POST-MORTEM ELECTRICAL STIMULATION OF CARCASSES

EFFECTS ON BIOCHEMISTRY, BIOPHYSICS, MICROBIOLOGY AND QUALITY OF MEAT

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-CONTENTS-

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Post-Mortem Electrical Stimulation of Carcasses

Effects on Biochemistry, Biophysics, Microbiology and Quality of Meat

A. Asghar and R.L. Henrickson
Animal Science Department

Introduction

Swammerdam probably was the first, according to Needham (1971), to make the empirical discovery in 1663 of the contraction of frog muscle on stimulation with bimetallic wire. Later, Luigi Galvani in 1791 described a similar phenomenon (Bockris and Reddy, 1964), which was noticed by his assistants. Then, in the 1830's, Du Bois-Reymond and Bernstein (1902) demonstrated the role of the muscle membrane in conducting an impulse, while Mateucci (1838) reported the mode of electrical potential transfer from stimulated muscle surface to the nerve (Needham, 1973; Bendall, 1980). In 1879 Hermann proposed a model for electrotonus in nerve which is still accepted with minor modification (Zierler, 1973). From this time onwards, scientific interest in electrical stimulation with respect to muscle physiology increased and led to many discoveries of fundamental significance (Goldman, 1943; Hodgkin and Katz, 1949).

The beneficial effect of electrical stimulation on the quality of turkey meat seemed to be known since the observation of Benjamin Franklin in 1749, according to Lopez and Herbert (1975). However, the limited application of electrical shock in the meat industry remained confined to pre-slaughter anaesthesia for some meat animal species until 1951, when Harsham and Deatherage employed electrical current to accelerate the glycolytic rate and the tenderization process in post-slaughter beef carcasses. A patent was granted to them for this process. At the same time, Rentschler (1951), a physicist by profession, was also granted a patent for designing equipment for the electrical stimulation of carcasses. However, no commercial use was made of either patent by the meat industry.

It was during the early seventies that meat scientists in New Zealand initiated extensive studies on electrical stimulation of lamb carcasses in an attempt to solve the problem of meat toughening caused by cold shortening. Later, several researchers in the United States, England, Australia, West Germany, Sweden, Belgium, and Russia directed their efforts toward exploring the effects of various electrical parameters on the physicochemical, ultrastructural, microbiological, and quality characteristics of meat. Recent developments in meat science revealed that electrical stimulation of the carcass provided many quality advantages. These facts motivated the meat industry in the United States, United Kingdom and New Zealand to adopt electrical stimulation unit operation in the slaughter of both beef and lamb. The current information on all aspects of electrical stimulation is presented in this review.

Effect On Biochemistry of Muscle

The effects of electrical stimulation on certain aspects of muscle biochemistry have been studied in relation to the onset of rigor mortis. The important findings are summarized below:

Rate of Glycolysis

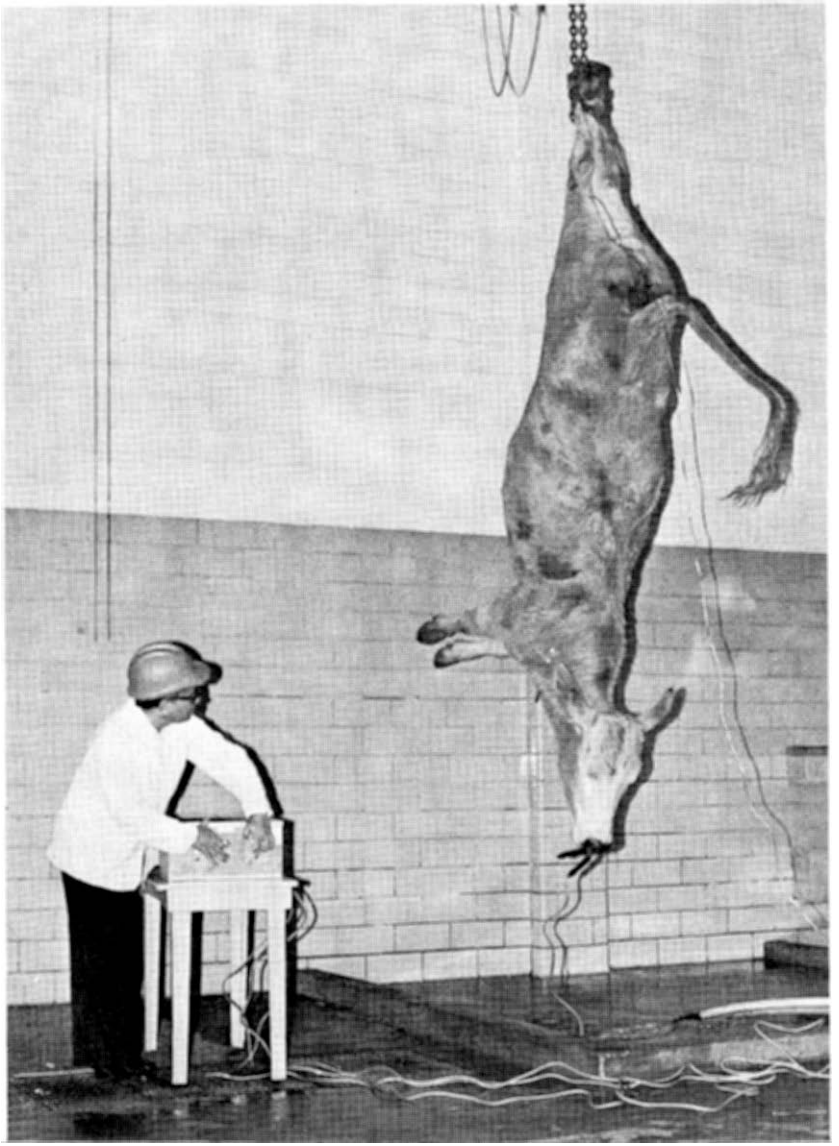
Meyerhof in 1921 demonstrated that glycogen was catabolized to lactic acid during muscle stimulation. He related these chemical changes to the amount of work done by the muscle. Based on this original finding, the post-mortem rate of glycolysis in muscle is generally expressed by recording the change in pH value of muscle at suitable intervals. Muscle pH declines soon after slaughtering the animal and continues over a period of about 24 hours or more until the ultimate pH is reached.

Numerous studies have shown that electrical stimulation accelerates the rate of post-mortem glycolysis in muscles (Harsham and Deatherage, 1951; Karpatkin et al., 1964; Hallund and Bendall, 1965; Forrest et al., 1966; Forrest and Briskey, 1967; McLoughlin, 1970) to replenish the adnosine triphosphate (ATP) which is rapidly catabolized during the stimulation process. This amounts to an increase in the biochemical reaction of the glycolytic pathway by 100 to 150 times due to the mechanical activity of the muscles. The overall process hastens the development of rigor mortis. When all the potential sources of ATP are exhausted, and the ATP level is reduced to below 0.1 mole/g and the extensibility of muscle is lost, it is said to be in rigor (Bendall, 1973).

Further studies have distinguished two distinct phases of glycolysis in muscles as a result of electrical stimulation. The first represents the electrical stimulation period (consisting of $\sim$2 minutes) during which time a very pronounced drop in pH of about 0.4 to 0.7 units occurs. This is designated as Δ pH (Bendall et al., 1976; Chrystall and Devine, 1978), the extent of which is highly dependent upon the stimulation parameters. The second phase symbolizes the post-stimulation period during which the rate of pH fall (expressed as dpH/dt) is relatively slower than the first phase, but still 1.5 to 2 times faster than the non-stimulated muscle (Chrystall and Devine, 1978). However, Bendall et al. (1976) concluded that the rate of pH fall after stimulation was identical to that in the non-stimulated muscle when corrections were made according to the temperature coefficient and rate of pH fall (Jeacocke, 1977) for the temperature difference in stimulated and unstimulated beef carcasses.

Chrystall and Devine (1978) re-examined this discrepancy by holding both stimulated and unstimulated carcass sides at isotemperature (36°C). At this temperature correction for temperature was not needed. The results from this study again did not agree with the observations of Bendall et al. (1976). Savell et al. (1977) reported that in the case of carcasses from heavy weight, grain-fed animals, the pH difference between stimulated and unstimulated sides was significant only at 1 and 6 hours post-mortem, thereafter the pH values became identical.

Information is not available on the response to electrical stimulation of carcasses from underfed animals. It has, however, been ovserved that the rate of glycolysis is relatively slower in carcasses from underfed lambs than in those from well-nourished animals (Asghar, 1969). Size of the glycogen granules in muscle has also been reported to influence the glycolysis rate (Lawrie et al., 1959). Small granules are catabolized at a slow rate (Larner et al., 1956). It is also well accepted that the decrease in pH adversely affects the activity of the glycolytic enzymes. It is, however, not known whether or not different isozymes of the glycolytic pathway have any bearing on the rate of pH fall per se in respect to electrical stimulation.

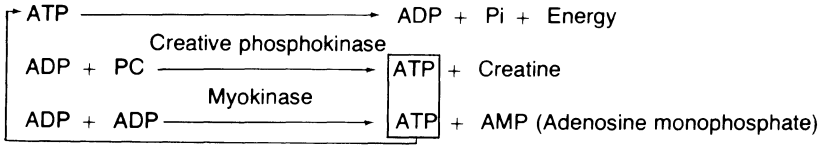


Post-Mortem Electrical Stimulation of a Beef Carcass

Energy-rich Phosphate Compounds

The content of energy-rich phosphate compounds, Adenosine Triphosphate (ATP) and phosphocreative (PC) in muscle is of fundamental significance to the concept of electrical stimulation, as they determine the response of muscle fiber shortening during chilling or freezing of muscle. From the fundamental discovery of Lohmann (1931), adenosine triphosphate (ATP) has been regarded as the primary source of energy for muscle contraction (Needham, 1973). It has been reported that 50% of the ATP disappears from muscle at about pH 6.0 and 90% at about 5.7 in 1 and 2 hours post-mortem, respectively, from an electrically stimulated carcass, whereas the unstimulated carcass takes 8.5 to 10.5 hours (Bendall et al., 1976). ATP turnover rate is very high during the first 15 seconds of stimulation (~2.8 mMole/g/sec.) for muscles in complete tetany, and the rate relatively decreased during subsequent periods of stimulation (Bendall et al., 1976).

The ATP content of muscle probably decreases little during the first few seconds of stimulation; because the ATP deficit is replenished by transphosphorylation of mainly adenosine diphosphate (ADP) and to some extent by another reaction (shown below) to maintain the energy supply for the activated muscle:



This accounts for the large reduction in phosphocreatine (PC) content of muscle during the first 30 seconds of stimulation (Bendall, 1976). However, under the same electrical stimulating conditions the decrease in PC content in different muscles was reported to be varied. For example, the PC decreased from an initial value of 14 μ Mol g^{-1} in the L. dorsi during electrical stimulations of the lamb carcass. During the same period, 9 μ Mol of PC per gram of biceps femoris was catabolized.

When the PC store has been exhausted and oxygen stores are depleted, further demand for the ATP of the activated muscle triggers the anaerobic glycolysis of the glycogen reserve which is then the only avenue for post-mortem muscle to procure ATP. Consequently, glycogen is converted to lactic acid. It has been shown that in the case of steer carcasses glycogen decreases from 25.5 mg to about 22 mg per gram of muscle during electrical stimulation for 90 seconds, with concomitant increase in lactic acid from about 25 μ Mol to 42 μ Mol per gram of muscle (Clarke et al., 1980). This suggests that the lactate production rate on the average was 11 μ Mol/min/g in stimulated muscle as compared to merely 0.1/μ Mol/min/g in unstimulated muscle. Bendall (1976) reported that a total of 50 and 70 μ Mol of energy-rich phosphate compounds per gram of L. dorsi and biceps femoris, respectively, were catabolized during electrical stimulation of lamb carcass for 120 seconds. This means that carcasses can be subjected to cooling or freezing temperature after dressing much earlier (~2 hours post-mortem) than in the conventional practice (~10 hour post-mortem) without risking cold induced toughening of meat.

Bendall (1976) reported a high correlation between ATP and lactate, which can be quite precisely reinterpreted in terms of pH value for lamb muscle as shown below:

a) *Longissmus dorsi muscles*
   Lactic acid (μ Mol g^{-1}) =62.5 (7.14 − pH)
   (S.E. of slope = ± 3.8; r= −0.951)

b) *Biceps femoris muscles*
   Lactic acid (μ Mol g^{-1}) = 55.2 (7.18 − pH)
   (S.E. of slope = ± 2.0; r = −0.98)

Intra-cellular Proteins

Sarcoplasmic Proteins

Based on histological observation, George et al. (1980) concluded that slow cooling of electrically stimulated carcasses causes denaturation and precipitation of sarcoplasmic proteins onto the myofibrils. If such a deposition occurs, it should be reflected in decreased solubility of sarcoplasmic proteins as reported in the case of PSE muscles (Briskey, 1964). However, our results on fast and slow cooling of electrically stimulated lamb carcasses did not show any difference in the solubility (Rashid et al., 1982b) nor in the SDS-electrophoretic profile pattern of sarcoplasmic proteins (Figure 1) as compared to those of non-stimulated carcasses (Asghar et al., 1981).

It should be emphasized that the glycolytic enzymes, which are sarcoplasmic originated, comprised about 20% of the muscle proteins (Czok and Bucher, 1960; Asghar and Pearson, 1980). Thus, it is important to realize the functional implications of denaturation and precipitation of sarcoplasmic proteins as assumed by some investigators. In this case one should expect a decrease in the glycolytic rate in muscles from electrically stimulated carcasses and an increase in toughness, much in the same manner as was reported in the case of PSE muscle (Bendall and Wismer-Pederson, 1962). In fact, the muscles in electrically stimulated carcasses always have a higher glycolytic rate (section II. A) and are generally more tender than those from unstimulated carcasses (section VIII. D).

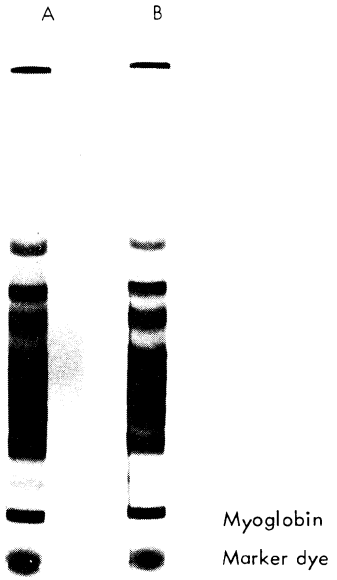


Figure 1. SDS-polyacrlamide gel electrophoretic profile of sarcoplasmic proteins of lamb *L. dorsi* muscle, excised 24 hours post-mortem from electrically stimulated (A) and unstimulated (B) carcasses. The proteins were extracted in deionized distilled water. From Asghar et al., 1981.

There is, however, a difference in the significance of reversible binding of some glycolytic enzymes to myofibril proteins during electrical stimulation. For example, it has been shown that aldolase (Starlinger, 1967), glyceraldehyde 3-phosphate dehydrogenase phosphofructokinase and pyruvate kinase (to some extent glucose 6-phosphate isomerase, phosphoglycerate kinase and lactate dehydrogenase) bound with actin filaments especially at the level of I-bands in electrically stimulated muscles (Clarke et al., 1980). Different explanations have been extended for those observations. Arnold et al. (1971) . . . suggested that low pH might be a factor, but Clarke et al. (1980) did not think it an adequate assumption. According to Clark a specific effect of metabolite concentration is also important. It is, however, not clear whether the increased enzyme binding was the cause or effect of the increased glycolytic rate in the electrically stimulated muscles, although kinetic action of several glycolytic enzymes is known to modify binding to particulate material (Walsh et al., 1977). The possibility that electrical stimulation activates the enzyme systems per se in muscle has not been ascertained.

Myofibrillar Protein

The ordered structure of myofibrils is now known to be composed of several proteins. The thick filaments contain myosin (light and heavy chains), M-protein, and C-protein, whereas actin, tropomyosin, troponin T, I, and C, and α-actinin are present in the thin actin filaments. The α-actinin and desmin or connectin are associated with the Z-disks. More detailed information on the muscle proteins is available in several recent reviews (Harrington, 1979; Goldman et al., 1979; Mani et al., 1980; Asghar and Pearson, 1980; Greaser, 1981).

In an earlier study, Seideman et al. (1979) reported that the amount of sarcoplasmic protein (soluble in 0.03 M PO_4^{-3} buffer), M-protein, C-protein and α-actinin in hot-boned (2-week conditioned) L. dorsi muscle from electrically stimulated sides was significantly higher than that from non-stimulated, cold-boned (2-week conditioned) muscle. The total amount of myofibrillar proteins, (soluble in 1.1 M KI-KPO_4 buffer), myosin, actin, troponin T, tropomyosin and myosin light chain contents were not different between the treatments.

It cannot be ascertained from the study of Seidman et al. (1979) whether or not the reported effects were the result of electrical stimulation. In this study the L. dorsi muscles from electrically stimulated sides were removed about 1 hour post-mortem, whereas those from unstimulated sides were excised 24 hr post-mortem. Thus, the muscles must have experienced different cooling rates, and the reported changes seem to be the result of third order interaction involving electrical stimulation, cooling rate and aging period. In another study, McKeith et al. (1980a) found no measurable difference in the solubility of myofibrillar protein of muscle from electrically stimulated and unstimulated steer carcasses.

Studies on electrical stimulation of lamb carcasses also revealed no significant effect on the solubility of different myofibrillar protein fractions either soluble in 0.3 M KCl or in 0.6 M KI-PO_4 buffer (Rashid et al., 1982a, b). There was also no apparent difference in the SDS-oylacrylamide gel electrophoretic profile of the myofibrillar proteins, extracted within 24 hours post-mortem, with 0.3 M NaCl in 0.1 M PO_4^{-3} buffer, 0.1 M tris buffer or with 0.06 M NaI-PO_4 buffer from electrically stimulated and unstimulated carcasses as shown in Figure 2. However, McKeith et al. (1980) noted a significant influence on the proportion of troponin-T in stimulated muscles from cows but none in the case of steers. Whether this effect was associated with sex or age difference was not ascertained.

By using the SDS-polyarylamide gel electrophoretic technique many investigators have claimed to identify the changes in myofibrillar proteins, especially troponin-T

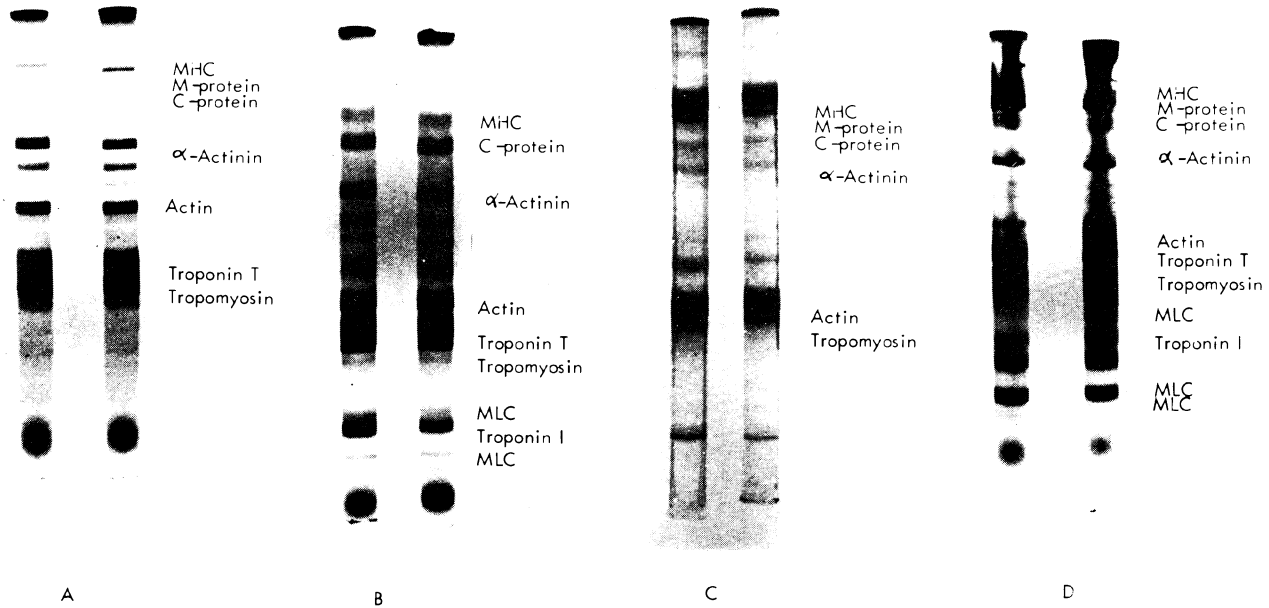


Figure 2. SDS-polyacrylamide gel electrophoretic profiles of myofibrillar proteins of lamb *L. dorsi* muscle from electrically stimulated and unstimulated (control) carcasses. The left side gel of each pairs represents electrically stimulated muscle and the other represent control. A) Myofibrillar proteins were extracted with 0.3 M NaCl-phosphate buffer; B) Myofibrillar proteins were extracted with 0.1 M tris solution; C) Myofibrillar proteins were extracted with 0.6 M Nal-phosphate buffer; D) Myofibrillar proteins were extracted with 6.0 M urea. All extraction were made 24 hours post-mortem. MHC, myosin heavy chain, MLC, myosin light chain. From Asghar et al., 1981.

during post-mortem aging of meat (Cheng and Parrish, 1978; Penny and Dransfield, 1979; Penny, 1980). However, the results are not always consistent (Parrish et al., 1981). Other workers found little relationship between changes in troponin-T and tenderness (George et al., 1980). Perhaps a refinement in the extraction of the various myofibrillar proteins, and/or in the SDS-polyacrylamide gel electrophoresis may resolve the problems. In this regard two dimensional SDS-polyacrylamide gel electrophoresis may be helpful in identifying the changes especially in those proteins which constitute the z-disks.

Extra-Cellular Proteins

Most of the studies on electrical stimulation of carcasses have so far been centered on the intra-cellular proteins of muscle. Very limited information is available on the influence of electrical stimulation of carcasses on the extra-cellular muscle proteins.

Collagen Solubility

Judge et al., (1980) investigated the effect of electrical stimulation on muscle protein from cattle managed under different nutritional regimens. They did not find any difference in the solubility characteristics of the perimysial collagen as estimated by the hydroxyproline measurement. A recent study by Rashid et al. (1982b) also showed no change in the solubility characteristics of L. dorsi's endomysial residue from electrically stimulated lamb carcasses as compared to those from the control. In this case solubility was examined with 0.1 M lactic acid and the protein content in the extract was estimated by the biuret method.

Collagen Cross-linkage

As mentioned above, Judge et al. (1980) found no increase in the solubility of the perimysial collagen from electrically stimulated muscle. However, their data using differential scanning colorimetery showed a significant decrease (0.6°C) in the thermal stability of the perimysial collagen of the L. dorsi muscle from electrically stimulated carcasses as compared to that from the control. Apparently this observation is at variance with no change in solubility characteristics of collagen from electrically stimulated carcasses. The disparity may be due to the estimation of hydroxyproline which is not precise enough to detect a small difference.

The swelling magnitude of a polymer such as collagen has been shown to depend primarily on the number of cross-links and the interaction constant between polymer and solvent according to the equation of Flory and Rehner (1943):

$$v = \left[\frac{Z \epsilon V}{M(1-2\mu)} \right] \quad 3/5$$

where v is the volume-fraction of the polymer at equilibrium; V, the molar volume of solvent; ϵ, the density of pure polymer; μ, an interaction constant between solvent and polymer; and M, the average molecular weight of the chains between cross-links. Thus in a particular solvent system, the extent of cross-linkages would determine the magnitude of swelling of a collagen. On the basis of this assumption, the swelling factor has been used as an indicator of change in the collagen cross-linkages (Eastor and Courts, 1963; Asghar and Yeates, 1979; Asghar et al., 1981). However, Rashid et al. (1982b) showed that a change in connective cross-linkages, if any, resulting from electrical stimulation of carcasses was not large enough to be detected by the swelling factor.

Effect on the Biophysics of Muscle

Muscle Tension

Physiologically two types of contractions have been recognized: isotonic and isometric (Zierler, 1973). If a muscle is allowed to contract against some fixed resistance, the distance between tendons decreases. It is called an isotonic contraction, that is, the shortening of muscle against a fixed load. On the other hand, if the muscle ends are fixed so that the muscle cannot change its overall length, but only exerts a force (i.e. pull against the fixed position) on stimulation, it is known as isometric contraction. This type of contraction mainly prevails in muscles of the intact carcass.

If a muscle is stimulated repetitively at a very low frequency, it responds with one twitch per stimulus. As the stimulation frequency is increased, the tension developed from one twitch adds to the tension developed by the preceding twitch and the mechanical response increases until it reaches a plateau (tetanus) which can be maintained for a finite period under proper environmental conditions. The mechanical response of muscle to electrical stimulation is expressed in terms of tension, which is a measure of the proportion of fibers activated by the electrical current (Chrystall and Devine, 1978). The area under the tension-time curve is a measure of developed tension and its stability (Figure 3). The decrease in tension, represented by the time it takes to fall below 50% of peak tension, is a measure of the decrease in stimulation effectiveness due to a fatigue and polarization effect.

By recording the tension-time curve during electrical stimulation, Chrystalland Devine (1978) concluded that pulse frequency exerted a greater effect on the tension-time curve than other electrical parameters. They noted a marked decrease in time for the peak tension to fall by 50% with an increase in frequency, although the peak tension

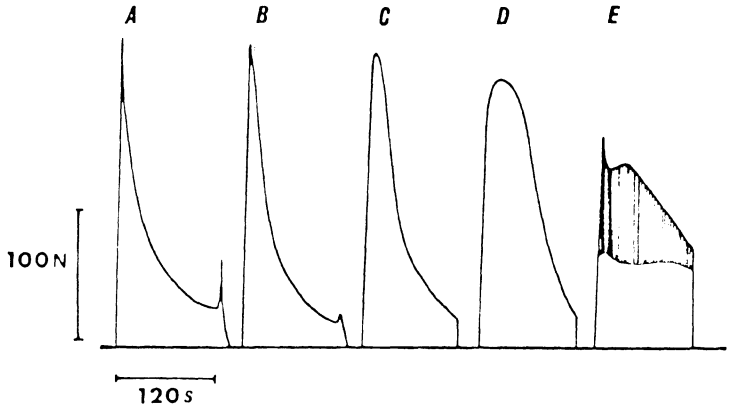


Figure 3. Tension-time curves of muscle strips (size, about 0.001 m^2), stimulated with 200 peak volt of direct current for 3 minutes at different pulse rates. A) 100 pulses per second, B) 50 pulses per second, C) 25 pulses per second, D) 12.5 pulses per second. The shaded area under curve E represents the range of repeated pulsation evident at this frequency. From Chrystall and Devine, 1978. Courtesy of Applied Science Publishers, Ltd. England, U.K.

slightly increased. Further, they found 5 to 16 pulses sec^{-1} equally effective in accelerating the rate of glycolysis. However, higher pulse values should be preferred for mechanical reasoning since much less muscle pulsation occurs. This supports the report of Bendall et al. (1976) that frequencies below 10 Hz caused the muscles to contract and relax in sequence with the sine wave, whereas frequencies above 10 Hz resulted in complete fusion of separate twitches into a tetanus. In the latter case, the carcass remained rigid in a contracted state. They considered twitching of the carcass undesirable, and hence did not recommend a frequency below tetanus fusion.

Physiology of Muscle Contraction

A brief account of the physiological events leading to muscle contraction and tension development is provided to formulate an appreciation for the status of the present knowledge. Extensive information is available on the mechanics of muscle contraction (Huxley, 1971, 1974; Weber and Murray, 1973; White and Thorson, 1973; Zierler, 1973; Cohen, 1975; MacLennan and Holland, 1975; Squire, 1975; Ackerman et al., 1979). Some of the generally accepted views on the mechanism of muscle contraction are herein summarized.

