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Title: PRE-RIGOR HIGH-PRESSURE TREATMENT EFFECTS ON SELECTED QUALITY CHARACTERISTICS OF BEEF SEMITENDINOSUS MUSCLE

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The effects of pre-rigor high-pressure treatment of beef semitendinosus muscle on tenderness, juiciness, and flavor were studied using both objective and subjective tests. Objective tests included Warner-Bratzler shear, myofibril protein solubility, total moisture, water-holding capacity, pH, total nitrogen, and fat content for raw and cooked samples. Total, evaporation, and drip cooking losses were determined also. Cooked samples were subjectively evaluated by a trained panel of eight judges. Samples were judged for tenderness, friability, juiciness, flavor, and number of chews before swallowing.

Paired-t statistical analysis of data indicated no significant ($P < 0.05$) difference between the pressure-treated and untreated meat in Warner-Bratzler shear values, myofibril protein solubility, total moisture, water-holding capacity, total and evaporation cooking losses, total nitrogen, and fat content. Pressure treatment resulted

in significantly less drip cooking loss and higher pH for the raw and cooked treated meat. Taste panel results showed no significant difference between the pressure-treated and traditionally-aged untreated meat in tenderness, juiciness, friability, flavor, and number of chews.

Pre-Rigor High-Pressure Treatment Effects on Selected Quality Characteristics of Beef Semitendinosus Muscle

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PRE-RIGOR HIGH-PRESSURE TREATMENT EFFECTS ON SELECTED QUALITY CHARACTERISTICS OF BEEF SEMITENDINOSUS MUSCLE

INTRODUCTION

Per capita consumption of meat in the United States is near an all-time high (USDA-ERS, 1976b), with expenditure of a large portion of the family food budget for meat and meat products (USDA-ARS, 1974). Growth in meat consumption has been accompanied by increased demand for convenience and prepared foods (USDA-ERS, 1976a). Prefabricated meat is one such convenience food which is being used increasingly in institutions and restaurants. Prefabricated meat is meat which has been processed into a variety of boneless trimmed cuts according to the tenderness of the muscle (Fielder et al., 1963). The cuts are portion-controlled with smaller cuts designed to provide one serving and roasts designed to provide a variable number of servings.

High meat prices are an economic concern to the consumer. Rising costs experienced by the livestock producer contribute to high meat prices. Additionally, they are partially a result of costs to the meat packing industry, which reported increases in raw material costs, energy costs, and operating expenses in 1975 (American Meat Institute, 1976). The trend toward more use of prefabricated meat may be a money-saving development, particularly for institutions.

Reduced meat costs, labor costs, workloads, and waste, and improved prediction of costs are suggested advantages of prefabricated meats (Kotschevar et al., 1953).

A recently-developed high-pressure treatment of hot-boned pre-rigor muscle (Macfarlane, 1973) shows the potential of yielding meat of desired quality suitable for prefabrication. This processed meat would have the advantage of possible energy and economic savings in production and transport in comparison to meat handled in the traditional way. These savings include less refrigeration space and input and lower transportation costs for meat from which the waste bone and fat have been removed prior to refrigeration and shipping (Kastner et al., 1973).

The objectives of this research were to study the effects of the high-pressure treatment of pre-rigor beef semitendinosus muscle on tenderness, juiciness, and flavor, using both objective and subjective methods. Objective methods included Warner-Bratzler shear, myofibril protein solubility, total moisture, water-holding capacity, cooking losses, total nitrogen, fat content, and pH. Subjective testing was performed using taste panel evaluation. Panelists evaluated samples for tenderness, friability, juiciness, flavor, and number of chews before swallowing.

REVIEW OF LITERATURE

Consumer acceptability of meat is greatly influenced by the conditions under which rigor mortis occurs, as well as other conditions of post-mortem handling of the meat. Post-mortem handling influences quality due to its effect on the metabolic events which occur around the time of rigor mortis (Briskey and Kauffman, 1971). Some knowledge of these metabolic events and how they influence the muscle cell is a requisite to insight on factors determining ultimate meat quality.

Sarcomere Components and the Contractile Mechanism

The structure of the sarcomere and the mechanism of muscle contraction have been summarized by Goll et al. (1974). The skeletal muscle myofibril is an array of two kinds of interdigitated protein filaments, thick ones containing myosin, and thin ones containing actin and some regulatory proteins. The thick and thin filaments are arranged in an order that gives a striated appearance to the myofibril and to the entire muscle cell. The dark, myosin-containing band is known as the A-band and the light, actin-containing band is the I-band. The ends of the myofibril are determined by the Z-disk, which is seen microscopically as a dark line perpendicular to the filaments. The myofibril from Z-disk to Z-disk is defined as

the sarcomere and is the basic repeating contractile unit.

The myosin filament has cross-bridges which project from the surface of the filament (Goll et al., 1974). Muscle contraction occurs when the projecting myosin cross-bridges contact the actin strands and push the opposed actin strands toward the center of the sarcomere. Since there is no change in length of the filaments, the change in muscle length upon contraction is felt to be due to further interdigitation of the thick and thin filaments from their pre-contraction state.

Adenosine triphosphate (ATP) supplies energy required for muscle contraction (Goll et al., 1974). Hydrolysis of ATP catalyzed by myosin is a major step in the generation of contractile force. ATP functions also to inhibit the myosin cross-bridge attachment to actin.

Calcium plays a major role in muscle contraction (Goll et al., 1974). In a relaxed living muscle cell, calcium is present at low levels in the sarcoplasm, the intracellular fluid of the muscle cell. However, with a nerve impulse to muscle, bound calcium is released from the sarcolemma to the sarcoplasm. In increased sarcoplasmic concentration, calcium effectively activates the actin filaments via a reaction with tropomyosin, a blocking regulatory protein. The activation facilitates the attachment of the myosin cross-bridges to the actin, resulting in the formation of actomyosin. Myosin

cross-bridges are in an energized state, with adenosine diphosphate (ADP) and phosphate bound tightly. The attached energized myosin bridge undergoes a conformational change, moving the actin filament along the myosin filament. Simultaneously, the ADP and phosphate are released from the myosin and replaced by ATP, causing dissociation of the myosin head from the actin. Bound ATP is then hydrolyzed to ADP and phosphate, the myosin cross-bridge reconnects with actin and the cycle repeats. The muscle is in an inextensible state when the cross-bridge is attached to actin as the actin and myosin filaments are not free to slide past each other. Muscle relaxation in the living cell is a result of sequestering of the calcium into the sarcolemma.

Each known muscle fiber protein is insoluble at its isoelectric point in the absence of a salt such as a phosphate or potassium salt. Actomyosin is the most insoluble of these proteins at its isoelectric point (Weber and Portzehl, 1952). In an ATP-free solution, Haga et al. (1965) observed that the amount of myofibril protein solubilized from rabbit skeletal muscle with Weber-Edsall solution (0.6M KCl, 0.04M $NaHCO_3$, 0.01M Na_2CO_3) increased with time of extraction up to eight hours and was constant thereafter. They proposed that myosin is initially extracted from the myofibrils and is then converted to actomyosin, with the conversion starting after about eight to ten hours of extraction and completed at about 20 hours. Mihalyi and Rowe (1966) also indicated that during extraction, the myofibril

structure is partially solubilized and the extracted protein is re-organized into actomyosin in the extract. They concluded from their work with rabbit muscle that the presence or absence of ATP determines if myosin or actomyosin is predominantly present in the extract and that the solubilization of actin is required for production of actomyosin. Their suggested model to account for this behaviour proposed that ATP interferes with the production of actomyosin, perhaps by stabilizing the actin filaments or their connection to the Z-disk, therefore making them insoluble, and not by retention of actin and myosin in a dissociated state.

In comparing stretched and unstretched beef sternomandibularis muscle, Cook (1967) found a significant increase in extractability of myofibril proteins per gram of post-rigor tissue in stretched muscle. His work supported the hypothesis that increased myofibril protein solubility is a result of prevention of complete interdigitation of actin and myosin (Khan and van den Berg, 1964). Correlation between nitrogen extractability and both sarcomere length and moisture press ratio lead Cook (1967) to the conclusion that there exists a relationship between protein solubility and the contractile state of the myofibrils.

Some Post-Mortem Phenomenon in Muscle

Slaughter of an animal marks the start of a series of biochemical and physiological changes in the muscle cell. With increasing

time after slaughter, certain cellular mechanisms fail to operate with their pre-slaughter efficiency, ultimately becoming non-functional (Goll et al., 1971). An example is the post-mortem loss of calcium-sequestering ability of the sarcolemma, possibly as a result of decrease in pH, loss of ATP, or uncoupling of the calcium pump by proteolysis. The result is continuous activation of the actin filaments.

