

## Effect of CD36 on vascular repair

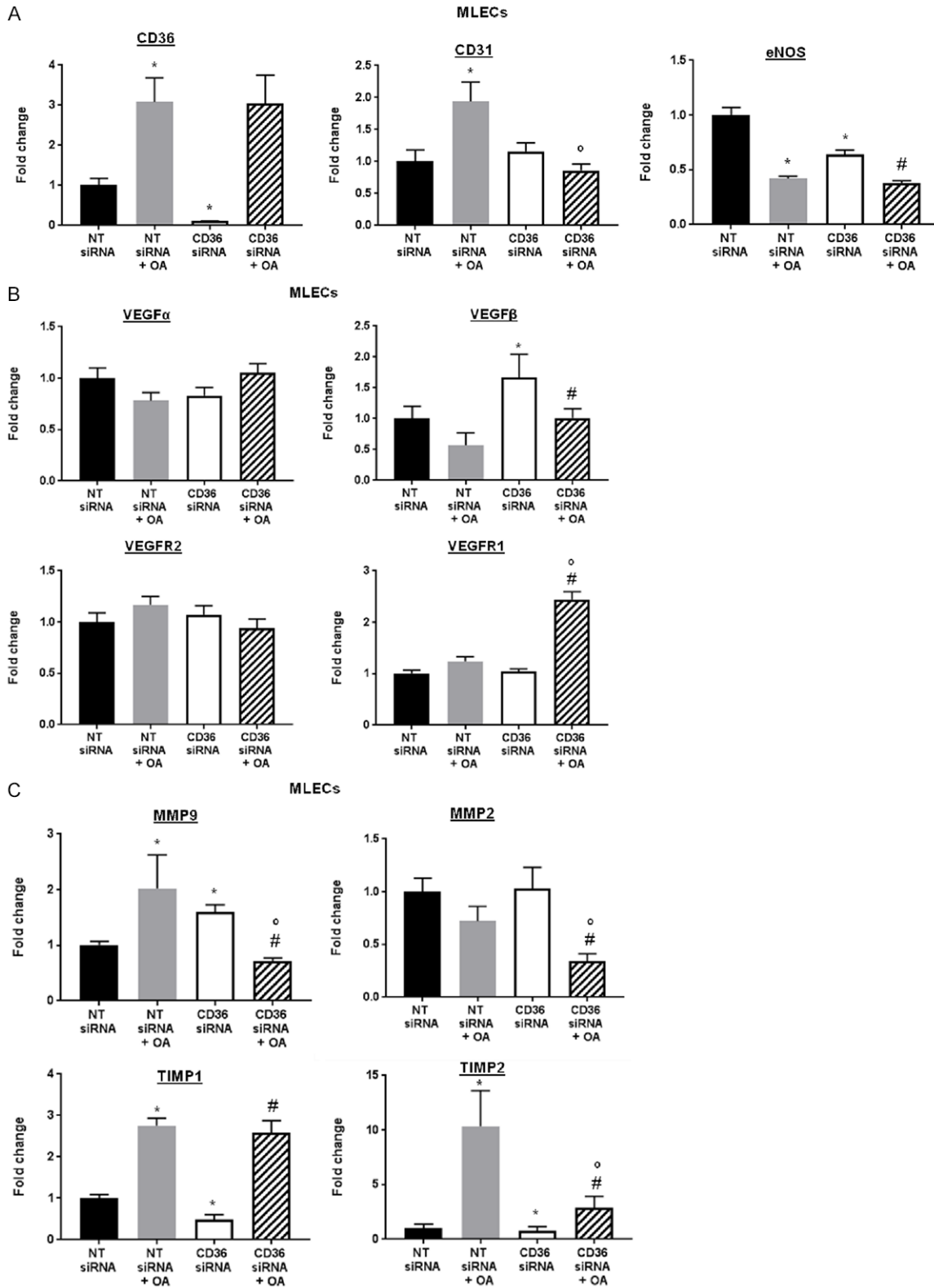
**Supplementary Table 1.** Mouse-specific primers for endothelial and angiogenic marker genes for qRT-PCR

