

# Supplemental data

**Supplemental Table 1.** Sequences of primers for RT-PCR on cultivated of EPCs

Transcript	Sequence
<i>Ccr1</i>	(F) 5'-TTTTAAGGCCCAAGTGGGAGTTCACTCACC-3'
	(R) 5'-TGGTATAGCCACATGCCTTTGAAACAGCTG-3'
<i>Ccr2</i>	(F) 5'-CCTGCAAAGACCAGAAGAGG-3'
	(R) 5'-GATGGCCAAGTTGAGCAGAT-3'
<i>Ccr5</i>	(F) 5'-ACGGTGTTCAATTTTCCAGC-3'
	(R) 5'-GCAAGAAGCGACTTTATGGC-3'
<i>Cxcr4</i>	(F) 5'-TCCTGCCCCACCATCTACTTC-3'
	(R) 5'-TTTCAGCCAGCAGTTTCCTT-3'
<i>Flk1</i>	(F) 5'-AGTGGCTCTGTCCTCCAAGA-3'
	(R) 5'-GCAAACCTTCCAAAACCAAA-3'
<i>Vegf</i>	(F) 5'-TCCAACATCACCATGCAGAT-3'
	(R) 5'-CATCTGCAAGTACGTTTCGTT-3'
<i>Tgfb1</i>	(F) 5'-GGGCAAGACAGTCATCGAAT-3'
	(R) 5'-TTGGTTTTTGGTCACGTTCA-3'
<i>Hgf</i>	(F) 5'-GCTTGGCATCCACGATGTTC-3'
	(R) 5'-CCCTCACATGGTCCTGATCC-3'
$\beta$ -actin	(F) 5'-TTCTACAATGAGCTGCGTGTGGC-3'
	(R) 5'-CTCATAGCTCTTCTCCAGGGAGGA-3'

(F): Forward primer, (R): Reverse primer

### **Legends to Supplemental Figures**

**Supplemental Figure 1.** The protein levels of CCL3, CCL4, and CCL5 at the wound sites of WT mice were determined by ELISA. All values represent mean  $\pm$  SEM (n=6). \*\*,  $P < 0.01$ , vs. uninjured skin (time=0).

**Supplemental Figure 2.** Hydroxyproline content in wound site of WT and *Ccr5*<sup>-/-</sup> mice. All values represent mean  $\pm$  SEM (n=6). \*,  $P < 0.05$ , vs. WT mice.

**Supplemental Figure 3.** Neovascularization was immunohistochemically evaluated with anti-VE-cadherin mAb at 6 days after injury. (A) Representative results from four animals are shown here. Original magnification, x 200. (B) The vascular areas were identified as VE-cadherin-positive areas with Adobe PhotoShop. Values represent mean  $\pm$  SEM (n=4). \*\*,  $P < 0.01$ , vs. WT mice.

**Supplemental Figure 4.** Hydroxyproline content in wound site of BM chimera mice. All values represent mean  $\pm$  SEM. \*,  $P < 0.05$ , vs. WT recipients transferred WT donor BM cells; #,  $P < 0.05$ , vs. *Ccr5*<sup>-/-</sup> recipients transferred *Ccr5*<sup>-/-</sup> donor BM cells.

**Supplemental Figure 5.** Quantitative evaluation of Flk-1<sup>+</sup>/CD34<sup>+</sup> EPCs by flow cytometric analysis at the indicated time intervals after injury. Representative results from six independent experiments are shown here; (A) BM, (C) peripheral blood, and (E) wound sites. Changes in percentage of Flk-1<sup>+</sup>/CD34<sup>+</sup> cells in BM (B), peripheral blood (D), and wound sites (F) are shown. Values represent mean  $\pm$  SEM (n=6). \*\*,  $P < 0.01$ , vs. WT mice.