Whitaker (1959) noted the importance of the following in development of rigor: 1) start of glycolysis at death and continuation until low pH prevents activity of glycolytic enzymes, 2) pH decrease due to lactic acid buildup, 3) decrease in creatine phosphate, 4) decrease in ATP level (at about 80% of the initial level, the muscle loses extensibility and contracts), 5) ammonia liberation. Bate-Smith and Bendall (1947) demonstrated that rigor onset was not a direct effect of lactic acid production from glycolysis. They proposed that pH and glycolysis rate do affect the time of rigor onset because these two factors determine the rate of ATP destruction. Busch et al. (1967) showed a correlation between ATP hydrolysis and pH fall in muscle. Post-slaughter ATP breakdown was shown by Bate-Smith and Bendall (1947, 1949) to proceed slowly at first and then increase. Rigor onset was observed to be simultaneous with the rapid ATP breakdown phase. Muscle shortening occurred during this phase in muscle with pH greater than 6.2, a condition characteristic of

muscle with low glycogen reserves. In addition to ATP breakdown, the anaerobic conditions brought about by the death of the animal caused a decrease in ATP synthesis due to decreased efficiency of the glycolytic ATP synthesis mechanism (Lister, 1970).

Bodwell et al. (1965) found in their studies on beef longissimus dorsi stored at 3 to 4° C post-mortem that rigor mortis onset occurred at about 12 to 15 hours post-mortem and development of rigor was complete at about 24 hours post-mortem. They detected negligible ATP and creatine phosphate content in 24-hour post-mortem muscle samples. pH was observed to decline initially with time post-mortem until about 96 hours and then to fluctuate slightly around the 96-hour pH value.

As well as biochemical changes, certain physiological changes are associated with rigor onset. Locker (1960) showed that unrestrained muscle shortens as it enters rigor, and the shortening was correlated with increased toughness. Marsh and Carse (1974) emphasized the role of actomyosin in meat tenderness and showed that the extent of actin and myosin interdigitation during rigor onset was correlated with toughness. Based on the findings of other workers, Goll et al. (1964b) suggested that rigor mortis is a result of irreversible binding of actin and myosin. Briskey (1970) mentioned the uncertainty regarding mechanisms of the increased tenderness which marks rigor resolution. He surmised the occurrence of movement of

the myosin cross-bridges. However, Wierbicki et al. (1956), using a protein solubility method could find no correlation between post-mortem actomyosin dissociation and rigor resolution.

Post-Mortem Treatments of Meat

Traditional treatment of the beef carcass post-mortem involves the aging or conditioning process, which Lawrie (1974) defined as storing at chill temperatures for 10 to 14 days. Davey and Gilbert (1969) suggested that the tenderization which accompanies aging may be a result of loss of adhesion between myofibrils due to disruption of the Z-disk of the myofibril, and also to decreased myofibrillar tensile strength. In tests on beef and mutton, Bouton and Harris (1972b) found aging-induced tenderness increase was only slightly if at all attributable to changes in connective tissue. DeFremery and Streeter (1969) came to this same conclusion with regard to chicken muscle. Carcass position during aging has been shown to affect sarcomere length and tenderness (Herring et al., 1965, Hostetler et al., 1970, Bouton et al., 1974), with greater muscle and sarcomere extension resulting in more tender meat.

In contrast to the traditional processing of beef in which primal cuts are excised from the post-rigor carcass, the concept of hot-boning the beef carcass involves pre-rigor excision of the muscle. Schmidt and Gilbert (1970) found that beef muscles excised two hours

after exsanguination and kept at 15° C for 24 hours were usually as tender as the paired muscle excised after 24-hour storage at 9° C. If the hot-boned muscles were kept at 15° C for 48 hours, certain muscles were found to be more tender than their paired muscle excised after 48-hour storage at 9° C. Using variations in excision and conditioning time for hot-boned muscle, Kastner et al. (1973, 1976), Kastner and Russel (1975), and Falk et al. (1975) found hot-boning to yield an acceptable product that was comparable in tenderness to a cold-boned control.

The effects of pre-rigor application of pressure to hot-boned warm excised ox and sheep muscle were studied by Macfarlane (1973). Muscle was removed from the carcass, sealed in a polyethylene bag and subjected to hydrostatic pressure in a chamber. The author found that the ox muscle samples pressurized at about 100 MNm^{-2} were more tender and more acceptable, but less juicy than the paired muscle left in the carcass and stored at 2° C. Warner-Bratzler shear values were consistently lower for pressurized ox muscle and lower in some cases for pressurized sheep muscle. Macfarlane (1973) suggested that this method would be conducive to rapid processing and tenderization of meat, avoiding the muscle-shortening-induced increase in toughness.

Application of pressure of the order of 100 MNm^{-2} for two and one-half minutes or longer to post-rigor beef muscle heated to

40-60° C was found by Bouton et al. (1977) to increase the tenderness of the cooked muscle. Post-rigor pressure-heat treated meat was compared with the pre-rigor pressurized beef. Both treatments were found to yield meat significantly more tender and acceptable and having lower shear values than untreated meat or post-rigor meat treated with heat or pressure only. There was no significant difference between the two treatments for these parameters. However, the pressure-heat treatment resulted in lower juiciness scores than post-rigor pressurization or no treatment. Sarcomere length measurements indicated that pre-rigor pressure treatment caused extreme contraction, but the pressure-heat treatment did not.

Cold-shortening of muscle is a treatment carried out by exposing excised fresh muscle to temperatures just above freezing, resulting in extreme shortening (Davey and Gilbert, 1974b). Marsh and Leet (1966) studied the effects of different degrees of shortening on tenderness of beef. They noted increasing toughness above 20 percent shortening until a peak was reached at 40 percent. With shortening beyond this, meat became more tender until at about 60 percent it was as tender as meat shortened less than 20 percent. In a later publication, Marsh et al. (1974) showed that increased tenderness with shortening greater than 50 percent is due to actual fracturing of many of the fibers. The toughness at 40 percent shortening was proposed by Davey et al. (1967) to be due to abuttment of the myosin

bands against the Z-disks.

High-Pressure Treatment of Muscle

The effects of pressure on muscle are by no means clearly defined at the present time. Macfarlane (1973) noted some variables that influence the reaction of a muscle to pressure. These included amount of pressure, rate of increase or decrease of pressure, length of pressure treatment, restriction on muscle contraction, time lapse between slaughter and pressurization, the muscle subjected to pressure treatment, and species of animal used as muscle source.

The effects of pressure on muscle may be observed from three different viewpoints: 1) effects on muscle tension and volume, 2) effects on the myofibril structure, 3) effects on the chemical and biochemical reactions in the muscle. Pressure-induced changes from each of these viewpoints are, of course, interrelated due to the integration of physiological and biochemical phenomena.

Effects on Muscle Tension and Volume

Murakami (1970) noted that several Japanese studies indicated that exposure to pressures of 4,300 psi or greater induced contraction in isolated skeletal, cardiac, and smooth muscle, and relaxation occurred upon release of the pressure. Brown (1934, 1934-1935) studied hydrostatic pressure-induced tension development in turtle auricle

muscle. In analyzing the experimental results of Brown, Johnson et al. (1954) concluded that the tension which developed was a function of temperature as well as pressure. They reported the greatest tension increase with increasing pressure up to 9,000 psi at 20° C and lower tension development at 15, 10, 5, and 1° C.

Based upon observed pressure-volume effects in muscle at different stages of contraction, Macfarlane (1973) suggested that applied pressure could possibly affect the contraction state of muscle. More research is needed on the effects of applied pressure on muscle and muscle volume. However, due to the thermodynamic relations between internal and external pressure and volume, a discussion of the more highly-studied area of internal pressure-volume phenomena in muscle is relevant.

Hotta and Terai (1966) observed a volume decrease of myofibrils in suspension upon addition of ATP. They concluded that volume change always occurs with ATP-induced myofibril shortening, and this may be due to a conformational change of the myofibril proteins. In related studies of the effects of ATP on myofibril volume, Baskin (1964) observed either a volume decrease or increase, depending upon the ATP concentration. He postulated that the observed volume changes were a result of either myofibril protein configuration changes or dissociation and association of the actin and myosin filaments during ATP-induced contraction or relaxation. He also noted

the similarities between the ATP- induced volume changes observed in his work with isolated myofibrils and the volume changes observed during a single twitch of frog sartorius muscle. Baskin suggested that the volume changes may have the same origin.

