COMPARISON OF HISTOLOGICAL CHARACTERISTICS WITH PALATABILITY FACTORS OF THE LONGISSIMUS DORSI MUSCLE FROM EWE, RAM, AND WETHER MARKET LAMBS

by

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INTRODUCTION

Kansas, Texas, and California are the 3 most important states for early lambs (Coppersmith, 1965). There has been severe price discrimination against ram lambs in the channels through which Kansas lambs are marketed. However, according to Kropf (1963) rams appear to have an advantage over ewes in a faster rate of gain, greater muscular development, and less fat deposition, but if lambs are marketed at a suitable weight, sex should not alter carcass quality characteristics. However, there has been conflicting evidence concerning effect of the quantity, distribution and composition of fat in muscle tissue on its organoleptic properties.

There are many reports in the literature on the effect of breed, feed, and age on the quality of lamb as measured by market grade. Only a few of these studies, however, give information on palatability of cooked lamb, and none were found that give information regarding histological characteristics of lamb muscle as related to palatability. So, it seemed important to compare the histological characteristics with the palatability of muscle from lambs of different sex and grade. In addition, microscopic examination of the muscle tissue will provide further information regarding lamb carcass quality characteristics.

The Departments of Animal Husbandry and Foods and Nutrition have conducted research to compare ewe, ram, and wether lambs relative to slaughter data, grade factors, muscling, cooking losses, and organoleptic characteristics. Samples of raw and cooked longissimus dorsi (LD) muscle from right loins of these lambs that varied in grade from Prime to Choice were available for histological study. Also, panel scores for tenderness, juiciness, and flavor; Warner-Bratzler shear values; and press fluid yields were available for the cooked loins from which the histological samples were taken. This study was carried out to meet the following objectives: (1) to estimate and compare the relative quantity of intramuscular fat and connective tissue, and the relative width of muscle fibers in the LD muscle, raw and cooked, from ewe, ram, and wether lambs, (2) to observe the microscopic appearance of muscle fibers and connective tissue, and (3) to study the relationship of the histological estimate of these factors to the data for palatability factors.

REVIEW OF LITERATURE

General Structure and Composition of Skeletal Muscle

A discussion of the structure and composition of skeletal muscle is presented in textbooks of histology such as those by Lambert (1944), Maximow and Bloom (1957), Finerty and Cowdry (1960), and Smith and Copenhaver (1964). According to these authors, skeletal muscle is made up of 3 types of tissue; namely, muscle fibers, connective tissue, and fat.

Muscle fibers. The smallest independent unit is the fiber. It is elongated, cylindrical, and multinucleated; and is enclosed in the sarcolemma, a thin, colorless, elastic membrane. The length of the fiber varies from 1 to 60 mm, and the diameter varies from 10 to 100 µ, according to muscle and species of the animal. The fibers are grouped together in bundles called fasciculi, and are characterised by cross and longitudinal striations. The cross striations are attributed to the regular alternation of light (I or J) and dark (A or Q) bands of myofibrils. There is a Z band in the middle of the light band. The portion between the 2 Z bands is known as a sarcomere, a functional and structural muscular unit. Myofibrils are composed of bundles of myosin filament and appear as long, parallel threads that do not branch, and which impart to the muscle fiber an appearance of longitudinal striations.

The thickness of the myofibrils, the contracting tissue fluctuates from 1 to 4 µ. The cytoplasm, which fills the space between myofibrils, is known as sarcoplasm. The amount of sarcoplasm varies from muscle to muscle, but generally red muscle contains more sarcoplasm than white muscle.

Myofibrils are composed of sarcoplasmic and contractile protein. Sarcoplasmic protein can be extracted from finely homogenized muscle by means of dilute salt solution. There are 20 or more enzymes in this type of protein that are involved in the anaerobic glycolytic cycle. Most of the enzymes are albumin in nature, and are easily denatured under mild acidic conditions. In addition to enzymes, mitochondrial fragments and particles of the sarcoplasmic reticulum also are present. Mitochondrial fragments are involved in the aerobic metabolism of muscle, whereas the particles of sarcoplasmic reticulum are associated with the system conducting impulses to the contractile filament (Bendall, 1964).

Contractile protein is made up of 2 proteins, myosin and actin. Also present, in a small quantity, in mammalian muscle is tropomyosin B, which resembles myosin in its chemical constituents, but differs markedly in most of its properties (Bailey, 1948). Huxley (1963), in electron microscopic studies on the structure of protein filament in striated muscle, observed tropomyosin in thin cross sections at the Z line.

Myosin is the major component (about 38% of the protein) of the thick filament of striated muscle. Several studies described by Szent-Györgyi (1951) indicated that myosin obtained by short extraction has a relative low viscosity, whereas myosin obtained by prolonged extraction has relative high viscosity. The latter proved to be a combination for a second protein, actin, with myosin thus forming the highly viscous actomyosin that is responsible for muscle contraction.

Szent-Györgyi (1953) reported that short trypsic digestion of myosin produced 2 subunits, light (L-) and heavy (H-) meromyosin. H-meromyosin retains the enzymic properties (Adenosine triphosphate-ATPase) and actin-combining-power of myosin, whereas L-meromyosin appears to be of purely structural importance. L-meromyosin is nearly 100% helical, whereas H-meromyosin is less than 45% helical. Most of the SH groups and proline residues are found in H-meromyosin.

Actin represents about 13% of the total muscle protein, or is present in muscle with respect to myosin in a ratio of 1:3.

Actin can exist in either of 2 forms, G-actin (globular), which is a monomer, or F-actin (fibrous), which is a polymer. Change occurs in the 2 forms during contraction; however, ATP is required for depolymerization.

Other constituents in muscle such as water, inorganic salts, pigments, and nitrogenous and non-nitrogenous extractives were reviewed by Lowe (1955, p. 209 to 212).

Connective tissue. Connective tissue in the animal body supports muscle fibers, and individual fibers are held in bundles or fasciculi by an extremely thin network of connective tissue called the endomysium. The fasciculi are surrounded by connective tissue, the perimysium, whereas the connective tissue enclosing each muscle is known as the epimysium.

All connective tissue contains collagenous fibers, which run in all directions, and their ends cannot be found. These fibers may be straight or wavy, and consist of bundles of parallel fibrils. Although the fibers branch considerably, the fibrils do not. Collagenous fibers are not elastic, but are quite flexible. A bundle of collagenous tissue appears white, and thus is called white fibrous connective tissue.

The main protein in connective tissue is collagen. The collagen molecule consists of a triple helic, about 2800 A° in length and 15 A° in diameter. The helical forms are held by intra- and interhelical cross bonds, which give high elasticity to collagen. It is believed that those cross-bonds are ester links between serine-OH residues and carboxyl groups. Collagen is high in the amino acids glycine, proline, and hydroxyproline, and contains varying amounts of all other amino acids except tryptophan and cystine.

