# THE INFLUENCE OF TENSION DURING CHILLING ON PRE-RIGOR EXCISED BOVINE MUSCLES

by

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THE INFLUENCE OF TENSION DURING CHILLING ON

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# INTRODUCTION

Post-mortem condition of the muscle fiber, as it relates to tenderness, has drawn considerable interest in recent years. Its structural condition has been shown to vary as to differences in contraction and distortion in rigor, due to temperature change, and as a result of different amounts of tension on the muscle.

With technological advancements in processing and preservation of meat and meat products more rapid and economical methods of meat fabrication may be advocated in the near future. The concept of pre-rigor fabrication of pork is one such method. Steak by the muscle and nearly 100% fabricated meat may be others. However, new ideas and methods are not without problems. One such problem often overlooked is the contractile state of the muscle when excised, whether removed intact or as a part of a retail cut, prior to the onset of rigor mortis. It has been shown that pre-rigor excised muscles of the bovine have incurred considerably less extensibility and tenderness than post-rigor excised muscles. In this regard it has been shown that different internal strains exist in muscles of the vertically suspended carcass, thus causing some muscles to be in a stretched-restrained condition whereas others are in a shortened condition with the onset of rigor, and ultimately effecting tenderness.

This study was designed to elucidate the relationship of muscle tension to tenderness, fiber diameter, sarcomere length, and the amount of fiber distortion resulting from various pre-rigor levels of tension.

The relationship of the previously mentioned structural characteristics to tenderness were also studied. The semitendinosus and semimembranosus muscles were used because of their parallel fiber orientation and relatively low amounts of intramuscular fat. The semitendinosus muscle was considered a muscle of high connective tissue content and representative of a locomotive muscle. The semimembranosus muscle was considered a muscle of moderate connective tissue content and representative of a support muscle.

# REVIEW OF LITERATURE

The information reported herein will be confined, in general, to (1) the gross anatomy of skeletal muscle, (2) the microstructure of skeletal muscle, (3) the mechanism of muscle contraction, (4) red and white muscle fibers, (5) connective tissue and fat, (6) inherent variations of fiber diameter, (7) post-mortem variations in fiber structure, and (8) structural variations of the muscle fiber in relation to tenderness.

## Gross Muscle Anatomy

Muscles are highly specialized organs, which are characterized by their ability to contract in a definite manner when stimulated (Sisson and Grossman, 1953). There are three main kinds of muscular tissue: (1) skeletal or striated, (2) smooth or non-striated, and (3) cardiac. Skeletal muscle will be considered in this review. Skeletal muscles are for the most part connected directly or indirectly to the skeleton; they cover the greater part of the skeleton, and play an important part in determining the form of the animal.

Muscles are generally red in color and the shade varies among different muscles and under various conditions. The form of a muscle is determined by its function, which, in turn, requires compromise between power, speed, and range of movement (Lockhart, 1960). The forms vary considerably, however are definite enough to be classified as triangular, quadrilateral, fusiform, etc.

Muscles attach to bones in most cases, but many muscles are attached to cartilage, ligaments, fascia, or the skin. The most proximal attachment, and in many cases the less motile, is referred to as the origin; and the distal attachment, often the more movable attachment, is referred to as the insertion. In all cases the attachment is made by a band or sheath of fibrous tissue (tendon or aponeurosis). The muscular tissue does not come into direct contact with the point of attachment.

Muscular action is found to be rather complex in some cases and relatively simple in others. Muscles can be classified as to type of action, that is, muscles which concur in action are termed synergists; those which have opposite actions are antagonists (Sisson and Grossman, 1953). In any case, muscular action is a result of contraction. It has been found that the muscle fiber in a fully stretched state can contract to 57% of its length (Haines, 1934), with this amount generally corresponding to the degree of movement permitted at the joint between the bones involved. However, some muscles are so short that they cannot execute full movement between their attachments. Some muscles often shorten while others become longer with the same movement of the limb, but in every case the importance of muscular movement in relation to gravity concerns every muscle and movement (Lockhart, 1960).

The consideration of structure involves the direction of the muscle fibers, the arrangement of the tendons, the synovial membranes, and any other accessory structures (Sisson and Grossman, 1953). In long muscles of the limbs, the number of origins (heads) will vary from one to several, and fusiform muscles may have either one or two bellies with an intermediate tendon. In most muscles the muscle fibers

join the tendon at an acute angle (pennate form). Unipennate, fibers arranged on one side of the tendon; bipennate, fibers arranged on both sides of the tendon; and multipennate muscles can be found.

The muscle blood supply is quite extensive, as might be expected. Blood vessels are most frequently found in the connective tissue septa of the muscle, but also invade the muscular mass of fibers.

The nerve supplying a muscle contains both motor and sensory fibers, with almost half of them sensory. Each muscle is a company of neuromotor units, with each unit comprising a nerve cell, its axon, and its group of muscle fibers. All efferent nerves to voluntary (skeletal) muscle stimulate the muscle to contract; there are no efferent nerves to produce relaxation. Relaxation is achieved by reducing or terminating the neuromotor stimuli (Lockhart, 1960). Vessels and nerves embedded in the connective tissue enter a muscle at a definite cleft, as a rule, the neurovascular hilum (Brash, 1955).

The accessory structures associated with the muscles are the synovial membranes and the fasciae. The synovial membranes are thin-walled sacs. Two forms are generally found: a synovial bursa which is a simple sac and interposed at a point of unusual pressure between a tendon or muscle and some underlying structure, most frequently a prominence of the skeleton; a synovial sheath where the sac is folded around the tendon so that two layers can be distinguished. An inner layer which is adherent to the tendon while the outer layer lines the canal in which the tendon lies. A synovial sheath may belong to two or more tendons in common; in such cases the synovial membrane is reflected from one tendon to another (Sisson and Grossman, 1953).

The fasciae are sheets of connective tissue, composed mainly of

bundles of white (collagenous) fibers, with a greater or less admixture of elastic fibers. Two layers: a superficial and a deep layer can usually be distinguished. The deep fascia may be slightly adherent to the underlying structures, but in many places it is attached to the skeleton, ligaments, and tendons. In many places laminae are given off from the deep fascia, pass between the muscles, and are attached to bones or ligaments, and make up intermuscular septa. Many fascia furnish origin or insertion to muscles and thus act as tendons (Sisson and Grossman, 1953).

## Microstructure of Skeletal Muscle

Muscle is separated into different levels of organization by the connective tissue components (Cassens, 1964). The muscle itself is surrounded by a relatively thick connective tissue sheath, termed the epimysium. The epimysial sheath extends into the muscle proper thus dividing it into different orders referred to as muscle bundles or fasciculie. The fasciculi are further subdivided into secondary and tertiary fasciculi. The connective tissue projections defining the fasciculi are referred to as the perimysium. The perimysium further extends into a delicate sheath surrounding the individual muscle fibers and is characterized by small collagenous fibrils. This connective tissue sheath is referred to as the endomysial reticulum (Venable, 1963). Many blood vessels, fat cells, and nerves are found throughout the connective tissue sheaths.

The muscle fiber has been described by Walls (1960) as a cylindrical, elongated, and multinucleated syncytium that generally does not branch, however Bardeen (1903) has shown that variations of

this description do exist. The fibers range from 10 to 100 microns or more in thickness and can be from 1 to 40 mm. in length (Cassens, 1964). It is generally agreed that in short muscles the fibers may run from the origin of the muscle to its insertion. With regard to the length of the fibers in long muscles, the consensus of opinion seems to be that the fibers do not run from end to end of the muscle (Walls, 1960).

The nuclei of the muscle fiber are of an elongated, ovoid shape, and are generally situated in the peripheral cytoplasm immediately beneath the sarcolemma.

Each muscle fiber is enclosed by a thin membrane called the sarcolemma. The sarcolemma is primarily connective tissue in nature and is often thought of as being structureless (Ham, 1957 and Bevelander, 1965). However, with advancement in electron microscopy and new methods of tissue preparation the sarcolemma has been found to consist of several relatively complex structures. The sarcolemma is not a perfectly smooth membrane, but it is marked by several small vesicles and caveolae (Porter and Palade, 1957). Further description by Bennett (1960) and Wang (1956) have noted that the cell boundary can be identified as, (1) the plasmalemma, the dark line limiting the cell, (2) a fuzzy shell of intermediate density closely paralleling the cell membrane, and (3) a less well defined amorphous matrix containing connective tissue fibrils. The proper nomenclature relative to the "sarcolemma" is still unsettled. In regards to the physical properties of this membrane it has been found that it possesses remarkable electrical properties and it appears to be elastic (Walls, 1960). Concerning the mechanical properties of the sarcolemma it has been suggested that the resting tension of the muscle fiber is governed by the

sarcolemma alone (Ramsay and Street, 1940). In this regard Banus and Zetlin (1938) observed that the connective tissue sheath of a whole muscle, when disected free, gave the same tension-length curve as the whole muscle. However, Sichel (1941) has suggested that the intact muscle fiber would resist extension by a tension fully twice that of the sarcolemma. This phenomenon must be considered in the realm of this experiment.

The cytoplasm of the muscle fiber consists of a fluid matrix made of various soluble proteins. This cytoplasm, termed sarcoplasm, contains mitochondria, myofibrils, fat droplets, Golgi apparatus, endoplasmic reticulum, and the various other inclusions and organelles characteristic of most cells. The main functions of the sarcoplasm are of myofibril nutrition and conduction of nerve impulses throughout the fiber (A.M.I.F., 1960).

An organelle of the muscular fiber which has received considerable attention due to is contractile properties is the myofibril. The myofibril is an elongated threadlike structure which measures approximately 1 micron or less in thickness; it is further made up of smaller elements referred to as myofilaments. There are two main kinds of myofilaments; a course filament approximately 100 angstroms in diameter and 1.5 microns in length, and a fine filament approximately 50 angstroms in diameter and 2 microns in length (Huxley, 1957). The thick filament is comprised of the protein myosin and makes up what is referred to as the A-band. The thin filament is comprised of the protein actin and makes up what is referred to as the I-band. Between adjoining I-bands there is another striation referred to as the Z-disc. When examined by polarized light the A-bands are birefringent and the

I-bands are weakly birefringent thus giving the muscle fiber its characteristic striated appearance. The distance from one Z-disc to another is referred to as the contractile unit of the muscle fiber, the sarcomere.

It has been noted (Ham, 1957 and Cassens, 1964) that the thick and thin filaments interdigitate in the vertebrate muscle (the A-band), the fine filaments are seen in a cross section to be arranged so as to form hexagons with a coarse filament in the center of each hexagon. The coarse filaments are arranged so as to form triangles, each of which has a fine filament in its center. It also has been noted that the thick filaments bear a large number of regularly spaced short projections referred to as "bridges", and that these bridges are instrumental in binding the thick filaments to the thin filaments (Huxley, 1963). Further investigations by Hanson and Lowry (1963) indicated that the thin filaments have a beaded appearance when viewed through an electron microscope.. They inferred that these filaments consisted of two helically wound strands composed of subunits which appear to be alike and approximately spherical. There is evidence that each of these globular subunits represent an action monomer (Cassens, 1964).

## Mechanism of Contraction

Studies of Huxley and Hanson (1960) indicated that the mechanism of muscle contraction envolves a sliding movement of the arrays of actin filaments inwards into the arrays of myosin filaments in the A bands, thus forming an actomyosin complex, and that extension of the muscle reverses this movement. As previously mentioned, the sites of

linkage between the actin and myosin molecules are presumably located where the lateral projections on the myosin filaments touch the actin filaments. With the linkage locked (no ATP), the fibril would be inextensible; with the lock open (ATP present and ATP-ase activity suppressed), the arrays of actin filaments could be pulled out of the A-bands and the muscle would be extensible. It will suffice to indicate at this point that muscles derive their energy to contract from the breakdown of (ATP) adenosine triphoshate to (ADP) adenosine diphosphate, and that ADP is resynthesized to ATP from the breakdown of carbohydrates (glycogen). The presence of ATP has also been found to prevent the actin and myosin filaments from combining. Hence, a muscle remains active for as long as ATP is being split, each of the cross-linkages going through one cycle of operation each time it captures a "quantum" of ATP and splits it. If the supply of ATP fails, the links lock and remain locked as in rigor mortis (Huxley and Hanson, 1960).

