SUPPLEMENTAL FIGURE LEGENDS

Supplemental Figure 1. Teff cells exhibit increased glucose uptake compared to Treg cells and selective accumulation in Glut1 transgenic mice *in vivo*. A-C. Wild-type $CD4^+$ T cells were cultured in conditions to enrich Teff (Th1, Th2, Th17) and Treg subsets. (A) Representative FACS staining for intracellular cytokine production and expression of FoxP3. (B-C) Glucose uptake in Teff, Treg, and naive $CD4^+$ T cell cultures. **D-E.** Immunoblot of (D) Glut1 expression and (E) glucose uptake in unstimulated $CD4^+$ T cells from Glut1- and non-transgenic littermates. **F.** $CD4^+$ $CD44^{high}$ T cells were enumerated by FACS analysis in the spleen, lymph node, and liver of greater than on year aged Glut1- and non-transgenic littermates (Aged) and 6-8 week old (young) wild-type mice. **G.** The presence of anti-dsDNA antibodies was detected by ELISA from greater than on year aged Glut1- and non-transgenic littermates (Aged) and 6-8 week old (young) wild-type mice. **H-I.** Representative FACS plots of $CD4^+$ T cells from greater than 1 year aged transgenic and non-transgenic mice stimulated overnight to determine the percentage of Th1, Th2, and Th17 cells as measured by intracellular cytokine staining for IFNγ, IL-4, and IL-17 respectively. The percentage of Treg cells in the (H) spleen and (I) thymus was assessed directly *ex vivo* by intracellular staining with anti-FoxP3. **J.** Wild-type $CD4^+$ T cells were stimulated in conditions to enrich for Treg and Th17 differentiation in the presence or absence of 5mM glucose followed by intracellular cytokine and FoxP3 staining. All results are representative of three independent experiments (n

of greater than 5 mice per group) and graphs are displayed as the average and standard deviation with *P* values determined using the Student's t test (*$P \leq 0.05$).

Supplemental Figure 2. Lipid oxidation and AMPK activation promote Treg generation in vitro and in a murine model of asthma. A. $CD4^+$ T cells were cultured to generate Th17 cells in the presence or absence of etomoxir and differentiation was assessed by intracellular cytokine staining. **B-C.** T helper cell subsets were generated *in vitro* as described, washed, and replated an additional two days in the presence or absence of FA. (C) T helper cell subsets were generated *in vitro* in the presence or absence of FA, washed, and replated an additional two days in the indicated cytokines and blocking antibodies without additional FA. Differentiation was assessed by intracellular cytokine and FoxP3 staining. **D.** Immunoblot of $CD4^+$ T cells untreated (PBS) or treated with metformin (Met) for 24 hours. **E.** Naïve $CD4^+$ T cells were activated with anti-CD3 + anti-CD28 in the presence or absence of 25nM Rapamycin (Rapa), 100μM metformin (Met), or 200μM Etomoxir (Etx) and palmitate oxidation was assessed after 24 hours. **F.** Mice were sensitized to ovalbumin in the presence of Met or a PBS followed by aerosol challenge 21 days later and the bronchial alveolar lavage (BAL) fluid was analyzed to quantitate the indicated cell populations. **G.** Model depicting Teff and Treg differentiation is partially regulated through mTOR driven glycolysis and AMPK induced fatty acid oxidation respectively. Results are representative of two

independent experiments and graphs are displayed as the average and standard deviation with *P* values determined using the Student's t test (*P ≤ 0.05).