At netural pH and physiological ionic strength collagen has a high and compact structure and great strength. By lowering both pH and ionic strength many types of collagens swell and finally dissolve, but the triple helices of the molecule are still intact as in the native form. However, the helices in solution unravel and denature at temperatures between 35° and 40°C. In this stage collagen loses its high elastic modules and breaks easily (Bendall, 1964). Collagen is readily attacked by pepsin but exhibits stability to trypsin.

Also, all connective tissue contains some elastic fibers. These fibers are homogeneous instead of fibrillar, are thinner than collagenous fibers and run in all directions. They appear straight under natural conditions, but when teased on to a slide they appear wavy or spiral. Elastic tissue stretches easily, and when many fibers are massed together, the tissue appears yellowish in color, and is called yellow connective tissue.

Elastin is a different type of protein from the native collagen found in connective tissue. It is less elastic than collagen, and the molecule is of indefinite size, consists of long polypeptide chains, held together sideways by cross links, about every 30 amino acid units. It occurs mainly in epi- and perimysium, and is resistant to pepsin.

The ground substance, a matrix for collagenous and elastic fibers, contains a mucopolysaccharide that varies from a fluid to a gel-like consistency. The composition of the ground substance varies with age and activity of the animal.

Fat. Fat is a special type of tissue in which the cells occur isolated or in groups in loose connective tissue. In certain places in the animal body, fat is present in large quantities, known as adipose tissue. Fat tends to be deposited first around organs and under the skin, then between the muscles, and finally intramuscularly (Lowe, 1955 p. 198-199). Most of the intramuscular fat is first deposited between fasciculi and then between muscle fibers. When intramuscular fat is visible without magnification, it is known as marbling (Blumer, 1963).

Fats are composed principally of the glycerol esters of the straight chain carboxylic acids, having an even number of carbon atoms. Triglycerides are predominate in meat fats, although small amounts of mono- and diglycerides may be present. Fats are chemically homogeneous, but in addition to the triglycerides, meat fats contain other substances in small amounts. Such components may be steroids, protein fragments, free fatty acids, water, and phospholipids in which phosphoric acid has replaced one of the fatty acids (Dugan and Slover, 1960).

Relationship of Histological Characteristics and Tenderness of Skeletal Muscle

The relationship of microscopic characteristics of muscle fibers, connective tissue, and fat to the tenderness of muscle has been investigated by many workers.

Muscle fibers. Length and diameter of muscle fibers vary with the muscle and species of the animal, (Maximow and Bloom, 1957). Several authors (Hammond, 1932; Hiner et al., 1953; and Ramsbottom et al., 1945) stated that as the animal matures the muscle fiber diameter increases and tenderness decreases. According to Hiner et al. (1953) fibers from less active muscle increase in diameter more than those from active muscles. In a study of beef muscle they found that, in terms of percentage, the psoas major (PM) muscle fibers increased in diameter the most and the serratus ventralis neck muscle fibers increased the least, as the animal grew.

The number of fibers per bundle in beef muscle adductor (AD), LD, semitendinosis (ST), and triceps brachi (TB) was counted by Brady (1937), who concluded that the number of fibers in a bundle could be used as a measure of texture of meat. He stated that the greater the number of fibers in a bundle the "finer" the texture; and since texture is an indication of tenderness, the "finer" the texture, the more tender the meat. Brady's (1937) work was done on muscles from cows and steers and on fresh and aged tissue.

Tuma et al. (1962) studied the relationship of fiber diameter to tenderness as influenced by age of animal. They used 33 Herefords from 5 age groups between 6 and 90 months old, and found a gradual increase in fiber diameter with increasing age of animal. They also observed a significant dorsal, middle, and lateral "position" effect for fiber diameter in the LD, in which the fiber diameter decreased from the dorsal to lateral position. They concluded that fiber diameter, age of the animal, and tenderness were interrelated; fiber diameter increased and tenderness decreased with increasing age of the animal. However, within any one age group there seemed to be little relationship between fiber diameter and tenderness.

Satorius and Child (1938) and Joubert (1956) used an ocular micrometer to measure the diameter of muscle fibers. Satorius and Child noted a decrease in fiber diameter when AD and LD muscles were heated from the raw state to an internal temperature of 58°C. Tenderness of the AD was not affected, whereas the LD became more tender with heating to 58°C. Diameter of muscle fibers decreased and tenderness increased in ST heated to 67°C; however, with further heating (between 67 and 75°C) the diameter of the muscle fibers increased slightly and tenderness decreased. From observations on the LD, Joubert (1956) concluded that tenderness was influenced by a complexity of factors apart from muscle fiber diameter.

Fiber extensibility was determined by Wang _et_ _al_. (1956) by measuring the force required to break single muscle fibers held by tweezers 5 mm apart under the microscope. The farther apart the tweezers could be moved before the fiber broke, the less tender the fiber. They found that the extensibility of fibers from cooked meat was related significantly ($P = 0.05$) to the shear strength and tenderness scores of the meat in 3 of the 5 carcass classes studied.

Connective _tissue_. The quantity and character of connective tissue is considered one of the factors that influence the tenderness of meat. In a histological study, Ramsbottom _et_ _al_. (1945) determined, by using a numerical scoring system, the relative quantity of elastic and collagenous connective tissue in certain beef muscles. They found that in superficial pectoral muscle there was large and well defined fasciculi with extensive connective tissue, which gave a coarse structure. The smooth, fine texture of the PM did not have enough connective tissue to divide the muscle fibers into distinct bundles, thus the muscle was extremely tender (organoleptic rating) and had a low shear reading. A better correlation was obtained between the quantity of connective tissue in raw muscle and shear readings than between the quantity of connective tissue in cooked muscle and shear readings.

Harrison et al. (1949) found an agreement between the numerical histological rating for quantity of collagenous and elastic tissue and tenderness in certain beef muscles. They listed several muscles in decreasing order of tenderness for cooked meat: PM, LD (rib section), ST, semimembranosus (SM), and LD (loin section).

Carpenter et al. (1963) studied the relationship of the quantity of collagenous and elastic tissue, as measured by histological methods, to the organoleptic properties of the LD muscle from 78 carcasses of pork animals varying in age. They found no significant relationship between tenderness measurement and total quantity of connective tissue, because most of the muscle contained a similar quantity of connective tissue. However, the LD muscles from the older animals contained thicker or coarser collagenous tissue strands than those from younger animals. The coarseness of the collagenous tissue strands was inversely related to tenderness, and with an increase in intramuscular fat, there was a significant decrease in the coarseness of connective tissue strands.

Strandine et al. (1949) classified representative beef muscles into 4 groups dependent on size of the fasciculi and thickness of the perimysium. There was a general correlation between fasciculi-connective tissue patterns and tenderness measurements of the muscles. Tenderness was associated with muscles having indistinct fasciculi.

Fat. It was pointed out by Wang *et al*. (1954) that it is not the total quantity of fat in a muscle, but the way it is distributed throughout the muscle that affects tenderness appreciably. In a histological study they measured the "linear" fat, i.e., the longest axis of each fat island in a microscopic section of raw beef, and found consistently that the quantity of "linear" fat of raw meat correlated with tenderness for cooked samples. The higher the "linear" fat content of beef, the more tender the cooked meat.

