## EFFECTS OF PAPAIN ON BEEF TENDERNESS

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#### Abstract

From practical stand point may be used many type of treatments to increase beef tenderness: electrical stimulation of carcass, aging of carcasses or cuts, mechanical tenderization of cuts, injection of cuts with tropical plant enzymes (papain, ficin and bromelin) or chloride salts (especially calcium chloride) and proper temperature, time and type of treatment. From all of these types of treatments we studied enzymatic tenderization.

I considered papain to be a proteolytic enzyme (derived from papaya), with positive effects on mature beef tenderness and the selection I made because it is used also in medical interests (like digestive enzyme, to defibrillate wounds in hospitals, to prevent cornea scar deformation, to treat edemas, inflammatory process and in the acceleration of wound healing).

In the present experiment, papain was added in injection brine and than the beef cuts were injected with a specific percent of brine. Experimental data indicate that the papain weaken beef meat structure producing improvement of beef tenderness. A significant increase in tenderness by rigidity index measurement was observed in the samples treated with papain as compared with the control. The palatability traits of beef treated with papain were better than control's. All of these data confirm that the meat tenderizer effects of papain.

**Keywords**: Tenderness, meat, papain, rigidity index.

## Introduction

The major palatability attributes of meat are tenderness, juiciness and flavor. Consumers consider tenderness as the most important factor in determining eating satisfaction of beef (Issanchou, 1996, Boleman, Miller, Taylor, Cross, Wheeler, Koohmaraie, Johnson and Savell, 1997). Obtaining a meat with high palatability characteristics for consumers satisfaction may seems easy at a first sight. When consumers demand is oriented to a dietetically nutrition, less rich in cholesterol, beef prevails on the market.

But beef may be sometimes difficult to chew due to its rather hard rigid texture.

Tenderness is defined as the ease of mastication, which involves the initial ease of penetration by the teeth, the ease with which the meat breaks into fragments and the amount of residue remaining after mastication (Lawrie, 1998). The two major determinants of meat tenderness are the content and state of the connective tissue and the structure and state of the myofibrils.

Meat tenderness depends on specie, breed, age, sex and individual skeletal muscle tissue of animal. Tenderness originates in structural and biochemical properties of scheletal muscle fibres, especially myofibrils and intermediate filaments, and of the intramuscular connective tissue, the endomysium and perimysium, which are composed of collagen fibrils and fibres (Takahashi, 1996). The mechanical stability of collagen fibrils increase markedly with chronological ageing. Meat produced from old animal is tough and has a lower eating quality. Improvement of meat tenderness of aged cattle is necessary to increase palatability and value of the meat (Shiba, 2004). Meat can be made tender in the following ways:

- *mechanical methods* mincing or hitting to crush the conjunctive and muscle tissue;
- chemical methods by injecting in the muscle solutions with chemical substances (salt, sodium polyphosphate, lactate potasium, diacetate sodium all dissolved into water),(R.K., 2003); by immersing into marinade, which can be acid (as lemon juice, vineagre, wine or tomatoe juice); milk products (yogurt and butter it seems that the calcium ions in the milk products activate endogenous enzymes of meat thus accelerating proteolysis);
- enzymatic methods used enzymatic preparates as papain, bromelin or ficin and also fruits containing such enzymes papaya, pineapples or figs as piuree). Enzymatic tenderization can be made in a humid way (by injecting in the muscle solutions with enzymatic preparates) or in a dry way (pressing tender mixtures on the meat surface).

The end of the tenderization process and the method employed for ageing and thermal processing should be established to get an optimum sensorial quality of the meat.

## **Experimental**

The study employed adult beef thigh post rigor (24 hours after slaughter), purchased from the commercial distribution in refrigerated form. Salt was of food-suitable purity, being a largely used additive in meat industryPapain Chilko P (Lay Condiments, Bucharest).

**Chemical Analyses.** The chemical composition of beef was determined by:

- *the water content* according to the AOAC- 1995 method, by hot drying for 24 hours at 100°C;
- *the total nitrogen content* and calculating the content of global proteins by multiplying the percentual content of total nitrogen by the proteic factor 6,25 was achieved according to the SR ISO 9037:2007 method;
- *the fat content* consisted in extraction of the free fat substances from the sample to be analysed with ethilic ether according to the AOAC, 1984 method:
- *the pH* was achieved according to the AOAC method, by preparing an extract (10 g ground sample and 90 ml distilled water) which was homogenised for 2 minutes, the meat suspension was filtered, and the pH determined with a Hanna digital pH-meter.

**Tenderness degree.** The degree of tenderness for the papaintenderized beef was assessed by determining the rigidity index, according to the method described by Ionescu, and al, 1992. the rigidity index was calculated by means of the formula:

$$I_r = a/g [cm^2/g]$$

where:

a = surface occupied by pressed meat by applying a weight of 1 kilo, for 10 minutes, cm<sup>2</sup>;

g = mass of the meat submitted to pressure, in g.

**Sensorial analysis.** The sensorial analysis of papain-tenderised meats was achieved by a team of samplers, made up of 5 specialists, using a 5-point scale (1-I dislike it; 2- I do not like or dislike it; 3- I like it; 4-I like it moderately; 5- I like it very much).