When a muscle is stimulated by electrical impulse, cold, heat, or mechanical pressure, the surface potential of the axon of the nerve fiber changes to action potential (it is also called spike potential—Ackerman et al., 1979). Depending on the diameter of the nerve fiber, the spike potential travels at a characteristic velocity along the axon in the direction of the muscle fiber, owing to the nature of synapses which provide a connection between neurons (the functional units of nervous system). As the muscle nerve fibers lack myelin which acts as an electrical insulator between nerve fibers (Ackerman et al., 1979), a spike potential along one axon may stimulate its neighbor. When the spike potential reaches the myoneural junction (neuromuscular junction) in vertebrates, a transmitter such as acetylcholine (ACh) is released from small vesicles of the nerve ending (Nachmansohn, 1973; Hoar, 1975; MacLennan and Holland, 1975).

Acetylcholine diffuses across the synapses and is absorbed by the motor end plate, where it alters the ionic permeability of the membrane to Na^+ ions. The action potential from the depolarized motor end plate is then transmitted to the sarcolemma, which, in turn, also becomes permeable to the Na^+ ions. (Zierler, 1973; Ackerman et al., 1979). The movement of Na^+ ions from the interstitial fluid across the sarcolemma changes the sign of electrical potential from negative to positive (with respect to extra-cellular space fluid).

After passing over the sarcolemma, the action potential travels along each transverse tubular system (T-system) through each Z-disk. Finally, the excitation signal reaches the longitudinal tubules (L-system) of the sarcoplasmic reticulum either by action potential propagated from the sarcolemma or by electrotonus (inward spread of current). This results in the release of bound Ca^{++} ions from the terminal cisteinae of the L-system of the sarcoplasmic reticulum (Wingard, 1968; Inesi and Malan, 1976; Endo, 1977; Hasselbach, 1977; Frank, 1980). Thus a concentration of free Ca^{++} ions in the sarcoplasm increases from about 10^{-7} Mole/l (initial value) to 10^{-5} Mole/l. Free Ca^{++} ions diffuse into the myofibril spaces to initiate contraction (Cohen, 1975; Endo, 1977; Ebashi et al., 1969). Some of the physiological events described above are depicted in Figure 4.

The ACh is ultimately catabolized by acetylcholinesterase located in the motor and plate (Ackerman et al., 1979). The mechanism by which Ca^{++} ions regulate the actin-myosin interaction perhaps varies for different types of muscles (Adelstein and Eisenberg, 1980). In the case of the vertebrate skeletal muscle troponin C binds the Ca^{++} ion to relieve the actin-activated ATPase activity which is inhibited by troponin-

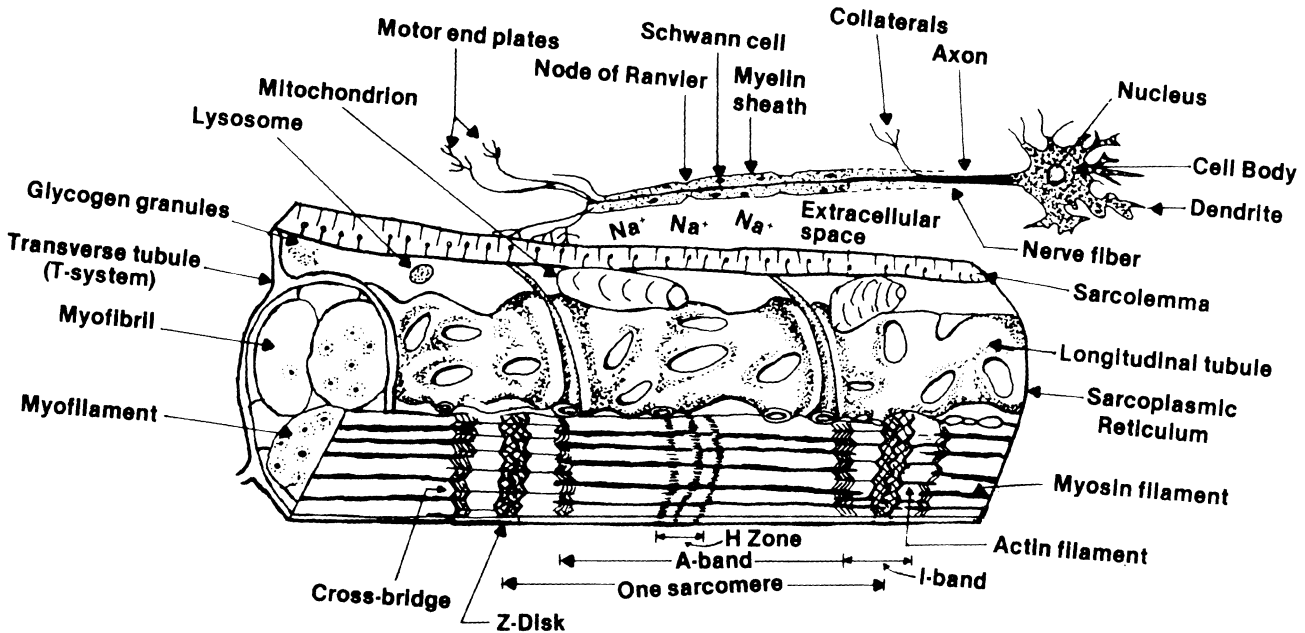


Figure 4. Diagramatic representation of muscle fibers at the cellular level, along with other organelles, and the nervous system associated with the action potential during muscle contraction.

tropomyosin complex in the resting state (Ebashi and Endo, 1968; Ebashi et al., 1969; Ebashi and Nonomura, 1973; Bremel and Weber, 1972). This results in activation of actin, and hence a reaction between actin and myosin is initiated. Consequently, a tension is developed along the long axis of the fibers and they may shorten depending on the external conditions. It may, however, be pointed out that some of these events are only speculative and need to be experimentally examined. Moreover, the stimulation of muscle at high voltages probably directly depolarizes the muscle membrane (section X. A).

If the muscle is not stimulated at a very high frequency, the membrane quickly repolarizes and the contraction events are reverted to the resting state. The Ca^{++} ions are recaptured from the sarcoplasm by the sarcotubules and the excess Na^{+} ions are pumped out. Some of the reactions of the contracting-relaxing process require energy expenditure. The primary source of energy in muscle is the free enthalpy of ATP splitting into ADP and inorganic phosphate (Szent-Gyorgyi, 1951; Curtin and Davies, 1973; Bagshaw and Trentham, 1974). Intensive efforts have been made to understand the mechanism of ATP metabolism at the molecular level during muscle contraction. The section on biochemical models provides a resume of the latest developments.

Biochemical Models of Contraction

During the past decade much progress has been made in understanding the biochemistry of muscle contraction, and the rate limiting reaction in the contracting-relaxing cycle. As a result, several biochemical models were proposed to relate the mechanism of muscle contraction with actonyosin ATPase, and to show whether ATP hydrolysis occurs with or without disassociation of the actomyosin complex accompanied by conformational changes in myosin. Some of the important ones are Eisenberg and Moos (1968) model, Lymn and Taylor (1971) model, refractory state model (Eisenberg and Kielley, 1972) and modified refractory state model (Stein et al., 1979). Adelstein and Eisenberg (1980) have provided a detailed description of these models. The important features of these models are shown in Figure 5. Eisenberg and Greene (1980) recently proposed a 'cross-bridge action model', which is based on the biochemical model of Stein et al. (1979), whereas Tregear and Marston (1979) presented a logical modified form of the cross-bridge model which is based on the latest biochemical, structural, and mechanical evidences.

Despite all of these advances, it is not yet clear as to how the kinetic mechanism of cyclic contraction is regulated. There also exist some disagreements as to which reaction in the ATPase cycle is rate limiting, and whether or not the actin and myosin must disassociate each time when an ATP molecule is catabolized. Apart from the interaction of the Ca^{++} ions with troponin C, some investigators believe in a Ca^{++}-dependent conformational change within the thick filament during contraction (Huxley, 1971; Haselgrove, 1975). Detailed descriptions of the biochemistry of muscle contractions are available in many excellent reviews (Korn, 1978; Lymn, 1979; Perry, 1979; Taylor, 1979; Harrington, 1979, 1981; Adelstein and Eisenberg, 1980).

Mechanical Models of Contraction

Huxley in 1957 first proposed a model, commonly known as the 'sliding filament' to explain the mechanics of the contraction process. Further progress on the structure and biochemistry of the myofibrillar proteins in muscle resulted in many refinements of Huxley's original model for coupling the new information on chemical and mechanical changes at the molecular level (Davies, 1963; Huxley, 1971; Harrington, 1971; Huxley and Simmons, 1971; Huxley, 1974; Hill, 1975; Huxley et al., 1981). In the revised models, sometimes referred to as the 'sliding filament-moving bridge' or 'cross-bridge'

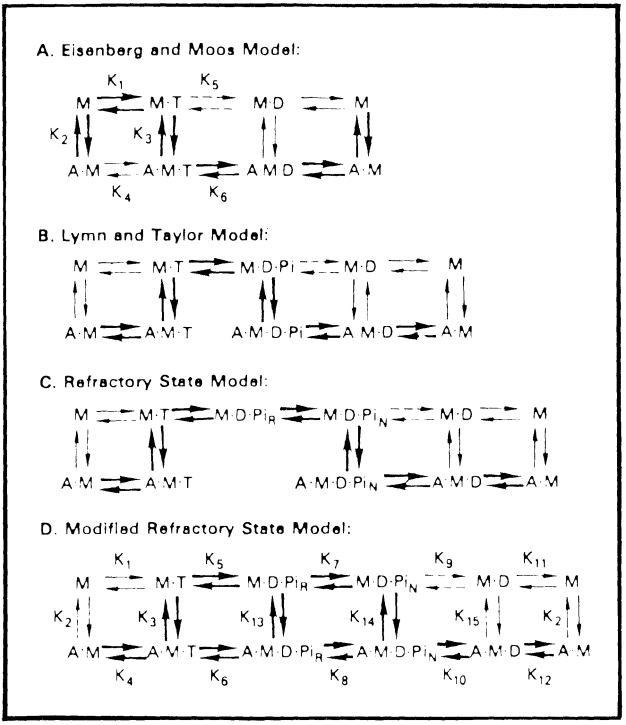


Figure 5. Biochemical models depicting the kinetic schemes for the hydrolysis of ATP by myosin and actomyosin. The heavy solid lines show the optimal pathway for the actomyosin ATPase cycle in each scheme. The dashed arrows indicate the rate-limiting steps in the myosin ATPase cycles in the absence of actin. M, a single myosin cross-bridge head (S-1 in vitro); A, an actin monomer in the F-actin filament; T, ATP; D, ADP; the R and N subscripts indicate the refractory and nonrefractory state, respectively. It is likely that all of the steps involving the binding or release of ATP, ADP, and Pi in these schemes are, in fact, at least two-step processes consisting of formation of a collision intermediate followed by a conformational changes (Bagshaw et al. 1974). In the present schemes, however, all of these two-step processes have been shown as single steps for simplicity. Finally, in one version of the original refractory state model, state M• D• Pi_R was shown as transforming to state M• D• Pi_N in the actoymyosin ATPase cycle, while in the myosin ATPase cycle state M• D• Pi_R transformed directly to state M• D, as originally proposed by Bagshaw et al. (1974). In the present scheme, for uniformity, no branches have been shown; state M• D• Pi_N occurs in both the myosin and actomyosin ATPase cycles. In all reactions where a ligand (A, T, D, Pi) binds to M, the equilibrium constants, $K_i = k_i/k_{-i}$, are defined as association constants. The other equilibrium constants are defined with product to the right of reactant. From Adelstein and Eisenberg, 1980. Courtesy of Palo Alto, California.

model, trypsin-sensitive and papain-sensitive regions of the myosin molecule were assumed to act as a flexible hinge for movement of the 'head of the myosin filament for making contact with actin. According to these models, muscle contraction consisted of the cyclic attachment and detachment of the globular head portion of myosin with the actin filaments, which resulted in the sliding of the filaments past each other.

While a majority of the investigators have accepted Huxley's model with certain modifications, some workers (Carlsen et al., 1961; Noble and Pollack, 1977) criticized it. McClare (1971, 1972) also pointed out the limitations of the cross-bridge model on the basis of thermodynamics, and suggested a molecular model to explain the mechanism of muscle contraction. In the model of Tsong et al. (1979), the helix-coil transformation of a part of the folded S-2 fraction of the myosin molecule was assumed to play a role in the cross-bridge cycle. The salient features of some popular models explaining the mechanism of muscle contraction are depicted in Figure 6. Further

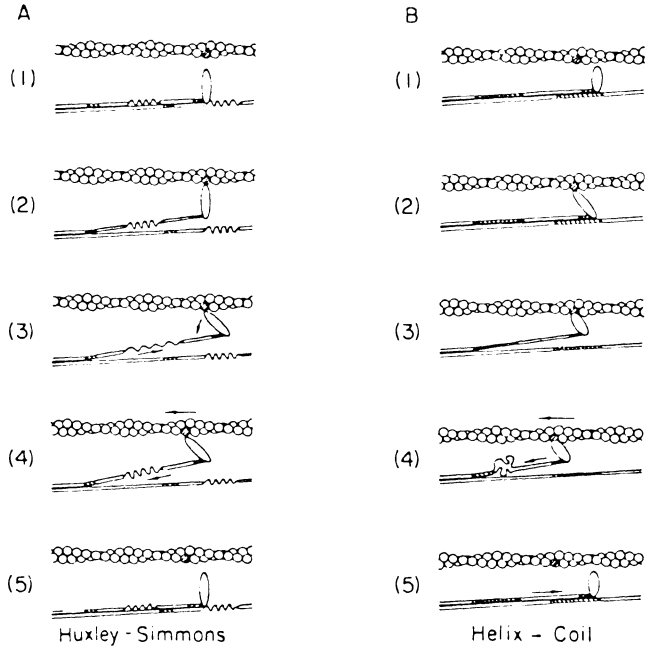


Figure 6. The salient features of fundamental steps in the Huxley-Simmond model and the helix-coil model. A. Huxley-Simmons Model: 1) Resting state; 2) Attachment of myosin cross-bridge head (S-1) to actin filament; 3) Rotation of S-1 while it is attached to the actin filament and simultaneous stretching of the spring-like elastic component in subfraction (S-2); 4) Power stroke resulting from retraction of elastic component 5) Return of cross bridge to resting state. B. Helix-Coil Model: 1) Resting state; 2) S-1 swivels to attach to actin filament; 3) Release of S-2 from thick filament surface; 4) Power stroke resulting from helix→coil transition in S-2; 5) Return of cross bridge to resting state. Long hatched region in (B) represents sticky "hinge" region of S-2 (Sutoh et al., 1978). From Tsong et al., 1979. Courtesy of Natl. Acad. Sci. U.S.A.

research is needed to ascertain which model correctly explains the contraction mechanism in muscle.

A few alternative hypotheses, mostly based on the electrostatic charge of the actomyosin filaments, have also been extended by different researchers (Spencer and Morthington, 1960; Elliott et al., 1970), but they received little experimental support from other studies (Marston et al., 1976, 1979).

Cold Shortening

When a carcass is subjected to chilling temperatures (below 0°C) immediately after dressing, muscles near the surface may cool below 15°C, while the ATP content is still high enough to cause fiber contraction. In such an environment, certain sarcomeres of a muscle fiber will contract up to 40% of their resting length (Marsh and Leet, 1966). This phenomenon, called 'cold shortening' causes considerable toughening of meat (Davey et al., 1976b).

Recently, Rashid et al. (1982b) studied the effectiveness of electrical stimulation in reducing the incidence of cold shortening. The data in Table 1 show that the excised semitendinosus muscle from electrically stimulated lamb carcass sides on rapid chilling shortens about 7.3% less in 24 hours than muscles from unstimulated rapid chilled sides. On the other hand, slow chilling (conditioning the excised muscles at 16° for 5 hours) of the semitendinosus muscle from electrically stimulated carcass sides caused only 4.5% shortening in 24 hours as compared with no change at 5 hours post-mortem.

Ca^{++} ion-induced shortening of the semitendinosus muscle (Figure 7) showed a more pronounced difference between electrically stimulated and control sides (Rashid et al., 1982a). The shortening in electrically stimulated muscle was about 13.2% less than in the control (32.7% *v* 19.5%). Since the muscles from electrically stimulated carcasses also cold shortened 19.5% by Ca^{++} ion injection, it suggested that stimulated carcasses still had some energy supply to support shortening. Hence, a risk of meat toughening will be involved if the carcass is hot-boned immediately after electrical stimulation and subjected to chilling.

Table 1. Effect of electrically stimulating and mode of chilling on the extent of fiber shortening in semitendinosus muscle strips from lamb carcasses

Treatment	Shortening*		Difference
	5 hours postmortem	24 hours postmortem	%
Stimulated + Slow chilled[a]	9.8	10.0	0.2
Stimulated + Rapid chilled[b]	12.3	13.6	1.3
Unstimulated + Slow chilled[a]	9.8	14.5	4.7
Unstimulated + Rapid chilled[b]	15.8	20.9	5.1

a) strips were conditioned 16°C for 5 hours; then they were subjected to 2°C for 19 hours.
b) strips were subjected to 2°C, immediately after being stimulated for 24 hours.

* The shortening at 5 hours and subsequent 19 hours were measured on the same strips. Each figure represents the average from 12 observations (Rashid et al., 1981 Personal Communication).

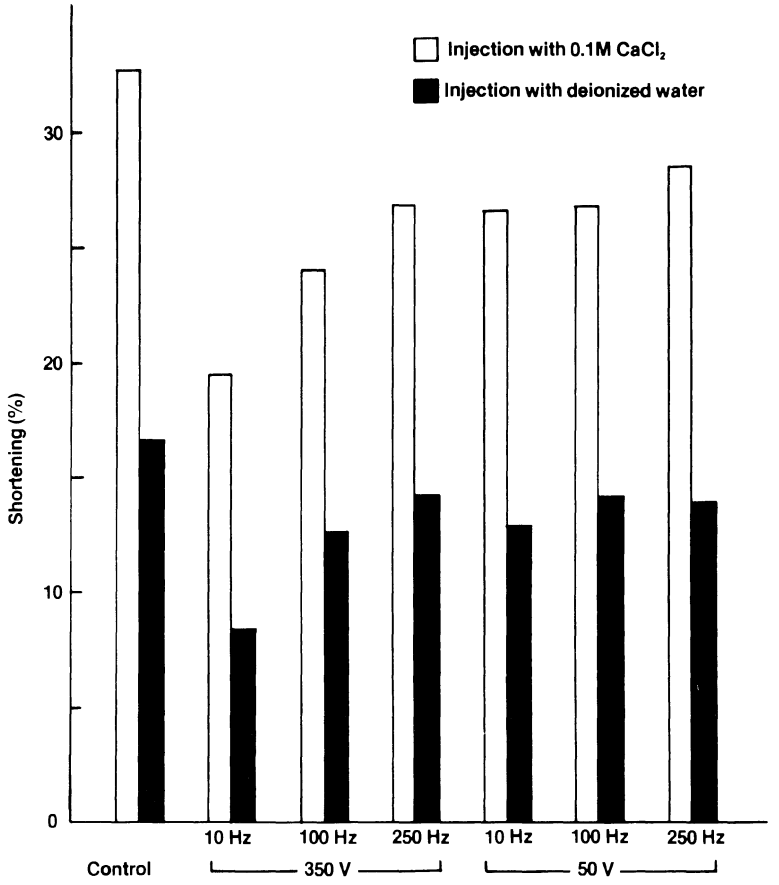


Figure 7. The effect of electrical stimulation at different voltages (V) and frequencies (Hz) of lamb carcasses on the extent of shortening of semitendenosus muscle strips. The muscle was exised soon after electrical stimulation and the strips were injected with the appropriate solution and subjected to 2°C for 24 hours. Courtesy of Rashid et al. (1981).

Muscle Temperature

Fenn (1923) stated that upon stimulation muscle shortens and does work. During this period an extra amount of energy is mobilized. This phenomenon is now known as the Fenn effect (Zierler, 1973). Hill (1965) provided a quantitative treatment of the Fenn observation, whereas Mommaerts (1969) has treated at length the issue of muscle heat and its genesis. Muscles produce heat (about 2.4×10^3 cal/gm/min) even in the basal condition. The total initial heat produced in response to an isometric twitch is called activation heat, while in an isometric tetanus, the total heat is designated as

maintenance heat which is cumulative activation heat produced by activation of muscle with each stimulus (Zierler, 1973).

Some heat is also produced by electrical current in the carcasses, the extent of which can be computed by the following equation (Marcus, 1964):

$$\text{Heat} = 0.24 \times I^2 \times R \times T$$

where heat is in calories, I is the current in amperes, R is the resistance in ohms and T is the time in seconds. According to Bendall (1980), although heat develops during a two minute stimulation period at 680 volts (peak is about 13 kj), it is distributed over the whole carcass mass and the mean temperature rise is probably less than 0.04°C. The temperature may be relatively higher close to the electrode. On the contrary, Carse (1973) reported a drop in carcass temperature from 40°C to 37°C during electrical stimulation for 30 minutes. This discounts the possibility that fast glycolysis causes a rise in muscle temperature due to passage of electricity.

Water-Holding Capacity (WHC)

Electrical stimulation did not effect the amount of free water (drip) in the package during retail display (Morgan, 1979; Jeremiah and Martin, 1980). Although rapid pH fall in muscle of some breeds of swine was associated with low water-holding capacity of meat (Briskey, 1964). Possibly, rapid conditioning changes in muscles from electrically stimulated carcasses increase the osmotic pressure in the intra-cellular space to offset the decrease in WHC of the muscle proteins (Bouton et al., 1958). Bendall (1980) also remarked that there are no appreciable differences between muscles from control and electrically stimulated sides either in the so-called 'bag-drip' or in the extra-cellular space. However, the bag-drip of the deep muscles of the round from the stimulated carcasses tended to be slightly higher than that from the control.

According to Honikel et al. (1981a) neither shortening nor the development of rigor was due only to pH fall, and independent of temperature. These conclusions seemed inconsistent with the earlier contention of Hamm (1975) according to which 2/3 of the decrease in WHC of post-mortem meat was associated with the reduction in ATP content. However, in another report, Honikel et al.(1981b) demonstrated that as much as 2/3 of the decrease in WHC of salted meat occurred due to rigor development and the remaining 1/3 was caused by the decrease in pH.

Effect on Micro- and Ultra-Structures of Muscle

Sarcomere Length

A number of studies revealed no difference in the sarcomere length of muscle fibers from electrically stimulated and non-stimulated beef (Smith et al., 1977; Savell et al., 1979; Demeyer and Vandenriessche, 1980; Elgasim et al., 1981; Salm et al., 1981), calves (Smith et al., 1979) and lamb carcasses (Savell et al., 1979). These observations led them to believe that increased muscle tenderness of electrically stimulated carcasses is not simply related to cold shortening. However, muscles from stimulated sides of goat carcasses were reported to have longer sarcomeres than those from the control sides (McKeith et al., 1979). This difference was explained on the assumption that inadequate subcutaneous fat cover on the goat carcass allowed more rapid cooling of the unstimulated side when muscle pH and ATP content are high enough to induce cold-shortening.

In an earlier study, Bouton et al. (1978) found no significance difference in the sarcomere length of muscle fibers from stimulated and unstimulated carcass sides.

However, their later observations did show markedly longer sarcomeres in the L. dorsi, biceps femoris, vastus lateralis, and gluteus medius muscles from electrically stimulated sides than from the control (Bouton et al., 1980a). Nichols and Cross (1980) and Whiting et al. (1981) also noted longer sarcomeres in fibers from muscles which were excised from stimulated carcasses and immediately frozen, than similarly treated muscles from unstimulated sides. The L. dorsi muscle from electrically stimulated intact carcasses was found to have longer sarcomeres than did L. dorsi muscle from electrically stimulated sides of carcasses from steers. The latter, however, did not differ from the control sides (McKeith et al., 1980a).

The disparity in results on sarcomere length among different investigators can be ascribed to many factors that were probably overlooked by most investigators. For example, the data in Table 1 indicates the extent of shortening that could be expected in the case of hot-boned meat. However, the situation with intact muscles in a carcass is quite different. In that case, each muscle maintained a particular overall length in the suspending carcass due to attachment of the terminal ends of the muscle to the skeletal system through tendons. The study by Marsh and Leet (1966) suggested that the cold shortening phenomenon in a muscle whose length has been fixed by clipping the ends, is localized in certain regions of the muscle, whereas overall length did not change during the chilling operation. If so, then a decrease in sarcomere length can be identified only in a particular region of muscle where cold shortening has occurred, while other regions probably would not show any change in sarcomere length.

Another important variable that may have a bearing on the variation in sarcomere length is the post-mortem time at which muscle samples are removed from the carcass. For example, muscles removed from electrically stimulated sides at 1 to 2 hours post-mortem were found to have markedly longer sarcomeres than those from unstimulated sides. However, muscles removed at 22 hours post-mortem from stimulated and control sides exhibit no difference in sarcomere length (Bouton et al., 1980a). The recent studies by Rashid et al. (1982b) clearly demonstrated that the semitendinosus muscle removed from electrically stimulated lamb carcasses and conditioned for 5 hours at 16°C before subjecting them to chilling temperature, shortened less than those which were immediately chilled after excision from the stimulated carcasses.

Honikel et al. (1981b) also reported an interesting relationship between sarcomere length and the temperatures at which meat is conditioned. They observed little change in sarcomere length on holding meat at temperatures between 8 to 16°C; beyond these limits sarcomeres decrease sharply with a change in temperature. Honikel et al. (1981b) also reported that a 0.5°C the initial pH drop is fast in the meat accompanied by cold shortening which occurred before the onset of rigor, followed by little rigor shortening. Above 16°C, rigor shortening increased with rising temperature without pre-rigor contraction.

Sarcomere length is generally measured either by microscopic technique or by laser procedure. Some workers have found a good correlation by these methods; others reported a poor relationship. George et al. (1980) obtained the measurements on sarcomere length by these two methods. The data collected by the laser method did not show a difference in sarcomere length of fibers between electrically stimulated and unstimulated carcasses; however, microscopic measurements showed significantly longer sarcomeres in muscle from stimulated than from unstimulated carcasses. Cross et al. (1981) have shown that for measuring the sarcomere length with 99% precision, 34 measurements were required in the case of the laser method. On the other hand, 45 and 66 estimations were needed to achieve the same precision by oil immersion microscopic technique using filar micrometer and shearicon size analyser methods respectively.

Ultra-structure

Several workers have examined the ultra-structural changes in muscle induced by electrical stimulation. The presence of 'contraction bands' was reported in muscles from electrically stimulated carcass sides (Savell et al., 1978a; McKeith et al., 1980a) to be similar to those muscles which eperienced rapid post-mortem glycolysis (Dutson et al., 1974; Abbott et al., 1977). However, muscles from intact electrically stimulated carcasses did not show such contraction bands (McKeith et al., 1980b). The contraction bands were assumed to be formed due to extreme shortening of the sarcomeres (Savell et al., 1978a). When muscles are restrained in the carcass, areas of contraction will likely be accompanied by areas of extended sarcomeres. Therefore, an average sarcomere length may show no overall difference.

Recently, Will et al. (1980) provided a detailed study on the ultra-structural changes in different muscles from electrically stimulated beef carcasses. Some of their findings agreed with those of Savell et al. (1978a). However, they observed contraction bands only in the L. dorsi and supraspinatus muscles, but none in the psoas major and semitendinosus. The electron micrographs in Figure 8 show the changes in muscle ultra-structure due to electrical stimulation of the carcass.

On the basis of histological evidence, George et al. (1980) also reported the presence of irregular bands, similar to contraction bands, on muscle fibers from slowly cooled, electrically stimulated carcasses. They assumed that these bands originated from the precipitation of the sarcoplasmic proteins onto the surface of the myofibrils, much in the same manner as was reported in the case of PSE muscle (Bendall and Wismer-Pederson, 1962).

