To the Graduate Council:

I am submitting herewith a thesis written by Joseph Edwin Marcy entitled "Certain Chemical, Physical, and Histochemical Characteristics of Three Quality Levels of Porcine Longissimus." I recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science with a major in Food Technology and Science.

Curtis C. Melton, Major Professor

We have read this thesis and recommend its acceptance:

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CERTAIN CHEMICAL, PHYSICAL, AND HISTOCHEMICAL CHARACTERISTICS OF THREE QUALITY LEVELS OF PORCINE LONGISSIMUS

A Thesis

Presented for the

Master of Science

Degree

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ABSTRACT

This study was conducted in two phases. In phase I, muscle pH, muscle color (subjective and objective), percentage of fiber types, number of giant fibers, and muscle fiber waviness were studied in three quality levels of porcine longissimus. Thirty-six loins were purchased at a local packing plant after a 20 hour postmortem chill, and placed into quality levels according to the muscle pH. Loins were classified as pale, soft and exudative (PSE) if the pH was below 5.30, normal if the pH was between 5.30 and 5.70, and dark, firm and dry (DFD) if the pH was above 5.70.

Of the 36 loins tested, 14 did not demonstrate typical fiber distribution or show reciprocal enzymatic activity between the oxidative and phosphatase enzyme stains. Subjective color score differed ($P < .01$) between the enzymatically normal and abnormal groups. The PSE group was lighter ($P < .01$) than the DFD group in subjective color. The only variance in measured objective color was due to the difference in lightness between quality levels. There was no visual difference in the chromaticity among quality level. Therefore, the differences perceived in the subjective color determination were the result of variation in lightness. The percentage of muscle fibers types was not affected by quality levels, but a greater number ($P < .05$) of giant fibers was found in the DFD group. The loins with abnormal enzymatic activity had a higher ($P < .01$) percentage of fibers that did not show reciprocal enzymatic activity. Waviness of muscle fibers

was found to be nonsignificant among quality levels.

In phase II, the longissimus of nine pork carcasses was sampled at 45 minutes, 90 minutes, six hours and 20 hours postmortem. Muscle pH and temperature, percentage of fiber types, and number of giant fibers were recorded for each time period. Muscle color was measured objectively and subjectively at 20 hours postmortem.

Five of the nine carcasses had abnormal enzymatic activity at each sampling time postmortem, three were normal at each sampling time, and one showed abnormal enzymatic activity only at 20 hours postmortem.

The percentage of fiber types did not significantly change with time postmortem, but was different ($P < .01$) among carcasses sampled.

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CHAPTER I

INTRODUCTION

Pale, soft, and exudative (PSE) pork is of importance to the pork industry because of the adverse economic effect that it has. At the 1972 Pork Quality Symposium it was reported that 18% of all pork carcasses are PSE. It was also estimated that 3-5% of all hogs die on the way to slaughter because of Pork Stress Syndrome (PSS), a disorder related to PSE. PSE carcasses will lose 2-10% more weight before processing than normal carcasses. An Iowa study reported 35% of all herds were affected by PSE. A nationwide survey showed 36% of United States hog producers have encountered PSE and their herds account for 44% of total market hogs.

PSE muscle has a lower water binding capacity which causes increased shrinkage in processed products. PSE muscle also cures differently, thus decreasing product uniformity. Although the data is conflicting, some researchers believe PSE muscle is less tender and it is generally agreed that tenderness of PSE muscle is decreased more than normal muscle by incorrect cooking or overcooking. Some also blame the generally lower consumer acceptance of pork to the variation in quality caused by PSE muscle.

Because of these adverse economic effects of PSE muscle, researchers have been trying to identify physical, chemical, and histochemical characteristics associated with PSE pork that could be used to select herd sires which were free from this condition. It has been noted that

a hog that tends to become PSE will react poorly to stress. When stressed the animal begins to twitch and the body temperature is elevated. Research has shown the animals have abnormal levels of adrenal hormones in the blood after stress. When the animal is slaughtered it will tend to go into rigor-mortis more rapidly. This is associated with low pH values (below 5.3) and increased lactic acid in the postmortem tissue.

Histologically the muscle is characterized by atrophied fibers and possible changes in the kinds of fibers present. Parks (1974) studied the histological traits of porcine longissimus and found six of 26 animals reacted abnormally for adenosine triphospatase (ATPase) stain and had heavier muscling that is associated with PSE pork.

The purpose of this research was to further elucidate the histochemical abnormality observed by Parks (1974), and to study the relationships of certain physical and chemical properties of porcine longissimus to the observed ATPase abnormality.

CHAPTER II

REVIEW OF LITERATURE

Relationship of pH and Temperature on PSE Development

Most researchers agree that below normal muscle pH and above normal muscle temperature postmortem are associated with PSE muscle in pork, but their interaction is still not completely understood (Wismer-Pedersen and Briskey, 1964; Sayre et al., 1964; Briskey, 1964).

Briskey and Wismer-Pedersen (1961a) report pH of PSE muscle attains its lowest point of 5.1 at 1-1/2 hours postmortem and then may rise to a pH of 5.3 to 5.6, while normal muscle shows a steady decline in pH for approximately eight hours with an ultimate pH of 5.3 to 5.7. Eikelemboom et al. (1973) found the 45-minute pH to be more highly correlated ($r = 0.69$) than ultimate pH ($r = 0.51$) with percent light transmission of the muscle fluid. A larger transmission value reflects a lower muscle quality. Martin et al. (1974) concluded that ultimate pH is a better indicator of pork quality than pH at one hour postmortem. Wismer-Pedersen and Briskey (1961b) reported a pattern of continuous pH depressions and elevations during rigor development. Bendall et al. (1962) reported holding a carcass at a temperature of 37° C during rigor mortis produced generally pale and watery pork. Bendall et al. (1965) concluded that muscle pH and deep muscle temperature were unrelated. Bodwell et al. (1966) showed that all

pork carcasses allowed to go through rigor at 37° C did not become PSE and concluded that pH and temperature alone could not explain this phenomenon. Borchert and Briskey (1965) were able to reduce the development of PSE in pork by quickly cooling a carcass in liquid nitrogen. Martin et al. (1975) found ultimate meat quality could be predicted with only moderate (R^2 = 26%) success with the knowledge of muscle pH and temperature up to two hours postmortem.

Color of PSE Pork

Muscle is most commonly classed as PSE on the basis of tissue color. The color of extreme PSE muscle has been described (Briskey, 1964) as gray, the same color as boiled meat, and very moist on the surface. As the muscle becomes more normal the typical pink color returns and the surface moisture decreases. There are various degrees of PSE between normal and very pale.

Several subjective scales for measurement of muscle color have been developed and widely used (Forrest et al., 1963; Sayre et al., 1963; Briskey, 1959). Other researchers have used objective measures such as reflectance (Kastenschmidt et al., 1965; Wismer-Pedersen and Briskey, 1961a) and transmission (Hart, 1962).

Histological Characteristics of PSE Muscle

In an effort to explain the occurrence of PSE researchers have studied the histological differences of normal and PSE pork. Several differences have been noted in the appearance of the longitudinal section of the muscle fiber. Cassens (1966) and Lawrie (1960) suggested

that twisting or kinking of the muscle fiber may be due to the violent rigor mortis of PSE muscle. Bendal (1960) concluded that the waviest fibers were actually forced that way because of actively contracting adjacent fibers. In a histological study of 200 porcine longissimus muscles Briskey (1964) reported the wavy condition described by Lawrie (1960) was found but they were unable to relate it consistently with pale muscle color or low muscle pH. Cassens (1963) was also unable to relate muscle fiber damage and PSE muscle.

Histochemical Differences of PSE Muscle

In the histochemical study of mammalian muscle it was recognized early that there was not a single fiber type, but the true number of fiber types is still uncertain. Dubowitz and Pearse (1960) found that muscle could be classified into two categories based on their reciprocal relationship of phosphorylase activity and activity of certain dehydrogenases. The smaller fibers were called red fibers and were found to be low in phosphorylase activity, high in oxidative enzyme activity, and contained large amounts of myoglobin. The larger fibers were called white fibers because they contained little myoglobin. White fibers were high in phosphorylase activity and low in oxidative enzyme activity. Dubowitz and Pearse (1960) also recognized that there could be intermediate activity for any given enzymatic reaction. Other authors have classified fibers into two types (Brody and Engel, 1964; Brooke and Engel, 1969; Brooke and Kaiser, 1970; Dubowitz, 1970; Engle and Brooke, 1967). Using the two fiber type system for classification, possible intermediate fibers were usually classified as red fibers.

Other authors suggest that there are three fiber types. Guth et al. (1970) classified red, white, and intermediate fibers as beta, alpha, and alpha-beta, respectively. Ashmore and Doerr (1971) proposed a classification of beta red, alpha white and alpha red fibers to correspond to red, white and intermediate fiber types. As many as eight possible fiber types were found by Romanul (1964) in rat muscle.

Fibers that have been classified as intermediate in porcine longissimus have been described in different ways. Anderson and Parrish (1972) described the intermediate fiber found in porcine longissimus as intermediate in color intensity for alkaline adenosine triphosphatase (ATPase), intermediate in color intensity for phosphorylase, and described the nicotinamide adenine dinucleotide tetrazolium-reductase (NADH - TR) reaction as formazan granules dense in subsarcolemmal regions, but absent or in small numbers in the center of the fiber. Swatland and Cassens (1973) also classified intermediate fibers as fibers of intermediate color reaction for alkaline ATPase and NADH - TR stains. Morita et al. (1969) classified intermediate fibers as intermediate color reaction for Benzidine and NADH - TR stains. Moody and Cassens (1968) found that not all fibers demonstrated the reciprocal enzymatic relationship between phosphorylase and oxidative enzymes described by Dubowitz and Pearse (1960). When serial sections of porcine longissimus stained for NADH - TR and amylophosphorylase were compared, the NADH - TR reaction showed more red fibers than the amylophosphorylase. The fibers that did not show the reciprocal relationship were classified as intermediate fibers. Cooper et al. (1969)

also classified intermediate fibers as fibers that did not show reciprocal enzymatic activity.

