ANTI-ISCHEMIC ACTION OF PERFLUOROCARBON EMULSION (PFCE) ON THE CANINE MYOCARDIUM

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Recent investigations of perfluorocarbon emulsions have shown that these preparations can protect the myocardium against ischemic and reperfusion-induced damage, caused by arterial obstruction [3, 11, 12].

The anti-ischemic effects of PFCE are associated mainly with the ability of the emulsion to increase gas transport and flowability of the blood [8, 9]. However, the contribution of the rheologic and gas-transport properties of PFCE to anti-ischemic protection of organs has not yet been finally settled. Furthermore, the importance of the gas-transport properties of PFCE for the oxygen supply of organs is still a matter for discussion [7, 13].

The aim of the present investigation was to determine the role of the rheologic and gas-transport properties of PFCE in protection of the heart against ischemia.

EXPERIMENTAL METHOD

Experiments were carried out on 22 mongrel male and female dogs weighing 15-20 kg. After premedication with trimeperidine (10 mg/kg, intramuscularly) anesthesia was induced with thiopental sodium (15 mg/kg), the trachea was intubated, after which artificial ventilation of the lungs was carried out with a mixture of air and oxygen (2:1) with a RO-2 volume respirator. Thoracotomy was performed in the 5th intercostal space, after which the pericardium was divided and a small segment of the anterior descending branch of the left coronary artery was mobilized in its upper third, and a screw clip was fixed around it [6]. In the zone of presumed ischemia, two nickel-coated electrodes for recording the ECG were sutured to the epicardium and inserted into the wall of the left ventricle to a depth of 6-8 mm, and pO_2- and pH-electrodes [5] were fixed to the epicardium to determine the myocardial pO_2 and pH (MpO_2 and MpH). Next, the great vein of the heart was isolated in the zone of ischemia and catheterized to measure the coronary blood flow (CBF), the carotid artery to measure the blood pressure (BP), and the jugular vein likewise to return the coronary blood to the animal. Blood samples were taken from the carotid artery and coronary vein for determination of pO_2 and pCO_2, and pH of the arterial (apO_2) and coronary venous (cpO_2) blood by means of a "Corning" gas analyzer (Hungary), and the hematocrit (Ht) on a hematocrit centrifuge, and the hemoglobin concentration (Hb) by Sahli's method. The viscosity of the arterial blood was determined on a VK-2 viscosimeter. At the end of the experiment samples of myocardial tissue were taken in the intact zone and subendocardium and in the subepicardium of the ischemic zone, and activity of the enzyme creatine phosphokinase (CPK) in them was determined. The animals were heparinized (250 U/kg body weight) 30 min before the experiment began. The myocardial oxygen consumption (MOC) and the coronary vascular resistance (CVR) were calculated by traditional methods. The experiment began with recording of the initial values of the test parameters, after which the coronary artery was occluded by means of the screw clip by 70-80% [6]. Next, either PFCE (n = 7) or the salt composition of PFCE (SC) (n = 7), or a 4% solution of the surface-active substance (SAS) proxanol in SC (n = 8) was injected in a dose of 10 ml/kg in the course of 30 min, 60 min after occlusion. The PFCE contained 10 vols.% of perfluorocarbons (PFC).

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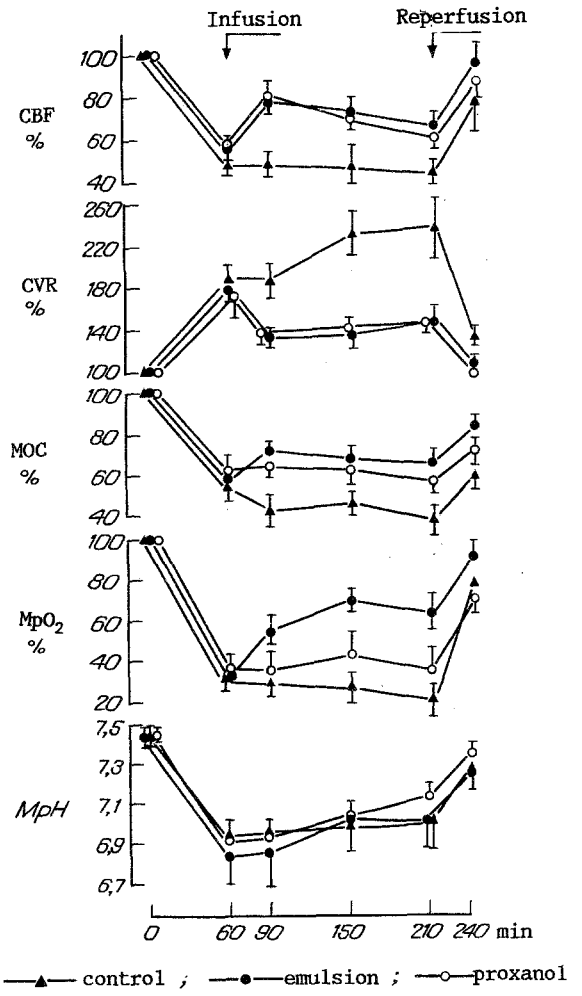

