Supplemental Materials

Antibodies.

The following anti-mouse monoclonal antibodies (mAb), were obtained from eBioscience (San Diego, CA): anti-FcR (CD16/CD32) (93), FITC-anti-CD3e (145-2C11), FITC-CD11b (M1/70), FITC-anti-CD86 (GL-1), FITC-anti-IFN-γ (XMG1.2), PE-anti-CD8a (53-6.7), PE-anti-CD25 (PC61.5), PE-anti-CD40 (1C10), PE-anti-CD44 (IM7), PE-anti-CD80 (16-10A1), PE-anti-CD107a (eBio1D4B), PE-anti-IL-4 (11B11), PE-anti-Gr-1 (RB6-8C5), PE-anti-GITR (DTA-1), PerCP-anti-CD4 (RM4-5), APC-anti-F4/80 (BM8), APC-anti-FoxP3 (FJK-16s), APC-anti-CD4 (RM4-5), APC-anti-CD62L (MEL-14), and APC-anti-IL-17 (ebio17B7).

The following anti-mouse mAb with a fluorescent conjugate were obtained from BioLegend (San Diego, CA): Alexa488-anti-CCR7 (4B12), FITC-anti-DX5 (DX5), PE/Cy7-anti-CD86 (GL-1), PerCP-anti-CD8 (53-6.7), PerCP-anti-CCR7 (4B12), APC-anti-CD3e (145-2C11), APC-anti-CD11c (N418), and APC/Cy7-anti-CD3e (145-2C11).

The following anti-mouse monoclonal functional grade purified antibodies, were obtained from eBioscience (San Diego, CA): anti-CD3e (145-2C11), anti-CD28 (37.51), anti-IL-4 (11B11) and anti-IFN-γ (XMG1.2).
Reverse transcription polymerase chain reaction (RT-PCR) analysis.

Total RNA was isolated from WEHI3B and WGM cells cultured in vitro for 2 weeks using Sepasol-RNA II Super (Nacalai Tesque). RNA was used to synthesize cDNA using the SuperScript™ III First-Strand Synthesis System for RT-PCR (Invitrogen, CA), and polymerase chain reaction (PCR) amplification was performed using KOD Fx (TOYobo, Osaka, Japan). For BLT1, the PCR conditions were as follows for 35 cycles: denaturation at 94 °C for 30 s, annealing at 58 °C for 30 s, elongation at 72 °C for 30 s. For β-actin, the PCR was performed as follows for 35 cycles: denaturation at 94 °C for 30 s, annealing at 58 °C for 30 s, elongation at 72 °C for 30 s. The following primers were used for PCR: BLT1 FW, 5’-AGGCATCTGGGTGTCTT-3’; BLT1 RV, 5’-GCGAAGGGCACAGGATAATGAG-3’; β-actin Fw, 5’-ATGGATGACGATATCGCTGC-3’; β-actin RV, 5’-GTAACGAGGAGGACTCGCTGC-3’. The pcDNA3-FLAG-mBLT1 plasmid was used as a positive control for BLT1. PCR products were detected by electrophoresis on 2.0% tris/borate/ethylenediaminetetraacetic acid (EDTA) (TBE)–agarose gels and ethidium bromide staining.

Enzyme-linked immunosorbent assay
Tumor cells \((2 \times 10^5 \text{ cells/ml})\) were plated on a 6 well plate in 5 ml of the culture medium and incubated for 24 h. Culture supernatants were harvested and assessed for GM-CSF production. The amount of secreted GM-CSF was determined by ELISA using OptEIA™ Set Mouse GM-CSF kit (BD Bioscience) according to the manufacturer’s instruction.

**Multifunctionality assay**

On day 46 after the first tumor challenge, TDLNs or splenocytes were harvested from WT/WGM and KO/WGM mice \((n=3-5\text{ per group})\), respectively. Cells were homogenized as described in Methods, and cultured in CM containing 2-mercaptoethanol \((50 \text{ nM})\) for 4 d, and then PMA \((10 \text{ ng/mL})\), ionophore A23187 \((250 \text{ mg/mL})\), and brefeldin A \((1 \text{ ng/mL})\) was added for 5 h. After washing, cells were pretreated with FeR block followed by staining with PerCP-anti-mouse CD4 mAb for 30 min. Subsequently, obtained cells were fixed with 2% paraformaldehyde and then stained intracellularly with FITC-anti-IFN-γ, PE-anti-IL-2, and APC-anti-TNF-α mAb in permeabilization buffer (eBioscience) for 30 min on ice. Stained cells were washed three times in FAB and analyzed by FACS Calibur and analyzed using the FlowJo software.
**Figure S1**

*In vitro* sufficient production of mouse GM-CSF from GM-CSF transduced tumor cells.

Mouse GM-CSF production levels from supernatants of $1 \times 10^6$ WEHI3B or WGM cells cultured *in vitro* for 24 hours were measured by ELISA.
Undetectable BLT1 mRNA expression on both WEHI3B and WGM cells.

Expression levels of mouse BLT1 mRNA in both WEHI3B and WGM cells were examined by RT-PCR analysis. RT(-) represents samples where reverse transcriptase was not included in the assay as controls. Representative data from three independent experiments with similar results are shown.
Positive effect of the loss of LTB1/LTB4 axis on expression levels of various maturation markers of DCs residing in TDLNs.

Two left axillary TDLNs were harvested from WT/WGM or KO/WGM mice on day 2 (CD80, CD86 and CCR7) or day 4 (CD40) after the FTC (n=3-5 per group). Shown are representative histogram plots that depict an MFI difference of CD40, CD80, CD86, and CCR7 in TDLNs-derived CD11c⁺ DCs between WT/WGM (open histograms with thin line) and KO/WGM mice (open histograms with bold line). Shaded histograms depict isotype control. The plots are gated on CD11c⁺ DCs.
Multifunctionality of CD4\(^+\) T cells in TDLNs.

Responses in WT/WGM and KO/WGM mice are color-coded according to the number of acquired functions (IFN-\(\gamma\), IL-2, TNF-\(\alpha\)) and summarized in the pie chart, where each wedge represents the frequencies of CD4\(^+\) T cells expressing following 3 cytokines such as IFN-\(\gamma\)^IL-2\(^+\)TNF-\(\alpha\)^ (3), or either 2 cytokines such as IFN-\(\gamma\)^IL-2\(^-\)TNF-\(\alpha\)^, IFN-\(\gamma\)^IL-2\(^-\)TNF-\(\alpha\)^, and IFN-\(\gamma\)^IL-2\(^-\)TNF-\(\alpha\)^ (2), or either one cytokine such as IFN-\(\gamma\)^IL-2\(^-\)TNF-\(\alpha\)^, IFN-\(\gamma\)^IL-2\(^-\)TNF-\(\alpha\)^, and IFN-\(\gamma\)^IL-2\(^-\)TNF-\(\alpha\)^ (1), or no cytokine (0). Results are representative of three independent experiments.