Dysbiosis of the Gut Microbiota Is Associated with HIV Disease Progression and Tryptophan Catabolism

Ivan Vujkovic-Cvijin,1,2* Richard M. Dunham,1* Shoko Iwai,3 Michael C. Maher,4 Rebecca G. Albright,1 Mara J. Broadhurst,1 Ryan D. Hernandez,4 Michael M. Lederman,5 Yong Huang,6 Ma Somsouk,1,3 Steven G. Deeks,7 Peter W. Hunt,7 Susan V. Lynch,3*† Joseph M. McCune1*†

Introduction

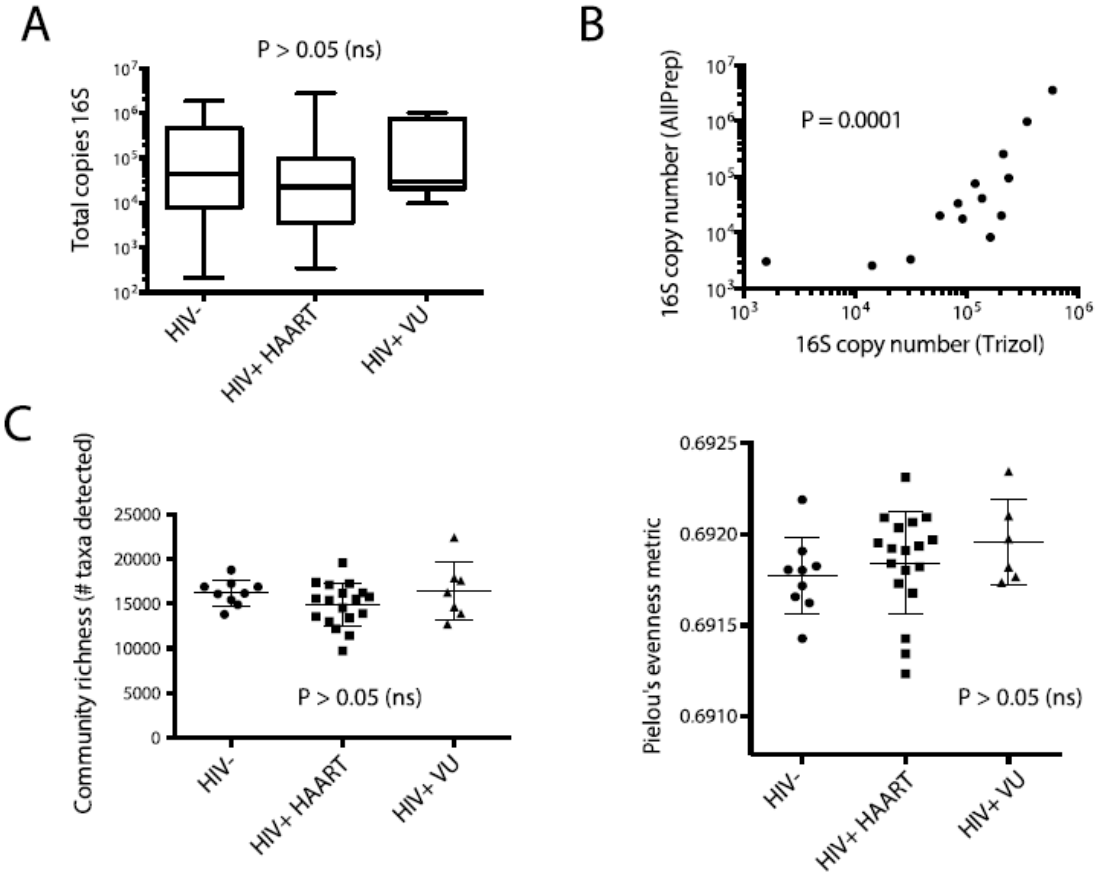
- Progression to AIDS during HIV infection is driven by chronically elevated T cell activation and chronic inflammation.
- Decreased gastrointestinal epithelial barrier integrity linked with accelerated disease.
- Increase in activation of the kynurenine pathway of tryptophan catabolism through IDO1 (indoleamin 2,3-dioxigenase 1) -> production of tryptophan catabolites that can inhibit differentiation of IL-17-secreting $CD4^+$ T cells.
- Highly active antiretroviral therapy (HAART) reduces viral load but often inflammation persists (IDO1, IL-6, interferon inducible protein 10 IP-10).

- **Is the gut microbiota altered during HIV infection?**
- **Are alterations in gut microbiota associated with HIV disease progression?**

Results

- Study cohort 34 male subjects
  - 6 viremic untreated (VU)
  - 18 virally suppressed (HAART) with var. $CD4^+$ T cell recovery
  - 1 HIV long-term non-progressor
  - 9 controls
- Analyzed: Rectosigmoid biopsis and peripheral blood
- Microbial profiling: 16s ribosomal RNA high-density microarray (G3 PhyloChip)

S 1



Results

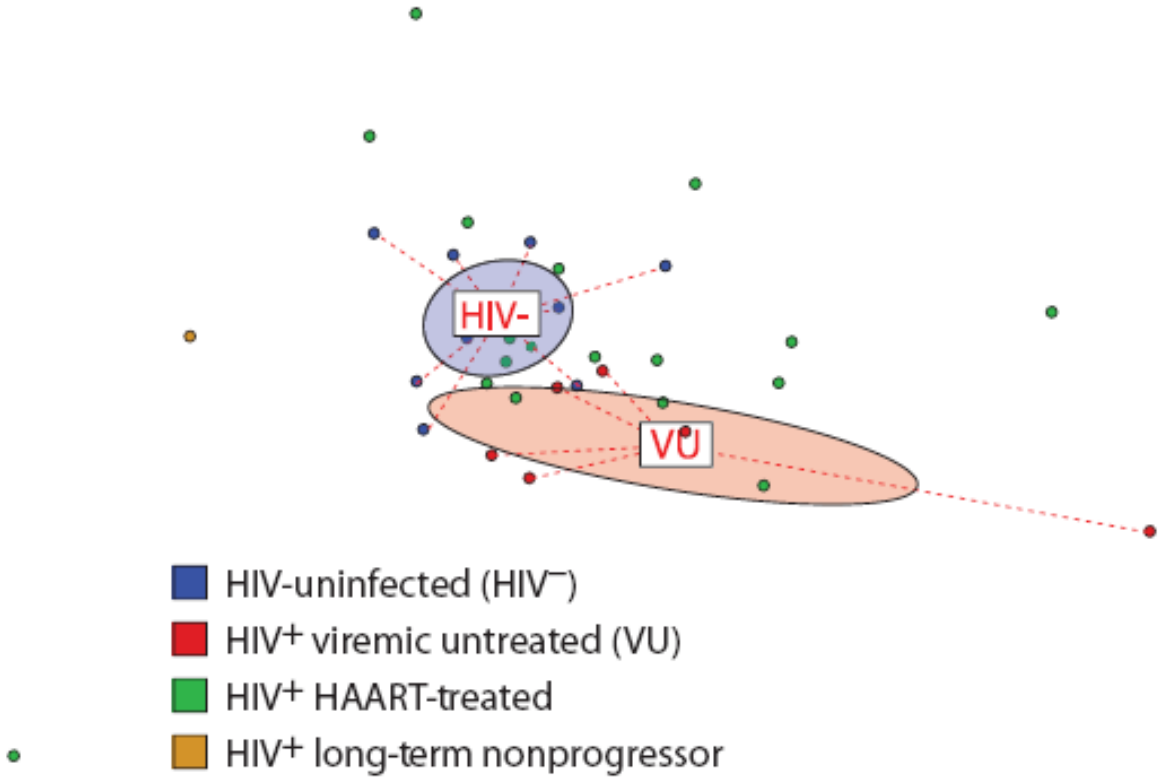


Fig. 1. Gut bacterial microbiota composition in HIV-infected VU subjects differs from that of HIV-uninfected risk-matched controls.

