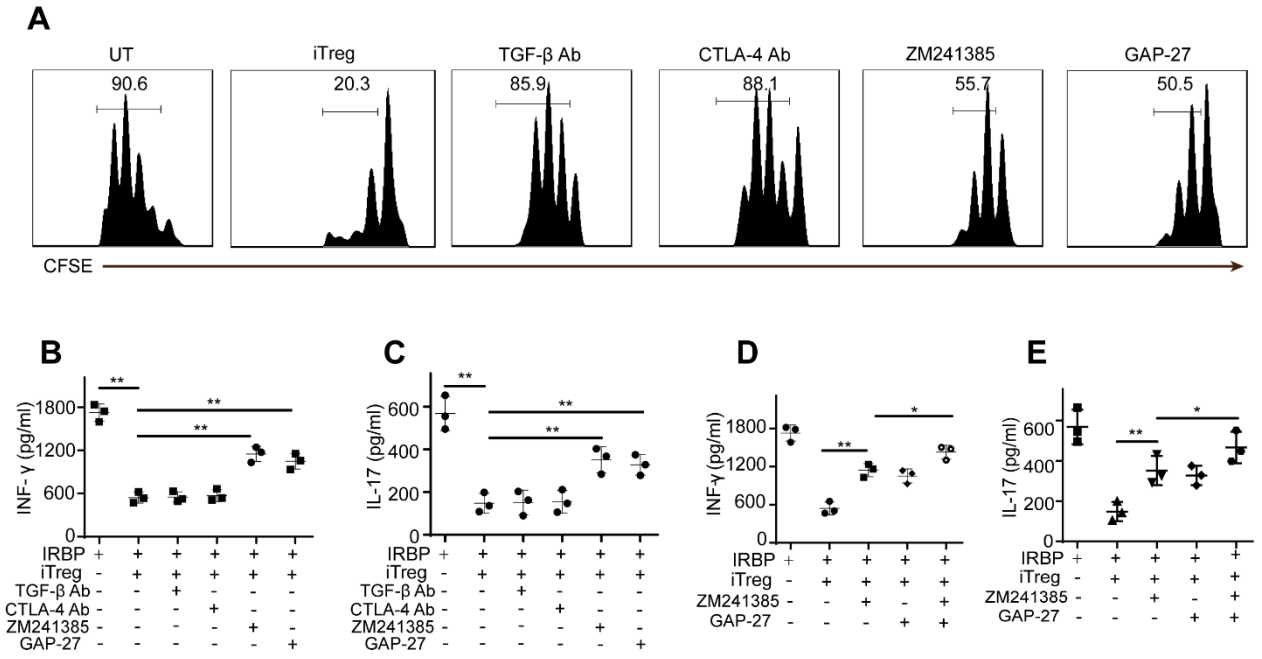

Figure S1. The gap junction inhibitor and A2AR inhibitor reversed iTregs-mediated inhibition on UTs obtained from EAU mice. (A-C): Both A2AR antagonists (ZM241385) and gap junctions inhibitor (GAP-27), significantly reversed the iTregs–mediated inhibitory effect on UTs proliferation and inflammatory cytokines production. (D-E): The combination of A2AR antagonists and gap junction inhibitor was more effective compared with either alone in reversing the iTregs–mediated inhibitory effect on UTs inflammatory cytokines production. The results were representative of three independent experiments. The data are presented as the means ± SDs. * P <0.05; **: P <0.01. (Between the indicated groups). Abbreviations: TGF-β Ab, TGF-β antibody; CTLA-4 Ab, CTLA-4 antibody. Data were analyzed using one-way ANOVA with Bonferroni correction.