The percent of fiber types in PSE longissimus has been studied and found to vary from normal, but because of differences in nomenclature of fiber types the results are confusing. Dildey et al. (1970) reported an increase in the number of red fibers in PSE longissimus. Cooper et al. (1969) and De Bruin (1971) suggested a higher red fiber content in PSE muscle because of more intermediate fibers. Merkel (1971) found an increase in white fiber content. Cooper et al. (1969) also reported finding giant fibers as described by Cassens et al. (1969) in PSE porcine longissimus. The giant fiber is usually larger than the surrounding fibers and usually compresses the surrounding fibers in an effort to conform to a round shape. The giant fiber is NADH - TR variable and alkaline ATPase positive.

Cooper et al. (1969) reported that 65-80% of the fibers that stained positive for NADH - TR also were positive for alkaline ATPase and amylophosphorylase. They concluded that these fibers could be responsible for PSE muscle because unlike true red fibers they have high ATPase and phosphorylase activity, enabling them to break down adenosine triphosphate (ATP) and accelerating glycolysis and thus triggering an accelerated glycolytic rate in the entire muscle. Swatland and Cassens (1973) also found that some PSE longissimus muscles deviated from the normal distribution pattern for red and white fibers. They found that in some fasiculi the centrally located fibers were alkaline ATPase positive while in others the fibers had

no special location. The pattern of normal tissue (Moody and Cassens, 1968) has a very consistent distribution of discrete clumps of five to seven red fibers towards the center of a fiber bundle surrounded by white fibers. Parks (1974) also found that PSE longissiums deviated from the normal fiber distribution pattern. He stained sections for alkaline ATPase (Padykula and Herman, 1955) and found a pattern of fibers that resembled acid stabile ATPase or showed no particular pattern. The percentage of fibers that stained positive for alkaline ATPase compared well with the percentage of red (negative staining) fibers in normal porcine longissimus.

CHAPTER III

MATERIALS AND METHODS

To fulfill the objectives of this study the data were collected in two phases.

Phase I.

Thirty-six pork loins were purchased from a local packing plant and transported to the University of Tennessee meats laboratory. All loins were removed from the chilled carcass at 20 to 24 hours post-mortem. The longissimus muscle was cut the tenth and eleventh ribs, allowed 30 minutes to bloom or oxygenate, then scored for subjective color. An objective color measurement was made 45 minutes after the longissimus muscle was cut. A one inch chop was removed from the eleventh rib location for pH and histological analysis. The histological sample was removed from the dorsal side of the longissimus muscle, one half the distance from the vertebrae to the lateral end of the muscle. This location would approximate the desirable surgical location on a live animal.

The histological tissue was cut into pieces one centimeter square with a clean, new razor blade. The pieces were then dipped in talcum powder and submerged in liquid nitrogen for quick freezing, then placed into an aluminum foil container and immediately immersed in a liquid nitrogen storage tank until sectioned for analysis. Data collected from the histological sample included: enzymatic activity, percent fiber type, number of giant fibers, and waviness of muscle fibers.

Methods for Data Collection

Subjective muscle color. Color of the longissimus muscle was measured subjectively 30 minutes after the muscle was cut using the Iowa State University scale (prepared by R. E. Rust and D. G. Topel, 1969). This is a scale of one to five, with one corresponding to very PSE, three is normal, and five is dark, firm, and dry (DFD). Muscle color was scored by two trained judges with a white cutting table as background. The room was illuminated by General Electric 40 watt cool white fluorescent lamps and was supplemented with northwest daylight.

Objective muscle color. The objective color was measured 45 minutes after the longissimus muscle had been cut, using a Hunterlab color/difference meter model D25D2. The color values were measured in International Commission on Illumination (C. I. E.) units. The chromaticity coordinates x and y were calculated as:

$$x = \frac{X + Y + Z}{x} \; ; \quad y = \frac{X + Y + Z}{Y} \; .$$

The reflectance of the muscle was calculated as: $L = 10\sqrt{Y}$.

Muscle pH. A ten gram sample of muscle was removed from the longissimus adjacent to the histological sample. The sample was blended with 50 milliliters of distilled water in an Oster blender model 847-05A at the lowest speed for one minute. The slurry was centrifuged for two minutes at 1,000 g. The pH of the supernatant was measured with an Orion Research Ionalyzer model 801 pH meter standardized to a

pH of 7.0 using commercially prepared buffer solution. On the basis of pH the loins were placed in categories as follows:

pH below 5.30 = PSE,

pH between 5.30 and 5.70 = normal,

pH over 5.70 = DFD.

Muscle fiber enzyme activity. The enzyme activity was measured as either normal or abnormal according to its reaction to histological staining. Following equilibration to -20° C. serial cross sections of the muscle were sectioned at a thickness of ten microns on a Lipshaw model 1800 cryostat. The sections were picked up on warm coverslips and allowed to dry for 30 minutes. These serial sections were then stained for NADH - TR and ATPase activity using the method of Engel and Brooke (1967), Appendix A. The same sections were used to determine percent fiber type and number of giant fibers. The tissue was judged normal if serial sections exhibited a reciprocal enzymatic relationship between the NADH - TR and ATPase reaction as described by Dubowitz and Pearse (1960). The tissue was judged abnormal if it did not show the reciprocal enzymatic relationship and if the typical distribution pattern of muscle fibers (Moody and Cassens, 1968) was not evident.

Percent muscle fiber type. Serial cross sections (ten microns thick) were cut on a Lipshaw model 1800 cryostat, placed on glass coverslips and allowed to dry 30 minutes. Serial sections were then incubated in NADH - TR and ATPase solutions as described by Engel

and Brooke (1967), Appendix A. The sections were mounted in glycerogel and photographed using a Wild M20 microscope with automatic camera attachment. Total magnification was 24 diameters (x). Negatives of the serial sections stained for ATPase and NADH - TR were framed and projected onto a 1200 square centimeter sheet of paper. Total magnification was 360x. To randomize the fibers that were counted, the paper was always placed in the same position and only the fibers that were projected onto the paper were counted. Fibers were classified as follows:

Type 1 - - - Positive for alkaline ATPase
Negative for NADH - TR

Type 2 - - - Negative for alkaline ATPase
Positive for NADH - TR

Type 3 - - - Positve for alkaline ATPase
Positive for NADH - TR

Type 4 - - - Negative for alkaline ATPase
Negative for NADH - TR

The number of giant fibers reported represented the total number of giant fibers that was projected on the 1200 square centimeter paper.

Muscle fiber waviness. Longitudinal sections (ten microns thick) were made on a cryostat. They were adhered to coverslips and allowed to dry 30 minutes. The sections were then stained for NADH - TR as previously described. The negatives were framed and projected onto a screen. A panel of four trained judges subjectively scored waviness

of the muscle fibers using the following scale.

1 = straight,

2 = slightly wavy,

3 = wavy,

4 = very wavy.

The judges were allowed to use any value between one and four that they considered appropriate, including fractions.

Phase II

In this phase the experimental data were collected from nine pork carcasses purchased from a local packing plant and chilled at 4° C. The longissimus muscle of the carcasses was sampled at four time periods postmortem.

The first time period was 45 minutes postmortem. At this time the core temperature of the longissimus was measured at the ninth rib location. The temperature determination of the longissimus was taken with a YSI model 42SC pyrometer. A stainless steel thermocouple was inserted approximately 1-1/2 inches into the approximate center of the longissimus muscle. A sample of the longissimus muscle was excised from the tenth rib location and immediately cut into one square centimeter pieces, dipped into talcum powder and frozen in liquid nitrogen. The frozen tissue was stored in liquid nitrogen until histological analysis was done. An adjacent portion of longissimus was also immediately placed in liquid nitrogen for pH determination. The histological sample was analyzed for enzyme

activity percent fiber, and number of giant fibers. These same data were also collected at 90 minutes and six hours postmortem. The sample of longissimus muscle for the 90 minute postmortem observation was removed from the eleventh rib location and the six hour sample was removed from the ninth rib location on the same longissimus muscle from which the first two samples were taken.

The 20 hour sample was removed from the opposite longissimus muscle of the carcass. The temperature of the longissimus muscle was measured at the ninth rib location. The loin was cut at the tenth to eleventh rib and a one inch chop removed from the eleventh rib location. The chop was exposed to the atmosphere 30 minutes before a subjective color measurement was made. At 45 minutes after cutting the longissimus objective color was measured. A histological sample was then removed as described in Phase I of this experiment. An adjacent sample was removed for a pH determination. Enzymatic activity, percent fiber type, and number of giant fibers were included in the histological data collected from the 20 hour sample.

In all parts of Phase II of this experiment the data were collected using procedures described in Phase I.

Statistical Analysis

The effect of quality levels and enzymatic activity of porcine longissimus on other muscle characteristics was estimated in Phase I. A randomized complete block design with unequal subclass numbers was used in the Analysis of Variance to estimate possible significant differences. Duncan's New Multiple Range Test was used to partition

the means. Correlation coefficients were calculated among certain variables. In Phase II the Analysis of Variance used equal subclasses to estimate the effect of time postmortem on other muscle characteristics.

In both phases, percent fiber type was manipulated by a constant, arc sine ($\sqrt{\text{percent fiber type}/100}$), to normalize the data before the Analysis of Variance was computed. The square root of the number of giant fibers was used in the Analysis of Variance to normalize the variance.

CHAPTER IV

RESULTS AND DISCUSSION

The means and standard deviation of all longissimus muscle characteristics studied in Phase I are shown in Table 1. All of the nine loins in the PSE quality level had abnormal for enzymatic activity. Four loins in the normal group and one in the DFD quality level had abnormal for enzymatic activity.

Phase I Muscle pH

Muscle pH was different ($P < .01$) among the three quality levels (Tables 2, 3) and was found to have a nonsignificant effect on enzymatic activity. This could be explained by the rebounding effect of muscle pH as described by Briskey and Wismer-Pedersen (1961) who found that pH of muscle fell during early rigor, but rose before complete rigor had developed. The simple correlation coefficients of muscle pH (Table 4) were highly significant ($P < .01$) for enzymatic activity ($r = .61$), subjective color ($r = .71$), and reflectance L ($r = .75$). Muscle pH was also correlated ($P < .01$) with the four fiber types. A weaker relationship ($P < .05$) was observed between muscle pH and number of giant fibers ($r = .36$).