Baskin and Paolini (1967) noted changes in striated frog muscle volume with contraction and attributed at least part of this to alterations in the contractile component of the muscle. However, they were not able to relate the volume changes to any specific molecular phenomena. As a result of non-parallel arrangement of muscle fibers, the development of internal pressure in the frog gastrocnemius muscle upon contraction was observed. These investigators proposed that the developed internal pressure caused a compression of muscle fluid.

Effects on Myofibril Structure

Using electron microscopy, Macfarlane (1973) noted marked breakdown of the myofibrils of pressure-treated meat and suggested that perhaps this occurrence and the resultant tenderization are due to the forcing of the myosin filaments into the Z-disks. In the same publication, Macfarlane observed that pressurized ox biceps femoris shortened by about 40 percent of its rest length. As previously mentioned, Davey et al. (1967) proposed that at 40 percent shortening, the myosin bands are abutting the Z-disks, an event which produces

toughness in non-pressure-treated meat, but would appear not to have this effect in the case of pressure-treated muscle (Macfarlane, 1973). This lack of toughness in pressure-treated meat at 40 percent shortening may be attributable to the disruption of the Z-disk by the myosin filaments so that the myofibril-anchoring function of the Z-disks is impaired.

Effects on the Chemical and Biochemical Reactions in the Muscle

Hamann (1957) discussed the effects of pressure on the formation and breakdown of the activated complex in the transition state theory of chemical reaction rates. He stated that a chemical reaction proceeds through a transition state which consists of a molecular species called the activated complex. The formation of the transition state involves a change in volume from the initial state. This change is known as $\Delta V^{\ddagger}$. $\Delta V^{\ddagger}$ is made up of two terms, $\Delta V_1^{\ddagger}$, the actual change in volume of the reacting molecules in going from the initial to the transition state, and $\Delta V_2^{\ddagger}$, the volume change in the solvent surrounding the reacting molecules. $\Delta V_2^{\ddagger}$ is large only in cases where there exists alteration of intermolecular forces, for example, charge development in the formation of the activated complex. Hamann stated that $\Delta V_2^{\ddagger}$ can be influenced by either steric forces, changed configuration of reactant molecules or change in binding strength

between solute and solvent, all of which may result in solvent molecule rearrangement.

Laidler and Bunting (1973) stated that pressure decreases the reaction rate if $\triangle V^{\ddagger}$ is positive, that is if there is a volume increase in the formation of the activated complex, and the opposite is true if $\triangle V^{\ddagger}$ is negative. Formation of the myosin-ATP activated complex, a complex involved in the contractile process, appears to involve charge neutralization, with resultant release of bound water and positive $\triangle V^{\ddagger}$ (Laidler, 1955), so application of pressure would appear to retard the progress of myosin-catalyzed ATP breakdown. Ikkai and Ooi (1969) proposed that in the absence of ATP, pressure does not induce the dissociation of an actin-heavy meromyosin (a component of myosin) complex, but in the presence of ATP this dissociation is possible.

Heat treatment of dissociated myofibril proteins was the basis of a scheme for the tenderizing effect of post-rigor high-pressure-heat treatment of beef muscle proposed by Bouton et al. (1977). They suggested that application of pressure to the associated undenatured proteins of the myofibril causes dissociation of the proteins. Subsequent application of heat plus pressure results in the irreversible denaturation of the proteins in the dissociated state.

As well as effects on dissociation of myofibril protein complexes, pressure has been shown to influence the extent of hydration

of these proteins. Macfarlane (1974) suggested that increased protein hydration, leading to more tightly bound water molecules, would decrease the volume of the water molecules, a reaction favored by pressure. Additionally, in some cases, pressure treatment increased water-holding capacity of meat homogenates in salt solution and the treatment resulted in less cooking loss.

The influence of pressure on pH has been noted by Johnson et al. (1954). They pointed out that a volume decrease usually accompanies ionization due to the closer attraction of the water molecules to the charged groups and ions. Therefore, pressure increase favors ionization. Johnson et al. (1954) also noted that the influence of pressure on large molecules such as proteins is greater than that on smaller molecules since the volume changes per ionizing group are additive.

Macfarlane (1973), using six different ox muscles, found an immediate post-pressurization pH drop. Although he found no difference between ultimate pH two days post-slaughter of the pressurized and non-pressurized muscle, ultimate pH was found to be lower than the immediate post-pressurization pH. He suggested that the pattern of pH changes observed indicated the near-completion of glycolysis soon after termination of the pressure treatment.

Quality Characteristics of Meat

Tenderness, juiciness, flavor, aroma, and color are generally accepted as important factors in determining the palatability or eating quality of meat (Bratzler, 1971). Objective and subjective procedures may be used to investigate the factors which influence these quality parameters.

Bouton et al. (1975) discussed the role of myofibril protein and collagen in meat tenderness and summarized some factors affecting these two muscle components. Myofibril contribution to toughness is affected by aging, increased muscle fiber diameter as a result of myofibril contraction, and heat-induced cooking changes, including moisture loss. Collagen contribution to toughness is influenced by orientation of collagen in relation to myofibril contraction state, changes in collagen structure related to animal age, and heat-induced changes in collagen. Some of the biochemical and physiological changes affecting myofibril contraction state and some aspects of aging have been discussed previously in this review and need not be repeated. The effectiveness of different measurements of tenderness are greatly affected by the relative influence of the various tenderness determining factors.

Tenderness has been shown to have a positive correlation with extractability of beef contractile proteins by a number of workers

(Hegarty et al., 1963; Kastner et al., 1976; Cook, 1967). Cook (1967) suggested that myofibril protein solubility is an indicator of the myofibril contraction state and therefore of the myofibril contribution to toughness.

Cross et al. (1973) found that percent soluble collagen was significantly related to panel evaluations of connective tissue contribution to toughness. They also observed that total collagen and elastin concentrations did not correlate with muscle tenderness scores. Goll et al. (1964a) showed that collagen from older beef animals was more extensively and strongly cross-linked than that from younger animals. They also suggested this observation may be a partial explanation of decreasing tenderness with increasing age of an animal. Application of sufficient heat to collagen causes breakdown of collagen to gelatin, producing increased tenderness (Goll et al., 1964b). However, application of heat to meat has the opposite effect in the heat-induced denaturation of actomyosin and myofibril shortening, resulting in decreased tenderness (Davey and Gilbert, 1974a). Davey and Gilbert (1975) distinguished between the mechanisms of cold shortening and cooking shortening. They showed two distinct stages of cooking shortening, one between 40° and 50° C due to actomyosin coagulation and the second between 65° and 75° C due to connective tissue shrinkage. They pointed out that denaturation of actomyosin in addition to the fact that the meat was in rigor at time

of cooking precluded the validity of a sliding filament model of cooking shortening.

The Warner-Bratzler shear device has been one of the instruments that has been most frequently reported for measuring mechanically the tenderness of meat (Szczesniak and Torgeson, 1965). This instrument measures the amount of force required for a blade to cut through a sample core of given diameter and fiber orientation. Differences in tenderness measured by this instrument are due to either connective tissue or myofibril properties (Bouton and Harris, 1972a). Davey and Gilbert (1975) noted the importance of consideration of the lateral transmission of forces of the shearing within and among the fibers. They also noted the importance of the number of fibers per unit of muscle cross-sectional area. They proposed that the lateral force transmission in meat is a result of lateral inter-fiber connections provided by the connective tissue as well as intra-fiber connections provided by the sarcoplasmic reticulum and transverse tubular system described by Bennett (1960).

Bourne et al. (1966) studied the adaptation of the Instron Universal Testing Machine (UTM) to food texture studies. In analyzing the feasibility of the attachment of the working parts of the Warner-Bratzler device to the UTM, they concluded that the food texture measurements performed by the Warner-Bratzler device could be duplicated on the UTM. These investigators suggested that the force-distance curve obtained using the UTM could be used to obtain

maximum force, slope, area, plateau height, relaxation, and recovery. This gives more information about the food texture than does the original Warner-Bratzler device. Bouton et al. (1975) came to the conclusion that peak or initial yield shear force values gave an accurate index of the myofibril component of meat toughness. They suggested that the coagulated myofibrils of cooked meat rather than the softened collagen bear the initial force applied by the shear blade, and the additional force registered by the UTM results from compression of connective tissue and myofibrils.

Subjective evaluation of friability of beef samples in studies on tenderness was used by Cover (1959). She defined friability as the aspect of tenderness measured by "the ease with which muscle fibers broke--whether they tended to be crumbly or rubbery." She found greater friability associated with greater connective tissue tenderness and with lower shear force values.

