

# Hindlimb ischemia

The murine hindlimb ischemia preparation is a model of peripheral arterial disease (PAD), and is useful for testing new therapies.

Previous studies have shown that human [urine derived stem cells](#) (USCs) have great potential as a cell source for [cytotherapy](#) and [tissue engineering](#) and that [extracellular vesicles](#) (EVs) secreted by USCs (USCs-EVs) can prevent [diabetes](#)-induced [kidney](#) injury in an animal model. The present study was designed to evaluate the effects of USCs-EVs on ischemia repair.

USCs-EVs were isolated and purified by a battery of centrifugation and filtration steps. The USCs-EVs were then characterized by [transmission electron microscopy](#), [western blot](#) and tunable resistive pulse sensing techniques. After intramuscularly transplanting USCs-EVs into an [hindlimb ischemia](#) mouse, we observed the therapeutic effects of USCs-EVs on perfusion by laser doppler perfusion imaging, angiogenesis and muscle regeneration by histology and immunohistochemistry techniques over 21 days. We subsequently tested whether USCs-EVs can induce the proliferation of a human microvascular endothelial cell line HMEC-1 and a mouse myoblast cell line C2C12 by cell counting kit 8 assay in vitro. Meanwhile, the potential growth factors in the USCs-EVs and supernatants of the USCs cultures were detected by enzyme-linked immunosorbent assay.

The USCs-EVs were spherical vesicles with a diameter of 30-150 nm and expressed exosomal markers, such as CD9, CD63 and Tsg101. Ischemic limb perfusion and function were markedly increased in the hind-limb ischemia (HLI) model after USCs-EVs administration. Moreover, angiogenesis and muscle regeneration levels were significantly higher in the USCs-EVs treatment group than in the PBS group. The in vitro experiments showed that USCs-EVs facilitated HMEC-1 and C2C12 cell proliferation in a dose-dependent manner.

These results revealed for the first time that USCs-EVs efficiently attenuate severe hind-limb ischemic injury and represent a novel therapy for HLI <sup>1)</sup>.

<sup>1)</sup>

Zhu Q, Li Q, Niu X, Zhang G, Ling X, Zhang J, Wang Y, Deng Z. Extracellular Vesicles Secreted by Human Urine-Derived Stem Cells Promote Ischemia Repair in a Mouse Model of Hind-Limb Ischemia. *Cell Physiol Biochem*. 2018 Jul 24;47(3):1181-1192. doi: 10.1159/000490214. [Epub ahead of print] PubMed PMID: 30041250.

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