Carpenter *et al*. (1961) studied intramuscular fat in microscopic sections and measured ether extractable material from pork loin. They found that with an increase in ether extract there was a highly significant increase in finely dispersed fat cells.

According to Goll *et al*. (1965) a fine texture and even distribution of fat was associated with increase in tenderness of beef muscle. Scores for marbling distribution and texture were significantly related to all measurements of tenderness conducted on cooked muscle.

Lewis et al. (1958a) used numerical scoring and photomicrographic methods to estimate the quantity and distribution of fat in the pectoralis major and gluteus primus muscles of turkey. They observed a slightly larger quantity and a wider distribution of fat in the gluteus primus than in the pectoralis major. Lewis et al. (1958b) reported tenderness scores for those muscles from the same birds used in the histological study. Over a 16 mo period of frozen storage, the palatability committee found little consistent difference in tenderness between the 2 muscles, and in general, the difference was not significant.

Relationship of Intramuscular Fat (Marbling) and Palatability of Skeletal Muscle

When fat is dispersed throughout the muscle it is referred to as marbling. There is controversy regarding marbling as a factor affecting the palatability of skeletal muscle.

Lowe (1948) listed the finish and distribution of fat in poultry as factors that may influence its flavor and juiciness. Gaddis et al. (1950) reported that data for 97 beef rib samples showed that with an increase in the percentage of fat in the press fluid, there was a tendency for percentage of total press fluid to decrease and for panel scores for quantity and quality of juice to increase.

Muscle tissue well marbled with fat has been associated with tenderness. Kauffman et al. (1964) studied pork loin and ham muscles from 439 carcasses for intramuscular fat and its relationship to organoleptic characteristics. They found that a large quantity of fat was associated with high flavor, tenderness, and especially high juiciness ratings of the cooked fresh product. The large quantity of fat also was accompanied by low curing and cooking shrinkage. Harrington and Pearson (1962) measured the tenderness of pork loin with various degrees of marbling by means of chew count, and found a significant correlation between mean chew count and marbling, the less marbled loins being somewhat tougher than those with a greater degree of marbling.

Paul et al. (1964) found that as the age of lamb increased, the marbling score and separable fat content also increased. Cuts from older animals tended to be more tender, juicier, finer in texture, and had a more mature flavor of the lean than cuts from younger animals.

A study by Cover (1956) indicated that marbling is only moderately associated with beef tenderness. Moreover, Gilpin et al. (1965) found that beef steaks from highly marbled carcasses scored only slightly higher in quality than those from less well marbled carcasses. The relationship between marbling of a muscle (LD or semitendinosus) and the palatability characteristics of tenderness, juiciness, and flavor of the muscle was generally inconsistent, and correlation coefficients were low.

The effect of marbling and maturity on beef muscle characteristics was studied by Walter et al. (1965). They found that marbling had no statistically significant effect on tenderness, but tenderness decreased with advancing carcass maturity.

Blumer (1963) concluded from data reported in the literature that if a reasonable estimation of association of marbling and panel tenderness scores is made, the range of values of 0.01 to 36% of the variance in tenderness of beef is accountable. He said that according to the number of samples represented among the several studies he reviewed, the average value would be about 5%.

Factors Affecting the Characteristics of Lamb Carcasses

Quality of the carcass of meat animals is important to consumers, the meat industry, and food science and animal science researchers. There are many factors, such as age, sex, breed, and fatness that vitally affect the growth and development of qualitative and quantitative characteristics of the lamb carcass.

Age. Age of the animal affects the percentage carcass yield as influenced by the development of various organs of the body and the proportion of the change, mainly by an increase of the muscle and fat in the carcass as the animal ages. Also, with increase in age there is slacking of the growth of certain organs (Hammond, 1932).

Carpenter and King (1965a) evaluated 259 ram, wether, and ewe carcasses for maturity, and measured the tenderness of the chops from these carcasses with the Warner-Bratzler shearing apparatus. They reported that with an increase in the age of the lamb the maturity score for the carcasses and tenderness of the chops increased, whereas cooking loss from the chops decreased.

Sex. Since sex has an effect upon growth, it also affects the carcass. Hammond (1932) compared ram, ewe, and wether lambs at 5, 7, and 14 months of age, and found that at all 3 ages the wethers produced a higher percentage carcass yield than ewes. Calculated on the basis of live weight of the animal, yields at 5 months of age were 53.7 and 50.7% for wethers and ewes, respectively. At 7 months the yields were 52.4 and 51.0% for wethers and ewes, whereas at 14 months yields were 51.5 and 51.1% for the 2 groups. He also found that at 5 months of age the carcass yield was higher for rams than for ewes.

Carpenter and King (1965a) evaluated the cutability of 314 carcasses from ram, ewe, and wether lambs ranging in weight from 35.4 to 56.2 kg. They found that rams were significantly higher in weight per day of age, yield of fore-saddle, loin-eye area, and retail rack than the ewes and wethers. Ram carcasses also had less fat over the loin-eye and a lower percentage of kidney fat than ewes.

In contrast to Carpenter and King's (1965a) work, Knight and Foote (1965) found differences between ewes and wethers were minimal when they studied selected characteristics of 20 wether and 20 ewe carcasses. Carcass weight for ewes was 25.56 kg, whereas for wethers it was 26.82 kg. Dressing percent was 55.6 and 55.2, and carcass length was 66.4 and 67.6 cm for ewes and rams, respectively.

Breed. Hammond (1932) suggested that since the percentage of carcass yield increases with age of the animal, early maturity gives a high percentage yield at a young age. He compared 2 early maturity breeds, (Suffolk and Southdown) with 2 late maturing breeds (Lincoln and Welsh) at 5 months of age. He found that late maturing breeds differed from the early maturing ones in development of muscle, bone, and fat in the carcass. Carcass yield calculated from live weight was 53.7% for Suffolk and 53.6% for Southdown, whereas the yield was 51.1 and 50.0% for Lincoln and Welsh, respectively.

Knight and Foote (1965) compared carcass measurements for 4 breeds (Rambouillet, Columbia, Hampshire-whiteface cross, and Suffolk-whiteface cross). They found that the characteristics considered indicative of carcass quality were similar. Carcass weight for Rambouillet was 22.95 kg, and for Columbia, Hampshire-whiteface cross and Suffolk-whiteface cross was 23.54, 24.30, and 24.16 kg, respectively.

Fatness of the animal. According to Hammond (1932), carcass yield, calculated on live weight, increased as the fatness of the animal increased. He determined carcass yield for "Smithfield" wethers at 11 months of age, and found a yield of 56.9% when caul fat averaged 1.8 lb per carcass and gut fat 1.3 lb. Carcass yield was 61.1% when caul and gut fat were 3.9 lb and 1.5 lb, respectively. Carpenter et al. (1961) reported the effect of fatness on carcass yield of retail cuts. As the carcass weight increased there was an increase in fatness, which resulted in a decrease in retail cuts.

