# **Research Article**



# **Tenderisation of Meat Using Bromelain from Pineapple Extract**

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#### ABSTRACT

The aim is to study the meat tenderising effects of bromelain obtained from pineapple extract. Tenderisation of meat is defined as the breaking down of collagen in meat to make it more palatable and bromelain does the same by degrading the collagen fibres of meat as bromelain is a mixture of two proteases. Bromelain is an enzyme found in pineapple that has uses in medicine and in the tenderisation of meat. It is commercially derived from the stems of pineapple. The objective is to assess the proteolytic activity and analyse the factors like concentration of bromelain, pH, water-holding capacity, moisture content which affect meat tenderisation by bromelain from pineapple extract. This study aids us to analyse the factors which affect the action of bromelain on meat and thus affect cooking time. The physicochemical characteristics of the meat samples treated with bromelain were determined. The pH of meat on addition of various concentrations of bromelain seemed to decrease thus, tenderising the meat as the acidity increased, with the lowest pH being 5.51. The water-holding capacity (WHC) gradually increased; however, there was a sudden drop in one of the bromelain treated meat samples (from 9% to 8%). The highest WHC was recorded to be 11% in the meat sample with the highest concentration of bromelain. The moisture content, too, was analysed and was found to be initially increasing and then decreased when the concentration of bromelain was the highest. Thus these results indicate that bromelain derived from pineapple extract can be used for the effective tenderisation of meat.

Keywords: Bromelain, Tenderising meat, pH, Water-holding capacity, Pineapple extract.

## INTRODUCTION

oughness is one of the most common quality characteristics of meat. Toughness is due to a range of factors including the amount of intramuscular connective tissue, intramuscular fat and the length of the sarcomere.<sup>1-2</sup> Thus, it is unsatisfactory to the consumer, if the meat is so tough that it is difficult to eat.<sup>3</sup> There have been various attempts to tenderise meat and thus make it more palatable. Different studies have made the use of calcium chloride,<sup>4</sup> salts, phosphates<sup>5</sup> and enzymes<sup>6-7</sup> and reduce the amounts of detectable connective tissue chemically. Physical methods have too been tried to tenderize meat by pressure treatments, electrical stimulation and blade tenderization.<sup>2</sup> Commonly used technique to tenderise meat is using enzymes such as ficin, derived from figs latex, bromelain, derived from pineapple stem and papain, derived from pineapple latex. These are proteolytic enzymes extracted from plants.<sup>8</sup> In this study, we will be using bromelain from pineapple extract to test its effectiveness in tenderising meat.

Bromelain is a complex mixture of proteinases that is derived from the stem of the pineapple plant, *Ananas comosus.*<sup>9</sup> The proteolytic enzymes are sulfhydryl proteases; a free sulfhydryl group of a cysteine amino acid side chain is required for function. Bromelain is reported to also be present in pineapple wastes such as in cores, peels and leaves. The bromelain in their waste materials are in relatively smaller quantities as compared to in the stems and fruits.<sup>10</sup> It is also used as a nutritional additive to assist digestive health, an anti-inflammatory, anti-oedematous, absorption facilitator of antibiotic

drugs, an anti-thrombotic, an inhibitor of tumour cell reproduction, a debrider and an immunogenic agent.<sup>8</sup> Bromelain acts on meat by breaking down the collagen fibres, i.e.; it shows hydrolytic activity on the connective tissue leading to the tenderisation of meat.

Action of bromelain on meat is affected by factors such as pH, water-holding capacity, moisture content and concentration thus this study's motivation was to study its proteolytic activity and the factors affecting it. This study investigates the effects of the bromelain extract on the physicochemical and quality properties of the samples.<sup>2</sup>

## MATERIALS AND METHODS

#### **Preparation of Meat**

Fresh boneless meat was obtained from a butcher shop and was sliced into several pieces of approximately the same size and weight. The meat was divided into two groups; one group was used for the experiment and the other used as control group. The experiment group consisted of 4 samples and the control group had one sample;

