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Physical Training Limits the Fall of Blood Pressure and the Endothelium Overactivation in Acute Myocardial Infarction

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Summary

Physical training (PT) is beneficial in cardiovascular diseases associated with NO deficiency such as coronary disease, hypertension, etc. However, it is not known whether PT can also prevent pathological conditions associated with excess NO and fall of blood pressure (BP) such as acute myocardial infarction (AMI). The aim was to compare the effect of AMI on BP and functional state of the endothelium in rats trained by swimming and in untrained animals. After AMI, BP fell from 110±2 to 74±4 mm Hg ($p<0.05$), the endothelium-dependent relaxation increased from 37±4 to 66±6 % ($p<0.05$) and the extent of contraction suppression by the endothelium was significantly greater than in the controls. PT itself increased the endothelium-dependent relaxation of rat aorta but left BP unaffected. PT limited the AMI-induced fall of BP to 87±3 mm Hg, the endothelium- dependent relaxation to 53±4 % and prevented the hyporesponsiveness of the aorta to norepinephrine. We suggest that the protective effect of PT is related to inhibition of inducible NO synthase by a negative feedback mechanism.

Key words

Nitric oxide – Endothelium-dependent relaxation – Exercise – Adaptation – Myocardial infarction – Hypotension

Introduction

Stepwise adaptation to environmental factors is a potent therapeutic and preventive means for many diseases and pathological conditions of the cardiovascular system. The best studied type of adaptation that has long been applied in experiments and clinics is the adaptation to exercise or physical training (PT). PT prevents the development of hypertension (Kaplan 1991), reduces the probability of coronary atherosclerosis (Kramsch *et al.* 1981), exerts a beneficial effect on ischaemic heart disease (Leon 1985) and limits the risk factors of cardiovascular diseases (Meerson and Pshennikova 1988). The pathogenesis of all these diseases is known to involve impaired endothelium-dependent vasodilator responses due to the decreased production of nitric oxide (NO) (Star 1993). Up to the present, however, it is still not known whether PT can also be beneficial in pathological states associated with excessive production of NO.

We have previously shown that other types of adaptation, such as adaptation to stress (Meerson *et al.* 1991) and adaptation to hypoxia (Manukhina *et al.* 1994), can appreciably restrict the fall of blood pressure (BP) related to overactivation of the endothelium, i.e. with an excessive increase in endothelium-dependent relaxation and suppression of vasoconstrictor responses by the endothelium in acute myocardial infarction (AMI).

Therefore, the aim of the present study was to investigate the possibility of such protection by PT. To this aim we compared the effect of AMI on blood pressure and the functional state of the endothelium in trained and untrained rats.

Methods

The study was conducted in conformity with the policies and procedures detailed in the "Guide for the Care and Use of Laboratory Animals".

Experiments were carried out on Wistar male rats weighing 230–260 g. PT consisted of 30 daily sessions of passive swimming without load at the water temperature of 32 °C. The 1st session lasted for 15 min, the 2nd for 20 min. etc. up to 60 min. The

animals were taken for the experiment 48 h after the last training session.

Experimental myocardial infarction was induced by ligation of the left coronary artery (Selye *et al.* 1960). BP was measured using the indirect tail-cuff method by Physiograph DMP-4F (USA) 3 h after AMI. We have previously shown that the maximal postinfarction fall of BP occurs at this time (Meerson *et al.* 1989).

Animals were decapitated and the ring of thoracic aorta was isolated and suspended in an organ bath according to the previous method (Manukhina *et al.* 1994, Meerson *et al.* 1991). Records from native and denuded preparations were made simultaneously using a two-channel recorder Gemini (Ugo Basile, Italy). The endothelium was removed mechanically using a special catheter. Contractions of the aorta were induced with norepinephrine (NE) ($3x10^{-8}$ to $5x10^{-7}$ M). The sensitivity of adrenoceptors was evaluated by the ED_{50} value computed by the program LIGAND. Endothelium-dependent relaxation was induced by acetylcholine (10^{-8} to 10^{-5} M) against the background of NE ($5x10^{-7}$ M)-induced contractions.

All data were expressed as mean ± S.E.M. Unpaired Student's t-test was used to compare the results of the two studied groups or the results between endothelium-intact and denuded vessel preparations. Differences were considered significant at $p<0.05$.