Results

- 33’951 Bacterial taxa identified
- 579 taxa enriched, 45 taxa depleted in VU subjects compared to HIV-

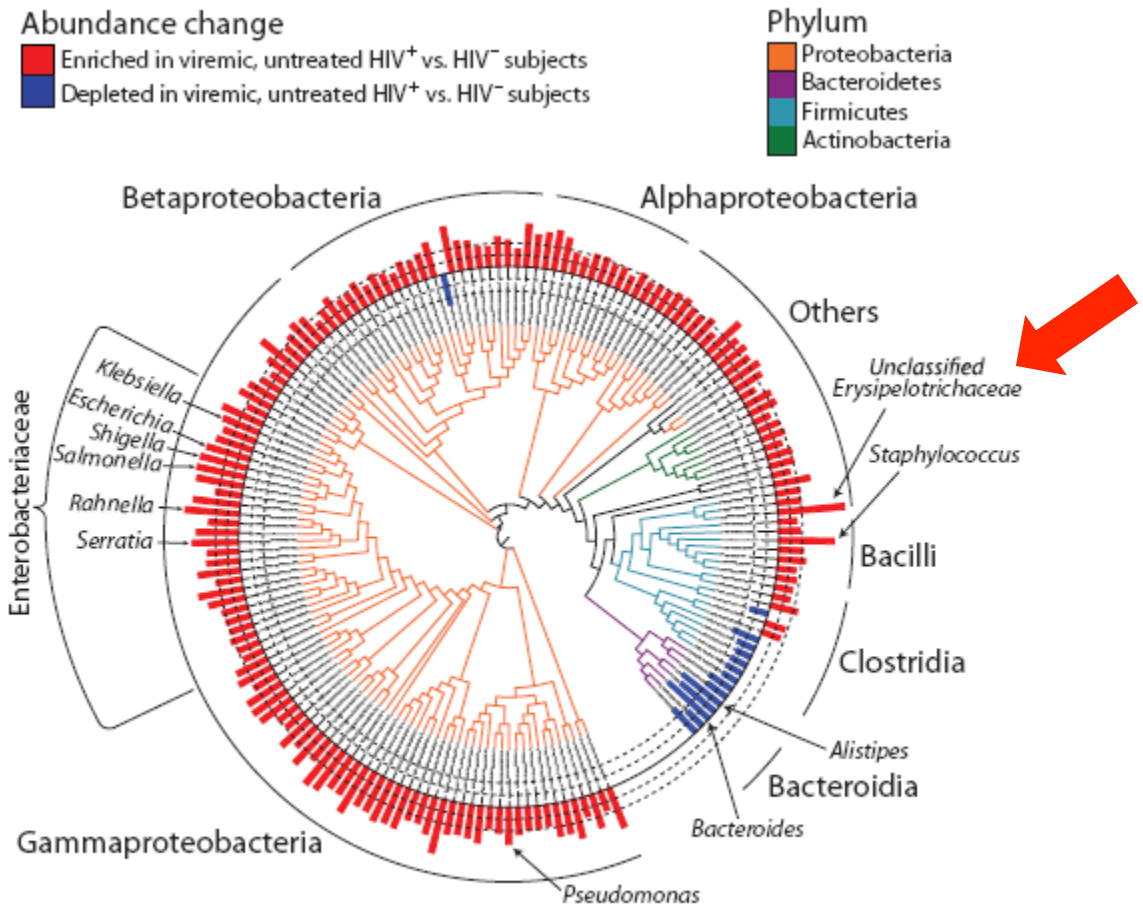


Fig. 2. Phylogenetic distribution of the DMC, defined as significantly depleted or enriched in VU subjects compared with HIV− subjects. Phy-

Results

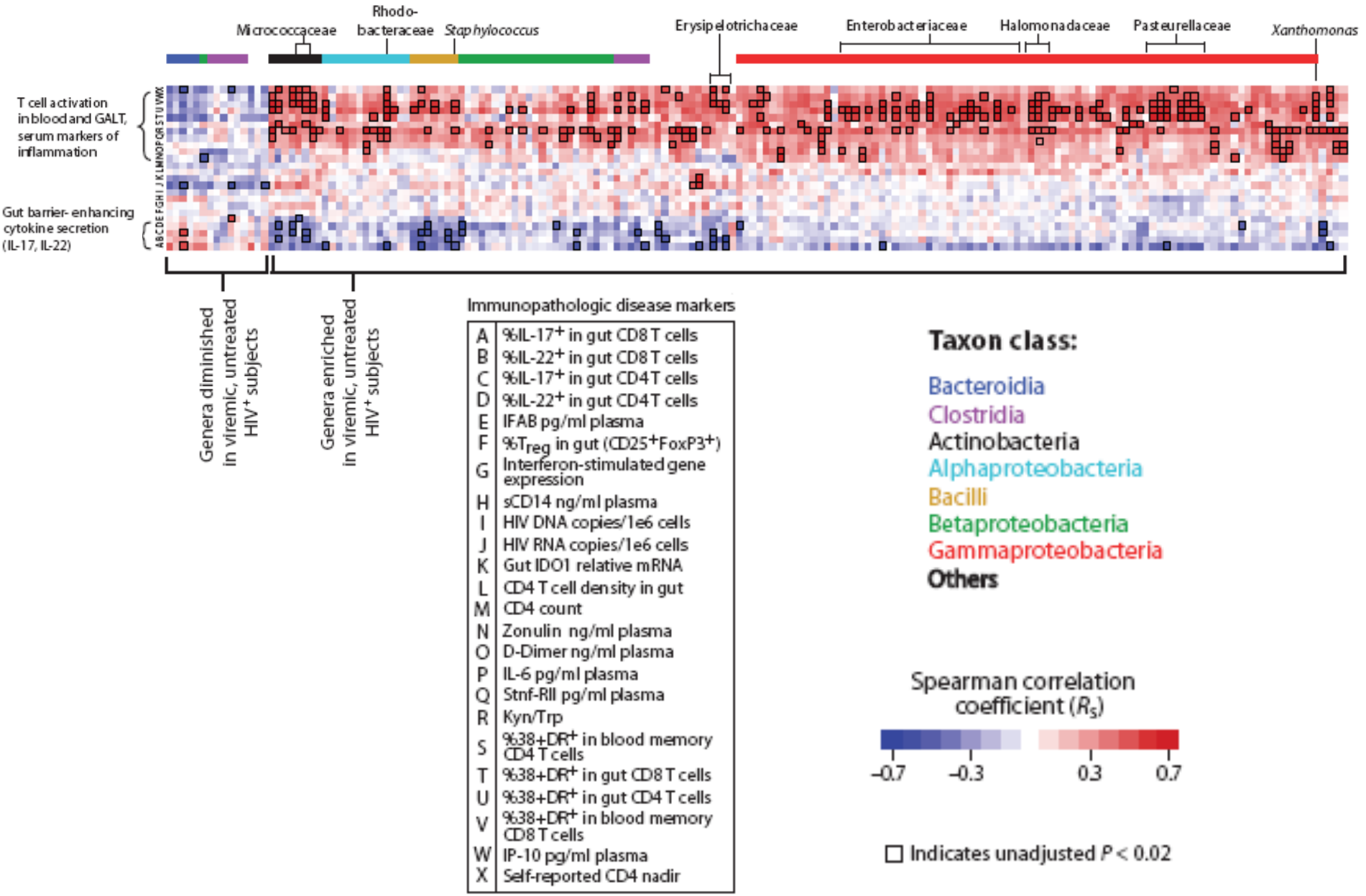


Fig. 3. Bacterial community enriched in untreated infection associates with immunopathologic markers of HIV disease progression within HIV-infected subjects. (A)