A correlation between cooked meat juiciness and water content with special emphasis on the extent of binding of the water to the coagulated tissue was reported by Hamm (1960). He described a scheme of water-binding to meat proteins. A first layer, the monomolecular layer, of water molecules one molecule thick is tightly bound to some hydrophilic protein groups, with an actual volume contraction of the bound water. A second layer, the multimolecular layer, is formed over the first. Some water is immobilized in

capillary condensation in the small spaces of muscle fibers. Hamm defines the true hydration water of muscle as that water which is bound in the mono- and multimolecular layers. The remaining water, the majority of the water in the tissue, is defined as free water, although it is immobilized in the fiber network. Hamm (1960) noted that the presence of ATP resulted in dehydration and myofibrillar contraction.

Hamm (1960) defined the water-holding capacity (WHC) as the ability of meat to hold fast to its own or added water during the application of any force. Hamm pointed out a positive correlation between WHC and juiciness of cooked meat, and noted increased juiciness associated with higher water content of meat and tighter binding of this water to tissue. Bouton et al. (1971) used a technique involving centrifugation of muscle tissue to determine WHC. Their definition of WHC was the fraction of the total moisture content remaining after deducting the total juice loss. Total juice loss included juice expressed upon centrifugation and cooking loss where applicable. Bouton et al. (1971) found an increase in WHC and decrease in cooking losses in beef with increasing pH.

Cover et al. (1962), in discussing juiciness in meat, expressed the view that both the bound water and immobilized water discussed by Hamm (1960) contribute to overall juiciness. Bratzler (1960) separates the sensation of juiciness of cooked meat into two factors:

the sensation of wetness produced by the release of fluid upon the first few chews, and, second, sustained sensation of juiciness as a result of release of serum and the saliva-stimulating effect of fat. Cover et al. (1962) noted that total moisture losses increased and subjective juiciness scores decreased with greater final cooking temperature of beef. Lower juiciness scores were correlated with cooking weight loss in beef by Cover et al. (1957).

Since the evaluation of flavor in meat is highly subjective in nature, factors affecting flavor are difficult to define. Feed ingredients, animal age and sex, and meat storage conditions may influence flavor (Bratzler, 1971). Lawrie (1974) associated flavor development with the aging process. Macy et al. (1964) and other workers (Hornstein et al., 1960; Kramlich and Pearson, 1958; Crocker, 1948) studied meat flavor. They concluded that flavor precursors of beef were water-soluble and resided in the juice of meat, both raw and cooked. Some of these water-soluble precursors are amino acids, peptides, and nucleotides, as well as water-soluble components from adipose tissue (Dwivedi, 1975).

Meat color is an important quality characteristic of meat. Myoglobin is the main pigment responsible for meat color, but hemoglobin in blood is present also. The oxygenated form of myoglobin gives a bright red color to raw meat, however, the oxidized form is brownish (Govindarajan, 1973). Denatured heme pigments,

decomposition and polymerization of carbohydrates, fats, and proteins all contribute to the brown color of cooked meat (Bratzler, 1960).

Bratzler (1960) emphasized the necessity of sensory evaluation of meat and other foods to evaluate tenderness, flavor, juiciness, and other palatability characteristics. He discussed the value of chemical, physical, and other meat analysis methods, but assigned human judgement as the ultimate test of quality.

Lowe (1949) developed the method of chew count before swallowing as an assessment of sample tenderness. Larmond (1976) expressed the view that this method may correlate well with instrumental tenderness measurements, but is not a true indicator of tenderness due to variation in force of chew by panel members.

Method of cooking of the samples is an important part of sensory testing, and Rogers and Ritchey (1969) recommended roasting as a method that is easy to standardize. Larmond (1976) routinely obtained samples from the same anatomical location for each treatment to decrease the effect of location upon the sample. Harries et al. (1963) concluded that greater tenderness discrimination was possible using cold meat samples, compared to hot samples.

Larmond (1976) discussed various scoring systems for sensory evaluation. She mentioned unstructured scales with verbal anchors at each end and the greater discernment of sample differences possible using this scale compared to category scales. Stanley et al.

(1972) used a five-inch linear rating scale, the ends of which represented opposite extremes of the meat parameter being scored. Anchor lines were located one-half inch from the ends. Their intent in use of this scale was to avoid some of the reluctance of the judges to use the extremes of the scale.

EXPERIMENTAL PROCEDURE

Pressure Treatment and Cooking Methods

Eight pairs of roasts of the semitendinosus muscle of good and standard grade beef were supplied and one of each pair was pressure-treated by the Animal Science Department of Oregon State University. One of the two roasts of each pair was hot-boned, immediately placed in a polyethylene bag and subjected to 15,000 psi (103.5 MNm^{-2}) pressure in a pressure chamber filled with water at 38° C. One minute was required for the pressure to reach the required 15,000 psi and the muscle was then held at that pressure for two minutes. The second roast of the pair was left in the beef carcass, which was hung in a cold room at 3° C and aged for ten days, at which time the roast was excised. Treated and untreated roasts were kept refrigerated for 3° C for 22±2 days after slaughter until testing.

Roasts were oven-roasted in a Magic-Chef gas oven at 160° C to an internal temperature of 70° C. Endpoint internal temperature of each roast was recorded using a Leeds and Northrup Speedomax W 12 recorder.

Objective Testing Methods

Sampling procedures and objective tests are summarized in Figure 1 and Table 1, respectively. For all objective tests except

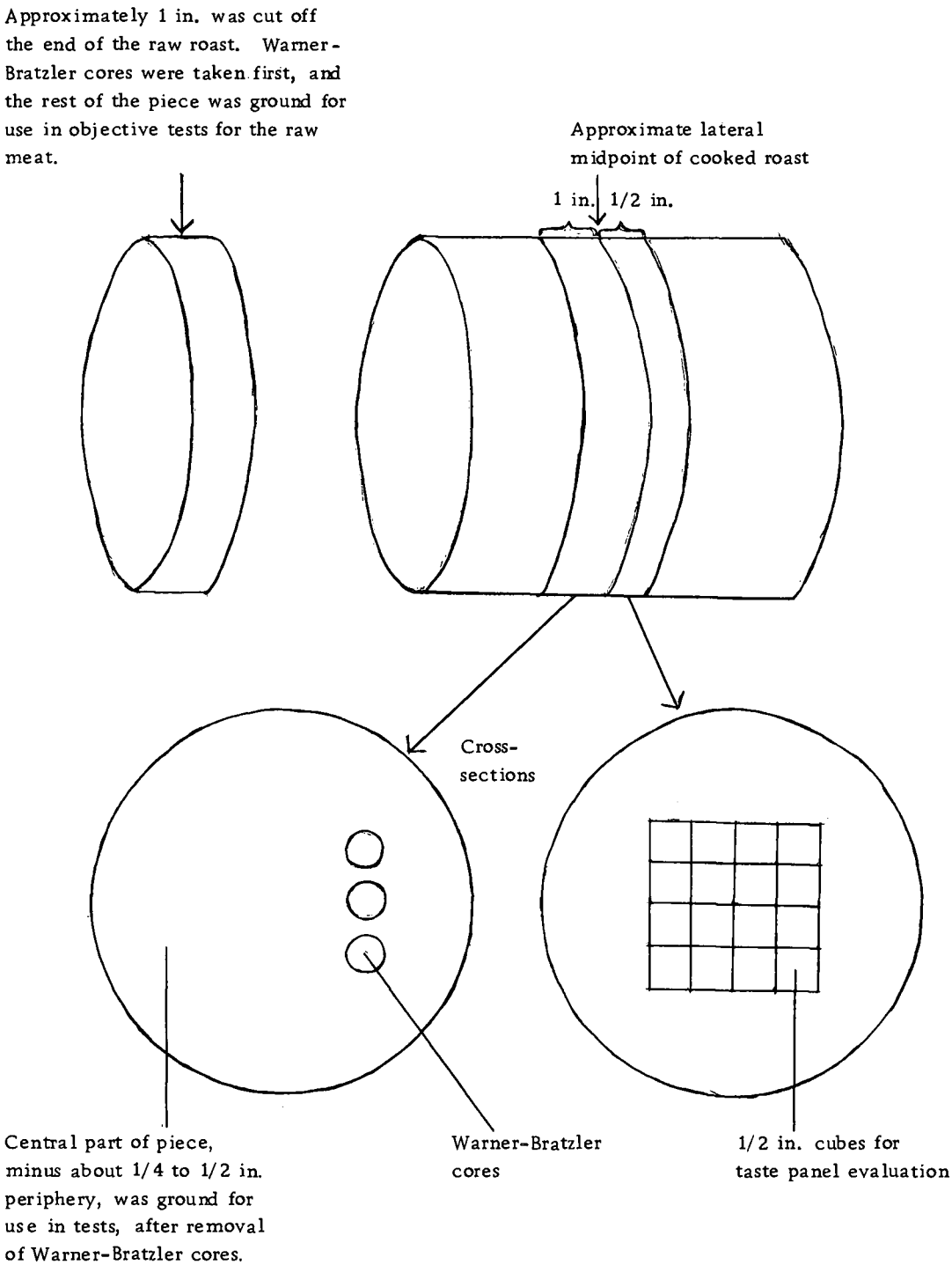

Figure 1. Sampling procedure for beef semitendinosus roasts.