Results

Three hours after AMI, BP of rats fell from 110±2 to 74±4 mm Hg ($p<0.05$). The PT itself did not influence BP (108±4 mm Hg) but significantly restricted AMI-induced hypotension because BP fell only to 87±3 mm Hg ($p<0.05$).

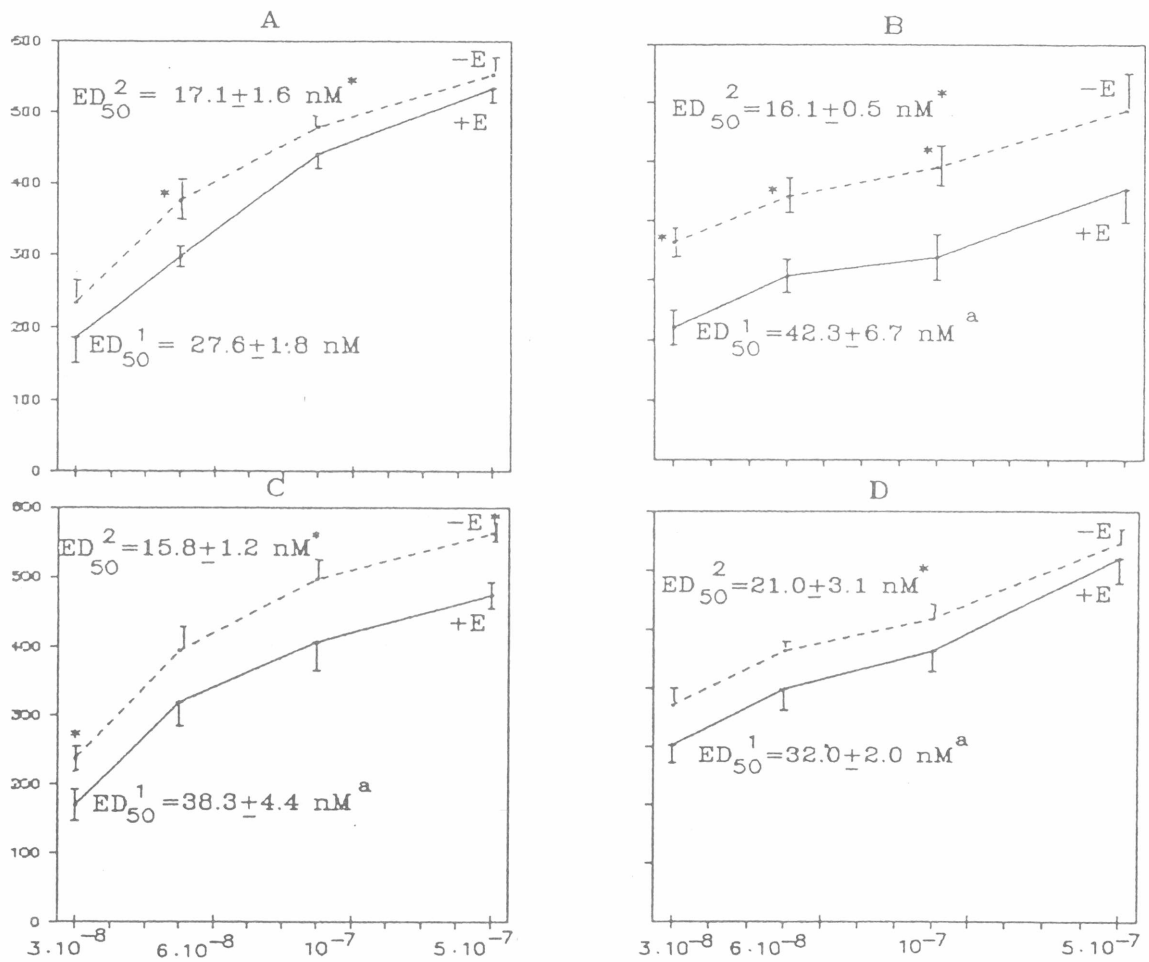

Fig. 1

*The effect of physical training on contraction force of endothelium-intact (+E) and endothelium-denuded (−E) rat aorta in control and acute myocardial infarction. A: control (n=12); B: acute myocardial infarction (n=9); C: physical training (n=11), D: myocardial infarction against the background of physical training (n=9). Abscissa: norepinephrine concentration (M); ordinate: contraction force, mg. ED_{50}^1 - for "+E" aorta; ED_{50}^2 – for "−E" aorta. a: significant differences from control; * significant differences between "+E" and "−E".*