An interesting observation by Huxley pointed out that the extent of the sliding of the actin filaments along the myosin filaments is much greater than the distance between any two lateral projections on the myosin filaments. Thus it is postulated that these lateral projections alternately disconnect and reconnect the myosin to new sites along the actin filament.

It seems appropriate to indicate that a"muscle relaxing factor" has been discovered (Marsh, 1951-52). In a more recent study, Marsh (1955) observed that homogenized muscle fibers swelled on addition of ATP when the factor was present, but shrank when the factor was absent. Swelling and shrinkage were correlated with lengthening and shortening

respectively, ATP-ase activity was lower in presence of the factor than in its absence. The exact nature of this factor has not yet been determined, however it may in time lead to some interesting results and perhaps fill some gaps in regards to the mechanism of muscle contraction.

## Red and White Muscle Fibers

In a study by Ranvier (1874) it was noted that both "red" and "white" muscle fibers could be observed in rabbit muscle. He indicated that the red fibers have a greater amount of sarcoplasm, more nuclei, and that the nuclei of the white fibers occurred at greater depths than the red. It was also noted that the red fibers had a slower contraction rate than the white fibers. Further investigation by Denny-Brown (1929) established the fact that the color of the muscle lean is dependent upon the proportion of red fibers in the muscle as compared to the proportion of white fibers. In a study by Beecher et al. (1965) using porcine muscle, red muscle fibers contained significantly higher ($P<.05$) concentration of myoglobin, the meat coloring pigment, thus accounting for the color difference. It also has been noted (Allen et al. 1967) that red muscle fibers generally contain more lipid material than the white fibers.

It will suffice to indicate that metabolic differences between red and white fibers do exist. Beecher et al. (1965) noted that the red fibers are characterized by high oxidative enzyme activity, while the white fibers are associated with a high glycolytic rate. This worker has also shown that rigor mortis develops earlier post-mortem in the white fibers.

## Connective Tissue and Fat

Although muscle fibers account for approximately 75 percent of the total muscle tissue, the connective tissue components and associated fat deposits have considerable influence on the quality and physical properties of muscle. These components have previously been indicated as the epimysium, perimysium, endomysium, endoplasmic reticulum, components of the sarcolemma, tendons, ligaments, and sheaths. Usually these areas are characterized by many blood vessels and fat deposits. The amount and character of the connective tissue has been found to vary considerably.

In a study by Wilson et al. (1954) no significant differences in the amounts of collagenous and elastic type connective tissues were noted between grades of beef, however, they did observe a significant difference within grades. It was also noted that no differences between the collagen and elastin content of samples from steers and cows were observed, but the samples from both of these older groups contained less collagen and elastin than veal. In this regard, Coolidge and Lightfood (1948) observed that there was a significant increase in percent collagen with an increase in animal age. Harrison et al. (1949) found that meat from yearling steers contained more elastin than meat from an eight year old cow. Most workers, however, are in general agreement that age does not have a great effect upon the amount of connective tissue in a muscle.

In regard to qualitative changes in connective tissue in the animal, it has been shown (Hiner et al. 1955) that the size of the elastin fibers in young veal and steer calves was smaller than in heifers and cows. As animals became more mature the size of the

elastin fibers increased and the number of collagenous fibers increased. In the case of elastin, it has been suggested (Gersh and Catchpole, 1949) that there may be a coalescing of fibers into larger fibers with age. Bray et al. (1951) found that the ash and moisture content of both elastin and collagen tended to decrease as the animal increased in age.

The amount of fat in an animal is influenced by many factors. It is generally accepted that when the lean tissue growth begins to slow down and all environmental conditions remain the same, the animal will begin to deposit intra- and intermuscular fat. It will suffice for this review to indicate that the amount of fat deposited is dependent upon factors such as level of nutrition, nutritive requirements of the animal body in terms of maintenance and production, and the animal's genetic character.

It is generally accepted that connective tissue has an effect on the physical properties of the muscle. Elastic fibers have been found to extend readily upon stretching, but return to their normal length when relaxed. Collagen fibers, on the other hand, are relatively inert with regard to elasticity.

Herring et al. (1965a) studied the effect of excision and handling on shortening of the semitendinosus (high connective tissue content) and the psoas major muscles (low connective tissue content). It was noted that the semitendinosus muscle shortened by 20-25 percent and the psoas major muscle shortened 10-20 percent, thus perhaps indicating the effect of connective tissue on shortening of these muscles. It was also found that when these two muscles were stretched mechanically the semitendinosus muscle could be stretched extensively without incurring

any damage to the muscle itself, and when relaxed it regained initial length. The psoas major muscle, on the other hand, encountered considerable tearing when stretched and did not return to its initial length when relaxed.

The relationship of kind and amount of connective tissue with tenderness has been studied quite extensively by many workers, however is still obscure. Ramsbottom et al. (1945) indicated that there was an inverse relationship between the quantity of connective tissue and tenderness. Many workers are in agreement with these findings, however Lorinez and Szeredy (1959) indicated that the quality rather than quantity of connective tissue determines tenderness.

The effects of intramuscular fat (marbling) on the physical properties of muscle have been studied. Most workers are in general agreement that the amount of intramuscular fat has little influence on tenderness (Blumer, 1965). However, some differences have been indicated in the case of an extreme abundance of fat, as compared to very little fat. It can be observed that muscles characterized by relatively high amounts of intramuscular fat are usually more firm than those with low amounts of fat. In a study by Pilkington (1960) it was noted that there was a positive correlation between firmness and tenderness, which can be attributed to the fact that less fibers were being sheared per unit area as a result of considerable intramuscular fat.

## Inherent Variation in Skeletal Muscle Fiber Diameter

There is considerable inherent variation in fiber diameter within a muscle, with a range of 10 to 100 microns commonly being accepted (Walls, 1960). Fiber size differs in the major animal classes with fish having the largest fibers and birds the smallest (Mayeda, 1890). The fibers of one muscle may be generally larger than those of another muscle in the same animal (Hammond and Appleton, 1932). In addition, the fiber diameters vary considerably within the same muscle (Swanson et al. 1965). Other factors such as specie, size, breed, age, sex, exercise, and level of nutrition also have been found to influence this variation.

Joubert (1956a) reported that muscle-fiber diameter had no clear relationship to size of species, however, an increase in fiber diameter was closely associated with a relative increase in body weight.

Using cattle, Joubert (1956a) studied the effect of breed on fiber diameter. He found that one breed of cattle, British Friesian, whether crossbred or purebred, had significantly larger fiber diamters than another breed, Dairy Shorthorn purebred and crossbred. This breed difference was not shown to be independent of body weight. Other workers; Adametz (1888), Hammond and Appleton (1932), Strateciuc (1933), Mehner (1938), and Glebina (1952) have supported the findings that interbreed differences do exist, however in most cases the difference is proportional to differences in body size.

A review of early studies caused Joubert (1956a) to conclude that males generally have thicker fibers than females. However, taking body size into consideration he found that there was a slight tendency for

females to have thicker fibers than males. Adametz (1888) found that muscle fibers of bulls were appreciably larger than of cows, but that only slight differences existed between bulls and steers. Other workers; Hammond and Appleton (1932) using sheep, Mehner (1938) with fowl, and Ishihara et al. (1953) in Japanese Black cattle have confirmed these results.

Contradictory evidence has been presented by Brady (1937), and Sartorius and Child (1938) who found that cows had significantly thicker muscle fibers than steers. The true relationship, if any, is still a matter of conjecture.

The effect of age on muscle fiber diameter has been studied extensively; McMeekan (1940-41), Thompson (1942), Meara (1947), Hiner et al. (1953), Joubert (1956a), Tuma et al. (1962), Carpenter et al. (1962), and Henrickson et al. (1963), and all are in agreement that fiber diameter increases from birth to maturity and that fibers rapidly increase in size while the animal is quite young and tend to level off as the animal approaches maturity.

Yeates (1964) studied starvation changes and subsequent recovery of adult beef muscle. The experiment revealed that with starvation of the adult animal the shrinkage in cross sectional areas of the muscles, after allowing for the loss of some intramuscular fat, was associated with the reduction in diameter of the individual fibers; thus, with repair of live weight, recovery both of whole muscle dimensions and muscle fiber diameter appeared to be complete.

An increase in muscle size with exercise is common knowledge, and it appears that this hypertrophy results from an enlargement of the diameter of the individual fibers due to the formation of more

sarcoplasm (Walls, 1960).

## Post-Mortem Structural Variation in Skeletal Muscle Fibers

Another cause of structural variation of the muscle fiber is the conditions to which it is subjected after the animal is slaughtered. Muscular contraction referred to as rigor mortis has received considerable attention by investigators. Rigor mortis is defined as the physical and chemical changes that take place after death of the animal. This discussion will be limited to the physical effects of rigor on the muscle fiber. The main observed physical change is from a highly extensive elastic condition of muscle of freshly killed animals to the inextensible and rigid condition of the muscle fiber in full rigor. This is a result of the actin filaments becoming bound to the myosin filaments thus greatly decreasing fiber extensibility. This actomyosin complex remains locked in a contracted state until rigor resolves (March, 1954). Along with this change in extensibility it has been noted that there is a gradual shortening of the sarcomere as rigor approaches, which leaves the muscle in a semi-contracted state (Locker, 1960). Another well known physical effect of rigor on the muscle fiber is the presence of rigor kinks found in localized areas along the lengths of some fibers.

The effect of temperature on the condition of the muscle fiber has been shown to have considerable influence. Locker and Hagyard (1963) have shown that shortening of the muscle fiber occurs when exposed to very cold temperatures. This phenomenon is referred to as "cold shortening." It is currently thought that cold shortening occurs simultaneously with the formation of cross-linkages in rigor; a degree

of internal strain or actual disorganization occurs, which actually increases the resistance of the muscle to cleavage. Herring et al. (1965a) reported that slightly more shortening, as indicated by sarcomere length, appeared to take place in the stretch-restrained muscle samples at one degree centrigrade than at five degrees centigrade. It was postulated in this experiment that some cold shortening may have occurred at this temperature as well. Cook and Wright (1966) using samples of unfrozen and prerigor frozen ovine semitendinosus muscle, incubated for twenty-four hours at six temperature levels between zero and 40 degrees centigrade, found that variations in temperature caused muscle fibers to be in various states of contraction. The variations in sarcomere length of unfrozen and pre-rigor frozen muscle did not follow any specific course in relation to temperature, but a difference was observed.

Harrison et al. (1949) noted that the difference between sections of raw and cooked muscle were slight, however, the cooked sections tended to have straighter fibers than the raw muscle sections.

Paul et al. (1944) reported that the structural appearance of the muscle fibers varied with aging. Harrison et al. (1949) found that freshly killed beef muscle showed poorly differentiated fibers which were straight to slightly wavy. After one day of storage at 1.7 degrees centigrade the fibers and cross striations were more distinct, and the longitudinal striations less distinct. Contracture nodes, kinks, and waves increased after four to nine days of storage. Disappearance of cross striae in small, infrequent areas of the fibers was noted on the second day of storage, and this disintegration tended to increase in frequency and extent as the storage time increased.

Younger and Baigent (1965) studying the effect of precooking on freeze-dried lamb have noted that uncooked freeze-dried meat appeared to suffer much more fiber damage, in terms of fiber distortion and shrinkage, than the cooked freeze-dried samples. When rehydrated, the samples revealed a similar pattern, the uncooked freeze-dried material revealed considerable distortion of the fibers and many of the fibers were smaller in diameter than normal. The cooked freeze-dried samples very closely resembled fresh meat. The fibers were restored almost completely in size and shape, and there were no abnormal spaces between the fibers.