The fact is that the contraction bands have been found only in the case of electrically stimulated carcasses, but also under several pathological conditions. For example, contraction bands in muscle structure have been reported in the case of malignant hyperthermia (Britt and Kalow, 1970; Isaacs et al., 1973; Fenoglio and Irey, 1977), ischemia, cardiomyopathy, catecholamine infusion (Adomian et al., 1976; 1978) and hemorrhagic shock (Leet et al., 1975). Fenoglio and Irey (1977) believed that these bands resulted from the degeneration of myofibrillar proteins per se. In the presence of all these evidences more studies are required to trace the etiology of the contraction bands.

It may also be emphasized that one of the important factors which is generally not taken into account in electron microscopic study is the size of the sample. This especially carries much significance in the case of a muscle which is not homogenous in texture and contains a structured pattern of variation from center to periphery and to terminal ends. Thus, a sample only a few microns in dimension, obtained from a particular location of a muscle can hardly be regarded as representative of the overall muscle structure by any logic, disregarding any statistical consideration. A slight deviation in the knife angle with respect to the longitudinal plane of the fibers during sectioning would show quite different muscle structural features from the normal. Hence, observations made on ultra-microscopic sections should be interpreted with reservation unless they are substantiated by other data on an overall representative muscle sample.

Sarcomere Integrity

Some researchers have indicated that electrical stimulation caused a considerable degree of physical disruption of the myofibrils (Savell et al., 1978a; McKeith et al., 1980b; Will et al., 1980). Contrary to this, another study by Savell et al. (1979) did not show a significant difference in the 'fragmentation index' of muscle from electrically

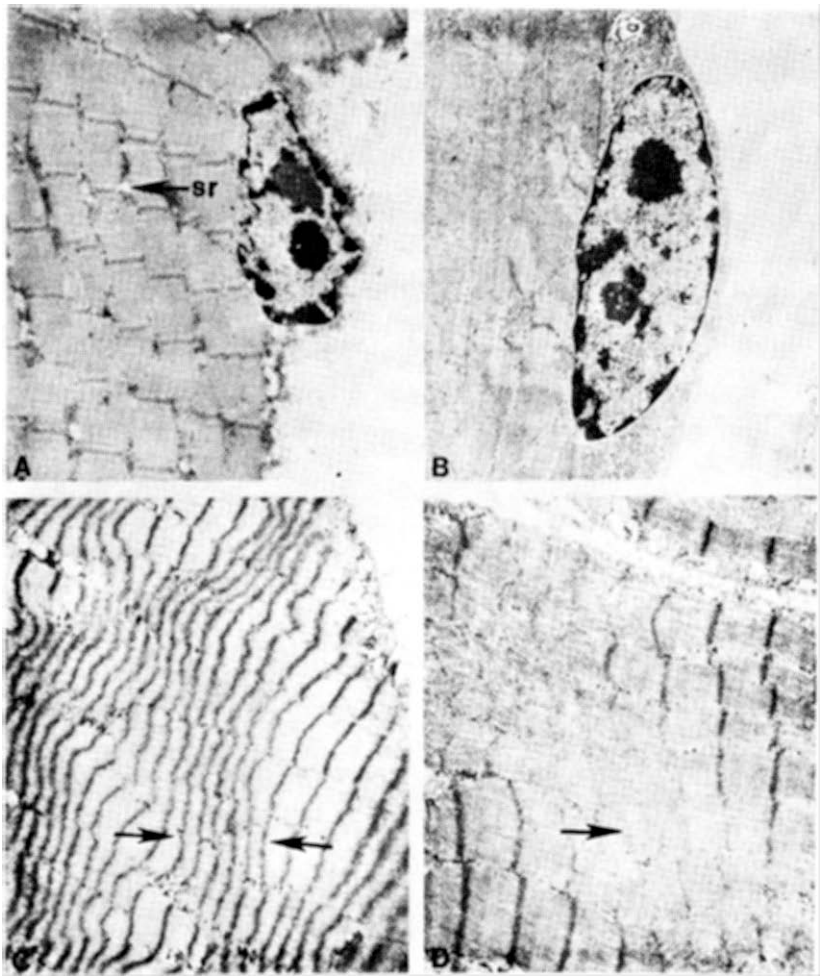


Figure 8. Electron micrographs of *L. dorsi* muscle from electrically stimulated and unstimulated (control) lamb carcasses 1 hour post-mortem. A) Nucleus in control muscle appears normal but sarcoplasmic reticulum (Sr) is slightly dilated (magnification × 9,500). B) Electrically stimulated muscle shows no change in nuclear morphology (magnification × 8,500). C) Electrically stimulated muscle showing contraction bands (arrows) and stretched areas on either side of a band (magnification × 8,000). D) Electrically stimulated muscle showing the disruption of saromeres (arrows) (magnification × 11,000). From Will et al., 1980. Courtesy of the Institute of Food Technologists, Chicago.

stimulated and unstimulated steer carcasses. Even the 'myofibril fragmentation index', which is claimed to be more precise than the fragmentation index (Olson and Parrish, 1977; Parrish et al., 1979), failed to show any structural damage of the myofilaments by electrical stimulation (Judge et al., 1980; Salm et al., 1981). Similarly, George et al. (1980) were unable to detect any gross damage to the sarcomeres by a detailed histological study.

Little information is available whether or not the sarcolemma remains intact in muscles from electrically stimulated carcasses. It is likely that a rapid drop of pH in stimulated carcasses may weaken the tensile strength of the sarcolemma, and this may account for some of the increase in tenderness. Physical disruption of the sarcolemma during electrical stimulation may not be occurring, otherwise a significant drip loss should be expected.

Effect on the Organelles of Muscle Syncytia

Sarcoplasmic Reticulum

Only limited information is available on the response of some organelles of muscle syncytia to post-mortem electrical stimulation. In this regard the research has been limited to the sarcoplasmic reticulum. Tume (1979) has shown that electrical stimulation affected the Ca^{++}-dependent ATPase (the enzyme which is located in the phospholipid vesicles of the sarcoplasmic reticulum and is responsible for driving Ca^{++} ions pumped across the membrane). Although the conformation and activity of Ca^{++}-dependent ATPase was altered by electrical stimulation, no change in the Ca^{++} ion transport ability was apparent. Joseph et al. (1980) also observed little effect of electrical stimulation on Ca^{++} ion capturing capacity of the sarcoplasmic reticulum.

Another study by Tume (1980) indicated that calsequestrin (a protein located in the lateral tubules of the sarcoplasmic reticulum which stores the Ca^{++} ions) was more exposed by electrical stimulation. Besides, a steady state concentration of phosphoenzyme and the ATP$\leftrightarrow$ Pi exchange reactions in the sarcoplasmic reticulum were also reduced by electrical stimulation. The significance of these changes and their relation to post-mortem rate of glycolysis in electrically stimulated muscle is not clear.

It is generally accepted that the Ca^{++} ion-binding ability of the sarcoplasmic reticulum starts decreasing with post-mortem time possibly due either to the direct influence of lactic acid accumulation (Greaser et al., 1969; Pearson, 1977) or indirectly. The low pH may be instrumental in releasing the cathepsins from lysosomes when muscle temperature is still high (Moeller et al., 1976a). The catheptic enzymes, in turn, may be impairing the ATPase and Ca^{++} ion accululating-ability of the sarcoplasmic reticulum (West et al., 1974; Fields, 1976). The whole system seems to operate like a 'feedback' mechanism.

Lysosomes

Many workers assumed simply on the basis of circumstantial evidence that electrical stimulation of carcasses accelerates the release of cathespins from the muscle lysosomes to increase tenderness (Harsham and Deatherage, 1951; Savell et al., 1977, 1978b; Smith et al., 1978; Will et al., 1980). Moreover, Sorinmade et al. (1978) and Dutson et al. (1980a) provided direct proof for the increase in free activity of muscle lysosomal enzymes caused by electrical stimulation of the ovine carcass. This suggested that the lysosomal membrane is ruptured much earlier in muscles from electrically stimulated carcasses than from unstimulated ones. The importance of the changes in lysosomes has been discussed in section VIII.D.2a.

Mitochondria

With regard to the effect on other organelles of muscle syncytia, Will et al. (1980) reported the presence of swollen mitochondria and T-tubules in muscles from electrically stimulated beef carcasses. The significance of these changes in post-mortem muscle with respect to meat quality is not known. However, some workers believe that the relative amount of mitochondria is directly associated with cold shortening (Cornforth et al., 1980). It is thought that mitochondria may release an overload of Ca^{++} ions at low temperatures, and thus saturate the sarcoplasmic reticulum so that excess free Ca^{++} ions initiate shortening when a muscle is subjected to a cold stimulus (Buege and Marsh, 1975; Cornforth et al., 1980). There is considerable evidence that the mitochondria binds Ca^{++} ions (Carafoli and Crompton, 1976; Bygrave, 1979; Saris and Ackerman, 1980). The concentration of the Ca^{++} ion carrier protein in mitochondria is extremely low as compared to that in the sarcoplasmic reticulum (Greaser, 1977).

Effect on the Microbiology of Meat

Very inconsistent information is available on the microbiology of meat from electrically stimulated carcasses. The first report provided by Raccach and Henrickson (1978) showed that electrical stimulation of beef carcasses for 15 minutes (using 300 volts of direct current with a square wave pulse of 60 Hz) caused the lag phase of the psychrotropic bacterial population to increase by 2 days (4-5 *v* 7-8 days respectively for control and stimulated samples). On the other hand, the growth rate during the log phase increased (12.2 *v* 9.9 generations formed respectively in the stimulated and control samples) significantly in meat from electrically stimulated beef carcasses.

By displaying lamb chops for 4 days, Riley et al. (1980a) found a significantly lower bacterial count on meat from electrically stimulated carcasses than from unstimulated ones. They were of the opinion that electrical stimulation might have had a deleterious effect on either the bacterial cells by affecting the viability, or on the meat as a growth medium by fast reduction in pH value. Another proposition was that the proteolytic enzymes, which are believed to be released during electrical stimulation, may be destroying the bacteria.

Mrigadet et al. (1980) have examined the bacteriological aspect of electrically stimulated and unstimulated rabbit, lamb, beef, and pork carcasses. They noticed a significant reduction in the count of *Pseudomonas putrefaciens, M. thermospactum,* and *Lactobacillus* species in muscle (115 min. post-mortem) from electrically stimulated rabbit carcasses. Although aerobic plate count (APC) of stimulated lamb and beef carcass muscles did not show any difference from those unstimulated, the APC of ground beef and pre-rigor fabricated steaks from electrically stimulated sides was often lower than that of respective unstimulated samples after 3 and 6 days of storage. They believed that the change in redox potential (ER) and presence of free radicals may be responsible for inactivating the bacterial cells in meat from electrically stimulated carcasses. However, no consistent and substantial change was noted in microbial types in ground beef, blade steak, T-bone steaks, or rib steaks from electrically stimulated carcasses as compared to those from unstimulated carcasses. In contrast to this, yet another study from the same laboratory (Hall et al., 1980) reported little significant difference in psychrotrophic bacterial counts either initially or at the termination of the display of electrically stimulated and unstimulated beef samples (steaks and ground beef).

A number of other workers failed to identify any difference in the psychrotrophic count of meat from electrically stimulated and unstimulated carcasses either initially

(Gill, 1980; Stern, 1980), or following the 4 day display period (Jeremiah and Martin, 1980). The study by Taylor et al. (1980b) also did not show any effect either on the initial total viable counts and number of *E. coli* cells, or after storage. Similar results were reported by Kotula (1980) and Kotula and Emswiler-Rose (1981a). On the contrary, Contreras and Harrison (1981) observed a lower microbial count in ground beef from electrically stimulated, hot-boned carcasses than from conventional chilled ones.

A critical evaluation of all these reports shows that apart from the dissimilarities in the specification of the electric current employed in each case, there were many differences in the post-stimulation handling of the meat such as storage temperatures, packaging material, sampling procedure and incubation temperature of agar plates. Further, while some workers contaminated the carcass or muscle sample with known flora, others relied on the naturally present microorganisms and made the erroneous assumption that microflora were uniformly present over the entire surface of the test material. These facts may explain the disparity between the results from different laboratories. However, if not for other reasons, one can at least expect that acceleration of post-mortem rate of glycolysis by electrical stimulation would permit rapid chilling of the carcasses. This would help in extending the lag phase of microbial growth in carcasses, thereby resulting in more wholesome product with a longer shelf life as compared to product from unstimulated carcasses which may be stored at a high temperature for a period before cooling to avoid cold shortening.

Effect on the Carcass Quality Characteristics

Heat-ring

Formation of "heat-rings" is sometimes a problem generally associated with cooling beef carcasses which have little subcutaneous fat cover. Thus, a differential cooling rate across the loin area results in the appearance of a dark area on the muscle surface. Savell (1977) preferred to call this phenomenon 'cold-ring' since it develops during the rapid chilling procedures. These dark areas are regarded as a disqualifying feature in the present carcass grading system.

Many workers have reported that electrical stimulation reduces the probability of heat-ring formation in the case of beef carcasses (Smith et al., 1977; Savell et al., 1978b, c, 1979; Cross, 1979; Henrickson, 1979; Salm et al., 1981). However, McKeith et al. (1980y, 1981) reported that electrical stimulation lessens this incident only in steer carcasses but cow carcasses were not affected. Another study by Cross et al. (1980) found no effect of electrical stimulation on the development of heat-ring.

Marbling

Several workers have claimed an increase in marbling score by 11-17% of meat from electrically stimulated carcasses over that from unstimulated carcasses, if evaluated within 48 hours post-mortem (Savell et al., 1978 b, c, 1979; Smith et al., 1980). This difference has been ascribed to the faster 'setting' of adipose tissue and firming of the lean in electrically stimulated carcasses than in conventionally cooled carcasses. This phenomenon is advantageous as it would permit early carcass grading. The ultimate marbling scores between meat from stimulated and non-stimulated carcasses, however, do not differ significantly (Cross et al., 1980).

Lean Firmness and Texture

Savell et al. (1978b) and Salm et al. (1981) have reported that electrical stimulation significantly improved the lean firmness and texture of beef, but the study by Cross et al. (1980) disputed this contention. It is likely that the difference in firmness and color of lean, marbling, and USDA quality grade may be apparent to some extent if the stimulated and control sites are evaluated 14-18 hours post-mortem; beyond that time the difference might start diminishing and become significant 48 hours post-mortem.

Carcass "Bloom"

Electrical stimulation is also believed to contribute to the "blooming" of the meat (Bendall, 1980). The chemical basis of this phenomenon is not clear. It is assumed to be the result of rapid depletion of muscle substrates and intermediate metabolites from the oxidative pathways, which, in turn, reduces the consumption of oxygen from the air and permits deep penetration of the available oxygen into the meat (Bendall, 1980). This may increase the depth of bright red oxymyoglobin layer. This proposition is substantiated by the findings of Tang and Henrickson (1980), who showed that meat from electrically stimulated beef carcasses had more oxymyoglobin than meat from unstimulated carcasses. Perhaps the rapid decrease in pH due to electrical stimulation of carcasses also increases the light scattering properties of the muscles, which may appear bright in color.

Effect on the Meat Quality Characteristics

The ultimate objective of most of the studies on electrical stimulation of carcasses has been to identify the improvement in quality characteristics of the meat. Consequently, extensive works have been reported on the quality characteristics of both raw and cooked meat as influenced by electrical stimulation of the carcass.

Raw Meat Color

Most of the studies, based on panel evaluation, have found the meat from stimulated carcasses generally to be brighter (Smith et al., 1977; Savell et al., 1978, 1979; Calkins et al., 1980; Riley et al., 1980b; Salm et al., 1981) and a more youthful lean color (McKeith et al., 1981) than that from unstimulated carcasses. These differences were identified when the carcasses were evaluated within 24 hours post-mortem (Savell et al., 1978b); beyond that time the difference in color of stimulated and unstimulated meat, if any, possibly levels off thereafter (Calkins et al., 1980). Probably, that is why several scientists from other laboratories did not find any improvement in lean color associated with electrical stimulation (Grusby et al., 1976; Stiffler and Ray, 1979; Strickland et al., 1979a; Nichols and Cross, 1980).

It is probable that the difference in pH value and temperature between stimulated and unstimulated carcasses may be the causative factors in lean color within 24 hours post-mortem. While H^+ ion concentration affects the spacial structure of myoglobin (Asghar and Pearson, 1980), the difference in cooling rate influences the light scattering and lightness of the meat surface (MacDougall, 1977).

None of the studies have mentioned the incidence of pale-soft, exudative (PSE) condition in electrically stimulated beef carcasses, despite the rapid rate of glycolysis. However, George et al. (1980) from England have claimed the occurrence of PSE muscle in electrically stimulated beef carcasses. The researchers in New Zealand (Chrystall and Hagyard, 1976) who have mainly dealt with beef and lamb carcasses,

have categorically refuted any evidence of PSE condition visible in stimulated carcasses at any stage of processing. Electrical stimulation rather seems to improve the color of dark cutting beef, though it may lower (Westervelt and Stouffer, 1978) or conversely improve the color (Thompson, 1981) in the case of pork, depending on the breed.

Recently, Contreras and Harrison (1981b) evaluated the color stability of ground beef from electrically stimulated and control carcasses. The data on reflectance, Hunter Lab spectrophotometer and Hunter a/b ratios suggested that ground beef from electrically stimulated carcasses was more sensitive to metmyoglobin formation than from the control, when both were exposed to radiant energy for 4 hours. However, our recent studies on electrical stimulation of lamb carcasses with varying voltages and frequencies of the pulses did not reveal any measureable difference in lean color of L. dorsi from stimulated and unstimulated carcasses (Rashid et al., 1982a). In this study color measurements were made 30 hours post-mortem using Hunger-Lab's L, a, b system. Even the display of L. dorsi chops from electrically stimulated and control sides for 4 days under 70 ft candle of florescent light at 2°C revealed no change in L, a, b parameters of the color (Rashid et al., 1982b).

Reflectance spectrophotometry has been employed to estimate the relative proportion of oxymyoglobin, metmyoglobin, and myoglobin in muscle (Snyder, 1965; Stewart et al., 1965; Snyder and Armstrong (1967). Whether or not the reflectance spectrophotometer method is more sensitive than that of Hunger to identify a change in meat color is not established. It would be worthwhile to determine which method yields better results.

Retail Case-Life

The round steaks from electrically stimulated carcasses were regarded to be acceptable by a taste panel up to 4 days while those from unstimulated carcasses became unacceptable after 3 days' storage (Savell et al. 1978c). In the study of Riley et al. (1980b) the panel also observed some difference in surface discoloration and overall appearance of the retail cuts from electrically stimulated and control carcass sides on the 4th day of display. However, an examination of meat color using the Hunter colorimeter values of lamb chops every 24 hours for 4 days by Rashid et al. (1982b) did not show any significant difference in L, a, b values of meat from stimulated and unstimulated sides. The chops were wrapped in Saran film and displayed under 70 ft candles of light to simulate commercial conditions.

Thawing and Cooking Losses

Savell et al. (1978c) and Riley et al. (1980b) observed lower thawing losses in meat from electrically stimulated than from unstimulated carcasses. However, the former workers noted a higher cooking loss from electrically stimulated meat. Bouton et al. (1980a) reported similar results on cooking losses, but several other researchers did not find any significant differences between stimulated and unstimulated meat (Riley et al., 1980b; Thompson, 1981; Rashid et al., 1982b). On the other hand, Elgasim et al. (1981) observed less cooking loss in meat from electrically stimulated beef carcasses than from unstimulated controls.

Tenderness

Most of the investigators have agreed that post-mortem stimulation of the carcasses in general produced a tenderizing effect on the musculature (Carse, 1973; Chrystall and Hagyard, 1976; Davey et al., 1976b; Grusby et al., 1976; Ray et al., 1978; Stiffler et al., 1978; Cross, 1979; Cross et al., 1979a; Nilsson et al., 1979; Savell et al., 1979; Smith

et al., 1979; Riley et al., 1980b; Bouton et al., 1980a; Taylor and Marshall, 1980a; McKeith et al., 1981; Valine, 1981). The meat from electrically stimulated carcasses is, on the average, 18-30% more tender than from unstimulated carcasses depending on species, grade, and the nutritional status of the animal (Smith et al., 1979). Rashid et al. (1982b) found about a 11% increase in tenderness in lamb meat by electrical stimulation. Different muscles from a carcass tenderize to a different degree by electrical stimulation. There are several reasons for the differential effect of electrical stimulation on different muscle. These have been discussed in section X.C.

George et al. (1980) have studied in detail the rate of change in shear force of meat from stimulated and unstimulated carcasses by following an exponential equation as applied by Dransfield et al. (1980);

$$F_t = F\infty + (F_o - F\infty)_e - kt$$

where F_t is the shear force (F) in kg at any post-mortem time (t); F_o is the computed shear force at zero time, and $F\infty$ is the shear force at time ∞; k is the rate of constant of the shear force decay in kg per day post-mortem. They have shown that, though shear force decreased relatively faster (0.37 kg/day) in the case of stimulated L. dorsi muscles than that of unstimulated ones (0.16 kg/day), the difference was not significant because of a high standard error. However, the shear force on the first day of aging decreased from 11 to 6 kg. in the stimulated carcasses. The semi-tendinosus muscle showed a similar but less marked effect, perhaps due to variation in current flow.

Bouton et al. (1980a) showed that stimulation of beef carcasses at high voltage (1100 V) resulted in more tender meat than that achieved by low voltage (45 V).

Stimulation of porcine carcasses at low voltage (110 V, 10 sec on, 5 sec off, 110 sec^{-1}) did not improve the tenderness of the L. dorsi muscle (Westervelt and Stouffer, 1978).

Mechanism of Tenderization

Three views so far have emerged to account for the mechanism by which electrical stimulation improves tenderness of meat. They include a reduction in cold shortening, physical disruption of fibers, and an increased acid protease activity.

Reduction in Cold Shortening: A review by Asghar and Pearson (1980) has discussed the influence of cold shortening (shortening of pre-rigor muscle due to cold stimulus) on the tenderness of meat. Many workers believed that the incidence of cold shortening is lessened in electrically stimulated tenderness (Chrystall and Hagyard, 1976; Davey et al., 1976a; Bouton et al., 1980b). The mechanism by which this happens is little understood. We have already discussed the biochemistry of muscle contraction in section III-A. In the light of those events the effect of electrical stimulation on meat tenderness may well be explained as follows.

During electrical stimulation of a carcass, the energy is derived from the catabolism of ATP content and Ca^{++} ions are released by the sarcoplasmic reticulum to support violent contraction of muscles. As soon as the current supply is terminated, the sarcoplasmic reticulum, which is very sensitive to low temperature, effectively recaptures the free Ca^{++} ions, since muscle temperature during stimulation changes little from physiological condition. Thus, the actomyosin ATPase activity ceases due to depletion of Ca^{++} ions from sarcoplasm, whereas the residual ATP content (or ATP formed during the subsequent glycolysis) possibly helps restore the relaxed state of the muscle fibers. With further lapse of time, muscles do not contract in response to cold stimulus or Ca^{++} ions as the pH conditions are not conducive and energy sources (ATP, PC) have been exhausted.

However, those researchers who did not find any significant differences in sarcomere length of muscle fibers between electrically stimulated and unstimulated carcasses (see section V-a) do not believe that inhibition of cold shortening is the major reason

to account for the increase in tenderness of meat by electrical stimulation. We have already stated in section V some of the variations in experimental procedures which possibly resulted in conflicting conclusions on the changes in sarcomere length of fibers.

Physical Disruption of Fibers: On the basis of the electron miscroscopic observation, Savell et al. (1978a) proposed that the massive contraction which occurs during electrical stimulation possibly causes disruption of the muscle fibers, and accounts for the increased tenderness of meat from stimulated carcasses. If this were the case, the 'myofibril fragmentation index' of electrically stimulated carcasses would be expected to be higher than that of unstimulated carcasses. However, in a subsequent report, Savell et al. (1979) have shown that the 'myofibril fragmenation index' of meat from stimulated and unstimulated carcasses was identical. Other workers have reported similar results (section IV). McKeith et al. (1980a) found no evidence of structural damage due to electrical stimulation of intact carcasses, although the meat from these carcasses was more tender than meat from the unstimulated carcass sides.

Recent histological studies by George et al. (1980) also showed no indication of gross structural damage due to electrical stimulation. Should the structural damage be the cause of increase in tenderness of stimulated carcasses, the subsequent chilling or freezing operations are not expected to decrease the tenderness of the electrically stimulated carcasses. All these facts do not lend credence to the hypothesis that physical disruption of fibers during electrical stimulation is the mechanism associated with increased tenderness of stimulated carcasses.

Increased Proteolase Activity: Those research workers who did not find any difference in fibers' sarcomere length between electrically stimulated and unstimulated carcasses believe that benefits of electrical stimulation are not simply related to the prevention of cold shortening (Smith et al., 1977; Savell et al., 1978a). These findings together with others led these investigators to accept that the enhanced activity of lysosomal enzymes in muscle from electrically stimulated carcasses possibly contributes in part to the tenderizing effect. For example, Sorinmade et al. (1978) and Dutson et al. (1980a) observed a significantly higher free activity and lower sedimentable specific activity of β-glu-curonidase (24%) and cathepsin C (30%) in muscle from electrically stimulated carcasses than that from unstimulated carcasses. These enzymes can cleave certain linkages in myofibrillar protein and possibly in connective tissue.

It is thought that a rapid drop in pH may facilitate the rupture of the lysosomal membrane and liberate the proteolytic enzymes while the muscle temperature is still high (Moeller et al., 1976a). High temperature is particularly important, since aging of meat has a high temperature coefficient with 61.5-63.0 kj/mole activation energy (Davey and Gilbert, 1976). George et al. (1980) determined the influence of temperature/pH history on the rate of tenderization of L. dorsi muscle from the activation energy (61.5 kj/mole) at different temperatures during aging of meat. The shear force decay rate was higher in stimulated than in unstimulated carcasses (4.0 v 2.3 kg/day).

Various hypotheses advanced, so far, to account for the chemical changes in meat during the post-mortem ripening process, have been discussed at length by Asghar and Yeates (1978). Another recent review (Asghar and Pearson, 1980) provided additional information on this issue. It appears that some researchers strongly believe that troponin T is the only regulatory protein which is degraded during conditioning of meat (Abbott et al., 1977; Cheng and Parrish, 1978; Yamamoto et al., 1979; Penny and Ferguson-Pryce, 1979). Penny and Dransfield (1979) have shown a relationship between the rate of decay of troponin-T and the shear force of meat.

Assuming the degredation of troponin T may also be responsible for the decrease in shear force of electrically stimulated carcasses, George et al. (1980) examined the

electrophoretic pattern of myofibrillar proteins of L. dorsi muscle from stimulated and unstimulated carcasses, at different intervals of time, and followed the changes in troponin T content and shear force. However, they did not find any relationship between the decay of shear force and decomposition of troponin T. Hence, they concluded that a large difference in shear force between electrically stimulated and unstimulated carcasses at one day post-mortem (8.0 kg *v* 13.3 kg) may be due to some other changes in muscle brought about by electrical stimulation. If so, then there is a need to search elsewhere rather than troponin T for the lower shear force of electrically stimulated meat.

Very recently, some new information on the changes in cytoskeletal protein during conditioning of meat has appeared in the literature. The study by Young et al. (1981) on the ripening of sternomandibularis muscle indicated the disappearance of the newly discovered muscle protein desmin (Lazarides and Hubbard, 1976), sometimes referred to as skeletin (Small and Sobieszek, 1977) or 10 nm filament protein (Robson et al., 1980). Desmin is believed to constitute the honeycomb-like structure in the periphery of the Z-disk (Goldman et al., 1979; Lazarides, 1980).