Organoleptic Characteristics of Lamb Muscle

Tenderness, juiciness, flavor, and color usually are considered when evaluating the organoleptic quality of lamb muscle. Those 4 factors are so closely related that it is difficult to separate them.

Tenderness. Tenderness is one of the most important characteristics influencing the palatability of meat, and it is affected by several factors, such as age and sex of the animal, ripening of the carcass, percentage of separable and intramuscular fat, and location of the muscle in the carcass. Paul et al. (1964) reported that as the age of lamb and the percentage of separable fat in the carcass increased, there was an increase in tenderness of the muscle. A comparison was made between lambs 10 to 12 months of age (old crop) with those 5½ months of age (new crop). The mean tenderness score for loin chops from new crop lambs was 4.8, whereas for the old crop, it was 5.8 on a 10 point scale. Mean tenderness scores were 5.1 and 6.8 when the values for separable fat ranged from 10.0 to 11.9% and from 40.0 to 49.9%, respectively.

Batcher et al. (1962) reported that panel scores for the rib-loin cut showed that lamb meat became significantly less tender as the age of animal increased. However, in leg cuts a decrease in tenderness scores with the increasing age of the animal was not significant. They suggested that lack of significant differences for leg cuts may be accounted for by the narrow range of scores at all ages. On a 10 point scale, mean scores ranged from 7.8 to 5.5 for rib-loin, and from 6.9 to 5.5 for leg cuts from animals ranging 4 to 14 months of age. Cuts with greater proportion of separable fat in relation to lean generally were no more tender than those with less separable fat.

Carpenter and King (1965b) measured tenderness by means of Warner-Bratzler shear and marbling scores, and found that oven-broiled and electronically-cooked rib chops with a marbling score of 5 (12 point scale was used, where 12 represented abundant and 1 devoid of marbling) were significantly more tender than those receiving a marbling score of 3. The difference was significant at the 5% level for oven-broiled chops and at the 0.1% level for electronically-cooked chops. These authors also reported that rib chops from wethers were slightly more tender than those from ewes, and rams provided less tender chops than the other 2 groups.

Cover et al. (1944) studied the effect of fatness of the lamb on tenderness of the meat from full-fed and limited-fed animals. Full-fed lambs were somewhat more tender than limited-fed animals. The mean shear score for SM muscle from full-fed animals was 21.3 lb, whereas for limited-fed, it was 23.0 lb.

Gifford (1965) investigated the tenderness of steaks from the upper- and lower-leg, and the sirloin slices from the same wholesale cut of lamb. Also, the effect of 3 methods of cookery (pan-broil, fry, and braise) were studied. He found no evidence of a significant effect of those 3 methods of cookery on tenderness. Upper-leg slices were most tender, but only slightly more so than sirloin slices. Steaks from the lower-leg were significantly less tender than those from the other 2 locations.

Juiciness. The quantity and quality of juice formed when meat is chewed are other factors considered when judging the overall quality of a piece of cooked meat. Paul et al. (1964) found that 3 cuts (loin chops, leg, and shoulder) from old-crop lambs tended to be juicier than those from new-crop lambs, especially at high levels of separable fat.

Batcher et al. (1962) reported that juiciness of either rib-loin or leg cuts was not associated consistently with age of the animal. However, juiciness scores were slightly higher for rib-loin than for leg cuts.

Flavor. Flavor is a subtle and complex sensation through which the presence and identity of foods are determined (Meyer, 1960). Gifford (1965) found that there was little difference in flavor of steaks from legs and sirloin of lamb as associated with the location of the slice, or with any of the 3 methods of cookery studied.

Batcher et al. (1962) studied the effect of fatness and age of the animal on flavor, and found that organoleptic scores were not consistently associated with age of the animal. Panel scores for rib-loin from animals 4-5 moths old averaged 7.3; 6-8 months, 6.7; and 11-14 months, 6.5. With leg cuts, a lower natural flavor was obtained for young than for older animals. According to these authors, the amount of separable and intramuscular fat did not affect the flavor of the cooked meat. Hofstrand and Jacobson (1960) concluded that the quantity of fat in lamb broth did not affect the flavor of the broth.

Color. Carpenter and King (1965a) investigated the interrelationship of color and tenderness of lamb rib-chops, and found that minor differences observed in tenderness were associated with variation in visual color scores.

Paul et al. (1964) reported that color scores for loin chops and legs did not show clear cut trends with age of the animal.

However, the color score for cooked chops increased with increasing fat in the raw chops.

PROCEDURE

A total of 152 samples of small blocks of raw and cooked LD muscle from ewe, ram, and wether lambs (Tables 5 and 6, Appendix), which were fixed in physiological salt solution and formalin, were prepared for microscopic study. The microscopic study included an estimation of the relative quantity of intramuscular fat and connective tissue, and the relative width of muscle fibers. Five slides were prepared from sections cut from each sample.

Sectioning. The tissue to be sectioned was removed from the fixative, blotted on a paper towel, and a piece (about 6 mm sq) was cut parallel to the long axis of the fibers; then, placed in a small dish filled with water for at least 3 min. By means of a glass rod, a few drops of gum arabic were put on the corrugated surface of the freezing plate of a clinical microtome. The block of tissue was laid on the gum arabic with forceps, and covered with another drop of gum arabic. The height of the freezing chamber was adjusted so that the upper edge of the tissue was level with the blade of the microtome knife, which was conditioned just before use by stropping. The automatic feed mechanism was set to cut sections 15 to 20 μ thick. The tissue was frozen by opening and closing the small valve at the chamber several times in quick succession until the gum arabic appeared white, instead of translucent.

A small dish of tap water was used to receive the sections as they were cut. Ice cubes were added to chill the water and prevent a sudden change from the frozen state, and to aid in preventing the sections from breaking. The sections were removed from the microtome knife by means of a small camel's-hair brush and transferred to the dish of cold water. The brush was wiped often transferring sections to prevent excess moisture accumulating on the knife blade, which would cause thawing of the frozen block.

If the block of tissue was frozen too hard, which caused the section to splinter, the tissue was allowed to thaw for a few minutes, or was rubbed lightly with the finger. If tissue thawed slightly and slipped, it was refrozen by turning the valve to the freezing chamber on and off a few times.

Staining. The sections were transferred through the staining solutions (freshly prepared Sudan IV and Harris' hematoxylin) in a small rectangular wire basket. Sudan IV imparted a bright red-orange to fat, whereas Harris' hematoxylin stained the muscle fiber a bluish purple. The connective tissue was darker bluish purple to gray.

The wire basket was lifted from the water and passed through reagents in the following manner for staining:

1. Dipped for 1 min in 30% ethyl alcohol
2. Dipped for 1 min in 50% ethyl alcohol
3. Dipped for 1 min in 70% ethyl alcohol
4. Stained in Sudan IV solution 3 min
5. Washed 1 min in 70% ethyl alcohol
6. Washed 1 min in 50% ethyl alcohol

7. Washed 1 min in 30% ethyl alcohol
8. Washed in distilled water 1 min
9. Stained in hematoxylin 30 sec
10. Washed throughly in tap water, through at least 2 baths, and transferred to a dish (5.5 in. diam) containing tap water.