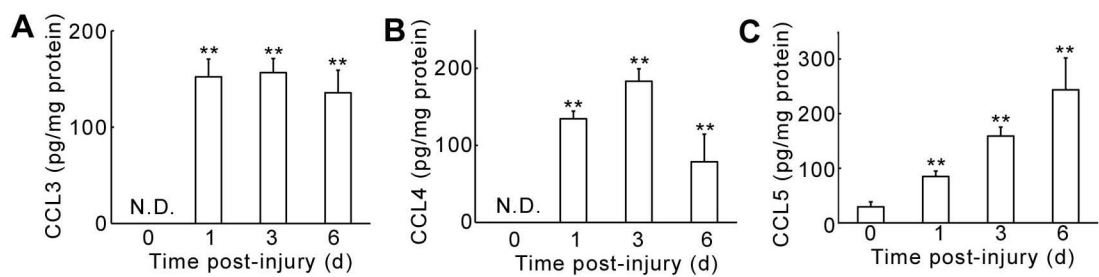
**Supplemental Figure 6.** Immunofluorescence analysis of c-Kit<sup>+</sup>/Tie-2<sup>+</sup> cells (left panel) and c-Kit<sup>+</sup>/Tie-2<sup>+</sup>/CCR5<sup>+</sup> cells (right panel) in the wound of WT mice at 4 days after injury. Representative results from six independent experiments are shown. Original magnification, x 400.

**Supplemental Figure 7.** (A) BM mononuclear cells were cultured as described in Materials and Methods. After a 4-day culture, adhered and spindle-shaped cells were recognized as EPCs and the cells reached confluent monolayer at 14 days after the culture. Original magnification, x 100. (B) On day 4, cultured EPCs were characterized with both AcLDL uptake (Dil) and BS-1 lectin binding (FITC). Original magnification, x 100. (C) To identify the characteristics of the cells, 4-day cultured cells were subjected to immunofluorescent staining with anti-Tie-2, together with AcLDL uptake and BS-1 lectin binding. Original magnification, x 200. (D) Double-color immunofluorescence analysis of the obtained EPCs using anti-Tie-2 and anti-CCR5, followed by DAPI. Some of cultivated Tie-2<sup>+</sup> EPCs expressed CCR5.

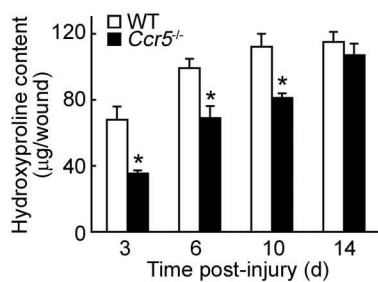
**Supplemental Figure 8.** The gene expression of *Vegf*, *Tgfb1* and *Hgf* in the resultant EPCs by RT-PCR analysis. Representative results from six independent experiments are shown here.

**Supplemental Figure 9.** The gene expression of *Ccr1*, *Ccr2*, *Ccr5*, *Cxcr4* and *Flk-1* in the resultant EPCs by RT-PCR analysis. Representative results from six independent experiments are shown here.

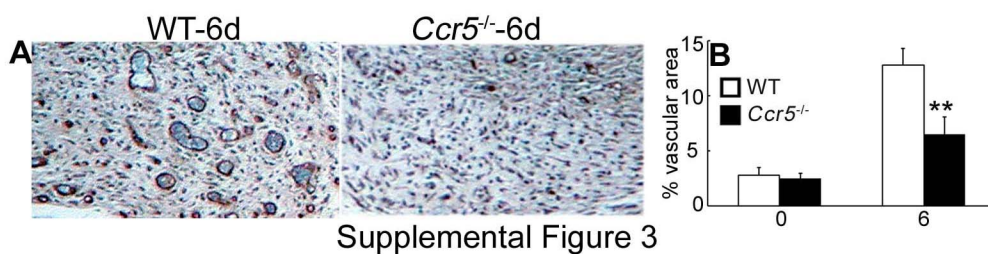
**Supplemental Figure 10.** (A) and (B) The mRNA expression of *Cxcl12* and *Ccl2* at the wound sites of WT and *Ccr5*<sup>-/-</sup> mice by quantitative RT-PCR analyses. (C) and (D) Protein levels of CXCL12 and CCL2 at the wound sites of WT and CCR5<sup>-/-</sup> mice by ELISA analyses. Values represent mean ± SEM (n=6).