Carcass position has been found to have a definite effect on sarcomere length and fiber diameter of the muscle fiber (Herring et al. 1965b). It has been found that by vertically suspending the carcass certain muscles are in a stretched state, as indicated by sarcomere length, while other muscles are in a shortened state. In general, the differences in sarcomere length of the muscle-fibers were associated with differences in diameter. When the muscles shortened, there was a corresponding decrease in sarcomere length and an increase in fiber diameter.

Recent work by Reddy and Henrickson (1967) studying the effect of pre-rigor excision of three bovine muscles on fiber diameter and amount of fiber distortion have indicated interesting relationships. The amount of fiber distortion, termed percent rigor, was found to be greater ($P<.10$) for the pre-rigor excised longissimus dorsi muscle than the post-rigor excised muscle. However, the opposite was found true for the gluteus medius muscle, thus supporting the postulation of Locker (1960) and the results of Herring et al. (1965b) that different

internal strains among muscles exist in the vertically suspended carcass. These workers also found that the diameters of the fibers of the pre-rigor excised semitendinosus muscle were greater than the fiber diameters of the post-rigor excised muscle, which was apparently due to the shortening of the muscle.

## Structural Variations of the Skeletal Muscle Fiber in Relation to Tenderness

In a study to determine the relationship of fiber diameter to tenderness, Hiner et al. (1953) indicated a definite curvilinear relationship. He found that up to a point, an increase in fiber diameter will result in an increase in shear force. Tuma et al. (1962) also reported that with an increase in fiber diameter there was a corresponding increase in shear force of meat from cattle of different age groups. However, when the effect of age was removed little relationship was noted. Carpenter et al. (1962) on the other hand found that with an increase in maximum fiber diameter there was a decrease in shear force of raw longissimus dorsi muscle. Opposite results were found for cooked longissimus dorsi muscles. These workers postulated that for a given size core there may be more small fibers per unit area than large fibers, therefore more of the sarcolemma and associated connective tissue was present, resulting in a less tender product.

Herring et al. (1965b) noted that as fiber diameter increased tenderness decreased whereas the opposite was true when fiber diameter decreased. It was also shown that a change in fiber diameter was related to a change in sarcomere length. In a previous experiment Herring et al. (1965a) found that by stretching a muscle there was

increased tenderness over a muscle that had not been stretched. The stretched muscle had smaller fiber diameters than the muscle that was not stretched and it was assumed that a greater number of fibers were being severed in the stretched sample per unit area. This assumption is in agreement with that of Carpenter et al. (1962) in that the greater number of smaller fibers should theoretically make the muscles less tender. However, the stretched muscle was more tender thus indicating that the thickness of the sarcolemma and associated connective tissue was reduced in thickness when the fibers were stretched (Casella, 1950).

Reddy and Henrickson (1967) have noted that the post-rigor excised semitendinosus muscles were more tender than the pre-rigor excised muscles. These post-rigor excised muscles also had smaller fiber diameters than the pre-rigor excised muscles, thus indicating perhaps the difference in fiber diameters accounted for the difference in tenderness.

The degree of muscular contraction as it effects tenderness has received considerable interest in recent years. In an experiment where Locker (1960) studied the effects of pre-rigor excision of several muscles he concluded that muscles in a relaxed state, as indicated by fibrillar pattern, are more tender than those partly contracted. In this regard March and Leet (1966) studied the effects of cold shortening on tenderness. They noted that with a decrease in length of up to approximately 20 percent caused little or no toughening, but with 20 to 40 percent shortening the toughness increased several fold. Beyond 40 percent shortening the meat became increasingly more tender, and at 60 percent it was cleaved almost as easily as meat in which

almost no shortening had occurred. These workers postulated that the 40 to 60 percent range may be a zone of progressive rupturing thus causing a rapid decrease in internal strain in this phase, with consequent realignment of previously distorted cleavage planes.

Structural variations of the muscle fiber, thus far mentioned, have been shown to effect the ultimate tenderness of muscle. It stands to reason that perhaps some of the commonly associated differences in tenderness with age, sex, and etc. may be due, in part, to the structural condition of the muscle fiber.

## MATERIALS AND METHODS

### Materials

Five steers (3 Hereford, 1 Angus, 1 Shorthorn) ranging in weight from 433.02 kg. to 501.56 kg. of comparable finish and grade were obtained from the Oklahoma Agricultural Experiment Station herd. The steers were from similar managerial backgrounds and ranged in ages from approximately 11 to 14 months. The animals were delivered to the Meat Laboratory where feed and water were withheld for approximately 15 hours prior to slaughter. The animals were slaughtered in accordance with the practices normally employed in the laboratory and by industry (Wellington, 1953). Following bleeding, dehiding, and eviscerating, the carcasses were split and washed thoroughly. Each side was then quartered and the hind quarters laid out on a cutting table. The semitendinosus and semimembranosus muscles were excised from both hindquarters approximately 45 minutes post-mortem, trimmed of excess external fat, and divided into two samples each of approximately (6 x 6 x 21 cm.), thus four samples per muscle. These samples were then subjected to four degrees of tension (0, 1000, 2500, 5000 gm.) by the use of a tensiometer (Figure 1) and held in this state at about one degree centigrade for 48 hours. Each sample was covered with a fibrous bologna casing to prevent excessive moisture loss. The samples were then removed from the tensiometer; histological core samples taken, and the remainder wrapped in aluminum foil, frozen in the blast freezer at

-20 degrees centigrade, and stored at -40 degrees centigrade until future shear determinations could be made.

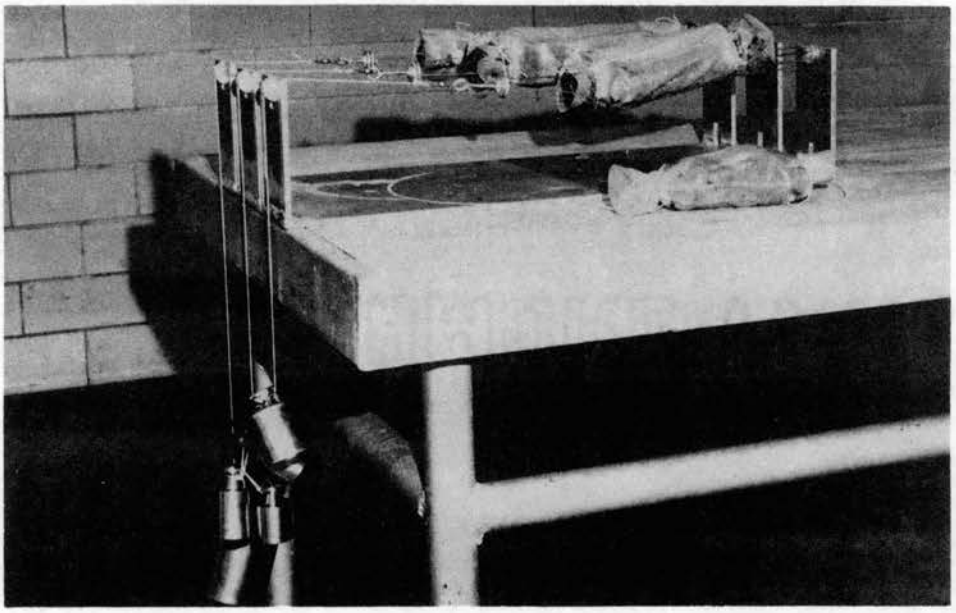


Figure 1. Tensiometer Apparatus

## Methods

The samples used for structural characterization consisted of removing two 1/4 x 1/2 inch cores from each end of the muscle section (Figure 2). These were put into a 10% buffered formalin fixative for approximately one week at about one degree centigrade. The microscopic study consisted of measuring fiber diameter, sarcomere length, and percentage rigor. For each of these measurements a small piece of tissue was randomly removed from each of the four cores (Figure 3).

Shear force determinations were made on one inch diamter cores removed from the center of each cooked sample. Six shear force readings were recorded for each core (Figure 4).

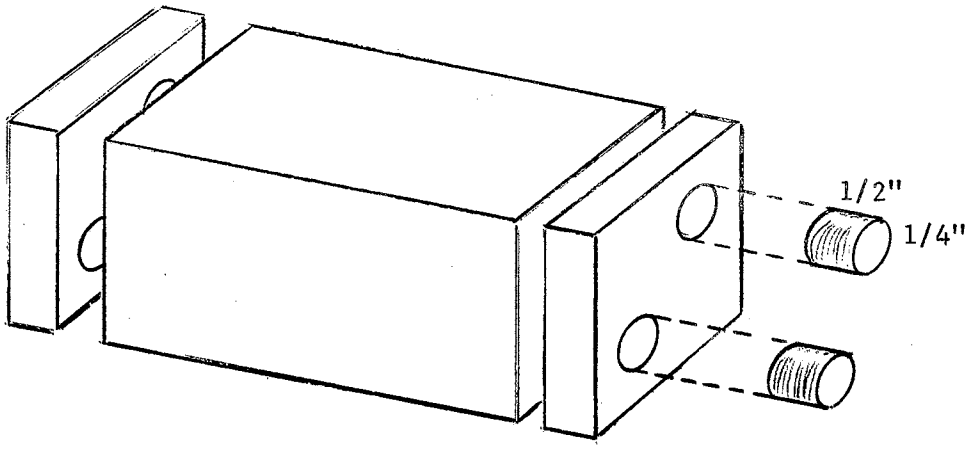



Figure 2. Histological Core Locations

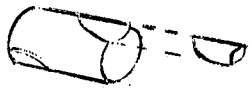


Figure 3. Removal of Tissue Sample for Measuring Histological Characteristics.

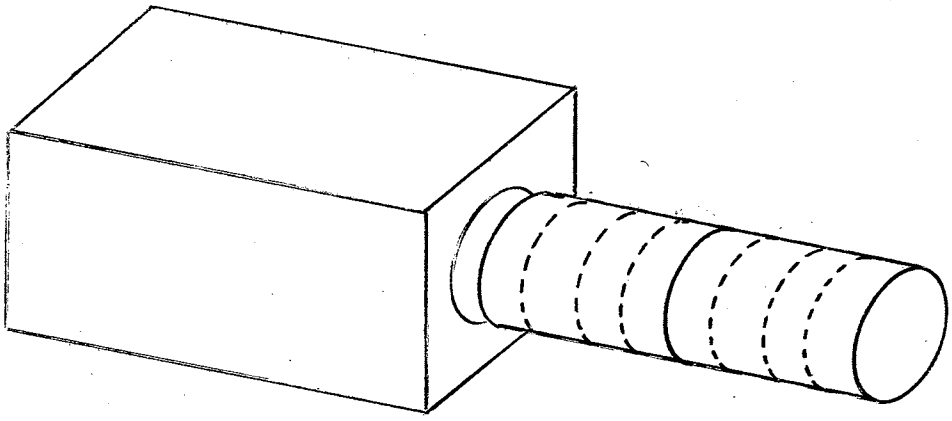


Figure 4. Shear Core Location and Shearing Sites.

## Fiber Diameter

A sample of tissue was removed from each fixed core and blended in a slow speed Waring blender, blades reversed, for one minute in a 10% formalin media to dislodge the fibers. The suspension of fibers, in the formalin media, was transferred into four 2-inch diameter petri dishes. Any remaining solution and fibers were discarded. The fibers were allowed to settle to the bottom of the petri dishes before being observed under the microscope. The petri dish was then placed on the mechanical stage of a Bausch and Lomb binocular light microscope. The microscope was equipped with an ocular micrometer in the 15X eyepiece, which was calibrated as 10 microns per division when the 10X objective was used. Artificial lighting provided the necessary illumination.

