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EFFECT OF CORDARONE ON CARDIOMYOCYTE ULTRASTRUCTURE IN EXPERIMENTAL MYOCARDIAL INFARCTION

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Cordarone (amiodarone) is widely used in the clinical treatment of ischemic heart disease (IHD). The mechanism of its antianginal action has been a frequent subject of research [2, 3, 5, 10, 14]. After the discovery that the drug had antiarrhythmic properties [8] indications for its use in the treatment of IHD widened considerably. However, despite many biochemical and electrophysiological investigations, the fine structural changes in the myocardium during the action of cordarone remain virtually unstudied.

The aim of this investigation was to study the dynamics of ultrastructural changes in muscle cells of the peri-infarct zone of the heart under the influence of cordarone at different times after acute experimental coronary occlusion.

EXPERIMENTAL METHOD

Experiments were carried out on 30 cats weighing 3-4 kg and divided into two groups: group 1 (control) — animals with experimental myocardial infarction (EMI) caused by ligation of the anterior descending coronary artery at the junction between its middle and lower thirds; group 2 consisted of cats with EMI receiving cordarone in a dose of 10 mg/kg intramuscularly, twice a day. The ECG of animals of both groups was recorded in standard lead II on the 3rd, 7th, and 15th days, after which the heart was removed under pentobarbital anesthesia (40 mg/kg, intravenously) for electron-microscopic investigation. Pieces of left ventricular myocardium were taken from the peri-infarct zone, fixed in 1% OsO_4 solution, dehydrated, and embedded in Araldite. The number of glycogen granules was determined morphometrically in electron micrographs obtained on the JEM-100B microscope.

EXPERIMENTAL RESULTS

Electron-microscopic investigation of the peri-infarct zone of myocardium in animals treated with cordarone revealed some differences in cardiomyocyte ultrastructure observed on the 3rd, 7th, and 15th days after ligation of the coronary artery.

Considerable degenerative changes were found in the structure of the cardiomyocytes on the 3rd day after acute coronary occlusion in the control animals. The muscle cells were edematous, the myofibrils separated, the myofilaments partially fragmented and, in some cases, in a state of over-contraction. The mitochondria were swollen and the cristae were vacuolated

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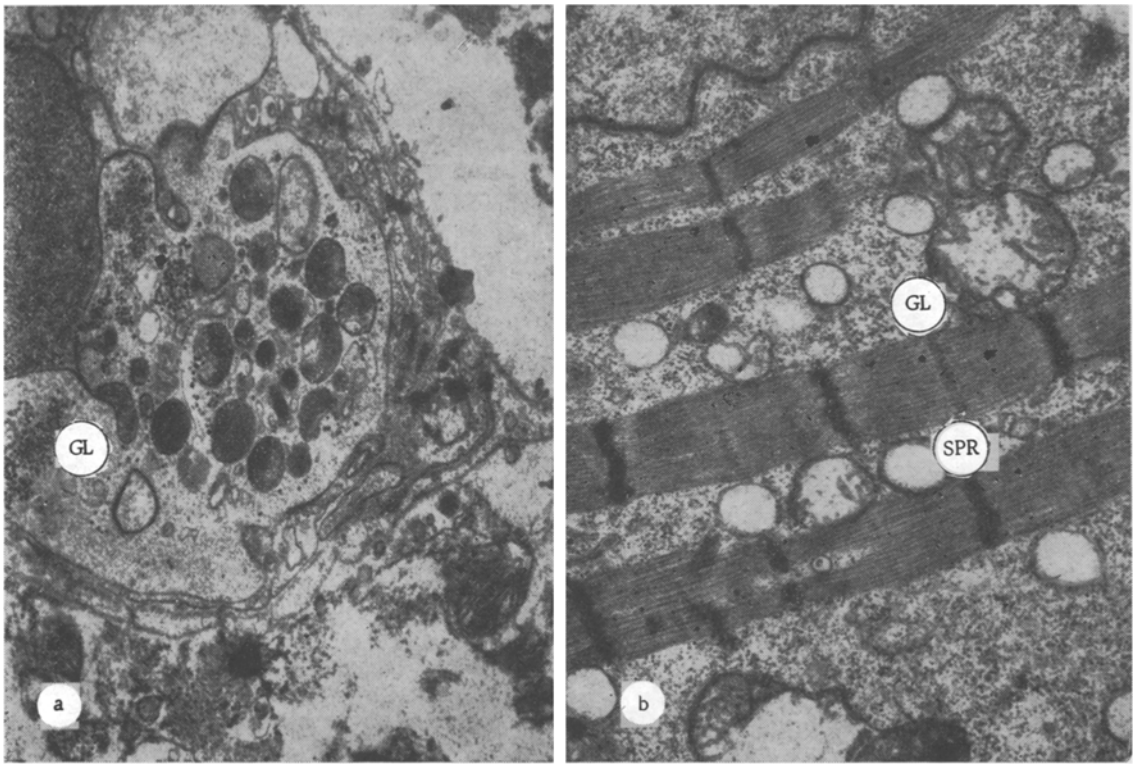