Results

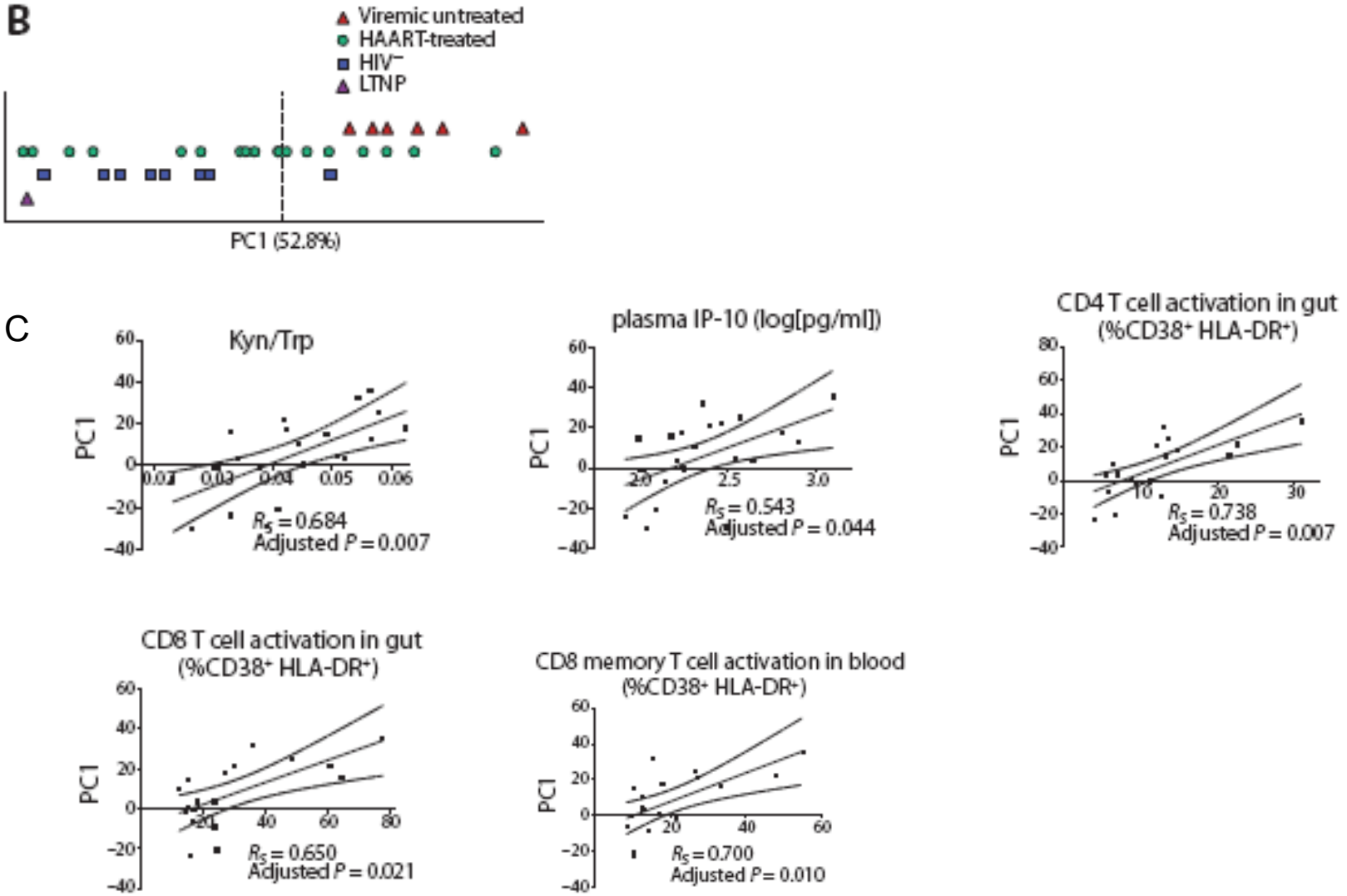


Fig. 3. Bacterial community enriched in untreated infection associates with immunopathologic markers of HIV disease progression within HIV-infected subjects. B + C

