

1 The immunosuppressive effect of the tick protein, Salp15,
2 is long-lasting and persists in a murine model of
3 hematopoietic transplant

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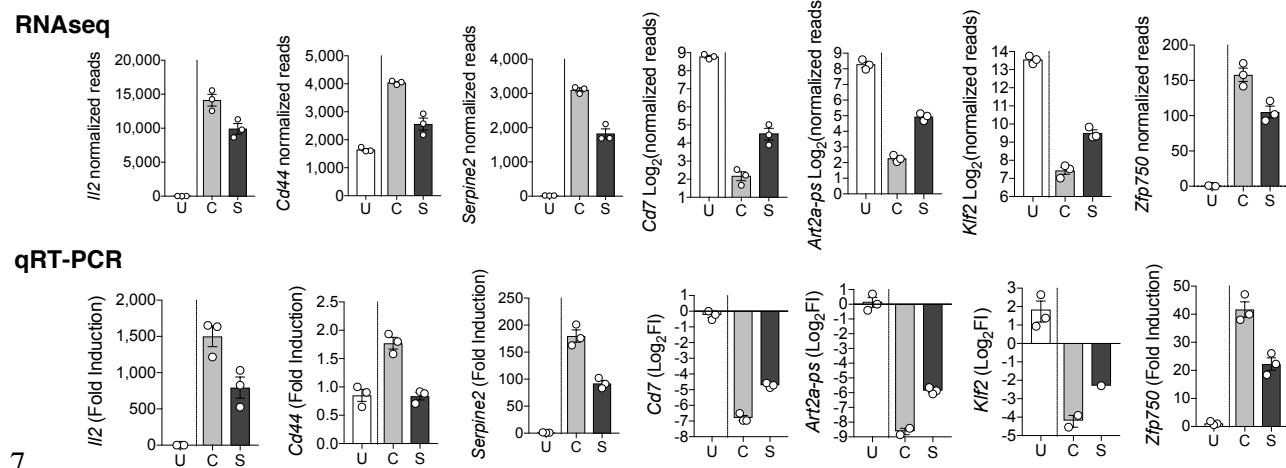
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1 **Supplementary Fig. S1. Validation of the RNAseq on selected genes.**

2 Normalized reads (top) and fold induction changes by qRT-PCR (bottom) of a
 3 group of selected genes differentially regulated in CD4 T cells activated with anti-
 4 CD3/CD28 for 2 days in the presence of Salp15 (S) or Salp15 Δ P11 (C). U: Non-
 5 stimulated CD4 T cells.

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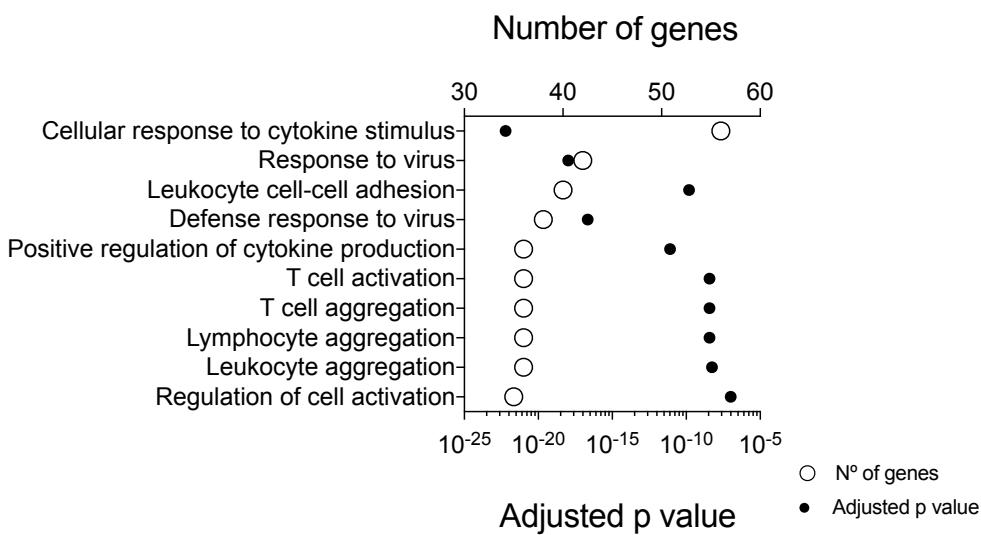
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1 **Supplementary Fig. S2. Gene Ontology analysis of genes affected by Salp15 at**
 2 **2 days.** Ten most over-represented Gene Ontology of Biological Processes (GOBP)

