Conclusions: Based on our experiences, the frequency of recanalization after endovenous ablation while on warfarin is not worse compared with that described in the literature. The population size of this subset was small, but it appears antiplatelet agents also had no significant impact on incidence of recanalization. Thus, we believe it is safe to perform endovenous ablation on systemically anticoagulated patients with no appreciable negative impact on short-term durability or effectiveness.

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C8i: Poster Session—Research (1)

PS194.

Silencing of Int6 Promotes Recovery of Blood Perfusion After Limb Ischemia by Stabilizing Hypoxia-Inducible Factor 2α

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Objectives: Hypoxia-inducible factors (HIFs) are transcription factors that transcribe a spectrum of genes during hypoxia and other stress conditions. In particular, the HIF-2α subtype is more stable than HIF-1α in mild hypoxia and plays an essential role in vascular remodeling by transcribing angiogenic factors. We previously observed that silencing of Int6, a hypoxia-independent regulator of HIF-2α protein, led to neoangiogenesis by facilitating HIF-2α activity in normoxia. The aim of current study was to test the hypothesis that silencing of Int6 in muscle may enhance the recovery of blood flow in ischemic limbs.

Methods: We used a small interfering RNA (siRNA) plasmid designed to inhibit the Int6 gene, and assessed the influence of Int6 silencing on the gene and protein expression of mouse myoblasts quantitative RT-PCR and Western blotting. In vivo, BALB/c mice were randomized to treatment and control groups. After unilateral femoral artery ligation, the Int6 siRNA plasmid was injected into the muscle near the ligation site. Tissue damage and loss of limb function were scored for 28 days. Serial measurements of limb perfusion were also obtained by laser Doppler perfusion imaging. A random siRNA plasmid was given to the control group.

Results: Silencing of Int6 in cultured myoblasts led to stabilization of HIF-2α protein, accompanied by upregulation of angiogenic genes, including basic fibroblast growth factor and platelet-derived growth factor-B compared with the control ($P < .05$). In a mouse model of hind limb ischemia, intramuscular injection of the Int6 siRNA plasmid significantly enhanced perfusion ($P < .05$ at days 7 and 14) and functional recovery ($P < .05$ at days 7, 14, and 21) of damaged limbs.

Conclusions: Silencing of Int6 in muscle led to enhanced perfusion and functional recovery in ischemic limbs. Int6 may serve as a therapeutic target to control angiogenesis in ischemic diseases.

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PS196.

Increased Adipose-Derived Mesenchymal Stem Cells Counts and Pro-B-Type Natriuretic Peptide in Patients With Critical Limb Ischemia

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Objectives: Mesenchymal stem cells (MSCs) have been shown to improve regeneration of injured tissues in vivo. Several in vitro studies and animal models have demonstrated improvement in MSCs paracrine effects under hypoxic conditions. Moreover, several studies suggested that the pro B-type natriuretic peptide (pro-BNP) could be involved in the stimulation of postischemic vascular regeneration. The purpose of this study was to investigate the effect of critical limb ischemia, in a human model, on in situ adipose-derived mesenchymal stem cells (ADMSCs) and to determine whether serum levels of N-terminal pro-BNP correlate with ADMSCs counts and associated paracrine effects.

Methods: Lipoaspirate samples of ≥10 mL were collected from ischemic limbs (ischemic group) and compared with control samples (without ischemia). MSCs were characterized by frequency, viability, differentiation potential, cytokines expression, and cell surface markers. Serum NT pro-BNP was measured as well.

Results: MSCs counts were ninefold to 10-fold higher in patients with ischemic limbs (mean 7952 ± 542 MSC/mL) than controls (mean 790 ± 65 MSC/mL). Pro-BNP levels (range, 1878-4757 pg/mL) were approximately eightfold to 26-fold higher than in age- and sex-matched controls. Furthermore, there were positive correlations between pro-BNP levels and MSCs counts in the ischemic group.

Conclusions: Patients with critical limb ischemia (CLI) have higher levels of pro-BNP and MSCs counts than controls. Increased levels of pro-BNP and MSCs counts can be considered humoral and cellular surrogates of ischemia and hypoxia in patients with CLI. This supports recent studies that suggest that the increase production of peripheral BNP may be a stem cells-mediated response to stimulate angiogenesis in the ischemic skeletal muscles.

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