef shank. This analysis provides insights into how chemical factors such as pH and WHC influence the physical characteristics of meat.

Table 3. Correlation between pH and WHC with texture profile.

Characteristic	pH	WHC
T0 (Control)		
Hardness	0.21	0.936*
Springiness	-0.12	0.49
Cohesiveness	-0.00	0.49
T1 (8% vinegar)		
Hardness	0.17	-0.58
Springiness	0.67	-0.59
Cohesiveness	0.04	0.60
T2 (16% vinegar)		
Hardness	-0.41	0.55
Springiness	-0.37	0.76
Cohesiveness	-0.61	0.56
All sample		
WHC	0.545*	-
Hardness	0.699*	0.508*
Springiness	0.29	0.480*
Cohesiveness	0.17	0.46

*Significant at $P < 0.05$.

The pH was very weakly correlated with all texture parameters (hardness, springiness, and cohesiveness) of beef shank in the control treatment (T0). In contrast, WHC exhibited a very high and highly significant correlation with hardness ($r = 0.936$, $P < 0.05$), suggesting that higher WHC contributes to greater hardness of the meat. were not statistically significant, but WHC ($r = 0.490$ and $r = 0.498$, respectively) also significantly correlated with springiness and cohesiveness.

In T1 treatment, pH correlated poorly with hardness ($r = 0.165$) and cohesiveness ($r = 0.040$), while strongly correlated with springiness ($r = 0.670$). This indicates that elasticity is more sensitive, than hardness or consistency of the meat in response to the changes in pH under T1. On the other hand, WHC showed a negative correlation with hardness ($r = -0.577$) and springiness ($r = -0.589$), which suggested that low WHC was associated with low hardness and elasticity. The WHC had a moderately strong positive correlation with cohesiveness ($r = 0.602$), which indicated that good WHC can maintain the consistency of meat.

For the T2 treatment, pH was often negatively correlated to texture profile. There was fairly poor correlation of pH with hardness ($r = -0.409$) and springiness ($r = -0.372$), while this was relatively strongly correlated with cohesiveness ($r = -0.611$). That a pH drop can decrease hardness and elasticity but has a larger effect on the consistency of the meat. Conversely, hardness ($r = 0.545$) and cohesiveness ($r = 0.555$) were relatively weakly positively correlated with WHC; while WHC was highly correlated with springiness ($r = 0.759$). These correlations indicate that WHC is among the main factors affecting meat texture after this type of treatment.

On the whole, according to the data of all the samples, WHC significantly ($P < 0.05$) has a strong positive relationship with pH ($r = 0.545$). The same thing was stated by Rahman *et al.* (2023). According Lucarini *et al.* (2020), factors that influence pH will also influence WHC. Furthermore, pH had a positively highly significant correlation with hardness ($r = 0.699$, $P < 0.05$), and very low correlation with springiness ($r = 0.286$) and cohesiveness ($r = 0.167$). It is a sign that hardness is more directly affected by pH than other texture parameters. There were several significant correlations between WHC and texture parameters, including hardness ($r = 0.508$, $P < 0.05$) and springiness ($r = 0.480$, $P < 0.05$), as well as a moderately strong positive correlation with cohesiveness ($r = 0.458$).

With decreasing WHC, more free liquid leaves the muscle tissue, creating a less tenacious structure that is more susceptible to bite. Wyrwiz *et al.* (2012) found that pH and WHC greatly affect the texture of certain muscle types. As the pH declines, denaturation of myofibrillar proteins can occur, which can in turn contribute to the effects of WHC and texture profile of meat (hardness and cohesiveness). According to Bulgaru *et al.* (2022), WHC is one of the major parameters that affect the meat texture. WHC not only strengthens the tissue structure by increasing the water holding capacity but also results in greater hardness. On the other hand, low pH weakens actomyosin bonds, thus softer and springier meat.

During the cohesion, the content of myofibrillar proteins is more important than WHC (Bulgaru *et al.*, 2022; Rahman *et al.*, 2023). Data shows moderate correlations between WHC and cohesiveness while the effect of pH is rather weak, as can reasonably be expected. Therefore this study provides supportive evidence that marinating with pineapple vinegar not only decreases meat hardness by conducting protein hydrolysis, indirectly affecting pH and WHC, and promoting comprehensive

Conclusion

Pineapple vinegar has the potential to be used as a natural marinade to improve the texture quality of shank beef shank cuts. The best concentration of pineapple vinegar is 16%. Between pH value and WHC has a close relationship, but WHC has a closer relationship to the texture of beef shank. This knowledge is important in improving the quality and processing of beef shank meat, and the importance of pH and WHC regulation to improve the quality of meat texture.

Conflict of interest

The authors declare that they have no conflict of interest.

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