Mounting. Microscope slides and cover glasses were cleaned with 95% ethyl alcohol, and a slide was held at a sharp angle with one end resting on the bottom of the dish containing the stained sections. One section was teased up on the slide with a fine camel's-hair brush, and water allowed to run off as the slide was lifted slowly from the dish so that the tissue laid flat without folds or wrinkles. If the tissue did not lay properly, a drop of water was placed on it, or the slide was redipped slowly into the water. Excess water was wiped off the slide with a lint free cloth.

Glycerine jelly, used for mounting, was kept the desired consistency by placing it in a hot water bath. A drop of warm glycerine jelly was put at one end of the tissue by means of a glass rod. Then a cover glass was set at the same end as the drop of glycerine jelly so that the glycerine jelly flowed along the edge of the cover glass. The cover glass was lowered slowly over the section to allow air bubbles to escape, and the glycerine jelly to run smoothly under the cover glass. If small bubbles appeared, the cover glass was gently pressed with the handle end of the camel's-hair brush near the bubble until it disappeared.

Microscopic study. Stained and mounted sections from the

LD muscle were studied (200x and 860x magnification) by 2 or 3 observers for differences among ewe, ram, and wether lambs in the histological estimate for quantity of fat and connective tissue, and for width of the fibers. For the former 2 factors, a numerical scoring system described by Ramsbottom et al. (1945) was used, and is given below:

Quantity	Score
Large	7
Medium	5
Small	3
None	1

The average score for 5 sections from each sample was calculated. Also, the pattern of distribution of fat within the muscle and size of the fat globules were noted, and the connective tissue was observed and described in terms such as wavy, straight, or granulated fibers.

The relative width of the muscle fibers was obtained by counting the number of fibers in an entire microscopic field (3 to 4 fields were used for each section) at a magnification of 860x. Also, the width of muscle fibers was measured by 1 observer with a calibrated ocular micrometer (procedure is given in Appendix p. 44). Three fibers in each section were measured. In addition, muscle fibers were observed, and described as straight, kinked or twisted, accordian plaited, or granulated (Form 1, Appendix).

RESULTS AND DISCUSSION

Histological methods are used to study fundamental structure of meat and changes that occur in muscle that affect the palatability of meat. However, there are limitations that must be faced with this type of work. Lowe (1948) pointed out that it must be kept in mind that only a small portion of the whole muscle appears on the slide, the original samples taken from the muscle represent only a small part of the whole, and different portions of the same muscle may appear to contain varying quantities of fat and connective tissue. According to Hulle et al. (1965), in spite of all limitations, histological study of frozen tissue is useful in studying the structure of meat.

In the study reported here, sections of LD muscle (raw and cooked) from ewes, rams, and wethers from lambs raised in 1964 and 1965 were examined for the quantity and distribution of fat, quantity and characteristics of connective tissue, and width and characteristics of muscle fibers. Also, the relationship of histological data to data for palatability factors was studied.

Fat

Appearance and distribution. Intramuscular fat in sections of raw and cooked LD muscle from ewes, rams, and wethers was similar in appearance. In general, the cells were medium in size (Table 1).

The distribution of fat was slightly different in rams from that in ewes or wethers. On the whole, fat in LD muscle of rams

Table 1. Average values for estimated quantity of fat and connective tissue, size of fat globules, and relative width of muscle fibers from raw and cooked LD muscle from lambs raised in 1964 and 1965.

Factors	Rams				Wethers				Ewes			
	1964		1965		1964		1965		1964		1965	
	Raw	Cooked	Raw	Cooked	Raw	Cooked	Raw	Cooked	Raw	Cooked	Raw	Cooked
Quantity of fat[a]	3.7	4.7	4.4	4.7	4.6	4.2	4.2	4.4	4.2	4.8	4.1	4.5
Fat globules, size[b]	4.9	4.7	4.8	4.6	5.2	4.4	4.8	3.9	5.3	4.7	4.9	4.7
Quantity of connective tissue[a]	4.5	5.0	4.6	4.8	4.9	5.0	5.1	3.8	4.8	5.0	4.9	4.7
Width of muscle fibers												
No. of fibers in field (x860)	3-4	---	3-5	---	3-4	---	3-5	---	3-4	---	3-5	---
Diameter, μ	41.7	39.7	41.2	37.9	40.4	38.8	40.5	38.3	39.9	39.2	42.6	37.9

Scoring key

a	Large	- 7	b	Large	- 7
	Medium	- 5		Medium	- 5
	Small	- 3		Small	- 3
	None	- 1			

occurred a little oftener in clustered than in scattered form, whereas in ewes and wethers it tended to be distributed throughout the sections (Table 2, Fig. 1a and b). Hammond (1932) also found that marbling in rams was coarser than in wethers, i.e., in rams, the fat was distributed in large thick patches between the bundles of fibers, whereas in wethers it was more finely divided.

Numerical estimate. The relative quantity of intramuscular fat was estimated by a numerical scoring system described by Ramsbottom *et al.* (1945). Average values for the estimated quantity of fat in raw and cooked LD tissue from rams, wethers, and ewes are given in Table 1. In general, the quantity of fat was not affected by sex or year. In raw LD muscle the average score (1964 and 1965) for quantity of fat was 4.0, 4.4, and 4.2 for rams, wethers, and ewes, respectively. There was no significant difference between rams (3.04%) and wethers (3.70%) for ether extract from the LD of the same animals used for the histological study, but a significantly ($P = 0.05$) higher value (4.53%) for ether extract was found for the LD from ewes (Hinnergardt, 1966). However, the marbling scores for rib-eye (Hinnergardt, 1966) differed significantly ($P = 0.05$) between rams and wethers, rams and ewes, and between wethers and ewes. Average marbling scores (1 to 11 range) were: rams, 4.50; wethers, 5.48; and ewes, 5.90.

Average values for the quantity of fat in the LD from 1964 and 1965 lambs, except for wethers, increased slightly with cooking. The average value for raw muscle from wethers was 4.4

Explanation of Fig. 1. Photomicrographs illustrating fat distribution and muscle fiber characteristics of lamb muscle. Procedure for photomicrography is in the Appendix, p. 44.

a - Section of cooked muscle from a wether in which the fat was distributed evenly throughout the section (150X).

b - Section of raw muscle from a ram in which the fat occurred in clustered form (150x). The dark spots are precipitated stain (Sudan IV).

c - Section of raw muscle from a ram in which the accordian plaited effect of the fibers occurred (150X).

d - Section of raw muscle from a ewe in which a twist and a kink (bottom middle and bottom right of the photomicrograph) occurred (200x).