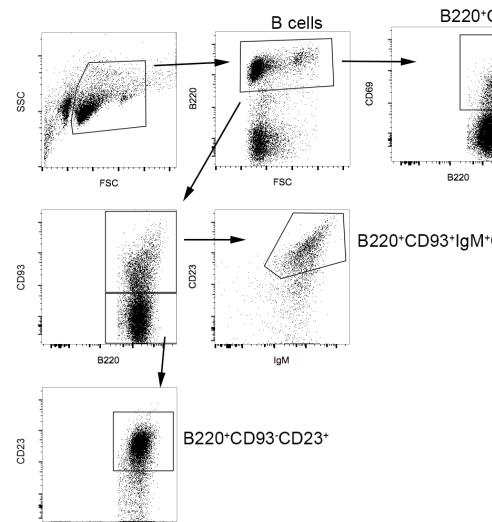
3 groups affected by Salp15 on activating CD4 T cells. The number of genes (white

4 circles) and the adjusted p values (black circles) are presented.

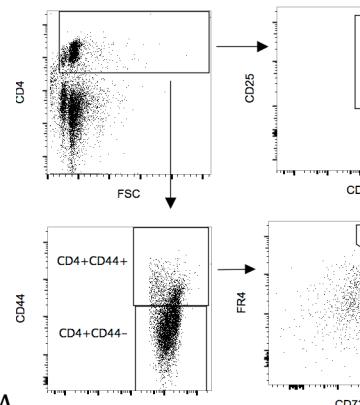
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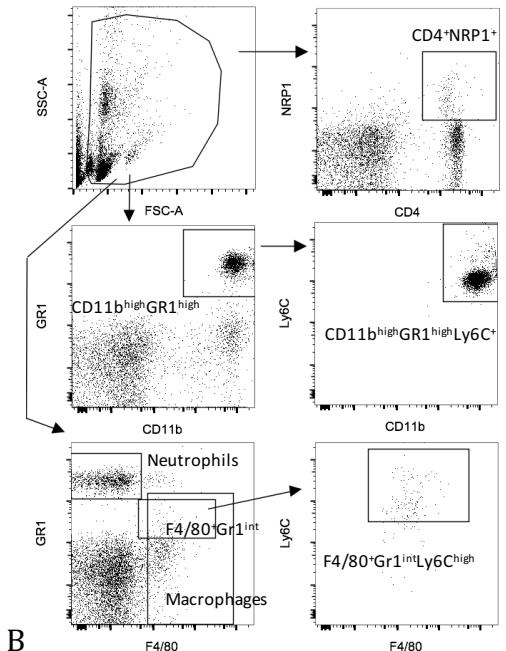
1 **Supplementary Fig. S3. Gating strategy to identify the populations in ovalbumin-immunized mice.** (A) B cell populations in the
 2 blood. (B) CD25 (top right blot), CD44 (bottom left blot) and anergic CD4 T cells (bottom right blot) in the spleen. (C) CD4+NRP1+
 3 (Tregs; top right blot) and myeloid populations in the spleen.



A



CD4+CD44highCD73highFR4high



B

C

4

1 **Supplementary Fig. S4. Salp15 does not affect Th1 or Th2 differentiation in**
 2 **immunized mice.** (A) Heatmap representing the genes associated with CD4 T cell
 3 differentiation according to the transcriptomic analysis in the presence of Salp15
 4 (S) or Salp15 Δ P11 (C) at 2 and 4 days of activation. (B) Ovalbumin/KLH-
 5 immunized mice (see Materials an Methods) were sacrificed at day 21 and whole
 6 splenocytes (3×10^6 /ml) were restimulated with ovalbumin (Ova) or KLH. The
 7 restimulation supernatants were analyzed for IFN γ and IL-4 by capture ELISA. The
 8 levels of IFN γ were below the detection limit. No differences were observed
 9 between Salp15- or Salp15 Δ P11 (control)-treated, immunized mice ($p > 0.05$, 2-
 10 way ANOVA). The data represent 10 mice in each group immunized with
 11 ovalbumin and 5 per group with KLH and are representative of 2 independent
 12 experiments.

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