Table 1. Summary of objective tests for beef semitendinosus.

The following tests were performed on both raw and cooked samples:

1. Warner-Bratzler shear
2. Myofibril protein solubility
3. Total moisture (%)
4. Water-holding capacity
5. Total nitrogen (%, dry weight)
6. Fat (%, dry weight)
7. pH

Cooking weight losses and weight of drippings were used to determine:

8. Total cooking loss (%)
9. Drip loss (%)
10. Evaporation loss (%)

the Warner-Bratzler shear and cooking losses, the sample was ground three times in an Oster electric food grinder. The sample was kept in a tightly-covered jar under refrigeration until testing, within ten hours after grinding sample. Samples for fat determination were frozen (-18° C) until analysis, within one month.

Tenderness was evaluated using the Warner-Bratzler shear test to determine the force required to cut through cooked and raw sample cores. This test was performed using the Warner-Bratzler attachment on the Instron Universal Testing Machine, model 1132. Crosshead speed and chart speed were, respectively, 20 inches per minute, and 12 inches per minute. Triplicate cylindrical sample cores of one-half inch diameter were used (Figure 1) and the peak height in pounds of force was recorded.

An indirect indication of tenderness was obtained from the myofibril protein solubility of both cooked and raw duplicate samples, using the method of Haga et al. (1965) (Appendix). Minced muscle was immersed in Weber-Edsall solution, a potassium chloride-bicarbonate-carbonate buffer for 20±4 hours at 3° C and extracted protein was precipitated by dilution with water. Extracted and total nitrogen was determined by the micro-Kjeldahl method (AOAC, 1970). Extracted nitrogen was expressed as a percentage of the total nitrogen content.

Water-holding capacity and total moisture of raw and cooked

meat and cooking losses, including total, drip, and evaporation were used to assess juiciness qualities. A modification of the method of Bouton et al. (1971) was used to determine water-holding capacity. Difference in sample weight before and after centrifugation at 22, 500XG for one hour was used to calculate expressed juice. Total cooking loss as well as expressed juice was used to calculate total juice loss, from which water-holding capacity was calculated. Water-holding capacity was determined in duplicate for each roast, raw and cooked (Appendix).

Total moisture was determined in duplicate for each roast, raw and cooked, using the method of AOAC (1970). Total, drip, and evaporation cooking losses were calculated using cooking weight loss of roasts and drip weight (Appendix).

The pH of raw and cooked samples was measured on a Beckman 2000 model G pH meter. Six grams of previously-ground sample were homogenized with 30 ml distilled water for 30 seconds at medium speed in a Virtis "45" homogenizer. The pH of the slurry was recorded.

Nitrogen content of each roast before and after cooking was analyzed in duplicate according to the micro-Kjeldahl method (AOAC, 1970). Fat content, using a micro-Soxhlet apparatus and the ether extract method of AOAC (1970), was determined in duplicate on raw and cooked samples from each roast. Nitrogen and fat analysis

results are in Appendix.

Subjective Testing Methods

Panelists were selected by their performance during screening sessions, and then trained for beef sensory evaluation, using the scoresheet shown in Figure 2. The panel of eight faculty and staff members of the Foods and Nutrition Department of Oregon State University evaluated the tenderness, friability, juiciness, and flavor of one sample of each roast (Figure 2). Number of chews was determined on a duplicate sample. The one-half inch cubes of each sample were evaluated at room temperature under red light to alleviate color bias. For the first four parameters, a 10 cm continuous line scoring scale was used, the ends of which were anchored by extremes of the quality parameter being scored. The judge marked the point on each line corresponding to her evaluation of the parameter for each sample. The distance of the mark from the lower end of the scale was measured to 0.01 cm with a ruler and the judge's evaluation was assigned that value for statistical analysis of the results.

Statistical Analysis

The paired-t test was used to compare the pressure-treated muscle with the untreated muscle for each of the parameters tested. The level of significance was established at 0.05 for the paired-t

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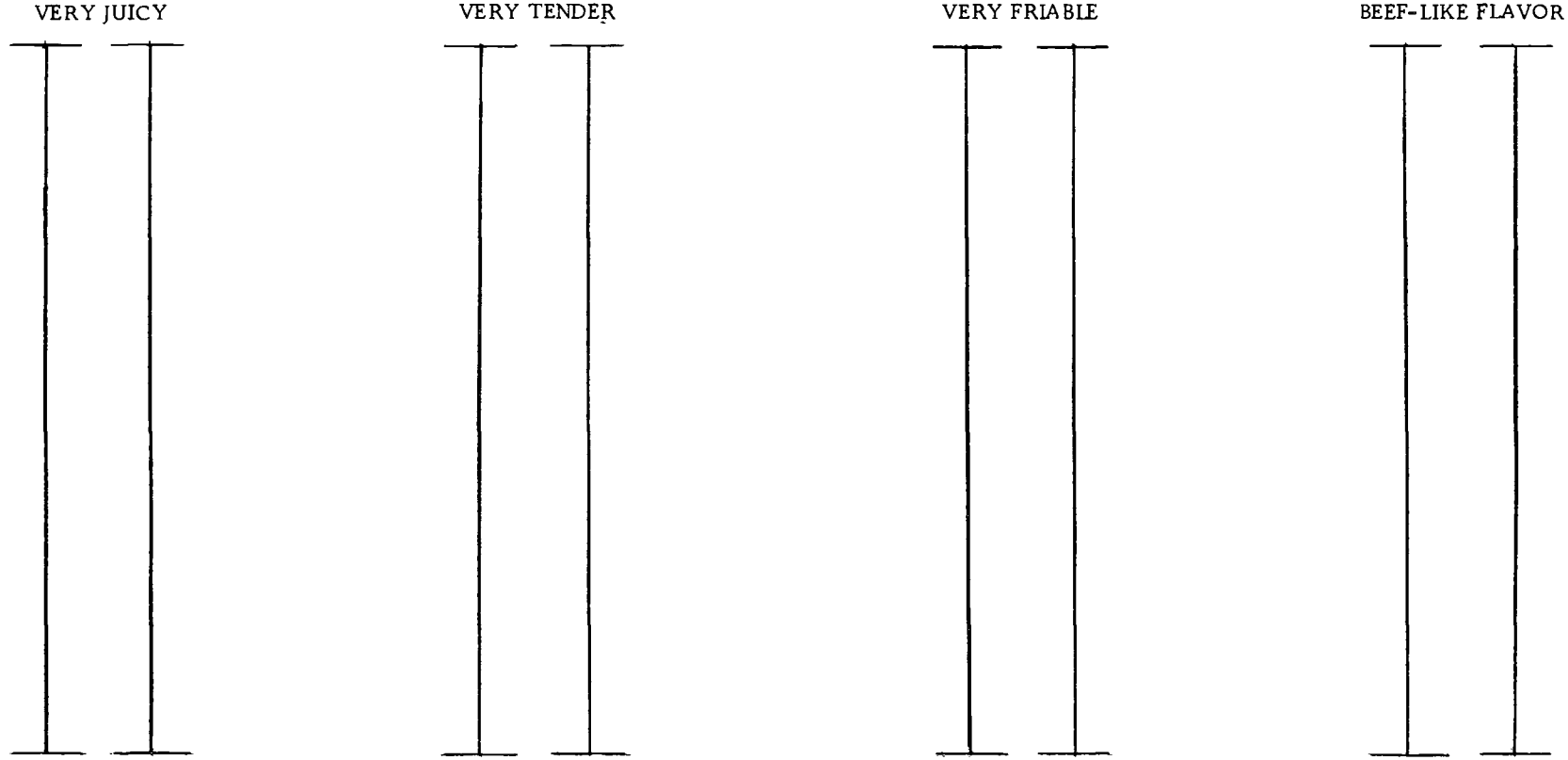
SENSORY EVALUATION OF PRESSURE-TREATED AND NON PRESSURE-TREATED BEEF



CODE___ ___ ___ ___ ___ ___

CODE NO. OF CHEWS

___ ___

___ ___

TWO CODED SAMPLES ARE PRESENTED: ONE FOR DETERMINATION OF NUMBER OF CHEWS, THE OTHER TO EVALUATE JUICINESS, TENDERNESS, FRIABILITY AND FLAVOR BY PLACING YOUR JUDGEMENT ALONG THE INDICATED LINE. PLEASE USE WARM-UP SAMPLE FROM THE CENTER OF THE DISH BEFORE BEGINNING EVALUATION OF CODED SAMPLES.