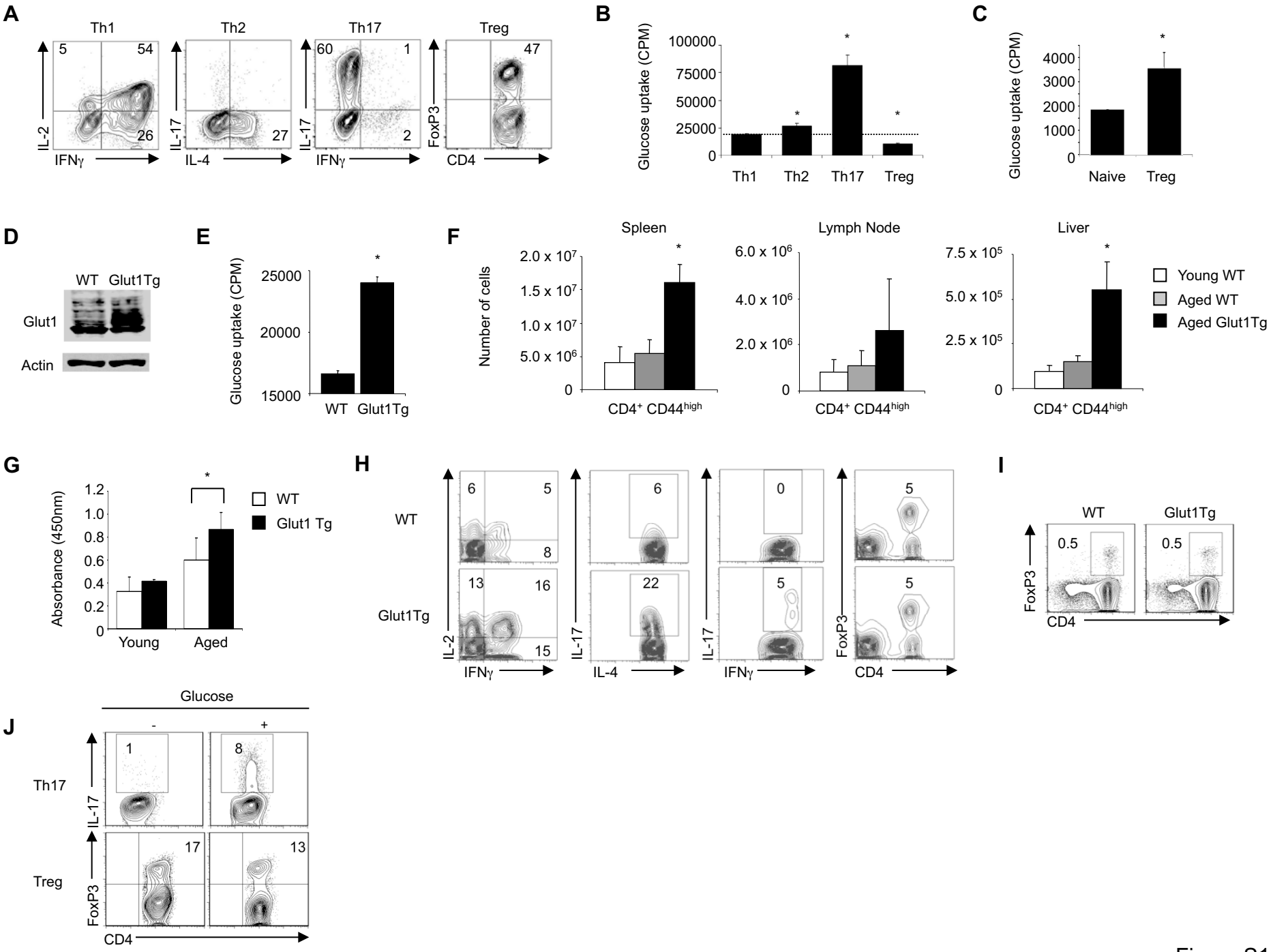

Figure S1

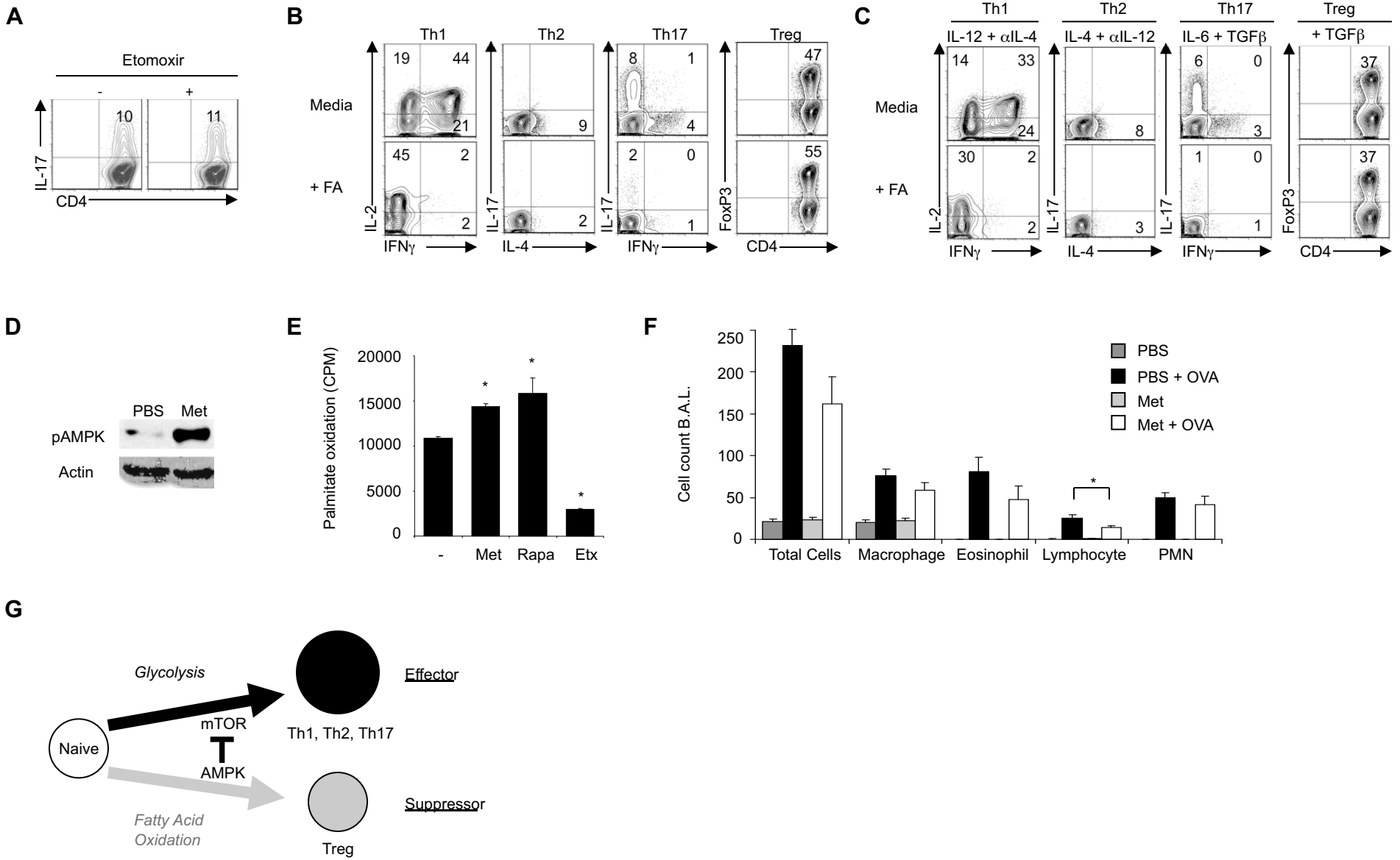

Figure S2