A muscle protein isolated from the Z-disk having similar functional characteristics as that of desmin was referred to as connectin by Maruyama et al. (1977) and Toyoda and Maruyama (1978). Locker and Daines (1979) are of the opinion that connectin possibly composes the 'gap filaments'. Some studies have shown that connectin reduces in amount during aging of meat (Takahashi and Saito, 1979; Young et al., 1981). Despite all these observations, Young et al. (1981) concluded that factors other than the changes in proteins are responsible for the toughness of cooked, cold-shortened meat.

Enzymology of Meat Conditioning

Various experimental evidences so far appearing in the literature suggest that endogenous proteolytic enzymes from two subcellular locations are involved in the post-mortem tenderizing process: one is the lysosomes and the other is the sarcoplasm.

Lysosomal Enzymes: So far over 60 enzymes have been reported to be present in the lysosomes from one or more cell types. They are comprised of several proteinases (commonly known as cathepsins), glycosidases, phosphatases, sulphatases, phospholipases, and nucleases (Dean and Barrett, 1976; Dingle and Dean, 1976; Dean, 1978). The pH optima of lysosomal enzymes is generally in the acid range with few exceptions such as cathepsin G, elastases and phospholipases which are active at neutral pH (Dean and Barrett, 1976). Lysosomal enzymes do not normally occur free in the cytoplasm. In vivo the release of neutrophils lysosomes is regulated by a fall in cAMP and a rise in cGMP and Ca^{++} ion-influx (Dean, 1978).

A list of some important cathepsins described by different workers is provided in Table 2. Among these, lysosomal proteinases, cathepsins BI, D, E, G, L, and S, and collagenase are endopeptidase in character, whereas cathepsin A, B2, C, and H are exopeptidases. In addition, Matsuda and Misaka (1974) have reported in rat liver the presence of several multiple forms (isozymes) of cathepsin A, namely A_I, A_{II} and A_{III}, of cathepsin B, designated B_I and B_{II}, and of cathepsin C, called C_I, C_{II}, and C_{III}. However, in skeletal muscle only the activity of a few lysosomal endopeptidases have been reported. They include cathepsin B1 (Landmann, 1963; Bouma and Gruber, 1964; Randall and MacRae, 1967; Parrish and Bailey, 1967; Caldwell, 1970; Caldwell and Grosjean, 1971) and cathepsin D (Bouma and Gruber, 1964; Iodice et al., 1966; Parrish and Bailey, 1967; Caldwell, 1970; Caldwell and Grosjean, 1971).

Cathepsin B and D have been shown to degrade myosin, actin, and troponin T (Hasselbach, 1975; Bird et al., 1977; Okitani et al., 1976; Schwartz and Bird, 1977) at low pH (5.2) and high temperature. Although cathepsin D is the main protease present

Table 2. Lysosomal Proteinases (Cathepsins)

Trivial Name	Functional Group	Mode of action	Optimum pH	Other Characteristics	References
cathepsin A (cathepsin 1) (carboxypeptidase A)	−COOH	Expopeptidase		Comparable to pepsin	Matsuda and Misaka, 1974
cathepsin B1 (cathepsin B-II) (cathepsin B')	−SH	Endopeptidase	5 - 6	Comparable to pepain, not trypsin as was originally thought	Barrett, 1978; Otto and Riesenkonig
cathepsin B2 (carboxypeptidase B)	−COOH	Expopeptidase			Ninjoor et al., 1974
cathepsin C (Dipeptidyl aminopeptidase-I)	−SH	Expopeptidase		Comparable to pencreatic chymotrypsin	Gutman and Fruton, 1948; McDonald et al., 1969
cathepsin D (carboxyproteinase) (acid protease)	−COOH	Endopeptidase	2.8 - 5.0	Pepsin-like	Bouma and Gruber, 1964
cathepsin E	−COOH	Endopeptidase	2 - 3		Lapresle and Webb, 1962
cathepsin F		Endopeptidase	4 - 5		Lebez and Kopitar, 1970
cathepsin G	−OH	Endopeptidase	6 - 8		Blow, 1977
cathepsin H	−SH	Endopeptidase	6		Kirschke et al., 1977
cathepsin L	−SH	Endopeptidase	5 - 6		Kirschke et al., 1977
cathepsin S	−SH	Endopeptidase	3		Turnsek et al., 1975
Lysosomal collagenase	$-Me^{++}$	Endopeptidase	7 - 8		Taller et al., 1975
Leucine aminopeptidase (cathepsin-III)	−SH	Exopeptidase			Taller et al., 1975
Carboxypeptidase		Exopeptidase			Taller et al., 1975

in muscle (Iodice et al., 1972), Okitani and Fujimaki (1972) do not think it plays a major role in the post-mortem aging process, whereas Schwartz and Bird (1977) have shown that cathepsin D affected more extensive degradation of myosin and to some extent of F-actin. Penny (1980) emphasized that the pH range within which these enzymes are active does not preclude them from being operative in post-mortem muscle, especially at high temperatures (Arakawa et al., 1976; Penny and Dransfield, 1979; Ouali and Valine, 1981). For example, cathepsin B and D exhibit optimum activity at pH 5.2 and 4.0, respectively. At post-mortem muscle pH (5.5-5.6) cathepsin B and D still have 50% and 30%, respectively, of their optimum activity (Bird et al., 1977).

The low pH and high temperature conditions of muscles have been found to be quite favorable for certain other lysosomal enzymes which catabolize mucopolysaccharides of the ground substance (Dutson and Lawrie, 1974) and some cross-linkages of collagen in the non-helical region (Etherington, 1976). The activity of β-galactosidase, β-glucuronidase, and acid ribonuclease have also been reported to increase in the soluble fraction of meat during aging (Eino and Stanley, 1973; Moeller et al., 1976b). According to Ono (1971) β-galactosidase plays a more important role than β-glucuronidase in the meat conditioning process.

Some evidence of exopeptidase cathepsin A (Bodwell and Pearson, 1964; Parrish and Bailey, 1967; Caldwell and Grosjean, 1971) and more of cathepsin C activity is also available in muscle (Landmann, 1963; Bouma and Gruber, 1964; Randall and MacRae, 1967; Moeller et al., 1976a, b; Caldwell and Grosjean, 1971). Apart from these exopeptidases, several investigators have indicated the presence of a dipeptidase, an aminopeptidase, an aminotripeptidase and leucine peptidase in skeletal muscle (Bury and Pennington, 1975; Parsons and Pennington, 1976; Parsons et al., 1979; Otsuka et al., 1980; Okitani et al., 1981). These peptidases have optimal activity at high pH as in DFD muscle (Okitani et al., 1973, 1981).

It should be emphasized that only endopeptidase can be expected to play a role in the post-mortem tenderizing process of meat, because their sites of action are within the protein molecules. In view of the existence of a large number of endopeptidases and their multiple forms in lysosomes of mammalian cells, it would be of interest to examine the role of different cathepsins and their isozymes in the conditioning process of meat. The exopeptidases, on the other hand, carry little significance so far as important in enhancing the flavor development during ripening of meat, because they act on the terminal ends of protein molecules and produce free amino acids and some peptides of small molecular weight which contribute to meat flavor.

Calcium-activated factor: In muscle sarcoplasm, Busch et al. (1972) reported the proteolytic property of a calcium-activated factor (CAF) which was originally described by Krebs and co-workers (Meyer et al., 1964) as kinase activating factor (KAF), since it activated the conversion of skeletal muscle glycogen phosphorylase b to a. However, Huston and Krebs (1968) had identified the KAF as a proteolytic enzyme. A recent study by Seperich and Price (1981) suggests that KAF and CAF are one and the same, although they were extracted by somewhat different procedures. The CAF is present either free in the sarcoplasm or adsorbed on the myofibrils (Dayton et al., 1976b; Reville et al., 1976). Ishiura et al. (1978) preferred to name it calcium-activated neutral protease (CANP), whereas others call it simply neutral protease (Kang et al., 1981).

Various studies have shown that CAF can digest a variety of muscle proteins. These include proteins of the Z-disk and M-line, C-protein, tropomyosin, and troponin T and I (Dayton et al., 1976a, b; Suzuki et al., 1977, 1978), but CAF has no effect on myosin, actin, and troponin C (Goll et al., 1978). It is probable that CAF may be degrading desmin which is associated with the Z-disk. On the other hand, Locker (1976) suggested that the gap-filaments may be digested by CAF.

Olson et al. (1976) and Olson and Parrish (1977) had associated the degradation of troponin T to 30,000 dalton with CAF. They believe that CAF is the major endogenous proteolytic enzyme which is responsible for the post-mortem tenderization action in meat. However, a very recent study from the same laboratory (Parrish et al., 1981) did not find a significant difference in CAF and free Ca^{++} in meat from tender and tough beef L. dorsi of A-maturity, although tender L. dorsi had a higher content of 30,000 dalton component in proportion to actin than that in tough one. On the other hand, both tender and tough L. dorsi muscles from E-maturity carcasses had the same amount of 30,000 dalton component.

Since CAF requires a high level (1 to 5 mMol) of Ca^{++} and pH in the range 7.0 to 7.5 (Dayton et al., 1976a, b; Ishiura et al., 1978) for optimum activity, the question arises that these conditions in post-rigor meat are not conducive to support the potential activity. Muscle contains about 1.5 mMol of Ca^{++}, possibly 1/3 of which becomes free in post-rigor meat (Hasselbach, 1975). This concentration can activate the CAF at pH 5.5 to about 6% of its maximum capacity. Recently, Penny (1980) compared the action of cathepsin B and CAF during conditioning of isolated myofibrils. CAF was found to digest the myofibrils in the region of C-protein and degrades unknown polypeptides with molecular weight 80,000 and 76,000. On the other hand, high concentrations of cathepsin B affected degradation of myosin into two fragments with molecular weights of 150,000 and 82,000 whereas in dilute solution only troponin T was degraded.

Despite some controversial evidences for and against the activity of lysosomal enzymes in meat discussed by Asghar and Yeates (1978), it appears that CAF and lysosomal enzymes may be working on the principle of division of labor in the post-mortem aging process, analogous to the coordinated activation of different bacteria in sauerkraut production, where each species becomes effective at different stages of the process, with the pH drop due to lactic acid formation from sugar by the preceding species. Thus, in the early stages of the meat conditioning process, when pH is high, CAF may be playing a major role. As the muscle pH drops below 6, the cathepsins may be more active to affect the ripening changes. Huang and Tapple (1971) also postulated a joint action of cathepsin D with other cathepsin for orderly protein degradation in living mammalian tissue.

The evidences discussed in the entire section VIII-D lead to the conclusion that electrical stimulation may have some direct degradation effect on muscle structure, especially at high voltage, but it contributes more by causing a fast drop in pH and hence early onsetting of rigor mortis when the carcass temperature is still high. These conditions permit an early commencement of the autolytic changes in muscle proteins since the ripening of meat has a high temperature coefficient. Besides, the super-contraction of fibers is not favored at low content of ATP and high H^+ ion concentration. However, if rapid chilling of the carcasses is performed at 0°C or less immediately after electrical stimulation, it may allow little tenderization to proceed beyond inhibition of cold shortening. On the other hand, delay cooling (i.e. holding carcasses at temperatures above 10°C for 8 hours) after stimulation would certainly permit additional improvement in tenderness by accelerating the aging process at high temperature. The fast drop in muscle pH by electrical stimulation presumably facilitates early release of catheptic enzymes from lysosomes to promote the aging reactions. The overall mechanism of the post-mortem conditioning process is perhaps much more complex, as described earlier by Asghar and Yeates (1978).

Juiciness

The findings on the effect of electrical stimulation of carcasses on the juiciness of meat seem inconsistent. Most of the studies found no effect on juiciness (Davey et al.,

1976a; Savell et al., 1977; Walker et al., 1977; Bouton et al., 1978; McKeith et al., 1979; Savell et al., 1979; Smith et al., 1979; Elgasim et al., 1981; Salm et al., 1981). However, the same group of workers sometimes reported the meat from the electrically stimulated carcass sides to be less juicy than from unstimulated sides (Savell et al., 1978c; Sorinmade et al., 1978; Bouton et al., 1980a).

Flavor

An improvement of 6-10% in flavor panel scores of meat from electrically stimulated beef carcasses has been reported as compared to that from unstimulated carcasses (Davey et al., 1976a, Savell et al., 1978a, c; Smith et al., 1980). The chemical basis for the improvement is not clear. Possibly, the accelerated aging process in stimulated carcasses is responsible for early development of characteristic meat flavor. The lysosomal expopeptidase may be playing a part in this regard.

Several workers, however, indicated no difference in the flavor intensity and desirability of the meat from stimulated and unstimulated lamb (Savell et al., 1977; Smith et al., 1979), although McKeith et al. (1979) reported a significant difference. A recent study by Elgasim et al. (1981) and Salm et al. (1981), however, revealed no difference in flavor of beef from electrically stimulated and unstimulated carcasses.

Effect of the Electrical Parameters on Meat

Different methods and electrical conditions for stimulating carcasses have been used to accelerate the rate of glycolysis in muscle. They vary from electrode-carcass contact to various parameters of electricity, such as type of current, voltage, frequency, pulse duration, pulse width, pulse shape, etc. The available information on these variables is presented herein.

Electrode-carcass Contact

The contact between the carcass and electrode system is an important feature which determines the effectiveness of stimulation at a particular voltage. In the case of low voltage, it is particularly essential that contact be made with muscle tissue rather than just with the hide. Positioning of electrodes also seems to be important especially if carcasses are stimulated 30 minutes post-bleeding, since delay seems to reduce the effectiveness of stimulation. (Rashid et al., 1982b).

Among different methods of electrode-carcass contact, some preferred to attach a pair of electrodes to the severed m. sternocephalicus and another pair to the muscles adjacent to the Achilles tendon on both sides of the hanging carcass (Bendall et al., 1976). Some inserted a plastic covered rod in the spinal column of the neck to connect high tension electrodes. Others applied the current via two multi-point surface electrodes: one electrode inserted at the distal end of the biceps femoris-semitendinosus muscle junction, and the second into the brachiocephalicus muscle (Shaw and Walker, 1977; Bouton et al., 1980b, c; Taylor and Marshall, 1980a). Still others used a rectal probe (Bouton et al., 1978), a bipolar pithing rod (Bendall, 1980) or rubbing electrodes (Chrystall and Hagyard, 1976).

The use of a rectal probe system is believed to stimulate the muscles indirectly via the nerve plexus in the pelvic area, but the forequarter muscles are not adequately stimulated (Bouton et al., 1980c). The bipolar pithing rod system is designed to stimulate the muscles via the spinal cord. Both these systems may be effective even with low voltage (Bendall, 1980) if they are applied within 5 minutes after bleeding the animal to allow stimulation of muscles in the remote areas. Rubbing and multi-point

electrode systems are useful for direct stimulation of muscles, and yield better results than other systems, especially at currents above 200 volts (Chrystall et al., 1980b). However, those muscles not laying in the direct pathway of current may not be adequately stimulated (Chrystall and Hagyard, 1976).

Bouton et al. (1978) compared the effect on pH drop and tenderness of two different methods of electrical stimulation and suspension of carcass sides by the Achilles tendon or by the sarcosciatic ligament (pelvic hung). They observed the rectal stimulation with or without side suspension to be more effective in reducing the pH of muscle than side suspension alone. Chalcroft and Chrystall (1975) have shown that stimulation of carcasses from one leg to neck resulted in an uneven current flow.

Type of Current

There are several kinds of electrical current essentially the same in nature, which may vary in their method of flow, direction, and current strength. The primary kinds of electrical current are direct, alternating, continous, pulsating, interrupted and oscillatory. The salient features of these currents are shown schematically in Figure 9.

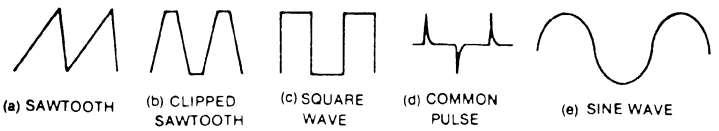
No detailed study seems to have been made to compare specifically the differential effect, if any, of the different kinds of electrical current in accelerating the post-mortem rate of glycolysis. Only a few details are available on this aspect.

Although Chrystall and Devine (1978) stated that alternating pulses gave a little greater $\triangle$ph value than unidirectional pulses (DC), the difference at any one frequency was not consistent. However, alternating current offers some other advantages. For example, there is no risk of electrode polarization and saturation of transformer core. Chrystall and Devine (1980) also stated that alternating polarity pulse, though only marginally superior in terms of pH, was markedly better than unidirectional pulse of the same frequency in making the lamb carcasses tender. A study by Strickland et al. (1979b) has shown that pulsating current (1 sec on; 0.5 sec off) decreased the muscle pH significantly, whereas continous current (1-2 amp) was not at all effective.

Voltage

Different researchers have employed different levels of voltage empirically rather than based on theoretical consideration to stimulate carcasses. The voltage (peak) included 3,600 V (Chrystall and Hagyard, 1976; Gilbert and Davey, 1976), 2,000 to 2,500 (Harsham and Deatherage, 1951), 1,600 V (Davey et al., 1976a), 1,100 V (Gilbert, 1978), 700 V (Bendall et al., 1976), 440 V (Savell et al., 1978b), 300 V (McCollum and Henrickson, 1977) and 250 V (Bendall, 1976). The fact that the application of such a high voltage in commercial practice would demand major safety considerations in the abattoir, motivated some workers to examine the effectiveness of low voltage ranging from 10 to 110 V for accelerating post-mortem glycolysis in musculature (Westervelt and Stouffer, 1978; Bouton et al., 1978; Jonsson et al., 1978; Fabiansson et al., 1979; Taylor and Marshall, 1980a). Most of these workers claimed that 100 V or even less is enough to achieve maximum pH fall in carcasses.

It should be emphasized that peak voltage values of alternating current (A.C.) as such cannot be compared with those of direct current (D.C.). For this purpose, only 'effective voltage', that is, 'root-mean-square' (RMS) voltage of A.C. can be used for computing the energy input if comparison is to be made with that of direct current (D.C.). The effective voltage (V_{eff}) and effective current (I_{eff}) of an alternating current



ELECTRICAL WAVE FORMS

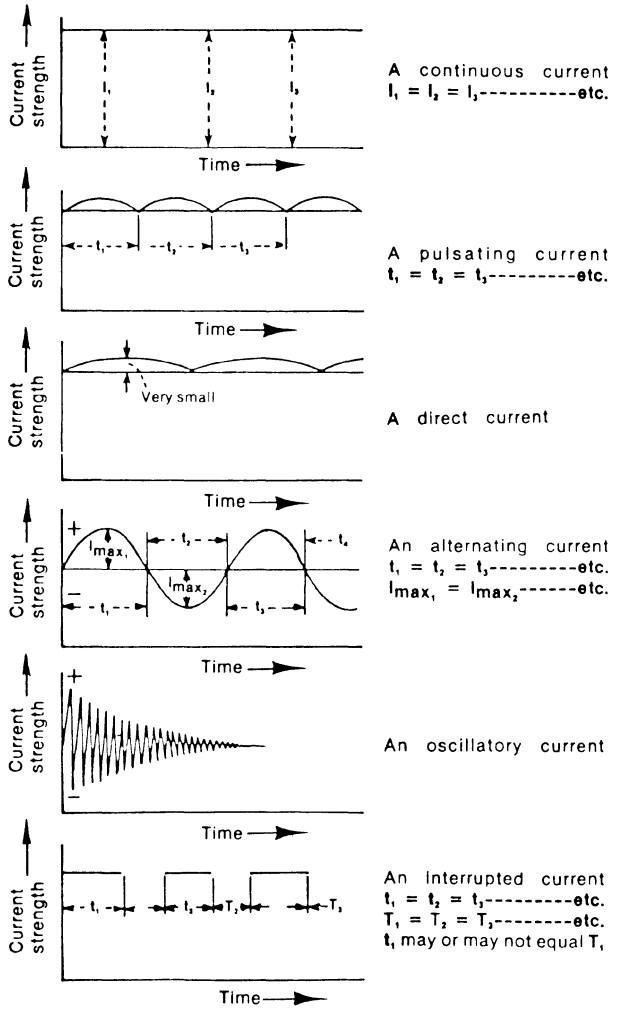


Figure 9. Types of electrical current.

consisting of any wave form can be calculated by solving the following differential equation (Kowalski, 1960):

$$V_{eff} \text{ or RMS} = \sqrt{\frac{1}{T}\int_0^T v^2 \, dt}$$

$$I_{eff} = \sqrt{\frac{1}{T}\int_0^T i^2 \, dt}$$

where T is the time in seconds of the cycle, v is the instantaneous potential drop and i is the instantaneous current. However, the following simplified form of the equations can be used in the case of the sinusoidal (sine) wave for deriving the effective voltage and effective current, and average voltage (V_{av}) and average current (I_{av}).

$$V_{eff} \text{ or RMS} = \frac{V_m}{\sqrt{2}} = 0.707 \times V_m$$

$$V_{av} = \frac{2}{\pi} V_m = 0.637 \times V_m$$

$$\text{Peak Voltage} = 1.414 \times V_{eff}$$

$$I_{eff} = \frac{I_m}{\sqrt{2}} = 0.707 \times I_m$$

$$I_{av} = \frac{2}{\pi} I_m = 0.637 \times I_m$$

Where V_m is the peak voltage, and I_m is the peak current.

Harsham and Deatherage (1951) stated that high voltage was necessary in the case of beef carcasses to achieve a potential difference throughout the carcass to affect contraction of all muscles. Bendall et al. (1976) also found less consistent results with stimulation at low voltage as compared to those obtained with high voltage. However, current density needs to be kept low in order to prevent heating at the electrodes. According to Katz (1967), the well known hyperbolic nature of current strength/duration curves suggests that the threshold current required to excite a membrane increases as the duration of the current pulse is shortened.

In a few cases only comparative studies have been made to examine the effect of different voltage on the rate of glycolysis in carcasses. For example, Carse (1973) compared the effect of interrupted current at zero to 250 V and pulses varying from 2 to 13.5 msec duration delivered at rates from 3 to 17.5 Hz on lamb carcasses. He concluded that of all variables, only voltage affected the rate of glycolysis significantly, the maximum being at 200 V. Stimulation for 30 minutes decreased the muscle pH to 6.0 in 3 hours, whereas unstimulated control took 16 hours.

Bendall's (1976) work also showed that 250 V gave adequate results and rigor was completed in the lamb carcass in about 3 hours. In another study, Bendall et al. (1976) reported that voltage has a highly significant effect both on the immediate pH fall (ΔpH) during stimulation and on the subsequent rate of glycolysis (dpH/dt). They indicated that the higher voltage (300, 700 V) was much more effective than 100 V. The difference between 300 and 700 V was relatively quite small. Low voltages gave less than optimal pH fall in the forelimb muscles such as triceps brachii.

According to Chrystall's (1978) study, early chilled lamb carcasses after stimulation with 800 V (RMS) were more tender than those stimulated with 400 V (RMS). Bouton et al. (1980b) compared the effectiveness of three electrical stimulation systems on beef carcasses. Their results indicated high voltage (1,100 V) to be more effective than low voltage (110 V) and extra-low voltage (45V) in accelerating the glycolysis in muscle, and that 90 seconds stimulation time was as good as 120 seconds. It may, however, be pointed out that wave form, pulse frequency, and carcass-electrode contact method were not the same in this study. This makes it difficult to generalize the interpretation of the results. Very recently, McKeith et al. (1981) found significantly more improvement in the quality characteristics of meat from carcasses stimulated at 550 V than at 150 V. However, duration of stimulation (1 to 2 minutes) did not evoke any difference in quality criteria of meat.

Our study on lamb carcasses compared the effect of two voltage levels (50; 350 V) on the rate of glycolysis. The results substantiated the view that high voltage is significantly more effective in accelerating the rate of post-mortem glycolysis in muscles (Rashid et al., 1982a). The studies by Houlier et al. (1980) have also clearly shown that ΔpH increases with the increase in the electrical field of 2 V per cm, depending on the stimulation time. This suggests that the voltage should not be less than 600 V for the stimulation of large beef carcasses to obtain overall satisfactory decreases in pH of the musculature. Bendall (1980) also advocates the use of at least 600 V (peak) if electrical stimulation is performed through electrodes on the severed neck region and Achilles tendon, and at least 1,000 V (peak) for the stimulation of lamb carcasses.

In contrast to all these evidences, Shaw and Walker (1977), who have tried 10 to 100 V direct current and pulse rate varying from 10 to 40 pulses per second for 1 to 4 minutes, found that voltage and stimulation time had little effect on the rate of pH fall. They found 20 V as effective as higher voltage in accelerating glycolysis. However, physiological considerations suggest that weak pulses are not affective to produce the required action potential, and a pulse above a sharp threshold around 18 pC/cm^2 generates the needed response (Ackerman et al., 1979).

Pulse Rate (Pulse Frequency)

Pulse rate denotes the number of cycles per unit time, and the number of cycles per second is called hertz (Hz). From the physiological viewpoint, the pulses (cycles) should consist of a few millisecond duration so as to produce effective depolarization of the nervous system and allow recovery of the membrane potential (repolarization). Harsham and Deatherage (1951) preferred to keep the frequency close to the physiological stimulation frequencies, that is, 60 pulses per second for stimulation of beef carcasses. Other workers have used as much as 400 cycles per second (McCollum and Henrickson, 1977; Raccach and Henrickson, 1978; Tang and Henrickson, 1980; Will et al., 1980).

Regarding the effect of frequency (number of pulses) on the rate of glycolysis, Bendall et al. (1976) reported that only the total number of pulses was an important factor. They found identical results whether 1,500 pulses were given in 1 or 2 minutes, that is, 25 or 12.5 Hz. The same results were found in the case when 3,000 pulses were

given in 2 or 4 minutes (25 and 12.5 Hz respectively), and pulses beyond 3,000 offered no advantages in increasing the rate of glycolysis.

However, the findings of Bendall et al. (1976) were at variance with those of Chyrstall and Devine (1978). The latter workers noted that ΔpH increased markedly with pulse frequency and reached its maximum (0.7 pH units) before the individual twitches summated into a tetanic contraction at frequency near 16.6 sec^{-1}. The ΔpH then decreased as the frequency increased above 20 pulses sec^{-1}. This means that 1,500 pulses delivered in 120 seconds (i.e. @ 50 pulses sec^{-1}) do not yield equivalent ΔpH. The energy input, though, continues increasing linearly with increasing frequencies at high voltage while the drop in pH starts declining beyond 16.6 pulses sec^{-1}. Consequently, more time was required for the muscle to attain pH 6.0 at higher frequencies. These results suggest that pH is more a function of pulse frequency than the total energy input as conceived by Bendall et al. (1976). However, dpH/dt was not affected by pulse frequency between 12.5 to 100 pulses per second (Chrystall and Devine, 1978).

The study by Rashid et al. (1981a) compared the effect of three levels of frequency (10, 100, 250 Hz) on post-mortem rate of glycolysis in lamb carcasses keeping the duty cycle constant at 20%. Their data (Table 3) partly disagrees with the finding of Chrystall and Devine (1978) that the rate of glycolysis declines as the frequencies increase above 10 Hz. Table 3 shows that the glycolytic rate in lamb carcasses was more a function of energy input per pulse than the total energy input. This suggested that the total energy was not the decisive factor in affecting the rate of glycolysis in muscle. It was, in fact, the energy content per pulse which seemed to be the rate determining factor. Thus, the interaction between voltage and pulse frequency has a significant influence in the glycolytic rate in the electrically stimulated carcasses. Although the optimum pulse rate is thought to lie in the range of 5 to 16.6 Hz (Chrystall and Devine, 1978), the findings of Rashid et al. (1981a) suggested a linear increase in rate of glycolysis with decrease in pulse frequency from 250 to 10 Hz. Therefore the optimum pulse rate may be less than 10 Hz to achieve the maximum rate of glycolysis in carcasses by electrical stimulation.