Gene	Primer (5'→3')
CD36 F	ATGGGCTGTGATCGGAACTG
CD36 R	GTCTTCCAATAAGCATGTCTCC
CD36 Ex4 F	AACACTGTGATTGTACCTG
CD36 Ex4 R	TCAATAAGCATGTCTCCGAC
MMP9 F	CTGTCCGGCTGTGGTTCAGT
MMP9 R	AGACGACATAGACGGCATCC
MMP2 F	GGGGTCCATTTTCTTCTCA
MMP2 R	CCAGCAAGTAGATGCTGCCT
VEGF $\alpha$ F	AATGCTTTCTCCGCTCTGAA
VEGF $\alpha$ R	GCTTCCTACAGCACAGCAGA
VEGFR2 F	TCCAGAATCCTCTTCCATGC
VEGFR2 R	AAACCTCCTGCAAGCAAATG
VEGF $\beta$ F	GTGAAGCAGGGCCATAAAAG
VEGF $\beta$ R	GAGCTCAACCCAGACACCTG
VEGFR1 F	AAGAGAGTCTGGCCTGCTTG
VEGFR1 R	CTGCTCGGGTGTCTGCTT
eNOS F	CCTAGGGGAGCTGTTGTACG
eNOS R	GACCAGCACATTTGGCAAT
CD31 F	CTTTTCGAGGTGGTGCTGAT
CD31 R	CCTCCAGGCTGAGGAAAAC
Ki67 F	CTGCCTGCGAAGAGAGCATC
Ki67 R	AGCTCCACTTCGCCTTTTGG
TIMP1 F	AGGTGGTCTCGTTGATTCGT
TIMP1 R	GTAAGGCCTGTAGCTGTGCC
TIMP2 F	GAATCCTCTTGATGGGGTTG
TIMP2 R	CGTTTTGCAATGCAGACGTA
TIMP3 F	TAGACCAGAGTGCCAAAGGG
TIMP3 R	CCAGGATGCCTTCTGCAAC
TIMP4 F	GGGCTCAATGTAGTTGCACA
TIMP4 R	AGAAACCAACAGTCACAAGCA
AMPK $\alpha$ F	ACAGGCCATAAAGTGGCAGTT
AMPK $\alpha$ R	AAAAGTCTGTCGGAGTGCTGA
ATGL F	CGCCTTGCTGAGAATCACCAT
ATGL R	AGTGAGTGGCTGGTAAAAGGT
FASN F	TTGCTGGCACTACAGAATGC
FASN R	AACAGCCTCAGAGCGACAAT
Cpt1a F	CATGTCAAGCCAGACGAAGA
Cpt1a R	TGGTAGGAGAGCAGCACCTT
Cpt1b F	GTCGCTTCTCAAGGTCTGG
Cpt1b R	GGTCTCATCGTCAGGGTTGT
GLUT1 F	GCTGTGCTTATGGGCTTCTC
GLUT1 R	CACATACATGGGCACAAAGC
GLUT4 F	ACTCTTGCCACACAGGCTCT
GLUT4 R	CCTTGCCCTGTCAGGTATGT
PLIN2 F	CTACGACGACACCGAT
PLIN2 R	CATTGCGGAATACGGAG

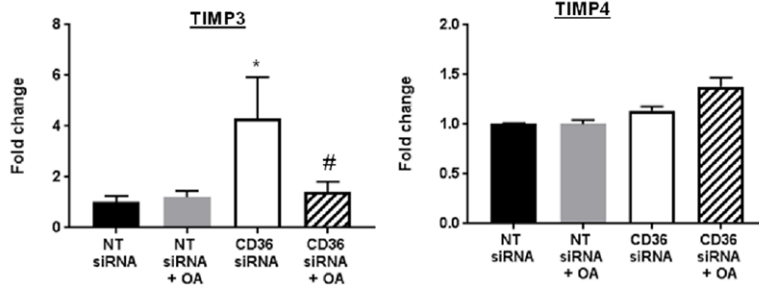
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RPS3 F	AGCTTCCCAGACACCACAAC
RPS3 R	ACAAACTCCTTGAGGGCTT
18S F	GTAACCCGTTGAACCCATT
18S R	CCATCCAATCGGTAGTAGCG

