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Effects of exercise training upon endothelial function in patients with cardiovascular disease

Axel Linke, Sandra Erbs, Rainer Hambrecht

University of Leipzig, Heart Center, Department of Cardiology, Leipzig, Germany

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1. ABSTRACT

Preservation of normal endothelial function depends on the bioavailability of nitric oxide (NO). In addition, accumulating evidence suggests that bone marrow-derived circulating progenitor cells (CPCs) are required to maintain the integrity of the vasculature. However, in patients with cardiovascular disease (CVD), the impairment of NO production in conjunction with excessive oxidative stress, results in a decline in NO bioavailability, promotes the loss of endothelial cells by apoptosis and, therefore, results in endothelial dysfunction. Moreover, functional alteration of CPCs might contribute to an impaired endogenous regenerative capacity and lead to further deterioration of vasomotion in different vascular beds in CVD. However, exercise training (ET) has assumed a role in cardiac rehabilitation in CVD since it reduces morbidity and mortality. This has been partially attributed to ET-mediated improvement of endothelial function. At the molecular levels, accumulating evidence suggests that regular physical activity restores the balance between NO production and NO inactivation by ROS. Moreover, ET might have the potential to restore the regenerative capacity of CPCs in CVD. Given the prognostic value of endothelial function further studies are necessary to elucidate whether the ET-induced correction of vasomotion is the key mechanism responsible for the decline in mortality in patients with CVD.

2. INTRODUCTION

2.1. The normal endothelium

2.1.1. Regulation of vasomotion: cross-talk between NO and reactive oxygen species

Since more than 2 decades ago, NO has been identified as the primary vasodilator in different vascular beds. (1) However, our understanding of endothelial vasomotion has been extended considerably since then: the NO precursor L-arginine is taken up by an active transmembraneous transport into endothelial cells, where it is either stored in intracellular vesicles or instantaneously converted into L-citrulline and NO. (2) The latter step is catalyzed by the endothelial NO synthase (eNOS), whose activity is influenced by different factors. (3) Appropriate expression and stability of the eNOS protein are key components, which modulate vascular production of NO. (3) In this regard, the promoter polymorphism (T-786C) of eNOS – representing a mutation in the 5´-flanking region of the gene – caused considerable attention, since this alteration was associated with a blunted eNOS protein expression and coronary artery spasm in humans.(4,5) Although controversially discussed, experimental data suggest that eNOS protein - transcribed from the gene in case of an homozygous genotype of the exon 7 polymorphism (G894T) - is more susceptible to proteolytic cleavage. (6) This was associated with reduced steady-

