Inhibition of Norepinephrine Reuptake and Size of Myocardial Infarction during Focal Ischemia and after Preconditioning

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Experiments on rats showed that blockade of norepinephrine reuptake in the early reperfusion period after focal myocardial ischemia aggravates myocardial injury and abolishes the protective effect of ischemic preconditioning.

Key Words: *norepinephrine; myocardial ischemia; ischemic preconditioning; desipramine*

The severity of injury in myocardial infarction (MI) is associated with norepinephrine (NE) release from the sympathetic nerve endings to the myocardial interstitium [5]. Reversion of NE reuptake during ischemia is a cause of significant increase in NE content in the myocardial interstitium. Under ischemic conditions, NE is transported from the cytoplasm to the interstitial space by the neuronal uptake carrier in the direction reverse of its normal transport [6].

An attempt to prevent NE accumulation in the interstitium by treatment with NE reuptake inhibitor before ischemia was successful. The severity of MI decreased under these conditions [4]. However, the effect of NE reuptake blockade on the size of MI during the reperfusion period corresponding to recovery of normal mechanism for NE reuptake remains unclear.

Here we studied the effects of blockade of NE reuptake during the reperfusion period on the size of MI during focal myocardial ischemia and after ischemic preconditioning (IPC).

MATERIALS AND METHODS

Experiments were performed on 27 male Wistar rats weighing 375.4±14.1 g. The animals were maintained in a vivarium under standard conditions. The experiment was conducted in accordance with the "Rules of Studies on Experimental Animals" (Supplement to the Order of the Ministry of Health of USSR, No. 755 of 12.08.1977).

The animals were subjected to tracheotomy under urethane anesthesia (2.10±0.06 mg/kg intraperitoneally). Artificial pulmonary ventilation was performed with the mixture of air and 20% O_2 on a Model 683 device (Harvard Apparatus). ECG was monitored during the experiment. Core temperature was measured in the rectum and maintained by heating of the surgical table. Left-sided thoracotomy was performed in the fifth intercostal space. After dissection of the pericardium, the left coronary artery was ligated with a nylon thread at a distance of 1-2 mm below the left auricle. Both ends of this loop were introduced into the holes of a plastic occluder.

The animals were randomized into 4 groups of 6-7 specimens each. Occlusion (30 min) and reperfusion (120 min) were performed in group 1 and 2 animals. Ringer's solution (1 ml) and desipramine (0.8 mg/kg) were injected intravenously to rats of groups 1 and 2, respectively, at the beginning of reperfusion. The animals of groups 3 and 4 were subjected to IPC (three episodes of 3-min occlusion at 3-min intervals of reperfusion). Occlusion (30 min) and reperfusion (120 min) were performed in the follow-up period. Ringer's solution (1 ml) and desipramine (0.8 mg/kg)

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were injected intravenously to rats of groups 3 and 4, respectively, at the beginning of reperfusion.

The perfusion zone (risk zone) was examined by the end of the experiment. The coronary artery was occluded repeatedly. Evans blue solution (2%, 4 ml) staining only the zone of myocardial perfusion was injected intravenously. The animals were euthanized by overdosing the anesthetic drug. The heart was isolated. The risk zone was excised (unstained area of the left ventricle). Then the risk zone was dissected to layers (1 mm in thickness). The plates were incubated in 1% triphenyltetrazolium at 37°C for 20 min. Under these conditions the intact myocardium is colored red, while MI zone remains unstained. After incubation, myocardial strips were scanned. The MI/risk zone ratio was calculated.

The results were analyzed by nonparametric Mann–Whitney test.

RESULTS

The area of MI in group 1 animals was 32.0±3.1% of the risk zone (Fig. 1). The area of MI in group 2 rats increased to 46.1±3.4% (p=0.006). IPC (group 3) was followed by a decrease in MI zone to 15.3±3.1% (p=0.008 compared to group 1). The area of MI in group 4 animals increased to 44.7±4.7% (p=0.0027 compared to group 3) and did not differ from that in group 2 rats (p>0.05).

Myocardial ischemia is accompanied by massive release of NE from the sympathetic nerve endings to the myocardial interstitium. NE content in the interstitial space increases manifold [3]. A specific feature of NE accumulation in the interstitium is a multistage nature of this process. At the beginning of ischemia, the increase in NE content is related to exocytosis and is limited by reuptake. After the 20th minute of ischemia, the mechanism of NE reuptake is reversed. This process is followed by a rapid and significant increase in NE content in the myocardial interstitium [6].

An increase in interstitial NE is a factor aggravating myocardial injury. NE has a dose-dependent adverse effect [5]. Some attempts were made to prevent or reduce NE accumulation in the myocardial interstitium. Treatment with NE reuptake inhibitor desipramine before the start of ischemia contributes to a decrease in the area of MI [4]. This positive effect is associated with a decrease in NE accumulation in the interstitium due to suppression of NE reuptake during ischemia.

Normal mechanism of reuptake is rapidly restored at the beginning of reperfusion. NE content in the interstitium decreases significantly under these conditions [1]. It can be suggested that blockade of NE reuptake in this period will contribute to ischemic injury. Our results indicate that blockade of NE reuptake during the reperfusion period increases the area of MI (group 2). This effect is probably associated with an increase in NE content in the myocardial interstitium due to rapid recovery of NE reuptake.

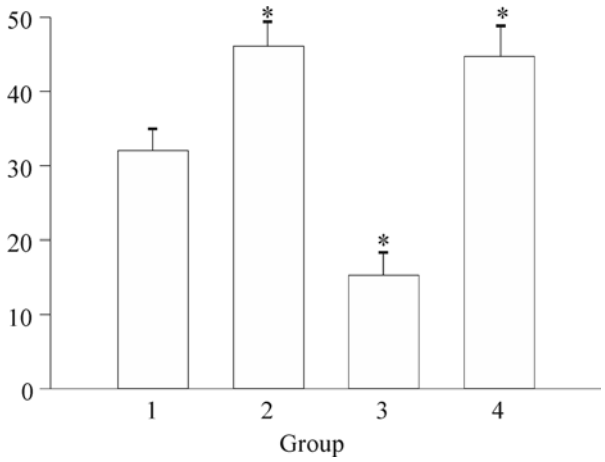

Fig. 1. Area of MI (% of the risk zone) during blockade of NE reuptake in myocardial ischemia and after IPC. *p<0.01 compared to group 1.

IPC contributes to a decrease in the severity of myocardial injury during ischemia. We showed that IPC in group 3 rats is followed by a twofold decrease in the area of MI (compared to group 1 animals). IPC results in a decrease in NE accumulation in the myocardial interstitium [7], which is probably related to a greater duration of NE reuptake [1]. Treatment with NE reuptake inhibitor was followed by an increase in the area of MI in IPC animals (group 4). This drug completely abolished the protective effect of IPC. The area of MI in these animals did not differ from that in group 2 rats.

Our results indicate that the inhibition of NE reuptake in the early reperfusion period after myocardial ischemia can produce an adverse effect. This effect is typical of tricyclic antidepressants for the treatment of depression. The use of tricyclic antidepressants is associated with high risk of acute MI [2]. However, the decrease in endogenous NE release from the sympathetic nerve endings after IPC probably has a greater role in the prevention of ischemic and reperfusion injuries.

Our results indicate that blockade of NE reuptake in the early reperfusion period after ischemia is followed by an increase in the severity of myocardial injury and abolishes the protective effect of IPC.

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