INVESTIGATION OF FOCAL METABOLIC LESIONS IN THE MYOCARDIUM BY POLARIZATION AND ELECTRON MICROSCOPY

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Investigations in polarized light show that 30-60 min after injection of novodrin (isopropylnoradrenalin sulfate) into animals, myocytolysis is present in the heart muscle cells. Investigations with the electron microscope have shown that the process starts with destruction of components of the sarcoplasmic reticulum and myofibrils, while the mitochondria, nucleus, and sarcolemma remain relatively intact.

Degenerative changes in heart muscle cells are always mosaic in character [3, 5, 6, 10, 11]. This mosaic pattern is based on variation in the degree and character of injury to individual cells and it suggests heterogeneity of the myocardial cell population, possibly in connection with asynchronous cycles of renewal of the intracellular structures [1, 2, 4, 6].

The writers have shown by the use of polarization microscopy that in the very early stages acute degenerative changes can be found in the heart muscle cells [3, 4, 6]. Two main types of lesions were distinguished: contractual and myocytolysis. In polarized light myocytolysis is characterized by the appearance of areas of disappearance of the anisotropy of the myofibrils in the muscle fibers. Examination in ordinary light shows that these areas take up dyes less strongly. Only a few cells with lesions of this type subsequently undergo colliquative necrosis, and most of them regain their normal structure within 48 h of the original injury [4, 7]. Myocytolysis is thus a special type of reversible injury to the myocardial cell, accompanied by partial destruction and subsequent restoration of intracellular structures. It was decided to study this process at the submicroscopic level.

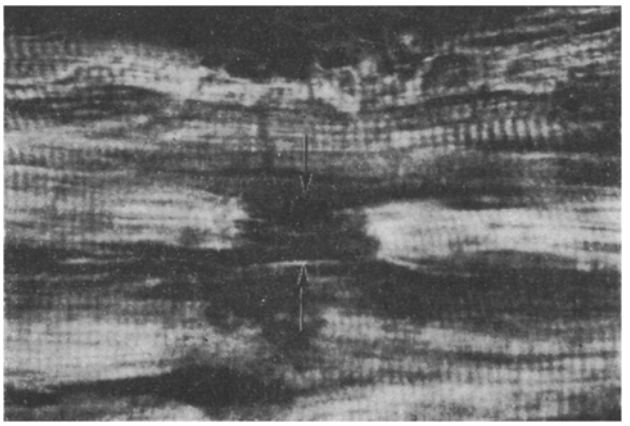

Fig. 1. Section through papillary muscle of rat sacrificed 1 h after injection of novodrin. Area of myocytolysis corresponding to electron micrographs in Figs. 2 and 3 is indicated by arrows. Photographed in polarized light, 1000×.

The initial stages of myocytolysis were examined in the investigation described below.

EXPERIMENTAL METHOD

Male and female Wistar albino rats weighing 120-180 g received a single subcutaneous injection of novodrin (isopropylnoradrenalin sulfate) in a dose of 0.8 mg/100 g body weight. The eight animals used

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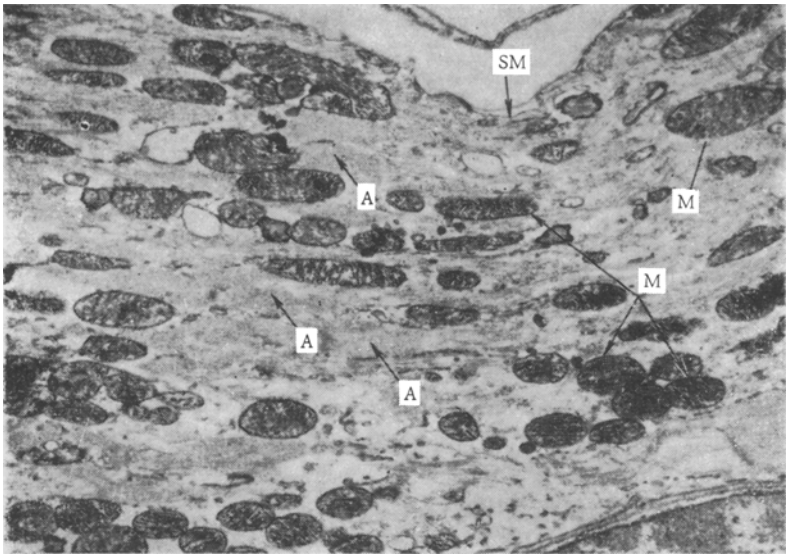

Fig. 2. Same area of myocytolysis on survey electron micrograph. Sarcolemma (SM) of muscle cell is intact, myofibrils are absent in center of focus, and intact A-disks of myofibrils (A) are visible at the periphery. Mitochondria (M) show no visible changes, 8200×.