Twenty-five fibers were measured per petri dish which gave a total of 100 fibers measured per sample, thus 400 measurements per muscle. Only the fibers that were observed in the field of a predetermined course (Figure 5) were measured. All fibers, broken and distorted, were measured at their widest diameter. If 25 fibers were not observed in this field, a new field was fixed parallel, on either side, of the previous course.

## Percentage Rigor

Percentage rigor, or the amount of kinkyness characteristic of the fiber in rigor, was determined at the same time as the fiber diameters were measured. For each fiber diameter measured a value was subjectively assigned to the condition of that fiber, this in turn was expressed on a percentage basis as shown in the following example:

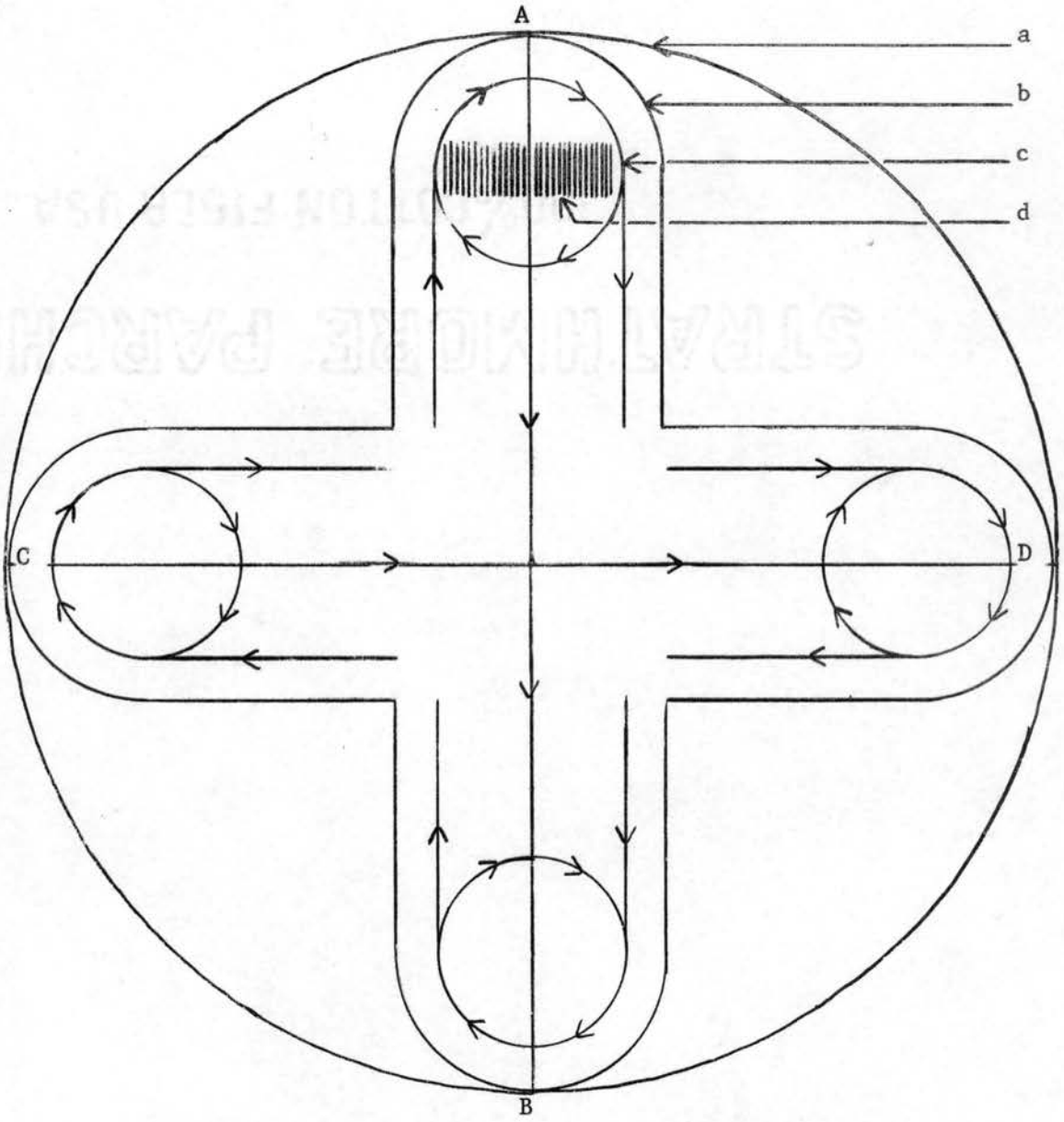



Figure 5. Illustration of Method for Microscopic Field Course. (Compliments, S. G. Reddy).

a. Receptacle
b. Field covered by objective lense
c. Field covered by the radius of the ocular micrometer
d. Ocular micrometer

Example - 100 fibers

| Condition of fiber | Number | Assigned Value | Product |
|---|---|---|---|
| Straight | 50 | 0 | 0 |
| Straight Plus | 20 | 1 | 20 |
| Wavy | 5 | 2 | 10 |
| Wavy Plus | 10 | 3 | 30 |
| Twisted | 5 | 4 | 20 |
| Twisted Plus | 6 | 5 | 30 |
| Kinky | 4 | 6 | 24 |
| Total | 100 | | 134 |

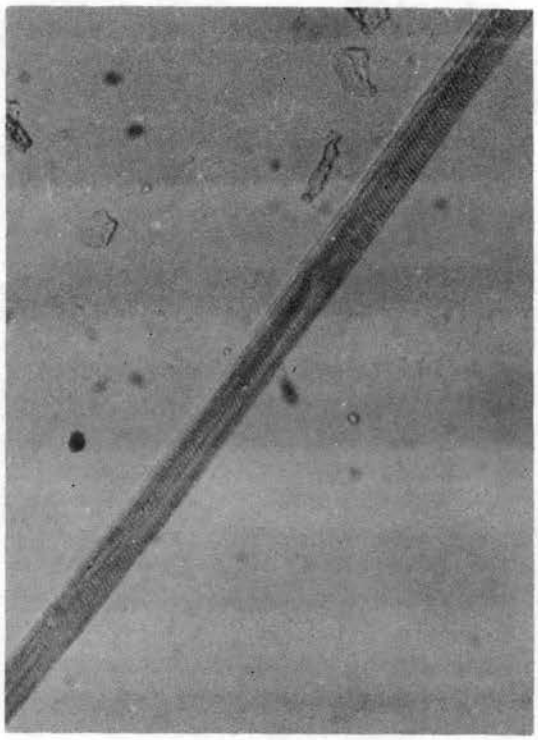
134/600 X 100 = 22.33% rigor

The number of fibers falling into the seven classifications were recorded and multiplied by an assigned value. The products were then added and divided by the total number possible (600), which permitted the degree of fiber kinking and distortion to be put on a percentage basis.

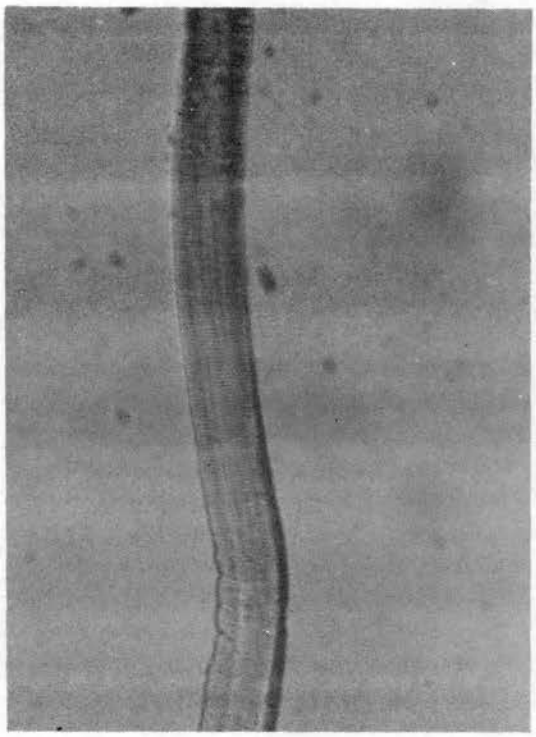
The examples of the various conditions in which the fibers were classified are shown in (Figure 6).

## Sarcomere Length

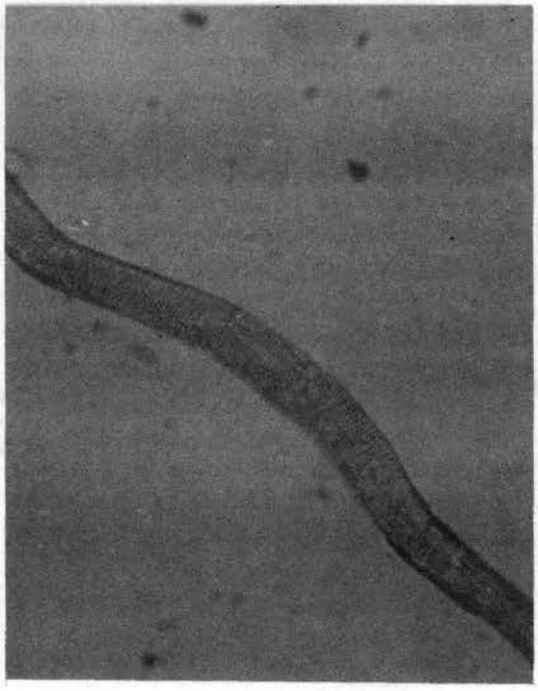
A small sample of the tissue was again removed from the formalin fixed cores and blended for one hour at about one degree centigrade in 10% formalin. A drop of the suspended myofibrils was then placed on a clean glass slide and held in place with a glass coverslip. No stain or mounting media was used. The slide was viewed under a Zeiss phase-contrast microscrope equipped with an ocular micrometer, calibrated as one micron per division, at a power of 1600X. The field course consisted of making three passes across the slide being careful not to allow fields to overlap. Sarcomere lengths were determined by measuring the length of 10 sarcomeres, collectively, on each of 25 myofibrils and