It follows from Figure 1 that denudation of the endothelium always shifted the "concentration-response" curve upwards. The area between the curves reflects the attenuating effect of endothelium on smooth muscle contractions. It may be seen that, after AMI (Fig. 1B), the increment of contraction force due to denudation was considerably greater than in the control (Fig. 1A). In PT, this shift was also slightly larger than in the control ($p>0.05$) (Fig. 1C). However, in AMI on the background of PT (Fig. 1D), the suppression of contraction force by the endothelium did not significantly differ from the control.

Therefore, PT completely prevented the increased endothelial suppression of NE-induced contractions of the aorta.

Quantitatively, AMI increased the ED_{50} value for the native aorta 1.5 times, i.e. decreased the sensitivity of alpha-adrenoceptors. The PT itself also significantly decreased the adrenoreactivity of native preparation although to a lesser extent. Furthermore, the ED_{50} value for the aorta with intact-endothelium from trained animals subjected to AMI did not significantly differ from the control. It is important that ED_{50} values for denuded preparations were virtually similar for all experimental groups.

The endothelium-dependent relaxation of the aorta was sharply increased in AMI as compared to the control (Fig. 2). For instance, the maximal endothelium-dependent relaxation induced by acetylcholine at a concentration of 10^{-6} M comprised 37.0 ± 2.2 % in control, while after AMI it increased to $66,0\pm6,0\%$ ($p<0.05$). The PT also somewhat increased this parameter by 47.0 ± 2.4 % in comparison to the control ($p<0.05$). In trained animals, AMI increased the endothelium-dependent relaxation only by 53.2 ± 4.0 %, i.e. to a lesser extent than in untrained rats. The protective effect of PT was statistically significant.

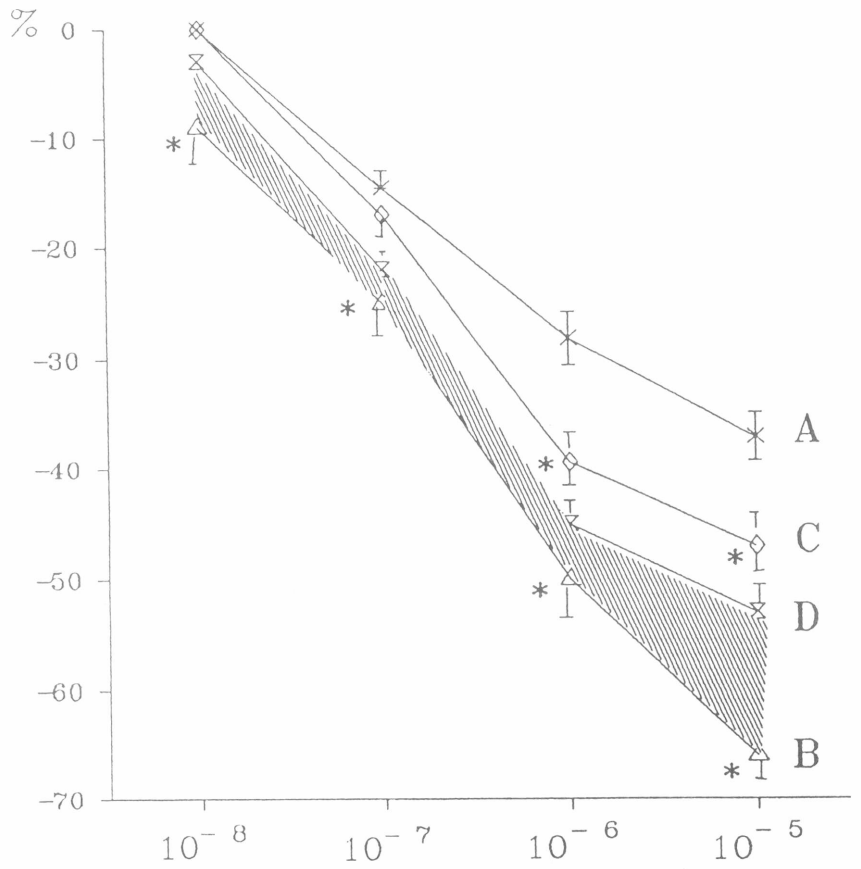

Fig. 2
*The effect of physical training on endothelium-dependent relaxation of isolated rat aorta in control and acute myocardial infarction. Abscissa: acetylcholine concentration (M); ordinate: relaxation, percentage of the contraction induced by norepinephrine ($5x10^{-7}$ M). A: control ($n=12$); B: acute myocardial infarction ($n=9$); C: physical training ($n=11$), D: myocardial infarction against the background of physical training ($n=9$). * significant differences from control. Hatched zone reflects the protective effect of physical training.*