Results

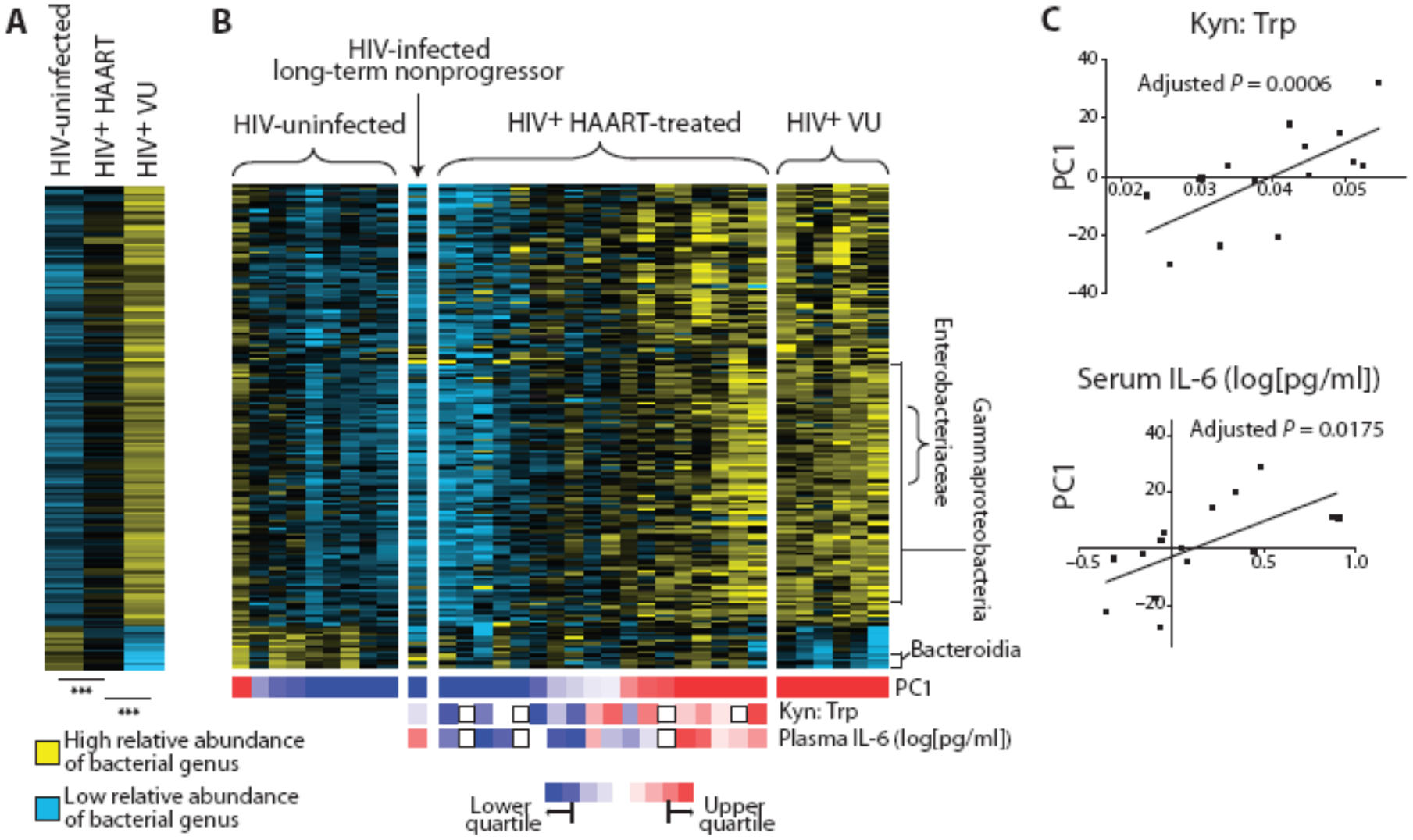


Fig. 4. Relative abundance of DMC members is diminished in HAART subjects compared to VU and falls along a spectrum of VU- or uninfected-like bacterial communities.

Results

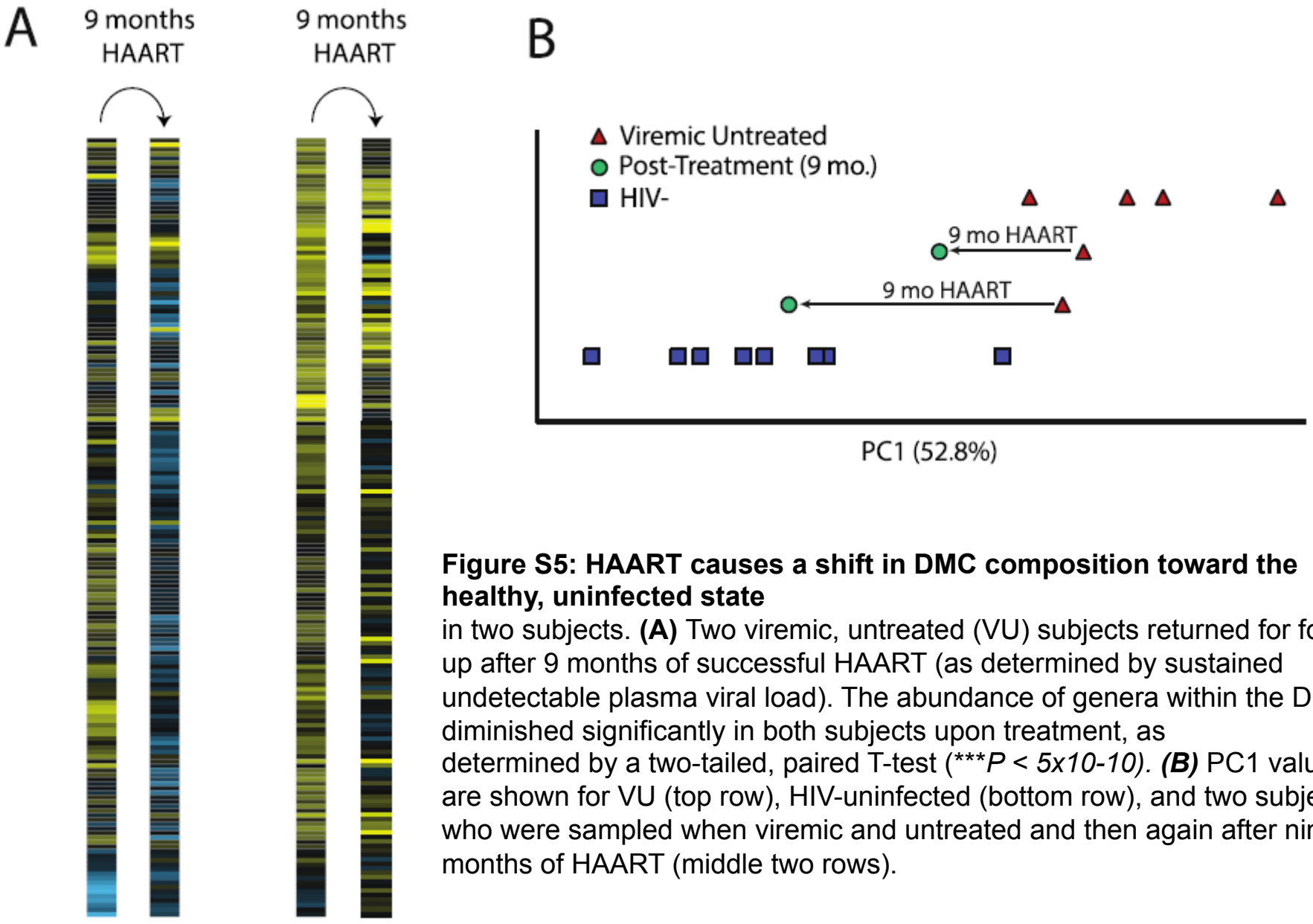
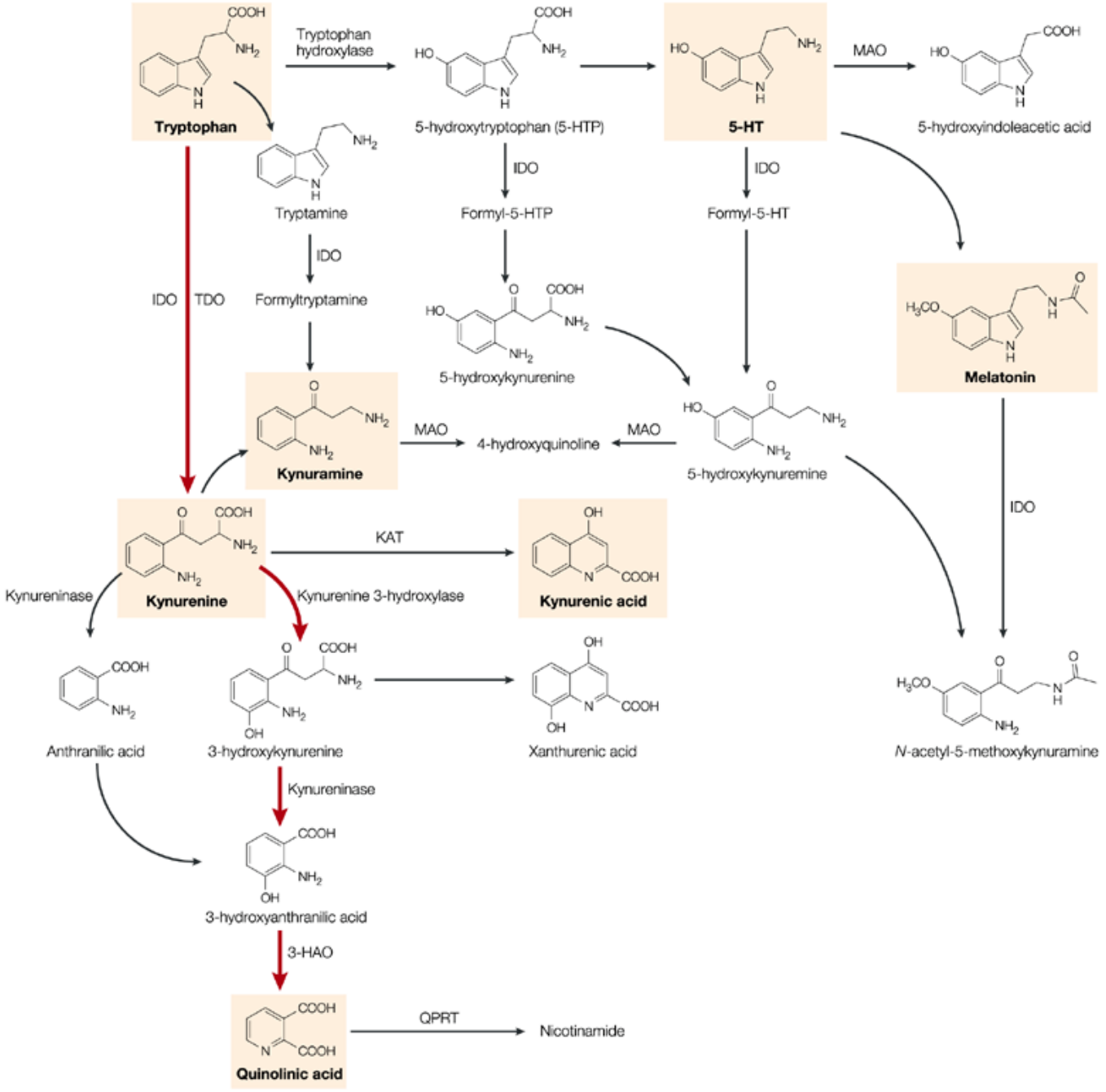