Phase I Muscle Color

Subjective muscle color, as shown in Table 2, was different ($P < .01$) among quality levels and enzymatic activity groups.

TABLE 1

PHASE I, MEANS AND STANDARD DEVIATION OF PORCINE *LONGISSIMUS* CHARACTERISTICS

Characteristic	Mean (N = 36)	Standard Deviation
Muscle pH	5.47	.35
Subjective color[a]	2.73	.49
Chromaticity x	.373	.005
Chromaticity y	.348	.005
Reflectance L	47.62	6.62
Number of giant fibers[b]	.58	1.16
Percent of fibers 1	52.19	35.42
Percent of fibers 2	13.42	10.20
Percent of fibers 3	8.33	11.41
Percent of fibers 4	26.06	35.12
Waviness of muscle fiber[c]	2.14	.93

[a]Iowa State 1-5 scale. 1 = PSE, 5 = DFD.

[b]Number per 1200 square cm. at 360x.

[c]Subjective 1-4 scale. 1 = straight, 4 = very wavy.

TABLE 2

PHASE I, SUMMARY OF ANALYSIS OF VARIANCE OF PORCINE LONGISSIMUS CHARACTERISTICS

Variable	Source	df	Mean Square	Error Mean Square
Muscle pH	Quality level[a]	2	.972**	.023
	Enzymatic activity[b]	1	.016	
Subjective color	Quality level	2	3.477**	.425
	Enzymatic activity	1	3.383	
Chromaticity x	Quality level	2	.00001	.00003489
	Enzymatic activity	1	.000014	
Chromaticity y	Quality level	2	.000114**	.00001589
	Enzymatic activity	1	.000054	
Reflectance L	Quality level	2	176.52**	14.514
	Enzymatic activity	1	296.045**	
Percent fiber 1	Quality level	2	0.074	.028
	Enzymatic activity	1	2.229**	
Percent fiber 2	Quality level	2	.013	.008
	Enzymatic activity	1	.436	
Percent fiber 3	Quality level	2	.002	.009
	Enzymatic activity	1	.776**	
Percent fiber 4	Quality level	2	.0430	.018
	Enzymatic activity	1	2.916**	
Waviness	Quality level	2	.849	.734
	Enzymatic activity	1	.325	

TABLE 2 (continued)

Variable	Source	df	Mean Square	Error Mean Square
No. giant fibers	Quality level	2	2.05**	.345
	Enzymatic activity	1	2.771**	

[a]The three quality levels are: PSE, Normal, DFD.

[b]Enzymatic activity was judged as either normal or abnormal.

*$P < .05$.

**$P < .01$.

TABLE 3

PHASE I, MEANS AND STANDARD DEVIATION FOR THREE QUALITY LEVELS OF PORCINE LONGISSIMUS MUSCLE CHARACTERISTICS SEPARATED BY DUNCAN'S NEW MULTIPLE RANGE

Characteristic	Quality Levels PSE(N = 9)	Normal (N = 17)	DFD (N = 10)
pH	$5.06^{a} \pm .12$	$5.43^{b} \pm .10$	$5.92^{c} \pm .23$
Subjective color[d]	$1.92^{a} \pm .06$	$2.58^{ab} \pm .06$	$3.73^{b} \pm .09$
Chromaticity x	$.373^{a} \pm .004$	$.373^{a} \pm .006$	$.375^{a} \pm .007$
Chromaticity y	$.351^{a} \pm .003$	$.350^{ab} \pm .003$	$.344^{b} \pm .006$
Reflectance L	$52.62^{a} \pm 4.07$	$49.16^{a} \pm 3.79$	$40.49^{b} \pm 6.65$
No. Giant Fibers[e]	$.33^{a} \pm .71$	$.35^{a} \pm .99$	$1.20^{b} \pm 1.55$
Percent fiber 1	$6.9^{a} \pm 13.4$	$61.3^{a} \pm 30.8$	$77.5^{a} \pm 10.8$
Percent fiber 2	$1.4^{a} \pm 2.7$	$17.6^{a} \pm 9.7$	$17.1^{a} \pm 6.2$
Percent fiber 3	$20.0^{a} \pm 10.04$	$6.1^{a} \pm 10.7$	$1.7^{a} \pm 3.5$
Percent fiber 4	$71.7^{a} \pm 18.6$	$15.0^{a} \pm 29.0$	$3.7^{a} \pm 11.1$
Waviness[f]	$2.86^{a} \pm .88$	$1.91^{a} \pm .90$	$1.90^{a} \pm .72$

Means with different superscripts (a,b,c) are significantly ($P < .05$) different.

[d] Iowa State Subjective 1-5 scale. 1 = PSE, 5 = DFD.

[e] Number per 1200 square cm at 360x.

[f] Subjective 1-4 scale. 1 = straight, 4 = wavy.

TABLE 4

PHASE I, SIMPLE CORRELATION COEFFICIENTS BETWEEN MUSCLE pH AND CERTAIN PORCINE LONGISSIMUS CHARACTERISTICS

Characteristic	Correlation Coefficient with Muscle pH
Enzymatic activity	.61**
Subjective color	.72**
Chromaticity x	.11
Chromaticity y	-.71**
Reflectance L	.75**
Percent fiber 1	.60**
Percent fiber 2	.42**
Percent fiber 3	-.55**
Percent fiber 4	-.64**
No. Giant Fibers	.36*

*P < .05.

**P < .01.

Table 3 shows subjective muscle color for DFD samples was darker ($P < .05$) than that of PSE but not darker than the normal quality level.

Chromaticity x differences were nonsignificant (Table 2) for both quality level and enzymatic activity. Chromaticity y was different ($P < .01$) between PSE and DFD quality levels (Table 3), but was nonsignificant for enzymatic activity.

Reflectance differed ($P < .01$) for both quality level and enzymatic activity. The statistical analysis (Table 3) indicated significant differences in both lightness or reflectance (L) and chromaticity (x,y). Obviously, some effort must be made to relate the relative differences to visually perceivable terms. The means for the chromaticity values among the three quality levels vary at most by 0.007 units with a standard deviation of 0.005 (Table 1). Plotting the mean coordinates and their standard deviation into a Munsell renotation chart (Newhall et al., 1943) for value 6, the value adjacent to the mean L value, showed a variation of 0.83 hue step and 0.17 chroma step. Conversion of the means of the L values for each quality level (Table 3) resulted in a Munsell value difference of 0.33 between normal and PSE and 0.86 between normal and DFD, while the difference in value between PSE and DFD was 1.19. A difference of one unit in any of the Munsell color attributes, hue, value, or chroma, is a fair approximation of a limen or <u>just noticeable difference</u>. Therefore, the only visual difference which can be substantiated by the data was a difference in lightness, or total reflectance between PSE and DFD.

The small standard deviation between quality levels and within quality levels indicates the lack of variance in chromaticity, whereas the large standard deviations for lightness or reflectance between and among quality levels indicates the degree of variability in the amount of total reflectance. A possible explanation for the lack of difference between normal and PSE reflectance is that four loins were categorized as normal by pH but showed typical PSE color, increasing the reflectance means of the normal group and the standard deviation.

Phase I Percent Fiber Type and Number of Giant Fibers

Among the three quality levels there was no difference observed for percent fiber types (Table 2, Page 18). Table 3 shows that there were large differences between the means of percent fiber types, but the differences were not significant because of the large variation within each quality level group as shown by abnormally large standard deviations. Loins with abnormal enzymatic activity were represented in all three quality levels, which could help explain the large within group variation.

Figure 1 shows normal enzymatic activity and typical fiber distribution of porcine longissimus stained for ATPase and NADH - TR. Also in Figure 1 is a giant fiber (Fiber A). The number of giant fibers was highly significant ($P < .01$) for both quality level and enzymatic activity (Table 2, page 18). The number of giant fibers in DFD porcine longissimus was shown to be significantly ($P < .05$) more abundant than in both normal and PSE quality levels.