Fig. 1. Effect of cordarone on cardiomyocyte ultrastructure in peri-infarct zone: a) glycogen granules (GL) in capillary lumen on 3rd day after acute occlusion of coronary artery, 20,000 ×; b) glycogen granules (GL) and dilatation of cisterns of sarcoplasmic reticulum (SPR) on 7th day of EMI, 25,000 ×.

or locally destroyed. The matrix of the mitochondria was much more transparent. Dense, irregularly shaped osmiophilic inclusions were observed in many mitochondria. Considerable changes were observed in the structure of the nucleus: peripheral condensation of chromatin, compaction and homogenization with loss of structure of the nucleolus. Cytogranules were absent in the sarcoplasm.

In animals treated with cordarone, degenerative changes in the state of the cardiomyocytes also were observed on the 3rd day: marked edema, swelling of the mitochondria with destruction and vacuolation of the cristae, and transparency of the mitochondrial matrix. However, unlike in the control, single granules of glycogen could be seen in the cardiomyocyte cytoplasm of animals receiving cordarone. Large concentrations of glycogen granules were observed also in the capillary lumen and in the cytoplasm of the blood cells (Fig. 1a).

On the 7th day of EMI the intracellular edema in myocytes of the control animals was reduced. However, the myofibrils were still separated, the mitochondria swollen with a transparent matrix, and single cytogranules (polysomes and glycogen granules) appeared in the cytoplasm (Fig. 2a).

On the 7th day considerable accumulation of glycogen granules was observed in the animals receiving cordarone, amounting to 12.3 ± 0.9 granules per conventional unit of area of muscle cell cytoplasm (normal 1.2 ± 0.2). These granules, 30-40 nm in diameter, were distributed diffusely among the myofibrils and mitochondria, forming large concentrations in the perinuclear zone and beneath the sarcolemma (Fig. 1b). The intracellular edema was considerably reduced. The myofibrils were in a relaxed state with clearly distinguishable A and I disks. Gross swelling of the mitochondria was observed with destruction of the cristae and the cisterns of the sarcoplasmic reticulum were appreciably widened (Figs. 1b and 2b) to reach a diameter of 8-10 μm.

On the 15th day a tendency toward normalization of cardiomyocyte structure was observed in the control (Fig. 3a). Edema of the myocytes was reduced even more, and the degenerative changes in the mitochondria were less marked. Glycogen granules (3.8 ± 0.4 per conventional unit of area), ribosomes, and lipid drops were found in the cytoplasm of the muscle cells.