- C Control (Sample without Bromelain)
- B1 Sample treated with 1% Bromelain
- B2 Sample treated with 2% Bromelain
- B3 Sample treated with 3% Bromelain
- B4 Sample treated with 4% Bromelain



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#### **Determination of pH**

Approximately 2 g of meat was taken in 5 test tubes. The tubes were incubated with 1%, 2%, 3% and 4% solutions of Bromelain in distilled water and a test tube without Bromelain, served as control. It was allowed to stand for 10 minutes and was homogenized by grinding followed by crushing mechanically using a mortar and pestle. The pH values were determined using a digital pH meter. The pH meter was calibrated using a standard sodium acetate buffer of pH 4 and the pH of the samples was measured.

# **Determination of Water-holding Capacity**

Water-holding capacity (WHC) was determined according to the procedure described by Wardlaw.<sup>11</sup> 6 g samples of meat (treated with 0%, 1%, 2%, 3% and 4% of Bromelain) were stirred for 1 min with 10 ml of 0.6 M sodium chloride (NaCl) solution in a 15 ml centrifuge tube.

The tube then was held at 4° C for 15 min, stirred again for 1 min and centrifuged at 10,000 rpm at 4° C for 15 min. After centrifugation, the volume of the supernatant was measured and the water-holding capacity of the meat treated with various concentrations of meat was calculated using the formula:

WHC (%) = ((Initial volume -Volume of supernatant)/Initial volume) x 100

# **Determination of Moisture Content**

5 g of meat (treated with 0%, 1%, 2%, 3% and 4% of Bromelain) was suspended in 10 mL of 0.6 M sodium chloride (NaCl) solution for 30 minutes. The weight of the meat was noted. They were placed on a glass plate and heated at 100°C for 20 minutes using a hot plate. The weight of the meat after removing moisture was determined. Percentage of moisture content in the meat samples were calculated as:

Percentage of moisture content = ((Initial weight – Final weight) / Initial Weight) x 100.

# **RESULTS AND DISCUSSION**

#### Assessment of pH

pH was determined using a pH meter and the resulting pH ranged from 5.51 to 6.93. The experiment was repeated 3 times. A significant reduction in pH was observed in all bromelain treated samples when compared to the control (C). In a normal living muscle, the pH is approximately 7.2.<sup>12</sup>

No significant difference was observed between the control C, B1, B2 and B3 as their average pH were approximately 6.

The peak in the difference of pH is observed in B4 (meat marinated in 40% bromelain) with the pH becoming 5.51 in comparison to the control group C. Moreover, bromelain extract hydrolysis of the muscle may result in releasing amino acids that can reduce the pH of the system.<sup>2</sup>

The pH value in the meat product is highly important because it has a major influence on other physicochemical and quality properties such as WHC, tenderness, and juiciness.<sup>13</sup>

In the current research, the greatest pH decrease is observed in B4 (meat marinated in 40% bromelain), which is the highest level of bromelain applied in this study.

The average pH for the treatments of C, B1, B2, B3 and B4 was 6.83, 6.31,6 and 5.51 respectively. With an increase in the concentration of bromelain, the pH of treated sample of meat decreased thus it became more acidic with the increase in the addition of bromelain on it.

Gault N.F. stated in his study, "Additionally, strong linear decreases in toughness were apparent over the pH range 4.6 to 4.1 for the three muscle types studied".<sup>14</sup>

The acidity of the meat denotes that the muscle fibres of the meat have been softened thus, lesser the pH, higher is the tenderization of the meat.