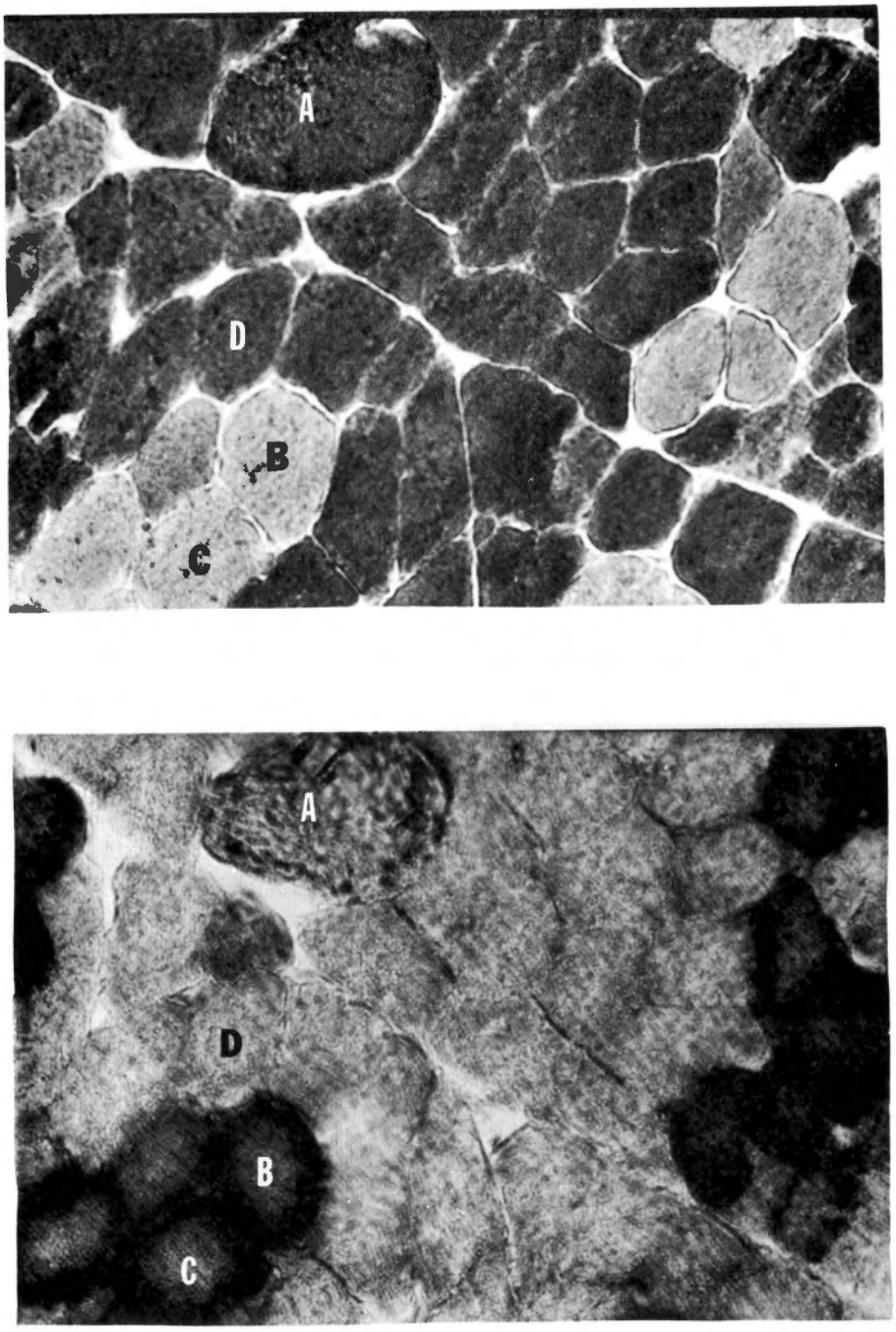


Figure 1. Serial cross sections (10μ) of porcine longissimus 20 hours postmortem from normal muscle.

Total magnification 230x. Tissue in the top photomicrograph was stained for ATPase and the bottom stained for NADH - TR (Engel and Brooke, 1967). Tissue was judged normal for enzymatic activity.

Fiber A demonstrates a giant fiber. Fiber B, C demonstrate fiber type 2. Fiber D demonstrates fiber type 1.

Photomicrographs of porcine longissimus with abnormal enzymatic activity are presented in Figure 2. It appeared the NADH - TR stain is normal because it demonstrated the normal fiber distribution pattern, but the ATPase appeared to be a complete reversal of the expected or normal alkaline AtPase pattern. The pattern of fiber distribution in Figure 2 is very similar to that shown in acid stabile ATPase staining (Guth and Samaha, 1969). Park (1974) reported the percentage of alkaline ATPase positive fibers for this abnormal type reaction compared well with the typical red fiber percentage in normal porcine longissimus.

The appearance of the negative ATPase staining fibers in the abnormal reaction was noticeably different from the tissue with a normal staining pattern.

The staining pattern of severe PSE muscle is shown in Figure 3. The enzymatic reaction for ATPase and NADH - TR was non-reciprocal. The number of positive fibers in both stains was reduced. The positive fibers of the ATPase reaction stained very strongly, while the positive NADH - TR fibers were only slightly positive. The obvious deterioration of the tissue shown in Figure 3 was typical of low pH (below 5.1) muscle.

Waviness

Waviness of muscle fibers was found to be nonsignificant (Table 2, page 18) for quality level and enzymatic activity.

In phase II of this study a total of nine carcasses were sampled at 45 and 90 minutes and again at six and 20 hours. Five carcasses were abnormal for ATPase and NADH - TR enzymatic activity at each

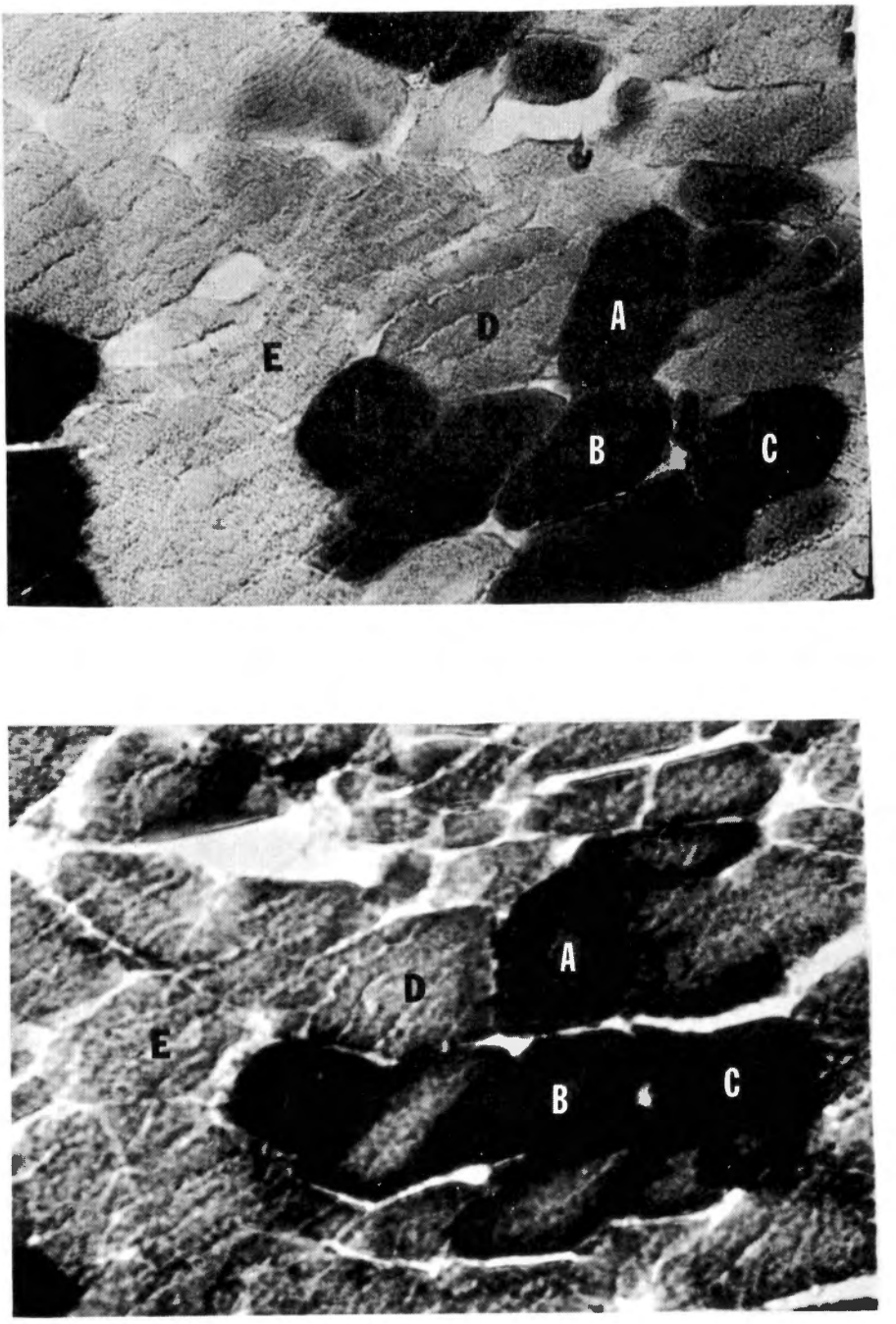


Figure 2. Serial cross sections (10μ) of porcine longissimus 20 hours postmortem from moderately PSE muscle.

Total magnification 230x. Tissue in the top photomicrograph was stained for ATPase and the bottom stained for NADH - TR (Engel and Brooke, 1967). Tissue was judged abnormal for enzymatic activity.

Fiber A, B, C demonstrate fiber type 3. Fiber D, E demonstrate fiber type 4.