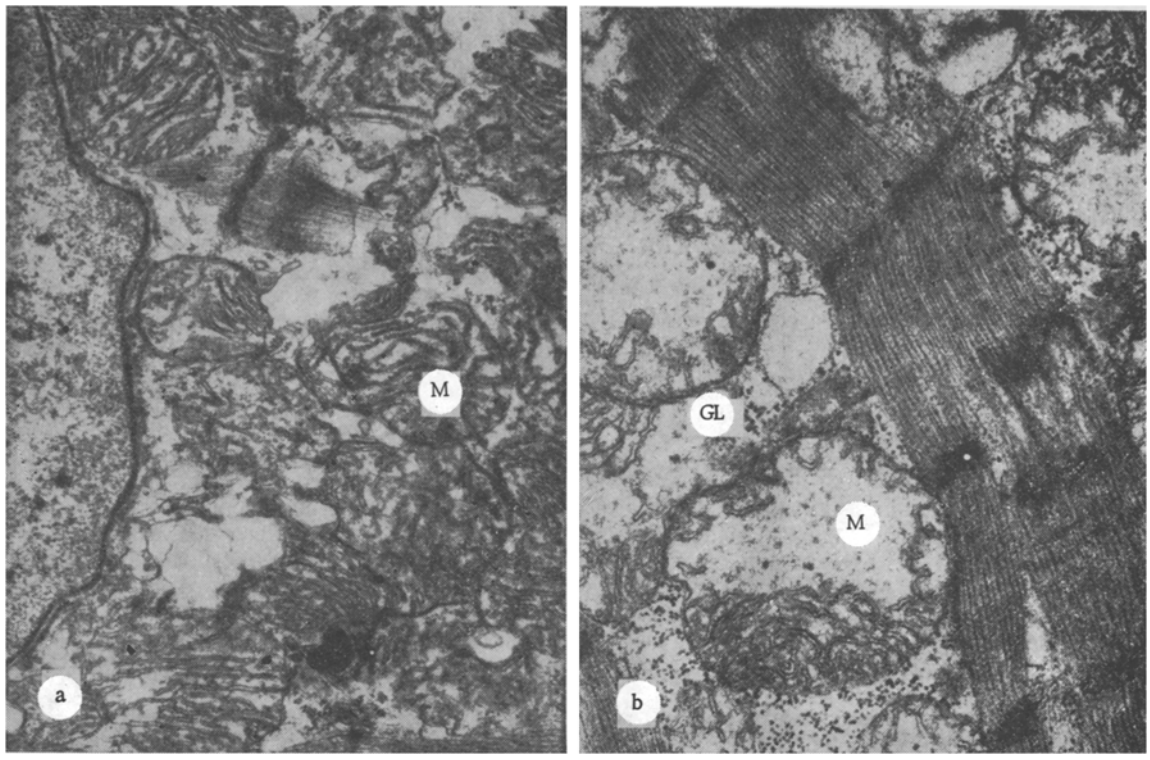

Fig. 2. Myocardial muscle cells of peri-infarct zone on 7th day: a) intracellular edema and swelling of mitochondria (M) in control, 30,000 ×; b) swelling of mitochondria (M), enlargement of glycogen granules (GL) in response to cordarone, 32,000 ×.