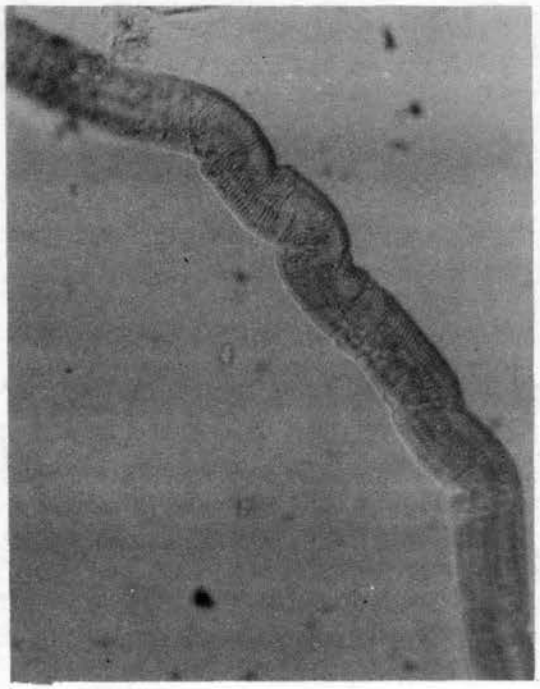
1. Straight Fiber



2. Straight Plus Fiber

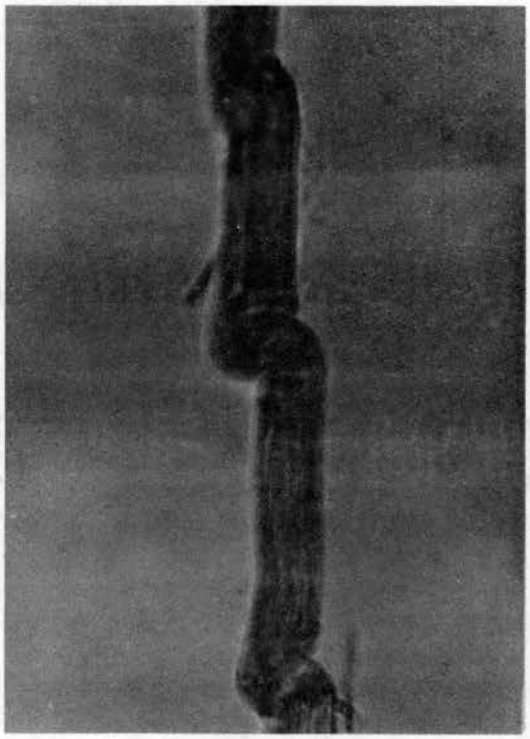


3. Wavy Fiber

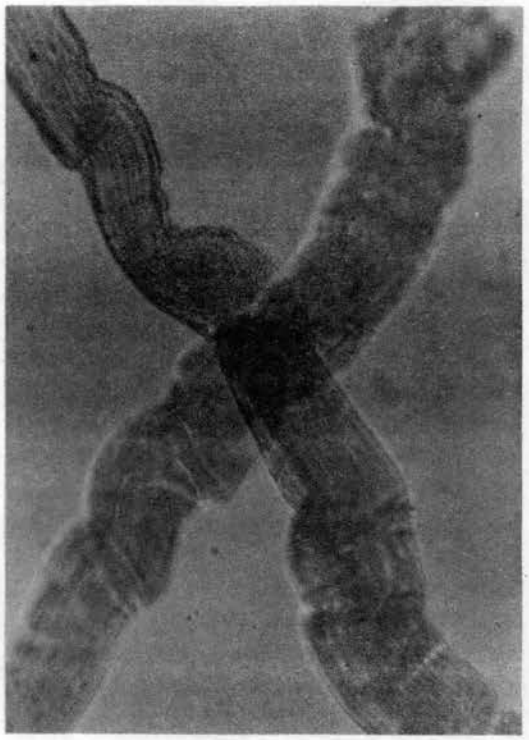


4. Wavy Plus Fiber

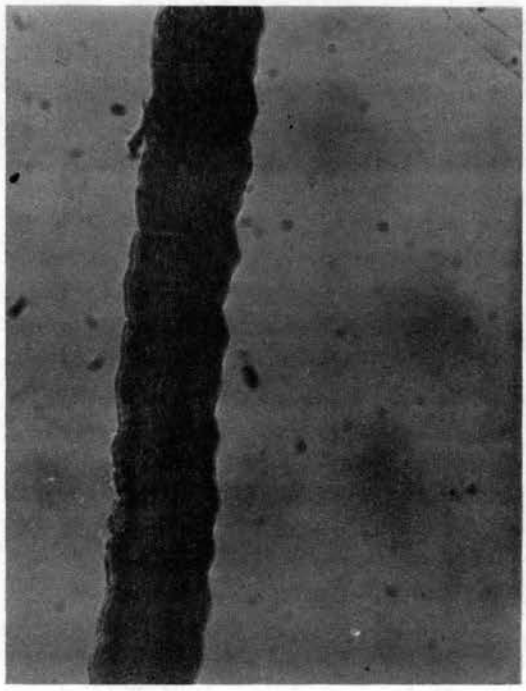
Figure 6. Classification of Muscle Fibers According to Their Condition in Rigor Mortis.

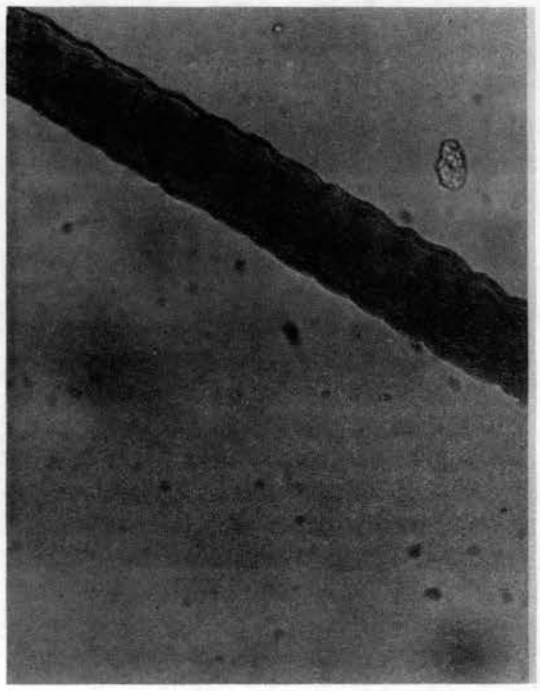


5. Twisted Fiber



6. Twisted Plus Fiber





7. Kinky Fibers

Figure 6. (Concluded).

taking an average of these. Twenty-five myofibrils were measured per sample, or 100 average measurements per muscle (Figure 7).

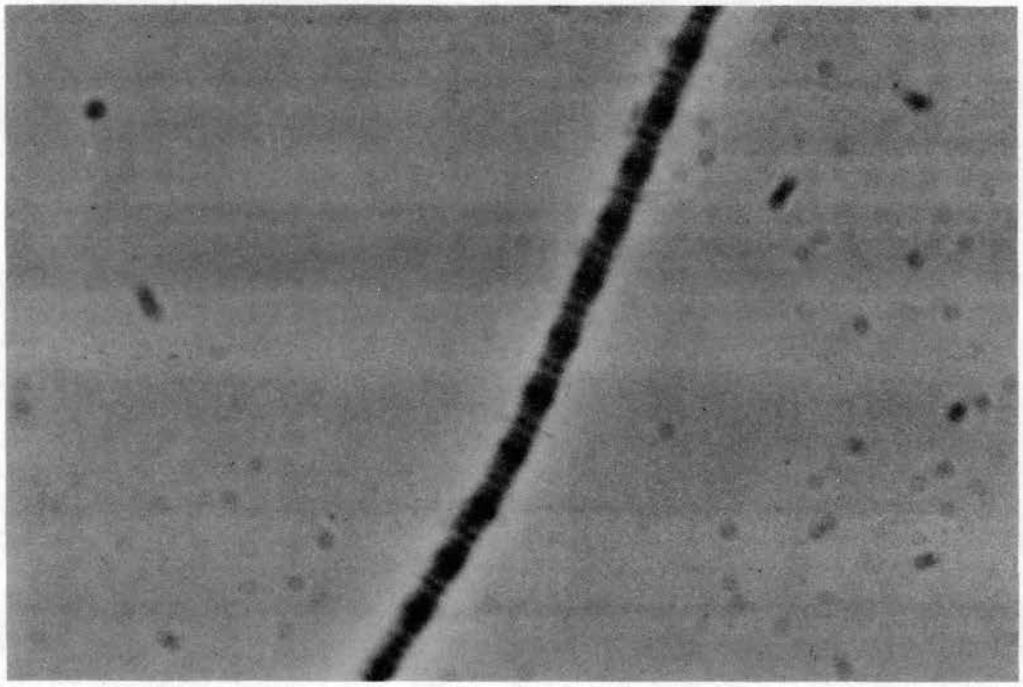


Figure 7. Longitudinal Photomicrograph of Myofibril

## Shear Force

The frozen muscle samples were allowed to thaw at 3.89 degrees centigrade for approximately 48 hours. They were then tagged and cooked in a deep-fat fryer with the cooking oil preheated to 135 degrees centigrade. The samples were allowed to attain an internal temperature of 68.33 degrees centigrade, as indicated by a conventional meat thermometer placed in the center of the muscle sample. They were then placed in the 3.89 degree cooler, covered, and allowed to equilibriate to an internal temperature of 3.89 degrees centigrade for approximately

24 hours. Six shear force determinations were recorded on the Warner-Bratzler shear machine for the one inch core. The core was removed from the center of the muscle sample by the use of a mechanical coring device mounted on a small drill press. A total of 24 shear determinations were made per muscle.

## Statistical Analysis

The effect of tension on the muscles with regard to tenderness, fiber diameter, sarcomere length, and percentage rigor was determined by using an Analysis of Variance, F-test, and Duncan's New Mulitple Range test (Steel and Torrie, 1960). The error term in the analysis used for testing was not a true estimate of experimental error *per se*, but a pooled estimated of block (animal) interactions.

## Experimental Design

Animal No. 1-5 (Blocks)

| Tension | ST Muscle | SM Muscle |
|---|---|---|
| 0 gms. | 6 shears<br>100 fiber diameters<br>250 sarcomere lengths<br>100 est. of % rigor | 6 shears<br>100 fiber diameters<br>250 sarcomere lengths<br>100 est. of % rigor |
| 1000 gms. | 6 shears<br>100 fiber diameters<br>250 sarcomere lengths<br>100 est. of % rigor | 6 shears<br>100 fiber diameters<br>250 sarcomere lengths<br>100 est. of % rigor |
| 2500 gms. | 6 shears<br>100 fiber diameters<br>250 sarcomere lengths<br>100 est. of % rigor | 6 shears<br>100 fiber diameters<br>250 sarcomere lengths<br>100 est. of % rigor |
| 5000 gms. | 6 shears<br>100 fiber diameters<br>250 sarcomere lengths<br>100 est. % rigor | 6 shears<br>100 fiber diameters<br>250 sarcomere lengths<br>100 est. % rigor |

## RESULTS AND DISCUSSION

### Fiber Diameter

Variation in fiber diameter was found to be highly significant ($P<.01$) for the four tension treatments (Table I). However, highly significant differences ($P<.01$) were also noted for muscles and blocks (animals). This was to be expected and is in agreement with the findings of most research workers. No significant interaction ($P<.05$) of tension and muscles was found, although an interaction of blocks with muscles and tension may have occurred.

Further analysis, using Duncan's New Multiple Range test, indicated that there was a highly significant decrease ($P<.01$) in fiber diameter with increasing tension to the 1000 gram pull treatment, then a gradual leveling off with succeeding increases in tension (Figure 8). This would seem logical in that as the individual muscle fibers were stretched there was a corresponding decrease in fiber diameter. The muscle fiber evidently approached a point of physical limitation with respect to stretching at the 1000 gram pull treatment, with only a slight decrease in diameter to the 2500 gram pull.

The semimembranosus muscle had consistently smaller fiber diameters at each degree of tension than the semitendinosus muscle thus indicating true inherent differences between the two muscles.

TABLE I

EFFECTS OF FOUR LEVELS OF TENSION ON FIBER DIAMETER IN TWO BOVINE MUSCLES

| Tension[a] | Semitendinosus[b] 0 | 1000 | 2500 | 5000 | Semimembranosus[c] 0 | 1000 | 2500 | 5000 | SE[d] |
|---|---|---|---|---|---|---|---|---|---|
| Animal 1[e] | 70.35[f] | 57.25 | 54.65 | 54.70 | 65.30 | 58.05 | 54.10 | 55.05 | 2.86 |
| Animal 2[e] | 68.20 | 53.02 | 56.75 | 57.50 | 57.05 | 54.60 | 56.47 | 50.00 | 2.86 |
| Animal 3[e] | 65.80 | 58.35 | 56.90 | 58.05 | 60.90 | 50.75 | 49.05 | 53.00 | 2.86 |
| Animal 4[e] | 67.02 | 66.83 | 64.13 | 59.01 | 67.93 | 54.43 | 56.27 | 53.80 | 2.86 |
| Animal 5[e] | 63.86 | 58.95 | 54.63 | 58.55 | 63.05 | 50.64 | 49.01 | 50.30 | 2.86 |
| Average[g] | 67.05 | 58.88 | 54.41 | 57.56 | 62.85 | 54.29 | 52.98 | 52.43 | 1.28 |

[a]Grams pull.

