Critical limb ischemia (CLI) is an advanced stage of peripheral artery disease (PAD) and a serious health problem worldwide, even in clinical settings with established endovascular therapy. Currently, the limited therapeutic options cannot overcome the high mortality and morbidity rates of such patients with severe atherosclerotic status. Supervised aerobic exercise therapy is strongly recommended by clinical guidelines and is the most fundamental intervention for patients with PAD. Aerobic exercise induces functional recovery through mechanisms that include hypoxia-induced angiogenic growth factor expression in skeletal muscle and endothelial nitric oxide synthase (eNOS) activation by increased shear stress. However, performing adequate aerobic exercise is difficult, and the beneficial effects are limited to patients with CLI. In contrast, resistance training has been presented as an alternative exercise intervention for PAD patients. This training aims to maintain and increase muscle volume and can be performed at the bedside even by patients with CLI.

Neovascularization is an adaptive response to alleviating ischemia in tissues with compromised arterial blood supply, and in the adult candidate, it is thought to be brought about by arteriogenesis, which is the formation of fully differentiated endothelial cells in situ. Subsequent studies have shown that circulating progenitor cells mobilized from the bone marrow contribute to blood flow recovery after ischemia by homing to sites of neovascularization, and
differentiating into endothelial cells in the underperfused tissue. Heme oxygenase (HO) catalyzes heme degradation, which produces antioxidant biliverdin and carbon monoxide, antagonizing disturbed flow-induced reactive oxygen species. Of the 2 isoforms of HO, HO-1 is expressed at low levels in most tissues and is rapidly induced by tissue ischemia. The inducible HO-1 isoform has been shown to inhibit atherosclerotic plaque formation, protect from myocardial ischemia-reperfusion injury, and inhibit cardiac hypertrophy in animal models. HO-1 plays a crucial role during angiogenesis by mediating vascular endothelial growth factor (VEGF)-induced endothelial cell proliferation and promoting progenitor cell mobilization, neovascularization, and functional recovery after critical hindlimb ischemia (HLI) in mice. Because Akt/PKB is rapidly activated in response to strong oxidants, similarly to eNOS, HO-1 activity is reported to be regulated through Akt-mediated phosphorylation in vitro and in vivo. Ouchi et al previously reported successful functional muscle hypertrophy without exercise training using muscle-specific Akt1 transgenic (Akt1-TG) mice. However, the mechanism of blood flow recovery is unclear in terms of the relationship between Akt1 and HO-1.

In this issue of the Journal, Onoue et al clearly show this relationship using this transgenic mouse. They clarify that HO-1 is the most upregulated protein in Akt1-TG mice over wild types. The strongest point of their study is that it is the first report to clarify that Akt1-mediated muscle growth enhances HO-1 expression not in the muscle cells, but in the neighboring endothelial cells and macrophages in ischemic limbs. This paracrine mechanism may provide a hint for the development of a promising interventional strategy in the future. Akt1 upregulation mediates diverse protein secretion from different cells; therefore, additional verification that HO-1 supplementation could be a therapeutic option is warranted. As the authors discuss, Akt1-induced muscle hypertrophy may, at least temporarily, induce a relative muscular ischemia because of insufficient blood supply, suggesting that ischemia-related proteins appear in an ischemic area and augment angiogenesis to achieve a balance between the demand and supply of oxygen. A possible mechanism is that VEGF receptor activation may lead to HO-1 upregulation through the Akt signaling pathway or eNOS/Nr2/HO-1 pathway, or muscle-derived VEGF may directly augment HO-1 expression (Figure).

Unfortunately, the findings from the present study were unable to explain how HO-1 simply induced angiogenesis and increased the formation of capillaries in the ischemic tissue, resulting in functional recovery. Although blood flow was significantly increased in the Akt1-TG mice compared with either wild type or Akt1-TG mice treated with HO-1 inhibitor, no difference was found in the capillary density at ischemic sites. The authors try to explain this phenomenon as increased arteriogenesis rather than angiogenesis in an acute phase. Moreover, the mechanism of HO-1 upregulation in Akt1-TG mice remains to be clarified: HO-1 inhibitor could not always suppress HO-1 protein production. Most reports using the HLI model have limited observation periods of up to 2-4 weeks after the ischemic insult. Prior diverse interventions achieved a modest improvement in blood flow recovery that appeared only in the acute phase; however, in the later phase, the difference generally was attenuated over the untreated model mainly because of vigorous angiogenesis in the non-atherosclerotic animals. Although a dramatic increase in blood flow was achieved by endovascular revascularization, it resulted in a low rescue rate of ischemic legs because of the high restenosis/reocclusion rate within a short time after revascularization. Therefore, as much blood flow as possible is essential, and if this alternative intervention can create rapid and plentiful blood flow to the ischemic feet, it will improve the very poor clinical outcomes of patients with CLI.

Disclosures

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References