1 Supplementary Materials

Figure S1

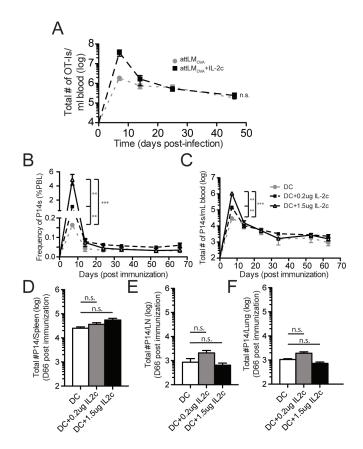


Figure S1. $3x10^4$ Thy1.1⁺ WT OT-I cells were adoptively transferred into naïve Thy1.2⁺ mice and infected with $5x10^7$ CFU attenuated LM-OVA₂₅₇ with or without IL-2c treatment from D4-6 post-infection. (A) Total number OT-I cells quantified per mL of blood across 46 days. (B) $3x10^4$ Thy1.1⁺ TCR transgenic P14 cells were adoptively transferred into naïve Thy1.2⁺ B6 mice and subsequently immunized with $5x10^5$ DC-gp33 followed by Rat Ig, $0.2\mu g$ or $1.5\mu g$ IL-2c on D4-6 post-DC immunization. Frequency of P14 cells quantified per mL of blood longitudinally across 66 days in all treatment groups. (C) Same as (B) except total number of P14 cells quantified per mL of blood. (D) Summary bar graph (mean \pm SEM) of total number of P14 cells harvested from the spleen at D66 post-DC immunization. (E) Same as (D) except total number of P14 cells in LN. (F) Same as (D) except total number of P14 cells in Lung. Data are representative of two experiments with at least n=5 mice/group/experiment. * = p<0.05; *** = p<0.005; *** = p<0.005; ns, not significant.

Figure S2

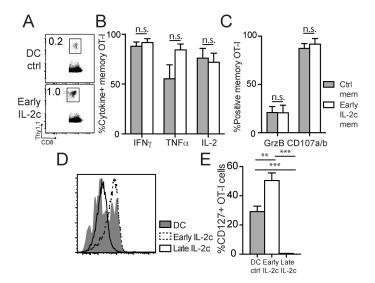


Figure S2. $3x10^4$ Thy1.1⁺ WT OT-I cells were adoptively transferred into naïve Thy1.2⁺ mice and immunized with $5x10^5$ DC-Ova followed by Rat Ig or IL-2c on D1-3 (Early) post-DC immunization. Spleens were harvested at D70 and splenocytes were stimulated with Ova₂₅₇ peptide for 5 hrs in the presence of Bfa. (A) Representative gating of memory OT-I cells. (B) Percent of memory OT-I cells expressing IFNγ, TNFα, or IL-2. (C) Same as (B) except GrzB and CD107a/b expression. (D) Same adoptive transfer setup as (A-C) with additional D4-6 (late) IL-2c treatment group. Expression of CD127 was measured on D7 post-DC immunization. Representative histogram depicting relative CD127 from OT-I cells among treatment groups. (E) Summary bar graphs (mean ± SEM) of %CD127+ OT-I cells among treatment groups. Data are representative of 2 independent experiments with at least n=5 mice/group. * = p<0.05; *** = p<0.0005.

35 Figure S3

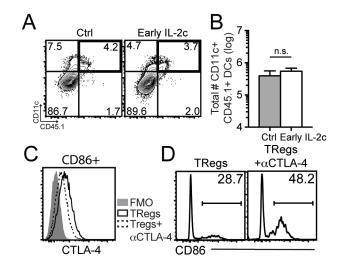


Figure S3. $3x10^6$ LPS-matured CD11c⁺ DCs from B16-flt3L inoculated CD45.1⁺ B6 mice were adoptively transferred into naïve CD45.2⁺ B6 recipients and subsequently administered Rat Ig or IL-2c for 2 days. Spleens were harvested and CD45.1+ DC population were quantified from both groups. (A) Representative flow plot depicting CD45.1+CD11c+ DC population. (B) Summary bar graph (mean \pm SEM) of total number of transferred DCs harvested from the spleen at 2 days post-immunization. (C) Treg cells were facs-sorted from FoxP3-GFP mice and incubated in a 1:2 ratio with LPS-matured DCs in the presence of 20nM bafilamycin A with or without 300µg α CTLA-4 for 3 hours. Representative histogram plot depicting CTLA-4 binding of CD86 ligand. (D) Representative histogram plots depicting CD86 expression on DCs in Treg and Treg+ α CTLA-4 treatment wells. Data are representative of two independent experiments with at least 3 biological replicates. n.s. = not significant.

51 Figure S4

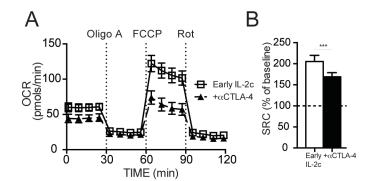


Figure S4. $3x10^4$ Thy1.1⁺ OT-I cells were adoptively transferred into naïve Thy1.2⁺ B6 mice and administered DC + IL-2c₁₋₃ or DC + IL-2c₁₋₃ + α CTLA-4 treatment. OT-I cells were purified from spleens at D6 post-DC immunization and assessed for metabolic function. (A) Time-course of oxygen consumption rate (OCR) in pmols/min from both treatment groups. (B) Same as (A) except summary (mean \pm SEM) of spare respiratory capacity (SRC) in both treatment groups normalized to percent of baseline. Data are representative of two independent experiments with at least 5 biological replicates/experiment. *** = p<0.0005.