2. V. N. Grigor'ev, in: Problems in Functional Lymphology [in Russian], Novosibirsk (1982), p. 57.
3. I. Rusznyak, M. Földi, and D. Szabo, Physiology and Pathology of the Lymphatic Circulation [in Russian], Budapest (1957).
4. M. R. Sapin, N. Ay. Yruina, and L. E. Étingen, The Lymph Node [in Russian], Moscow (1978).
5. V. A. Shakhlamov, Capillaries [in Russian], Moscow (1971).
6. J. H. Cox and W. L. Ford, Cell. Immunol., 66, 407 (1982).
7. N. V. Ohmann, Cell. Tissue Res., 212, 455 (1980).
8. H. Osatake, K. Atoh, A. Mitsushima, et al., J. Electron Microsc., 29, 72 (1980).

EFFECTIVENESS OF MYOCARDIAL PROTECTION AGAINST ISCHEMIA BY NORMOTHERMIC CARDIOPLEGIC SOLUTION WITH CREATINE PHOSPHATE

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KEY WORDS: isolated rat heart; cardioplegic protection; use of creatine phosphate and adenosine triphosphate.

Recently a number of natural metabolic substrates whose action is directed toward regeneration and preservation of high-energy phosphates (HEP) have begun to be used as components of cardioplegic solutions [1, 3-5, 10]. As a rule the so-called metabolic protection of the myocardium is effected under hypothermic conditions, i.e., when activity of the metabolic processes on which these components of the cardioplegic solution must act is considerably depressed.

The aim of this investigation was to study the anit-ischemic action of creatine phosphate (CP) and adenosine triphosphate (ATP) as components of potassium-based cardioplegic protection, and to compare the protective effect of CP under normo- and hypothermic conditions.

EXPERIMENTAL METHOD

Experiments were carried out on a model of the isolated working rat heart [9]. Male Wistar rats weighing 220-350 g were used. Heparin was injected intraperitoneally into the animals in a dose of 3000 U with thiopental sodium in a dose of 0.2 mg/g body weight. The heart was excised from the anesthesized animals 5-8 min later and placed in cardioplegic solution which was cooled on ice. After the heart stopped beating the aorta was cannulated and retrograde perfusion of the aorta was carried out for 10 min by Langendorff's method with modified Krebs–Henseleit buffer [9] under a pressure of 82 cm water. In the course of this time the left atrium (LA) was cannulated and the left ventricle (LV) punctured to record the left-ventricular pressure (LVP) and heart rate (HR) by means of a P-50 electromanometer, coupled to a power supply, SP-1405 amplifier, and SP-2009 recorder (Gold Statham, USA). The heart was then perfused for 10 min through LA under a pressure of 17 cm water (Neely's method), and during this time LV, which contracted spontaneously, worked against a resistance and expelled 25-30 ml/min of perfusate. After the 10-min control period the cardioplegic solution was injected into the aorta for 3 min and the heart was exposed to the action of normothermic (36-36.5°C) ischemia for 30 min, after which retrograde reperfusion of the aorta for 10 min and reperfusion through LA by Neely's method for 30 min were carried out. During perfusion through LA, HR (beats/min), LVP (in mm Hg), the aortic ejection — AE (in ml/min), and the coronary blood flow K (in ml/min) were measured in the control and reperfusion period. Parame-

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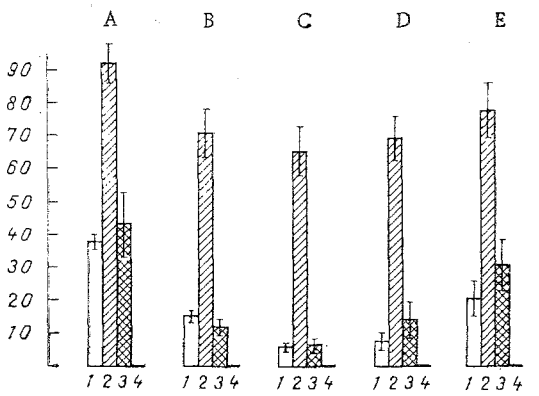

Fig. 1. Functional recovery of the heart (in % of control). A) HR; B) LVP; C) IWC: D) CO; E) SE.

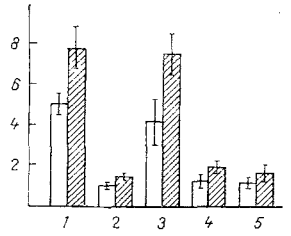

Fig. 2. Concentrations of ATP (unshaded columns) and CP (shaded columns) in myocardium (in μmoles/g weight of tissue). Here and in Fig. 3: 1) control, 2) normothermic potassium cardioplegia, 3) normothermic potassium cardioplegia with CP, 4) hypothermic potassium cardiopolegia with CP, 5) normothermic potassium cardioplegia with ATP.

ters such as the index of work of the heart (IWC = HR × LVP × 10^{-3}, cardiac output (CO = AE + K (in ml/min), and the stroke ejection SE = CO - HR (in μl/beat) also were calculated.