**Sample preparation.** The adult beef thigh separated from the gross conjunctive tissue and fat was cut into pieces of the same size in length and thickness, weighing approximately 130 g, cut along the muscular fibers. The meat pieces were then divided into four groups and were used for a certain treatment. They were injected with brine made up of: salt 2 g and water 98 g

to which various amounts of papain were added. For each treatment series were constituted, consisting of:

- Control sample (M), the pieces of meat were injected with 10% brine without papain addition;
- Sample A the brine was completed with papain to a concentration of 0.002 mg/100 g meat. The injection was 10%;
- Sample B the brine was completed with papain to a concentration of 0.004 mg/100 g meat. The injection was 10%;
- Sample C the brine was completed with papain to a concentration of 0.006 mg/100 g meat. The injection was 10%.

The injection was performed manually by means of a single-needle injector, so that the entire brine quantity could be uniformly pumped into the whole muscular mass. The eliminated brine was reinjected. The injected meats were wrapped with a polyethilene film and stored at 4°C for 24-48 hours aiming to achieving an uniform that diffusion of brine in the muscular tissue with or without papain addition and to deploying the activity of exogenous proteolitic enzymes.

After maturation, the meats were boiled in hermetically sealed test tubes on a water bath by gradual heating (about 1°C/minute) up to reaching the thermal centre of the temperature of 83°C, which was maintained for 10 minute. After boiling, the samples were immediately cooled on water bath cooled by means of ice, after which they were stored at refrigeration temperature over night. The boiled meats brought to room temperature were carefully removed from the test tubes and were weighed after being tapped with filter paper. The juice expressed at boiling was collected, weighed and used for various analyses. The juice percentage expressed at thermal treatment was calculated with the formula:

% Expressed juice = [Mass of meat after injection and maturation – Mass of boiled and cooled meat]  $\times$  100 / Mass of meat after injection and maturation

### **Results and Discussions**

The study used adult beef thigh post rigor (24 hours after slaughter), purchased from specialized stores in refrigerated state. Initially the chemical composition of the purchase meat was analyzed. The data obtained (Table 1.) showed a relatively lean meat (5.82%  $\pm$  1.4% fat), with an average protein content (17.2%  $\pm$  1.08%) and water content (75.8%  $\pm$  1.03), the

average values and errors being calculated with the statistic program Sigma plot 2001 for five different lots of beef.

Table	1 Chemical	components	of beef

Chemical	Content		
components	g%	g% s.u.	
Moisture	75.8	-	
Dry substance	24.2	-	
Total nitrogen	2.72	11.24	
Total proteins	17.2	71.07	
Fats	5.82	24.04	
Non-protein nitrogen	0.212	0.876	
Aminic nitrogen	0.066	0.272	
Ammonia	0.019	-	
pН	6.02	-	

The degree of rigidity was used as a measure of assessing the degree of tenderness of beef enzymatically tenderised with papain. The rigidity index represents the resistence opposed by meat to compression, both of raw meats and boiled meats.

The increase of the level of papain added as well as the increase of the duration of the enzyme activity led to a considerable increase of the values of the rigidity index both in raw samples and in thermally treated samples by boiling (figure 1, 2). The lowest values of the rigidity index were observed for the control (injected only with brine with no papain addition) as compared to samples tenderised with bromelain.

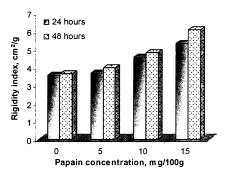


Fig 1: Influence of the papain level on the rigidity index of thermally treated beef by boiling

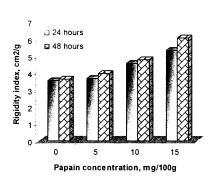


Fig. 2: Influence of papain on the rigidity index of raw beef

No matter the thermal treatment applied to beef, there are a series of more or less intense chemical and physical alterations. The meat texture is altered during the thermal treatment as a result of protein denaturation, meat dehydration, colagen hydrolisis, fat expulsion from fat cells and their dispersion into the meat mass.

The study on papain-tenderised meat registered losses at thermal treatment, they being maximal for the control sample (fig. 3). The increase of the papain level added and the period of enzyme activity, losses went down.

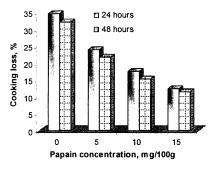


Fig. 3: Influence of papain enzymatic tenderisation on losses at thermal treatment

Sensorial analysis consisted in assessment of palatablity characteristics of enzymatically-tenderised beef thermally treated by boiling.

By the increasing of the level of enzyme added and by the increasing of duration from 24 to 48 hours, a considerable fragmentation of muscular fibers was seen, part of the enzymatically treated meat became soft after boiling, with very low resistence to mastication. Also, some samples exhibited, after thermal treatment, areas with texture similar to a paste probably due to the inappropriate distribution of the injected solution. The flavour of the thermally treated meat was not significantly affected. The incease of the papain dosages led to a slightly unpleasat taste that can be masked by spicing. Due to the small losses at thermal treatment the papaintenderised samples were juicier as compared to the control sample.

## **Conclusions**

Papain is a powerful proteasic preparation, with great under-layer specificity, catalyzing the breaking of the peptidic bonds in the protein molecules and their degradation poducts to aminoacids. Also, papain has a collagenasic activity, turning collagen soluble, collagen being a major component of the conjunctive tissue. The increase of the papain level as well as the increase of the duration of the enzyme activity led to a considerable weakening of the meat structure. Papain tenderisation led to improving flavour, juiciness, tenderness in adult beef.

It is recommended to use papain doses as low as possible, in order to avoid advanced tenderisation and obtaining meats with soft structure, with very low resistence to mastication and paste texture.

All the results obtained in the pesent study prove the tenderising effect of papain.

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