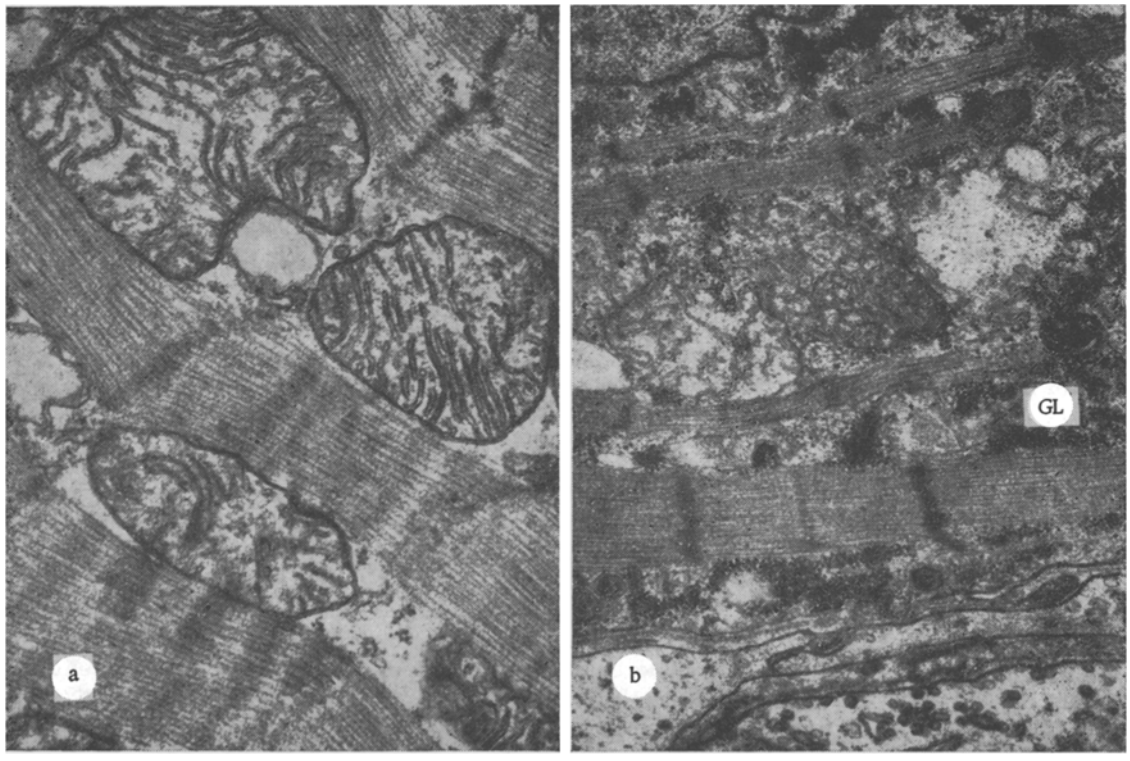

Fig. 3. Myocardial muscle cells of peri-infarct zone on 15th day: a) reduction in degree of intracellular edema and swelling of mitochondria in control, 40,000 ×; b) accumulation of glycogen granules (GL) in cytoplasm of cardiomyocytes in response to cordarone, 22,000 ×.

In animals receiving cordarone considerable accumulation of glycogen granules was observed on the 15th day in the muscle cell cytoplasm (15.1 ± 1,7 granules per conventional unit of area), to form extensive electron-dense fields (Fig. 3b). Meanwhile the structure of the mitochondria was still not fully restored: The mitochondria were enlarged, their matrix translucent, and their cristae fragmented. The structure of the nucleus was indistinguishable from normal. Myofibrils were in a state of relaxation and had the normal structure. Cisterns of the sarcoplasmic reticulum were greatly dilated.

The results of this electron-microscopic study thus showed that cordarone can exert a definite positive influence on the dynamics of repair processes in the peri-infarct zone of the heart. This was reflected in the more rapid disappearance of the intracellular edema, restoration of the structure of the myofibrils, and accumulation of glycogen granules in the myocytes than in the control, the same as is observed during the beneficial therapeutic effect of several antianginal agents [5, 6]. Meanwhile the normal structure of the mitochondria was restored more slowly than in the control and dilatation of the cisterns of the sarcoplasmic reticulum persisted for a long time.

Comparative analysis of the ECG of animals of the control group and those receiving cardarone showed only a minimal therapeutic effect of the drug.

The increase in the number of glycogen granules observed in the sarcoplasm of the myocytes in the peri-infarct zone under the influence of cordarone is in agreement with the results of biochemical investigations [11], possibly because of a decrease in excitation of adrenergic receptors and a fall in the cAMP level, caused by cordarone [7]. In turn, this effect may lead to slowing of glycolysis in the myocardium and to glycogen accumulation.

The considerable widening of the cisterns of the sarcoplasmic reticulum may indicate accumulation of calcium ions in the cardiomyocytes and calcium binding by these intracellular structures (removal from the actomyosin complex). The presence of myofibrils in the relaxation stage is indirect evidence of a fall in the calcium level in the tropomyosin complex. The ability of cordarone to lengthen the plateau of the action potential of myocardial cells and to affect the calcium current is known to be among the principal properties of the drug [13]. It is these properties which justified its inclusion in a special (the third) group of antiarrhythmic drugs.

The more marked swelling of the mitochondria after administration of cordarone than in the control may be evidence of slowing of repair processes in the peri-infarct zone, which is difficult to explain unambiguously. One cause of this swelling may be accumulation of calcium ions by these organelles. During active accumulation of Ca^{++} by the mitochondria their matrix increases in volume and translucency [1]. Mitochondria, together with the sarcoplasmic reticulum, play an important role in the calcium transport system in heart muscle, which has a high calcium capacity [9]. It has been shown that the ability of mitochondria to accumulate Ca^{++} predominates over their "trigger" function— the realization of oxidative phosphorylation [12]. However, an increase in the calcium content in the mitochondria leads to a decrease in coupling of oxidation with phosphorylation and in ATP formation, and this may make the work of the mitochondria and of the heart as a whole less efficient.

The data described above reflect the divergent character of the action of cordarone on ultrastructure of the myocardial muscle cells in the peri-infarct zone of the heart and make an unequivocal assessment of its efficacy impossible.

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