Figure S5: HAART causes a shift in DMC composition toward the healthy, uninfected state
in two subjects. **(A)** Two viremic, untreated (VU) subjects returned for follow-up after 9 months of successful HAART (as determined by sustained undetectable plasma viral load). The abundance of genera within the DMC diminished significantly in both subjects upon treatment, as determined by a two-tailed, paired T-test (****P < 5x10-10). **(B)** PC1 values are shown for VU (top row), HIV-uninfected (bottom row), and two subjects who were sampled when viremic and untreated and then again after nine months of HAART (middle two rows).

Results

Of the dietary tryptophan that is not used in protein synthesis, 99% is metabolized along the kynurenine pathway (red arrows). Alternative pathways are the conversion of tryptophan to 5-hydroxytryptamine (5-HT) and then to melatonin, or to tryptamine and then to the kynuramines (or kynurenamines). 3-HAO, 3-hydroxyanthranilic acid oxidase; IDO, indoleamine 2,3-dioxygenase; KAT, kynurenine aminotransferase; MAO, monoamine oxidase; QPRT, quinolinic-acid phosphoribosyl transferase; TDO, tryptophan 2,3-dioxygenase.



Results

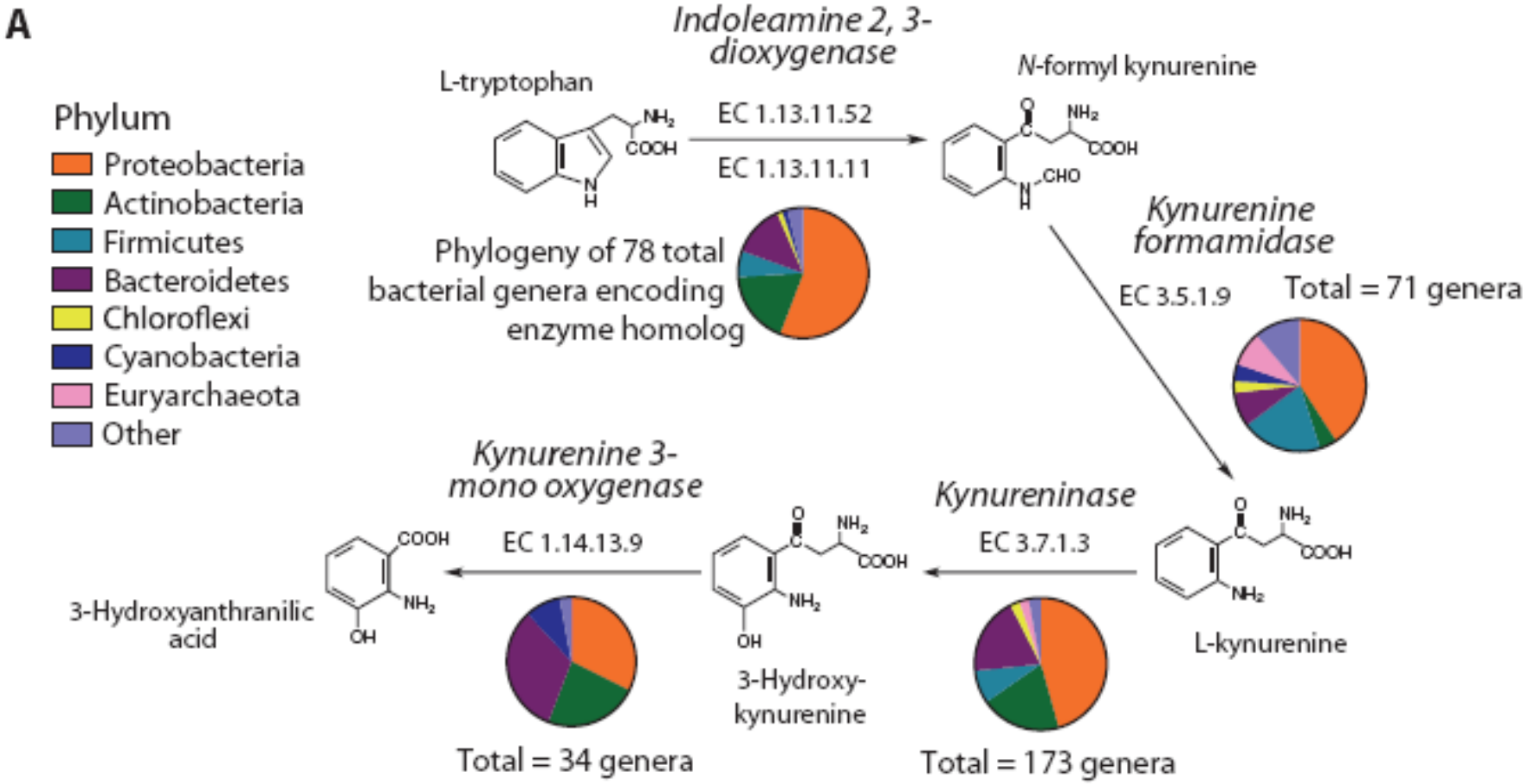
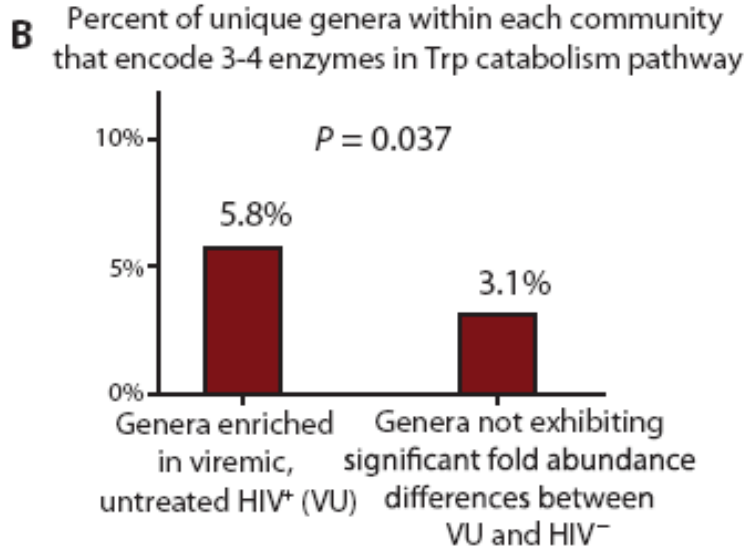
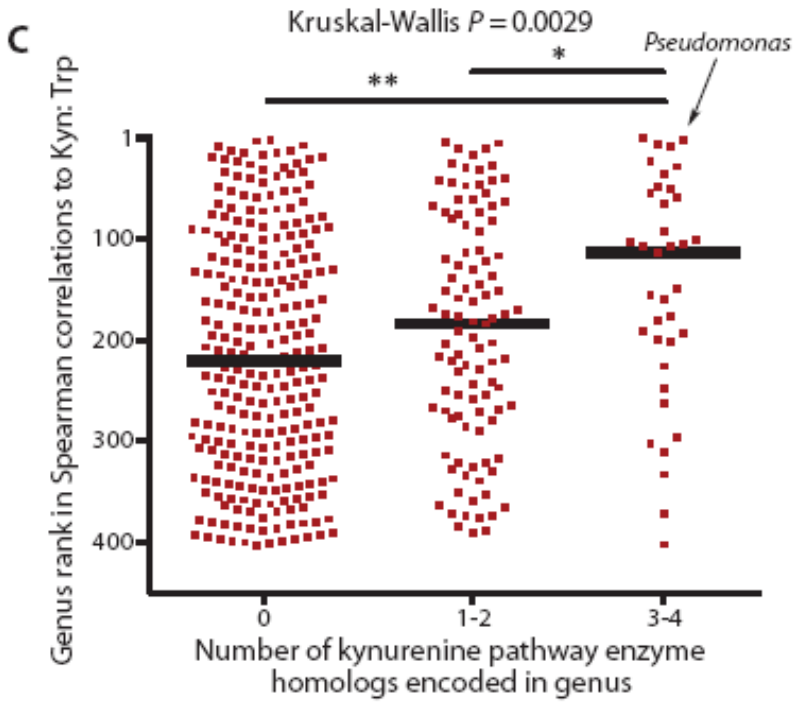