The pulse rate at which twitching of individual muscles in a carcass fuse fully into a tetanus seems to vary with type of carcasses (Bendall, 1976). For example, in the case of rabbit the contractile process is faster than beef, and twitching of individual muscle can be observed even at 25 pulses per second (pps). Bendall (1980) stated: "The pulse rate at which complete fusion of muscle twitches into a tetanus occurs is an approximate inverse measure of the time to peak twitch tension. Thus, in beef muscles where fusion occurs at 12 pps, the time to peak is about 80 msec. Any stimuli arriving at the motor end-plate within this time will, therefore, be ineffective. With rabbit white muscle, the time to peak at 38°C is less than 20 msec." The physical response of the lamb carcasses during electrical stimulation at different voltage and pulse frequencies is shown in Table 4.

Pulse Duration

Pulse duration, also called pulse width or pulse length, is expressed in seconds or milliseconds (msec). Sometimes this is described in terms of duty cycle or duty factor, which is the ratio of pulse width to the pulse period between successive pulses, and usually is expressed in percentage as shown below:

$$\% \text{ Duty Cycle or Factor} = \frac{\text{Pulse width (in sec)}}{\text{1/Frequency (in Hz)}} \times 100$$

Duty cycle denotes the percentage of the time for which the current remained on per period. For example, 20% duty cycle means that for a pulse period of 100 msec, current remained on for 20 msec and off for the remaining 80 msec. Most of the research workers have not correctly reported these parameters, making comparisons difficult.

Table 3. Relationship between energy per pulse and time to reach pH 6.0 in the L. dorsi and semimembranous muscles on post-mortem electrical stimulation of lambs sides at different voltages and pulse frequencies for 4 minutes using a square wave direct current with 20% duty cycle

Voltage[a]	Pulse frequencies	Resistance carcass side Ohm.[b]	Output current amperes (A)	Power watts (W)	Total electrical energy output (kj)	Energy per pulse	Time required to reach pH 6.0	
							L. dorsi hr.	Semimembranous hr.
Unstimulated control side	—	—	—	—	—	—	12.33	11.42
50	10	287.35	0.17	8.5	0.418	0.17	8.91	8.47
50	100	287.35	0.17	8.5	0.418	0.02	8.72	8.70
50	250	287.35	0.17	8.5	0.418	0.01	10.07	9.77
350	10	269.23	1.30	455.0	21.84	9.10	3.96	3.91
350	100	269.23	1.30	455.0	21.84	0.91	7.12	7.81
350	250	269.23	1.30	455.0	21.84	0.63	7.97	8.57

a) Voltages were recorded by Fluk 8000 A digital multimeter; b) Resistance of the carcass side was measured by Simpson digital multimeter Model 464 (Rashid et al., 1981).

Table 4. Physical response of lamb carcass sides during electrical stimulation at different voltages and pulse frequencies*

Electrical Parameters		Initial responses of	Twitching of muscles	Duration of
Voltage (V)	Frequency (Hz)	the carcass sides		Twitching
50	10	Cervical and thoracic regions bend laterally outward moderately	Fast twitching of most of the muscles	180 - 240 sec.
50	100	Cervical and thoracic regions bend laterally outward moderately	Very slight twitching in few muscles in the leg and neck regions, as separate twitches fused into tetanus	60 - 120 sec.
50	250	Cervical and thoracic regions bend laterally outward moderately	No obvious twitching of muscles, and the muscles immediately go into tetanus	—
350	10	Cervical and thoracic regions bend laterally outward vigorously	Fast and vigorous twitching of most muscles	120 - 180 sec.
350	100	Cervical and thoracic regions bend laterally outward vigorously	Very slight twitching in few muscles in the leg and neck regions, as separate twitches fused into tetanus	20 - 80 sec.
350	250	Cervical and thoracic regions bend laterally outward vigorously	No obvious twitching of muscles, and the muscles immediately go into tetanus	—

*From Rashid et al., 1981.

Pulses as long as 10 sec (Cross et al., 1979a) and 0.5 to 1.0 sec duration (Savell et al., 1979; Seideman et al., 1979) and as short as 2.0 to 2.5 msec duration (Tang and Henrickson, 1980; Will et al., 1980; Demeyer and Vandenriessche; 1980) have been used for stimulation of beef carcasses. Bendall (1980) does not think that pulse width carries any significance in affecting the rate of glycolysis in carcasses, except when very short pulses (<5 msec) are used. Although a comparative study was not available, short pulses may not stimulate all muscle fibers in the long patyway of a beef carcass. The recent study by Rashid et al. (1982b), however, demonstrated that a pulse duration of 100 msec was significantly more effective in accelerating the post-mortem rate of glycolysis in lamb carcasses than 10 and 4 msec pulse duration when the duty cycle was fixed constant at 20%.

Pulse Shape (Wave Form)

The interrupted current with different wave forms can be generated in an electronic circuit (Slurzberg and Osterheld, 1965). Most common forms are the sine waves, square waves, sawtooth waves, and train waves (Figure 9). More complex wave forms can be generated than these fundamental wave forms.

Very limited information is available on the influence of wave forms on post-mortem glycolysis rate during electrical stimulation of carcasses. From the available data, it appears that pulse shapes also have a bearing on the rate of glycolysis. Chrystall and Devine (1978) observed that 12.5 pulses per second of sinusoidal shape (sine wave) resulted in somewhat greater ΔpH of muscle than those of square wave pulses, although the energy input in the latter case was considerably greater than the former. However, the area under the tension-time and the time for tension to fall (to 50% of peak tension) were slightly more for square than for sinusoidal pulses, whereas peak tension was less.

Some studies at the Swedish Meat Research Institute reported the use of square pulses of low frequency and very low voltage for stimulation of beef carcass (Fabiansson et al., 1979; Nilsson et al., 1979). But a later study by Ruderus (1980) failed to reproduce the earlier findings. (Possibly error arose in the early experiments due to some faulty instrumentation.). Ruderus (1980) then rechecked the results on different pulse types in relation to pH fall during stimulation. According to his data, stimulation of beef carcasses with square wave pulses at 20 V or 40 V did not affect the rate of pH fall as compared to that of unstimulated carcasses. Pulses consisting of a sine wave 100 Hz (at 65 or 80 V, 60 msec, 1 sec) and 14 Hz (at 80 V) were almost equally effective, whereas continuous stimulation with frequency of 14 Hz (at 80 V, with the individual peak being 5 msec) gave the highest reduction in pH fall during carcass stimulation.

The literature reviewed in section II to VIII showed that a large number of research workers have contributed information on electrical stimulation of carcasses. A careful evaluation of experimental parts of their respective studies showed that while most of them failed to mention the complete conditions of their electrical stimulation parameters, others loosely used electrical parameter terminology so as to make interpretation confusing. These difficulties limited our attempts to compare the results of various studies concerning the influence of different electrical parameters on the glycolytic rate in carcasses. It is suggested that all the electrical terms used in scientific papers be defined in the standard electrical terminology Institute of Electrical, Electronic Engineers (IEEE) for better understanding of the information.

Mode of Current Flow Through the Carcass

The Theories of Impulse Transmission

The ionic basis of electrical impulse in an isolated muscle fiber is well documented by Hodgkin (1965), while Katz (1967) has narrated the mechanism of membrane depolarizing effect through liberation of acetylcholine. It seems that the membrane may exhibit a specific resistance in the order of 100 ohm/cm.

Lange et al. (1973) studies the internal electrical resistance of 3-10 year old cows by varying DC voltage, condensor discharge, and the routes of current. They concluded that at a low voltage (10V) the internal resistance of the body responds like an ohm conductor, whereas a higher voltage (up to 400 V) causes a sudden drop in the internal resistance which is only slightly influenced by a voltage of 400 V. This behavior seems to conform with the rules of a spatial electrical field, and the fall in resistance may be due to a state of biological super-conductivity.

As to the mode of electrical stimulation of a carcass, the studies by Bendall (1976) and Swatland (1977) suggested that a stimulus operates indirectly through nerve pathways. That is, the effect of stimulation may be due to impulses arriving from the motor nerves and ventral roots of the motor end plates. A single motor nerve fiber can activate through the single end plates on each fiber as many as 100 muscle fibers in a

so-called motor unit (Katz, 1967). There is no direct stimulation of the muscle membrane, which probably becomes more sensitive to the passage of an interrupted current in an animal curarized with pancuronium drug. This seems logical in view of the very high resistance of the muscle membrane. Devine et al. (1979) showed that muscle still responded to direct high voltage stimulation even though the neuromuscular transmission was blocked by administering curare to the animal before slaughtering.

It has been shown that nervous pathways remain viable only about 15 minutes post-mortem (Swatland, 1976a), and response to nerve stimulation is ceased after 30 minutes delay in the case of lambs (Chrystall et al., 1980b). Thus, it is likely that direct stimulation applied early post-mortem is effective partly by electricity responses from neuromuscular junctions (Swatland, 1977; Chyrstall et al., 1980b). The neuromuscular junction can no longer be excited 30 minutes post-mortem with low voltage ($\sim$12 V) to bring about muscle depolarization. It is, however, not yet clear whether lack of response through the nervous system 30 minutes post-mortem is due to conduction failure or neuromuscular failure.

Ackerman et al. (1979) stated that fatigue sets in at synapses in the central nervous system before the myoneural junction which, in turn, fatigues before the muscle does. If this were so, then it would not be feasible to achieve on line stimulation of beef carcasses, by blocking the motor end plates on the acetylcholine receptors before electrical stimulation of muscles, Devine et al. (1979) have shown that a functioning nervous system is not essential for achieving the optimum effect by direct stimulation of the carcasses at high voltages. The high voltages ($<$200 V) possibly give sufficient current density to directly depolarize the membrane and produce a physical and chemical response.

Carcass Impedance

The impedance (Z) is defined as the total resistance offered by a circuit to the flow of alternating current at a given frequency, that is

$$Z \text{ (ohms)} = \frac{\text{potential difference across a circuit (V)}}{\text{current of the circuit (I)}}$$

The impedance is directly related to the length and inversely to the cross-sectional area of a conductor. That is

$$Z = \frac{K \times L}{A}$$

where L is the length; A is the area; and K is the specific resistance of the conductor. Bendall's (1980) observations conform with this generalization, and showed that undressed carcasses offered less resistance than the dressed ones (0.65 *v* 1.02 ohms/cm), possibly due to large cross-sectional area and the presence of wet viscera. The impedance of sides was higher than intact dressed carcasses (1.30 *v* 1.02 ohm/cm) because of less cross-sectional area in the former case.

An increase in resistance causes a decrease in current flow according to Ohm's Law (i.e. current = voltage/resistance). Bendall (1980) also recorded that the lamb and rabbit carcasses offered much higher resistance (3.30 and 1.50 ohm/cm respectively), even though small mean cross-sectional areas of the carcasses were partly compesated by short length. These data suggest that small carcasses (e.g. lamb) would require much higher voltage than the large one (e.g. beef) to provide equivalent flow of current. In other words, a peak voltage of 680 V would give peak current to 5.2 A with undressed carcasses, 3.3 A with dressed carcasses, 2.4 A with sides, length being 200 cm in each case using a moderate size carcass of 300 kg.

Bendall (1980) also reported some data on voltage drop in various regions of the carcass during electrical stimulation. Accordingly, the mid lumbar area of the L. dorsi muscles showed 2.2 V/cm drop as compared to 3.0 V/cm of the entire length of the carcass on stimulation at 600 V (peak). In the forelimb muscles (e.g. Triceps brachii) which are not directly in the current pathway, the drop was less than 0.4 V/cm. Similarly, if the electrode was connected to only one leg during stimulation, the voltage drop in the other leg was merely 0.11 V/cm as compared to 4.3 V/cm in the former. Moreover, the specific resistance along the length of the muscle fibers is reported to be far less than across the fibers (450 *v* 9,000 ohm/cm of semitendinous muscle), because of the impedance of cell membrane. Thus, the rate of pH fall in muscle increased when the current was applied along the fibers, but no influence on pH when current was applied at the same voltage across the fibers.

The impedance of a carcass can also be affected by a change in its temperature. Generally, a rise in temperature increases the vibrations of the atoms in the conductor and it becomes difficult for electrons to pass through due to the continuous changes in the relative spacing. Hence impedance increases with temperature according to the following relationship:

$$R_F = R_i + [R_i \times T_c \times (t_f - t_i)]$$

where R_F is the final resistance; R_i is the initial resistance; T_c is the temperature coefficient of the conductor; t_i is the initial temperature; and t_f is the final temperature.

This relationship does not hold true, however, for those materials (e.g.) carbon which exhibit a negative temperature coefficient of resistance. In those cases the impedance increases with temperature possibly due to rhythmic oscillation of atoms which may be helping instead of resisting the flow of electrons. Some substances show little change in impedance with a change in temperature. Whether or not the impedance of a carcass is influenced by variation in its temperature has not been reported. This information would be very important in manipulating the post-mortem conditions to post-mortem carcass stimulation to achieve the best results.

Differential Response of Muscles

It has been observed that various muscles in a carcass responded differently to electrical stimulation (McCollum and Henrickson, 1977; George et al., 1980). Biceps femoris and M. triceps brachii showed identical response whether the carcass received 1,500 or 3,000 stimuli, whereas 1,500 stimuli were ineffective in the case of L. dorsi and semimembranous muscles, due possibly to more complex innervation or higher resistance in the electrical pathway (Bendall et al., 1976). This suggested that the variation in response of different muscle would be larger when lower voltage is applied. Bouton et al. (1980a) observed a fast pH decline in the L. dorsi on electrical stimulation with 14.3 pulses sec^{-1}, than in semimembranous muscle with 40 pulses sec^{-1}. However, the type of current, pulse width and shape were not identical in this study. Consequently, some difference might be associated with interaction of these variables. In another study, Bouton et al. (1978) reported that electrical stimulation has less effect on the forequarter muscle (deep pectoral, triceps brachii) than those of the hindquarter (semimembranous, L. dorsi, gluteus medius).

Several explanations have been extended to account for the differential response of various muscles to different electrical stimulation conditions. First, muscles might be responding differently per se to different wave forms. Second, the different wave forms may be transmitted with varying degrees of effectiveness along nerve-muscle pathways, and hence, effective stimulus received by a particular muscle may vary from the others. Swatland (1976b, 1977) related the high scatter in the post-stimulation pH values to the metabolic and contractile characteristics of the muscle fibers. Table 5 depicts some

Table 5. Relative difference in various characteristics of white and red muscle fibers.

Characteristics*	White fibers (or Type A or Type II or Fast)	Red fibers (or Type B or I or Slow)
1. Microscopic and ultrastructural		
Fiber diameter	Large	Small
Z-disk	Narrow	Broad
Post-mortem Z-disk degradation rate	Fast	Slow
2. Chemical		
Myoglobin content	Low	High
Glycogen content	High	Low
Lipid content	Low	High
3. Biochemical		
ATP content	High	Low
Creative phosphate content	High	Low
Glycolytic metabolism	High	Low
Oxidative metabolism	Low	High
ATP-splitting capacity	High	Low
Protein turnover rate (in living muscle)	Low	High
RNA content	Low	High
4. Enzymic		
Phosphorylase	High	Low
Succinate dehydrogenase	Low	High
Fructose 1, 6-diphosphate	High	Low
-glycerophosphate dehydrogenase	High	Low
Hexokinase	Low	High
Acid hydrolases	Low	High
Lipase	Low	High
Lactate dehydrogenase (LDH)	High	Low
Isozymes of LDH	LDH_4, LDH_5	LDH_1, LDH_2, LDH_3
5. Physiological		
Capillary density	Low	High
Contraction speed	Fast, shot duration	Slow, tension sustained
Contractile action	Phasic	Tonic
Blood supply	Low	High
6. Cellular		
Sarcoplasmic reticulum	Well-developed	Less-developed
Mitochondrial size	Small	Large
Number of mitochondria	Low	High
Mitochondrial distribution	Subsarcolemmal	Diffuse-subsarcolemmal

* The information has been derived from various sources such as Brooke, 1970; Gauthier, 1970; Beatty and Bocek, 1970; Peter et al., 1972; Verity and Coleman, 1973; Abbott et al., 1977; Gann and Merkel, 1978; Cornforth et al., 1980.

of the chemical, biochemical, physiological, enzymic, and structural differences between so-called white and red muscle fibers.

The study by Houlier et al. (1980) substantiates Swatland's proposition. Their study clearly showed that muscles having different fiber composition responded differently to electrical stimulation. The fast-twitching muscle, mainly composed of white type fibers (e.g. tensor, fasciae latae) gave high response to electrical stimulation, whereas slow-twitching muscle, composed of β-red type fibers (e.g. supraspinatus) responded the least, particularly to weak stimulation. The fast-twitching muscle, composed of α-red type fibers (e.g. Longissimus dorsi) gave intermediate response. This may explain why slow-twitching muscles remain sensitive to cold shortening even after electrical stimulation of the carcasses.

Commercial Application

Appropriate Time for Post-mortem Stimulation

McKeith et al. (1979, 1981) have shown that electrical stimulation of carcasses performed at any of several stages between post-bleeding and after splitting the sides improved equally well the tenderness of goat meat. However, if the effectiveness of electrical stimulation is judged from the rate of pH fall in muscles, then it seems logical on the basis of muscle physiology that the carcass be stimulated soon after bleeding the animal, when the nervous system is still viable. The response of beef carcasses declines markedly 50 minutes post-bleeding and that of lamb carcasses even quicker (Bendall, 1980). Harsham and Deatherage (1951) also advocated that carcasses should be stimulated immediately after bleeding. In any case, stimulation should not be delayed more than 40 minutes post-mortem for beef and 30 minutes for lamb.

Chrystall and Devine (1978) recorded very high $\triangle$pH values ($>$0.5) when the pre-electrical stimulation pH value of the carcass was above 7.0. The $\triangle$pH reduced less than 0.2 of a unit, when the carcass had already attained pH 6.5 at the time of stimulation, and the $\triangle$pH was almost zero when the carcass was stimulated at pH 6.3. The delay in electrical stimulation of a carcass after bleeding is disadvantageous in several ways (Chrystall and Devine, 1980a). First, as pointed out earlier, the nervous system becomes ineffective, and one will have to rely on direct stimulation. Second, the magnitude of $\triangle$pH continues to decrease with any delay in stimulation after bleeding. Third, muscle temperature starts falling off from bleeding time onward. This carries a practical significance since glycolysis in muscle has a high temperature coefficient (Jeacocke, 1977). Hence, low carcass temperature is likely to lessen the effect of electrical stimulation on the rate of glycolysis, and ultimately on the meat conditioning process.

Duration of Electrical Stimulation

The duration of electrical stimulation of carcasses may vary between 1.5 and 4 minutes depending upon the voltage (Bendall, 1980). The output of a slaughtering plant is an important factor to be taken into account while deciding the stimulation duration. Generally speaking, 4 minutes duration schedule may be followed with low voltage ($\leq$100 RMS) with a moderate dressing line. However, 1.5 to 2 minutes stimulation time at high voltage ($\geq$600 V peak) would be adequate for a modern dressing line with output of up to 50 carcasses/hour.

Safety Considerations

Man is sensitive to very low electrical current because of his highly developed nervous system. Although it is well known that it is the "current that kills," electrical safety regulations are generally worded in terms of voltage. The effect of an electrical shock depends, to a great extent, upon the path of the current flow, length of exposure, and type of contact, whether the contacts are firm and involve an appreciable area, or whether they are point contacts. A small current such as 10 milliamps is unpleasant (Seippel, 1974), and may produce a piercing pain or shock causing an involuntary movement or loss of balance, and the ensuing fall may result in injury. Currents large enough (20 milliamps) to stop breathing may cause excessive contraction of the chest muscles producing temporary paralysis by action on the nerves. Thirty milliamps current can cause damage to brain tissue and blood vessels (Seippel, 1974). When the current pathway is sufficient to stop breathing, it may also produce ventricular fibrillation, uncoordinated heart beats, or damage to the vital parts of the nervous system. When currents reach 250 milliamps, death usually occurs. Using 500 ohms as the human body resistance, Dalziel (1956) found the fibrillation threshold to be 13.5 watt seconds.

Practical implementation of electrical stimulation to the meat animal carcass requires electrical safety. Effective stimulation must include some type of pulsed excitation. Steady direct current has very little effect. A pulsed wave, whether of constant polarity (pulsed D.C.) or alternating polarity (pulsed A.C.) is similar in risk of electrocution at the same peak voltage. It is pulsing, not the alternating polarity, which increases the risk of fibrillation. The maximum peak voltage for a pulsed wave form to be classed as extra low voltage is 45 volts. Such a level is generally accepted as being safe to touch.

Before attempting to utilize electrical stimulation, care should be taken to assure worker safety. The quantitative effects of electrical current on man are shown in Table 6. The let-go threshold for alternating 60 cycle current is approximately 16 milliamperes. Increasing the cycles to 10,000 per second increases the let-go threshold to 75 milliamperes. Thus, it is necessary to utilize great precautions (Anderson, 1980). Care should be taken to have the stimulation unit adequately grounded. One should use polarized grounded outlets and make sure the case is grounded. Some commercial stimulation units will have a dead man switch. Where high voltage is used, stimulation must be done in a restricted or fail-safe area. A fail-safe area may take the forms of a single carcass cabinet, single manual probe, or a personnel-proof tunnel area equipped with plastic rail, warning lights, insulated high voltage circuits, automatic ground fault

Table 6. Quantitative Effects of Electric Current on Man*

	Milliamperes		
		AC	
Effect	DC	60 Cycle	10,000 Cycles
No hand sensation	1.0	0.4	7
Perception threshold	5.2	1.1	12
Shock - not painful	9.0	1.8	17
Shock - painful	62.0	9.0	55
Let-go threshold	76.0	16.0	75
Severe shock - muscle contraction	90.0	23.0	94
Certain Death (3 seconds)	1375.0	275.0	1375

*Dalziel, C.F. 1956.

control, automatic conveyor, and a contact bar. A live electrode, in the form of a broad stainless steel strip, makes rubbing contact at the brisket/shoulder region of the carcass. The return path of the current is through both hocks to the earthed rail.

When the electrical current is applied improperly, the carcass may jerk away from the electrode or jump from the rail. The location of the electrode must also be positioned to accommodate a wide range of animal size. Any high voltage system implies an enclosed cabinet or tunnel. This will exclude personnel or automatically shut off the electrical current when someone enters the area. Hand or foot operated safety switches help to reduce risk of injury.

Hot-Boning

Hot-boning generally implies the excising of muscles and/or muscle systems from the carcass within 1-2 hours post-mortem, vacuum packing, and conditioning them at 15°C for 24 to 48 hours or at 1°C for 8 days (Schmidt and Gilbert, 1970; Schmidt and Keman, 1974). Removing the muscles after holding the carcasses at 15°C for 6-10 hours is also considered hot-boning (Kastner et al., 1973, 1976). In many countries of the world, where refrigeration facilities are lacking, it is a common practice to cook the meat in a pre-rigor state (hot-boned).

The meat industry in the western countries started realizing the need for processing hot-boned meat as it offers several advantages over that of cold-processing. These advantages include reduction in refrigeration, transportation, and less weight loss during cooling, which amounts to a significant saving in processing cost, labor, time, and energy (Henrickson, 1975; Ferguson and Henrickson, 1979; Cuthbertson, 1980; Erickson et al., 1980). The thickness of the hindquarters limits the cooling rate and the geometry of the carcass precludes the efficient use of space in the chilling room and uniform circulation of cooling air. It has been estimated that the edible part of an average choice grade beef carcass requires only 20% (Henrickson and McQuiston, 1977) or 25% (Cuthbertson, 1980) of the space needs for the whole carcass. Thus, hot-boning offers the solution for some of the engineering problems of the meat industry and results in about a 15% saving in capital cost of refrigeration (Cuthbertson, 1980) with a 50% saving in energy (Henrickson and McQuiston, 1977).

Hot-processing of ground beef has also been found to be superior to cold processed minced beef in physical, chemical, cooking, and sensory properties (Cross et al., 1979b, 1980; Berry and Stiffler, 1981; Cross and Tennent, 1981; Jacobs and Sebranek, 1980; Will et al., 1980). These advantages have been assigned to the high content of ATP and pH value of hot-processed meat (Hamm, 1976; Honikel and Hamm, 1978). Other benefits identified in the case of hot-curing of meat are lower drip and cooking losses (Weiner et al., 1966), more tender (Mandigo and Henrickson, 1966) and bacteriologically sound end product (Barbe and Henrickson, 1967), a greater rate of cure diffusion, better development and more stable color formation (Arganosa and Henrickson, 1969; Henrickson et al., 1969) than curing of post-rigor meat. In addition, the pre-rigor beef mince has been claimed to have more emulsification capacity than post-rigor mince in sausage making (Trautman, 1964; Acton and Saffle, 1969).

Despite all these merits, especially in the case of fabricated products, the conventional hot-boning practice is not without some potential disadvantages. For example, hot-boned meat is likely to be tough due to cold-shortening if conditioned at 1°C (Marsh and Leet, 1966; Davey et al., 1976b; Kastner et al., 1973, 1976; Gilbert et al., 1976b; Cuthbertson, 1977; Griffin et al., 1979; Cross et al., 1980). Hot-boned meat tends to be susceptible to bacterial proliferation if conditioned at a high temperature (Schmidt and Gilbert, 1970; Follet et al., 1974; Dransfield and Jones, 1978; Fung et al., 1980; Kotula and Emswiler-Rose, 1981; Fung et al., 1981), although the study by

Taylor et al. (1980b) does not support this contention. Inconvenience in cutting, maintaining the shape of the cuts and difficulties in synchronizing slaughtering, grading hot-boning and manufacturing line are some of the other drawbacks associated with conventional hot-boning practice.

Many investigators agree that most of the problems arising from conventional hot-boning practice can be solved by introducing electrical stimulation unit operation in the carcass processing line (Davey et al., 1976a; Gilbert and Davey, 1976a; Gilbert et al., 1976b; Cross et al., 1979a; Seidemanet al., 1979; Kastner et al., 1980; Corte et al., 1980; Zakharov et al., 1980). Despite the fact that electrical stimulation depletes the ATP content and lowers muscle pH, Cross and Tennent (1981) did not find any ill effect on the chemical, physical, cooking and sensory quality characteristics of ground beef patties made from such meat. Recent studies by Contreras et al. (1981b) substantiate these findings, except that patties made with electrically stimulated, hot-boned ground beef showed slightly higher cooking losses and more intense beef flavor than that of control. Berry and Stiffler (1981) found more loss of fat during cooking of patties containing ground beef from electrically stimulated carcasses. The emulsion stability of frankfurters made from electrically stimulated carcasses was claimed to be improved over those made from unstimulated meat (Whiting et al., 1981).

Bouton et al. (1980a) have shown that stimulation of carcasses at very low voltage with a rectal probe, in general is not effective for use with hot-boning. It is necessary to employ high voltage for stimulation if carcasses are to be hot-boned. This may explain why Demeyer and Vandenriessche (1980) and Ray et al. (1980a, b) did not find any improvement in tenderness of hot-boned (muscle excised one hour post-mortem) beef carcasses that were stimulated by 110 volts. Electrical stimulation indeed reduces the intensity of cold shortening, but the risk of some toughening will be involved if the carcasses are chilled or frozen immediately after electrical stimulation.

The question then arises: "What is the appropriate time to initiate the chilling or freezing of carcasses after electrical stimulation?" It is well accepted that cold shortening does not occur to any significant degree if muscles have attained a pH 6.0 (Goll, 1968; Bendall, 1973). Some recent studies further substantiate this view (Rashid et al., 1982b; Elgasim et al., 1981). Hence, pH 6.0 can well be used as a criterion to determine the time for subjecting the carcasses to rapid cooling temperature. If blast-freezing of hot-deboned meat is designed, the muscle pH should preferably be allowed to reach 5.7 before tranferring the meat to the freezer (Bendall, 1973). This may take 3 to 4 hours if the carcasses are held at 18°C after electrical stimulation.