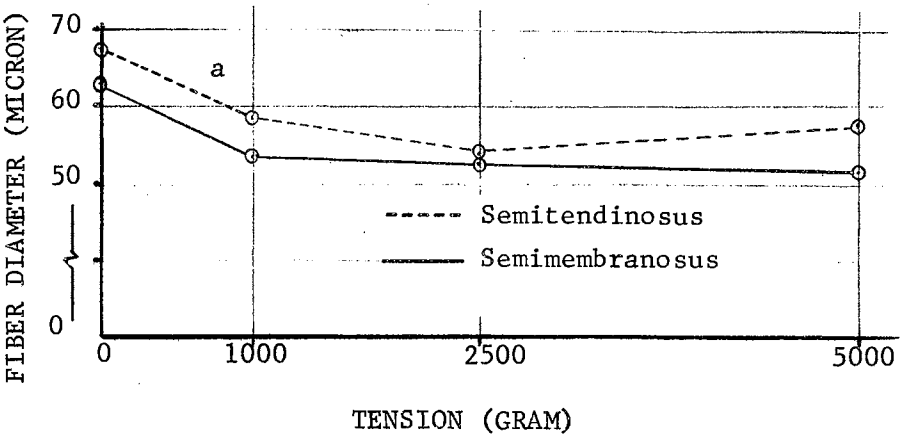
[bc]Highly significant differences (P<.01) between muscles.

[d]Standard error.

[e]Significant difference (P<.05) among animals.

[f]Diameter in microns.

[g](ST) 67.05> 58.88 (P<.01)
(SM) 62.85> 54.29 (P<.01).




a - Ranges differ ($P<.01$) for both muscles

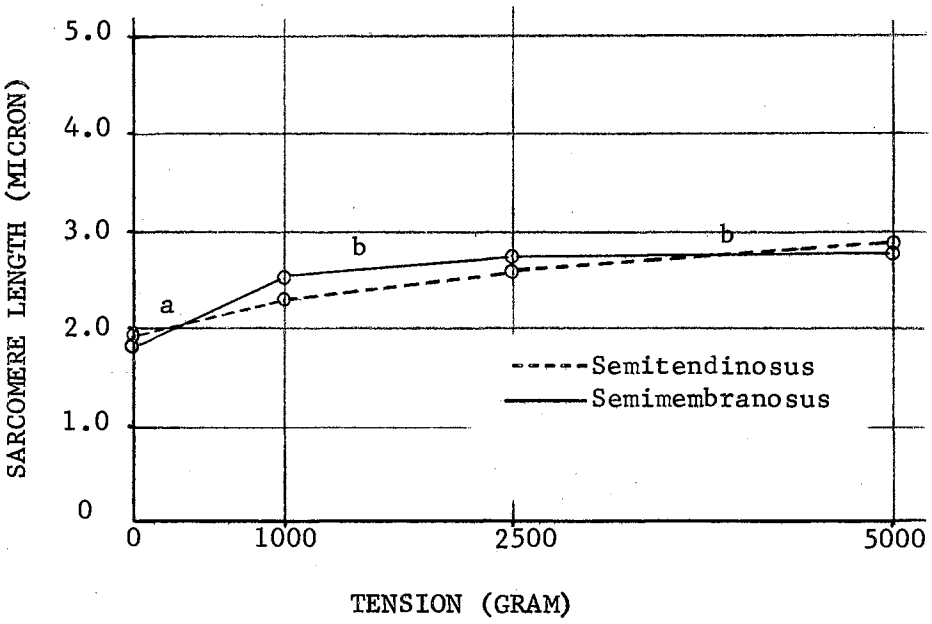
Figure 8. The Effect of Tension on Fiber Diameter.

## Sarcomere Length

Highly significant differences ($P<.01$) in sarcomere lengths from the two muscles were noted for the tension treatments (Table II). Significant differences ($P<.05$) were also found amongst blocks (animals). It can be said, with some degree of confidence, that the difference which was found between blocks was probably a result of sampling and that the muscles were in different physiological states. Little work has been conducted regarding inherent variations in sarcomere length; however the size of the actin and myosin filaments within the sarcomere has been found to be relatively specific, thus indicating that the maximum length of the sarcomere probably is limited.

Further analysis clearly indicated that with succeeding increases in tension above 1000 grams, a corresponding increase in sarcomere length occurred (Figure 9). Duncan's test indicated that all ranges

were highly significant ($P<.01$) except the ranges 1000-2500 and 2500-5000 which were significant at ($P<.05$). These findings are in general agreement with the work done by Herring et al. (1965a) who noted that sarcomere length of the semitendinosus muscle shortened as a result of pre-rigor excision, but that the pre-rigor excised, stretch-restrained semitendinosus muscles generally exhibited longer sarcomeres than the control samples.




a - Ranges differ ($P<.01$) for both muscles.

b - Ranges differ ($P<.05$) for both muscles.

Figure 9. The Effect of Tension on Sarcomere Length.

TABLE II

EFFECTS OF FOUR LEVELS OF TENSION ON SARCOMERE LENGTH IN THE FIBERS OF TWO BOVINE MUSCLES

| Tension[a] | Semitendinosus 0 | 1000 | 2500 | 5000 | Semimembranosus 0 | 1000 | 2500 | 5000 | SE[b] |
|---|---|---|---|---|---|---|---|---|---|
| Animal 1[c] | 1.80[d] | 2.22 | 2.58 | 2.84 | 1.96 | 1.92 | 2.43 | 2.29 | .17 |
| Animal 2[c] | 1.84 | 2.32 | 2.62 | 2.77 | 1.98 | 2.61 | 2.62 | 2.95 | .17 |
| Animal 3[c] | 1.97 | 2.40 | 2.64 | 2.96 | 1.75 | 2.72 | 2.72 | 2.62 | .17 |
| Animal 4[c] | 2.12 | 2.31 | 2.71 | 2.90 | 2.01 | 2.41 | 2.91 | 3.00 | .17 |
| Animal 5[c] | 1.98 | 2.43 | 2.43 | 2.92 | 1.83 | 2.57 | 2.97 | 3.26 | .17 |
| Average[e] | 1.94 | 2.34 | 2.60 | 2.88 | 1.91 | 2.45 | 2.73 | 2.82 | .08 |

[a]Grams pull.

[b]Standard error.

[c]Significant difference ($P<.05$) amongst animals.

[d]Length in microns.

[e](ST) $2.88 > 2.34 > 1.94$ ($P<.01$)
$2.88 > 2.60 > 2.34$ ($P<.05$)

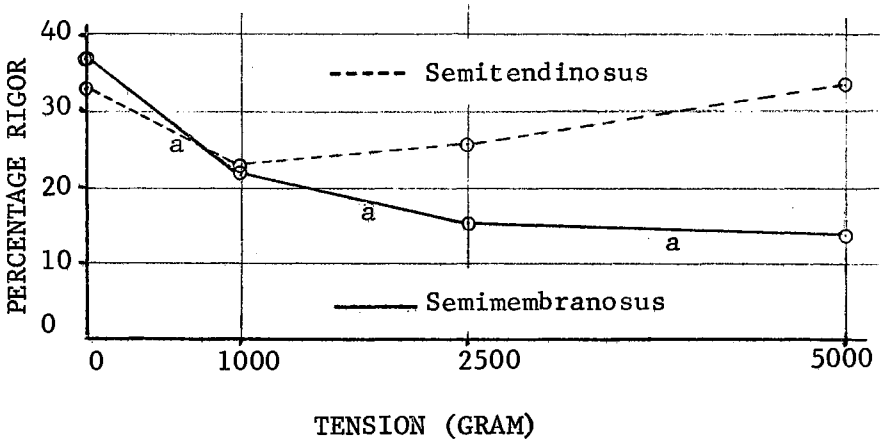
(SM) $2.82 > 2.45 > 1.91$ ($P<.01$)
$2.73 > 2.45$ ($P<.05$).

## Percentage Rigor

Variation in percentage rigor was found to be highly significant ($P<.01$) for the different degrees of tension (Table III). Significant differences ($P<.05$) were noted between muscles and for a muscle by tension interaction. The differences between muscles and the significant interaction can probably be accounted for by differences in the physical properties of the two muscles. Herring et al. (1965a) reported that the semitendinosus muscle was able to regain initial length after being stretched, whereas the psoas major muscle was not. His evidence along with the findings in this investigation indicate differences in the physical properties of the two muscles. In this experiment, there was some difference in connective tissue content of the muscles (Lee and Henrickson, 1967) which feasibly could have had an effect on the amount of fiber kinking and distortion. The exact cause of rigor kinks and nodes is not known, however it has been postulated that connective tissue does have considerable effect on the fibers ability to contract and become distorted. In this regard the differences in permeability of the sarcotubules throughout the length of the fiber may be one reason why rigor kinks and nodes only occur in localized areas. It is quite feasible that with the formation of lactic acid in post-mortem metabolism and subsequent lowering of muscle pH there is a change in permeability of the sarcotubule membranes. This in turn would allow some myofibillar calcium ions to be exuded in the cellular fluids, thus inactivating the "muscle relaxing factor" and activating ATP-ase activity and subsequent contraction of the sarcomere. This postulation is similar with that of Bendall (1960) on thaw rigor. It seems logical that if some areas of the fiber undergo more contraction than others,

a kinking and noding effect may occur. In any event this area does warrant more investigation.

As can be seen in Figure 10 both muscles decreased in percentage rigor to the 1000 gram pull treatment. The semimembranosus muscle tended to decrease and the semitendinosus muscle tended to increase in percentage rigor with increasing amounts of tension. This is in agreement with the findings of Reddy and Henrickson (1967) who noted that the pre-rigor excised longissimus dorsi muscle had a higher percentage rigor than the post-rigor excised muscle, thus indicating that in the post-rigor excised muscle the strains induced by vertical suspension of the carcass did inhibit kinking of the fibers to a degree.




a - Ranges differ ($P<.01$) for semimembranosus muscle.

Figure 10. The Effect of Tension on Percentage Rigor.

TABLE III

EFFECTS OF FOUR LEVELS OF TENSION ON PERCENTAGE RIGOR IN TWO BOVINE MUSCLES

| Tension[a] | Semitendinosus[b] 0 | 1000 | 2500 | 5000 | Semimembranosus[b] 0 | 1000 | 2500 | 5000 | SE[d] |
|---|---|---|---|---|---|---|---|---|---|
| Animal 1 | 26.83[e] | 27.50 | 23.50 | 30.16 | 47.67 | 35.83 | 28.00 | 14.67 | 8.30 |
| Animal 2 | 27.83 | 15.50 | 17.17 | 26.00 | 40.00 | 13.67 | 14.00 | 10.50 | 8.30 |
| Animal 3 | 43.00 | 15.33 | 39.33 | 46.00 | 38.00 | 17.00 | 9.83 | 7.33 | 8.30 |
| Animal 4 | 34.83 | 17.50 | 26.67 | 37.67 | 27.33 | 25.17 | 9.17 | 5.00 | 8.30 |
| Animal 5 | 31.83 | 35.67 | 22.83 | 27.17 | 29.83 | 17.33 | 18.00 | 31.67 | 8.30 |
| Average[f] | 32.86 | 22.30 | 25.90 | 33.40 | 36.57 | 21.80 | 15.80 | 13.83 | 3.71 |

[a]Grams pull.

[bc]Significant difference ($P<.05$) between muscles.

[d]Standard error.

[e]Percent rigor.

[f](SM) 36.57 > 21.80 ($P<.01$)

## Shear Force

Shear force was found to differ significantly ($P<.05$) for the tension treatments (Table IV). Significant differences ($P<.05$) were also noted between animals. Shear force is a highly variable measure and it is difficult to pick up small differences without running many shear determinations. A more precise measurement would have been desirable for the study of this relationship.