Fig. 5. Bacterial tryptophan catabolism machinery is genetically and functionally homologous to IDO1 enzymatic activity and is enriched in the DMC. (A)

Results





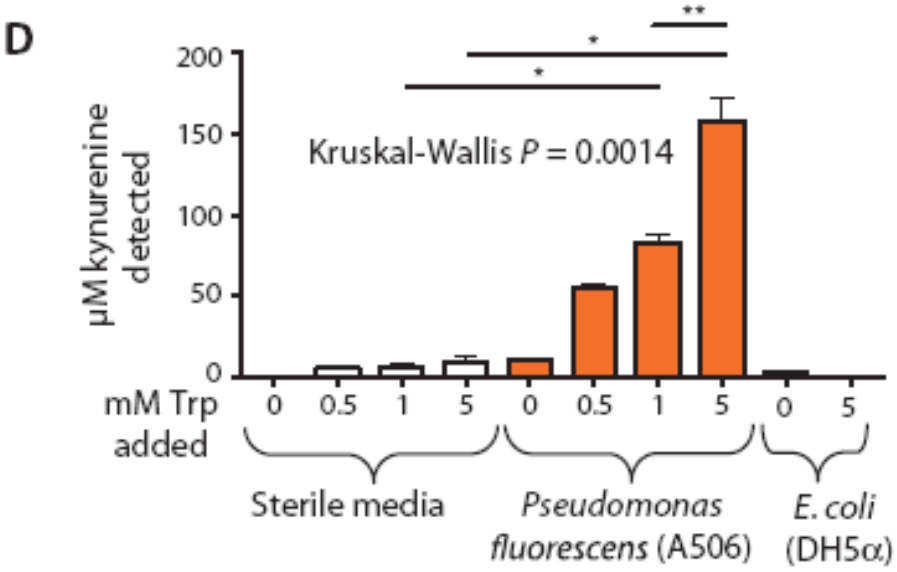


Fig. 5. Bacterial tryptophan catabolism machinery is genetically and functionally homologous to IDO1 enzymatic activity and is enriched in the DMC. B + C + D

Discussion /Conclusions

- Alterations in gut flora could be shown.
- Correlation of microbiota with disease marker. -> Mechanisms stay unclear
- Influence of reactive oxygen species, disturbance in microbial pattern recognition and immune cell dysregulation during HIV infection are not considered.
- Bacteria can catabolize tryptophan to immunomodulatory kynurenine derivatives.
- Potential role of kynurenine for barrier disruption.
- Role of host tryptophan metabolism? (Vitamin B6?)

T cell regulation mediated by interaction of soluble CD52 with the inhibitory receptor Siglec-10

Esther Bandala-Sanchez1,2,5, Yuxia Zhang1,2,5, Simone Reinwald1,2, James A Dromey1,4, Bo-Han Lee1,2, Junyan Qian1,2,4, Ralph M Böhmer1,2 & Leonard C Harrison1–3

Introduction

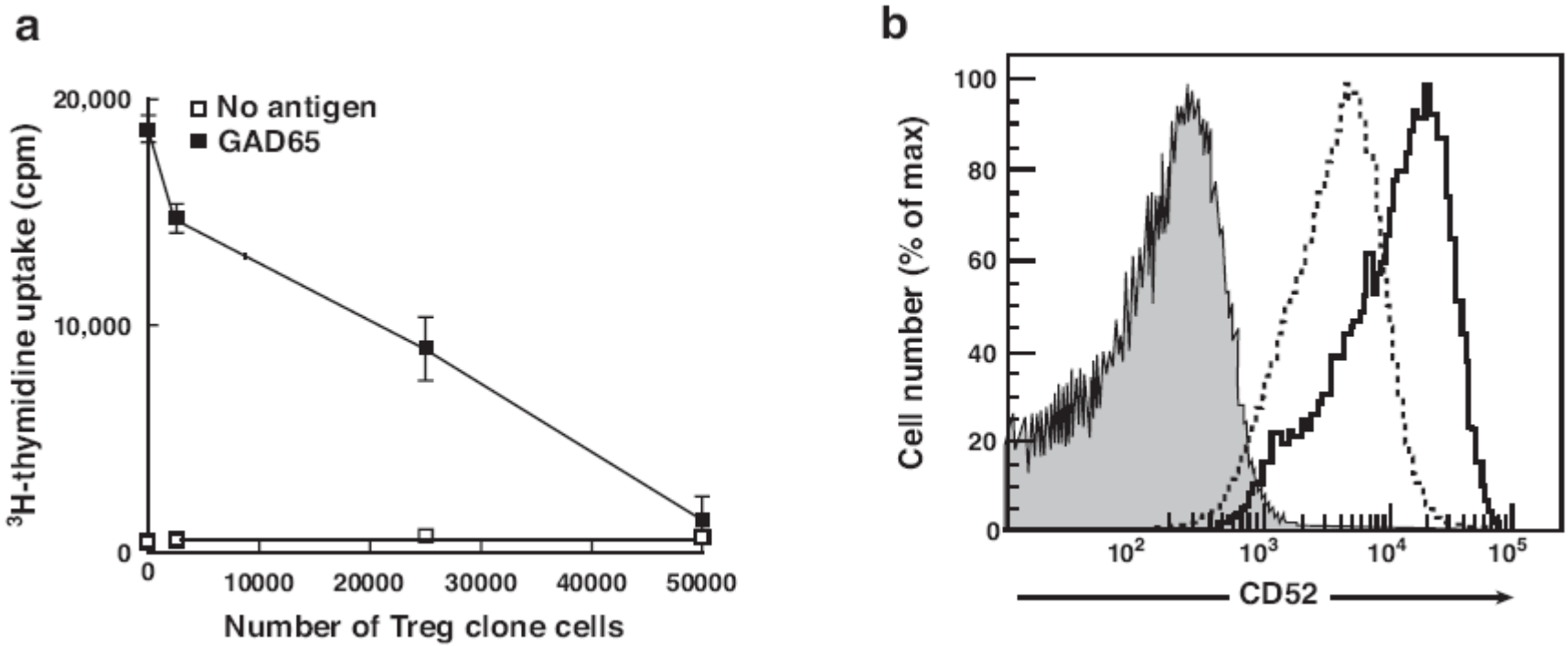
- New suppressor T cell -> $CD52^{hi}$ $CD4^{+}$
- CD52: CAMPATH-1 antigen, expressed on mature lymphocytes, monocytes/ macrophages, dendritic cells, eosinophils, mast cells, epithelial cells lining the male reproductive tract and sperm cells.
- Anti-CD52 antibody Alemtuzumab kills target cells, used to treat chronic lymphocytic leukemia.
- Siglec10: Sialic acid-binding Ig-like lectin 10, expressed on monocytes, eosinophils, B cells, ITIM domain.
- Expressed on T cells?