COMMENTS:

Figure 2. Scoresheet for sensory evaluation of beef semitendinosus roasts.

values, which were calculated using the Hewlett-Packard 9810A Model 10 Calculator with Statistics Block (Hewlett-Packard Co., 1971). Correlation coefficients were determined for selected experimental parameters using the OS3 Statistical Interactive Programming System (SIPS) (Guthrie et al., 1974). Calculated paired-t values and correlation coefficients are in Appendix.

RESULTS AND DISCUSSION

Myofibril protein solubility values (Table 2) were not significantly influenced by pressurization of the semitendinosus muscle. Interpretation of the myofibril protein solubility scores indicated no significant difference in the contractile state and the extent of actin-myosin interdigitation between the pressure-treated and untreated muscle. Myofibril protein solubility was suggested as an indicator of contractile state of myofibril proteins by Cook (1967). During cooking of the pressure-treated and untreated semitendinosus muscle in the current experiment, myofibril protein solubility decreased. Hamm and Deatheridge (1960) also reported decreased solubility of fibrillar proteins as a result of application of heat to beef muscle.

Warner-Bratzler shear values (Table 3) tended to be lower for pressure-treated than for untreated semitendinosus muscle, but the difference was not significant. These values indicated a slight increase in tenderness of the pressure-treated meat. The results of this research are similar to those observed in pressurization of ox semitendinosus muscle (Macfarlane, 1973). Macfarlane reported lower Warner-Bratzler shear values with pressure-treated versus untreated raw and cooked ox. Lower Warner-Bratzler values for cooked than raw samples in this current research, both pressure-treated and untreated, suggested that a portion of the raw meat

Table 2. Extracted myofibril nitrogen (% of total nitrogen content) of beef semitendinosus roasts, raw and cooked.

Replication	Raw		Cooked	
	Untreated	Pressure-Treated	Untreated	Pressure-Treated
1	2.19	3.49	0.59	0.26
2	3.34	3.96	0.21	0.53
3	2.63	2.18	0.22	1.23
4	2.75	4.43	1.42	0.89
5	1.85	2.61	0.61	0.26
6	2.64	1.20	0.66	1.43
7	1.89	2.90	1.10	0.83
8	1.62	1.60	0.48	0.42
Mean	2.36	2.80	0.66	0.73
Standard Deviation	0.58	1.13	0.42	0.44

Table 3. Warner-Bratzler shear peak heights (pounds) for beef semitendinosus roasts, raw and cooked.

Replication	Raw		Cooked	
	Untreated	Pressure-Treated	Untreated	Pressure-Treated
1	7.49	12.22	4.64	4.52
2	13.61	11.38	7.68	7.74
3	13.47	11.44	5.92	9.20
4	10.99	10.81	13.47	6.75
5	10.95	7.99	7.81	6.86
6	14.61	12.22	9.76	12.34
7	13.83	10.09	11.83	8.10
8	14.27	11.14	9.00	8.63
Mean	12.40	10.91	8.76	8.02
Standard Deviation	2.43	1.37	2.92	2.26

toughness is contributed by collagen, which is softened during cooking (Goll et al., 1964c).

Myofibril protein and collagen are the components measured by the Warner-Bratzler shear device (Bouton and Harris, 1972a). In this current research the Warner-Bratzler shear values were significantly correlated ($r = +.79$, $r = +.82$; $P < 0.05$) with myofibril protein solubility for both pressure-treated and untreated cooked meat. These unexpected contradictory correlations may be due to the less tender nature of the semitendinosus muscle (Ginger and Weir, 1958). Perhaps the high collagen content of the tough semitendinosus was the major factor evaluated by the Warner-Bratzler device. The heat-induced softening of the collagen may have more than compensated for the toughening effect of the myofibril protein denaturation.

Hegarty et al. (1963) found a negative correlation between myofibril protein solubility and shear force and a positive correlation between the former and panel tenderness evaluations of beef longissimus dorsi, a tender muscle. This research did not yield the same significant correlations.

Panel scores for tenderness and number of chews (Table 4) did not indicate a significant difference between pressure-treated and untreated beef. These two parameters were significantly correlated ($r = +.82$, $P < 0.05$) for the untreated cooked meat. Number of chews was found by Lowe (1949) to give an evaluation of comparative tenderness in different samples. Macfarlane's (1973) taste panel results

Table 4. Panel evaluation of tenderness and number of chews before swallowing of beef semitendinosus roasts.

Replication	Tenderness		Number of Chews	
	Untreated	Pressure-Treated	Untreated	Pressure-Treated
1	5.77	3.27	39	31
2	3.92	4.54	30	38
3	2.83	4.27	34	39
4	7.96	2.53	47	32
5	2.44	4.17	27	30
6	3.06	4.81	36	41
7	3.24	7.73	24	26
8	3.16	2.73	34	33
Mean	4.05	4.26	34	34
Standard Deviation	1.88	1.64	7.20	5.12

showed greater tenderness of pressure-treated ox longissimus dorsi compared to the paired untreated muscle.

Friability panel scores (Table 5) showed no significant difference between muscles from pressure-treated versus untreated beef. Friability is an indication of one aspect of tenderness, the relative ease of breaking of the meat fibers (Cover, 1959). In the current research, panel friability values were correlated to two tests which evaluated tenderness. Friability and panel scores for tenderness of cooked untreated meat were significantly ($r = +.86$; $P < 0.05$) related as were friability and Warner-Bratzler value of the cooked pressure-treated meat ($r = +.73$; $P < 0.05$). The positive Warner-Bratzler-friability correlation may be explained by the high collagen content of the semitendinosus. This would be the same mechanism previously suggested for the significant positive Warner-Bratzler-myofibril protein solubility correlation for cooked meat. The tenderness-related panel observations were in agreement with the results of the objective tenderness tests, the Warner-Bratzler shear and myofibril protein solubility, in showing no significant effect of pressure-treatment on tenderness.

The objective water-holding capacity and total moisture values (Tables 6 and 7) showed no significant difference between the pressure-treated and untreated meat, raw and cooked. However, treated cooked meat had a slightly higher mean water-holding capacity.

Table 5. Panel evaluation of friability of beef semitendinosus roasts.

Replication	Untreated	Pressure-Treated
1	6.98	4.43
2	4.76	4.38
3	5.75	4.90
4	7.94	5.40
5	4.27	5.14
6	4.24	7.00
7	3.14	5.37
8	4.30	4.63
Mean	5.17	5.16
Standard Deviation	1.88	0.84

Table 6. Water-holding capacity of beef semitendinosus roasts, raw and cooked.

Replication	Raw		Cooked	
	Untreated	Pressure-Treated	Untreated	Pressure-Treated
1	.5885	.5788	.4772	.5167
2	.6028	.6386	.5878	.5403
3	.6126	.5893	.4703	.4946
4	.6187	.6948	.4499	.5449
5	.5814	.5288	.4724	.4942
6	.6179	.6542	.4734	.4514
7	.6618	.6427	.3751	.3925
8	.6704	.7190	.4721	.4941
Mean	.6193	.6308	.4723	.4911
Standard Deviation	.0319	.0626	.0577	.0497

Table 7. Total moisture (% of total weight) of beef semitendinosus roasts, raw and cooked.

Replication	Raw		Cooked	
	Untreated	Pressure-Treated	Untreated	Pressure-Treated
1	73.98	74.47	70.32	69.77
2	74.44	67.15	68.60	68.97
3	71.70	73.45	67.73	70.72
4	72.44	73.25	71.60	69.45
5	73.77	72.58	69.12	70.51
6	73.58	74.29	69.97	71.07
7	75.02	76.54	74.39	73.88
8	72.57	75.24	68.38	69.41
Mean	73.44	73.37	70.01	70.47
Standard Deviation	1.12	2.80	2.15	1.56

Hamm (1960) positively correlated water-holding capacity with cooked meat juiciness. He suggested that juiciness is a function of the total moisture content of the meat as well as how tightly-bound this water is to the tissue. This current research did not indicate a significant water-holding capacity-juiciness correlation.