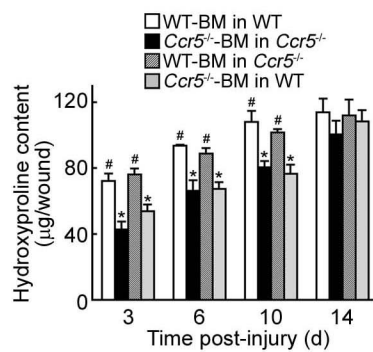
Supplemental Figure 1



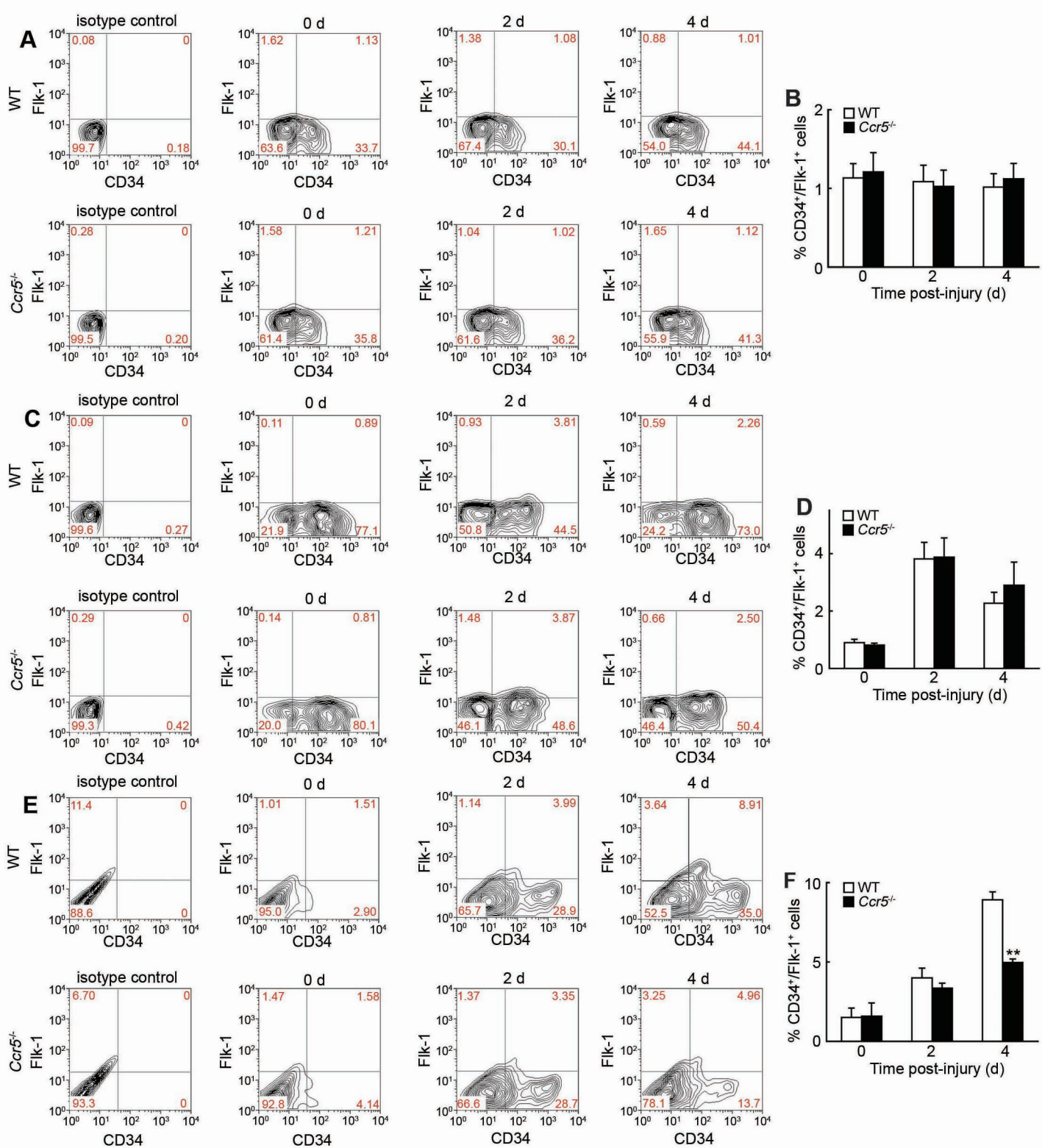
Supplemental Figure 2



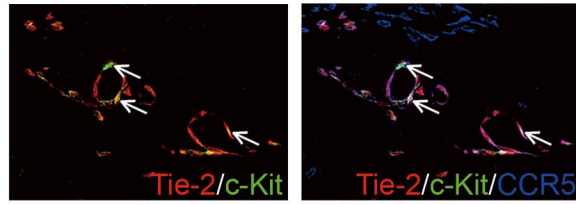
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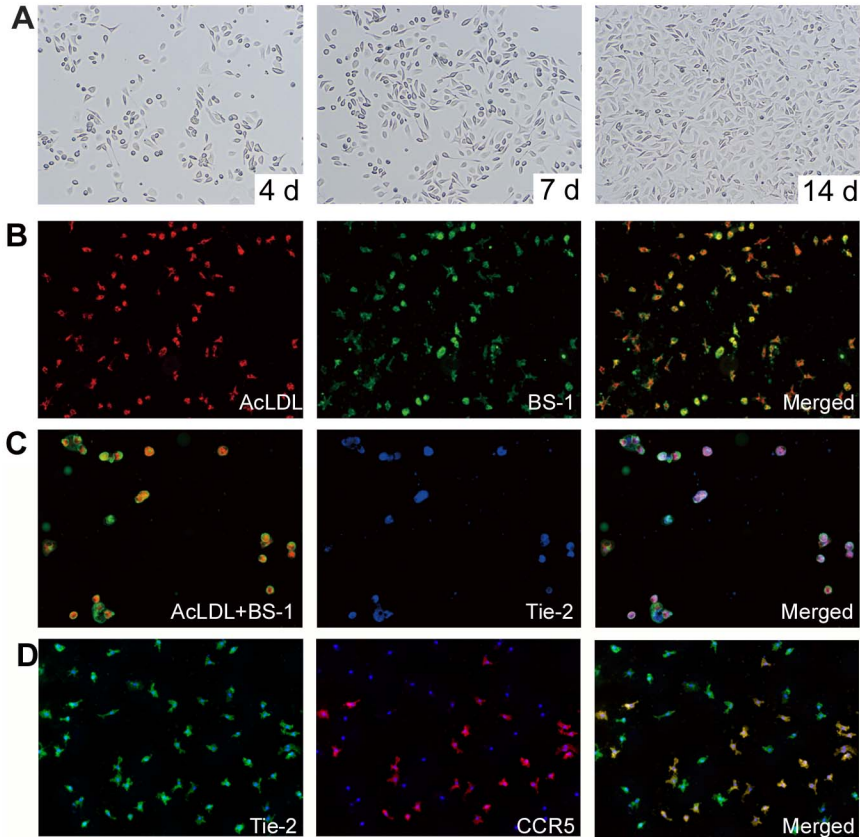
Supplemental Figure 4



