Figure S2

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MHCII

Figure. S2. Enhanced maturation of Runx3 KO BMDC and similar expression of TLR2 and TLR4 in WT and KO DC. (A) Day 11 BMDC from cultures not treated with LPS were gated as high forward scatter/CD11c⁺ cells (R2) and assessed for expression of CD80 and MHC II. Bold lines, Runx3 KO; thin lines, WT littermates. Enhanced spontaneous maturation of the KO DC is evidenced by a larger proportion of cells with high CD80 and MHC II. (B) Day 7 WT and KO BMDC were grown in the presence of TGF- β (10 ng/ml) and treated (bold line) or untreated (broken line) for 4h with LPS (1µg/ml) prior to FACS analysis with anti CD11c and anti MHCII. The ratio of mature MHCII^{high} (M2) DC to immature MHCII^{low} (M1) is indicated above the histogram. Note that TGF- β inhibited maturation of WT BMDC as early as 4h after LPS treatment, but failed to do so in the KO BMDC (C) Expression of TLR2 (left) and TLR4 (right). TLR2, isolated WT and KO splenocytes were stained with anti CD11c and anti TLR2 (PE conjugated, clone 6C2, 12-9021, eBioscience) and analyzed by FACS. CD11c⁺ DC were gated and level of TLR2 expression was measured. TLR4, day 7 WT and KO BMDC were treated for 16h with LPS (1µg/ml) before RNA extraction and RT-PCR analysis as described in Figure 3D. TLR4 was determined 25 at and 27 cycles with the primers, F: CCTGCATAGAGGTAGTTCCTA; R: TAAGCCATGCCATGCCTTG yielding a 220bp fragment.