Panelists in the current study did not differentiate between the pressure-treated and untreated meat with regard to juiciness (Table 8). Lower taste panel ranking for juiciness of pressure-treated ox longissimus dorsi than untreated muscle evaluated two days post-slaughter has been reported by Macfarlane (1973). Hamm (1960) attributed part of the aging-induced rise in water-holding capacity to pH increase of the meat which accompanies aging. Perhaps the similarity in juiciness of the pressure-treated and untreated meat evaluated 22±2 days post-slaughter observed in this research, in contrast to Macfarlane's findings, is partially due to greater post-treatment increase in hydration of the pressure-treated meat proteins. Greater hydration increase of the pressure-treated meat proteins could conceivably equalize juiciness between the treated and untreated meat, despite possibly lower initial juiciness of the treated meat. This increase in hydration could be associated with the development of significantly ($P < 0.05$) greater ultimate pH 22±2 days post-slaughter with pressure-treated meat, a suggestion supported by the correlation ($r = +.83$; $P < 0.05$) between the water-holding capacity and pH of the

Table 8. Panel evaluation of juiciness of beef semitendinosus roasts.

Replication	Untreated	Pressure-Treated
1	2.11	3.70
2	5.35	3.71
3	2.84	4.30
4	3.19	2.48
5	3.15	3.93
6	2.05	3.12
7	4.10	2.87
8	4.28	2.89
Mean	3.38	3.38
Standard Deviation	1.13	0.62

treated raw meat.

The pH for the pressure-treated meat, both raw and cooked, was significantly ($P < 0.05$) higher than for the untreated meat (Table 9). Macfarlane (1973) found no difference in ultimate pH between pressure-treated and untreated ox muscle. However, his pH values were obtained two days post-slaughter, whereas, the pH measurements in this research were taken on the cooking day, 22±2 days post-slaughter. Perhaps the difference in ultimate pH between pressure-treated and untreated meat observed in this research was at least partially a result of pressure-induced changes in the proteins. These pressure-induced changes may have affected the course of pH change during the time before testing was performed. Macfarlane (1973) suggested that pressurization may lead to the presence of more hydrophilic groups in the structural proteins. This condition could possibly affect subsequent pH changes. Aging brings about an increase in pH (Hamm, 1960), but the explanation for the increase is not clear. Proteolytic cleavage of meat peptide bonds resulting in net charge increase was suggested by Hamm (1960) as one possible cause of the pH increase during aging. In contrast, Goll et al. (1970) concluded that post-mortem proteolysis does not occur in a significant amount in muscle and therefore is not a major cause of aging-induced changes in proteins. Perhaps pressurization facilitates proteolysis during aging, resulting in greater pH increase, a

Table 9. pH of beef semitendinosus roasts, raw and cooked.

Replication	Raw[a]		Cooked[a]	
	Untreated	Pressure-Treated	Untreated	Pressure-Treated
1	5.62	5.70	5.77	5.93
2	5.65	5.74	5.86	5.98
3	5.59	5.78	5.80	5.98
4	5.62	6.02	5.71	6.06
5	5.60	5.70	5.73	5.99
6	5.60	5.81	5.72	5.99
7	5.79	6.04	5.81	6.12
8	5.62	6.21	5.79	6.02
Mean	5.64	5.88	5.77	6.01
Standard Deviation	0.06	0.19	0.05	0.06

[a]Treated roasts significantly different ($P < 0.05$) from untreated roasts.

suggestion requiring experimental verification.

Bouton et al. (1971) found correlations between increase in pH and both increase in water-holding capacity and decrease in cooking loss with beef. This current research found a significant correlation ($r = +.83$; $P < 0.05$) between water-holding capacity and pH for the raw pressure-treated meat. The higher pH ($P < 0.05$) and trend toward higher water-holding capacity and decreased drip loss for the pressure-treated meat in this research support Bouton et al.'s (1971) association of the three parameters.

Total cooking loss and evaporation cooking loss values (Table 10) were not significantly different when the pressure-treated and untreated roasts were compared. However, drip cooking loss values (Table 10) were significantly ($P < 0.05$) less for the pressure-treated roasts. This observation may be interpreted as greater capacity for retention of water in the pressure-treated roasts during the application of heat. This interpretation is supported by the trend toward higher water-holding capacity in pressure-treated cooked meat. Pressure treatment would not appear to be an influential factor in loss of volatiles during cooking. Macfarlane (1973), using cooking and storage weep loss data, found less moisture loss for pressure-treated ox samples than untreated samples, despite higher pressure-treated meat storage weep loss. He suggested that this may be a result of pressure-induced exposure of hydrophilic groups on the

Table 10. Total, evaporation, and drip cooking losses of beef semitendinosus roasts (% of initial weight of roast).

Replication	Total		Evaporation		Drip[a]	
	Untreated	Pressure-Treated	Untreated	Pressure-Treated	Untreated	Pressure-Treated
1	14.55	17.62	13.23	16.17	1.32	1.45
2	14.62	15.23	10.83	13.98	3.79	1.25
3	20.98	14.49	18.32	13.71	2.66	0.78
4	11.20	16.22	10.11	14.11	1.09	2.11
5	16.93	15.74	13.30	15.33	3.63	0.41
6	17.91	12.41	14.26	12.01	3.65	0.40
7	15.25	17.21	13.10	15.70	2.15	1.51
8	19.33	14.88	15.43	13.96	3.90	0.92
Mean	16.35	15.48	13.57	14.37	2.77	1.10
Standard Deviation	3.10	1.65	2.57	1.33	1.44	0.59

[a]Treated roasts significantly different ($P < 0.05$) from untreated roasts.

myofibril proteins, leading to hydrogen-bonding of water with these groups.

Panel flavor evaluations (Table 11) tended towards higher flavor scores for the pressure-treated meat in comparison to the untreated meat. Drip loss and flavor in the untreated meat were negatively correlated, although not significantly. Macy et al. (1961) and a number of other workers (Hornstein et al., 1960; Kramlich and Pearson, 1958; Crocker, 1948) determined that the flavor precursors of beef were water-soluble and resided in the juice of meat, both raw and cooked. Some of these water-soluble precursors are amino acids, peptides, and nucleotides as well as water-soluble components from adipose tissue (Dwivedi, 1975). With less cooking loss of water, and therefore supposedly less loss of the water-soluble flavor precursors, the likelihood of a more flavorful pressure-treated product seems possible. Perhaps the higher flavor score tendencies associated with the pressure-treated meat in the current study are at least partially a result of the lower drip loss with the pressure-treated meat.

The relatively long time interval after slaughter until the time of testing may have reduced the number of significant differences in some of the test results between the pressure-treated and untreated meat. Aging-associated changes in the meat, including tenderization of the untreated meat due to loss of adhesion between

Table 11. Panel evaluation of flavor of beef semitendinosus roasts.

Replication	Untreated	Pressure-Treated
1	5.06	4.78
2	4.02	6.04
3	4.16	4.98
4	5.82	4.96
5	4.24	5.33
6	5.30	7.23
7	6.43	5.89
8	4.40	5.11
Mean	4.93	5.54
Standard Deviation	0.88	0.82

the myofibrils or possible loss of fiber tensile strength (Davey and Gilbert, 1969), may be the main cause of lack of differences. Perhaps testing sooner after slaughter would have facilitated detection of any possible differences between the pressure-treated and untreated meat which may have become minimal with increasing time post-slaughter.

It is suggested that the main pressure-induced changes detected as a result of this research are related to increased exposure of hydrophilic protein groups (Macfarlane, 1973), causing a significant alteration of pH and alteration of the capacity of the cooked meat tissue to bind water. Any pressure-induced changes in the myofibril proteins which might theoretically affect the tenderness characteristics were too small to be significant.

More research is definitely needed in the area of the effects of applied pressure on muscle and resultant effects on meat quality. Information about the actual contractile state of the myofibril proteins during and after pressurization and their water-binding characteristics is needed. Pressure-induced changes in muscle cell metabolism, including ATP metabolism and ATP interaction with myofibril proteins during and after pressure treatment merit investigation.

SUMMARY

The effects of pre-rigor high-pressure treatment on tenderness, juiciness, and flavor of beef semitendinosus muscle were investigated. Objective tenderness indicators, Warner-Bratzler shear and myofibril protein solubility, as well as panel evaluations of tenderness, friability, and number of chews showed no significant difference in tenderness between the pressure-treated and untreated meat.