The anit-ischemic action of a normothermic potassium solution, the normothermic potassium solution with CP, hypothermic (6°C) potassium solution with CP, and normothermic (potassium solution with ATP) was investigated.

CP and ATP were added in concentrations of 10 mM to the cardioplegic solutions 10 min before they were injected into the aorta. Concentrations in the cardioplegic solutions were: Na^+ 111 mM, K^+ 30 mM, Ca^{++} 1 mM, Mg^{++} 1.2 mM, glucose 11 mM, and mannitol 60 mM; the osmolarity was 360 milliosmoles/liter, pH 7.7. The ionic composition of all the solutions used was verified by flame photometry on an IL-543 flame photometer (USA) and pH by means of a pH-meter (Corning, USA). The temperature of the solutions injected and of the surface of the myocardium was verified with a model 91 electrothermometer (Markson, USA).

At the end of each experiment a piece of myocardium was taken from the region of the apex of the heart and frozen with solid CO_2. Histological sections were stained by the PAS method in McManus modification. The glycogen concentration in the myocardium in each field of vision of the microscope under a magnification of 20 × 10 × 1.5 was estimated on a 7-point scale: 0) glycogen absent, 1 point) low glycogen concentration, 3 points) average, 5 points) considerable, and 7 points) high concentration: 2, 4, and 6 points corresponded to intermediate degrees of glycogen concentration. The remaining heart tissue was quickly frozen in liquid nitrogen, after which ATP and CP in the myocardium were determined by an enzymic method [7].

EXPERIMENTAL RESULTS

The most complete recovery of cardiac function was observed after myocardial protection with normothermic potassium solution with CP (Fig. 1). In this case LVP in the reperfusion period was 70.6 ± 6.7%, IWC was 65.1 ± 7.3%, CO 69.7 ± 6.1%, and SE 77.5 ± 8.2% of their control values. HR showed the highest level of recovery at 91.9 ± 5.7%. In other investigations

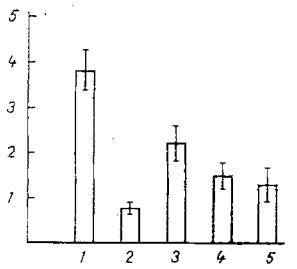

Fig. 3. Glycogen concentration in myocardium (in conventional units).

marked depression of the functional parameters of the heart was observed in the postischemic period, especially after normothermic potassium cardioplegia with ATP, which led to the development of myocardial contracture at the end of the reperfusion period. Recovery in experiments in which the myocardium was perfused by normothermic potassium cardioplegic solution and hypothermic potassium cardioplegic solution with CP was at a low level, virtually identical for the two series of experiments. These investigations showed that CP has a significant coronary dilator action (16.8 ± 1.4 ml/min). However, this effect was completely abolished by the use of CP as a component of the hypothermic cardioplegic solution (6.5 ± 1.1 ml/min; $P < 0.001$). During normothermic potassium cardioplegia the coronary blood flow was 13.2 ± 1.5 mg/min. The lowest coronary blood flow was observed after the use of the normothermic potassum solution with ATP (3.6 ± 0.9 ml/min).

Control values of the HEP concentration in the myocardium were determined in unperfused hearts. The ATP and CP levels were 5.04 ± 0.61 and 7.85 ± 1.02 μmoles/g wet weight of tissue, respectively. It will be clear from Fig. 2 that the HEP level after normothermic potassium cardioplegia with CP showed very little change (ATP 4.20 ± 1.22 μmoles/g, CP 7.56 ± 0.90 moles/g). In all the remaining experiments marked exhaustion of ATP and CP took place ($P < 0.001$).

The glycogen concentration in the myocardium of the control hearts and of hearts exposed to ischemia with cover of different forms of cardioplegic protection is shown in Fig. 3. Glycogen exhaustion was most effectively prevented by the use of cardioplegic solutions containing CP. The use of CP in a normothermic cardioplegic solution gave a more marked effect on maintenance of the glycogen level than the use of CP in hypothermic cardioplegic solution. Some investigations demonstrated that certain substrates of energy metabolism increased tolerance of the myocardium to ischemia. These metabolites are either precursors of ATP synthesis or they act on certain pathways of energy formation in the myocardial cell. One such compound is CP, which is directly linked with ATP formation and, according for data in the literature [2, 8, 5], has an anti-ischemic action. This investigation showed that CP, under normothermic conditions, gives a positive effect on preservation of myocardial contractility and maintenance of a sufficiently high glycogen and ATP level. It also has a significant coronary dilator action. Unlike CP, addition of ATP to the cardioplegic solution has no protective action on the myocardium, probably due to the more rapid degradation of this compound on the surface of the cell membrane or to differences in the ability of ATP and CP to pass through the membrane to the sites of their action in the cardiomyocytes.