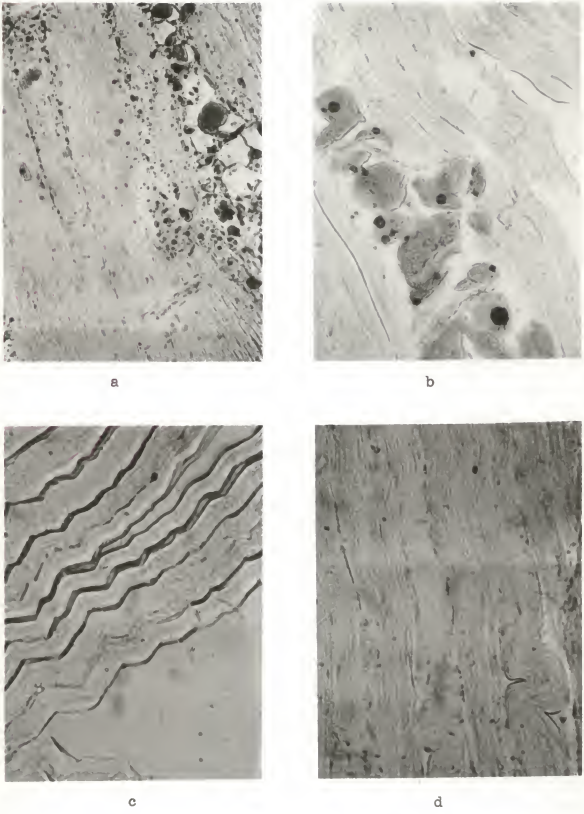

Fig. 1. Photomicrographs illustrating fat distribution and muscle fiber characteristics of lamb muscle.

Table 2. Average values[a] for descriptive characteristics of raw and cooked LD muscle from lambs raised in 1964 and 1965.

Characteristics	Rams				Wethers				Ewes			
	1964		1965		1964		1965		1964		1965	
	Raw	Cooked	Raw	Cooked	Raw	Cooked	Raw	Cooked	Raw	Cooked	Raw	Cooked
Fat												
Distribution												
Clumped together	0.7	0.9	0.7	0.8	0.9	0.8	0.8	0.5	0.9	0.8	0.8	0.6
Scattered or bead like	0.6	0.8	0.8	0.6	0.8	0.8	0.9	0.8	0.8	0.8	0.9	0.8
Connective tissue												
Type												
Wavy fiber	0.5	0.1	0.5	0.0	0.6	0.1	0.4	0.1	0.5	0.1	0.4	0.1
Straight fiber	0.2	0.7	0.2	0.5	0.4	0.6	0.4	0.2	0.2	0.6	0.2	0.3
Granulated	0.8	1.0	0.9	1.0	0.7	1.0	0.9	0.9	0.9	1.0	0.9	1.0
Muscle fiber												
Type												
Straight	1.0	1.0	1.0	0.9	0.9	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Kinked or twisted	0.9	0.8	0.9	0.8	0.9	0.4	0.9	0.9	1.0	0.7	0.9	0.8
Accordian plaited	0.5	0.3	0.5	0.1	0.4	0.3	0.4	0.4	0.5	0.3	0.2	0.2
Granulated	0.1	0.3	0.1	0.0	0.1	0.3	0.1	0.1	0.2	0.3	0.1	0.1

[a]Determined by averaging the number of times the particular characteristic was checked by the observers.

and for cooked muscle, 4.3; whereas, for rams it was 4.0 and 4.7; and for ewes 4.2 and 4.6 for raw and cooked muscles, respectively.

Connective Tissue

In the prepared sections of LD muscle, connective tissue appeared dark blue to gray. Harris' hemotoxylin was used to stain the muscle fibers, and according to Lowe (1948), when using this stain, connective tissue, depending on the thickness of the section and the magnification used for study, is sometimes discernible, but frequently it is not noticeable.

Descriptive characteristics. Average values for descriptive characteristics, obtained by subjective evaluation, are given in Table 2. In general, the degree of waviness or straightness of the connective tissue fibers was not affected by sex or year.

There was some difference between raw and cooked muscle in the waviness and granulation of connective tissue fibers. Raw muscle had more wavy connective tissue fibers than cooked muscle, and connective tissue in cooked muscle appeared granulated oftener than that in the raw (Table 2). Winegarden et al. (1951) reported that fibers in heated collagenous connective tissue were straighter, less distinct than fibers from raw tissue, and appeared to be fused or merged in some areas.

Numerical estimate. A numerical estimate of the quantity of the connective tissue in sections of lamb LD was made by the same scoring system as that used for fat. Quantity of connective tissue was not affected by sex or year (Table 1). In raw LD muscle from 1964 and 1965 lambs, average scores for the quantity of connective tissue were 4.6, 5.0, and 4.7 for rams, wethers, and ewes, respectively.

For all 3 sex groups, the average scores for quantity of connective tissue in raw muscle, except sections from 1965 wethers and ewes were slightly lower than those for cooked muscle (Table 1). Skelton et al. (1963) found that the quantity of collagenous connective tissue in beef appeared to be greater in cooked than in raw samples. For this, they gave 2 explanations. One explanation was that with heating the collagenous tissue swells, becomes granular, and fills the spaces between the muscle fibers. Thus, the swollen collagenous tissue gives the appearance of more collagenous in cooked than in raw muscle. The other explanation was that with cooking there is a redistribution of collagenous tissue, which appears as long fiberous strands in sections of raw muscle, and as masses of granular tissue dispersed throughout the section of cooked tissue.

Muscle Fibers

Descriptive characteristics. Data for descriptive characteristics of muscle fibers from 1964 and 1965 lambs are given in Table 2. Most of the terms that will be used to describe the histological appearance of the LD muscle fibers were defined by Lowe (1948). The term kink refers to a suddenly rounded curve, and a multiple kink is referred to as a twist. Accordian plaited is used to describe sharp, angular bends in the fiber that give an accordian plaited effect. The term granulated is used for the disappearance of the cross striae of the muscle fibers, which were replaced by a granulated substance.

In general, the microscopic appearance of muscle fibers was not affected by sex, year, or cooking. Some kinks and twists occurred, but not to a large extent (Table 2, Fig. 1d). However, when they did occur, a larger number were found in the raw tissue than in the corresponding cooked sample. Lowe (1948) reported that kinks and twists in the fibers of poultry disappeared with cooking. Accordian plaited muscle fibers were found more in raw than in cooked muscle (Table 2, Fig. 1c).

Both cross and longitudinal striae were visible in fibers of all sexes. However, the cross striations were more distinct in the cooked meat. Lowe (1948) made a similar observation in a study of chicken muscle. She explained this may be attributed to partial dehydration of the fibers during the cooking process.

Granulated fibers were observed in only a few sections. However, when noted, there seemed to be a tendency for more granulation in cooked than in raw muscle (Table 2).

<u>Width of fibers</u>. The relative width of muscle fibers was obtained by counting the number of fibers in a completely filled microscopic field (860x magnification). Also, the width was measured with a calibrated ocular micrometer.

The number of fibers in a microscopic field was counted only for raw tissue. Average values indicated no difference in the size of fibers attributable to sex, but the range in the number of fibers per field averaged 1 more fiber in muscle from 1965 lambs than in that from 1964 lambs (Table 1).