Fig. 1. Effect of infusion of PFCE, SAS, and SC (in doses of 10 ml/kg) on parameters of coronary blood flow, oxygen supply, and acid-base balance of the ischemic myocardium. Triangles indicate control, filled circles — emulsion, empty circles — proxanol.

The composition of SC was as follows (in mM): NaCl 102, KCl 5, $MgSO_4$ 1.2, $NaHCO_3$ 15, NaH_2PO_4 1.2, and glucose 11. The coronary artery was reperfused 120 min after infusion for 30 min by removing the screw clip from it. The test parameters were recorded again 60, 90, 150, 210, and 240 min after occlusion of the coronary artery.

EXPERIMENTAL RESULTS

The mean BP (BP_m) and apO_2 in all three groups did not change significantly in the course of the experiments, and amounted on average to 98 mm Hg and 120 mm Hg. By the 60th minute of ischemia, a significant fall of the CBF level on average to 55% of its initial level was observed, MOC fell to 59%, MpO_2 to 34%, and MpH from 7.4 to 6.9; meanwhile, CVR increased by 81% (Fig. 1). These changes were accompanied by the appearance of marked depression of the ST segment on the epicardial ECG (—0.38 mV), evidence of ischemic damage to the subendocardium [2].

Infusion of the dogs at the 60th minute of ischemia with the salt composition had no significant effect on the course of ischemia. During the next 2 h after infusion, a significant increase in CVR (up to 268% of the initial level), depression of the ST-segment (to —0.51 mV), and reduction of MOC (to 39.7%) were observed. The remaining test parameters did not change significantly.

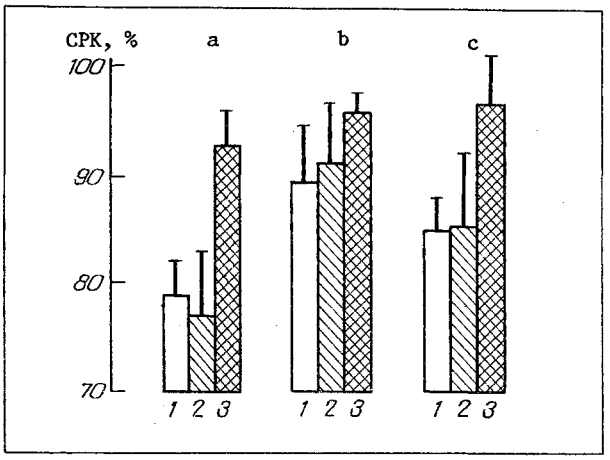

Fig. 2. Ratio of creatine phosphokinase (CPK) activity in subendocardium (END) and supepicardium (EPI) of ischemic zone to CPK activity in intact zone (INT). a) END/INT, b) EPI/INT, c) END/EPI. 1) Control, 2) proxanol, 3) emulsion.

Reperfusion of the coronary artery restored CBF (to 78%), MOC (to 62%), MpO_2 (to 79%), CVR (to 133%), and MpH (from 7.02 to 7.28); under these circumstances, cpO_2 increased to 143% but depression of the ST segment was reduced to —0.29 mV. However, these parameters (except MpH) differed significantly from the initial level at the end of the reperfusion period.

Changes in most parameters after injection of SAS and PFCE were similar in direction. In both these groups a significant decrease in blood viscosity (by 12%) and reduction of HR (by 10%) ($p < 0.05$) were observed. This was accompanied by a significant reduction of CVR (to 135%) and by an increase in CBF (to 80%) and cpO_2 (to 115%). Injection of PFCE also caused a significant increase in MOC and MpO_2 to 72% and 71%, respectively. A significant increase in MpH from 6.84 to 7.01 and a decrease in depression of the ST segment from 0.43 to 0.29 mV were observed 1 h after injection of PFCE.