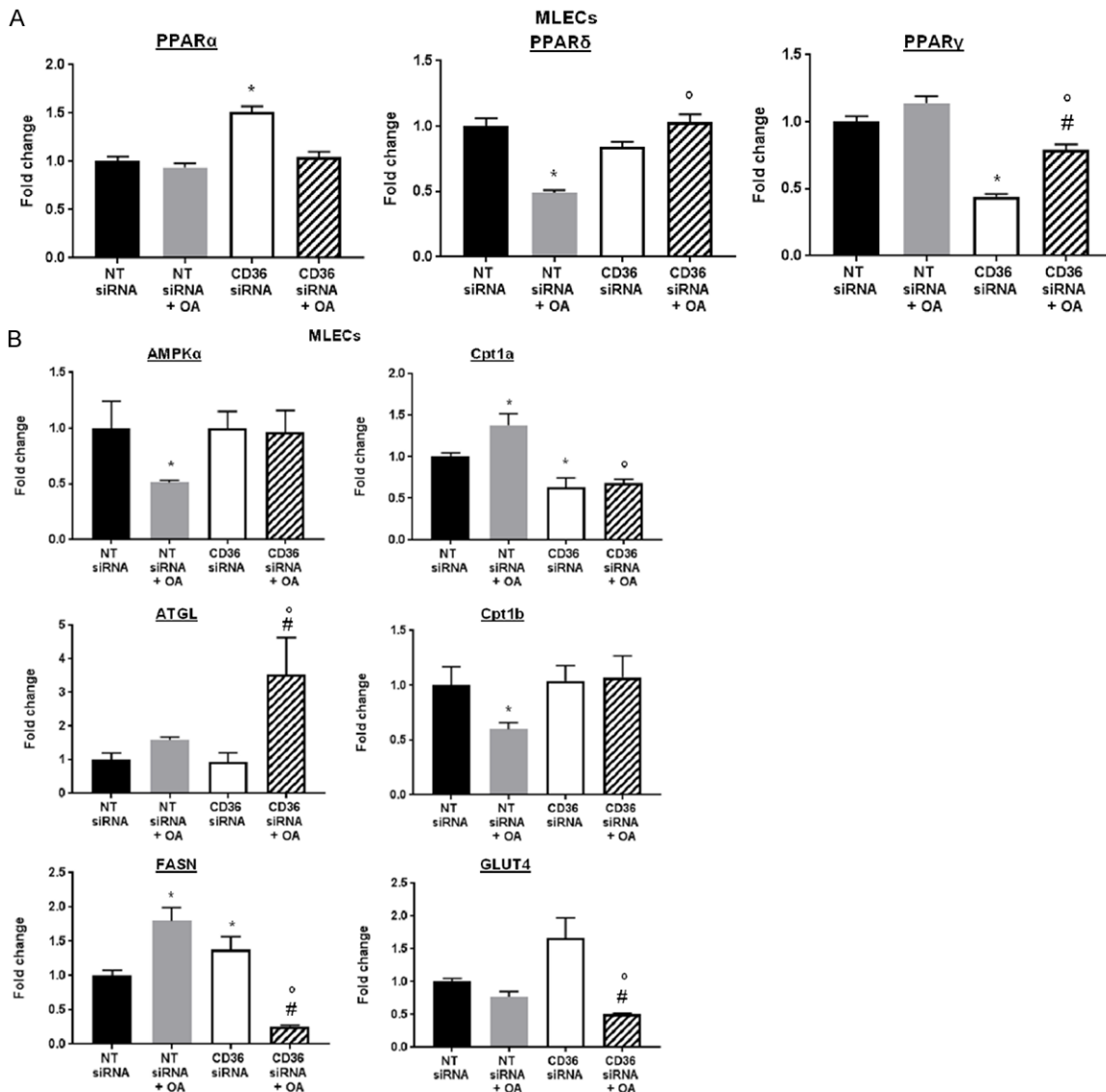
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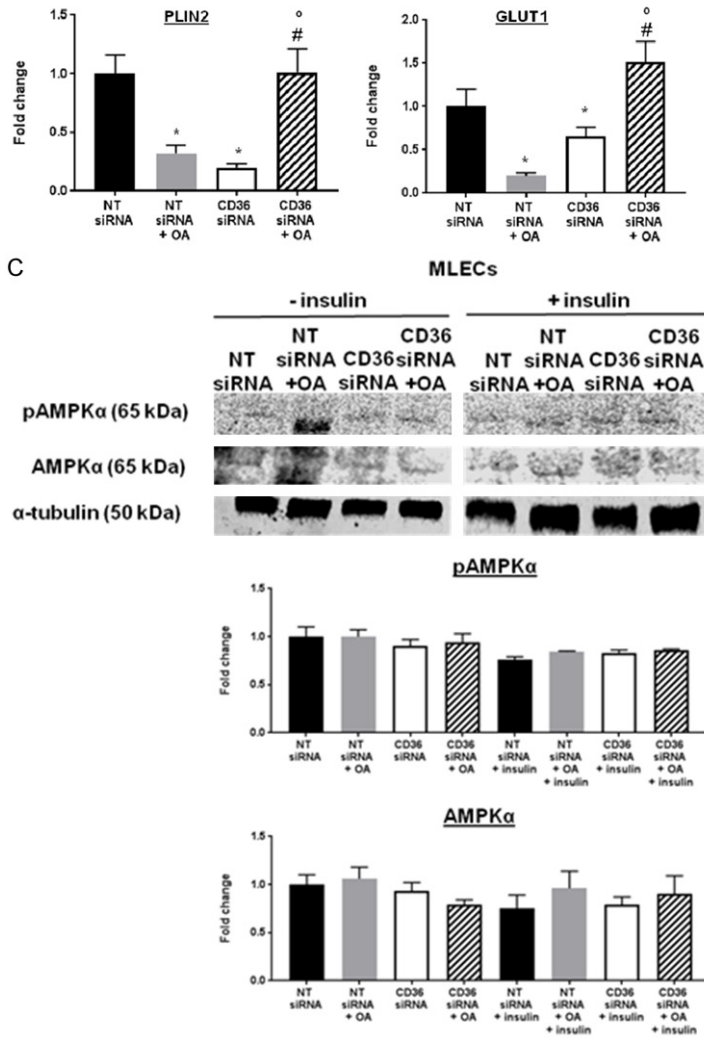
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**Supplementary Figure 1.** Effect of oleic acid on gene expression of endothelial and angiogenic markers in CD36-deficient MLECs. Effect of oleic acid (OA, 300  $\mu$ mol/L) and siRNA-mediated CD36 knockdown on MLEC endothelial and angiogenic marker mRNA expression: (A) CD36, CD31, eNOS (B) VEGF $\alpha$ , VEGFR2, VEGF $\beta$ , VEGFR1 and (C) MMP9, MMP2, TIMP1, TIMP2, TIMP3, TIMP4 assessed by quantitative realtime PCR (qRT-PCR) normalized to 18S mRNA expression. Histograms show fold change in mRNA expression compared to NT siRNA ECs. Data represent mean  $\pm$  SEM, n=6, \*p < 0.05 vs. NT siRNA, °p < 0.05 vs. NT siRNA + OA, #p < 0.05 vs. CD36 siRNA. Two-way ANOVA statistical tests were used to determine statistical significance.

