spinal asymmetry in the cerebellar (45 min) and cortical (24 h) models are noteworthy. Considering that in the first case short-axon intracerebellar and cerebellolumbar pyramidal projections are involved, it can be tentatively suggested that the signal heralding the onset of injury to nerve cells in a particular "center" is effective in the first relay of the damaged neurons, and, judging from its realization time, on account of the rapid axonal current. However, this suggestion requires direct experimental proof.

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EXPERIMENTAL PROTECTION OF THE MYOCARDIUM AGAINST ANOXIC INJURIES WITH A WEAK SOLUTION OF FORMALIN

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In modern cardiovascular surgery the advantages of operations on the arrested heart are well known — the heart is bloodless, relaxed, can easily be retracted, and so on [2, 6]. Existing methods of cardioplegia, despite their great diversity (anoxic, hypothermic, pharmacological, combined methods) are not without disadvantages. That is why the search for new methods of prolonged ischemic cardiac arrest is still an urgent problem in cardiac surgery and transplantology.

It was shown previously [5] that the use of formaldehyde solutions in low concentrations to preserve bone, cartilage, skin, blood vessels, and cardiac valves prevents proteolysis, inhibits energy metabolism, and reversibly inhibits enzymes, thus contributing to preserve the viability of the tissues. These properties of formaldehyde are due to its ability to take part in chemical dissociating reactions with organic substances of all classes possessing a mobile H^+ ion, through the formation of unstable methylol bonds RNH=CH OH. On hydrolysis of methylol compounds $RN\begin{matrix}H\\H\end{matrix} + HO{=}CH_2{=}OH$ the functional properties of proteins, lipids, and carbohydrates participating in metabolic processes are restored [3, 4, 8, 9].

It was shown previously [7] that intravenous injection of 1% formalin solution into animals causes cardiac arrest and disappearance of contractility in response to direct electrical

KEY WORDS: cardiac ischemia; cardioplegia; formaldehyde; protection of the myocardium.

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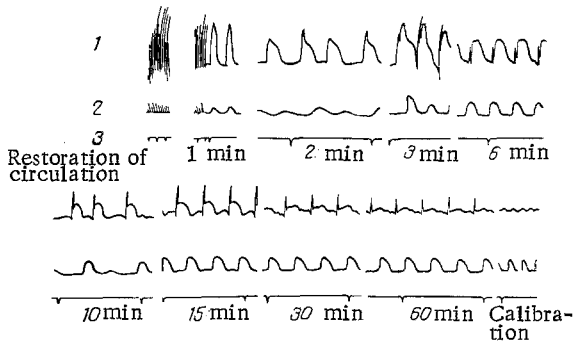

Fig. 1. ECG of transplanted rabbit's heart after cardioplegia produced by 0.25% formalin solution and ischemia lasting 1.5 h (standard lead II). 1) ECG; 2) pressure in left ventricle; 3) time marker (1 sec). Amplitude of calibration signal 1 mV.

stimulation. Resumption of the circulation in such a heart leads to restoration of its functional activity (excitability, contractility) and to normalization of metabolism. On the basis of these facts, in the investigation described below the possible use of formaldehyde solutions for cardioplegia was studied and the protective properties of this method were assessed.

EXPERIMENTAL METHOD

Experiments were carried out on 80 albino rats and 135 chinchilla rabbits. The cardioplegic solution, namely, 1, 0.5, 0.25, and 0.1% solutions of formalin in Ringer's solution (pH 7.3-7.5), was injected through a catheter which was passed up to the orifices of the coronary arteries while the venae cavae were compressed. The catheter was passed through the brachiocephalic trunk. Distal to the catheter a clamp was applied to the aorta. Formaldehyde was injected by means of a Unita automatic syringe for measured injection of solutions at the rate of 4-5 ml/min until the pressure within the left ventricle fell to zero. Cardiac activity was resumed and the viability of the myocardium verified by resuscitation perfusion of the heart, according to a model of heterotopic transplantation to the recipient's abdominal vessels. In a control series of experiments the heart was stopped by compressing the aorta and venae cavae (anoxic arrest). The electrical excitability of the myocardium was determined by direct stimulation with needle electrodes from a T-2 electrical stimulator (from Medicor, Hungary), inserted into the muscle of the left ventricle. The ECG, arterial pressure and respiration of the recipient and ECG of the transplanted heart were recorded on a Biograph polygraph (from Harvard Apparatus, USA). At the end of the experiment pieces of myocardial tissue were taken for histological, enzyme-histochemical, and electron-microscopic investigation.