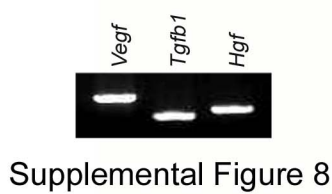
Supplemental Figure 5



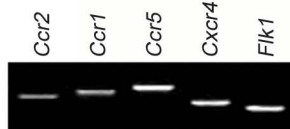
Supplemental Figure 6



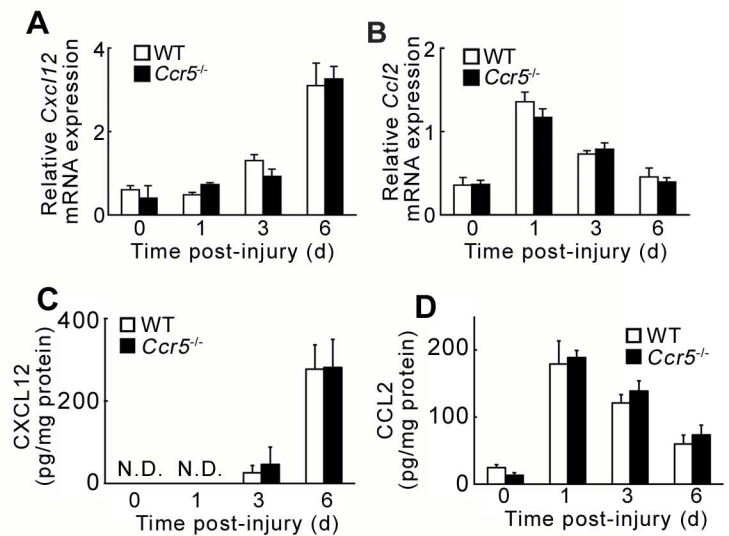
Supplemental Figure 7



Supplemental Figure 8



Supplemental Figure 9



Supplemental Figure 10