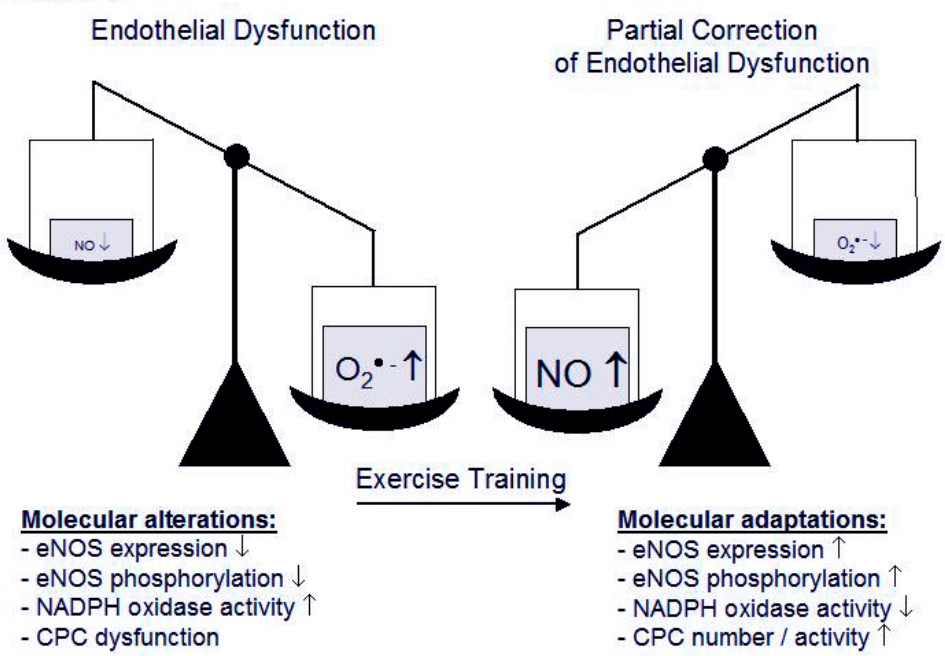

Figure 1. Molecular alteration in endothelial dysfunction and molecular adaptations in response to exercise training in CVD. In patients with cardiovascular disease, endothelial NO generation is primarily attenuated due to a decreased eNOS expression and AKT-mediated eNOS phosphorylation at serine 1177. The angiotensin II-induced activation of the NAD(P)H oxidase and, hence, the increase in ROS generation leads to further decline in NO bioavailability. In addition, reduction in the number of circulating progenitor cells (CPCs) and impairment in their function has been linked to endothelial dysfunction in CVD. Exercise training reduces angiotensin II subtype I receptor expression and decreases the NAD(P)H oxidase-derived production of ROS. In addition, exercise training significantly increases eNOS expression, AKT-mediated eNOS phosphorylation at serine 1177 and, hence, re-adjusts the balance between NO generation and NO inactivation by ROS. The augmented NO bioavailability leads to an improved vasodilatory capacity in patients with CVD. Moreover, accumulating evidence suggest that the training intervention might have the potential to induce endogenous endothelial regeneration through activation of circulating progenitor cells in CVD.

state levels of eNOS protein and, hence linked to a blunting of vascular NO production, occurrence of endothelial dysfunction, coronary artery disease, and myocardial infarction, respectively. (7,8) Moreover, conformational changes of eNOS protein are known to influence activity of the respective protein, and consequently NO generation. (3,9,10) In addition, it has been elucidated that serine/threonine protein kinase Akt or protein kinase A-dependent phosphorylation of eNOS at Ser^{1177} (human sequence) is leading to an increase in eNOS activity and, consequently, an elevation of endothelial NO production. (11,12) On the other hand, activation of protein kinase C lowers eNOS activity – and hence NO generation - through phosphorylation at the threonine 495 residue of eNOS. (3) In addition, protein kinase A (PKA) is known to regulate eNOS activity, in particular in response to pharmacological stimuli - but clear data proofing a contribution of PKA in shear-mediated activation of eNOS is missing in humans so far. (3) (Figure 1)

However, NO bioavailability is not only a function of NO production but also of NO degradation by ROS. In this regard, the NAD(P)H oxidase, xanthine oxidase, cytochrome P450, myeloperoxidase, heme oxygenase, glucose oxidase, lipoxygenase, enzymes of the respiratory chain and an uncoupled eNOS itself have been identified as potential sources of reactive oxygen species in the vasculature. (13-16) (Figure 1) Nevertheless, in a vessel of a healthy human being ROS are produced in small amounts to function primarily as signaling molecules. (13) In addition, cells are protected from ROS-mediated damage by a variety of non-enzymatic and enzymatic radical scavenger systems, e.g. the different forms of the superoxide dismutase, the glutathione peroxidase, catalase, and the thioredoxin system. (13) According to the literature, the endothelial isoform of superoxide dismutase (ecSOD) seems to be of importance for the instantaneous inactivation of vascular ROS. (14) Moreover, a feed-forward mechanism between eNOS-derived NO production and ecSOD expression suggests that NO controls the rate of superoxide degradation by ecSOD and, hence, its own bioavailability. (17) (Figure 1) Once NO is produced – and not inactivated by ROS - it diffuses to the vascular smooth muscle, induces a relaxation through cyclic guanosine monophosphate-dependent second messengers, and hence, causes a vasodilatory response. (2)