EXPERIMENTAL RESULTS

The experiments showed that injection of 1.0, 0.5, 0.25, and 0.1% solutions of formalin in all experiments arrested cardiac activity, which was confirmed by the absence of electrical activity and disappearance of electrical excitability in response to stimulation (150 V).

The cardiac arrest following injection of 1% formalin solution was irreversible, as shown by absence of cardiac contractions after coronary perfusion with donor's blood for 1 h. Resumption of the circulation in the heart after injection of 0.5% formalin solution was effective if ischemia did not exceed 30 min. Cardioplegia induced by injection of 0.25% formalin solution was reversible in all experiments, and was confirmed by restoration of the cardiac rhythm by perfusion with donor's blood after ischemia for 3 h (in the control cardiac activity did not recover after ischemia lasting 1.5 h). The use of 0.1% formalin solution also caused reversible cardioplegia, but the process of arrest followed a much longer course; according to Burakovskii et al. [1] this is an undesirable feature and it adversely affects restoration of cardiac activity and the functional integrity of the myocardium. The dynamics of recovery of cardiac activity after cardioplegia produced by 0.25% formalin solution is illustrated in Fig. 1. Histological examination of the control hearts after ischemia for 1.5 h showed swelling of the myocardial muscle fibers and focal disappearance of cross-striation of the myofibrils. Investigation of succinate dehydrogenase activity showed the diffuse character of distribution of the diformazan granules; in most muscle fibers the diformazan granules were greatly enlarged, and in some fibers enzyme activity was considerably reduced. The changes revealed are evidence of a marked disturbance of mitochondrial function in the cardiomyocytes. Investigation of mitochondrial α-glycerophosphate dehydrogenase also showed

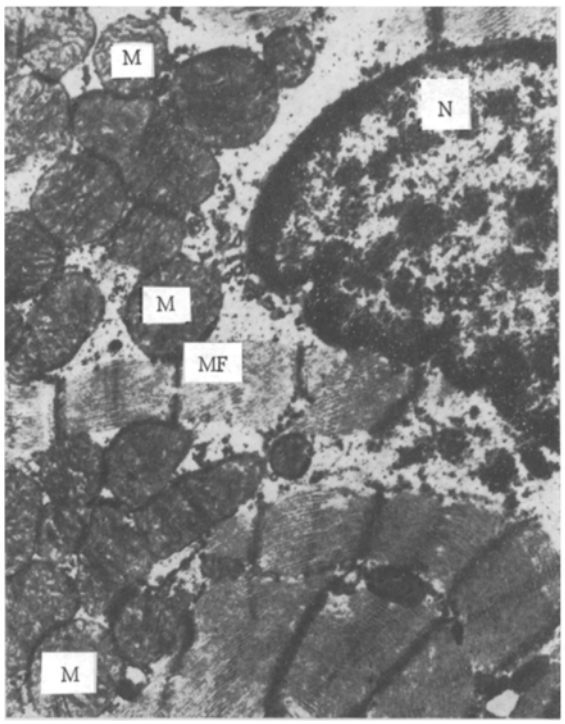

Fig. 2

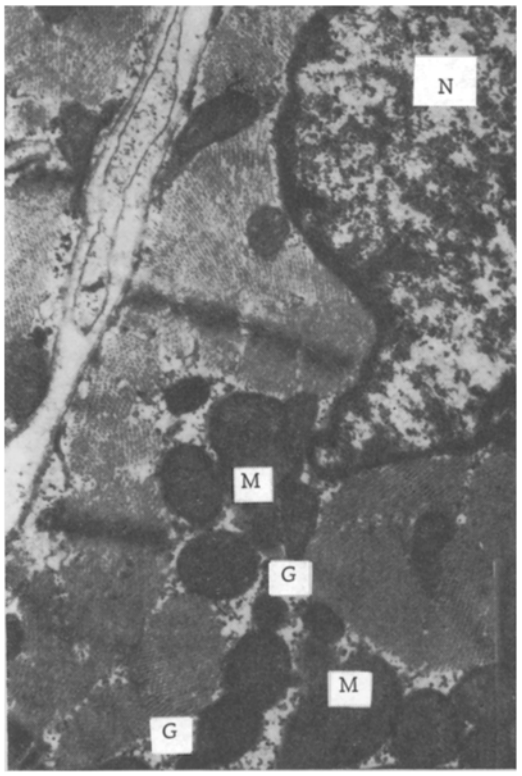

Fig. 3

Fig. 2. Cardiomyocyte after ischemia for 1.5 h. N) Nucleus; M) mitochondria; MF) myofibrils; 85,500 ×.