Energy Conservation

Many changes in fabricating the animal carcass for consumer size cuts have taken place in the past decade. Traditionally, sides of beef are chilled 18-24 hours at the slaughter facility before being quartered or divided into primal cuts and boxed for shipment. Centralized processing of meat is a growing trend which is likely to continue. Current energy and labor cost have dramatically increased, forcing the meat industry to adopt economical management. Prudent energy practices during the past four years have provided more than a 27% reduction in energy consumption. Further energy curtailment can be achieved through process innovation and the use of economic alternative types of fuel (Henrickson, 1981).

A warm beef carcass (600 lb) moving from the slaughter floor at 38°C internal temperature contains approximately 31,825 BTU's of energy. The lean portion (62%) would possess 21,500, fat 5,700, and bone 4,400 BTU's. When the bone is removed (18%) and the surplus fat trimmed (10%), the remaining edible portion will have about 24,300 BTU's. Thus, the energy requirement just to remove the heat from the

edible portion would be 25-30% less (Henrickson, 1981) than chilling the intact carcass. The two main sources of energy savings from hot-boning are cooling and transportation. Using the 1978 United States beef slaughter data, Henrickson et al. (1981) found maximum energy reduction potential of 5.08×10^{12} BTU's or 5.08 trillion BTU's of source energy. This amounted to 1.62×10^{7} BTU/head or 205 BTU's per pound of beef processed in the United States.

The refrigerated space usually provided for chilling a beef carcass is 86,400 cu. in. The space above and below the hanging carcass would usually take another 34,000 cu. in. making a per carcass total of 120,400 cu. in. An equal amount of holding space may also be provided, thus making a combined space allocation of 240,800 cu. in. per carcass. The space for a boxed carcass (bone-in primals) was estimated to be 90,000 cu. in. The edible portion by contrast may be cooled in less than 25,000 cu. in. of space either by a conveyor belt or on shelves. On this basis, the cooling space may be as much as 80% less for hot-boning (Henrickson, 1981).

Conventional chill rooms with hanging carcasses are inefficient in terms of cooling time and cost because the meat is soaking in cold air. Still air is a poor heat tranfer medium; therefore, air is blasted into the space in order to speed up the cooling process. Both of these heat transfer parameters lead to high cost operation (low refrigeration efficiency and high fan power). Since hot-boned meat is in relatively small pieces and easy to handle, the likelihood of a steady flow through a conveyorized coolor is feasible. This permits controlling the cooling media (air or liquid) so that the heat transfer rate can be enhanced. With these parameters in mind, a counter flow conveyorized cooler was conceived (Clary et al., 1968; Nelson et al., 1968; Kastner et al., 1970; Ganni, 1979). The cooler was visualized to be a circular duct with a conveyor belt traveling the centerline carrying the meat while air or liquid flows in the duct in the opposite direction. In a conventional cool room the heat transfer process is controlled by heat transferring from the meat surface to the air and the heat must be transferred from the center of the carcass to the surface by conduction. In the model conveyor, the whole process is speeded up to where heat conduction from the meat becomes the controlling factor.

Using the conveyorized chilling model with 520 head/day at an average carcass weight of 560 pounds, one would expect a 66% energy saving by cooling hot-boned meat (Ganni, 1979). Thirty-two percent was due to a reduction in mass (37.6% energy saving by cooling hot-boned meat (Ganni, 1979). Thirty-two percent was due to a reduction in mass (37.6%) and the remainder due to the improved cooling system. The capacity of the required refrigeration equipment was nearly the same for hot and cold processing. However, there was a 25-30% reduction in peak power demand due to a reduction in fan power requirements for the hot-boned system. The reduction in total energy requirements were the following:

1. About 32% of the energy reduction due to less mass
2. Nearly 90% reduction in fan power
3. Use of higher cooling air temperature
4. Only about 1/3 of the building space was required

Another important benefit of the cooling process is in reduction of inventory. It is a common practice for carcasses to hang in the chill cooler and holding cooler for a total of 24 to 48 hours. Using the conveyorized cooling system, the product may be cooled, boxed, and shipped within a 12 hour period. The benefits offered by hot processing of bovine are available only if the process can be adopted and implemented. The decision to convert to a new procedure requires careful analysis by management of relevant factors as they affect their individual firms. There appears to be sufficient evidence to encourage management to give serious consideration to the desirability of hot processing. The benefits to be gained include reduced energy, more effective use of existing

plant space, a more favorable internal rate of return on investment, and increased product yield. However, in order to obtain these benefits, capital funds are required, investment cost must be calculated, and temporary problems of process changes are inherent whenever altered operating procedures are introduced into a system. However, the benefits appear to be worth the temporary disadvantages.

Even though industry has been mainly concerned with the energy aspects of pre-rigor meat, it seems wise to point out other important factors. Some of these are plant layout, design or retrofit, labor requirement, and the distribution system. In a conversion to hot-boning, management will give first consideration to the economic impact resulting from a processing change. Dut to the variability of conditions in processing methods, equipment, plant size, land cost, etc., comparative costs would need to be made for each plant. An energy comparison using 18 departments was made by Fertuson and Henrickson (1979), using a computer model.

Research Needs

The experimental evidences discussed in this manuscript are relative to the effects of post-mortem electrical stimulation of carcasses on the biochemical, biophysical, ultrastructural, microbiological, and quality characteristics of meat. Some areas have already been pointed out in the text where the information is either lacking altogether or where more research is required to resolve the discrepancies. Apart from those, there are several other aspects of post-mortem electrical stimulation of carcasses on which the available information is merely suggestive rather than conclusive, and hence further studies are needed.

For example, the electrical parameters consisting of direct or alternating current with varying pulse duration, pulse frequencies, and wave forms were used empirically to accelerate the post-mortem glycolytic rate in the carcasses. Although some studies (Bendall, 1976; Chrystall and Devine, 1978; Ruderus, 1980; Rashid et al., 1982a) have attempted to optimize some of the electrical parameters for accelerating the glycolytic rate in carcasses, only in a few cases has the experimental design been well-planned statistically to partition the effect of different independent variables. Further studies are needed to ascertain the effect of duty cycle and of other electrical parameter interactions on the rate of glycolysis. Since the regulatory interaction at the muscle membrane surface has both potential and kinetic functions associated with specific biological events, only the specified electrical parameters (current density, pulse rate, wave form) probably would modify the kinetics of the desirable electrochemical reactions. Apart from academic interest, these studies would be of practical significance in further reducing the post-stimulation holding time of carcasses at a high temperature before chilling or hot boning to optimize tenderness.

The section (VIII-D. la) on post-mortem conditioning of meat provided the various experimental evidences which, on the basis of SDS-polyacrylamide gel electrophoresis, correlated the degradation of troponin T with an increase in tenderness of meat during aging (Olson and Parrish, 1977; Penny and Dransfield, 1979; Yamamoto et al., 1979). However, there is no apparent reason why the disappearance of a regulatory globular protein should weaken the structure of the myofibril. On the basis of its location in the myofilament and its physiological role in the contraction process, troponin T at most can be expected to affect the binding of troponin I and C to tropomyosin. Moreover, George et al. (1980) did not find any association between the post-mortem tenderizing rate of meat and the degradation rate of troponin T. These considerations suggest a need to search somewhere else than troponin T to resolve this issue.

Electron microscopic studies by many workers have indicated conspecious changes in the Z-disk of the myofibril as reviewed by Asghar and Yeates (1978) and Asghar and Pearson (1980). However, the constitution of the Z-disk was not as clearly understood then as now. The protein desmin (Lazarides, 1980) and/or connectin (Toyoda and Maryuama, 1978) is believed to constitute the network of intermediate filaments which are responsible for the structural continuity and act as a mechanical integrator in the skeletal muscle. It is likely that these filaments may be degraded due to massive contraction during electrical stimulation and during post-mortem conditioning of meat. Perhaps conventional electron microscopic techniques may not reveal these changes in desmin and/or connectin filaments at the Z-disk level, possibly due to superimposition of the tubular system of the sarcoplasmic reticulum. Differential extraction procedures may be helpful in removing the interfering elements before a detailed study of the desmin and/or connectin components can be made in relation to electrical stimulation or to the aging process of meat.

A considerable body of literature is available on the electrical control of cell function (Presman, 1970; Brighton et al., 1979) which is not pertinent to electrical stimulation of carcasses, may be useful in understanding the mechanisms of certain changes in tissues associated with electrical stimulation. We believe that electrical stimulation of carcasses does much more at the cellular level than some researchs originally thought (section VIII-D. la). Since muscle contains a host of dielectric compounds saturated with ionic liquid, they may be playing a role during electrical stimulation. A pulse shorter than 2 milliseconds is capable of generating within the cell an electrical field that may produce a polarization effect. This affect may create a much higher electrical field in its intimate environment to affect the cellular activity. The electrical potential perhaps modifies the charged species' interaction at the cell surface in the regulatory processes such as absorption, regulation of bound ions or dipoles across the membrane and the conformational changes in the molecular entities. In addition, it is known that some enzymatic reactions proceed through electrochemical mechanisms analogous to local cell theory of corrosion. The activation of certain enzymes in muscles by electrical stimulation is also probably. Future studies will clarify these speculations.

References

Abbott, M.T., Pearson, A.M., Price, J.F., and Cooper, G.R. 1977. Ultrastructural changes during autolysis of red and white porcine muscle. *J. Food Sci. 42*:1185.

Ackerman, E., Ellis, L.B., and Williams, L.E. 1979. *Biophysical Science.* p. 78, 151, Prentice-Hall, Inc., Englewood Cliffs, New Jersey.

Acton, J.C., and Saffle, R.L. 1969. Preblended and prerigor meat in sausage emulsions. *Food Technol. 23*:367.

Adelstein, R.S., and Eisenberg, E. 1980. Regulations and kinetics of the actin-myosin-ATP interaction. *Ann. Rev. Biochem. 49*:921.

Adomian, G.E., Laks, M.M., and Swan, H.J.C. 1976. The significance of contraction bands in the normal heart. *34th Ann. Proc. Electron Microscopy Soc. Amer.,* p. 312.

Adomian, G.E., Laks, M.M., and Billingham, M.E. 1978. The incidence and significance of contraction bands in endomyocardial biopsies from normal human hearts. *Am. Heart J. 95*(3):348.

Anderson, R.W. 1980. Commercial application of electrical stimulation—safety and regulatory aspects. *Proc. 26th European Meeting of Meat Research Workers,* Vol. 2, p. 102, Colorado Springs, Colorado.

Arakawa, N., Inagaki, C., Kitamura, T., Fujiki, S., and Fujimaki, S. 1976. Some possible evidences for an alteration in the actin-myosin interaction in stored muscle. *Agri. Biol. Chem. 40*:1445.

Arganosa, F.C., and Henrickson, R.L. 1969. Cure diffusion through pre- and post-chilling porcine muscle. *Food Technol. 23*:1061.

Arnold, H., Henning, R., and Pette, D. 1971. Quantitative comparison of the binding of various glycolytic enzymes to F-actin and the interaction of aldolase with G-actin. *Eur. J. Biochem. 22*:121.

Asghar, A. 1969. Biophysical, biochemical, and micro-structural aspects of lambs muscle, grown under different nutritional states, with particular reference to meat quality. Ph.D. Thesis, University of New England, Australia.

Asghar, A., and Yeates, N.T.M. 1974. Systematic procedure for the fractionation of muscle proteins, with particular reference to biochemical evaluation of meat quality. *Agri. Biol. Chem. 38*:1851.

Asghar, A., and Yeates, N.T.M. 1978. The mechanism for the promotion of tenderness in meat during post-mortem aging process. *Crit. Rev. Food Sci. Nutr. 8*(3):1.

Asghar, A., and Yeates, N.T.M. 1979a. Muscle characteristics and meat quality of lambs grown on different nutritional planes. 2. Chemical and biochemical effects. *Agri. Biol. Chem. 43*:437.

Asghar, A., and Yeates, N.T.M. 1979b. Muscle characteristics of meat and quality of lambs grown on different nutritional planes. 3. Effect on ultra-structure of muscle. *Agri. Biol. Chem. 43*:445.

Asghar, A., and Pearson, A.M. 1980. Influence of ante- and post-mortem treatments on the chemical composition and meat quality. *Adv. Food Res. 26:*:355.

Asghar, A., Rashid, N., and Henrickson, R.L. 1981. Unpublished data.

Bagshaw, C.R., and Trentham, D.R. 1974. The characterization of myosin-product complexes and product release steps. *Biochem. J. 141*:331.

Barbe, C.D., and Henrickson, R.L. 1967. Bacteriology of rapid cured ham. *Food Technol. 21*:1267.

Barrett, A.L. 1978. Lysosomal proteinases and their specificity. In *Protein Turnover and Lysosomal Function* (H.L. Segal and D.J. Doyle, eds.) p. 295, Academic Press, New York.

Beatty, C.H., and Bocek, R.M. 1970. Biochemistry of red and white muscle. In *Physiology and Biochemistry of Muscle as a Food* (E.J. Briskey, R.G. Cassens, and B.B. Marsh, eds.), Vol. 2, p. 155, University of Wisconsin Press, Madison.

Bendall, J.R. 1973. Postmortem changes in muscle. In *The Structure and Function of Muscle* (G.H. Bourne, ed.), Vol. 2, Academic Press, New York.

Bendall, J.R. 1976. Electrical stimulation of rabbit and lamb carcasses. *J. Sci. Food Agri. 27*:819.

Bendall, J.R. 1980. The electrical stimulation of carcasses of meat animal. In *Development of Meat Science–1*, p. 37. Applied Science Publishers Ltd., London.

Bendall, J.R., and Wismer-Pederson, J. 1962. Some properties of the fibrillar proteins of normal and watery pork muscles. *J. Food Sci. 27*:144.

Bendall, J.R. and Rhodes, D.N. 1976. Electrical stimulation of beef carcass and its practical application. *European Meats Conf.*, London B2:3.

Bendall, J.R., Ketteridge, C.C., and George, A.R. 1976. The electrical stimulation of beef carcasses. *J. Sci. Food Agri. 27*:1123.

Bernstein, J. 1902. Regarding the temperature coefficient of muscle energy. *Arch. Ges. Physiol. (Pfluger) 92*:521.

Berry, B.W., and Stiffler, D.M. 1981. Effects of electrical stimulation, boning, temperature, formulation, and rate of freezing on sensory, cooking and physical properties of beef patties. *J. Food Sci. 46*:1103.

Berry, B.W., Ray, E.E., and Stiffler, D.M. 1980. Effects of electrical stimulation and hot boning on sensory and physical characteristics of prerigor cooked beef roasts. *Proc. 26th European Meeting of Meat Research Workers*, Vol. 2, p. 61, Colorado Springs, Colorado.

Bird, J.W.C., Schwartz, W.M., and Spanier, A.M. 1977. Degradation of myofibrillar proteins by cathepsin B and D. *Arch. Biol. Med (Ger.) 36*:1587.

Blow, A.M.J. 1977. Action of human lysosomal elastase on the oxidised chain of insulin. *Biochem. J. 161*:13.

Bockris, J.O.'M. and Reddy, A.K.M. 1964. *Modern Electrochemistry*. Vol. 1, p. 13, Plenum Press, New York.

Bodwell, C.E., and Pearson, A.M. 1964. The activity of partially purified bovine catheptic enzymes on various natural and synthetic substrates. *J. Food Sci. 29*:602.

Bohley, P., Kirschke, H., Langner, J., Riemann, S., Wiederanders, B., Ansorge, S., and Hanson, H. 1978. Protein catabolism in rat liver cells. In *Protein Turnover and Lysosomal Function* (H.L. Segal and D.J. Doyle, eds.), p. 379, Academic Press, New York.

Bouma, J.M.W., and Gruber, M. 1964. The distribution of cathepsin B and C in rat tissue. *Biochem. Biophys. Acta. 89*:545.

Bouton, P.E., Howard, A., and Lawrie, R.A. 1958. Studies on beef quality. *Spec. Rept. Food Invest. Board*, London, 67.

Bouton, P.E., Ford, A.L., Harris, P.V., and Shaw, F.D. 1978. Effect of low voltage stimulation of beef carcasses on muscle tenderness and pH. *J. Food Sci. 43*:1392.

Bouton, P.E., Ford, A.L., Harris, P.V., and Shaw, F.D. 1980a. Electrical stimulation of beef sides. *Meat Sci. 4*:145.

Bouton, P.E., Weste, R.R., and Shaw, F.D. 1980b. Electrical stimulation of calf carcasses: Response of various muscle to different wave forms. *J. Food Sci. 45*:148.

Bouton, P.E., Shaw, F.D., and Harris, P.V. 1980c. Electrical stimulation of beef carcasses in Australia. *Proc. 26th European Meeting of Meat Research Workers*, Vol. 2, p. 23, Colorado Springs, Colorado.

Bremel, R.D., and Weber, A. 1972. Cooperation within actin filaments in vertebrate skeletal muscle. *Nature, New Biol. 238*:97.

Brighton, C.T., Black, J., and Pollack, S.R. (eds.). 1979. *Electrical properties of bone and cartilage*. Grune and Stratton, New York.

Briskey, E.J. 1964. Etiological status and associated studies of pale, soft, exudative porcine muscular tissue. *Adv. Food Res. 13*:90.

Britt, B.A., and Kalow, W. 1970. Malignant hyperthermia: aetiology unknown. *Can. Aesth. Soc. J. 17*:316.

Brooke, M.H. 1970. Some comments on neural influence on the two types of muscle fibers. In *Physiology and Biochemistry of Muscle as a Food* (E.J. Briskey, R.G. Cassens, and B.B. Marsh, eds.), Vol. 2, p. 131, University of Wisconsin Press, Madison.

Buege, D.R., and Marsh, B.B. 1975. Mitochondrial calcium and post-mortem muscle shortening. *Biochem. Biophys. Res. Commun. 65*:478.

Buja, L.J., and Roberts, W.C. 1974. The coronary arteries and myocardium in acute myocardial infarction and shock: pathologic aspects. In *Shock in Myocardial Infarction* (R.F. Gunnar, ed.), Grune and Stratton, New York.

Bury, A.F., and Pennington, R.J.T. 1975. Hydrolysis of dipeptide 2-naphthylamides by human muscle enzymes. *Biochem. J. 145*:413.

Busch, W.A., Stromer, M.H., Goll, D.E., and Suzuki, A. 1972. Ca^{2+}—specific removal of Z-lines from rabbit skeletal muscle. *J. Cell Biol. 52*:367.

Bygrave, F.L. 1979. Mitochondrial calcium transport. *Cur. Top Bioenerg. 6*:260.

Caldwell, K.A. 1970. Autolytic activity in aqueous extracts of chicken muscle. *J. Agri. Food Chem. 18*:276.

Caldwell, K.A., and Grosjean, O.K. 1971. Lysosomal cathepsins of chicken skeletal muscle. *J. Agri. Food Chem. 19*:108.

Calkins, C.R., Savell, J.W., Smith, G.C., and Murphey, C.E. 1980. Quality-indicating characteristics of beef as affected by electrical stimulation and postmortem chilling time. *J. Food Sci. 45*:133.

Canonico, P.G., and Bird, J.W.C. 1970. Lysosomes in skeletal muscle tissue. Zonal centrifugation for multiple cellular sources. *J. Cell Biol. 45*:321.

Carafoli, E., and Crompton, M. 1976. Calcium ions and mitochondria. In *Calcium in Biological Systems Symposium, Soc. Exp. Biol. 30*:89.

Carlsen, F., Knappeis, G.G., and Buchtal, F. 1961. Ultrastructure of the resting and contracted striated muscle fibre. *J. Biophys. Biochem. Cytol. 11*:95.

Carse, W.A. 1973. Meat quality and the acceleration of post-mortem glycolysis by electrical stimulation. *Food Technol. 8*:163.

Chalcroft, J.P.C., and Chrystall, B.B. 1975. Current distribution in carcasses during electrical stimulation. *Meat Ind. Res.* New Zealand Annual Report, p. 32.

Cheng, C.S., and Parrish, F.C., Jr. 1978. Molecular changes in the salt-soluble myofibrillar proteins of bovine muscle. *J. Food Sci. 43*:461.

Chrystall, B.B. 1978. The 24th European Meeting of Meat Research Workers. Kulmbach *E.7*:3.

Chrystall, B.B., and Hagyard, C.J. 1976. Electrical stimulation and lamb tenderness. *New Zealand J. Agri. Res 19*:7.

Chrystall, B.B., and Devine, C.E. 1978. Electrical stimulation, muscle tension and glycolysis in bovine sternomandibularis. *Meat Sci. 2*:49.

Chrystall, B.B., and Devine, C.E. 1980a. Electrical stimulation developments in New Zealand. *Proc. 26th European Meeting of Meat Research Workers*, Vol. 2, p. 104, Colorado Springs, Colorado.

Chrystall, B.B., Devine, C.E., and Davey, C.L. 1980b. Studies in electrical stimulation: post-mortem decline in nervous response in lambs. *Meat Sci. 4*:69.

Clarke, F., Shaw, F.D., and Morton, D.J. 1980. Effect of electrical stimulation post-mortem of bovine muscle on the binding of glycolytic enzymes. *Biochem. J. 186*:105.

Clary, B.L., Nelson, G.L., and Smith, R.E. 1968. Heat transfers from hams during freezing by low temperature air. *Transactions ASAE. 11*:496.

Cohen, C. 1975. The protein switch of muscle contraction. *Scientific Am. 233* (November), 36.

Contreras, S., Harrison, D.L., Kropf, D.H., and Kastner, C.L. 1981a. Electrical stimulation and hot boning: cooling losses, sensory properties and microbial counts of ground beef. *J. Food Sci. 46*:457.

Contreras, S., and Harrison, D.L. 1981b. Electrical stimulation and hot boning: color stability of ground beef in a model system. *J. Food Sci. 46*:404.

Cornforth, D.P., Pearson, A.M., and Merkel, R.A. 1980. Relationship of mitochondria and sarcoplasmic reticulum to cold shortening. *Meat Sci. 4*:103.

Corte, O.O., Cia, G., Picchi, V., Procknor, M.L.S.C., and Delazzari, I. 1980. Electrical associated with chilling and freezing. *Proc. 26th European Meeting of Meat Research Worker,* Vol. 2, p. 53, Colorado Springs, Colorado.

Crenwelge, D.D., Terrell, R.N., Dutson, T.R., Smith, G.C., and Carpenter, Z.L. 1980. Electrical stimulation and postmortem chilling effects on pork quality. *Proc. 26th European Meeting of Meat Research Workers,* Vol. 108, Colorado Springs, Colorado.

Cross, H.R. 1979. Effects of electrical stimulation on meat tissue and muscle properties—a review. *J. Food Sci. 44*:509.

Cross, H.R., Tennent, I., and Muse, D. 1980. Storage properties of hot and cold boned beef primals. *J. Food Qual. 4*:289.

Cross, H.R., Tennent, I. 1981. The effect of electrical stimulation and postmortem boning time on sensory and cookery properties of ground beef. *J. Food Sci. 46*:292.

Cross, H.R., Smith, G.C., Kotula, A.W., and Muse, D.A. 1979a. A research note. Effects of electrical stimulation and shrouding method on quality and palatability of beef carcasses. *J. Food Sci.* 44:1560.

Cross, H.R., Berry, B.W., and Muse, D. 1979b. Sensory and cooking properties of ground beef prepared from hot and chilled beef carcasses. *J. Food Sci. 44*:1432.

Cross, H.R., West, R.L., and Dutson, T.R. 1981. Comparison of methods for measuring sarcomere length in beef semitendinosus muscle. *Meat Sci. 5*(4):261.

Culler, R.D., Parrish, F.C., Jr., Smith, G.C., and Cross, H.R. 1978. Relationship of myofibril fragmentation index to certain chemical, physical, and sensory characteristics of bovine longissimus muscle. *J. Food Sci. 43*(4):1177.

Curtin, N.A., and Davies, R.E. 1973. ATP breakdown following activation of muscle. In *The Structure and Function of Muscle* (G.H. Bourne, ed.), Vol. III, P. 472, Academic Press, New York.

Cuthbertson, A. 1977. Hot boning of beef carcasses. *The Institute of Meat Bulletin,* Meat and Livestock Commission, Great Britain.

Cuthbertson, A. 1980. Hot processing meat. A review of the rationale and economic implication. In *Developments in Meat Science–I* (R.A. Lawrice, ed.), p. 61, Applied Science Publishers Ltd., London.

Czok, R., and Bucher, Th. 1960. Crystallized enzymes from the myogen of rabbit skeletal muscle. *Adv. Protein Chem. 15*:315.

Dalziel, C.L. 1956. Effects of electric shock on man. IEEE:*Transactions and Biomedical Engineering,* p. 44.

Davey, C.L., and Gilbert, K.V. 1976. The temperature coefficient of beef aging. *J. Sci. Food Agri. 27*:244.

Davey, C.L., Gilbert, K.V., and Carse, W.A. 1976a. Carcass electrical stimulation to prevent cold shortening toughness in beef. *New Zealand J. Agri. Res. 19*:13.

Davey, C.L., Niederer, A.F., and Graafhuis, A.E. 1976b. Effects of aging and cooking in the tenderness of beef muscle. *J. Sci. Food Agri. 27*:251.

Davies, R.E. 1963. The molecular theory of muscle contraction. *Nature 199*:1068.

Dayton, W.R., Goll, D.E., Zeece, M.G., Robson, R.M., and Reville, W.J. 1976a. A Ca^{2+}—activated protease possibly involved in myofibrillar protein turnover. Purification from porcine muscle. *Biochemistry 15*:2150.

Dayton, W.R., Reville, W.J., Goll, D.E., and Stromer, M.H. 1976b. A Ca^{++} activated protease possibly involved in myofibrillar protein turnover. Partial characterization of the purified enzyme. *Biochemistry 15*:2159.

Dean, R.T. 1978. Cellular Degradative Processes. p. 17. Chapman and Hall, London.

Dean, R.T., and Barrett, A.J. 1976. Lysosomes. Essays in *Biochemistry 12*:1.

Demeyer, D., and Vandenriessche, F. 1980. Low voltage electrical stimulation of beef carcasses: distribution of tenderizing effect in the carcass and relation to changes in sarcomere length. *Proc. 26th European Meeting of Meat Research Workers,* Vol. 2, p. 6, Colorado Springs, Colorado.

Denny-Brown, D.E. 1929. The histological features of striped muscle in relation to its functional activity. *Proc. Roy. Soc. London.* Series 12, *104*:371.

Devine, C.E., Chrystall, B.B., and Davey, C.L. 1979. Studies in electrical stimulation: effect of neuromuscular blocking agents in lamb. *J. Sci. Food Agri. 30*:1007.

Dingle, J.T., and Dean, R.T. 1976. *Lysosomes in Biology and Pathology.* Vol. 5, p. 349, North Holland Publishing Company, Amsterdam.

dos Remedios, C.G., and Gilmour, D. 1978. Is there a third type of filament in striated muscles. *J. Biochem. 84*:235.

Dransfield, E., and Jones, R.C.D. 1978. Effect of rate of chilling on the variability in texture of beef round. *J. Sci. Food Agri. 29*:601.

Dransfield, E., Jones, R.C.D., and MacFie, J.G. 1980. Quantifying changes in tenderness during storage of beef. *Meat Sci. 5*:131.

Du Bois-Reymond (1830) cited by Needham, D.M. 1971. In *Machine Carnis*, Cambridge Univ. Press.

Dubowitz, V. 1970. Differentiation of fiber types in skeletal muscle. In *Physiology and Biochemistry of Muscle as a Food* (E.J. Briskey, R.G. Cassens and B.B. Marsh, eds.), Vol. 1, p. 87, University of Wisconsin Press, Madison.

Dutson, T.R., and Lawrie, R.A. 1974. Release of lysosomal enzymes during post-mortem conditioning and their relation to tenderness. *J. Food Sci. 45*:1097.