Tests used to assess juiciness qualities, total moisture, water-holding capacity, total and evaporation cooking loss, and panel juiciness scores showed no significant difference as a result of the treatment. However, drip cooking loss was found to be significantly less in the pressure-treated meat. pH values for the pressure-treated meat, both raw and cooked were significantly higher.

Panel flavor evaluations showed no difference as a result of pressurization. Pressure treatment was found to result in meat that was comparable to unpressurized, traditionally aged meat in tenderness, juiciness and flavor.

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APPENDIX

Details of some Objective Testing Methods

Extraction of Myofibril Protein

Method of Haga et al. (1965). One gram of accurately-weighed minced muscle was mixed with 4.4 ml of Weber-Edsall solution (0.6M KCl, 0.04M $NaHCO_3$, 0.01M Na_2CO_3) and extracted for 20±4 hours at 3° C. The mixture was stirred occasionally. After the extraction period, the suspension was centrifuged at 5,000×G in a Sorvall Superspeed Model RC2-B refrigerated centrifuge at 3° C for ten minutes. The supernatant was poured off and was diluted with 34 ml cold water and neutralized with cold 0.02N HCl to pH 7.0. Upon dilution and neutralization, a precipitate formed which was separated by centrifugation at 5,000×G for ten minutes at 3° C. The supernatant was discarded and the precipitate was dissolved in 5 ml cold 0.6M KCl and the solution was clarified by centrifugation at 10,000×G for 30 minutes at 3° C. The supernatant was transferred to a 30 ml micro-Kjeldahl digestion flask and nitrogen content of the supernatant was determined using the micro-Kjeldahl method described in AOAC (1970). The extracted nitrogen was expressed as a percentage of the total nitrogen of the original weight of the meat sample extracted, also determined by the AOAC (1970) micro-Kjeldahl method.

Measurement of Water-Holding Capacity

Modification of Method of Bouton et al. (1971). Two grams of ground muscle were accurately weighed into a plastic centrifuge tube. Tubes were centrifuged at 22, 500XG for one hour in a Sorvall Superspeed Model RC2-B refrigerated centrifuge at 3° C. After centrifugation, the muscle pellet was carefully removed from the tube with a spatula, blotted with tissue to remove excess water and weighed. Liquid loss was determined by the difference in pre- and post-centrifugation weight.

Expressed juice (EJ) was the loss in weight after centrifugation expressed as a percentage of the initial weight of the uncooked sample. Expressed juice and percent total cooking loss were added to give the total juice loss (TJL). Water-holding capacity, calculated as shown below, was the fraction of the total moisture content (TW) of the uncooked meat remaining after subtraction of the total juice loss.

$$WHC = \frac{(TW - TJL)}{TW} = 1 - \frac{TJL}{TW}$$

In the case of raw meat, where there was no cooking loss, the following equation applied:

$$WHC = 1 - \frac{EJ}{TW}$$

The fat content and total moisture of each roast, raw and cooked,

were determined by the standard method of AOAC (1970), and a correction was made so that the TJL and TW were on a fat-free basis.

Calculation of Cooking Losses

Cooking losses were calculated using the following formulae:

1. $$\text{\% Total cooking loss} = \frac{(\text{weight raw roast} - \text{weight cooked roast})}{\text{weight raw roast}} \times 100$$

2. $$\text{\% Drip cooking loss} = \frac{(\text{final pan weight} - \text{initial pan weight})}{\text{weight raw roast}} \times 100$$

3. $$\text{\% Evaporation cooking loss} = \text{\% Total cooking loss} - \text{Drip cooking loss}$$

Table 12. Fat content (% of dry weight) of beef semitendinosus roasts, raw and cooked.

Replication	Raw		Cooked	
	Untreated	Pressure-Treated	Untreated	Pressure-Treated
1	11.34	5.25	12.23	15.12
2	7.74	30.10	9.87	21.10
3	13.87	9.71	17.63	17.49
4	13.49	13.04	13.16	18.27
5	9.61	17.94	18.00	17.80
6	12.41	9.72	13.17	21.32
7	0.60	6.09	1.82	2.93
8	13.07	9.43	17.87	13.94
Mean	10.27	12.66	12.96	16.00
Standard Deviation	4.43	8.09	5.41	5.86

Table 13. Nitrogen content (% of dry weight) of beef semitendinosus roasts, raw and cooked.

Replication	Raw		Cooked	
	Untreated	Pressure-Treated	Untreated	Pressure-Treated
1	13.87	14.30	13.48	13.03
2	13.69	9.77	14.36	11.96
3	13.00	13.45	12.61	12.98
4	13.72	13.05	14.01	12.50
5	13.69	13.27	12.60	12.44
6	13.44	13.89	13.59	11.93
7	15.25	15.09	15.50	15.01
8	13.34	14.26	12.27	13.04
Mean	13.75	13.39	13.55	12.86
Standard Deviation	1.41	1.60	1.08	0.98

Table 14. Paired-t statistics to evaluate differences between the untreated and pressure treated beef semitendinosus roasts.

Parameter	t-statistic	Significance
Warner-Bratzler		
raw	-1.54	N. S.
cooked	-0.65	N. S.
Myofibril protein solubility		
raw	1.20	N. S.
cooked	0.35	N. S.
Total moisture		
raw	-0.06	N. S.
cooked	0.84	N S.
Water-holding capacity		
raw	0.74	N. S.
cooked	1.27	N. S.
Cooking losses		
Total	-0.58	N. S.
Drip	-2.85	$P < 0.05$
Evaporation	0.72	N. S.
Total nitrogen		
raw	-0.67	N. S.
cooked	-1.81	N. S.
Fat		
raw	0.71	N. S.
cooked	1.75	N. S.
pH		
raw	3.82	$P < 0.05$
cooked	7.02	$P < 0.05$
Tenderness	0.19	N. S.
No. of Chews	0	N. S.
Friability	-0.02	N. S.
Juiciness	-0.02	N. S.
Flavor	1.59	N. S.

Table 15. Coefficients of correlation between selected parameters used for evaluation of beef semitendinosus roasts.

Correlated Parameters	Coefficient of correlation	Significance
Warner-Bratzler shear (cooked) & panel tenderness		
untreated	+.33	N. S.
treated	+.28	N. S.
Warner-Bratzler shear (raw) & myofibril protein solubility (raw)		
untreated	+.10	N. S.
treated	+.10	N. S.
Warner-Bratzler shear (cooked) & myofibril protein solubility (cooked)		
untreated	+.82	P <0.05
treated	+.79	P <0.05
Warner-Bratzler shear (cooked) & friability		
untreated	.00	N. S.
treated	+.73	P <0.05
Myofibril protein solubility (cooked) & panel tenderness		
untreated	+.61	N. S.
treated	+.28	N. S.
Myofibril protein solubility (raw) & pH (raw)		
untreated	+.20	N. S.
treated	+.14	N. S.
Myofibril protein solubility (cooked) & pH (cooked)		
untreated	+.56	N. S.
treated	+.22	N. S.
Panel tenderness & number of chews		
untreated	+.82	P <0.05
treated	+.24	N. S.
Panel tenderness & friability		
untreated	+.86	P <0.05
treated	+.27	N. S.
WHC (raw) & juiciness		
untreated	+.39	N. S.
treated	-.81	P <0.05
WHC (cooked) & juiciness		
untreated	+.34	N. S.
treated	+.18	N. S.
WHC (raw) & pH (raw)		
untreated	+.53	N. S.
treated	+.83	P <0.05
WHC (cooked) & pH (cooked)		
untreated	+.37	N. S.
treated	-.54	N. S.
WHC (raw) & flavor		
untreated	+.40	N. S.
treated	+.16	N. S.
WHC (cooked) & flavor		
untreated	-.76	P <0.05
treated	-.46	N. S.
% drip cooking loss & flavor		
untreated	-.62	N. S.
treated	-.42	N. S.
% drip cooking loss & juiciness		
untreated	+.38	N. S.
treated	-.53	N. S.
% drip cooking loss & pH (raw)		
untreated	-.21	N. S.
treated	+.34	N. S.
% drip cooking loss & pH (cooked)		
untreated	+.26	N. S.
treated	+.41	N. S.
% total cooking loss & pH (raw)		
untreated	-.30	N. S.
treated	+.05	N. S.
% total cooking loss & pH (cooked)		
untreated	+.17	N. S.
treated	+.22	N. S.