Fig. 3. Cardiomyocyte after cardioplegia produced by 0.25% formalin solution and ischemia lasting 1.5 h. G) Glycogen; 12,500 ×. Remainder of legend as to Fig. 2.

that the activity of this enzyme is "mosaic" in character — ranging from considerable enhancement to severe weakening. Acid phosphatase activity was increased in discrete foci. Staining with Sudan black revealed fine grains of lipids. No glycogen could be detected in the muscle fibers of the myocardium. The intima of the small vessels was greatly swollen. Perivascular and interstitial edema were present to a considerable degree.

Electron-microscopic investigation revealed translucency of the sarcoplasm of the cardiomyocytes. No glycogen granules could be detected. Clusters of ribosomes and polysomes were seen in the perinuclear zone of the sarcoplasm. Foci of lysis of myofilaments and of overcontraction of myofibrils were seen. Redistribution of chromatin and partial lysis were seen in the nuclei. Most mitochondria were deformed, their matrix was pale, and their cristae were partially fragmented and destroyed (Fig. 2). The changes described are evidence of severe disintegration of metabolic processes in the ischemized heart and they are the structural basis for absence of recovery of cardiac activity.

Compared with the heart in the control series of experiments, the perivascular and interstitial edema in the myocardium after cardioplegia with 0.25% formalin solution and 1.5 h of ischemia were much less marked. Activity of the mitochondrial enzymes was preserved at a higher level. Electron-microscopic examination revealed many glycogen granules in the muscle cells. Most mitochondria had a dense matrix and their membranes were preserved. The nuclear chromatin was uniformly distributed. The myofibrillary system was normal. The membranes of the sarcoplasmic reticulum and intercalated disks retained their structure (Fig. 3).

After heterotopic transplantation of such a heart, unlike in the control, cardiac activity was resumed. In this series of experiments it was virtually never necessary to resort to defibrillation of the heart. After short periods of fibrillation arising during the first 1-2 min of restoration of the blood flow, spontaneous restoration of cardiac contractions took place in the sinus rhythm. The pressure in the left ventricle reached 68 mm Hg. Morphological investigation revealed pericapillary hemorrhages and stasis of erythrocytes in the capillaries. High succinate dehydrogenase and α-glycerophosphate dehydrogenase activity was preserved in

most muscle fibers. Under the electron microscope, against the background of a translucent sarcoplasm, preserved mitochondria with an electron-dense matrix and parallel arrangement of the cristae could be seen. No changes were observed in the nuclei.

The experiments thus showed that cardioplegia followed by restoration of cardiac activity can be produced with the aid of 0.25% formalin solution. Formaldehyde, by blocking processes of cell metabolism, inhibits proteolysis and promotes preservation of the structure and viability of the heart under ischemic conditions.

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EFFECT OF HYPOXIA AND FENIGIDIN ON ACTION POTENTIAL DURATION AND CONTRACTILITY OF THE FROG MYOCARDIUM

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Under hypoxic conditions a reduction in the force of contraction of the myocardial fibers is accompanied by a decrease in the duration of action potentials, which is determined by the inward current through the slow calcium channels and the outward potassium current [2, 5-7].

However, changes in which of these two currents lead to a decrease in the duration of the action potentials is not clear.

Accordingly, the aim of the investigation described below was to identify the principal factors determining the duration of action potentials during hypoxia. To examine this problem the action of hypoxia and of the specific calcium channel blocker fenigidin (known in the literature as BAU-1040, nifedipine, and adalat) [1, 3, 4], on the duration of action potentials and the force of contraction of a strip of the frog ventricle was studied.

Experiments were carried out in strips of frog ventricle 3-5 mm long, placed in a continuous flow chamber 10 ml in volume. Mechanical activity of the strip was recorded by means of the 6M × 2B mechanotron and N-338-2 automatic writer; square pulses 15-30 msec in duration and with a frequency of 0.2-0.3 Hz, generated by an ÉSL-1 stimulator, were applied; the intensity of the stimuli was 3 to 4 times greater than the threshold. The transmembrane action potentials were recorded by glass microelectrodes. The duration of the action potentials

KEY WORDS: hypoxia; fenigidin; duration of action potential; contractility; myocardium.

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