When muscle fiber diameter was measured with a calibrated ocular micrometer, there was little difference in diameter of fibers from 1964 and 1965 animals when raw muscle was compared with raw muscle and cooked muscle with cooked muscle (Table 1). Differences between raw and cooked tissue were greatest for rams in 1964 and for ewes in 1965, whereas differences between raw and cooked tissue were smallest for ewes in 1964 and for wethers in 1965. However, differences between raw and cooked muscle were greater for all 3 sexes in 1965 than in 1964.

The diameter of fibers varied among the 3 sex groups. Fibers (raw and cooked) of rams (40.12 μ) were slightly larger than those of wethers (39.4 μ), and those of the ewes (39.9 μ) were intermediate. Hammond (1932) reported that raw fibers of rams (39.7 μ) were slightly larger than those of ewes (37.9 μ) and those of wethers (38.4 μ) were intermediate in size.

Cooking the muscle caused a decrease in the fiber diameter. The average diameter for raw muscle from ewes 1964 and 1965 animals was 40.5 μ, whereas for cooked muscle it was 38.4 μ. The average diameter of raw muscle fibers from wethers was 41.3 μ, and for cooked, it was 38.6 μ, whereas for raw and cooked muscle from rams it was 41.5 and 38.8 μ, respectively. Satorius and Child (1938) found with beef ST that the diameter of the muscle fibers shrunk (64.57 to 52.39 μ) with increase in temperature from unheated tissue to 67°C, then there was no change between 67 and 75°C. Cooked tissue used for the study reported here was from muscle heated to an internal temperature of approximately 75°C.

The Relationship of Histological Characteristics to Palatability Factors

Data for histological study, palatability factors, and total cooking losses of loin roasts from 1964 and 1965 animals were averaged and are presented in Tables 3 and 4.

Tenderness. There were significant differences in tenderness, as measured by panel scores and Warner-Bratzler shear values, attributable to sex. Panel scores indicated significant ($P = 0.05$) differences between rams and wethers, rams and ewes, and between wethers and ewes; whereas shear values measured significant ($P = 0.05$) differences only between rams and ewes and rams and wethers. Fiber diameter was inversely related to tenderness scores and directly related to shear values (Table 3). Tuma *et al*. (1962) and Carpenter *et al*. (1963) reported similar results for beef and pork, respectively.

Muscle from rams contained more connective tissue and received lower tenderness scores and higher shear values than that from wethers or ewes (Table 3). The relationship between quantity of fat and tenderness was not consistent for the 3 sex groups. The histological estimate for fat in the LD from rams was slightly greater than that for wethers or for ewes, but

Table 3. Average values for histological estimate of quantity of fat and connective tissue, width of muscle fibers, and subjective and objective measures of tenderness for cooked lamb LD muscle (1964 and 1965).

Sex	Histological Estimates			Tenderness Measurement[b]		
				Subjective[c]		Objective
	Fat[a]	Connective tissue[a]	Muscle fiber diameter, µ	Initial score	Chew score	Warner-Bratzler shear, lb/1/2 in. core
Rams	4.7	4.9	38.8	5.4	5.2	8.8
				*	*	*
Wethers	4.3	4.4	38.6	* 5.6	* 5.5	* 7.8
				*	*	
Ewes	4.6	4.7	38.4	5.8	5.7	7.2

[a]Scoring key-quantity 7-1 (7, large and 1, none).

[b]Unpublished data, Department of Foods and Nutrition.

[c]Scoring range 7-1 (7, very desirable and 1, undesirable).

*, Significant at the 5% level.

Table 4. Average values for total cooking loss and panel scores for juiciness and intensity and desirability of flavor (1964 and 1965)[a].

Sex	Total cooking loss (%)[b]	Juiciness[b]	Intensity of flavor[b]	Desirability of flavor[b]
Rams	11.7	6.0	4.7	5.5
	ns	ns	ns	ns
Wethers	12.3	6.0	4.5	5.6
	ns	ns	ns	ns
Ewes	12.0	6.0	4.6	5.6

[a]Unpublished data, Department of Foods and Nutrition.

[b]Scoring range, 7-1.

ns, Not significant.

muscle from rams was significantly (P = 0.05) less tender (panel scores and shear values) than muscle from either wethers or ewes (Table 3). Batcher et al. (1962) found that the quantity of intramuscular fat did not affect the tenderness of lambs. In the present study, fat in the LD from rams tended to occur more in clustered than in scattered form, whereas in wethers and ewes it was distributed throughout the section. This observation supports the view of Wang et al. (1954) that it is not the total fat in a muscle, but the way it is distributed within the muscle that affects tenderness.

Juiciness and flavor. There were no significant differences among the 3 sex groups for juiciness, intensity and desirability of flavor, and total cooking losses. There appeared to be no relationship between the estimated quantity of fat and juiciness or flavor scores. Wethers contained slightly less fat, but did not differ in juiciness or flavor from rams or ewes. Also, total cooking losses did not affect juiciness (Table 3 and 4). Batcher et al. (1962) reported that juiciness and flavor in lamb meat were independent of the quantity of intramuscular fat.

SUMMARY

This study was conducted to estimate and compare the relative quantity of intramuscular fat and connective tissue, and the width of muscle fibers in raw and cooked LD muscle from ewe, ram, and wether lambs raised in 1964 and 1965; and to study the relationship of the histological data for these factors to data for palatability factors. Also, the microscopic appearance of muscle fibers and connective tissue was observed.

In general, size of the fat globules, estimated quantity of fat or connective tissue, waviness or straightness of connective tissue fibers, and the characteristic appearance of muscle fibers such as degree of straightness, kinkiness, and granulation were not affected by sex or year. However, distribution of fat varied some among the 3 sex groups. Fat tended to occur in muscle from rams in clustered form, whereas in ewes and wethers it tended to be more evenly distributed throughout the muscle. Also, fiber diameter varied among the sex groups. The diameter of fibers in ram muscle was slightly larger than that in wether muscle, whereas fiber diameter of ewe muscle was intermediate.

Cooking had an effect on the microscopic appearance of connective tissue and muscle fibers. Waviness of connective tissue fibers and muscle fiber diameter decreased with cooking. Average fiber diameters for ewes were 40.5 and 38.4; for wethers, 41.3 and 38.6; and for rams 41.5 and 38.8 µ for raw and cooked muscles, respectively.

Fiber diameter was inversely related to panel tenderness scores and directly related to Warner-Bratzler shear values. Muscle from rams contained more connective tissue and received lower tenderness scores and higher shear values than that from ewes or wethers. The estimated quantity of fat did not affect tenderness, juiciness, or flavor scores.

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APPENDIX

Table 5. Samples (animal Code No.) from the longissimus dorsi muscles of lamb used for histological study.