Loss of Siglec expression on T lymphocytes during human evolution

Dzung H. Nguyen*†, Nancy Hurtado-Ziola*†‡, Pascal Gagneux*, and Ajit Varki*§

- **T cell regulation preventing autoimmune diseases?**

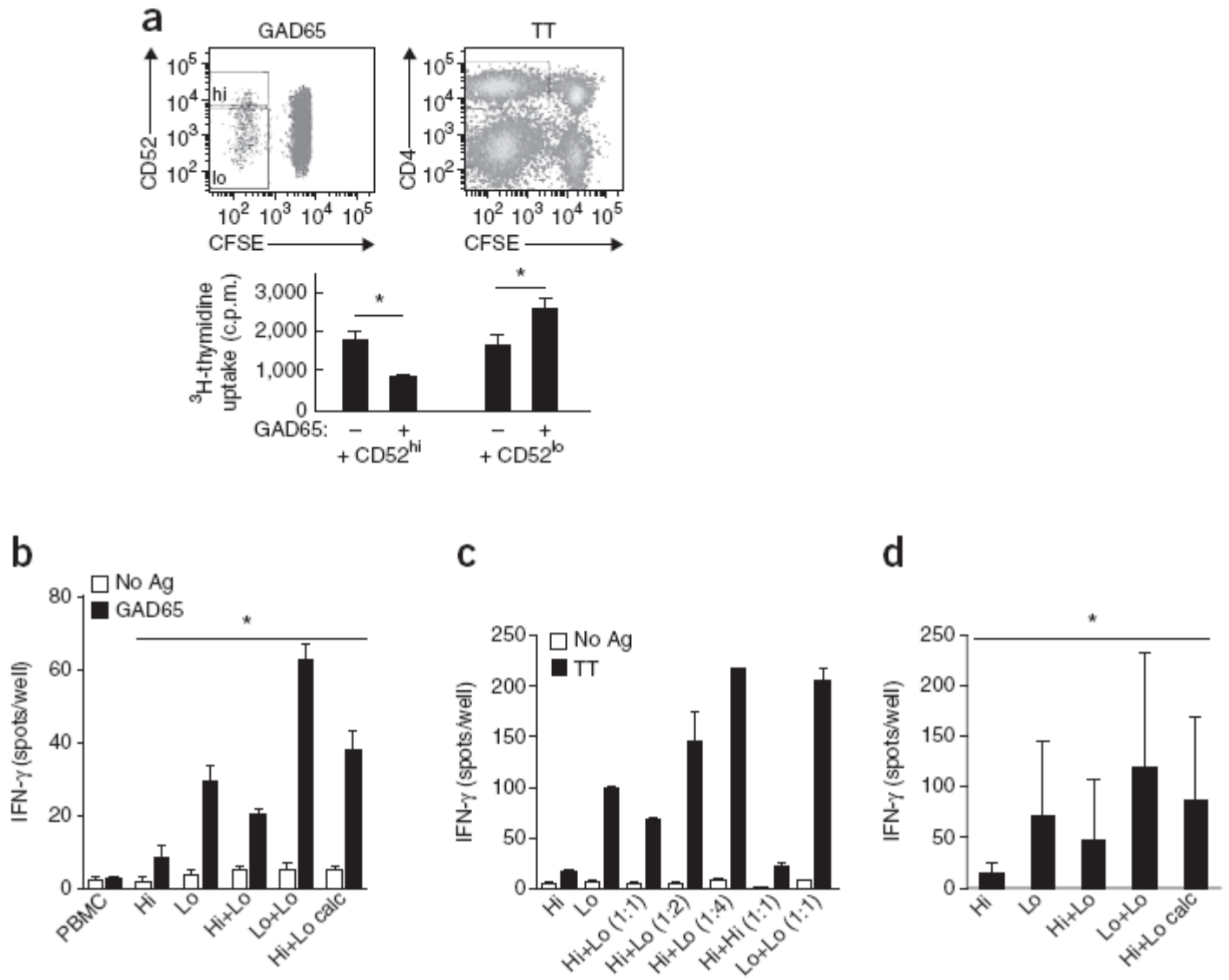
Results



Supplementary figure 1: CD52 expression distinguishes GAD65-specific CD4+ suppressor T cell clones**. a,** Proliferation of a GAD65-specific T cell clone (# 1.4) in the presence of an autologous GAD65-specific suppressor clone (# 3.19). A fixed number (25000) of GAD65-specific non-suppressor clone cells was co-cultured in 200 µl in round bottom 96-well plates with increasing numbers of an autologous GAD65-specific suppressor clone and irradiated PBMCs (1x105) as antigen presenting cells, in the presence or absence of GAD65. 3H-thymidine uptake was measured after 72 h. The result is representative of multiple autologous suppressor and non-suppressor clone pairs as previously described14. **b, Flow cytometric** histograms of CD52 expression by autologous GAD65-specific suppressor (solid line) and non-suppressor (dashed line) clones after overnight stimulation by plate-bound anti-CD3 antibody (1µg/ml). Staining by isotype control antibody is depicted in solid grey. The result is representative of clone pairs from 3 healthy individuals.