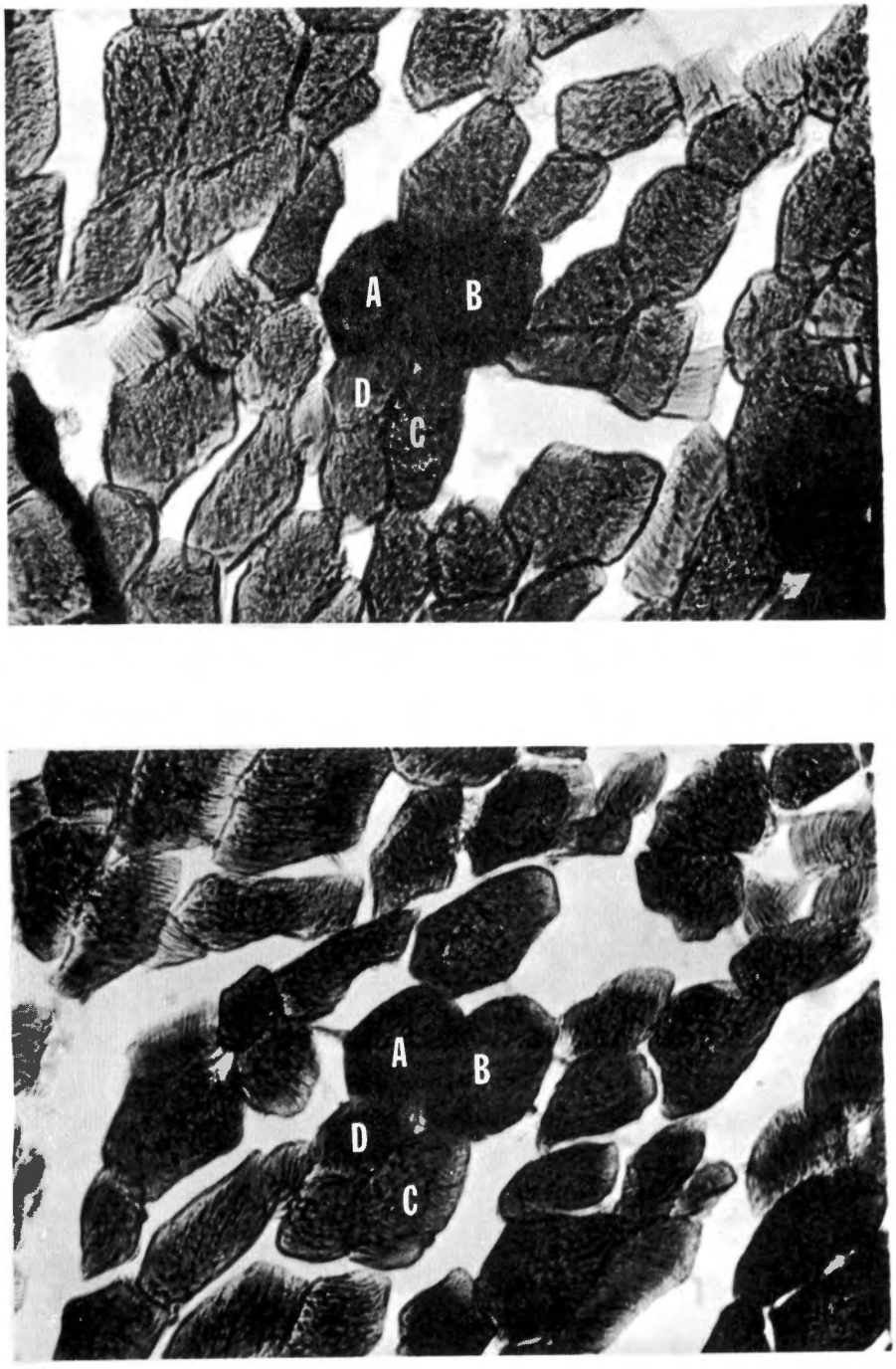


Figure 3. Serial cross sections (10μ) of porcine longissimus 20 hours postmortem from extremely PSE muscle.

Total magnification 230x. Tissue in the top photomicrograph was stained for ATPase and the bottom stained for NADH - TR (Engel and Brooke, 1967). Tissue was judged abnormal for enzymatic activity.

Fiber A, B demonstrate fiber type 3. Fiber C, D demonstrate fiber type 4.

of the four time periods postmortem, three were normal at each of the sampling times, and one was abnormal only at the 20 hour postmortem sampling.

Phase II Muscle pH and Temperature

In the Analysis of Variance (Table 5) significant ($P < .01$) carcass variation was observed for both muscle pH and temperature. Muscle temperature decreased ($P < .01$) with time postmortem (Tables 5, 6). Table 7 shows the carcasses that had normal enzymatic activity had their lowest pH at six hours postmortem. Carcasses with abnormal enzymatic activity showed a gradual decrease in pH from 45 minutes to the 20 hour postmortem sampling time (Table 7).

Phase II Muscle Color

One of the nine carcasses was not judged for subjective color because the longissimus muscle was heterogeneous. One-half of the muscle was very pale in color and the other half was slightly darker than normal. The different areas were divided by a distinct line with no intermediate color evident. Although not statistically tested the color data in Table 8 appears to be very similar to the data collected in phase I (Table 1, page 17). The same extremely low standard deviations in chromaticity and large standard deviations in reflectance are found in Table 8 as previously noted in phase I.

Phase II Percent Muscle Fiber Type and Number of Giant Fibers

The percentage of each fiber type did not change with time postmortem (Table 5). Each fiber type except fiber 3 was different

TABLE 5

PHASE II, SUMMARY OF ANALYSIS OF VARIANCE OF PORCINE *LONGISSIMUS* CHARACTERISTICS

Variable	Source	df	Mean Square	Error Mean Square
Muscle temperature	Carcass	8	18.57**	4.264
	Time	3	2611.96**	
Percent fiber 1	Carcass	8	.149**	.012
	Time	3	.013	
Percent fiber 2	Carcass	8	.051**	.011
	Time	3	.014	
Percent fiber 3	Carcass	8	.015	.015
	Time	3	.025	
Percent fiber 4	Carcass	8	.092**	.024
	Time	3	.021	
No. giant fibers	Carcass	8	.055	.080
	Time	3	.111	
Muscle pH	Carcass	8	.125**	.012
	Time	3	.029	

*$P < .05$.

**$P < .01$.

TABLE 6

PHASE II, MEANS AND STANDARD DEVIATION OF PORCINE LONGISSIMUS CHARACTERISTICS AT FOUR TIME PERIODS POSTMORTEM

Characteristics	45 Minutes	90 Minutes	Six Hours	20 Hours
Muscle temperature[a]	41.58 ± 2.69	33.53 ± 4.88	11.72 ± 1.72	4.44 ± 0.73
No. giant fibers[b]	0.0 ± 0.0	0.0 ± 0.0	0.11 ± 0.33	0.22 ± 0.44
Muscle pH	5.70 ± 0.26	5.65 ± 0.22	5.57 ± 0.17	5.59 ± 0.34
Percent fiber 1	55.3 ± 23.7	54.8 ± 27.5	49.1 ± 30.7	48.7 ± 27.4
Percent fiber 2	17.7 ± 10.8	16.4 ± 9.2	12.5 ± 10.1	11.7 ± 8.8
Percent fiber 3	6.0 ± 8.6	8.7 ± 9.9	11.8 ± 13.7	15.6 ± 14.5
Percent fiber 4	21.0 ± 25.8	20.1 ± 26.4	26.6 ± 26.8	23.9 ± 26.4
Chromaticity x				.373 ± .004
Chromaticity y				.350 ± .005
Reflectance L				50.63 ± 8.15
Subjective color[c]				2.81 ± .97

[a]Degrees Celsius.

[b]Number per 1200 square cm. at 360x.

[c]Mean includes eight observations.

TABLE 7

PHASE II, MEAN AND STANDARD DEVIATION OF MUSCLE CHARACTERISTICS OF NORMAL AND ABNORMAL TISSUE AT FOUR TIME PERIODS POSTMORTEM

Characteristic	45 Minutes	90 Minutes	Six Hours	20 Hours
Muscle temperature °C				
Normal[ac]	42.63 ± 1.60	35.25 ± 3.22	12.75 ± .96	4.33 ± .59
Abnormal[bd]	40.74 ± 3.25	32.16 ± 5.88	10.90 ± 1.82	4.5 ± .84
Muscle pH				
Normal	5.88 ± .28	5.79 ± .20	5.67 ± .16	5.89 ± .50
Abnormal	5.58 ± .16	5.53 ± .16	5.48 ± .14	5.42 ± .11
Percent fiber 1				
Normal	74.3 ± 4.0	77.7 ± 2.9	78.0 ± 7.4	79.1 ± 4.4
Abnormal	40.1 ± 21.0	36.3 ± 23.7	25.8 ± 18.4	33.1 ± 18.3
Percent fiber 2				
Normal	24.8 ± 3.2	22.2 ± 2.9	21.8 ± 5.0	20.4 ± 4.8
Abnormal	11.9 ± 11.6	12.6 ± 8.9	5.5 ± 5.8	11.8 ± 10.3
Percent fiber 3				
Normal	0.9 ± 1.8	0.0 ± .0	0.0 ± .0	0.0 ± .0
Abnormal	10.1 ± 9.9	15.6 ± 7.9	21.2 ± 11.2	23.4 ± 10.8

TABLE 7 (Continued)

Characteristic	45 Minutes	90 Minutes	Six Hours	20 Hours
Percent fiber 4				
Normal	0.0 ± .0	0.0 ± .0	1.7 ± 1.4	0.0 ± 0.0
Abnormal	37.7 ± 23.3	36.1 ± 25.7	47.2 ± 15.2	35.9 ± 24.5

[a]Tissue was judged normal for enzymatic activity if it showed reciprocal enzymatic activity and normal distribution of fibers.

[b]Abnormal tissue did not show reciprocal enzymatic activity and normal distribution of fibers.

[c]Means composed of five observations.

[d]Means composed of four observations.

TABLE 8

PHASE II, MEANS AND STANDARD DEVIATION OF PORCINE LONGISSIMUS COLOR CHARACTERISTICS AT 20 HOURS POSTMORTEM

Enzymatic Activity	Subjective Color[a]	Objective Color		
		Chromaticity		Reflectance
		x	y	L
Normal[b]	3.69 ± .99	.375 ± .002	.348 ± .009	42.90 ± 9.70
Abnormal[c]	2.29[d] ± .49	.372 ± .004	.352 ± .003	54.50 ± 3.87

[a]Subjective color measured with Iowa State 1-5 scale.

[b]Means composed of three observations.

[c]Means composed of six observations.

[d]Means composed of five observations.

($P < .01$) among carcasses sampled. Table 5, page 29, also indicated no differences in the number of giant fibers in either carcasses sampled or time postmortem.

The muscle judged abnormal for enzymatic activity was consistently higher in percentage of fiber type 3 and 4 and lower in percentage of fiber type 1 and 2 (Table 5, page 29) than normal tissue. Figures 4-7 show the differences in enzymatic activity from 45 minutes to 20 hours postmortem of longissimus from the same carcass at four times postmortem. The 45 minute postmortem histochemical section of longissimus (Figure 4) that became abnormal for enzymatic activity at 20 hours postmortem, had the reciprocal enzymatic activity and typical fiber distribution at 45 minutes postmortem. Figure 5 is a photomicrograph of the 90 minute postmortem muscle sample from the same tissue as Figure 4. An increase in the space between fibers can be noted. The tissue in Figure 5 still shows typical fiber distribution. Figure 6 illustrates the histochemical activity of the same longissimus at six hours postmortem. Although the typical fiber distribution pattern still is present, fiber D and E of Figure 6 do not demonstrate the reciprocal enzymatic relationship. Fiber D and E of Figure 6 are not strongly positive for NADH - TR and in some classification systems would be called intermediate fibers. In Figure 7 the abnormal enzymatic activity of the 20 hour longissimus sample from the same carcass as used in Figures 4 through 6 is shown. The fiber distribution appears typical in the NADH - TR stain, but is without a typical pattern in the ATPase reaction.