Discussion

Pronounced hypotension and vascular hyporesponsiveness to constrictor stimulation characteristic of the acute period of myocardial infarction are important links in the pathogenetic chain of cardiogenic shock (Ross 1974) (Fig. 3). It is known that a major role in the postinfarction fall of blood pressure is due to excessive production of NO which reduces the vascular tone and peripheral vascular resistance (Vanin *et al.* 1994). In the present study we show that PT considerably limits the increased endothelial suppression of smooth muscle contractions, the enhanced endothelium-dependent relaxation and the related systemic hypotension.

There are several mechanisms by which NE responses may be decreased in the endothelium of the aorta from rats subjected to myocardial infarction.

The first possible mechanism is a decreased smooth muscle sensitivity to NE. This mechanism seems unlikely because the ED_{50} values for the denuded aorta were virtually similar in all the experimental groups (Fig. 1).

Second, NE can induce vasodilation through the α_2-adrenoceptor endothelium-mediated NO pathway (Richard *et al.* 1990). It follows from Figure 1 that all the revealed changes in adrenoreactivity occurred at the level of the endothelium rather than at the level of the smooth muscle. The mechanism of these changes could be related to an increased sensitivity of endothelial α_2-adrenoceptors with the unchanged sensitivity of α_1-adrenoceptors. This phenomenon is known to be characteristic of various types of stress (Shirinyan *et al.* 1988). Therefore, an increase in α_2-adrenoreactivity seems to be another possible mechanism of the decreased NE responsiveness in aortas from rats with myocardial infarction.

The third possible mechanism is an increase in either stimulated or basal NO production. The increased contractile response of the aorta due to denudation of the endothelium reflects the basal release of NO, whereas the increase in endothelium-dependent relaxation reflects its stimulated release. The contribution of either of these mechanisms cannot be excluded because myocardial infarction in our experiments enhanced both the endothelial suppression of aortic contractions and potentiated the endothelium-dependent relaxation.