Dutson, T.R., Smith, G.C., and Carpenter, Z.L. 1980a. Lysosomal enzyme distribution in electrically stimulated ovine muscle. *J. Food Sci. 45*:1097.

Dutson, T.R., Smith, G.C., Savell, J.W., and Carpenter, Z.L. 1980b. Possible mechanisms by which electrical stimulation improves meat tenderness. *Proc. 26th European Meeting of Meat Research Workers.* Vol. 2, p. 84, Colorado Springs, Colorado.

Eastoe, J.E., and Courts, A. 1963. *Practical Analytical Methods for Connective Tissue Proteins.* p. 111, E and F.N. Spon. Ltd. London.

Ebashi, S., and Endo, M. 1968. Calcium ions and muscle contraction. *Prog. Biophys. Mol. Biol. 18*:123.

Ebashi, S., and Nonomura, Y. 1973. Proteins of the myofibrils. In *The Structure and Function of Muscle* (G.H. Bourne, ed.), Vol. III, p. 286, Academy Press, New York.

Ebashi, S., Endo, M., and Ohtsuki, I. 1969. Control of muscle contraction. *Q. Rev. Biophys. 2*:351.

Eino, M.F., and Stanley, D.W. 1973. Catheptic activity, textural properties and surface ultrastructure of postmortem beef muscle. *J. Food Sci. 38*:45, 51.

Eisenberg, E., and Moos, C. 1968. The adenosine triphospate activity of acto-heavy meromyosin. A kinetic analysis of actin activation. *Biochemistry 7*:1486.

Eisenberg, E., and Kielley, W.W. 1972. Evidence for a refractory state of HMM and S-1 unable to bind to actin in the presence of ATP. In "Cold Spring Harbor Symposium." *Quant Biol. 37*:145.

Eisenberg, E., and Hill, T.L. 1978. A crossbridge model of muscle contraction. *Prog. Biophys. Mol. Biol. 33*:55.

Eisenberg, E., and Greene, L.E. 1980. The relation of muscle biochemistry to muscle physiology. *Ann. Rev. Physiol. 42*:293.

Eitenmiller, R.R. 1974. Cathepsin activity of penaeus setiferus muscle *J. Food Sci. 39*:6.

Elgasim, E.A., Kennick, W.H., McGill, L.A., Rock, D.F., and Soeldner, A. 1981. Effects of electrical stimulation and delayed chilling of beef carcasses on carcass and meat characteristics. *J. Food Sci. 46*:340.

Elliott, G.F., Rome, E.M., and Spencer, M. 1970. A type of contraction hypothesis applicable to all muscles. *Nature 226*:400.

Endo, M. 1977. Calcium release from sarcoplasmic reticulum. *Physiol. Rev. 57*:71.

Erickson, D.B., J.H. McCoy and J.B. Riley 1980. Hot Processing Economic Feasibility of Hot Processing Beef Carcasses. Kansas State University, Agr. Expt. Sta. Bul. 639.

Etherington, D.J. 1976. Bovine spleen cathepsin B1 and collagenolytic cathepsin: a comparative study of the properties of the two enzymes in the degradation of native collagen. *Biochem J. 153*:199.

Fabiansson, S., Jonsson, G., and Ruderus, H. 1979. Optimum conditions for very low voltage electrical stimulation of beef carcasses. *Proc. 25th Meeting of European Meat Research Workers*, Budapest.

Fenn, W.O. 1923. A quantitative comparison between the energy liberated and the work performed by the isolated satorius muscle of the frog. *J. Physiol. 58*:175.

Fenoglio, J.J., Jr., and Irey, N.S. 1977. Myocardial changes in malignant hyperthermia. *Am. J. Pathology 89*:51.

Ferguson, E.J., and Henrickson, R.L. 1979. Final report on energy conservation in the Meat Processing Industry. U.S. Dept. of Energy contract EY-76-5-05-5097.

Flory, P.J., and Rehner, J. 1943. Statistical mechanics of cross-linked polymer networks. II. Swelling. *J. Chem. Phys. 11*:521.

Fields, P.A. 1976. Proteolysis of the sarcoplasmic reticulum by cathepsin B_1. Ph.D. Thesis, Texas A&M University, College Station, TX.

Follett, M.J., Norman, G.A., and Ratcliffe, P.W. 1974. The ante-rigor excision and air cooling of beef semimembranous muscles at temperatures between −5°C and +15°C. *J. Food Technol. 9*:509.

Forrest, J.C., and Briskey, E.J. 1967. Response of striated muscle to electrical stimulation. *J. Food Sci. 32*:483.

Forrest, J.C., Judge, M.C., Sink, J.D., Hoekstra, W.G., and Briskey, E.J. 1966. Prediction of the time course of rigor mortis through response to muscle tissue to electrical stimulation. *J. Food Sci. 31*:13.

Frank, G.B. 1980. The current view of the source of trigger calcium in excitation-contraction coupling in vertebrate skeletal muscle. *Biochem. Pharmacol. 29*(18):2399.

Fung, D.Y.C., C.L. Kastner, M.C. Hunt, M.E. Dikeman and D.H. Kropf. 1980. Mesophile and psychrotroph populations on hot-boned and conventionally processed beef. *J. Food Prot. 43*:547-550.

Fung, D.Y.C., C.L. Kastner, C.Y. Lee, M.C. Hunt, M.E. Dikeman and D.H. Kropf. 1981. Initial chilling rate effects on bacterial growth on hot-boned beef. *J. Food Prot.* 44-No. 7, 539-544.

Galvani, L. 1791. De viribus electricitatus in motu musculari commentarius. Cited by Bockris, J.O'M., and Reddy, A.K.N. (1974). In *Modern Electrochemistry* 1:13.

Gann, G.L., and Merkel, R.A. 1978. Ultrastructural changes in bovine Longissimus muscle during post-mortem aging. *Meat Sci. 2*:129.

Ganni, V. 1979. Design procedure for conveyorized chilling and freezing of hot boned beef. Ph.D. Thesis, Oklahoma State University, Stillwater, Oklahoma.

Gauthier, G.F. 1970. The ultrastructure of three fiber types in mammalian skeletal muscle. In *Physiology and Biochemistry of Muscle as a Food* (E.J. Briskey, R.G. Cassens, and B.B. Marsh, eds.), Vol. 2, p. 103, University of Wisconsin Press, Madison.

George, A.R., Bendall, J.R., and Jones R.C.D. 1980. The tenderizing effect of electrical stimulation of beef carcasses. *Meat Sci 4*:51.

Gilbert K.V. 1978. Accelerated aging of beef. *Proc. 20th Ann. Meat Industry Research Conf.* New Zealand.

Gilbert, K.V., and Davey, C.L. 1976a. Carcass electrical stimulation and early boning of beef. *New Zealand J. Agri. Res. 19*:429.

Gilbert, K.V., Davey, C.L., and Newton, K.G. 1976b. Electrical stimulation and the hot boning of beef. *New Zealand J. Agri. Res. 20*:139.

Gill, C.O. 1980. Effect of electrical stimulation on meat spoilage floras. *J. Food Protec. 43*:190.

Goldman, D.E. 1943. Potential, impedance, and rectification in membranes. *J. Gen Physiol. 43*:37.

Goldman, R.D., Milsted, A., Schloss, J.A., Starger, J., and Yerna, M.J. 1979. Cytoplasmic fibers in mammalian cells: cytoskeletal and contractile elements. *Ann Rev. Physiol. U1*:703.

Goll, D.E. 1968. Resolution of rigor mortis. *Proc. 21st Ann. Reciprocal* Meat Conf., p. 16.

Goll, D.E., Okitani, A., Dayton, W.A., and Reville, W.J. 1978. A Ca^{2+}− activated muscle protease in myofibrillar protein turnover. In *Protein Turnover and Lysosomal Function* (H.L. Segal and D.J. Doyle, eds.), p. 587, Academic Press, New York.

Granger, B.L., and Lazarides, E. 1978. The existence of an insoluble Z-disk scaffold in chicken skeletal muscle. *Cell 15*:1253.

Greaser, M.L. 1977. Mechanism of calcium uptake and binding of sarcoplasmic reticulum and mitochondria. *Am. Meat Sci. Assoc. Conf.*,p. 149.

Greaser, M.L. 1981. Muscle proteins. Proc. Reciprocal Meat Conf. *Am. Meat Sci. Assoc. 31* (in press).

Greaser, M.L., Cassens, R.G., Briskey, E.J., and Hoekstra, W.G. 1969. Postmortem changes in subcellular functions from normal and pale, soft, exudative porcine muscle. 2. Electron microscopy. *J. Food Sci. 32*:125.

Griffin, C.L., Stiffler, D.M., Ray, E.E., Ridenour, K.W., Berry, B.W., Noble, R., and Rierson, R. 1979. Effects of stimulation on hot-boned pre-rigor chilled post-rigor industrially and microwave cooked biceps femoris roasts. *Proc. West Sec. Am. Soc. Anim. Sci. 30*:102.

Griffin, C.L., Stiffler, D.M., Ray, E.E., and Berry, B.W. 1981. Effects of electrical stimulation, boning time, and cooking method on beef roasts. *J. Food Sci. 46*:987.

Grusby, A.H., West, R.L., Carpenter, J.W., and Palmer, A.Z. 1976. Effects of electrical stimulation on tenderness. *J. Anim. Sci. 42*:253. (Abstr.).

Gutman, H.R., and Fruton, J.S. 1948. On the proteolytic enzymes of animal tissues. *J. Biol. Chem. 174*:851.

Hall, L.C., Savell, J.W., and Smith, G.C. 1980. Retail appearance of electrically stimulated beef. *J. Food Sci. 45*:171.

Hallund, O., and Bendall, J.R. 1965. The long term effect of electrical stimulation on the post-mortem fall of pH in muscles of Landrace pigs. *J. Food Sci. 30*:296.

Hamm, R., 1975. Water-holding capacity of meat. In *Meat* (D.A.A. Cole and R.A. Lawrie, eds.), p. 32, Butterworths, London.

Hamm, R. 1976. New results in meat biochemistry, changes after slaughter. *Fleishwirtschaft 56*:79.

Harrington, W.F. 1971. A mechanochemical mechanism for muscle contraction. *Proc. Natl. Acad. Sci. USA 68*:685.

Harrington, W.F. 1979. Contractile proteins of muscle. In *The proteins* (H. Neurath and R.H. Hill, eds.), Vol. 6, p. 246. Academic Press, New York.

Harrington, W.F. 1981. Muscle contraction. *Carol. Biol. Readers 114*:31.

Harsham, A., and Deatherage, F.E. 1951. Tenderization of meat. U.S. Patent 2544581.

Haselgrove, J.C. 1975. X-ray evidence for conformational changes in the myosin filaments of vertebrate striated muscle. *J. Mol. Biol. 92*:113.

Hasselbach, W. 1975. In Molecular Basis of Motility (L.M.G. Heilmeyer, J.C. Ruegg, and R.L. Wieland, eds.), p. 81, Heiedelberg, New York.

Hasselbach, W. 1977. The sarcoplasmic calcium pump—a most efficient ion translocating system. *Biophys. Struct. Mechanism 3*:43.

Henderson, D.W., Goll, D.E., and Stromer, M.H. 1970. A comparison of shortening and Z-line degradation in postmortem bovine, porcine, and rabbit muscle. *Am. J. Anat. 128*:117.

Henrickson, R.L. 1975. Hot boning. *Proc. of the Meat Ind. Res. Conf.*, p. 25.

Henrickson, R.L. 1979. Electricity and beef tenderness: a paper presented at the American Association of Meat Processors, Caesars Palace, Las Vegas.

Henrickson, R.L. 1981. Energy aspects of prerigor meat. *Proc. Reciprocal Meat Conf., Am. Meat Sci. Assoc. 31* (in press).

Henrickson, R.L., and McQuiston, F.C. 1977. A study of hot-beef boning for energy conservation. In *Energy Conservation in Meat Plants. Am. Soc. Heating, Refrig. Air Condo. Engin. Semiannual Meet. Chicago.*

Henrickson, R.L., Parr, A.F., Cagle, E.D., Arganosa, F.C., and Johnson, R.G. 1969. *Proc. 15th European Congress of Meat Research Workers,* Helsinki.

Henrickson, R.L., Farmer, D.M., Okras, M.R., and Wilson, P.W. 1981. Energy requirements for meat production and distribution, section G. In *Processing and Utilization of Red Meat and Meat Products.* Handbook of Agriculture, CRC Press, Inc., Boca Raton, Florida.

Hill, T.L. 1975. Theoretical formalism for the sliding filament model of contraction of striated muscle. *Prog. Biophys. Mole. Biol. 28*:269.

Hoar, W.S. 1975. *General and Comparative Physiology*. p. 848. Englewood Cliffs, Prentice-Hall, Inc., New Jersey.

Hodgkin, A.L. 1965. *The conduction of the nervous impulse*. Liverpool University, Liverpool, U.K.

Hodgkin, A.L., and Katz, B. 1949. The effect of sodium ions on the electrical activity of the giant axon of the squid. *J. Physiol. 108*:37.

Honikel, K.O., and Hamm, R. 1978. Influence of cooling and freezing on minced pre-rigor muscle on the breakdown of ATP and glycogen. *Meat Sci. 2*:181.

Honikel, K.O., Fischer, C., Hamid, A., and Hamm, R. 1981a. Influence of post-mortem changes in bovine muscle on the water-holding capacity of beef. Postmortem storage of muscle at 20°C. *J. Food Sci. 46*:1.

Honikel, K.O., Hamid, A., Fischer, C., and Hamm, R. 1981b. Influence of post-mortem changes in bovine muscle on the water-holding capacity of beef. Post-mortem storage of muscle at temperature between 0 and 30°C. *J. Food Sci. 46*:23.

Houlier, B., Valin, C., Monin, Sale, O. 1980. Is electrical stimulation efficiency muscle dependent? In *Proc 26th European Meeting of Meat Research Workers*, Vol. 2, p. 81, Colorado Springs, Colorado.

Huang, F.L., and Tappel. A.L. 1971. Action of cathepsins C and D in protein hydrolysis. *Biochim. Biophys Acta. 236*:739.

Hubbard, B.D., and Lazarides, E. 1978. Co-purification of actin and desmin from chicken smooth muscle. *J. Cell Biol. 79*:273a (Abstr. No. MI 1747).

Huston, R.B., and Krebs, E.G. 1968. Activation of skeletal muscle phosphorylase kinase by Ca^{++}. II. Identification of the kinase activating factor as a proteolytic enzyme. *Biochemistry 7*:2116.

Huxley, A.F. 1957. Muscle structure and theories of contraction. *Prog. Biophys. Biophysical Chem. 7*:255.

Huxley, A.F. 1974. Muscular contraction. *J. Physiol. 243*:1.

Huxley, A.F., and Simmons, R.M. 1971. Proposed mechanism of force generation in striated muscle. *Nature 233*:533.

Huxley, H.E. 1971. The structural basis of muscular contraction. *Proc. Royl. Soc. London. B. 160*:442.

Huxley, H.E., Simmons, R.M., Farugui, A.R., Kress, M., Bordas, J., and Koch, M.H.J. 1981. Millisecond time-resolved changes in x-ray reflections from contracting muscle during rapid mechanical transients, recording using synchroteon radiation. *Proc. Natl. Acad. Sec. USA 78*(4):2297.

Inesi, G., and Malan, N. 1976. Mechanisms of calcium release in sarcoplasmic reticulum. *Life Sci. 18*:773.

Iodice, A.A., Leong, V., and Weinstock, I.M. 1966. Separation of cathepsins A and D of skeletal muscle. *Arch. Biochem. Biophys. 117*:477.

Iodice, A.A., Chin, J., Parker S., and Weinstock, I.M. 1972. Cathepsins A, B, C, D and autolysis development of breast muscle of normal and dystrophic chickens. *Arch. Biochem. Biophys. 152*:166.

Isaacs, H., Frere, G., and Mitchell, J. 1973. Histological, histochemical and ultramicroscopic finding in muscle biopsies from carriers of the trait for malignant hyperpyrexia. *Br. J. Anaesth. 45*:860.

Ishiura, S., Murofushi, H., Suzuki, K., and Imahori, K. 1978. Studies of a calcium-activated neutral protease from chicken skeletal muscle. *J. Biochem. 84*:225.

Jacobs, D.K., and Sebranek, J.G. 1980. Use of prerigor beef and frozen ground beef patties. *J. Food Sci. 45*:648.

Jeacocke, R.E. 1977. The temperature dependence of anaerobic glycolysis in beef muscle held in a linear temperature gradient. *J. Sci. Food Agri. 27*:551.

Jeremiah, L.E., and Martin, A.H. 1980. The effects of electrical stimulation on retail acceptability and case-life of beef. *Proc. 26th European* Meeting of Meat Research Workers, Vol. 2, p. 30, Colorado Springs, Colorado.

Jonsson, G., Fabiansson, S., and Nilsson, H. 1978. In *Proc. 24th European Meat Research Workers Congress,* E10, 1. Kulmbach, Germany.

Joseph, A.L., Dutson, T.R., and Carpenter, Z.L. 1980. Morphology and calcium uptake of bovine sarcoplasmic reticulum as affected by electrical stimulation and time postmortem. *Proc. 26th European Meeting of Meat Research Workers,* Vol. 8, p. 77, Colorado Springs, Colorado.

Judge, M.D., Reeves, E.S., and Aberle, E.D. 1980. Effect of electrical stimulation on thermal shrinkage temperature of bovine muscle collagen. In *Proc. 26th European Meeting of Meat Research Workers,* Vol. 2, p. 74, Colorado Springs, Colorado.

Kang, C.K., Donnelly, T.H., Jodlowski, R.F., and Warner, W.D. 1981. Partial purification and characterization of a neutral protease from bovine skeletal muscle. *J. Food Sci. 46*:702.

Karpatkin, S., Helmreich, E., and Cori, C.F. 1964. Regulation of glycolysis in muscle. II. Effect of stimulation and epinephrine in isolated frog sartorius muscle. *J. Biol. Chem. 239*:3139.

Kastner, C.L. 1977. Hot processing: update on potential energy and related economics. *Proceedings of the Meat Industry Research Conference, American Meat Institute Foundation* in cooperation with American Meat Science Association, Chicago, IL, p. 43.

Kastner, C.L., and Russell, T.S. 1975. Characteristics of conventionally and hot-boned bovine muscle excised at various conditioning periods. *J. Food Sci. 40*:747.

Kastner, C.L., Henrickson, R.L., Clary, B.E. 1970. Cooling of porcine ham by oil immersion. *J. Food Sci. 35*:673.

Kastner, C.L., Henrickson, R.L., and Morrison, R.D. 1973. Characteristics of hot-boned bovine muscles. *J. Anim. Sci. 36*:484.

Kastner, C.L., Sullivan, D.P., Ayaz, M., and Russell, T.S. 1976. Further evaluation of conventional and hot-boned bovine longissimus dorsi muscle excised at various conditioning periods. *J. Food Sci. 41*:97.

Kastner, C.L., Dikeman, M.E., Nagele, K.N., Lyon, M., Hunt, M.C., and Kropf, D.H. 1980. Effects of carcass electrical stimulation and hot boning on selected beef muscles. In *Proc. 26th European Meeting of Meat Research Workers,* Vol. 2, p. 40, Colorado Springs, Colorado.

Katz, B. 1967. *Nerve, muscle and synapse.* McGraw Hill Book Company, New York.

Keiloa, H., and Keil, B. 1969. Isolation and specificity of cathepsin B. *FEBS Letters 4*:295.

Kirschke, H., Langner, J., Wiederanders, B., Ansorge, S., and Bohley, P. 1977. A new proteinase from rat liver lysosomes. *Eur. J. Biochem. 74*:293.

Kopp, J., and Valine, C. 1981. Can muscle lysosomal enzymes affect collagen post-mortem. *Meat Sci. 5*(4):319.

Korn, E.D. 1978. Biochemistry of actomyosin-dependent cell molitity. *Proc. Natl. Acad. Sci. USA 75*:588.

Kotula, A.W. 1980. Bacteria associated with electrically stimulated and hot boned meat. In *Proc. 26th European Meeting of Meat Research Workers,* Vol. 2, Colorado Springs, Colorado.

Kotula, A.W., and Emswiler-Rose, B.S. 1981. Bacteriological quality of hot-boned primal cuts from electrically stimulated beef carcasses. *J. Food Sci. 46*(2):471.

Kowalski, S.J. 1960. *Electrotechnology.* John F. Rider Publishing, Inc., New York.

Landmann, W.A. 1963. Enzymes and their influence on meat tenderness. In *Proceeding Meat Tenderness Symposium,* p. 87, Campbell Soup Company Camden, New Jersey.

Lange, W., Matthaus, H.U., and Radtke, H.J. 1973. Internal electrical resistance in the body of cattle. (Ger.). *Arch. Fur Experimentelle Veterinarmedizin 27*(4):653.

Lapresle, C., and Webb, T. 1962. The purification and properties of a proteolytic enzyme, rabbit cathepsin B, and further studies on cathepsin D. *Biochem. J. 84*:455.

Larner, J., Ray, B.R., and Crandall, H.F. 1956. Pattern of action of crystalline muscle phosphorylase on glycogen as determined from molecular size distribution studies. *J. Am. Chem. Soc. 78*:5890.

Lawrie, R.A., Manners, D.J., and Wright, A. 1959. -1-4 glucosans. Glycogen structure and rigor mortis in mammalian muscles. *Biochem. J. 73*:485.

Lazarides, E. 1980. Intermediate filaments as mechanical integrators of cellular space. *Nature 283*:249.

Lazarides, E., and Hubbard, B.D. 1976. Immunological characterization of the subunit of the 100 A filaments from muscle cells. *Proc. Natl. Acad. USA. 73*:4344.

Lebez, D., and Kopitar, M. 1970. Leucocyte proteinases. Low molecular weight cathepsins F and G. *Enzymologia 39*:271.

Lee, K.S., Ladinsky, H., Choi, S.J., and Kasuya, Y. 1966. Studies *in* vitro interaction of electrical stimulation and Ca^{++} movement in sarcoplasmic reticulum. *J. Gen. Physiol. 49*:689.

Leet, N.G., Devine, C.E., Law, N.G., and Davey, A.B. 1975. The Meat Industry Research Institute, New Zealand Ann. Res. Retit. MIRINZ *511*:41.

Locker, R.H. 1960. Degree of muscular contraction as a factor in tenderness of beef. *Food Res. 25*:304.

Locker, R.H. 1976. Meat tenderness and muscle structure. *Proc. New Zealand Meat Industry Research Conference 18*:1.

Locker, R.H., and Hagyard, C.J. 1963. A cold shortening effect in beef muscles. *J. Sci. Food Agri. 14*:787.

Locker, R.H., and Daines, G.J. 1974. Bioscience Division. The *Meat Industry Research Institute of New Zealand.* Annual Research Report MIRINZ *443*:33.

Locker, R.H., and Daines, G.J. 1979. In *Fibrous proteins scientific, industrial and medical aspects* (D.A.D. Parry and L.K. Creamer, eds.). Vol. 2, Academic Press, London.

Lohmann, K. 1931. Preparation of adenylpyrophosphoric acid from muscle. *Biochem. Z. 233*:460 (*Chem. Abstr. 25*, 3681, 1931).

Lopez, C.A., and Herbert, E.W. 1975. *The Private Franklin, The Man and His Family.* 1st ed., p. 44, W.W. Norton and Company, New York.

Lymn, R.W. 1979. Kinetic analysis of myosin and actomyosin ATPase. *Ann. Rev. Biophys. Bioeng. 8*:145.

Lymn, R.W., and Taylor, E.W. 1971. Mechanism of adenosine triphosphate hydrolysis by actomyosin. *Biochemistry 10*:4617.

MacDougall, D.B. 1977. Colour in meat. In *Sensory properties of foods* (G.G. Birch, J.G. Brennan, and K.J. Parker, eds.), 59.

MacLennan, D.H., and Holland, P.C. 1975. Calcium transport in sarcoplasmic reticulum. *Ann. Rev. Biophys. Bioeng. 4*:337.

Maier, D.M., and Zaiman, R. 1965. The development of lysosomes in rat skeletal muscle in trichinous myositis. *J. Histochem. Cytochem. 14* (5):396.

Mandigo, R.W., and Henrickson, R.L. 1966. Influence of hot-processing pork carcasses on cured ham. *Food Technol. 20*:538.

Mani, R.S., Herasymowych, O.S., and Kay, C.M. 1980. Physical, chemical and ultrastructural studies on muscle M-line proteins. *Intl. J. Biochem. 12*(3):333.

Marcus, Abraham 1964. Basic Electricity, 2nd ed. Prentice-Hall, Inc., Englewood Cliffs, N.J. Pg. 264.

Marsh, B.B., and Leet, N.G. 1966. Studies in meat tenderness. III. The effects of cold shortening on tenderness. *J. Food Sci. 31*:450.

Marsh, B.B., Woodhams, P.R., Leet, N.G. 1968. Studies in meat tenderness. 5. The effects on tenderness of carcass cooling and freezing before the completion of rigor mortis. *J. Food Sci. 33*:12.

Marston, S.B., Rodger, C.D., and Tregear, R.T. 1976. Changes in muscle crossbridges when β, ν-imido ATP binds to myosins. *J. Mol. Biol. 104*:263.

Marston, S.B., Tregear, R.T., Rodger, C.D., and Clarke, M. 1979. Coupling between the enzymatic site of myosin and mechanical output of muscle. *J. Mol. Biol. 128*:111.

Maruyama, K., Matsubara, S., Natori, R., Nonomura, Y., Kimura, S., Ohashi, K., Murakami, F., Handa, S., and Eguchi, G. 1977. Connectin, and elastic protein of muscle. *J. Biochem. 82*:317.

Maruyama, K., Kimura, S., Toyoda, N., and Ohashi, K. 1979. In *Fibrous proteins: scientific industrial and medical aspects* (D.A.B. Parry and L.K. Creamer, eds.), Vol. 2, Academic Press, London.

Mateucci, (1838) cited by Needham, D.M. 1971. *Machina Carnis*, Cambridge University Press, Cambridge.

Matsuda, K., and Misaka, E. 1974. Studies on cathepsins of rat liver lysosomes. 1. Purification of multiple forms. *J. Biochemistry 76*:639.

McClare, C.W.F. 1971. Biochemical machines, Maxwell's demon and living organisms. *J. Theor. Biol. 30*:1.

McClare, C.W.F. 1972. A molecular energy muscle model. *J. Theor. Biol. 35*:569.

McCollum, P.D., and Henrickson, R.L. 1977. The effect of electrical stimulation on the rate of postmortem glycolysis in some bovine muscles. *J. Food Qual.* 1.

McDonald, J.K., Zeitman, B.B., Reilly, T.J., and Ellis, S. 1969. New observation on the substrate specificity of cathepsin C (Dipeptidyl aminopeptidase I). *J. Biol. Chem. 244*:2693.

McKeith, F.K., Savell, J.W., Smith, G.C., Dutson, T.R., and Shelton, M. 1979. Palatability of goat meat from carcasses electrically stimulated at four different stages during the slaughter-dressing sequence. *J. Anim. Sci. 40*:972.

McKeith, F.K., Smith, G.C., Dutson, T.R., Savell, J.W., Hostetler, R.L., and Carpenter, Z.L. 1980a. Electrical stimulation of intact or split steer and cow carcasses. *J. Food Protec. 43*:795.

McKeith, F.K., Smith, G.C., Savell, J.W., Dutson, T.R., Carpenter, Z.L., and Hammons, D.R. 1980b. Electrical stimulation of mature cow carcasses. *J. Anim. Sci. 50*:694.

McKeith, F.K., Smith, G.C., Savell, J.W., Dutson, T.R., Carpenter, Z.L., and Hammons, D.R. 1981. Effects of certain electrical stimulation parameters on quality and palatability of beef. *J. Food Sci. 46*:13.