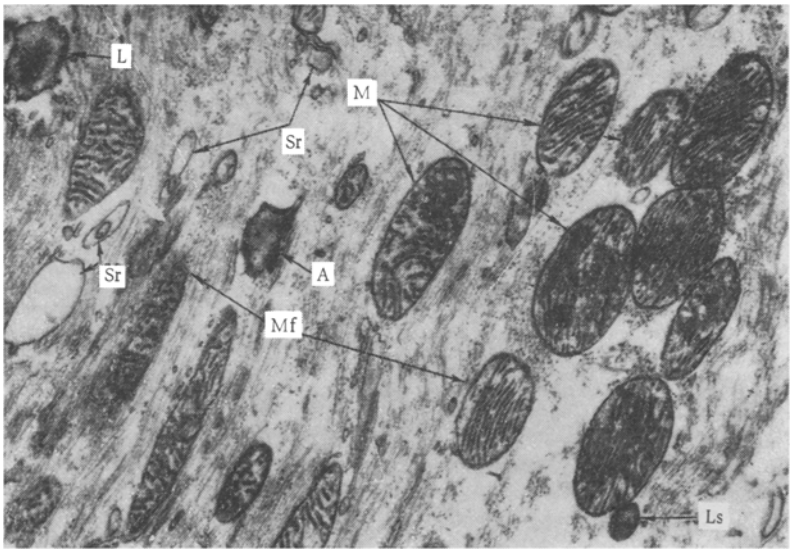

Fig. 3. Fragment of zone of myocytolysis under higher power. Fragments and bundles of myofilaments (Mf), lipid droplets (L), a lyosome (Ls), and solitary dilated tubules of sarcoplasmic reticulum (Sr) visible between mitochondria (M), 26,000×.

in the experiments were decapitated 20, 30, 45, and 60 min after the injection. The heart was stopped by lowering the temperature, the left ventricle was opened, and the papillary muscles were excised. The longitudinally divided papillary muscles were fixed at 4° in Caulfield's osmium fixative [12], dehydrated in alcohols of increasing concentration, and embedded in a mixture of methacrylates (1 : 6). For a preliminary study of the arrangement of the muscle fibers and the character of lesions in the muscle cells, sections 3-4 μ in thickness were cut from the blocks and examined in polarized light. The pyramids were centered accurately over particular groups of longitudinally arranged normal or injured muscle cells. To compare the electron micrographs with the picture obtained in polarized light, before and after the cutting of ultrathin sections on the JUM-5A ultratome, sections 1μ in thickness were obtained and photographed in polarized

light. Ultrathin sections 600-900 A in thickness were stained with lead salts [13, 14], and examined in the Tesla BS-513 electron microscope.

EXPERIMENTAL RESULTS

Unlike in experiments with adrenalin injury to the myocardium, in which foci of myocytolysis were observed to be formed during the first 15-20 min, in the case of novodrin administration myocytolysis appeared between 30 and 60 min after the injection. As a rule, in this period, myocytolysis affected small areas of the injured cell, frequently in the intercalated disk. In polarized light the zone of myocytolysis was revealed by the absence of the normal anisotropy of the A-disks of the myofibrils (Fig. 1). With the phase contrast method, lysis of the myofibrils was confirmed by translucency of the sarcoplasm and the irregular distribution of granules corresponding in their staining properties to sarcosomes [4, 7, 9], between which isolated longitudinal fibrils which had lost their cross-striation were sometimes visible.

Under low power of the electron microscope the picture of myocytolysis corresponded in its general features with that obtained in phase contrast. The structure of the myofibrils was blurred or indistinguishable, and only the groups of mitochondria, arranged in mosaic pattern, were clearly visible (Fig. 2). Whereas at the periphery of the focus of myocytolysis individual A-disks could still be seen, separated by wide spaces containing fragments of Z-bands, in the center of the focus, under high power, only detached fragments and bundles of myofilaments forming A-disks were visible. Mitochondria in the zone of myocytolysis before 1 h after injection in most cases appeared unchanged, occasionally with the matrix slightly translucent.

The almost complete absence of structures of the sarcoplasmic reticulum (Fig. 3) in the zone of the myocytolysis must be particularly noted. All that remained were single dilated tubules, mostly arranged longitudinally. Diads and triads of the sarcoplasmic reticulum, which are visible in the normal myocardium in nearly every Z-band of myofibrils, were very rare in the zone of myocytolysis.

Fragments and single intact myofilaments of A-disks of the myofibrils detected by the electron microscope were evidently the structures responsible for diffuse luminescence of the labeled antimyosin serum which was observed in an immunotopographic investigation of the early stages of myocytolysis [8].

The initial stages of myocytolysis are thus characterized by primary destruction of the sarcoplasmic reticulum and myofibrils at the Z-bands, with relative integrity of the sarcolemma, mitochondria, and nucleus.

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