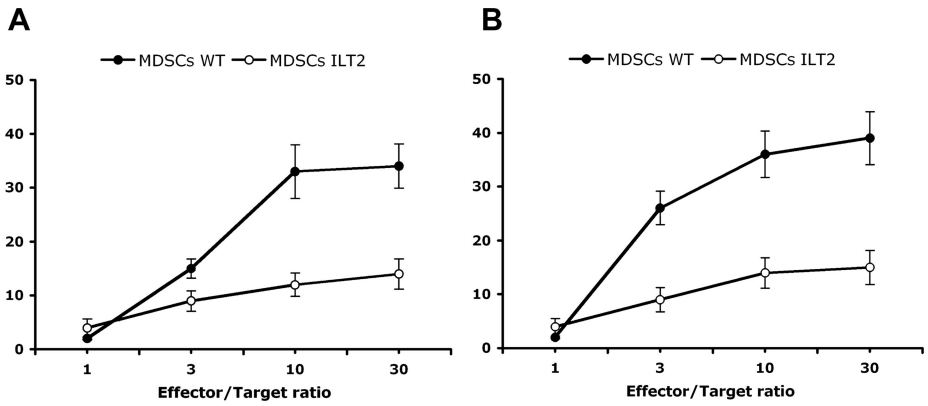
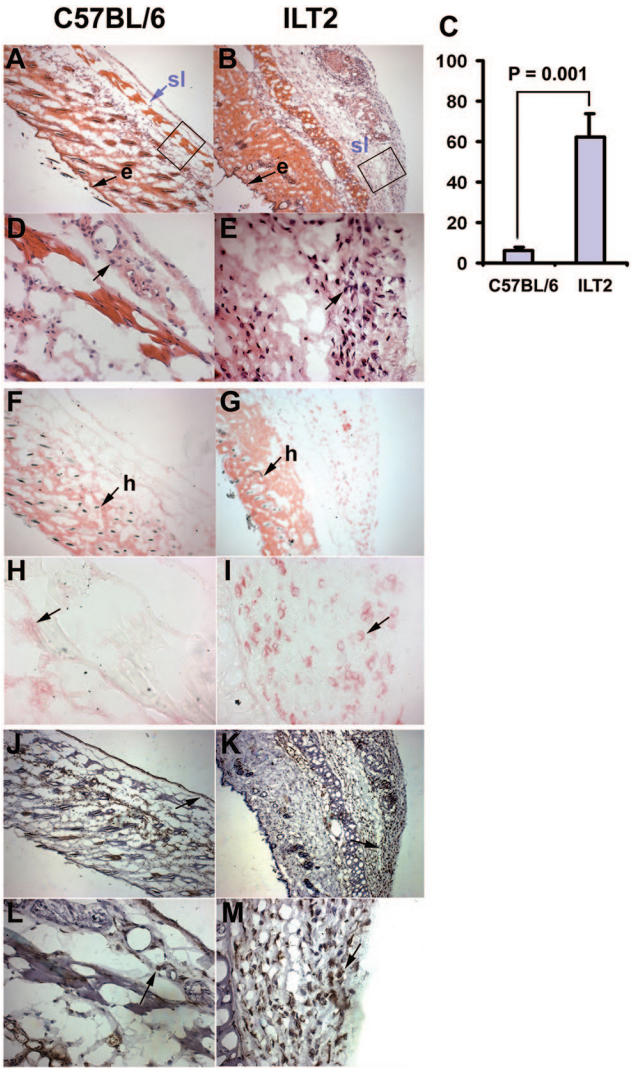


SUPPLEMENTARY FIGURE 1. Adoptive transfer of MDSCs from ILT2 mice decreases cytotoxic T lymphocyte (CTL) responses in recipients of MHC class II-disparate (bm12) allogeneic skin transplant. Splenocytes from WT mice that received MDSCs from ILT2 (š) or wild-type (~) mice and were transplanted with skin from bm12 mice were analyzed for CTL activity on days 13 (A) and 20 (B). T-cell blasts from donor mice were used as targets in ^{51}Cr release assay. Cytotoxic activity was assessed as described in Materials and Methods section. Error bars indicate SE of triplicate. Data shown are representative of three separate experiments.



SUPPLEMENTARY FIGURE 2. $CD11b^+/Gr\text{-}1^+$ myeloid cells from ILT2 mice are recruited to the site of the allogeneic skin graft. Skin graft histology and immunohistochemistry analysis. Adoptive transfers of MDSCs from C57BL/6 mice or ILT2 mice have been performed on recipient C57BL/6 mice grafted with allogeneic skin from bm12 mice as described in Materials and Methods section. Allogeneic bm12 skin was harvested on day 5 after transplantation. For immunohistochemical staining, frozen skin sections were fixed in ice-cold acetone and incubated with a rat anti-CD11b (M1/70) monoclonal antibody (F, G, H, I), or rat biotin-conjugated anti-Gr-1 (RB6–8C5) monoclonal antibody (J, K, L, M). For staining with primary anti-CD11b mAb, a biotinylated secondary antibody (donkey anti-rat IgG) was applied, followed by incubation with streptavidin-conjugated AP (Sigma). Alkaline phosphatase activity was localized with Fast Red Tablets (Roche Applied Science). For staining with biotin-conjugated anti-Gr-1 primary mAb, a streptavidine-conjugated HRP (BioGenex) was applied. Peroxidase activity was localized with diaminobenzidine (Vectastain ABC kit, Vector Laboratories). The epidermal layer (e), the subcutaneous layer (sl), and the hair follicles (h) are indicated. An accumulation of cells can be seen in the skin graft site of mice with adoptive transfers of MDSCs from ILT2 mice (B, H&E, original magnification, 10×; and E, H&E, original magnification, 40×) compared with the mice with adoptive transfers of MDSCs from C57BL/6 mice (A, H&E, original magnification, 10×; and D, H&E, original magnification, 40×). Quantitative data for the presence of MDSCs in the site of the graft is shown in Supplementary Figure 2C. Seven areas of site of the skin graft from three mice in each group were analyzed. Immunohistochemical analyses confirmed that cells are CD11b-positive (panels F and G, original magnification, 10×; and panels H and I, original magnification, 40×). In addition, these cells are positive for the Gr-1 marker as shown in panels J and K, original magnification, 10×; and panels L and M original magnification, 40×).