2.1.2. Contribution of bone marrow-derived progenitor cells to endothelial homeostasis

During aging, a proportion of endothelial cells lining the vasculature is lost due to apoptosis. (18) In the past, it was believed that this damage is primarily resolved by outgrowing resident endothelial cells. (18) However, recent studies support the hypothesis that the integrity of the vessel primarily depends on number and function of circulating bone marrow-derived progenitor cells. (19-22) A subset of these cells, so called circulating progenitor cells (CPC), are distinct from mature endothelial cells: they express CD34, CD133, and the receptor of vascular endothelial growth factor receptor (KDR) on their surface; they bind lectin, take up acetylated low-density cholesterol and are known to adapt the phenotype of mature endothelial cells in later stages of their development thereby promoting vascular repair. (22-25) In addition, in clinical pilot studies, the intracoronary infusion of these cells was shown to improve endothelial function and increase ejection fraction. (26,27) Nevertheless, further studies are necessary to elucidate how much these cells contribute to endothelial regeneration in healthy individuals.

2.2. Endothelial dysfunction in cardiovascular disease

2.2.1. Impairment of vasomotion: dysbalance between NO and ROS

In patients with cardiovascular disease, an impairment of NO bioavailability represents the primary reason for endothelial dysfunction in different vascular beds. (28) On one hand, this is the consequence of a blunted NO production due to reduced concentrations of L-arginine, changes in eNOS protein expression, conformation, stability and phosphorylation. (3,29-31) (Figure 1) In addition, increased concentrations of the endogenous eNOS inhibitor asymmetric dimethylarginine (ADMA), which is known to decline eNOS activity, might be responsible for the aggravation of endothelial dysfunction. (32) On the other hand, the biological half live of NO is shortened due its inactivation by ROS. In this regard, the nicotinamide adenin dinucleotide phosphatase (NADPH) oxidase, which is activated by angiogensin II, mechanical stretch and proinflammatory cytokines, has been recognized as a major producer of ROS in the vasculature, in particular in patients with coronary artery disease (CAD) and chronic heart failure (CHF). (33,34) Moreover, ROS derived from the xanthine oxidase - that is also expressed in the endothelium - appear to deteriorate vasomotion in the setting of CAD, since its activity was inversely related to endothelium-dependent vasodilatation. Besides the two above-mentioned key players in the circle of radical producing enzymes, myeloperoxidase, enzymes of the mitochondrial electron transport chain, cytochrome P450 isoenzymes, lipoxygenase, cyclooxygenase, heme oxygenase and glucose oxidase have been linked to an increase in vascular oxidative stress and, hence, the occurrence of endothelial dysfunction, at least in experimental settings. (13,34) Nevertheless, ROS generated by the above-mentioned enzymes will also promote the oxidation of tetrahydrobiopterine (BH_4), which is an essential co-factor for eNOS-derived NO production. (35) Oxidation of BH_4 ultimately results in "uncoupling" of eNOS that starts to produce ROS instead of NO. However, ROS-mediated cellular alterations are usually prevented or at least reduced by enzymatic and non-enzymatic radical scavenger enzyme systems. (33) In this regard, the ecSOD activity is of seminal importance in CAD, since Landmesser *et al.* were recently able to show that a reduced ecSOD activity is associated with an impairment of endothelium-dependent, NO-mediated vasomotion. (14,36) In summary, in patients with cardiovascular disease, the decline in vascular NO bioavailability is the consequence of a multifactorial process, which is associated with an aggravation of endothelial dysfunction that might lead to a reduction in organ perfusion.

2.2.2. Impairment of vasomotion: progenitor cell dysfunction?

In patients with atherosclerosis or cardiovascular risk factors, repair of endothelial damage by progenitor cells appears to be attenuated due to a decrease in their number and an attenuation of their regenerative capacity. (19,21) At the molecular level, this has been predominantly attributed to cellular senescence and impairment of proliferation. (21,37) In addition, it is conceivable that inflammatory cytokines – that are circulating in high concentration in particular in patients with end-stage CHF – exert myelosuppressive effects and, thereby, preclude the release of CPCs that are urgently needed for vascular regeneration. (38,39) Alternatively, the accelerated vascular damage might lead to an exhaustion of competent circulating progenitor cells in the blood and the bone marrow in patients with cardiovascular disease. (40) However, further studies are required to shed more light on progenitor cell-mediated vascular regeneration in CAD and CHF. (41)

