

Myeloid hypoxia inducible factor-1 α is essential for skeletal muscle regeneration in mice

Supplementary material

Supplementary figure 1

HIF-1 α in skeletal muscle cells did not influence the acute severity of trauma and was not required for adequate skeletal muscle regeneration. **(A and B)** Neither the extent of traumatic lesion nor the serum creatine kinase levels differed significantly between HIF-1 $\alpha^{+/+}$ and MCKCre/HIF-1 $\alpha^{+/+}$ mice. **(C and D)** There were neither differences in the cross-sectional area of regenerating myofibres nor in the extent of necrotic cell debris at 7d after injury between HIF-1 $\alpha^{+/+}$ and MCKCre/HIF-1 $\alpha^{+/+}$ mice. Each group included four to five mice. Data are mean \pm SEM. XSA=cross-sectional area.

Supplementary figure 2

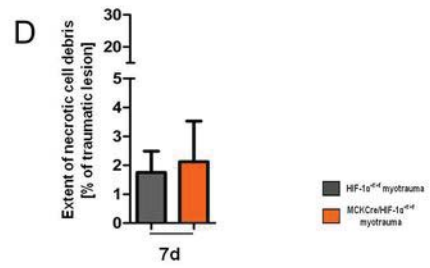
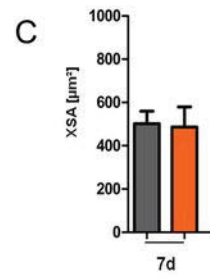
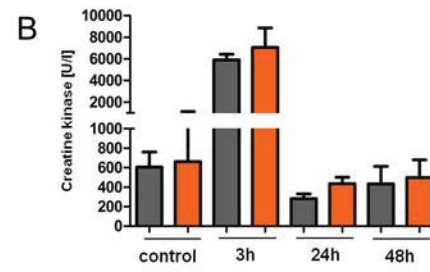
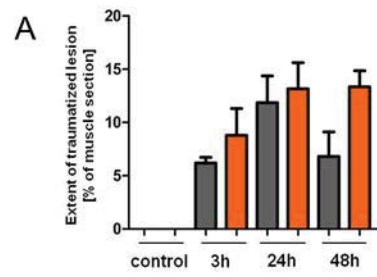
Myeloid-specific knock-out of VEGF did neither impair regeneration of muscle tissue nor of vasculature after skeletal muscle injury. **(A)** The extent of necrotic cell debris was not changed in mice with a VEGF knock-out in myeloid cells. **(B)** Numbers of CD34-positive blood-endothelial cells were not decreased at the injury site of LysMCre/VEGF $^{+/+}$ mice. Each group included five to six mice. Data are mean \pm SEM.

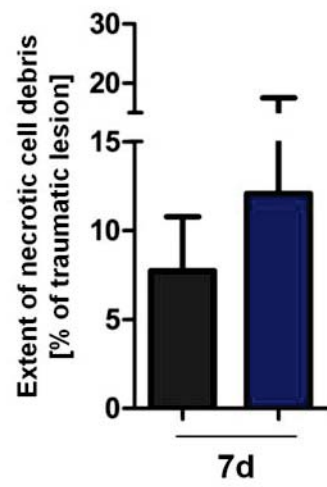
Supplementary figure 3

Myeloid cells, isolated from injured skeletal muscle, showed high purity and high deletion efficiency for HIF-1 α . **(A)** The purity of isolated myeloid cells accounted more than 80 % and was examined by FACS-analysis with a CD11b antibody. **(B)** The HIF-1 α deletion efficiency of myeloid cells, isolated from LysMCre/HIF-1 $\alpha^{+f/+f}$ mice, was approximately 75 %. Each group included five to six mice. Data are mean \pm SEM.

Supplementary figure 4

Macrophages of injured skeletal muscles of HIF-1 $\alpha^{+f/+f}$ and LysMCre/HIF-1 $\alpha^{+f/+f}$ mice showed a M2-phenotype. **(A and B)** Macrophages, isolated from injured skeletal muscles, showed approximately 100 times higher expression of arginase-1 mRNA than iNOS mRNA. **(C)** Representative iNOS- and YM-1-immunostaining. Both wild-type and HIF-1 α -deficient macrophages showed low expression of the M1-marker iNOS, but high expression of the M2-marker YM-1. Each group included five to nine mice. Scale bars: 50 μ m. Data are mean \pm SEM. YM-1=T-lymphocyte-derived eosinophil chemotactic factor.



A**B**