

Supplementary information, Figure S4. LKB1 signaling in DCs controls Th17 cell differentiation, in part, by shaping IL-6-STAT3 signaling. a, Real-time PCR analysis of Stk11 mRNA expression in CD4<sup>+</sup>CD44<sup>hi</sup>CCR6<sup>+</sup> Th17 cells from WT and LKB1<sup>△DC</sup> mice. **b**, Analysis of *Ifng* and *Il17* mRNA expression in popliteal draining lymph node (DLN) and splenic cells from KLH immunized WT and LKB1<sup>ΔDC</sup> mice after ex vivo stimulation with KLH for 48 h. c, Real-time PCR analysis of 116 and Tgfb2 mRNA expression in splenic  $CD8\alpha^+$  cDCs and  $CD8\alpha^-$  cDCs from WT and LKB1<sup>ΔDC</sup> mice. d, Cytokine analysis in the supernatant from naïve OT-II CD4<sup>+</sup> T cells co-cultured with WT or LKB1-deficient splenic DCs for 3 days. e, Relative thymidine incorporation of WT Tregs co-cultured with splenic DCs from WT and LKB1<sup>ΔDC</sup> mice in the presence or absence of anti-TGFB (values cultured with WT thymic DCs without antibody treatment was set as 1). f, Flow cytometry analysis of splenic Foxp3<sup>+</sup>CD4<sup>+</sup> Tregs from WT, LKB1<sup>ΔDC</sup>, *Il6<sup>-/-</sup>*, or LKB1<sup>ΔDC</sup>*Il6<sup>-/-</sup>* mice. g, Flow cytometry analysis (left) and statistics (right) of IL-17 expression in naïve OT-II CD4<sup>+</sup> T cells cultured with splenic DCs from WT, LKB1<sup> $\Delta DC$ </sup>, *Il6*<sup>-/-</sup>, or LKB1<sup> $\Delta DC$ </sup>*Il6*<sup>-/-</sup> mice for 5 days. **h**, Tumor growth curve in WT (*n* = 28), LKB1<sup> $\Delta DC$ </sup> (n = 21), Il6<sup>-/-</sup> (n = 5), or LKB1<sup> $\Delta DC$ </sup>Il6<sup>-/-</sup> (n = 19) mice following inoculation of MC38 tumor cells. i, WT and LKB1<sup>ΔDC</sup> mice were inoculated with MC38 tumor cells and treated with or without 1 mg/kg/d Stat3 inhibitor JSI-124 for 13 days started from day 4 for tumor growth curve analysis (n = 5, JSI-124-treated WT; n = 6 for other groups). Statistics was shown for LKB1<sup> $\Delta DC$ </sup>-vehicle group vs LKB1<sup> $\Delta DC$ </sup>-JSI-124 group. Data in plots indicate the means  $\pm$  s.e.m; each symbol represents an individual mouse. Numbers in gates indicate percentage of cells. NS, not significant; \*P < 0.05, \*\*P < 0.01, \*\*\*P < 0.001; \*\*\*\*P < 0.0001; two-tailed unpaired Student's t test (b-e) or two-way ANOVA (h, i). Data are from two (a-d, f, g, i) or three (e, h) independent experiments.