Year and grade	Sex						Number of samples	
	Ewe		Ram		Wether			
1964	Raw	Cooked	Raw	Cooked	Raw	Cooked	Raw	Cooked
Prime	626	625			653	653		
	625	626						
	627	627						
	658	658						
							5	5
Prime$^-$	615	615	503	500	501	501		
	616	616	610	503	502	502		
	621	621		610	618	618		
	629	629			623	623		
							10	11
Choice$^+$	609	609	505	504	608	608		
	624	622	504	611				
			611	614				
			612	624				
			614					
							8	7
Choice	619	619	607	607	678	678		
	622	680	613	612				
	680		617	613				
			631	617				
			632	631				
			646	632				
				646				
							10	10
Choice$^-$			660	660	669	669		
			671	671				
			675	675				
			677	677				
			681	681				
							6	6
Number of samples:	13	12	18	19	8	8	39	39

Table 6. Samples (animal Code No.) from the longissimus dorsi muscle of lamb used for histological study.

Year and grade	Sex: Ewe		Ram		Wether		Number of samples	
	Raw	Cooked	Raw	Cooked	Raw	Cooked	Raw	Cooked
1965								
Prime	736	736			721	721		
					747	747		
							3	3
$^{-}$Prime	707	707	700	700	704	704		
	708	708			722	720		
	712	712						
	713	713						
	714	714						
	717	717						
	720	720						
							10	10
$^{+}$Choice	703	703	701	701	711	711		
	716	716	702	702	715	715		
			705	705	724	724		
			709	709	741	741		
			710	710				
			718	718				
			719	719				
			723	723				
							14	14
Choice	725	706	733	706	743	743		
	727	725	745	733				
	735	727	748	745				
	759	735	749	748				
		759		749				
							9	11
Number of samples:	14	15	13	14	9	9	36	38

Histological Study Check Sheet

Form 1

Year ____________

Name ____________
Sample No. ____________
Kind (raw or cooked)

Factors	Slide No. 1	2	3	4	5	Total	Average
FAT							
Quantity[a]							
Distribution -							
Clumped together							
Scattered or beadlike							
Globules[b]							
CONNECTIVE TISSUE							
Quantity[a]							
Type -							
Wavy fiber							
Straight fiber							
Granulated							
MUSCLE FIBERS							
Width -							
Number							
Diameter in mm. a							
b							
c							
Type - check when observed in a section							
Straight							
Kinked or twisted							
Accordian plaited							
Granulated							

Scoring keys

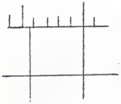
[a]		[b]	
Large	- 7	Large	- 7
Medium	- 5	Medium	- 5
Small	- 3	Small	- 3
None	- 1		

Procedure for Measuring Muscle Fiber Diameter (Width)

Calibration of Ocular Micrometer

Insert stage micrometer (the slide) on the stage of microscope under high power (43X objective, 10X eyepiece). Set Dynazoom knob in microscope at position 2 to give a magnification of 860X. Match a line of the scale on the stage micrometer with a line on the squared scale of the ocular (eyepiece) micrometer.

Example:

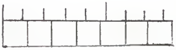


Stage scale

Squared ocular scale

Count the number of stage units and number of ocular units, where the stage line coincides with the ocular line.

Example:



6 stage units
5 ocular units

Give the distance covered by the stage units, its numerical value (each stage unit = 0.01 mm, see slide). Divide this value by the number of ocular units, thus finding the value of each ocular unit.

Example:

6 stage units = 6x 0.01 mm or 0.06 mm.

$\frac{0.06}{5}$ = 0.012 mm/ocular unit or 1 ocular unit = 0.012 mm.

Measurement of Fiber Diameter

Remove the stage micrometer and replace it with the slide to be studied. Measurement of the muscle fibers is made by using the value assigned to the ocular units. This value is good only as long as high power is used. Convert mm value to μ.

Note: Through the center of the eyepiece, the ocular units are further divided into 5 parts. These may be used in measurements for greater accuracy.

The eyepiece can be turned in the tube, thus turning the ocular scale. In this way, fibers can be measured even though they do not lie in a perfectly vertical or horizontal direction. For each section, select at random 3 fibers, and measure.

Photomicrography

A Polaroid Land Camera (model 80B, attachable to the microscope) was used for taking photomicrographs. First, the camera was loaded with Type 32 film that had an Exposure Index of 400 daylight. For loading, the directions given in the--"How to make good pictures with your Polaroid Highlander Land Camera"--instruction book were followed. For attaching the camera to the Dynazoom microscope, a straight vertical tube was screwed into the microscope, and the camera set into the sleeve, adjusted to the preferable position, and locked in place with the lock screws provided. Next, the slide was placed on the platform of the microscope, and the field to be photographed was located. Then, the prism control knob was turned to "out" position. The zoom knob on the microscope was set at either 1.5X or 2X, and the 10X objective was used to give a magnification of 150X or 200X. The Base Illuminator dial was set at 4, and the exposure time in the camera at 1/10 of a second. The picture was taken, and for developing it, the directions given in the Polaroid instruction book were followed. The developing time was 10 sec.

COMPARISON OF HISTOLOGICAL CHARACTERISTICS WITH PALATABILITY FACTORS OF THE LONGISSIMUS DORSI MUSCLE FROM EWE, RAM, AND WETHER MARKET LAMBS

by

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There are few reports in the literature giving information on the palatability of cooked lamb relative to sex of the animal, or that give information regarding histological characteristics of lamb muscle as related to palatability. Therefore, this study was conducted to estimate and compare the relative quantity of intramuscular fat and connective tissue, and the width of muscle fibers in raw and cooked LD muscle from ewe, ram, and wether lambs raised in 1964 and 1965; and to study the relationship of the histological data for these factors to data for palatability factors. Also, the microscopic appearance of muscle fibers and connective tissue was observed.

Small blocks (152) of raw and cooked LD muscle from 3 sex groups that were fixed in physiological salt solution and formalin were prepared for microscopic study. Longitudinal sections were cut 15-20 µ thick on a clinical microtome, and stained with Harris' hematoxylin and Sudan IV. The sections were washed in tap water and mounted on slides in glycerine jelly. Sudan IV imparted a bright red-orange to fat, whereas Harris' hematoxylin stained the muscle fiber a bluish purple. The connective tissue was darker bluish purple to gray.

From histological study, it was found that in general, size of the fat globules, estimated quantity of fat or connective tissue; waviness or straightness of connective tissue fibers; characteristic appearance of muscle fibers such as degree of straightness, kinkness, and granulation were not affected by sex or year. However, distribution of fat varied among the 3 sex groups. Fat tended to occur in muscle from rams in clustered form, whereas in ewes and wethers it tended to be more evenly distributed throughout the muscle. Also, the fiber diameter varied among the sex groups. The diameter of fibers in ram muscle was slightly larger than that in wether muscle, whereas fiber diameter of ewe muscle was intermediate.

Cooking had an effect on the microscopic appearance of connective tissue and muscle fibers. Waviness of connective tissue fibers and muscle fiber diameter decreased with cooking. Average fiber diameters for ewes, were 40.5 and 38.4; for wethers, 41.3 and 38.6; and for rams 41.5 and 38.8 µ for raw and cooked muscles, respectively.

Fiber diameter was inversely related to tenderness scores (panel) and directly related to Warner-Bratzler shear values. Muscle from rams contained more connective tissue and received lower tenderness scores and higher shear values than that from ewes or wethers. The estimated quantity of fat did not affect tenderness, juiciness, or flavor scores.