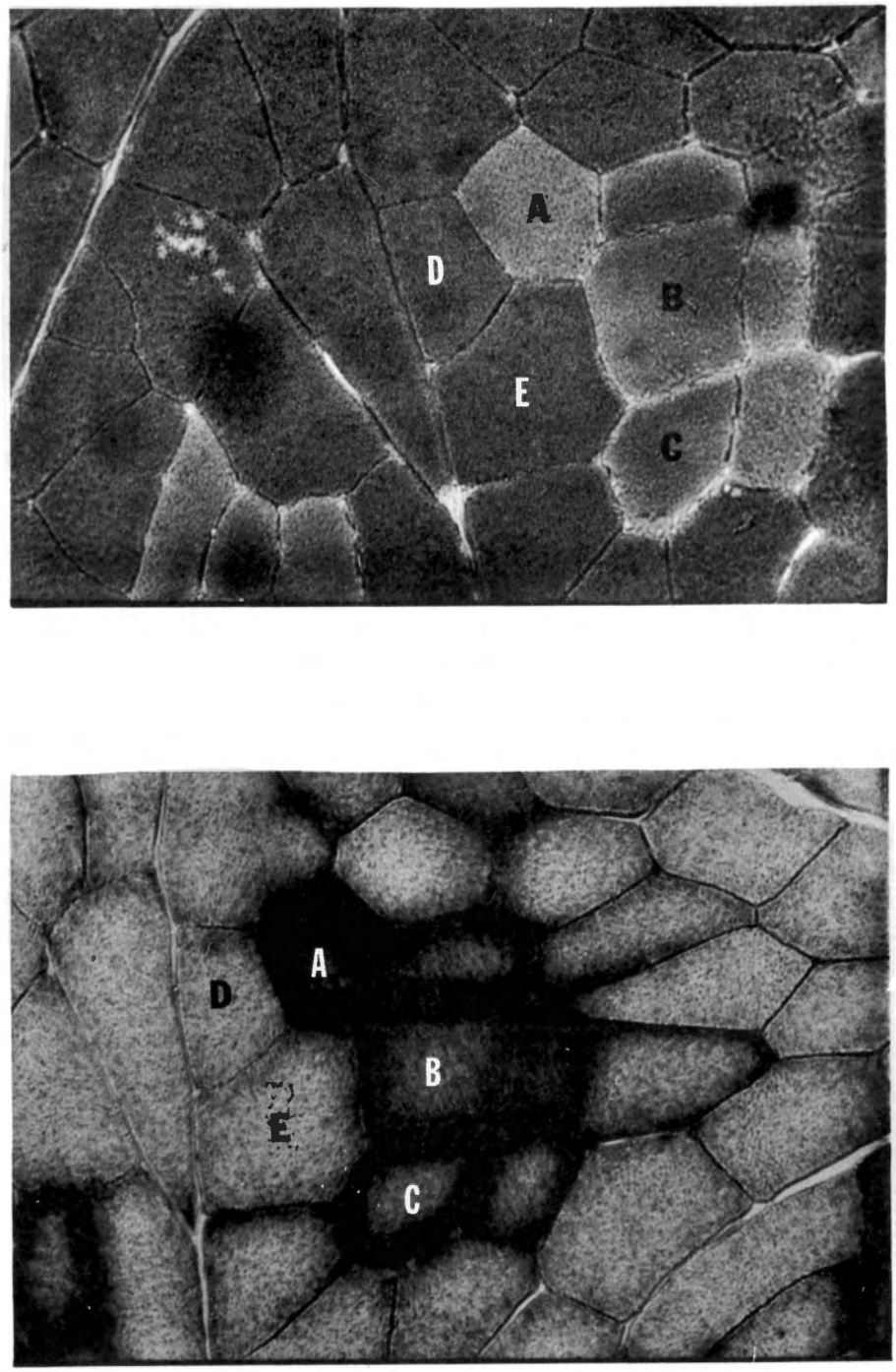


Figure 4. Serial cross sections (10μ) of porcine longissimus 45 minutes postmortem.

Total magnification 230x. Tissue in the top photomicrograph was stained for ATPase and the bottom stained for NADH - TR (Engel and Brooke, 1967). Tissue was judged normal for enzymatic activity.

Fiber A, B, C demonstrate fiber type 2. Fiber D, E demonstrate fiber type 1.

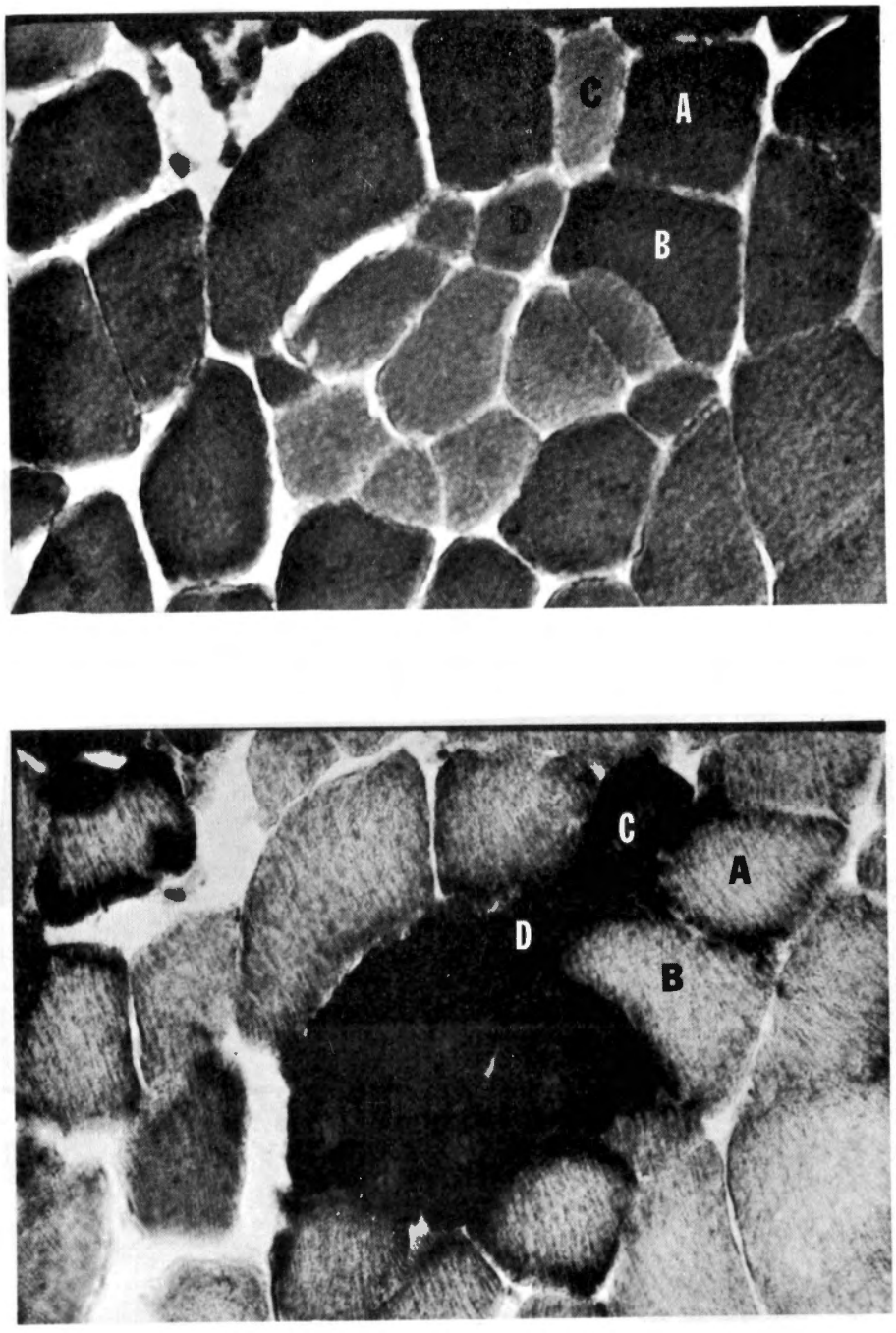


Figure 5. Serial cross section (10μ) of porcine longissimus 90 minutes postmortem.

Total magnification 230x. Tissue in the top photomicrograph was stained for ATPase and the bottom stained for NADH - TR (Engel and Brooke, 1967). Tissue was judged normal for enzymatic activity.

Fiber A, B demonstrate fiber type 1. Fiber C, D demonstrate fiber type 2.