Unlike PFCE, injection of SAS did not change the oxygen supply of the myocardium, and caused significant increase in MpH from 6.93 to 7.11 and reduced depression of the ST segment from 0.38 to 0.26 mV, only 2 h after injection.

Reperfusion restored CBF, CVR, and MpH to the initial level in both groups, whereas MOC was significantly lower. In the group receiving SAS residual depression of the ST segment (0.19 mV) was observed, whereas in the PFCE group, depression was completely abolished.

Analysis of CPK activity at the end of the experiment demonstrated a significant decrease in the subendocardium of the ischemic zone in the SC and SAS groups and in the subepicardium in the SC group. The enzyme activity in the ischemic zone in the PFCE group did not differ from that in the intact zone (Fig. 2).

The results are evidence that infusion of PFCE during ischemia reduces the ischemic disturbances of the oxygen supply and acid-base balance of the myocardium and prevents reperfusion-induced damage to the myocardium. It can be tentatively suggested that the anti-ischemic effect of PFCE is due to an increase in the coronary blood flow as a result of improvement of the rheologic properties of the blood. The rheologic effects of PFCE are connected with one of its components (SAS), for infusion with the same dose of a 4% solution of SAS, reduced the relative viscosity of the blood and CVR but increased CBF in a manner similar to PFCE. However, despite the improvement of CBF after injection of SAS, the oxygen deficiency in the ischemic myocardium was not abolished (MOC and MpO_2 were not significantly changed).

This difference in the action of the substances on the oxygen supply of the myocardium, despite identical effects on CBF, might apparently be easily explained by the presence of PFC in the emulsion, which dissolve oxygen several times more readily than plasma. However, calculation of the contribution of the different components of the blood to oxygen transport after infusion of 10 ml/kg of PFCE under the present experimental conditions shows that erythrocytes carry 98.3% of the total blood oxygen, plasma carries 1.2%, and PFC only 0.5%. It will be evident that the role of PFC in supplying oxygen is negligibly small compared with that of erythrocytes. Despite this, it must be recalled that under these experimental conditions (myocardial ischemia) some capillaries are impermeable for erythrocytes because of tissue edema, spasm of the precapillary sphincters, and occlusion of the capillaries with hypoxic erythrocytes, whereas PFCE particles, because of their small size (0.1 μm) can pass

through such capillaries. Under the conditions of an erythrocyte-free mediu8m the role of PFC in oxygen transport is greatly increased. Even with a fuorocrit of 1.4% the fraction of oxygen dissolved in PFC is 27%, and the plasma oxygen capacity is increased by 38%.

Meanwhile, PFCE improve the oxygen supply to the tissues by intensifying oxygen extraction from the erythrocytes at the critical oxygen supply level [10]. As the authors cited suggest, this effect of PFCE is connected with improvement of oxygen diffusion from the erythrocytes into the tissue through the plasma, and also through a considerable diffusion barrier, namely the vascular endothelium, where PFC particles may accumulate [12].

Improvement of oxygen diffusion in the medium containing PFC may be connected with differences in the Krogh diffusion constants for oxygen in PFC and water. Krogh's diffusion constant for oxygen is an order of magnitude greater in PFC than in water [4].

It is evident that improvement of the oxygen supply to the ischemic myocardium after infusion of PFCE is due mainly to its oxygen-transport function, for which the presence of PFC in the emulsion is responsible. Thanks to this function of PFCE, unlike SAS, prevented irreversible myocardial damage (absence of loss of CPK and of depression of the ST segment at the end of the reperfusion period). Under these circumstances, as our investigations showed, for the realization of this function there is no need to increase the PFC and oxygen concentrations in the blood up to the limit, as many investigators have done [8, 11, 12]. It must also be pointed out that PFC not only are responsible for the oxygen transport function of the emulsion, but by dissolving in cardiomyocyte membranes, they increase the resistance of the myocardium to ischemic and reperfusion damage [1].

Thus, it is the PFC in the emulsion, with which its gas-transport function is linked, that play the principal role in anti-ischemic protection of the heart.

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