Further analysis indicated that there was a significant decrease ($P<.05$) in shear force for an average of the two muscles to the 1000 gram pull treatment, then it leveled off (Figure 11). This type of response was also characteristic of several of the previously mentioned variables, thus indicating that a relationship of these variables with shear force may exist. In this regard, scattergrams were plotted. The scattergrams were derived by plotting the specific values (not random) for each tension, muscle, and animal combination. It should be noted that the relationships indicated are not true correlations, as an unknown amount of variation within the variables being measured can probably be attributed to the tension treatments. The scattergram (Figure 12) indicates that a trend may exist. With an increase in fiber diameter a slight decrease in shear force was noted. The relationship of sarcomere length to shear is shown in (Figure 13). As the sarcomere length increased there was a considerable decrease in shear force. The observations are in agreement with those of Herring et al. (1965a,b). It was also noted that with an increase in percentage rigor there was a gradual increase in shear force (Figure 14).

TABLE IV

EFFECTS OF FOUR LEVELS OF TENSION ON SHEAR FORCE OF TWO BOVINE MUSCLES

| Tension[a] | Semitendinosus 0 | 1000 | 2500 | 5000 | Semimembranosus 0 | 1000 | 2500 | 5000 | SE[b] |
|---|---|---|---|---|---|---|---|---|---|
| Animal 1[c] | 44.10 [d] | 28.50 | 25.20 | 26.40 | 26.90 | 37.90 | 24.90 | 30.10 | 4.61 |
| Animal 2[c] | 40.40 | 27.18 | 21.75 | 25.07 | 26.68 | 21.21 | 23.44 | 21.48 | 4.61 |
| Animal 3[c] | 27.51 | 24.05 | 31.32 | 26.05 | 30.43 | 24.75 | 22.35 | 24.18 | 4.61 |
| Animal 4[c] | 18.77 | 19.75 | 27.79 | 25.76 | 25.70 | 22.52 | 24.95 | 22.33 | 4.61 |
| Animal 5[c] | 36.96 | 31.13 | 25.83 | 30.67 | 36.54 | 27.00 | 26.25 | 27.50 | 4.61 |
| Average [e] | 33.55 | 26.12 | 26.39 | 26.79 | 29.25 | 26.68 | 24.38 | 25.12 | 2.06 |

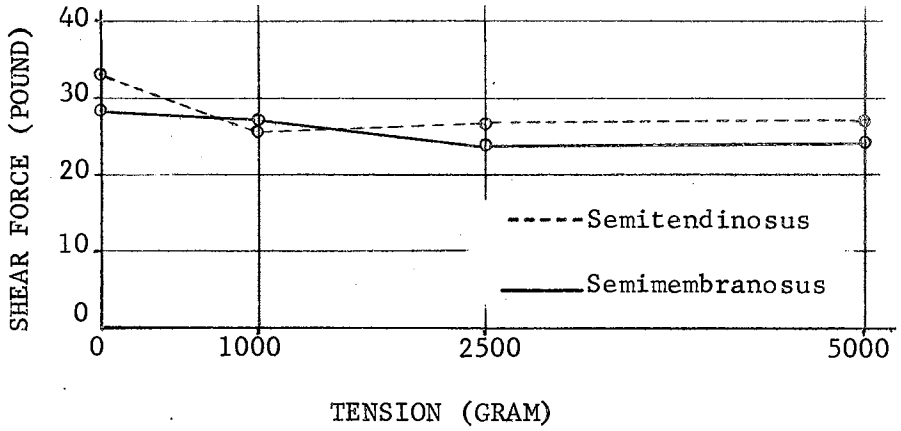
[a] Grams pull.

[b] Standard error.

[c] Significant difference ($P<.05$) amongst animals.

[d] Pounds shear.

[e] (ST) $33.55 > 26.79$ ($P<.05$)




a - Range differs ($P<.05$) for 0-2500 for average of both muscles.

Figure 11. The Effect of Tension on Shear Force.

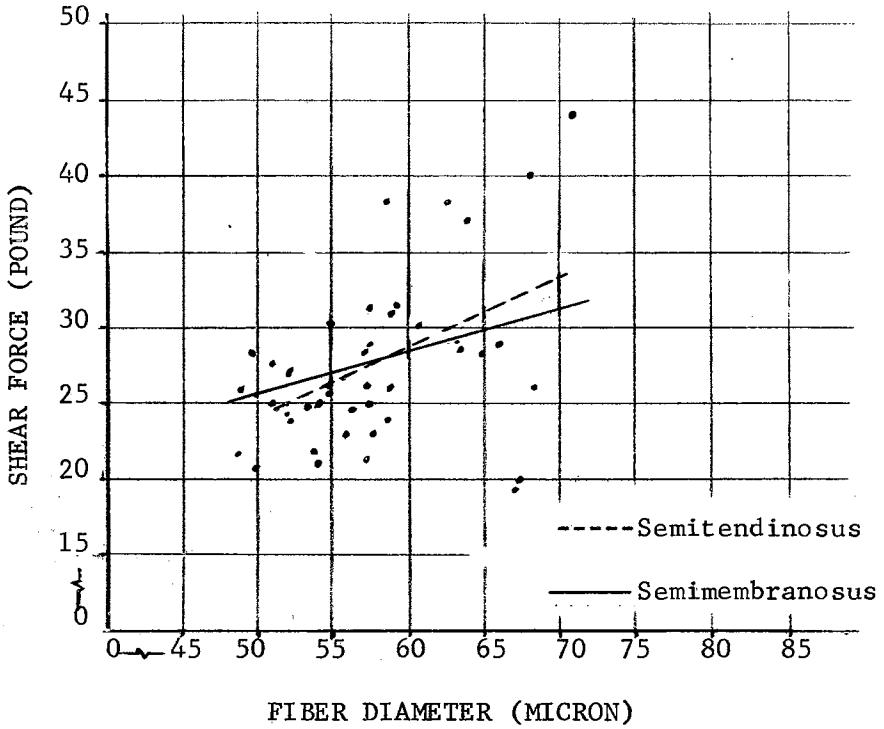



Figure 12. Relationship of Shear Force and Fiber Diameter.

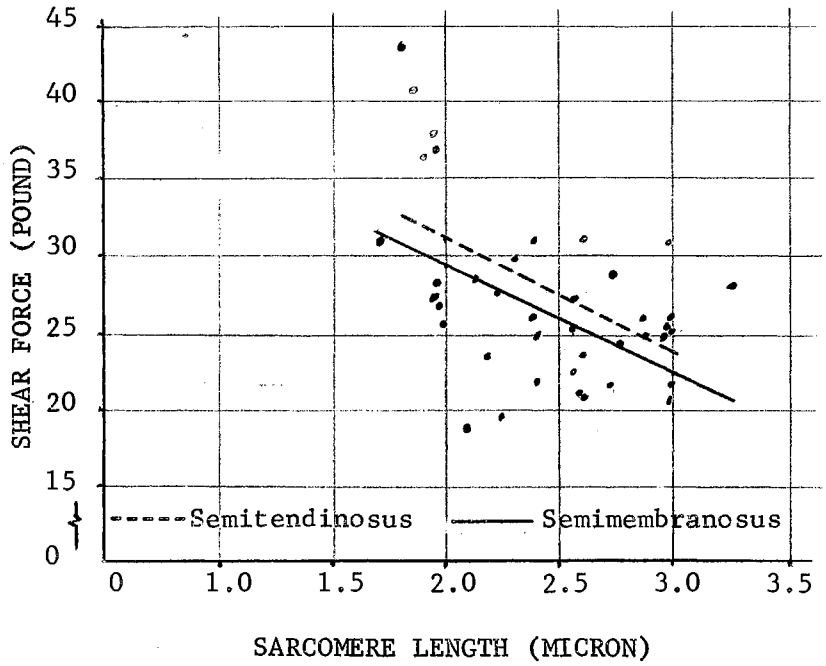



Figure 13. Relationship of Shear Force and Sarcomere Length.

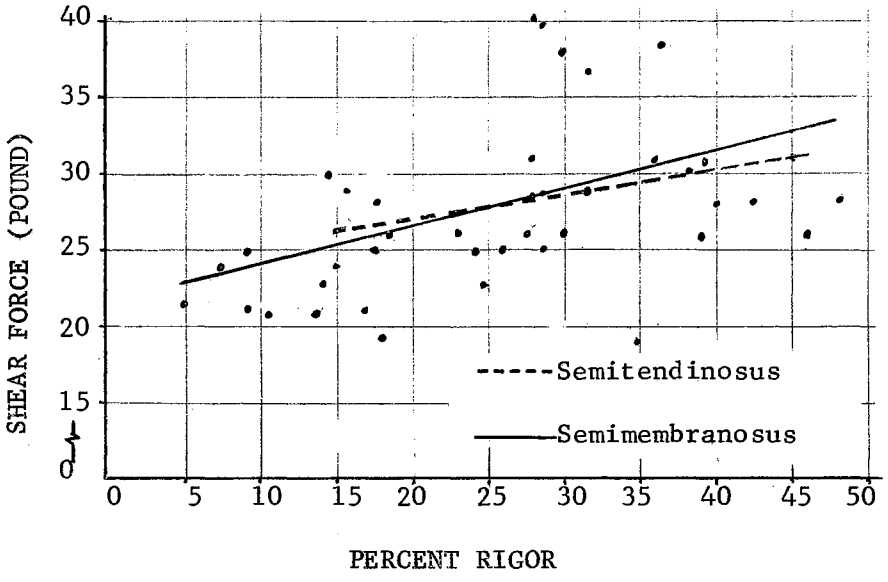



Figure 14. Relationship of Shear Force and Percentage Rigor.

## Relationship of Variables

A definite trend was noted for the relationship of sarcomere length and fiber diameter (Figure 15). As sarcomere length increased, fiber diameter decreased. This is in agreement with the work of Herring et al. (1965b) who found a negative correlation of -.82 between fiber diameter and sarcomere length. It seems logical that when the muscle fiber is in a stretched state the myofilaments are held apart, thus resulting in a smaller fiber diameter than if it was contracted and the myofilaments combined.

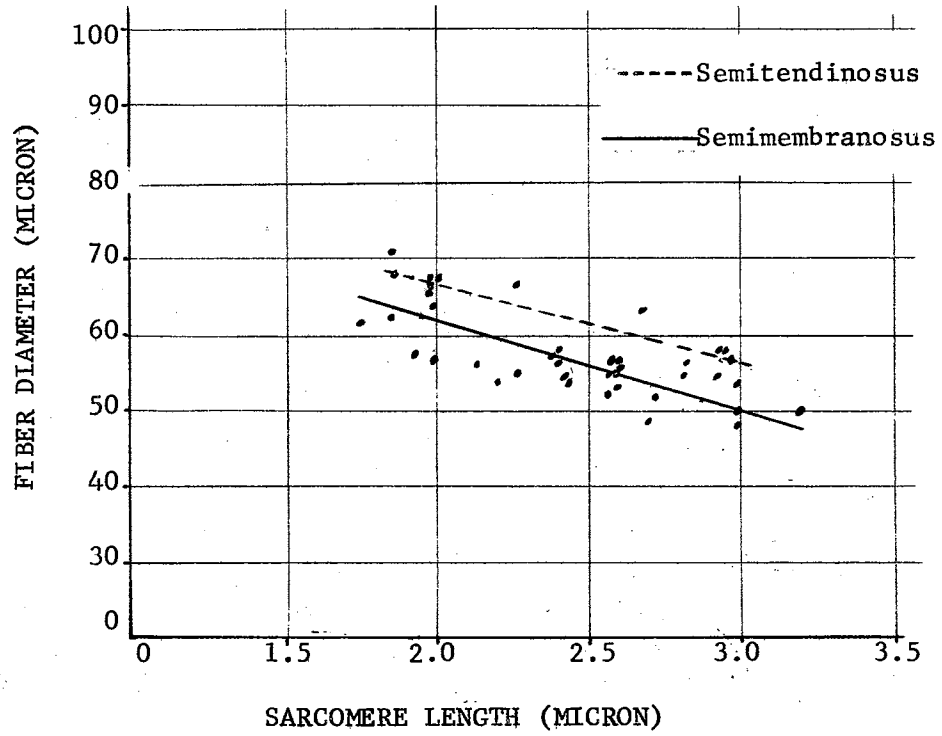



Figure 15. Relationship of Fiber Diameter and Sarcomere Length.