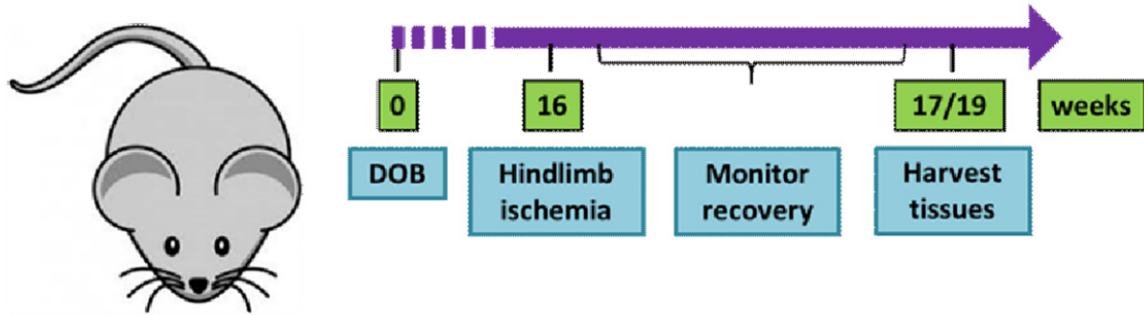


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**Supplementary Figure 2.** Effect of oleic acid on gene expression of metabolic markers and protein expression of AMPK in CD36-deficient MLECs. Effect of oleic acid (OA, 300  $\mu$ mol/L) and siRNA-mediated CD36 knockdown on MLEC metabolic gene mRNA expression: (A) PPAR $\alpha$ , PPAR $\delta$ , PPAR $\gamma$  (B) AMPK $\alpha$ , ATGL, FASN, PLIN2, Cpt1a, Cpt1b, GLUT4, and GLUT1. Histograms show fold change in mRNA expression compared to NT siRNA ECs. Data represent mean  $\pm$  SEM, n=5, \*p < 0.05 vs. NT siRNA, <sup>o</sup>p < 0.05 vs. NT siRNA + OA, #p < 0.05 vs. CD36 siRNA; (C) Effect of oleic acid (OA, 300  $\mu$ mol/L) and insulin (50 units/mL) and siRNA-mediated CD36 knockdown on MLEC intracellular signaling proteins as assessed by western blot. Images of SDS-PAGE gels represent protein expression of phospho-AMPK, AMPK and housekeeping gene  $\alpha$ -tubulin. Histograms show fold change in protein expression compared to NT siRNA ECs. Data represent mean  $\pm$  SEM, n=3. Two-way ANOVA statistical tests were used to determine statistical significance.

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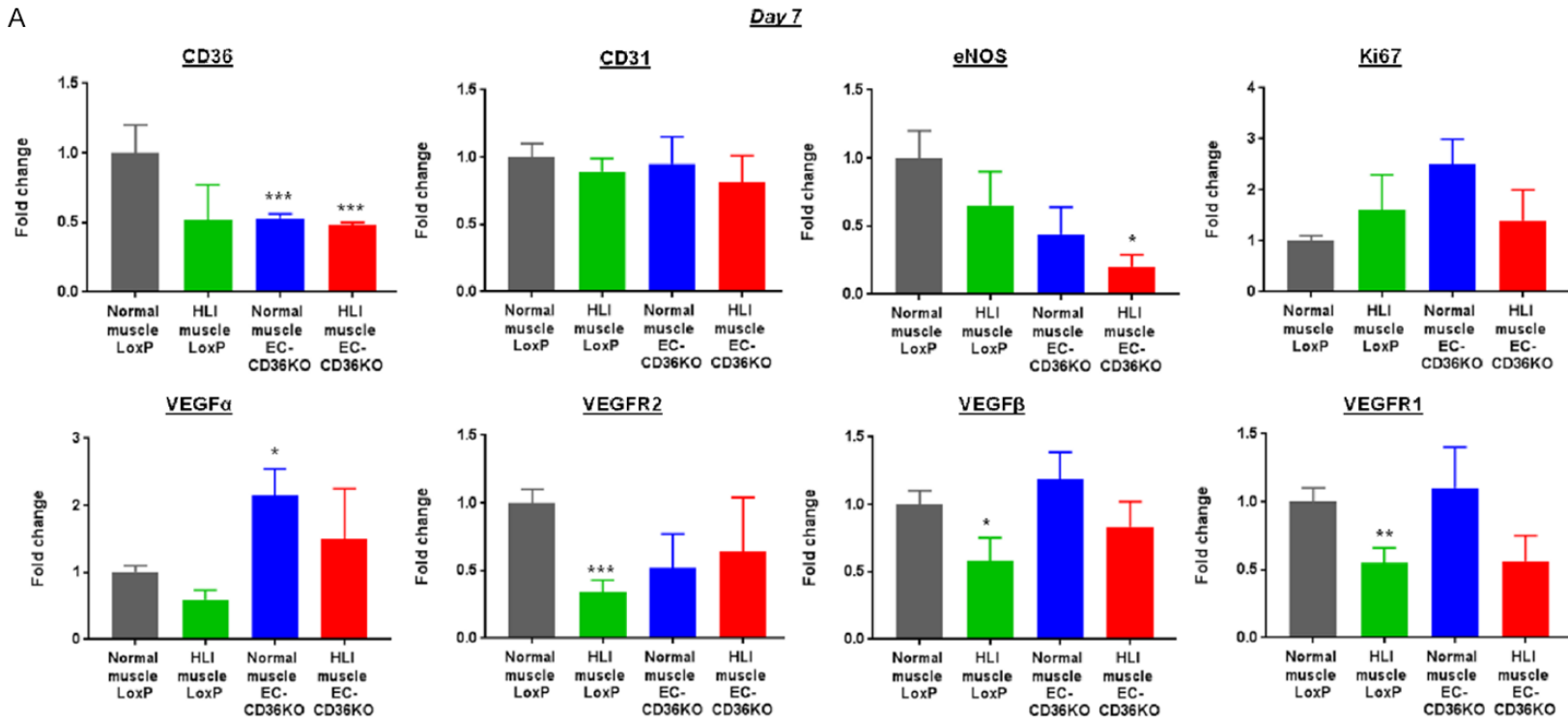