McLoughlin, J.V. 1970. Muscle contraction and post-mortem pH changes in pig skeletal muscle. *J. Food Sci. 35*:717.

Metrione, R.M., Neves, A.G., and Fruton, J.S. 1966. Purification and properties of dipeptidyl transferase (cathepsin C). *Biochemistry 5*:1597.

Meyer, W.L., Fischer, E.H., and Krebs, E.G. 1964. Activation of muscle phosphorylase B kinase by Ca^{++}. *Biochemistry 3*:1033.

Meyerhof, O. 1921. Energy exchange in muscle. IV. Lactic acid formation in minced muscle. *Arch. Ges. Physiol.* (Pfluger's) *188*:114.

Miyamoto, H., and Kasai, M. 1973. Re-examination of electrical stimulation on sarcoplasmic reticulum fragments *in vitro*. *J. Gen. Physiol. 62*:773.

Moeller, P.W., Fields, P.A., Dutson, T.R., Landmann, W.A., and Carpenter, Z.L. 1976a. Effect of high temperature conditioning on subcellular distribution and levels of lysosomal enzymes. *J. Food Sci. 41*:216.

Moeller, P.W., Fields, P.A., Dutson, T.R., Landmann, W.A., and Carpenter, Z.L. 1976b. High temperature effects on lysosomal enzyme distribution and fragmentation of bovine muscle. *J. Food Sci. 42*:510.

Mommaerts, W.F.H.M. 1969. Energetics of muscular contraction. *Physiol. Rev. 49*:427.

Morgan, W. 1979. The effects of electrical stimulation and conventional handling of beef on beef quality. *J. Sci. Food Agri. 30*:1103 (Abstr.).

Mrigadat, B., Smith, G.C., Dutson, T.R., Hall, L.C., Hanna, M.O., and Vanderzant, C. 1980. Bacteriology of electrically stimulated rabbit, pork, lamb and beef carcasses. *J. Food Protec. 43*:686.

Murray, J.M., and Weber, A. 1974. The cooperative action of muscle protein. *Scientific American 230* (Feb.):58.

Nachmansohn, D. 1973. The neuromuscular junction—The role of acetylcholine. In *The structure and Function of Muscle* (G.H. Bourne, ed.), Vol. III, p. 32, Academic Press, New York.

Needham, D.M. 1971. *Machina Carnis*. Cambridge University Press, Cambridge, U.K.

Needham, D.M. 1973. Biochemistry of muscle. In *The Structure and Function of Muscle* (G.H. Bourne, ed.), Vol. III, p. 364, Academic Press, New York.

Nelson, G.L., Clary, B.L., Smith, R.H., and Henrickson, R.L. 1968. Heat transfer in commercial cuts of meat. *Proc. Meat Ind. Res. Conf. Am. Meat Assoc.*, Litho, Ill.

Nichols, J.E., and Cross, H.R. 1980. Effect of electrical stimulation and early post-mortem excision on pH decline, sarcomere length and color in beef muscles. *J. Food Protec. 43*:514.

Nilsson, H., Ruderus, H., and Fabiansson, S. 1979. Meat quality characteristics of very low voltage stimulated beef carcasses. *Proc. 25th European Meeting of Meat Research Workers*, Budapest, Hungary.

Ninjoor, V., Taylor, S.L., and Tappel, A.L. 1974. Purification and characterization of rat liver lysosomal cathepsin B2. *Biochem. Biophys. Acta. 370*:880.

Noble, M.I.M., and Pollack, G.H. 1977. Molecular mechanisms of contraction. *Circ. Res. 40*:333.

Okitani, A., and Fujimaki, M. 1972. A neutral proteolytic system responsible for post-mortem proteolysis in rabbit skeletal muscle. *Agri. Biol. Chem. 36*:1265.

Okitani, A., Suzuki, A., Yang, R., and Fujimaki, M. 1972. Effect of cathepsin D treatment in ATPase activity of rabbit myofibril. *Agri. Biol. Chem. 36*(7):2135.

Okitani, A., Shinohara, K., Sugitani, M., and Fujimaki, M. 1973. A relation between the rate of increment in nonprotein nitrogenous compounds and muscle pH during post-mortem storage. *Agri. Biol. Chem. 37*:321.

Okitani, A., Otsuka, Y., Sugitani, M., and Fugimaki, M. 1974. Some properties of neutral proteolytic system in rabbit skeletal muscle. *Agri. Biol. Chem. 38*:573.

Okitani, A., Goll, D.E., Stromer, M.H., and Robson, R.M. 1976. Intracellular inhibitor of a Ca^{++} −activated protease involved in myofibrillar protein turnover. *Fed. Proc. 35*:1746.

Okitani, A., Otsuda, Y., Katakai, R., Kondo, Y., and Kato, H. 1981. Survey of rabbit skeletal muscle peptidase active at neutral pH region. *J. Food Sci. 46*:47.

Olson, D.G., and Parrish, J.C., Jr. 1977. Relationship of myofibril fragmentation index to measures of beef steak tenderness. *J. Food Sci. 42*:506.

Olson, D.G., Parrish, J.C., and Stromer, M.G. 1976. Myofibril fragmentation and shear resistance of three bovine muscles during post-mortem storage. *J. Food Sci. 41*:1036.

Ono, K. 1971. Lysosomal enzyme activation and proteolysis of bovine muscle. *J. Food Sci. 36*:838.

Otsuka, Y., Okitani, A., Kondo, Y., Kato, H., and Fujimaki, M. 1980. Further characterization of the aminopeptidase of rabbit skeletal muscle aminopeptidases. *Agri. Biol. Chem. 44*:1617.

Otto, K., and Riesenkonig, H. 1975. Improved purification of cathepsin B2. *Biochim. Biophys. Acta. 379*:462.

Ouali, A., and Valine, C. 1981. Effect of muscle lysosomal enzymes and calcium activated neutral proteinase on myofibrillar ATPase activity: relationship with aging changes. *Meat Sci. 5*:233.

Parsons, M.E., and Pennington, R.J.T. 1976. Separation of rat muscle aminopeptidases. *Biochem. J. 155*:375.

Parsons, M.E., Godwin, K.O., and Pennington, R.J.T. 1979. Further studies on aminopeptidases of rat muscle. *Intl. J. Biochem. 10*:217.

Parrish, F.C., Jr., and Bailey, M.E. 1967. Physicochemical properties of bovine muscle particulate cathepsin. *J. Agri. Food Chem. 15*:88.

Parrish, F.C., Jr., Vandell, C.J., and Culler, R.D. 1979. Effect of maturity and marbling on the myofibril fragmentation index of bovine longissimus muscle. *J. Food Sci. 44*:1668.

Parrish, F.C., Jr., Selvig, C.J., Culler, R.D., and Zeece, M.G. 1981. CAF activity, calcium concentration, and the 30,000-dalton component of tough and tender bovine longissimus muscle. *J. Food Sci. 46*:308.

Pearson, A.M. 1977. Effects of pH and temperature on calcium uptake and release by sarcoplasmic reticulum. *Proc. Recip. Meat Conf. 30*:155.

Pellegrino, C., and Franzini, C. 1963. An electron microscope study of denervated atrophy in red and white skeletal muscle fibers. *J. Cell Biol. 17*:327.

Penny, I.F. 1980. The enzymology of conditioning. In *Developments in Meat Sci. 1*, p. 115. Applied Science Publishers, Ltd., London.

Penny, I.F., and Ferguson-Pryce, R. 1979. Measurement of autolysis in beef muscle homogenates. *Meat Sci. 3*:121.

Penny, I.F., and Dransfield, E. 1979. Measurement of autolysis in beef muscle homogenates. *Meat Sci. 3*:121.

Perry, S.V. 1979. The regulation of contractile activity in muscle *Biochem. Soc. Trans. 7*:593.

Peter, J.B., Barnard, R.J., Edgerton, V.R., Gillespie, C.A., and Stempel, K.E. 1972. Metabolic profile of three types of skeletal muscle in guinea pigs and rabbits. *Biochemistry 11*:2627.

Poldosky, R.J., Onge, R., Yu, L., and Lymn, R.W. 1976. X-ray diffraction of activity shortening muscle. *Proc. Natl. Acad. Sci. USA 64*:504.

Pollard, T.D., Weihing, R.R. 1974. Actin and myosin and cell movement. *CRC Crit. Rev. Biochem. 2*:1.

Presman, A.S. 1970. *Electromagnetic Fields and Life.* Plenum Press, New York.

Press, E.M., Porter, R.R., and Cebra, J. 1960. The isolation and properties of a proteolytic enzyme, cathepsin D from bovine spleen. *Biochem. J. 74*:501.

Raccach, M., and Henrickson, R.L. 1978. Storage stability and bacteriological profile of refrigerated ground beef from electrically stimulated hot-boned carcasses. *J. Food Protec. 41*:957.

Randall, C.J., and MacRae H.F. 1967. Hydrolytic enzymes in bovine skeletal muscle. II. Proteolytic activity of the water-soluble proteins separated by Starch gel electrophoresis. *J. Food Sci. 32*:182.

Ranvier, L. 1874. De quelques faits relatifs a l'histologie et a la physiologie des muscles stries. *Arch. Physiol. Normal et Phathol. 1*:5.

Rashid, N., Asghar, A., Henrickson, R.L., and Claypool, P.L. 1982a. Evaluation of certain electrical parameters for the stimulation of lamb carcasses. *J. Food Sci.* (under preparation).

Rashid, N., Asghar, A., Henrickson, R.L., and Claypool, P.L. 1982b. The biochemical and quality characteristics of ovine muscles as affected by electrical stimulation. *J. Food Sci.* (under preparation).

Ray, E.E., Stiffler, D.M., Benge, A., and Berry, B.W. 1978. Influence of electrical stimulation, insulation, and high temperature chilling upon muscle pH, temperature and palatability factors of lightweight beef carcasses. *Proc. West. Sec. Am. Soc. An. Sci. 29*:131.

Ray, E.E., Stiffler, D.M., and Berry, B.W. 1980a. Effects of electrical stimulation and hot boning upon physical changes, cooking time and losses, and tenderness of beef roasts. In *Proc. 26th European Meeting of Meat Research Workers*, Vol. 2, p. 26, Colorado Springs, Colorado.

Ray, E.E., Stiffler, D.M., and Berry, B.W. 1980b. Effects of hot boning and cooking methods upon cooking time and losses, and tenderness of roasts from electrically stimulated beef carcasses. In *Proc. 26th European Meeting of Meat Research Workers*, Vol. 2, p. 90, Colorado Springs, Colorado.

Reddy, M.K., Etlinger, J.D., Rabinowitz, M., Fischman, D.A., and Zak, R. 1975. Removal of Z-lines and -Actinin from isolated myofibrils by a calcium-activated neutral protease. *J. Biol. Chem. 250*:4278.

Rentschler, H.C. 1951. Apparatus and method for the tenderization of meat. *U.S. Patent* 2,544,724.

Riley, R.R., Savell, J.W., and Smith, G.C. 1980a. Storage characteristics of wholesale and retail cuts from electrically stimulated lamb carcasses. *J. Food Sci. 45*:1101.

Riley, R.R., Savell, J.W., Smith, G.C., and Shelton, M. 1980b. Quality, appearance and tenderness of electrically stimulated lamb. *J. Food Sci. 45*:(1):199.

Reville, W.J., Goll, D.E., Stromer, M.H., Robson, R.M., and Dayton, W.R. 1976. A Ca^{++}-activated protease possibly involved in myofibrillar protein turnover. *J. Cell. Biol. 70*:1.

Robson, R.M., Stromer, M.H., Huiatt, T.W., O'Shea, J.M., Hartzer, M.K., Richardson, F.L., and Rathbun, W.E. 1980. Biochemistry and Structure of desmin and the recently discovered muscle cell cytoskeleton. In Proc. 26th European Meeting of Meat Research Workers, Vol. 1, p. A6, Colorado Springs, Colorado.

Ruderus, H. 1980. Low voltage electrical stimulation of beef. Influence of pulse types on post mortem pH fall and meat quality. In *Proc. 26th European Meeting of Meat Research Workers*, Vol. 2, p. 96, Colorado Springs, Colorado.

Salm, C.P., Mills, E.W., Reeves, E.S., Judge, M.D., and Aberle, E.D. 1981. Effect of electrical stimulation on muscle characteristics of beef cattle fed a high energy diet for varying lengths of time. *J. Food Sci. 46*:1284.

Saris, N.E., and Ackerman, K.E.O. 1980. Uptake and release of bivalent cations in mitochondria. *Curr. Top. Bioenerg. 10*:103.

Savell, J.W., Smith, G.C., Dutson, T.R., Carpenter, Z.L., and Suter, D.A. 1977. Effect of electrical stimulation on palatability of beef, lamb and goat meat. *J. Food Sci. 42*:702.

Savell, J.W., Dutson, T.R., Smith, G.C., and Carpenter, Z.L. 1978a. Structural changes in electrically stimulated beef muscle. *J. Food Sci. 43*:1606.

Savell, J.W., Smith, G.C., and Carpenter, Z.L. 1978b. Effect of electrical stimulation on quality and palatability of light-weight beef carcasses. *J. Anim. Sci. 46*:1221.

Savell, J.W., Smith, G.C., and Carpenter, Z.L. 1978c. Beef quality and palatability as affected by electrical stimulation and cooler aging. *J. Food Sci. 43*:1666.

Savell, J.W., Smith, G.C., Carpenter, Z.L., and Parrish, F.C. 1979. Influence of electrical stimulation on certain characteristics of heavy-weight beef carcasses. *J. Food Sci. 44*:911.

Schmidt, G.R., and Gilbert, K.V. 1970. The effect of muscle excision before the onset of rigor mortis on the palatability of beef. *J. Food Technol. 5*:331.

Schmidt, G.R., and Keman, S. 1974. Hot boning and vacuum packaging of eight major bovine muscles. *J. Food Sci. 39*:140.

Schwartz, W.N., and Bird, J.W.C. 1977. Degradation of myofibrillar proteins by cathepsin B and D. *Biochem. J. 167*:811.

Scopes, R.K. 1974. Studies with a reconstituted muscle glycolytic response to stimulated tetanic contraction. *Biochem. J. 138*:119.

Seidman, S.C., Smith, G.C., Dutson, T.R., and Carpenter, Z.L. 1979. Physical, chemical and palatability traits of electrically stimulated, hot-boned, vacuum-packaged beef. *J. Food Protec. 42*:651.

Seippel, R.G. 1974. *Fundamentals of Electricity.* p. 364. American Technical Society, Chicago.

Seperich, G.J., and Price, J.F. 1981. The similarities between the activities of the calcium-activated sarcoplasmic factor and the kinase activating factor from rabbit skeletal muscle. *Meat Sci. 5*:17.

Shaw, F.D., and Walker, D.J. 1977. Effect of low voltage stimulation of beef carcasses on muscle pH. *J. Food Sci. 42*:1140.

Slurzberg, M., and Osterheld, W. 1965. *Essentials of Electricity–Electronics*, p. 40. McGraw-Hill Book Company, New York.

Small, J.V., and Sobieszek, A. 1977. Studies on the function and composition of the 10 nm filaments of vertebrate smooth muscle. *J. Cell Sci. 23*:243.

Smith, B. 1965. The enzyme histochemistry of experimental myopathy. *Res. Muscular Distrophy Proc. 3rd Symp.*, p. 133.

Smith, G.C., Dutson, T.R., Carpenter, Z.L., and Hostetler, R.L. 1977. Using electrical stimulation to tenderize meat. *Proc. Meat Ind. Res. Conf. Amer. Meat Inst. Found. Chicago. 29*:147.

Smith, G.C., Dutson, T.R., Carpenter, Z.L. 1979. Electrical stimulation of hide-on and hide-off calf carcasses. *J. Food Sci. 44*:335.

Smith, G.C., Savell, J.W., Dutson, T.R., Hostetler, R.L., Terrell, R.N. Murphey, C.E., and Carpenter, Z.L. 1980. Effects of electrical stimulation on beef, pork, lamb and goat meat. In *Proc. 26th European Meeting of Meat Research Workers,* Vol. 2, p. 19, Colorado Springs, Colorado.

Snyder, H.E. 1965. Analysis of pigments at the surface of fresh beef with reflectance spectrophotometry. *J. Food Sci. 30*:457.

Snyder, H.E., and Armstrong, D.J. 1967. An analysis of reflectance spectrophotometry as applied to meat model systems. *J. Food Sci. 32*:241.

Sonaiya, E.B., and Stouffer, J.R. 1981. Tensioning of electrically stimulated carcasses for improved tenderness. *J. Food Sci.* (in press).

Sorinmade, S.O., Cross, H.R., and Ono, K. 1978. The effect of electrical stimulation on lysosomal enzyme activity, pH decline and beef tenderness. *Proc. 24th Meeting of the European Meat Research Workers,* Vol. 2, E-9, Kulmbach, Germany.

Spencer, M., Morthington, C.R. 1960. An hypothesis of contraction in striated muscle. *Nature 187*:388.

Squire, J.M. 1975. Muscle filament structure and muscle contraction. *Ann. Rev. Biophys. Bioeng. 4*:137.

Starkey, P.M., and Barrett, A.J. 1976. Neutral proteinases of human spleen. Purification and criteria for homogeneity of elastase and cathepsin G. *Biochem. J. 155*:255.

Starkey, P.M., Barrett, A.J., and Burleigh, M.C. 1977. The degradation of articular collagen by neutrophil proteinases. Biochem. Biophys. Acta. *483*:386.

Starlinger, V.H. 1967. Uber die Bindung der Muskeladolase an grobdisperse partikeln in Homogenaten erregter Musckeln. *Hoppe-Selyer's Physiol. Chem. 348*:864.

Stein, L.A., Schwartz, R., Chock, P.B., and Eisenburg, E. 1979. The mechanism of the actomyosin ATPase: evidence that ATP hydrolysis can occur without dissociation of the actomyosin complex. *Biochemistry 18*:3895.

Stern, N.J. 1980. Effect of boning, electrical stimulation and medicated diet on the microbiological quality of lamb cuts. *J. Food Sci. 45*:1749.

Stewart, M.R., Ziper, M.W., and Watts, B.M. 1965. The use of reflectance spectrophotometry for the assay of raw meat pigments. *J. Food Sci. 30*:464.

Stiffler, D.M., and Ray, E.E. 1979. Effect of electrical stimulation on muscle pH and shear force for steers and short-scrotum carcasses. *Proc. West Sec. Am. Soc. Anim. Sci. 20*:64.

Stiffler, D.M. Ray, E.E., and Harp, R.M. 1978. Effects of electrical stimulation on beef muscle pH and tenderness. *Proc. West. Sec. Am. Soc. An. Sci. 29*:151.

Straus, W. 1967. Lysosomes, phagosomes and related particles. In *Enzyme Cytology* (D.E. Roodyn, ed.), p. 239, Academic Press, New York.

Strickland, P.E., Lester, T.I., and West, R.L. 1979a. Effects of electrical stimulation and aging on beef tenderness and processing characteristics. *J. Anim. Sci. 49,* Suppl. 1:18 (Abstr. No. 46).

Strickland, P.E., West, R.L., and Palmer, A.Z. 1979b. Comparison of pulsatory and continous electrical stimulatin treatments on intact and split beef carcasses. *J. Anim. Sci. 49,* Suppl. 1:18 (Abstr. N. 47).

Sutoh, K., Sutoh, K., Karr, T., and Harrington, W.F. 1978. Isolation and physicochemical properties of a high-molecular weight subfragment-2 of myosin. *J. Mol. Biol. 126*:7.

Suzuki, A., Saito, M., Sato, H., and Nonami, Y. 1977. Reconstitution of the Z-disk. *Agri. Biol. Chem. 41*:1095.

Suzuki, A., Saito, M., Sato, H., and Nonami, Y. 1978. Effect of materials released from myofibrils by Ca^{++} -activated factor on Z-disk reconstitution. *Agri. Biol. Chem. 42*:2111.

Swatland, H.J. 1976a. An electrocorticographic study of necrobiosis in the brains of electrically stunned and exasanguinated pigs. *J. Anim. Sci. 43*:577.

Swatland, H.J. 1976b. Motor unit activity in excised prerigor beef muscle. *Can. Inst. Food Sci. Technol. J. 9*:177.

Swatland, H.J. 1977. Sensitivity of prerigor muscle to electrical stimulation. *Can. Inst. Food Sci. Technol. J. 10*:280.

Szent-Gyorgyi, A. 1951. *Chemistry of muscular contraction.* Academic Press, New York.

Takahashi, K., and Saito, H. 1979. Post-mortem changes in skeletal muscle connectin. *J. Biochem. 85*:1539.

Tallan, H.H., Jones, M.E., and Fruton, J.S. 1952. On the proteolytic enzymes of animal tissues. X. Beef spleen cathepsin C. *J. Biol. Chem. 194*:793.

Tang, B.H., and Henrickson, R.L. 1980. Effect of post-mortem electrical stimulation and bovine myoglobin and its derivatives. *J. Food Sci. 45*:1139.

Tappel, A.L. 1969. In *Lysosomes in Biology and Pathology.* (J.T. Dingle and H.B. Fell, eds.), Vol. 2, p. 167, Elvesiver, North-Holland, Amsterdam.

Taylor, E.W. 1979. Mechanism of actomyosin ATPase and the problem of muscle contraction. *CRC Crit. Rev. Biochem. 6*:103.

Taylor, D.G., and Marshall, A.R. 1980a. Low voltage electrical stimulation of beef carcasses. *J. Food Sci. 45*:144.

Taylor, A.A., Shaw, B.G., and MacDougall, D.B. 1980b. Hot deboning beef with and without electrical stimulation. In *Proc. 26th European Meeting of Meat Research Workers*, Vol. 2, p. 45, Colorado Springs, Colorado.

Thompson, J.T. 1981. The effect of electrical current on hot boned pork quality. *M.S. Thesis*, Oklahoma State University, Stillwater, Oklahoma.

Toyoda, N., and Maruyama, K. 1978. Fine structure of connectin nets in cardiac myofibrils. *J. Biochem. 84*:239.

Trautman, J.C. 1964. Fat-emulsifying properties of pre-rigor and post-rigor pork proteins. *Food Technol. 18*:1065.

Tsong, T.Y., Karr, T., and Harrington, W.F. 1979. Rapid helix-coil transitions in the S-2 region of myosin. *Proc. Natl. Acad. Sci. USA 76*:1109.

Turnsek, T., Kregar, I., and Lebez, D. 1975. Acid sulphydryl protease from calf lymph nodes. *Biochem. Biophys. Acta. 403*:514.

Tregear, R.T., and Marston, S.B. 1979. The crossbridge theory. *Ann. Rev. Physiol. 41*:723.

Tume, R.K. 1979. Post-mortem electrical stimulation of muscle and its effects on sarcoplasmic reticulum adenosine triphosphatase. *Aust. J. Biol. Sci. 32*:163.

Tume, R.K. 1980. Effect of post-mortem electrical stimulation on ovine sarcoplasmic reticulum vesicles. *Aust. J. Biol. Sci. 33*:43.

Turina, M., and Jenny, E. 1968. Studies on the *in vitro* uptake of Ca^{++} in sarcoplasmic reticulum of rabbit skeleton. Influence of monophasic electrical stimulation of exactly defined strength and of cardiac glycosides. *Cariologia 53*:193.

Valine, C. 1981. Tenderizing meat by electrical stimulation (Fr.). *Resherches 12*(122):612.

Van Fleet, J.F., Hall, B.V., and Simon, J. 1968. Vitamin E deficiency. A sequential light and electron microscopic study of skeletal muscle. Degeneration in wealing rabbit. *Amer. J. Pathol. 52*(4) 1067.

Varity, M.A., and Coleman, R.F. 1973. Histoenzymatic methods applied to human striated muscle disease. In *The striated muscle* (C.M. Pearson and F.K. Mostofi, eds.), p. 28, The Williams and Wilkins Co., Baltimore.

Walker, D.J., Harris, P.V., and Shaw, F.D. 1977. Accelerated processing of beef. *Food Technol. Australia 29*:504.

Walsh, T.P., Clarke, F.M., and Masters, C.J. 1977. Modification of the kinetic parameters of aldolase on binding to the actin-containing filaments of skeletal muscle. *Biochem. J. 165*:165.

Weber, A., and Murray, J.M. 1973. Molecular control mechanisms in muscle contraction. *Physiol. Rev. 53*:612.

Weber, H.H., and Portzehl, H. 1952. Muscle contraction and fibrous muscle proteins. *Adv. Protein Chem. 7.*

Weiner, P.D., Kropf, D.H., Mackintosh, D.L., and Koch, B.A. 1966. Effect on muscle quality of processing pork carcasses within one hour post-mortem. *Food Technol. 20*:541.

Wells, L.H., Berry, B.W., and Douglass, L.W. 1980. Effects of grinding and mechanical desinewing in the manufacture of beef patties using conventionally chilled and hot boned and rapidly chilled mature beef. *J. Food Sci. 45*:163.

West, R.L., Moeller, P.W., Link, B.A., and Landmann, W.A. 1974. Loss of calcium accumulating ability in the sarcoplasmic reticulum following degradation by cathepsins. *J. Food Sci. 39*:29.

West, R.L., Langston, D.L., and Oblinger, J.L. 1980. Storage stability and tenderness of top round roasts cooked pre- or post-rigor following electrical stimulation. In *Proc. 26th European Meeting of Meat Research Workers*, Vol. 2, p. 57, Colorado Springs, Colorado.

Westervelt, R.G., and Stouffer, J.R. 1978. Relationship among spinal cord severing, electrical stimulation and postmortem quality characteristics of the porcine carcass. *J. Anim. Sci. 46*:5.

White, D.C.S., and Thorson, J. 1973. The kinetics of muscle contraction. *Prog. Biophys. 27*:175.

Whiting, R.C. 1980. Calcium uptake by bovine muscle mitochondria and sarcoplasmic reticulum. *J. Food Sci. 45*:288.

Whiting, R.C., Strange, E.D., Miller, A.J., Bendict, R.C., and Mozersky, S.M., and Swift, C.E. 1981. Effect of electrical stimulation on the functional properties of lamb muscle. *J. Food Sci. 46*:484.

Wilkie, D.R. 1975. *Muscle as a thermodynamic machine.* CIBA Foundation *Symposium 31*(NS):327.

Will, P.A., Henrickson, R.L., Morrison, R.D., and Odell, G.V. 1979. Effect of electrical stimulation on ATP depletion and sarcomere length in delay-chilled bovine muscle. *J. Food Sci. 44*:1646.

Will, P.A., Ownby, C.L., and Henrickson, R.L. 1980. Ultrastructural postmortem changes in electrically stimulated bovine muscles. *J. Food Sci.* 45:21.

Wingard, S. 1968. Intracellular calcium movements of frog skeletal muscle during recovery from tetanus. *J. Gen. Physiol. 51*:65.

Yamamoto, K., Samejima, K., Yausi, T. 1979. Changes produced in muscle proteins during incubation of muscle homogenates. *J. Food Sci. 44*:51.

Yang, R., Okitani, A., and Fujimaki, M. 1978. Post-mortem changes in regulatory proteins of rabbit muscle. *Agri. Biol. Chem. 42*:555.

Young, O.A., Graafhuis, A., and Davey, L. 1981. Post-mortem changes in cytoskeletal proteins of muscle. *Meat Sci. 5*:41.

Zakharov, R.S., Mager, H., Moiseenko, E.N., Yatsenko, A.P. 1980. Increase in the efficiency of meat technological treatment produced by electrophysical methods used in a post-slaughtering period. In *Proc. 26th European Meeting of Meat Research Workers*, Vol. 2, p. 11, Colorado Springs, Colorado.

Zierler, K.L. 1973. Some aspects of teh biophysics of muscle. *In The Structure and Function of Muscle* (G.H. Bourne, ed.), Vol. III, p. 118, Academic Press, New York.