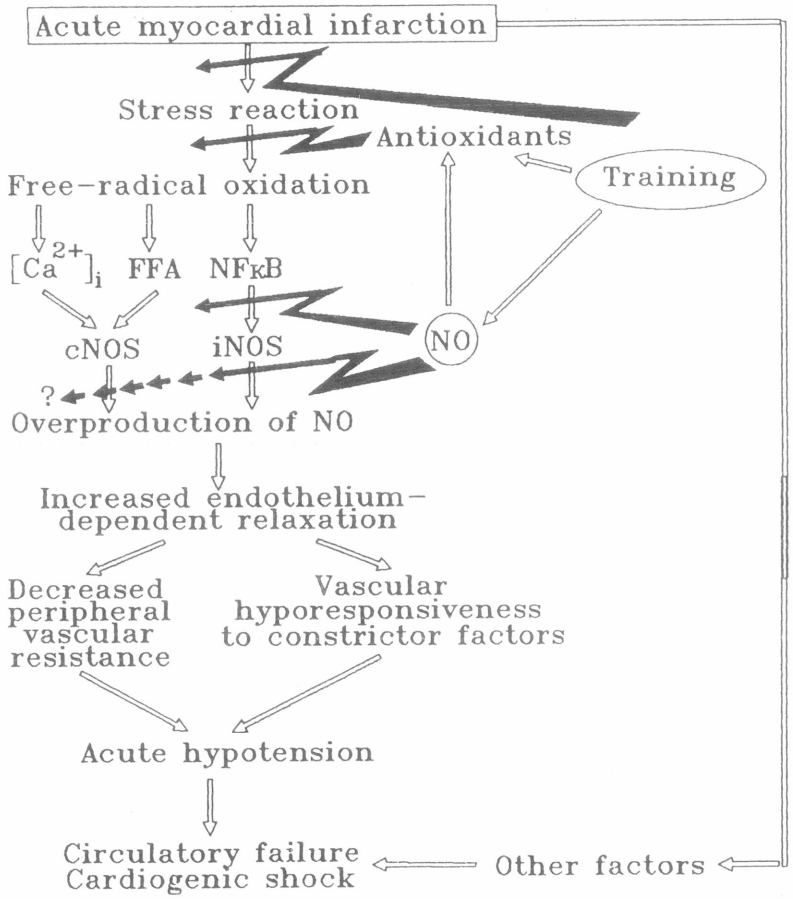

Fig. 3
Hypothesis on the protective effect of physical training against NO overproduction in acute myocardial infarction. For explanation see the text.

The mechanism by which myocardial infarction stimulates NO production remains obscure in many respects. It is possible that the stress reaction accompanying acute myocardial infarction activates free-radical processes in the organism (Meerson 1984). Oxygen radicals promote the expression of inducible NO synthase through a transcription factor NFκB and thereby sharply elevate NO production (Xie and Nathan 1994). As a result, the blood pressure falls and blood vessels become hyporesponsive to sympathetic stimulation (Bassenge 1994), which makes the hypotension irreversible (Fig. 3).

Some investigators believe that shock hypotension is due to the enhanced formation of NO by constitutive (cNOS) rather than by inducible NO synthase (iNOS) (Szabo *et al.* 1993). Free radical oxidation may stimulate NO production by endothelial NO synthase, for example, by the cleavage of phospholipids with the formation of free fatty acids (Ignarro 1989). In addition, the activation of lipid

peroxidation enhances intracellular Ca^{2+} due to disorders of Ca-ATPase mechanisms and disturbances of membrane structure and Ca-binding capacity (Meerson *et al.* 1981). This may activate Ca^{2+}-dependent constitutive NO synthase in the endothelium (Fig. 3).

Therefore, myocardial infarction may cause overproduction of NO by both cNOS and iNOS. It should be noted that the latter has been suggested to contribute to the basal NO production along with cNOS (Busse *et al.* 1993). In the present study we have demonstrated that PT itself enhances the suppression of aortic contractions by the endothelium and potentiates the endothelium-dependent relaxation. This is in agreement with our previous data that PT increases the NO production in different organs of rats as measured by the electron paramagnetic resonance-assay (Manukhina *et al.* 1996). Perhaps the expression of cNOS is due to chronic application of increased shear stress during the exercise-induced increase in blood flow (Miller and Burnett 1992). The produced NO can be sequestrated in the stable form of dinitrosyl iron complexes with thiol ligands (Mulsch *et al.* 1991) or in the less stable form of S-nitrosothiols (Stamler 1994). The sequestration of NO stimulates the catalytic activity of soluble guanylate cyclase and thus promotes relaxation of smooth muscles (Liu *et al.* 1993).

Since PT limits infarction-induced disorders of blood pressure and endothelial function the result would seem to be paradoxical: the factor which itself increases NO production prevents the detrimental effects of NO abundance. Similar phenomena were observed by Patel *et al.* (1993) and Kita *et al.* (1994) who have prevented myocardial damage induced by ischaemia and reperfusion, which is known to be related to a considerable extent with the overproduction of NO (Patel *et al.* 1993), both with NO synthase inhibitor L-NNA and with NO precursor L-arginine.

The mechanisms underlying the protective effect of PT are hypothetical as follows from Figure 3. The major pathway of iNOS inhibition is negative feedback by NO, its own product (Assreuy *et al.* 1993). It has been suggested that NO may bind to the haeme moiety of iNOS or it can control iNOS gene expression (Weiss *et al.* 1993). This is probably why PT increasing the NO production limits infarction-related disorders.

Furthermore, the protective effect of PT may also be due to the PT influence on other links in the pathogenetic chain of cardiogenic shock. Prior PT is known to limit the stress reaction of the trained organism (Meerson and Pshennikova 1988) and to potentiate its antioxidant defense (Jenkins *et al.* 1983). Besides, NO itself can serve as an antioxidant (Kanner *et al.* 1991).

In conclusion, physical training prevents the fall of blood pressure to a considerable extent and normalizes the functional state of the endothelium in acute myocardial infarction. In this connection such type of adaptation is of special interest as a factor of prevention and treatment of pathological states related with both deficiency and abundance of NO.

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