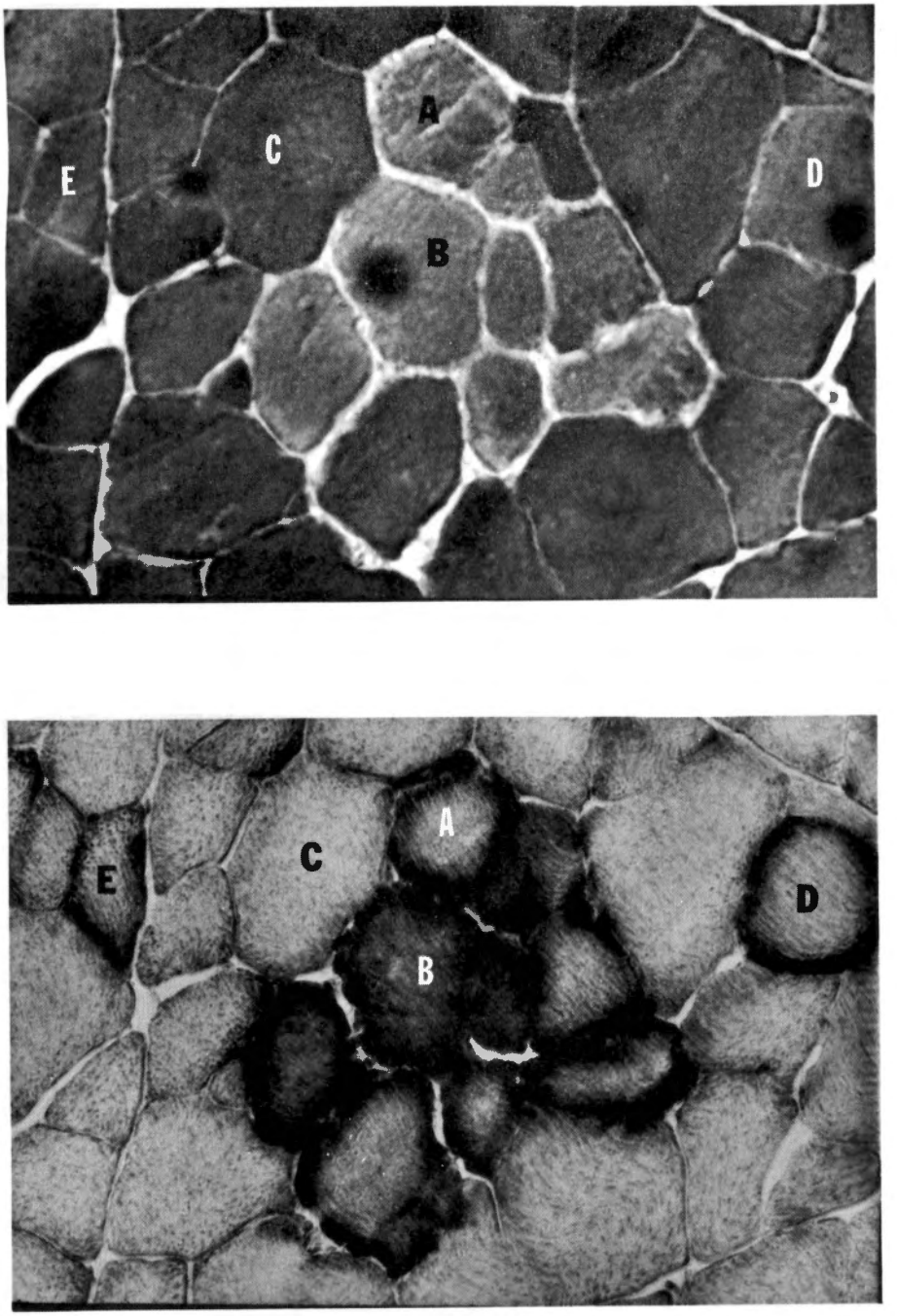


Figure 6. Serial cross sections (10μ) of porcine longissimus six hours postmortem.

Total magnification 230x. Tissue in the top photomicrograph was stained for ATPase and the bottom stained for NADH - TR (Engel and Brooke, 1967). Tissue was judged normal for enzymatic activity.

Fiber A, B demonstrate fiber type 2. Fiber C demonstrates fiber type 1. Fiber D, E demonstrate fiber type 3.

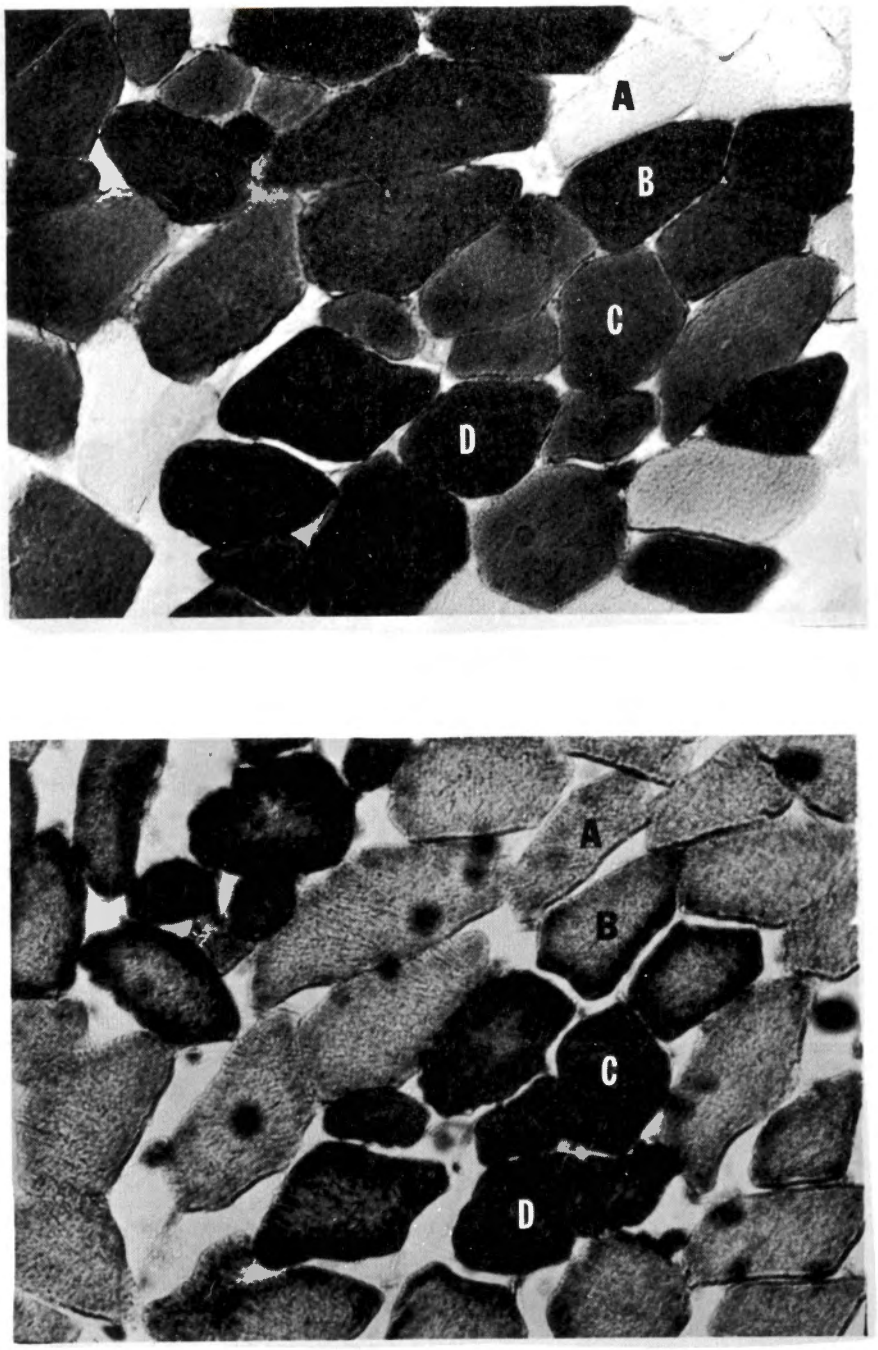


Figure 7. Serial cross sections (10μ) of porcine longissimus 20 hours postmortem.

Total magnification 230x. Tissue in the top photomicrograph was stained for ATPase and the bottom stained for NADH - TR (Engel and Brooke, 1967). Tissue was judged abnormal for enzymatic activity.

Fiber A demonstrates fiber type 4. Fiber B demonstrates fiber type 2. Fiber C, D demonstrate fiber type 3.

This series of figures suggests the reciprocal enzymatic activity of ATPase and NADH - TR was affected by postmortem changes in the muscle and that the observed abnormality in staining pattern is not the result of four different fiber types. Because the pattern of fiber distribution in the NADH - TR reaction appeared normal in all but the most severe PSE samples, it would appear the ATPase reaction is being altered by the postmortem environment of the tissue. For this reason, ATPase muscle staining reactions could be misleading in fiber classification, but could be a histochemical indicator of PSE muscle.

As there are various degrees in the severity of PSE porcine muscle, there likewise has been noted different patterns of abnormal ATPase reactions in this study. The pattern of ATPase stain shown in Figure 7 is typical of that judged only slightly PSE by subjective color. Figure 2, page 26, shows the pattern that is typical of muscle judged moderately PSE. The muscle that is judged extremely PSE tends to produce the pattern of muscle fiber staining shown in Figure 3, page 27.

CHAPTER V

SUMMARY

This study was conducted in two phases. In phase I, muscle pH, subjective muscle color, objective muscle color, percentage of fiber types, number of giant fibers and the waviness of muscle fibers were studied in three quality levels of porcine longissimus. Thirty-six loins were purchased at a local packing plant after a 20 hour post-mortem chill and placed into quality levels according to muscle pH. Loins were classified as PSE if the pH was below 5.30, normal if pH was between 5.30 and 5.70, and DFD if pH was above 5.70. Of the 36 loins tested 14 did not demonstrate typical fiber distribution or show reciprocal enzymatic activity between the oxidative and phosphatase enzyme stains.

There was a significant ($P < .01$) difference in subjective color between the enzymatically normal and abnormal groups. The DFD group was darker ($P < .01$) than the PSE group in subjective color. The variation in chromaticity x and y was very small and did not represent a visually significant difference between quality levels, while there was measured a visually significant difference in lightness. The observed differences in subjective color were caused by variation in the lightness or total reflectance of the muscle. The percentage of muscle fibers was not affected by quality levels, but there was an increase ($P < .05$) in the number of giant fibers found in the DFD

group. The loins with abnormal enzymatic activity had higher ($P < .01$) percentages of fibers that did not show reciprocal enzymatic activity.

Waviness of muscle fibers was found to be nonsignificant among quality levels. In phase II, nine pork longissimus were sampled at 45 minutes, 90 minutes, six hours, and 20 hours postmortem. Muscle pH and temperature, percentage of fiber type, and number of giant fibers were recorded for each time period. Color was measured objectively and subjectively at 20 hours postmortem.

Of the nine carcasses sampled, five were abnormal for enzymatic activity at each sampling time postmortem, three were normal at each sampling time, and one showed abnormal enzymatic activity only at the 20 hour postmortem time.

The percentage of fiber types did not change with time postmortem, but was different ($P < .01$) among carcasses sampled. Large fiber type mean differences were observed between the normal and abnormal enzymatic groups, but the differences were statistically nonsignificant because of large within group variation.