Figure 1: Effect of bromelain on pH of treated sample of meat

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Samples	С	B1	B2	B3	B4
	6.93	6.25	6.29	5.91	5.54
	6.64	6.54	6.11	6.2	5.47
	6.91	6.13	6.14	5.89	5.51
Average	6.83	6.31	6.18	6.00	5.51



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	Water-holding Capacity				
Samples	Initial Volume (mL)	Volume of supernatant (mL)	Water-holding Capacity (%)		
С	10	9.2	8		
B1	10	9.2	8		
B2	10	9.1	9		
B3	10	9.2	8		
B4	10	8.9	11		

Table 2: Effect of Bromelain on WHC of the treated sample of meat

 Table 3: Effect of Bromelain on Moisture content of the treated sample of meat

Moisture content								
Samples	Initial weight (g)	Final weight (g)	Moisture content (%)					
С	7.2	5.38	25.28					
B1	7.15	5.34	25.31					
B2	6.89	5.11	25.83					
B3	7.34	5.44	25.89					
B4	7.51	5.61	25.30					

# Assessment of Water-holding Capacity

The water-holding capacity (WHC) of foods can be defined as the ability to hold its own and added water during the application of forces, pressing, centrifugation, or heating.<sup>15</sup> It can also be defined as a physical property and is the ability of a food structure to prevent water from being released from the three-dimensional structure of the protein.<sup>16</sup>

The WHC of meat is very important since many physical properties such as colour, texture and firmness are partially dependent on the WHC.<sup>2</sup>

Water-holding capacity was measured and it was 8% for the control group C. There wasn't any difference between B1 and C as both their water-holding capacities were measured to be 8%. A significant increase in the waterholding capacities is seen only in B2 and B4 being 9% and 11% respectively.

Much of the water in the muscle is entrapped in structures of the cell, including the intra- and extramyofibrillar spaces; therefore, key changes in the intracellular architecture of the cell influence the ability of muscle cells to retain water.<sup>17</sup>

There was a gradual increase in the water-holding capacity of the meat as the concentration of bromelain increased with an exception being B3 as its water-holding capacity was the same as control group C. The water-holding capacity is highest for B4.

Upon increasing or decreasing pH, the net charge increases.

The mutual repulsion of fibrillar proteins causes the swelling of the matrix and the water-holding capacity increases.  $^{18}\,$ 

The pH of beef muscle affected water-holding capacity (WHC) and tenderness.

When the pH was brought to 4.0 and below by immersion in citric acid or lactic acid or brought to  $\sim$ 7.0 after base immersion, WHC and tenderness of beef muscle increased.<sup>19</sup>

The higher WHC in the control sample may be due to the overall reduction in the protein reactive group that is available for water binding.<sup>20</sup>

Low pH meat had the lowest water-holding capacity compared with normal and high pH meat as shown by the increase in cooking loss, which can be explained by factors other than protein denaturation.<sup>21</sup>



Figure 2: Effect of Bromelain on WHC of the treated sample of meat



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# **Assessment of Moisture Content**

Water content, or moisture content, is a measurement of the total water contained in a food. It is usually expressed as a percentage of the total weight.

The percentage of moisture content was calculated and it did not vary significantly among treatments.

The moisture content of the samples ranged from 25.28% to 25.89%. A significant peak in moisture content is observed in B2; 25.83%.

The moisture content initially seemed to increase till B3 and then decreased rapidly from 25.89% to 25.30% in B4.



Figure 3: Effect of Bromelain on Moisture content of the treated sample of meat

# CONCLUSION

Our outcomes on the effect of bromelain on physicochemical properties such as pH, concentration of bromelain, water-holding capacity and moisture content of meat have indicated the effective utilization of bromelain extract from pineapple in tenderizing tough meat.

These factors also affect proteolysis of key cytoskeletal proteins in postmortem muscle.  $^{\rm 17}$ 

This study shows how tough meat can be easily softened by the use of a natural product, pineapple, without compromising the quality of the meat. Even though India is a multi-cultural country the meat industry is huge valuing upto ₹148,954 crores (US \$ 31,000 Million).

Thus, natural tenderizers such as bromelain, ficin, etc., are very much in demand. It sheds light on the several uses of bromelain in our everyday life.

Bromelain has an economic importance as well, as the enzyme is obtained from pineapple parts including its stem and wastes and the technology for applying this enzyme is easily and cheaply available and can be exploited at the household or industrial level for tenderizing tough meat, and it can be used as a better alternative to chemical tenderizers or other plant proteases.<sup>2</sup>

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