3. IMPACT OF EXERCISE TRAINING ON THE VASCULATURE IN PATIENTS WITH CARDIOVASCULAR DISEASE

Exercise training is an established intervention in primary and secondary prevention of cardiovascular disease. (42-44) It increases exercise capacity, enhances myocardial and peripheral perfusion, but most importantly exercise training reduces morbidity and mortality in patients with CAD and – maybe – also in CHF. (42,45) However, the basic mechanisms behind this positive effects are poorly understood. Given the prognostic value of coronary and peripheral endothelial dysfunction, it is conceivable that exercise-mediated improvement in central and peripheral vasomotion is reflected in the improvement in prognosis. (41)

3.1. Improvement of endothelial dysfunction by exercise training

3.1.1. The coronary circulation

3.1.1.1. Conduit vessels

It was not clear for a long time, how exercise training influences coronary endothelial function in patients with CAD. Therefore, we randomly assigned patients with CAD to aerobic bicycle ergometer training or an inactive control group to address this issue.(28) Endothelium-dependent vasomotion of the epicardial coronary vessels in

response to acetylcholine was assessed at study begin, after 4 weeks and after 6 months, respectively. (46) The vessel diameter was measured by quantitative angiography and the average peak velocity was determined applying a Doppler wire. At study begin, all of the patients demonstrated coronary vasoconstriction after intracoronary infusion of acetylcholine as a sign of severe endothelial dysfunction. This vasoconstrictive response most likely results from a blunted endothelial NO production and a disruption of the vascular integrity due to a loss of endothelial cells by apoptosis. At 4 weeks, acetylcholine-mediated coronary vasoconstriction had declined by 54% in response to 7.2 µg/min of acetylcholine in patients of the training group, whereas no change was evident in the physically inactive controls. In addition, average peak velocity after stimulation with acetylcholine was found to be improved from 78% at study begin to 142% at 4 weeks as a result of the supervised, aerobic exercise training. (28) Therefore, this study provides proof-of-concept that 4 weeks of exercise training partially corrects endothelial dysfunction of conduit vessels in humans with CAD. However, so far the trial does not give an answer on the question of whether these positive treatment effects persist over time and whether the continuation of exercise training leads to a further augmentation of coronary vasomotion. Therefore, after 4 weeks in-hospital, patients in the training group were equipped with a bicycle ergometer and asked to continue exercise training at home. In these patients, a strong correlation between the change in average peak velocity and daily training duration was observed within the study period of 6 months. Although improvements with regard to coronary vasomotion observed after 4 weeks of in-hospital exercise training slightly deteriorated over time, endothelium-dependent vasomotion in response to acetylcholine was still better at 6 months as compared to study begin. This clearly implies exercise training – even on a lower level – leads to a persistent improvement of endothelial function in patients with CAD. (46)

3.1.1.2.Microcirculation/resistance vessels

Resistance of the vascular tree depends on the width of epicardial vessels, small arteries, arterioles, and the intramyocardial capillary system. Coronary flow reserve represents a combined measure of the above-mentioned resistance components in response to hyperemic stimulation, e.g. after infusion of adenosine. However, in the absence of overt arteriosclerosis, resistance of the epicardial coronary arteries is almost negligible and small arteries and arterioles with a diameter of >400µm have only minimal resistance as well. Therefore, small coronary arterioles with a diameter of < 400µm and capillaries mainly determine coronary vascular resistance and, consequently, coronary flow. (47) In animal studies, short-term physical activity improved the sensitivity of resistance vessels to adenosine; in contrast, long-term physical activity is associated with structural changes of the vascular tree, in particular an increase in the cross-section of the microcirculation. (48) Besides the above-mentioned exercise training-induced changes in epicardial coronary vessels, we recently documented an augmentation in coronary flow reserve after stimulation with adenosine by 30% after 4 weeks of in-hospital exercise training in patients with CAD. These data suggest that the responsiveness of the microcirculation to adenosine improves as a result of exercise training. (28) In addition, this finding supports the hypothesis that the cross-sectional area of the microcirculation has increased as a result of the exercise training intervention in patients with CAD, possible due to the formation of new capillaries.

