

## Supplemental material

## Yu et al., https://doi.org/10.1084/jem.20180397



Figure S1. **Expressions of T reg cell related genes in** *Traf3ip3*<sup>+/+</sup>*Foxp3*-Cre and *Traf3ip3*<sup>fl/fl</sup>*Foxp3*-Cre T reg cells. Splenic T reg cells from 8-wk-old *Traf3ip3*<sup>+/+</sup>*Foxp3*-Cre and *Traf3ip3*<sup>fl/fl</sup>*Foxp3*-Cre mice were isolated for quantitative RT-PCR analysis. Data are representative of at least three independent experiments and are presented as mean ± SEM. ns, not statistically significant; \*\*, P < 0.01. Student's *t* test.



Figure S2. **Cytokine expression of gut T reg cells.** Flow cytometric analysis of IFN- $\gamma$  and IL-17 expression in gut T reg cells from 8-wk-old *Traf3ip3*<sup>+/+</sup> *Foxp3*-Cre and *Traf3ip3*<sup>fl/fl</sup>*Foxp3*-Cre mice stimulated for 24 h with antibodies against CD3 and CD28. Data are representative of three independent experiments and are presented as mean ± SEM. \*\*, P < 0.01. Student's t test.



Figure S3. **Expressions of metabolism related genes in** *Traf3ip3<sup>+/+</sup> Foxp3-Cre* and *Traf3ip3<sup>fl/R</sup> Foxp3-Cre* **T** reg cells. Splenic *Traf3ip3<sup>+/+</sup> Foxp3-Cre* and *Traf3ip3<sup>fl/R</sup> Foxp3-Cre* **T** reg cells were stimulated for 0 (unstimulated; UT) or 24 h (stimulated; ST) with antibodies against CD3 and CD28 following quantitative RT-PCR analysis of glucose transporter GLUT1 (murine gene name *Slc2a1*), glycolytic enzyme hexokinase 2 and metabolic master regulator HIF1- $\alpha$  expression. Data are representative of three independent experiments and are presented as mean ± SEM. \*, P < 0.01; \*\*, P < 0.01. Student's *t* test.