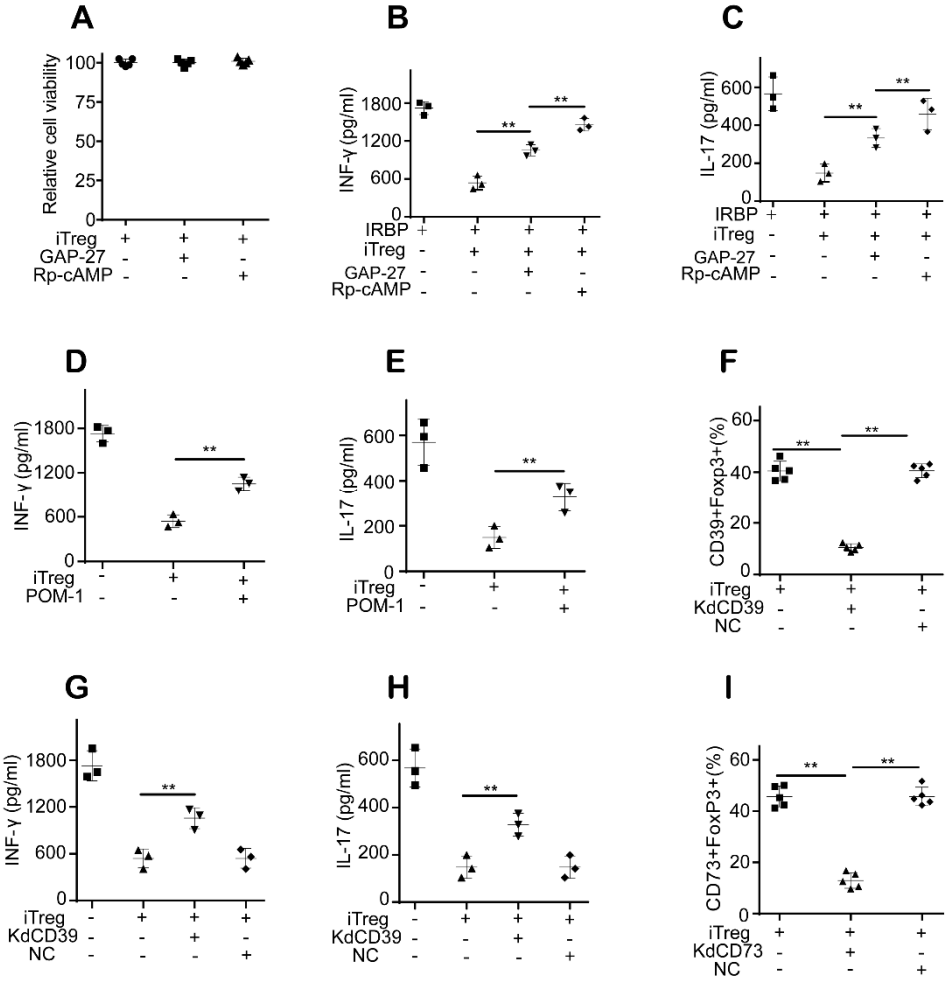

Figure S2. Intracellular cAMP and CD39 expression of iTregs is essential for iTregs-mediated inhibition on inflammatory cytokines production of UTs. (A): Cell viability was assessed using Trypan blue exclusion and examined using phase contrast microscopy. (B-C): Pretreatment of iTregs by Rp-cAMPs was more effective than gap junction inhibitor in reversing iTregs-mediated suppression on inflammatory cytokines production of UTs. (D-E): POM-1 pretreatment partially reversed iTregs-mediated suppression on inflammatory cytokines production of UTs. (F): CD39 siRNA but not control siRNA decreased CD39 frequency in iTregs. (G-H): CD39 siRNA but not control siRNA knockdown in iTregs also decreased the inhibitory capability of iTregs on inflammatory cytokines production of UTs. (I): CD73 siRNA but not control siRNA decreased CD73 frequency in iTregs. The results were representative of three independent experiments. The data are presented as the means ± SDs. **: P <0.01. (Between the indicated groups). Data were analyzed using one-way ANOVA with Bonferroni correction.

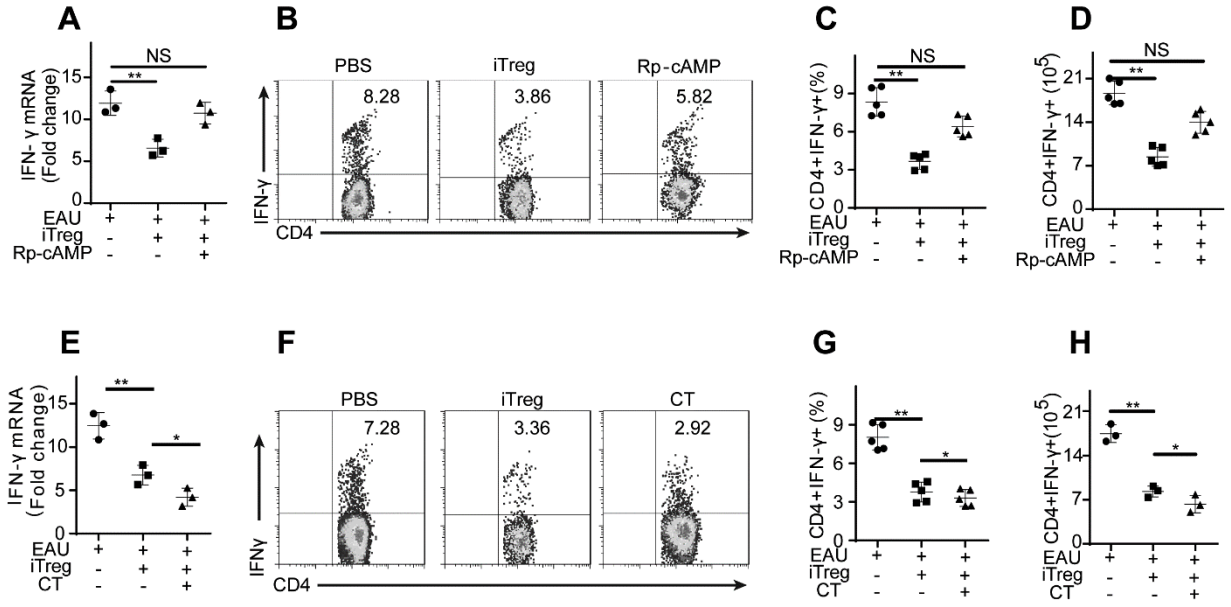

Figure S3. cAMP is critical for iTregs-mediated inhibition on IFN-γ production in established EAU. (A-D): iTregs preincubation with Rp-cAMPs also significantly decreased their inhibitory capability with regard to IFN-γ expression (n=3) and the frequency and number of CD4+ IFN-γ+ T cells in mice with EAU (n = 5). (E-H): CT pretreatment also improved iTregs' capability in inhibiting IFN-γ expression (n=3) and the frequency and number of CD4+ IFN-γ+ T cells in EAU mice (n = 5). The results were representative of three independent experiments. The data are presented as the means ± SDs. NS: $P > 0.05$; *: $P < 0.05$; **$P < 0.01$. (Between the indicated groups). Data were analyzed using one-way ANOVA with Bonferroni correction.