3.1.1.3.Collateral formation and regression of coronary atherosclerosis

In the past, it was believed that the exercise training-mediated reduction in risk profile and the partial correction of endothelial dysfunction might be associated with a regression of coronary stenosis. However, the exercise training-induced regression of coronary atherosclerosis was negligible in the majority of the trials and, therefore, most likely does not account for the relief in symptoms and the improvement of myocardial perfusion in patients with CAD undergoing exercise training. (49,50)
Besides the regression of atherosclerosis, the formation of collaterals has been considered a mechanism explaining the increase in myocardial perfusion in response to exercise training in CAD. This anticipation was supported by animal studies, showing an enhanced exercise training-induced growth of collaterals, in particular in dogs. (51) However, studies in humans revealed conflicting results. In one trial, increased collateral formation was observed in selected patients with ischemic heart disease after exercise training. (52) In contrast, the Heidelberg Regression Study failed to document collateral formation as determined by angiography after one year of regular physical activity in patients with significant CAD. (53) However, one has to keep in mind that angiography might be to insensitive to document small collaterals that are not necessarily recruited at rest and that more sophisticated methods to detect collateral flow might have revealed other results.

3.1.1.4.The peripheral circulation

Endothelial dysfunction is not only a feature of the coronary circulation in patients with cardiovascular disease, but also affects peripheral arteries supplying blood to the working skeletal muscle in particular in CHF. (54,55) In patients with CHF, peripheral vasoconstriction has been attributed to the activation of the sympathetic nervous system, the renin-angiotensin system and the pituitary-vasopression axis. In addition, an impaired release of NO in response to endocrine mediators e.g. acetylcholine or bradykinine and mechanical activation – in particular changes in blood flow velocity or shear stress – is known to contribute to a further aggravation of endothelial dysfunction and, hence, peripheral perfusion. (56,57) Therefore, the maladaptations leading to endothelial dysfunction in the coronary and peripheral circulation are very similar. However, there seem to be quantitative differences of the diverse alterations, which are depending on the type of the vascular bed as well as on the kind and severity of cardiovascular disease.

Exercise training – as a therapeutic intervention – has been shown to enhance maximal and submaximal exercise capacity, not only in patients with CAD but especially in CHF. (42-45) The corresponding

improvement in symptoms in the latter group is the result of a mutifactorial process, which involves a blunting of the activation of the renin-angiotensin system, a reduction in sympathetic drive, and correction of pulmonary and skeletal muscle alteration, respectively. However, it was not clear for a long time, whether the above-mentioned functional adaptation has its seeds in an exercise training-mediated improvement of peripheral vasomotion. This prompted us to elucidate the effects of systemic exercise training on peripheral endothelial function in CHF. In this study, patients with CHF and a severely compromised left ventricular ejection fraction (LVEF: 24±4%) were randomized to 6 months of exercise training or a physically inactive control group. (54) At study begin and after 6 months, the vessel diameter of the superficial femoral artery (SFA) was assessed by quantitative angiography; average peak velocity was determined by a Doppler wire, and leg blood flow was calculated. In this trial, exercise training had no effects on endothelium-independent vasomotion induced by nitroglycerine. However, peripheral blood flow in response to 90 μg/min acetylcholine increased significantly by 203% from 152±79 mL/min at begin to 461±104 mL/min after 6 months of exercise training. Intraarterial co-infusion of the eNOS-inhibitor L-NMMA revealed an augmentation in NO-dependent vasodilatation by 174% as a results of the exercise training intervention. (54) Interestingly, there was a strong correlation between the change in peripheral blood flow and peak oxygen uptake suggesting that the improvement of endothelial function is directly reflected in a gain of exercise capacity in CHF. (54)