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APPENDIXES

APPENDIX A

HISTOCHEMICAL PROCEDURES

NADH - TR Stain (W. K. Engel and M. H. Brooke, 1967)

Reduced disphosphopyridine nucleotide-tetrazolium reductase (NADH - TR) Solution:

0.2 M tris buffer (ph 7.4) - 10 ml

Nitro blue tetrazolium - 10 mg

Reduced diphosphopyridine nucleotide - 8 mg

The coverslips to which tissue sections are adhered are:

1. Incubated in the above solution for 30 minutes at 36° C.
2. Taken through acetone-water mixtures of the following strengths in order given: 30/70, 60/40, 90/10, 60/40, 30/70.
3. Rinsed in distilled water.
4. Mounted in glycerogel.

ATPase Stain (W. K. Engel and M. H. Brooks, 1967)

Adenosine triphosphatase (ATPase)

Solution (prepare fresh):

0.1 M sodium barbital (2.06 G in 100 ml distilled water) - 2 ml

0.18 M calcium chloride (1.998 G in 100 ml distilled water) - 1 ml

Distilled water - 3ml

Adenosine triphosphate - 25 mg

Adjust pH to 9.4 with 0.1 N NaOH

Make up to 10 ml and filter.

The coverslips to which the sections are adhered are:

1. Preincubate using 1% $CaCl_2$ at 36°C for 10 minutes.
2. Rinse distilled water.
3. Incubated for 10-20 minutes in the above solution at 36° C.
4. Washed in three changes of 1% calcium chloride for a total of 10 minutes.
5. Transferred to 2% cobaltous chloride for three minutes.
6. Washed in distilled water.
7. Immersed in 1% yellow ammonium sulfide for 30 seconds.
8. Rinsed in water.
9. Mounted in glycerogel.

APPENDIX B

RAW DATA COLLECTED

The raw data collected in this study and used for all analysis are shown in Tables 9, 10 and 11.

TABLE 9

PHASE I, DATA COLLECTED

Observation	pH	Enzymatic Activity[a]	Subjective Color	Chromaticity x	Chromaticity y	Reflectance L	No. Giant Fiber	No. Fiber 1	No. Fiber 2	No. Fiber 3	No. Fiber 4	Waviness
1	5.84	1	.300	.385	.346	39.62	0	177	47	0	0	2.25
2	5.64	1	.200	.372	.352	48.99	0	266	69	0	0	1.63
3	5.52	1	.200	.364	.351	46.48	0	259	87	0	0	1.00
4	5.62	1	.225	.376	.342	45.61	0	219	63	0	0	1.13
5	5.71	1	.350	.377	.344	44.50	0	385	109	0	0	2.55
6	5.10	2	.100	.372	.345	45.61	0	0	0	25	385	4.00
7	5.19	2	.225	.374	.350	52.54	0	0	0	22	340	2.50
8	5.42	1	.285	.370	.346	43.01	0	244	60	0	0	2.88
9	5.79	1	.325	.374	.340	45.06	0	224	65	0	0	1.25
10	5.30	2	.225	.375	.348	52.73	0	0	0	55	183	3.50
11	6.33	1	.420	.382	.334	37.28	0	250	48	0	0	1.05
12	5.71	1	.350	.376	.346	33.91	3	182	46	0	0	1.00

TABLE 9 (Continued)

Observation	pH	Enzymatic Activity[a]	Subjective Color	Chromaticity x	Chromaticity y	Reflectance L	No. Giant Fiber	No. Fiber 1	No. Fiber 2	No. Fiber 3	No. Fiber 4	Waviness
13	5.75	2	.210	.367	.355	55.32	1	101	11	23	74	2.38
14	5.77	1	.350	.381	.349	40.74	0	191	39	0	0	2.50
15	5.21	2	.290	.376	.352	49.80	0	54	1	116	146	2.63
16	5.50	2	.240	.374	.353	55.59	1	21	11	90	169	2.00
17	5.40	1	.310	.365	.347	49.80	0	230	48	0	0	1.30
18	5.53	2	.185	.372	.350	51.86	4	84	23	69	120	1.13
19	5.35	1	.255	.382	.356	51.10	0	233	74	0	0	3.50
20	6.14	1	.500	.376	.343	34.93	3	146	47	0	0	2.38
21	5.39	1	.260	.384	.354	50.60	0	163	78	0	0	2.00
22	6.22	1	.475	.367	.336	33.02	4	192	22	8	4	2.63
23	5.43	1	.275	.372	.349	46.90	0	231	78	0	0	1.75
24	4.90	2	.225	.378	.354	55.14	0	0	0	67	176	1.73
25	5.40	1	.300	.375	.350	48.17	0	229	67	0	0	1.68

TABLE 9 (Continued)

Observation	pH	Enzymatic Activity[a]	Subjective Color	Chromaticity x	Chromaticity y	Reflectance L	No. Giant Fiber	No. Fiber 1	No. Fiber 2	No. Fiber 3	No. Fiber 4	Waviness
26	4.98	2	.225	.377	.352	.52.35	0	0	0	106	335	4.00
27	5.30	1	.450	.385	.353	.42.07	0	150	57	3	0	1.38
28	5.31	2	.180	.372	.353	.55.86	1	0	0	58	228	1.65
29	5.47	1	.300	.368	.350	.47.54	0	234	48	0	0	1.13
30	5.05	2	.100	.367	.352	.60.25	1	5	0	58	271	2.50
31	5.91	1	.450	.366	.343	.40.50	1	240	36	6	0	1.00
32	5.20	2	.235	.370	.351	.51.48	0	143	18	56	143	3.88
33	4.99	2	.140	.371	.352	.51.09	0	2	0	71	166	2.55
34	5.37	1	.250	.366	.349	.49.90	0	220	30	8	1	3.60
35	4.96	2	.190	.368	.351	.55.32	2	8	18	43	186	1.93
36	5.38	1	.250	.370	.350	49.50	0	200	61	3	0	1.15

[a]Enzymatic activity was coded so that 1 = normal, 2 = abnormal.

TABLE 10

PHASE II, COLOR DATA COLLECTED

Carcass	Chromaticity x	Chromaticity y	Reflectance L	Subjective Color
1	.379	.353	54.22	1.50
2	.367	.350	58.56	2.55
3	.376	.339	31.78	4.80
4	.376	.357	49.60	2.90
5	.373	.355	49.09	2.80
6	.375	.354	52.73	2.35
7	.369	.348	53.01	2.25
8	.373	.349	47.33	3.35
9	.371	.350	59.36	Not Scored

TABLE 11

PHASE II, DATA COLLECTED

Observation	Carcass	Time[a]	Temperature	No. Fiber 1	No. Fiber 2	No. Fiber 3	No. Fiber 4	No. Giant Fiber	pH	Enzymatic Activity[b]
1	1	0	42.0	142	61	4	40	0	5.65	1
2	1	1	37.0	212	68	38	26	0	5.67	1
3	1	2	13.0	147	40	49	117	0	5.52	1
4	1	3	6.0	94	39	50	156	0	5.52	1
5	2	0	35.0	52	0	38	85	0	5.46	1
6	2	1	22.0	179	69	30	45	0	5.44	1
7	2	2	8.0	145	9	39	122	0	5.43	1
8	2	3	4.0	125	7	74	103	0	5.42	1
9	3	0	45.0	148	55	0	0	0	6.27	2
10	3	1	38.5	227	74	0	0	0	6.04	2
11	3	2	12.0	173	69	0	7	0	5.78	2
12	3	3	5.0	199	65	0	0	1	6.42	2

TABLE 11 (Continued)

Observation	Carcass	Time[a]	Temperature	No. Fiber 1	No. Fiber 2	No. Fiber 3	No. Fiber 4	No. Giant Fiber	pH	Enzymatic[b] Activity
13	4	0	42.0	169	60	0	0	0	5.66	2
14	4	1	37.5	190	54	0	0	0	5.64	2
15	4	2	14.0	194	63	0	0	0	5.55	2
16	4	3	5.0	96	41	32	67	0	5.44	1
17	5	0	42.0	219	55	0	0	0	5.70	2
18	5	1	33.0	252	56	0	0	0	5.62	2
19	5	2	12.0	255	38	0	0	0	5.52	2
20	5	3	4.0	201	36	0	0	0	5.42	2
21	6	0	43.0	30	1	75	283	0	5.43	1
22	6	1	35.0	8	2	98	251	0	5.32	1
23	6	2	11.5	0	3	76	187	0	5.28	1
24	6	3	4.0	0	0	61	185	0	5.27	1

TABLE 11 (Continued)

Observation	Carcass	Time[a]	Temperature	No. Fiber 1	No. Fiber 2	No. Fiber 3	No. Fiber 4	No. Giant Fiber	pH	Enzymatic Activity[b]
25	7	0	42.2	198	75	0	66	0	5.68	1
26	7	1	33.0	88	4	62	152	0	5.52	1
27	7	2	11.0	66	0	107	113	1	5.52	1
28	7	3	4.0	91	4	73	0	0	5.45	1
29	8	0	41.5	159	58	8	0	0	5.90	2
30	8	1	32.0	165	52	0	0	0	5.88	2
31	8	2	13.0	246	61	0	0	0	5.84	2
32	8	3	4.0	189	52	0	0	1	5.82	2
33	9	0	41.5	158	43	27	106	0	5.55	1
34	9	1	33.8	93	43	28	108	0	5.68	1
35	9	2	11.0	62	39	44	175	0	5.66	1
36	9	3	4.0	72	21	39	63	0	5.57	1

TABLE 11 (Continued)

[a]Time periods were coded as follows:

0 = 45 minutes postmortem.

1 = 90 minutes postmortem.

2 = six hours postmortem.

3 = 20 hours postmortem.

[b]Enzymatic activity was coded as follows:

1 = normal.

2 = abnormal.

VITA

Joseph Edwin Marcy, son of Mr. and Mrs. Charles S. Marcy, Jr., was born August 26, 1952 in Bristol, Virginia. He graduated from Central High School, Sullivan County, Tennessee, in June of 1970. The following September he entered the University of Tennessee, and in August of 1974, received his Bachelor of Science in Agriculture with honors. August of 1974 he accepted a graduate research assistantship at the University of Tennessee in Food Technology and Science and began his study towards a Master's degree. He received his degree in June of 1976. He is a member of Alpha Zeta and Gamma Sigma Delta.

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