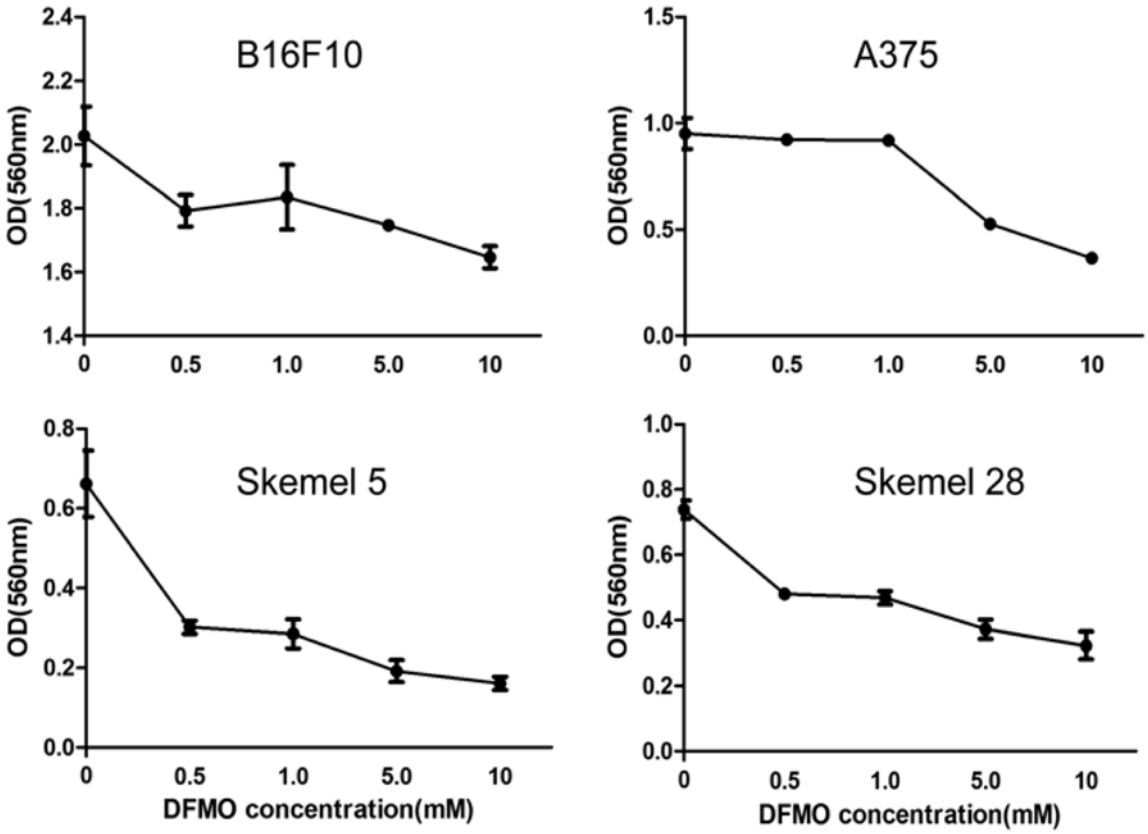
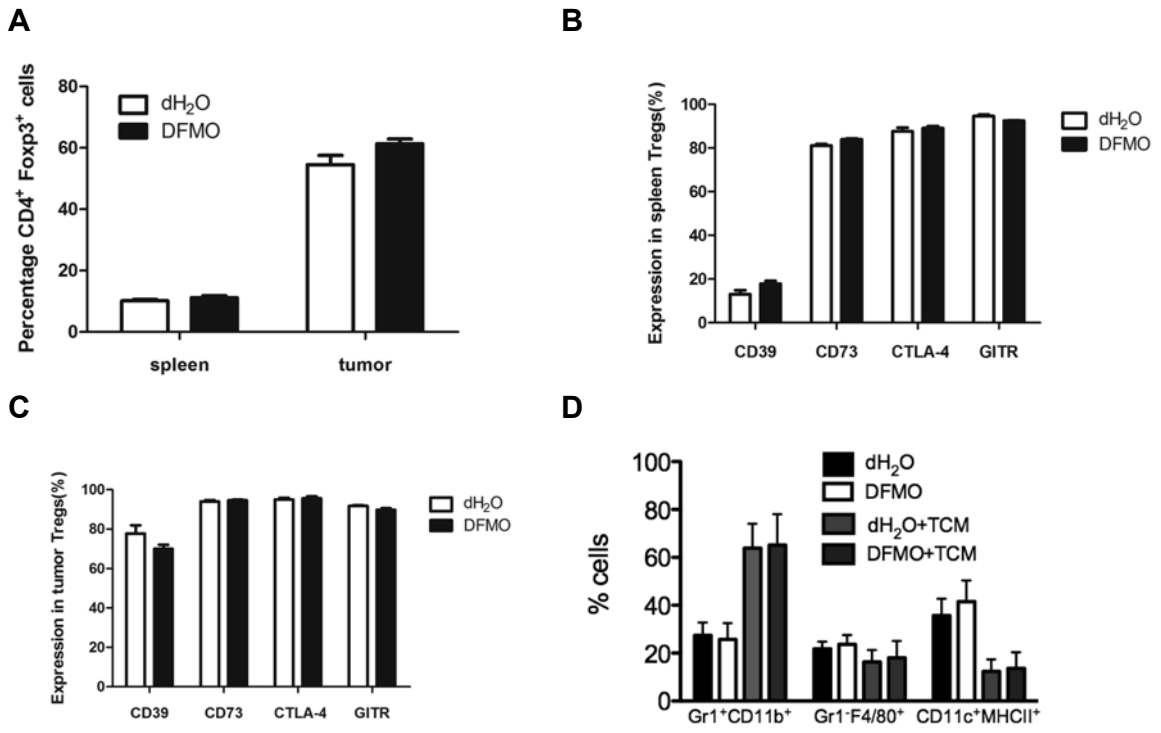
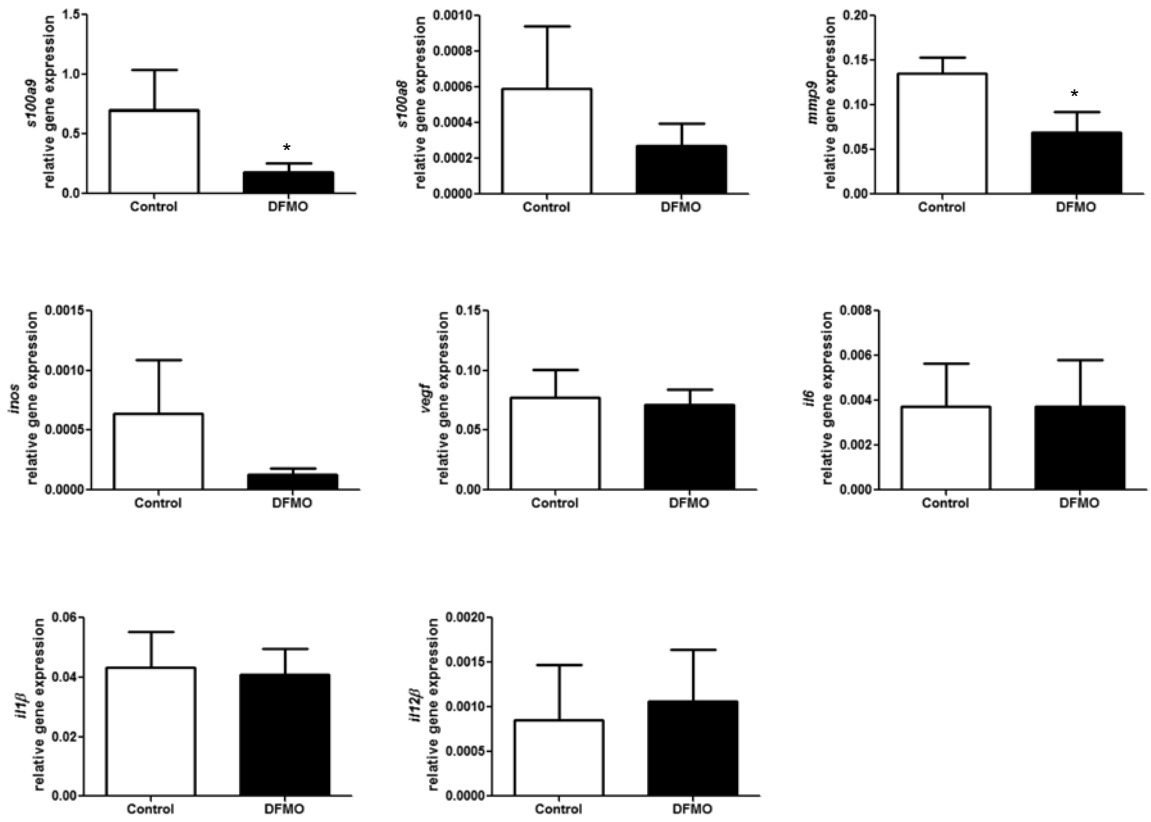


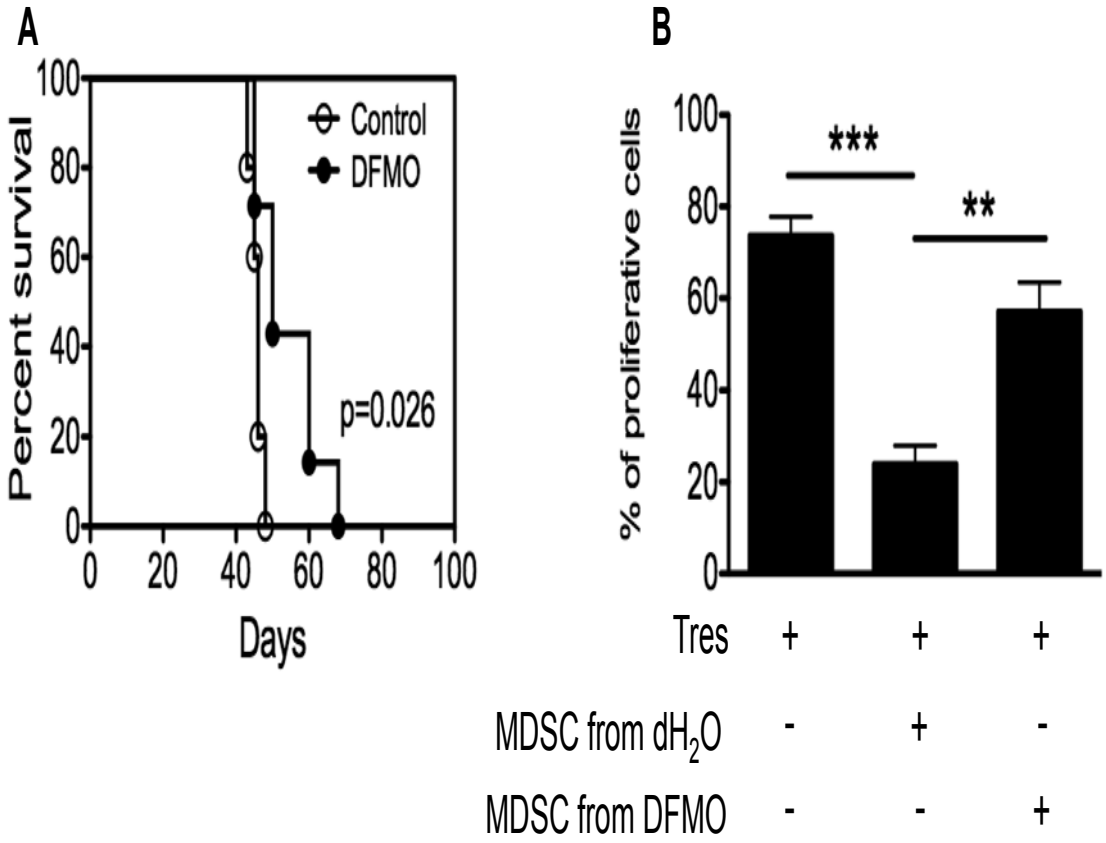
Supplemental Figure 1. *In vitro* growth inhibition of melanoma cells by DFMO. Murine B16F10, human A375, Skemel 5 and Skemel 28 melanoma cells were incubated for 3-5 days in culture with DFMO at the indicated concentrations. Cell growth was determined by the MTT assay from triplicate wells. Data shown are mean ± SD.



Supplemental Figure 2. The effect of DFMO on tumor-associated CD4$^+$Foxp3$^+$ regulatory T cells (Tregs) and MDSC differentiation. (A) Percent CD4$^+$Foxp3$^+$ T cells in spleen and tumor infiltrates. Percent CD39$^+$, CD73$^+$, CTLA-4$^+$ and GITR$^+$ cells among Tregs in spleen (**B**) and tumor infiltrates (**C**). Cells were collected from B16F10-bearing DFMO treated or control mice 14 days after tumor inoculation (3 mice per group). (**D**) BM cells were cultured with GM-CSF and IL-4 for 5 days in complete culture medium or in the TCM in the presence of DFMO or dH_2O as controls. The cell phenotypes were examined by flow cytometry. Data (mean ± SEM) are representative of 2 independent experiments.



Supplemental Figure 3. Altered gene expression in MDSCs following DFMO treatment. Bone marrow cells were cultured with GM-CSF and IL-6 in the presence or absence (as control) of DFMO at 10 mM for 4 d, and $Gr1^{+}CD11b^{+}$ MDSCs were selected using anti-CD11b magnetic beads. Expression levels of a series of indicated genes associated with MDSC activity were measured by real-time quantitative RT-PCR. GAPDH expression was measured for normalization. One representative of two experiments is shown.



Supplemental Figure 4. DFMO treatment increases the survival of ID8-bearing mice. (**A**) ID8-OVA cells (10^7) were injected i.p. into C57BL/6 mice (8 mice per group). DFMO was administered as a 1% solution in drinking dH_2O to mice starting 7 days after tumor injection. Mice fed with dH_2O without DFMO were used as controls. Mice survival was monitored every 3 days. (**B**) Suppressive activity of MDSCs as shown by quantification of eFluor450-labeled $CD4^+$ T responder cells (Tres) cocultured with the indicated $Gr1^+CD11b^+$ MDSCs from DFMO-treated tumor-bearing mice versus control mice. The ratio of T cell/MDSC was 2:1. **, $p<0.01$; ***, $p<0.001$ (3 mice per group).