Nevertheless, there was a continuing debate, of whether there is a critical proportion of whole body muscle mass that has to be trained to achieve systemic effect on endothelial function. Indeed, it appears that local exercise training of the lower leg has the potential to exert systemic effects on vasomotion, not only in healthy individuals (58) but in particular in individuals with CHF: in the latter study, patients suffering from CHF (LVEF 26±3%) were subjected to 4 weeks of in-hospital bicycle ergometer training or a sedentary lifestyle. (55) At begin and after 4 weeks, endothelium-dependent vasodilatation in response to acetylcholine, endothelium-independent vasomotion following intraarterial infusion of nitroglycerine and flow-mediated vasodilatation were assessed using a high-resolution ultrasound system. In patients with CHF, lower limb exercise training significantly improved endothelium-dependent and flow-mediated dilatation of the radial artery by 284% and 52%, respectively. Again, endothelium-independent vasomotion in response to nitroglycerine did not change, neither in the training nor in the control group. Importantly, the exercise training-mediated correction of endothelial dysfunction was closely linked to the improvement in peak oxygen uptake as a measure of exercise capacity. (55) Since the beneficial effects of exercise training were not confined to the trained extremity, the study implicates systemic effects of the training intervention on vasomotion as long as a critical muscle mass is activated at a level, which results in an increase in blood pressure amplitude and shear stress.

However, many patient with end-stage heart failure having cardiac cachexia due to a considerable loss of skeletal muscle mass might be just to weak to tolerate systemic exercise training. In those patients a local exercise training that only involves small muscle groups in the absence of systemic effects might lead to an increase in local muscle mass and improvement in endothelial function of the corresponding vasculature. This intervention has potential to provide the "equipment" for systemic exercise training once the patient is locally conditioned with regard to muscle and endothelium. Indeed, local hand grip exercise training was shown to enhance acetylcholine-induced and flow-mediated vasodilatation of the radial artery in patients with CHF. (31,59) However, the oral supplementation of the eNOS precursor L-arginine on top of local exercise training led to a further improvement of endothelial function clearly indicating that not of all the alterations on the vascular level can be completely normalized by the training intervention. (31)

3.2. Molecular mechanisms behind the exercise training-mediated improvement of endothelial function in cardiovascular disease

As described above, exercise training partially corrects endothelial dysfunction in patients with CAD and CHF but the exact mechanisms are only partially understood given the difficulties to harvest vascular tissue samples. Indirect evidence suggests that the exercise training-mediated improvement in endothelial function is at least partially the result of an enhanced eNOS-derived NO production and decline in oxidant stress. (Figure1) In order to understand the exercise training-mediated adaptations at the molecular level, patients with CAD having an indication for elective aortocoronary artery bypass grafting (CABG) were randomized to either 4 weeks of in-hospital regular aerobic exercise training or to physical inactivity. (29) Vasomotor function of the left internal mammary artery (LIMA) in response to acetylcholine was invasively measured using a Doppler wire at study beginning and after 4 weeks. eNOS protein expression and AKT-mediated eNOS phosphorylation were determined in LIMA tissue samples, which were collected at the time of bypass surgery. Exercise training enhanced eNOS protein expression and AKT-mediated eNOS phosphorylation at serine 1177 2fold and 4 fold, respectively. This was associated with a significant increase in average blood flow peak velocity (APV) – as a measure of endothelial function - in response to acetylcholine. The linear correlation between the level of phospho-eNOS^{1177} and change in APV observed in this study is consistent with the notion that endothelium-dependent vasodilatation partially recovers due to a shear stress-induced/Akt-dependent phosphorylation of eNOS at Ser^{1177}. (29) (Figure1) However, the individual response to the training intervention seems to be also a function of genetic alterations of the eNOS gene. This assumption is supported by a study showing that 4 weeks of exercise training caused comparable increase in APV by more than 80 % in WT and exon 7 polymorphism-positive patients, but only by 36% in carriers of the promoter polymorphism. (60) These results are supported by cell culture experiments clearly revealing

a blunted eNOS protein expression in the presence of the promoter polymorphism.