- **EC-CD36 KO mice (EC-*Cd36*<sup>-/-</sup>): C57BL/6J-*CD36*<sup>fl/fl</sup>/Tie2-cre**
- **LoxP control mice (EC-*Cd36*<sup>fl/fl</sup>): C57BL/6J-*CD36*<sup>fl/fl</sup>**
- **Hindlimb ischemia (HLI): ligation of femoral artery**
- **Recovery monitored over 7 and 21 days**

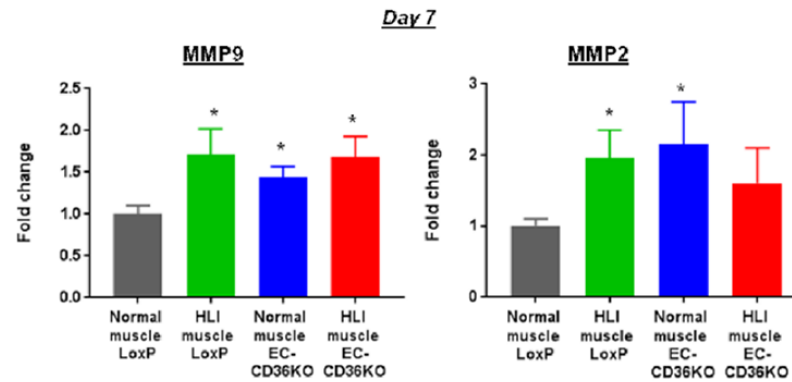
**Supplementary Figure 3.** *In vivo* hindlimb ischemia (HLI) mouse model of peripheral vascular disease (A) Figure shows the timeline of surgery and recovery of LoxP control mice (EC-*Cd36*<sup>fl/fl</sup>) and EC-CD36 KO mice (EC-*Cd36*<sup>-/-</sup>) whereby recovery was monitored over 7 and 21 days.