Both fiber kinking and muscular contraction are common physical effects of rigor. In an attempt to indicate a relationship of these variables a scattergram was plotted (Figure 16). No definite trend was noted due to the variability of the data, in fact, the two muscles indicated opposite directions of association.

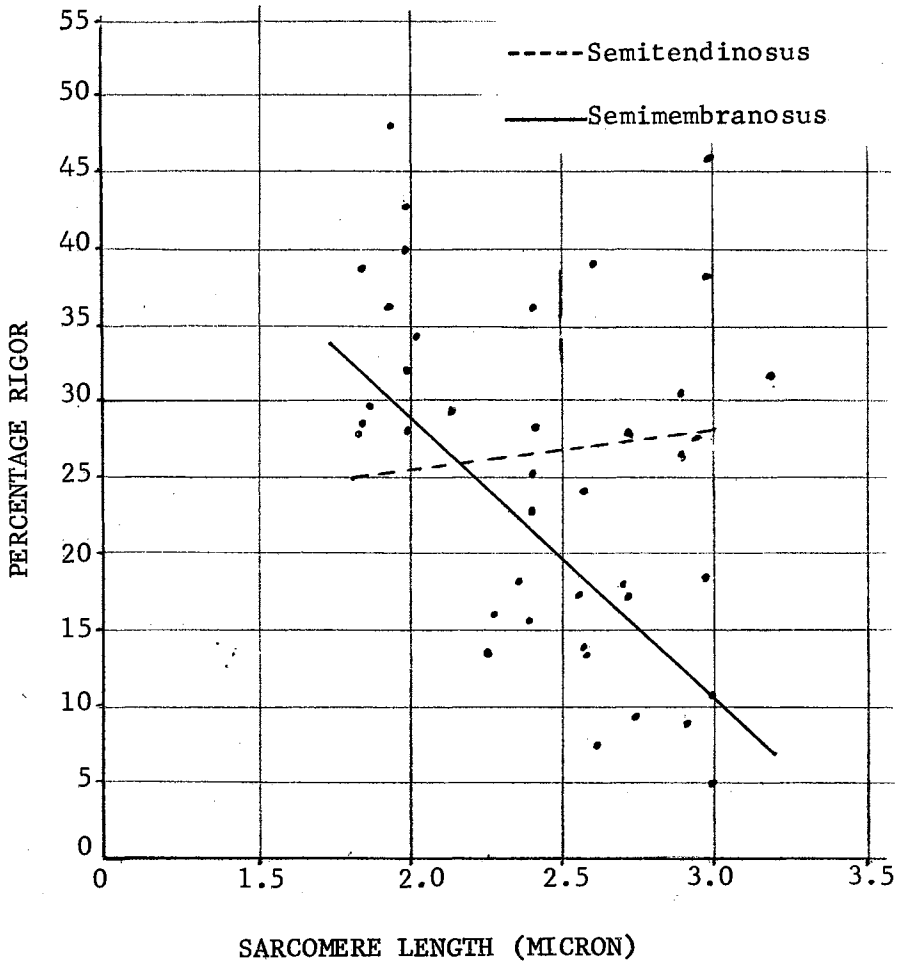



Figure 16. Relationship of Percentage Rigor and Sarcomere Length.

As indicated in (Figure 17) fiber diameter increased as did percentage rigor, thus indicating that the larger fibers may be subject to greater kinking and distortion.

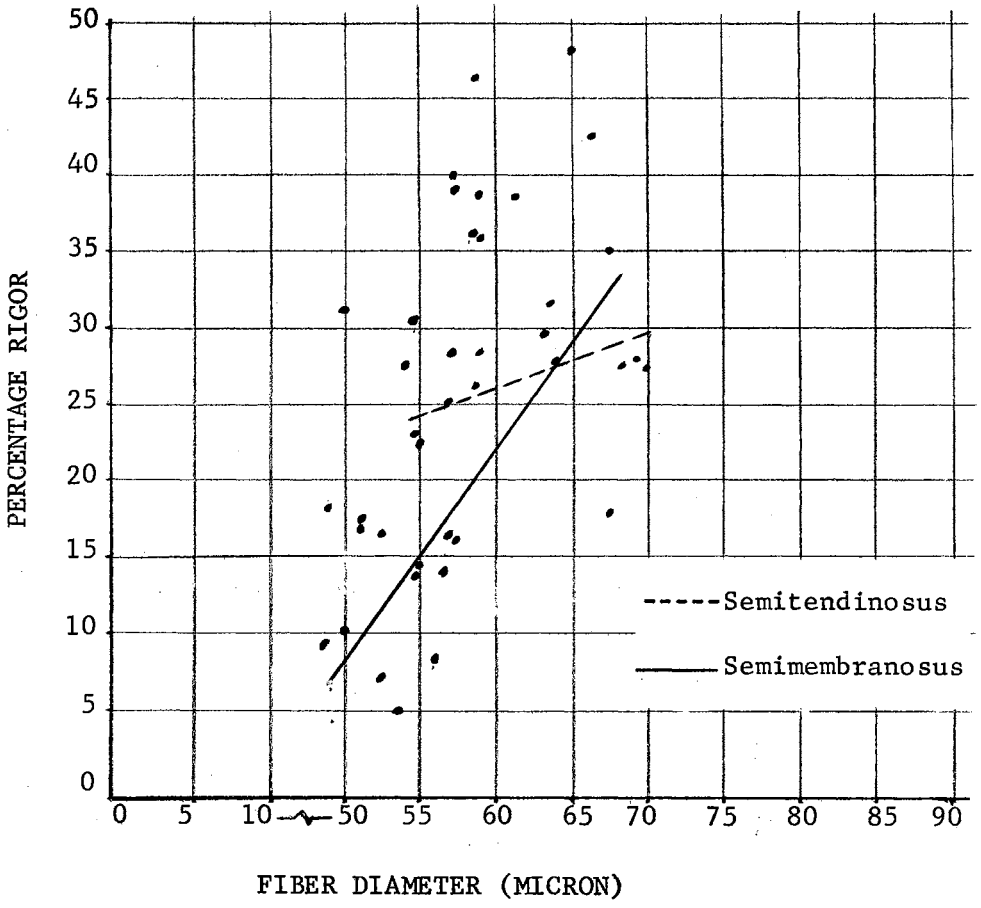



Figure 17. Relationship of Percentage Rigor and Fiber Diameter.

## SUMMARY AND CONCLUSIONS

Five steers ranging in weight from 433.02 kg. to 501.56 kg. of comparable finish and grade were obtained from the Oklahoma Agricultural Experiment Station herd. The steers were from similar managerial backgrounds and ranged in age from approximately 11 to 14 months. The animals were slaughtered in the meat laboratory in accordance with those practices normally employed. The semimembranosus and semitendinosus muscles were excised from the hindquarters approximately 45 minutes post-mortem, divided into four samples each (6 x 6 x 21 cm), and subjected to four degrees of tension(0,1000, 2500, 5000 gms) for 48 hours at 1.11 degrees centigrade. Histological and shear samples were then removed.

Histological testing consisted of measuring fiber diameters, sarcomere lengths, and percentage rigor, which was accomplished by the use of the regular light and phase-contrast microscopes. Shear force determinations, as an indication of tenderness, were made using the Warner-Bratzler shear machine.

A highly significant decrease ($P<.01$) in fiber diameter for both muscles was noted for the 1000 gram pull treatment, and no significant changes were found with succeeding increases in tension. Sarcomere length was found to increase significantly ($P<.01$) to the 2500 gram pull treatment and then evidently leveled off ($P<.05$). Both muscles exhibited less percentage rigor to the 1000 gram pull treatment, however after this point the semitendinosus muscle tended to increase

whereas the semimembranosus tended to decrease. In like manner, a considerable decrease was noted in shear force to the 1000 gram pull treatment for both muscles, but after this point a slight increase was noted for the semitendinosus muscle whereas the semimembranosus muscle did not change. Thus indicating that a relationship between percentage rigor and shear force may exist. In this regard a scattergram indicated that with an increase in sarcomere length there was a corresponding decrease in fiber diameter and shear force. No conclusions could be drawn as to the relationship of percentage rigor to sarcomere length and fiber diameter, because of variation in the data. However, a trend was noted in that the larger fibers (diameter) tended to encounter greater kinking and distortion. In all cases, the relationship of variables could not be interpreted as true correlations as the scattergrams were uncorrected for the tension treatments.

Conclusions from this investigation suggest that some muscle tension is necessary for maximum tenderness, and that there is a definite relationship of the structural condition of the muscle fiber to tenderness. Further work appears necessary as to how these structural conditions can be controlled or altered, and how rigor kinks and nodes are formed in the muscle fiber.

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# APPENDIX

TABLE V

ANALYSIS OF VARIANCE FOR FIBER DIAMETER OF TWO BOVINE MUSCLES AS INFLUENCED BY TENSION

| Source | df | SS | MS | F |
|---|---|---|---|---|
| Total | 39 | 1274.09 | - | - |
| Blocks | 4 | 161.87 | 40.47 | 4.935* |
| Groups | 7 | 882.74 | - | - |
| Muscles | 1 | 210.45 | 210.45 | 25.665* |
| Tension | 3 | 671.11 | 223.70 | 27.280* |
| Mus. x Ten. | 3 | 1.18 | 0.39 | 0.048 |
| Error | 28 | 229.48 | 8.20 | - |

* $P<.01$

TABLE VI

ANALYSIS OF VARIANCE FOR SARCOMERE LENGTH OF TWO BOVINE MUSCLES AS INFLUENCED BY TENSION

| Source | df | SS | MS | F |
|---|---|---|---|---|
| Total | 39 | 6.31 | - | - |
| Blocks | 4 | 0.46 | 0.12 | 4.000* |
| Groups | 7 | 4.95 | - | - |
| Muscles | 1 | 0.02 | .02 | 0.667 |
| Tension | 3 | 4.86 | 1.62 | 54.000** |
| Mus. x Ten. | 3 | 0.07 | .02 | 0.667 |
| Error | 28 | 0.90 | .03 | - |

* $P<.05$
** $P<.01$

TABLE VII

ANALYSIS OF VARIANCE FOR PERCENTAGE RIGOR OF TWO BOVINE MUSCLES AS INFLUENCED BY TENSION

| Source | df | SS | MS | F |
|---|---|---|---|---|
| Total | 39 | 4783.82 | - | - |
| Blocks | 4 | 389.75 | 97.44 | 1.415 |
| Groups | 7 | 2465.38 | - | - |
| Muscles | 1 | 437.72 | 437.72 | 6.335* |
| Tension | 3 | 1218.40 | 406.13 | 5.896** |
| Mus. x Ten. | 3 | 809.26 | 269.75 | 3.916* |
| Error | 28 | 1928.69 | 68.88 | - |

* $P<.05$
** $P<.01$

TABLE VIII

ANALYSIS OF VARIANCE FOR SHEAR FORCE OF TWO BOVINE MUSCLES AS INFLUENCED BY TENSION

| Source | df | SS | MS | F |
|---|---|---|---|---|
| Total | 39 | 1183.64 | - | - |
| Blocks | 4 | 292.81 | 73.20 | 3.440* |
| Groups | 7 | 395.12 | - | - |
| Muscles | 1 | 34.38 | 34.38 | 1.616 |
| Tension | 3 | 231.18 | 77.06 | 3.621* |
| Mus. x Ten. | 3 | 29.56 | 9.85 | 0.462 |
| Error | 28 | 595.71 | 21.38 | - |

* $P<.05$

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