The results of this investigation of the efficiency of CP in hypo- and normothermic cardioplegia showed that the protective action of CP depends on the temperature at which it is used, and that it is most effective under normothermic conditions. On addition of CP to the hypothermic cardioplegic solution IWC was 11 times lower, and the ATP and CP levels 2.7 and 3.3 times lower, respectively, than after its addition to the normothermic solution.

The results showing how the action of CP depends on temperature suggests that normothermia is essential for all metabolites used in cardioplegic protection of the myocardium for the optimal realization of their metabolic protective effect. Under hypothermic conditions, which depress both the utilization and formation of HEP [6], the use of metabolic substrates is ineffective because the reactions which they stimulate are inhibited. This is in agreement with observations by other workers [12] who found almost complete recovery (87%) of the "energetically exhausted heart" during protection of the myocardium by cold blood cardioplegia, if the cardioplegic infusion was carried out initially under normothermic conditions. Peyton et al. [11], during normothermic perfusion with cardioplegic solution containing glucose, also observed recovery of the depressed ATP and CP levels to the control values in hypertrophied rat hearts.

The present investigation confirmed that the CP increases the efficacy of cardioplegic protection and demonstrated that the most favorable conditions for metabolic cardioplegic protection of the myocardium are provided by normothermic injection of a cardioplegic solution with CP. Hence this leads to the conclusion, of great practical importance, that no useful purpose can be served by the use of hypothermia during cardioplegic protection of the myocardium, with the aim of preserving the existing solution and to ensure the most effective recovery of the exhausted HEP pool.

LITERATURE CITED

1. O. I. Pisarenko, E. S. Solomatina, I. M. Studneva, et al., Grudnaya Khir., No. 5, 10 (1982).
2. O. Faglemi, K. A. Rane, and I. R. Parratt, J. Cardiovasc. Pharmacol., 4, 53 (1982).
3. S. Z. Goldhaber, G. H. Ponost, R. A. Kloner, et al., Circulat. Res., 51, 181 (1982).
4. D. I. Hearse, D. A. Stewart, and M. V. Braimbridge, Circulation, 54, 193 (1976).
5. R. N. Jones, K. A. Reimer, M. L. Hill, et al., J. Molec. Cell. Cardiol., 14, Suppl. 3, 123 (1982).
6. W. Lamprecht, F. Stein, L. Iloin, et al., in: Methods of Enzymatic Analysis, (H. U. Bergmeyer, ed.) Vol. 4, Weinheim (1974), p. 1778.
7. R. J. Marshall and J. R. Parratt, Arch. Pharmacol., 281, 437 (1974).
8. J. R. Neely, H. Leibermeister, E. J. Battersby, et al., Am. J. Physiol., 212, 804 (1967).
9. M. K. Pasque, T. L. Spray, G. L. Pellom, et al., J. Thorac, Cardiovasc. Surg., 83, 390 (1982).
10. R. B. Peyton, P. Van Trigt, G. L. Pellom, et al., J. Thorac, Cardiovasc. Surg., 84, 11 (1982).
11. E. R. Rosenkranz, J. Vinten-Johansen, G. D. Buckberg, et al., J. Thorac. Cardiovasc. Surg., 84, 667 (1982).

BLOOD FLOW VELOCITY — A CONSTANTLY ACTING FACTOR IN DILATATION OF LARGE ARTERIES

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KEY WORDS: blood flow; sensitivity; velocity; diameter of artery.

The lumen of many main arteries of the systemic circulation in dogs and cats varies in response to a change in velocity of the blood flow [2, 4, 7, 9, 11]. However, in all investigations so far undertaken in which the sensitivity of arteries to blood flow velocity has been studied, dilatation of arteries was observed only in response to a considerable increase in blood flow velocity. It is therefore not yet clear whether there is continuous, uninterrupted regulation of the lumen of arteries in accordance with the blood flow velocity or whether changes in diameter take place only in response to considerable changes in blood flow.

The aim of this investigation was to determine whether arteries "monitor" changes in blood flow velocity continuously in the form of changes in their own diameter, and within what range of blood flow rates they exercise this property. The investigation was carried out on both the femoral and carotid arteries of cats, in which the presence of sensitivity to blood flow has been established [2, 9], and also on the renal artery, in which this property has not previously been found.

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