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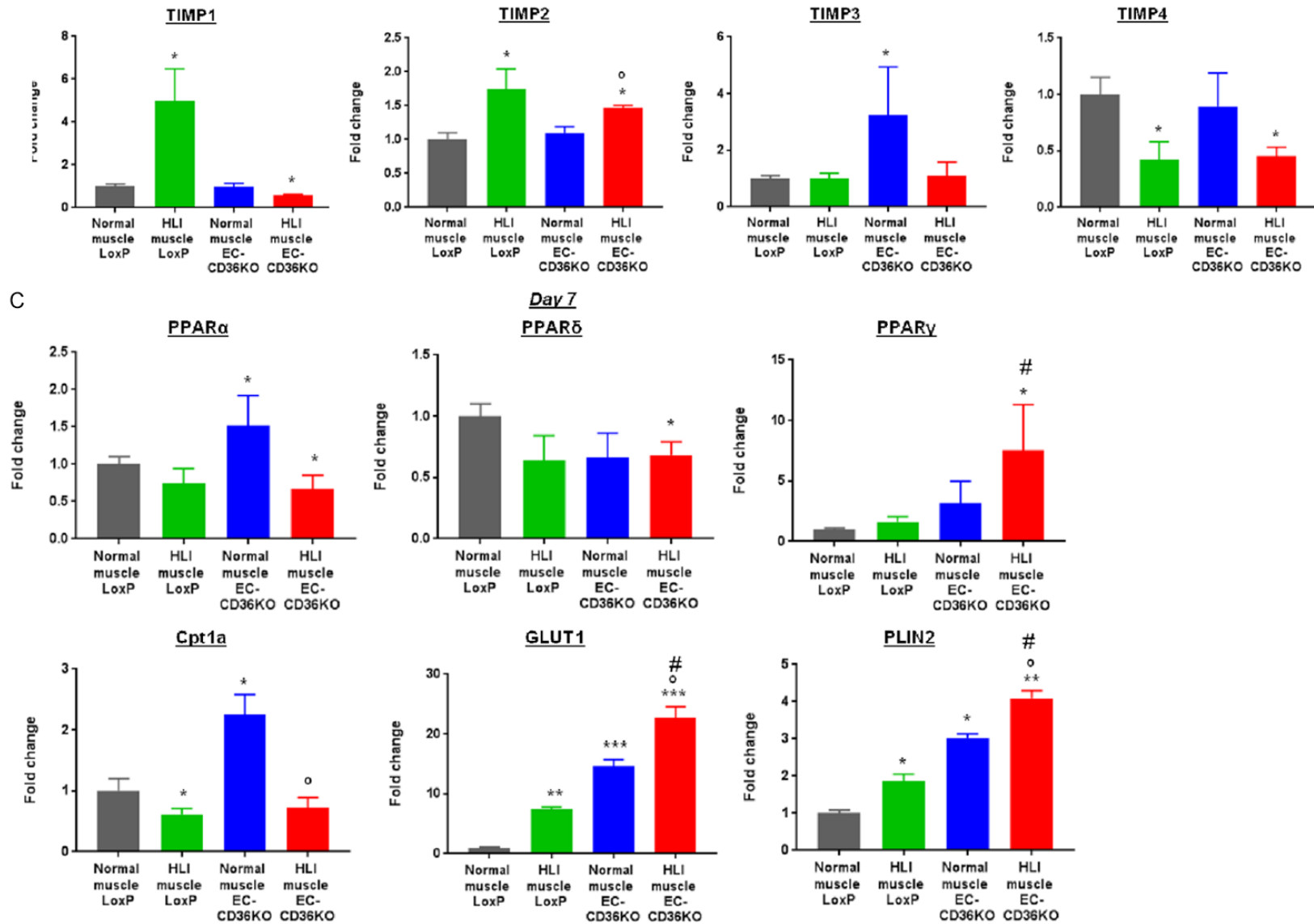
A



B



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**Supplementary Figure 4.** Effect of EC-CD36 knockout in HLI mouse muscle tissues on endothelial, vascular and metabolic gene expression at 7 days post-HLI. *In vivo* hindlimb ischemia (HLI) mouse tissue mRNA expression in normal and HLI muscle tissues of LoxP and EC-CD36KO mice of (A) day 7 post-ischemia mRNA expression of CD36, CD31, eNOS, Ki67, VEGF $\alpha$ , VEGFR2, VEGF $\beta$ , and VEGFR1. Histograms represent fold changes in mRNA expression normalized to RPS3 mRNA expression. Data represent mean  $\pm$  SEM, n=8, \*p < 0.05, \*\*p < 0.01, \*\*\*p < 0.001 vs. normal muscle LoxP; (B) day 7 post-ischemia mRNA expression of MMP9, MMP2, TIMP1, TIMP2, TIMP3, and TIMP4. Histograms represent fold changes in mRNA expression normalized to RPS3 mRNA expression. Data represent mean  $\pm$  SEM, n=8, \*p < 0.05, \*\*p < 0.01 vs. normal muscle LoxP, °p < 0.01 vs. normal muscle EC-CD36KO; (C) day 7 post-ischemia mRNA expression of Cpt1a, GLUT1, and PLIN2. Histograms represent fold changes in mRNA expression normalized to RPS3 mRNA expression. Data represent mean  $\pm$  SEM, n=8, \*p < 0.05, \*\*p < 0.01 vs. normal muscle LoxP, °p < 0.01 vs. normal muscle EC-CD36 KO, #p < 0.01 vs. HLI muscle LoxP. Two-way ANOVA statistical tests were used to determine statistical significance.