chloride). The rate and extent of superprecipitation of natural actomyosin from white fibres were 25–50% greater than those of natural actomyosin from red fibres. It remains to be established how the contraction times of the red fibres and the white fibres are related to their myosin adenosine triphosphatase activities.

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Post-Mortem Autolysis in Bovine Muscle

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Muscle becomes hard and inextensible during the onset of rigor (Bendall, 1960), but during subsequent storage at 2°C there is a progressive increase in tenderness. The ability to maintain isometric tension is lost (Jungk, Snyder, Goll & McConnell, 1967), but the muscle does not regain its extensibility. These changes during storage are believed to be caused by post-mortem autolysis. A study of the myofibrillar ultrastructural changes over the storage period essential to an understanding of the nature of this autolysis is reported. Proteolysis in the sarcoplasmic fraction and release of lysosomal cathepsin D were also investigated.

Bovine semitendinosus muscle was sampled immediately *post mortem* and after 24h, 4 days and 10 days storage at 2°C for light and electron microscopy. The supernatants obtained after centrifuging homogenates from 0h, 2-day and 15-day post-mortem samples for 30min at 1000g (r_{av} 7.62cm) at 4°C were applied to an exponential sucrose density gradient in the BXIV aluminium zonal rotor and centrifuged for 45min at 43000g (r_{av} 4.7cm) at 5°C. Collected fractions were analysed for protein (Schuel & Schuel, 1967) and cathepsin D activity (Anson, 1937).

The phase-contrast microscope showed a progressive increase in incidence of transverse fibre breaks during the storage period. These breaks always occurred in the I band.

Pre-rigor myofibrils in the electron microscope had the appearance of resting muscle (Huxley, 1960). The Z lines showed a well-defined fibrillar

pattern (Landon, 1970), which gradually turned amorphous during subsequent storage. The other components of the myofibril were not significantly affected by storage for 10 days. The ultrastructural locus of the fibre breaks seen in the light microscope was the Z line/I band junction (Reville, Joseph & Harrington, 1971).

The amount of sedimentable protein decreased during storage. In pre-rigor samples 52.5% of the total protein was sedimentable, whereas 2-day samples showed 27.5% and 15-day samples only 18% sedimentable protein.

The total activity of cathepsin D did not change appreciably over the storage period, but of this 84.5% was sedimentable in pre-rigor samples and 40.7% after 15 days.

The limited and specific ultrastructural disruption of the myofibrillar fraction occurring during storage would undoubtedly result in increased tenderness and loss of ability to maintain isometric tension. The sarcoplasmic fraction undergoes a more extensive autolysis, but all the cathepsin D activity does not seem to be released during storage. This agrees with Valin (1970).

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Light-Scattering Rayleigh Linewidth Measurements on G-Actin

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Light-scattering by solutions has been used as a method for finding macromolecular parameters for the past 25 years. During the past decade, however, it has become possible, largely owing to the advent of the laser, to investigate the spectral distribution of the scattered light (see Cummins, Knable & Yeh, 1964; Dubin, Lunacek & Benedek, 1967; French, Angus & Walton, 1969; Rimai, Hickmott, Cole & Carew, 1970; Pecora, 1970; Ford, Karasz & Owen, 1970; Sellen, 1970; Foord

et al. 1970). The spectral linewidth, which is of the order of 100 Hz (about 10^{-9} Å), arises from the thermal motion of the macromolecules. Translational diffusion, which makes the major contribution, leads to a random variation in phase of the scattered light, and this corresponds to a Lorentzian distribution about the incident light-frequency of half-width $(16\pi^2/\lambda^2) \cdot D \cdot \sin^2\theta/2$, where D is the translational diffusion constant, λ the wavelength and θ the angle of scatter. In addition, broadening may result from random variations in particle scattering factor as a result of rotation or changes of configuration, or of rotational variation of polarizability with optically anisotropic molecules (Pecora, 1970, and references cited therein).

Investigation of such small frequency shifts is impossible by conventional optical techniques, and the optical homodyne, made possible by the long coherence length available from a laser, is the most common technique used. In this case the quantity actually measured is (Sellen, 1970):

$$\overline{s_B^2} = 2c^2 \cdot \gamma^2 \cdot \Delta B \int_0^\infty I_{B/2-\Omega} \cdot I_{B/2+\Omega} \cdot d\Omega$$

where s_B is the reading obtained from a wave analyser at frequency B , $c^2 \cdot \gamma^2 \cdot \Delta B$ is a constant associated with the optical and electrical parameters, and I the intensity distribution function of the scattered light. In the simple case of translational diffusion $\overline{s_B^2}$ is a Lorentzian distribution of beat frequencies of twice the spectral width. However, for reasons given by information theory, it is preferable to use a correlation-function computer rather than a wave analyser. This yields the Fourier transform of the beat signal, which is the same as the square of the Fourier transform of I , and is given by $e^{-2K^2 D \tau}$ where $K = (4\pi/\lambda) \cdot \sin\theta/2$ and τ is the correlation-function delay time.

An apparatus suitable for work of this kind has been set up in this laboratory. Some preliminary results for G-actin are presented, and the particular difficulties involved with this material are discussed.

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Uptake of Calcium Ions and the β -Adrenergic Receptor Site in Cardiac Muscle

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The generation of systole and diastole in the heart presents an alternative system to skeletal muscle in the study of excitation-contraction coupling (Gillibrand, 1970). As in skeletal muscle, transport of Ca^{2+} relative to membranes is closely involved with myocardial contractility (Naylor, 1967), and Ca^{2+} flux is regulated by the sarcoplasmic reticulum (Porter & Franzini-Armstrong, 1965). In the heart both contractility and rate are augmented by noradrenaline, through the β -adrenergic receptor site. This consists minimally of a structurally organized enzyme system, adenylate cyclase and phosphodiesterase, which are known to be membrane-bound (Robison, Butcher & Sutherland, 1970). Rasmussen & Tenenhouse (1968) have proposed theoretically that Ca^{2+} transport can be linked to cyclic AMP (adenosine 3':5'-cyclic monophosphate), but the mechanism of coupling requires further investigation.

In order to demonstrate enzymic activity associated with Ca^{2+} uptake, preparations of fresh fragmented sarcoplasmic reticulum in 5 mM-dithiothreitol (Van der Kloot, 1969) were obtained by using the method of Gertz, Hess, Lain & Briggs (1967). The presence of theophylline, which inhibits the specific phosphodiesterase breaking down cyclic AMP (Butcher & Sutherland, 1962), gave a much higher Ca^{2+} uptake than in the control. Both noradrenaline and cyclic AMP also increased Ca^{2+} uptake significantly. Ouabain, which stimulates myocardial contractility in the failing heart, gave a lower value, but this was significantly above the control value. Both propranolol, a specific β -receptor-blocking drug, and practolol, which has some sympathomimetic activity, significantly decreased the Ca^{2+} uptake relative to control. Nicotine, an inhibitor of adenylate cyclase, gave the largest decrease.

By using the method of Krishna, Weiss & Brodie (1968), it was found that these membrane preparations had a high specific adenylate cyclase activity. Addition of noradrenaline significantly increased the total amount of cyclic AMP formed.

Alteration of Ca^{2+} uptake by drugs known to affect adenylate cyclase and phosphodiesterase activities indicates that the energy for Ca^{2+} uptake by cardiac microtubules may be regulated by membrane-bound adenylate cyclase. The data for cyclic AMP are consistent with the view that this nucleotide could form part of the carrier system