Nevertheless, endothelial function is influenced by eNOS-derived NO production as much as ROS-mediated NO inactivation. In patients with CAD and CHF, activation of the local and global renin-angiotensin-aldersterone system has been linked to endothelial dysfunction. (61) According to animal and *in vitro* studies, angiotensin II (Ang II) has direct vasoconstrictive properties through binding to the predominant Ang II type 1 receptor (AT1-R) at the surface of smooth muscle cells. However, the net effect of Ang II on vasomotion depends on the ratio between the AT1-R and AT2-R, since activation of the AT2-R can also cause vasodilatation. (47) In addition, Ang II has been found to augment ROS generation due to an activation of the NAD(P)H oxidase, which comprises of different subunits. (62) In order to elucidate the role of angiotensin-meditated vasomotion and ROS production and its modulation by exercise training, the expression of the Ang II receptors and the NAD(P)H oxidase were measured in the LIMA of patients with CAD that had been randomized to 4 weeks of in-hospital exercise training or a physically inactive control group.(30) In patient of the training group, the expression of the NAD(P)H oxidase subunits pg91phox and p22phox was significantly lower as compared to the inactive controls. (Figure1) The above-mentioned adaptations in the training group were associated with a considerably lower NAD(P)H oxidase activity and, hence vascular ROS generation. (30) Moreover, the improvement of endothelium-dependent vasodilatation in response to the training intervention was found to be the result of a blunted Ang II-induced vasoconstriction and a attenuated Ang II-mediated activation of the NAD(P)H oxidase. Therefore, in patients with CAD, regular physical activity not only enhances the vascular production of NO, exercise training also augments the NO bioavailability, since it blunts the activation of ROS-producing enzymes. (30)

3.3. Contribution of progenitor cells to exercise training-mediated improvement of endothelial function in cardiovascular disease

Besides angiogenesis, the formation of new-blood vessels through vasculogenesis might lead to an increase in the cross sectional area of the vascular tree in response to exercise training. (63) However, the amount of data with regard to the exercise-training mediated release of progenitor cells from the bone marrow and their involvement in vascular repair as well as vasculogenesis in humans with cardiovascular disease is limited. (64) Nevertheless, animal studies suggest that regular physical activity considerably increases the number of CPCs, since exercise training enhances the NO-mediated liberation from the bone-marrow and reduces the apoptosis of circulating CPCs. (64) These CPCs enhanced neoangiogenesis and attenuated neointima formation after vascular injury in the exercise-trained animals. (64) The hypothesis of a exercise training-mediated increase in CPCs is supported by studies in humans, showing 4fold higher CPC levels in runners than in inactive healthy control subjects. (65) Nevertheless, in patients with cardiovascular disease, the data are conflicting. Laufs and co-workers recently reported an exercise training-mediated augmentation in the number of CPCs in patients with CAD in the absence of an exercise induced ischemia. (64) On the contrary, in the setting of CAD Sandri *et al.* failed to demonstrate a rise in CPC levels in response to an exercise training program that was shown to enhance vascular NO bioavailability. (29,66) In this study, an increase in CPC levels was only present, when patients were subjected to a single bout of exercise, which induced myocardial ischemia. (24) However, the functional capacity of CPCs improved significantly as a result of an aerobic exercise training intervention, which was partially the consequence of an enhanced expression of the homing factor CXCR4. (66) These data are consistent with the notion that an exercise training-mediated increase in NO bioavailability does not necessarily augment the number of CPCs in patients with CAD. (Figure1) One might speculate that the exercise training-mediated modulation of CPC function and count is associated with vascular repair, an improvement in endothelial function and hence organ perfusion, not only in patients with CAD but also in CHF. However, further studies are necessary do address this issue.

4. PERSPECTIVE

In patients with cardiovascular disease the decline in NO bioavailability, the loss of endothelial cells by apoptosis and functional alteration of CPCs are responsible for the deterioration of vasomotion in different vascular beds in CVD. However, exercise training restores the balance between NO production and NO inactivation by ROS, since it increases eNOS expression/phosphorylation but also reduces the NAD(P)H oxidase-derived generation of ROS. Moreover, ET might have the potential to restore the regenerative capacity of CPCs. Given the prognostic value of endothelial function further studies are necessary to elucidate whether the correction of vasomotion is the key mechanism responsible for the ET-mediated decline in mortality and morbidity in patients with CAD and CHF.

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Send correspondence to: Axel Linke, MD, University of Leipzig, Heart Center, Department of Internal Medicine and Cardiology, Struempellstrasse 39, 04289 Leipzig, Germany, Tel: 49-341-865-1672 Fax: 49-341-865-1461, E-mail: linkea@medizin.uni-leipzig.de