Supplemental Figure 1

Neutrophil response to MI in P1 and P14 mice. (A) Relative percentages of neutrophils in the heart before MI (Pre) or at the specified days after MI at P1 or P14. Neutrophils (CD11b+Ly-6G+) were identified according to representative plots in Figure 1. (B) NIMP-R14+ staining to identify neutrophils at 7 days following MI at P1 or P14. Arrows highlight positively stained cells. Inset shows higher magnification image of positively stained neutrophils. Scale bar: 200µm (C) Quantification of NIMP-R14+ neutrophils spanning the entire heart section at day 7 post MI. Data represent mean ± SEM. ns=not significant. **P<0.01, ***P<0.001, ns=not significant.
Supplemental Figure 2
Splenic mononuclear phagocyte response to MI at P1 and P14. (A-B) Spleen cells isolated from uninjured mice or at the indicated days following MI at P1 or P14 were stained with anti-CD11b, -Ly-6G, -Ly-6C, -F4/80, -CD11c, -I-A^b mAbs and analyzed by FACS. (A) The percentage of mononuclear phagocytes (CD11b^+Ly-6G^-) was quantified relative to the percent enriched leukocyte gate (n=3-5/time-point). (B) Relative percentage of Ly-6C^hi and Ly-6C^lo splenic monocytes are shown for indicated times following MI at P1 or P14 (n=3-5/time-point). Data are mean ± SEM. **p<0.01, ***p<0.001.
**Supplemental Figure 3**

Lymphocyte response to MI at P1 and P14. (A) Representative dot plots depict FACS analysis of pooled spleens isolated from uninjured mice or at the indicated days following MI at P1 or P14 and stained with anti-B220 and -TCRβ mAbs. B-lymphocytes were identified as (B220+TCRβ) and all TCRβ+ cells included as T-lymphocytes. Percentages of cells are indicated for the representative dot plots. (B) TCRβ+ T-lymphocytes were stained with anti-CD25 and -CD4 to further delineate into CD25+CD4- (upper left), CD25+CD4+ (lower right) or CD25-CD4+ (upper right). Percentages of cells are indicated for the representative dot plots. (C) The percentage of splenic B- or T-lymphocytes was quantified relative to the percent enriched leukocyte gate before injury or at the indicated days following MI at P1 or P14 (n=3-5/time-point). (D) T-lymphocyte subsets were quantified and are expressed as a relative percentage of all TCR+ cells (n=3-5/time-point). Data are mean ± SEM. *P<0.05, **P<0.01, ***P<0.001.
**Supplemental Figure 4** Mononuclear phagocyte sorting from hearts following MI at P1 and P14

**A** Cell suspensions from hearts infarcted at P1 or P14 (3 days post MI) were stained with antibodies to distinguish mononuclear phagocytes. The population used for gene expression analysis by microarray (Pos) was compared to the remaining cells (Neg) for expression of macrophage and fibroblast markers by quantitative realtime RT-PCR analysis. **B** Definition and fold enrichment in P1 compared to P14 cardiac macrophages is presented for the most robustly regulated genes of known function (cutoff value is 2.0). Bold indicates soluble factors.
Supplemental Figure 5
Cardiomyocyte proliferation in monocyte/macrophage depleted neonates. (A) Heart weight/body weight (HW/BW) ratios in control and Mo/MΦ depleted mice measured at 7 and 21 days following MI at P1. (n=4 Sham and 10 MI/group). Data are mean ± SEM. All comparisons were not significant. (B) Representative images of pH3 (green) and troponin T (red) co-immunostaining of sections from control and Cl2MDP-L treated hearts at day 7 after MI to assess cardiomyocyte proliferation. (C) Quantification of serial sections beginning at the ligature and progressing toward the apex in 100µm intervals (3-4 sections per heart). (n=3 mice/group and is representative of 3 independent experiments) Scale bar: 100µm. Data are mean ± SEM. ns=not significant.