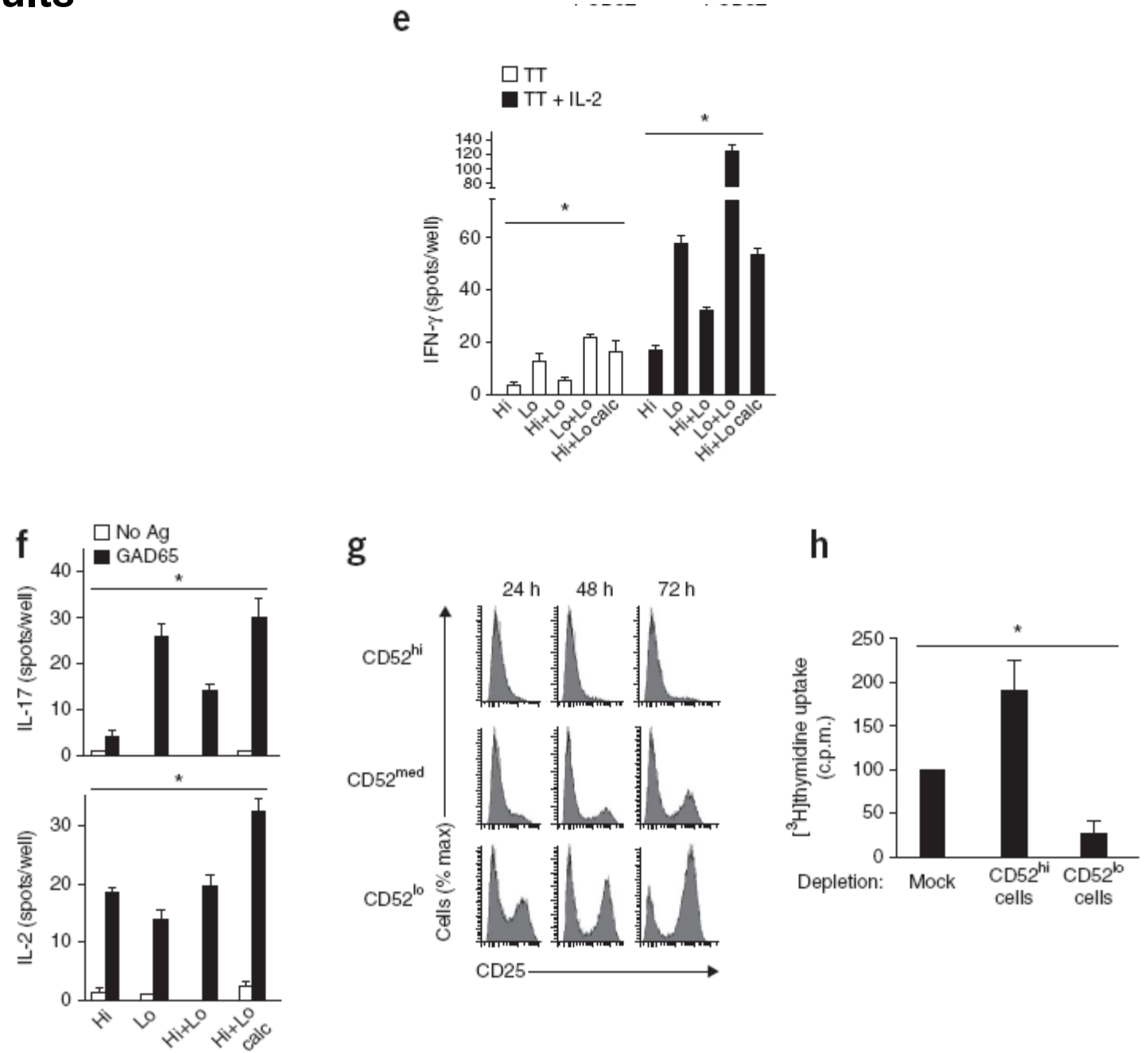
Results

Fig. 1



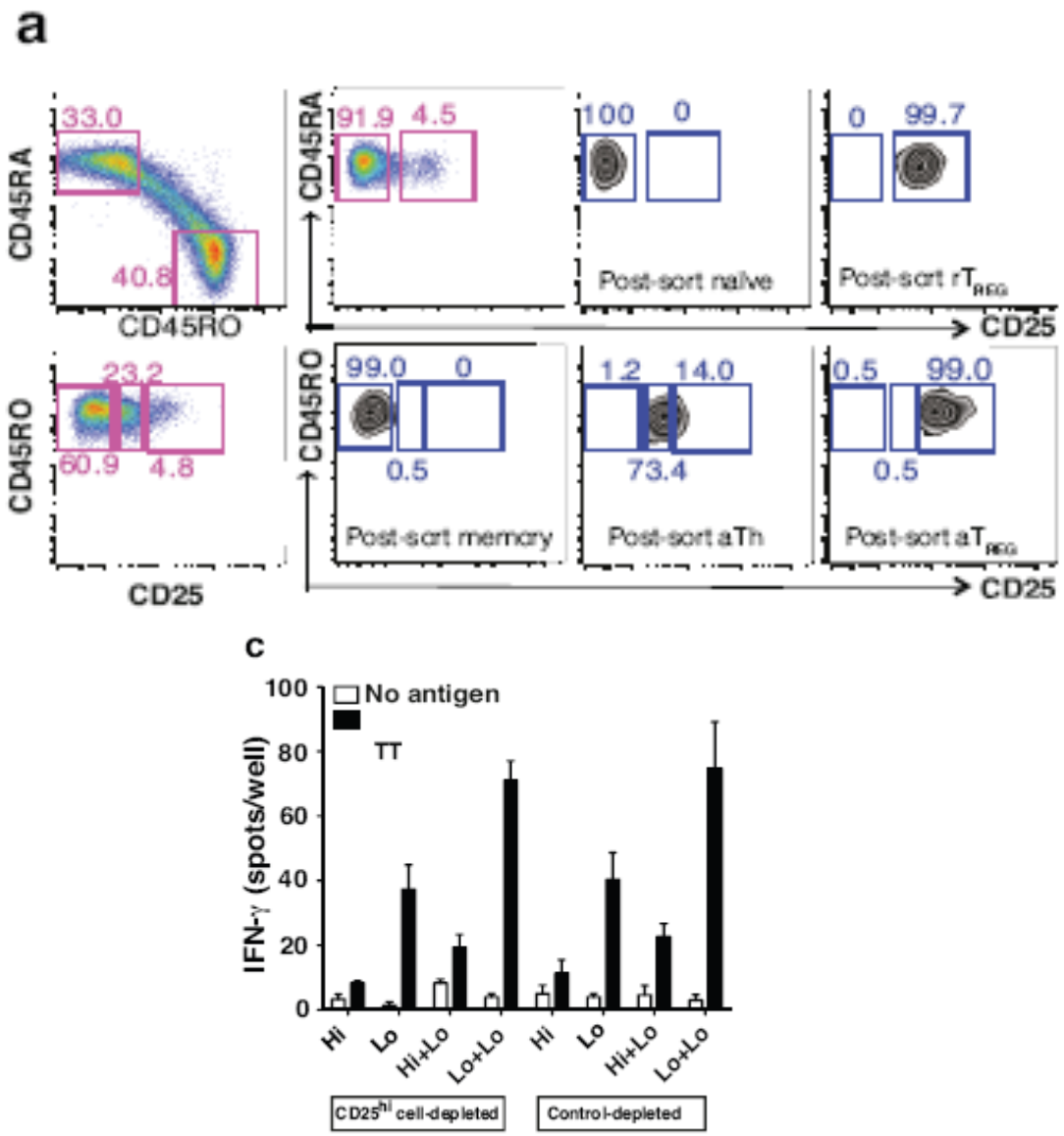
Results

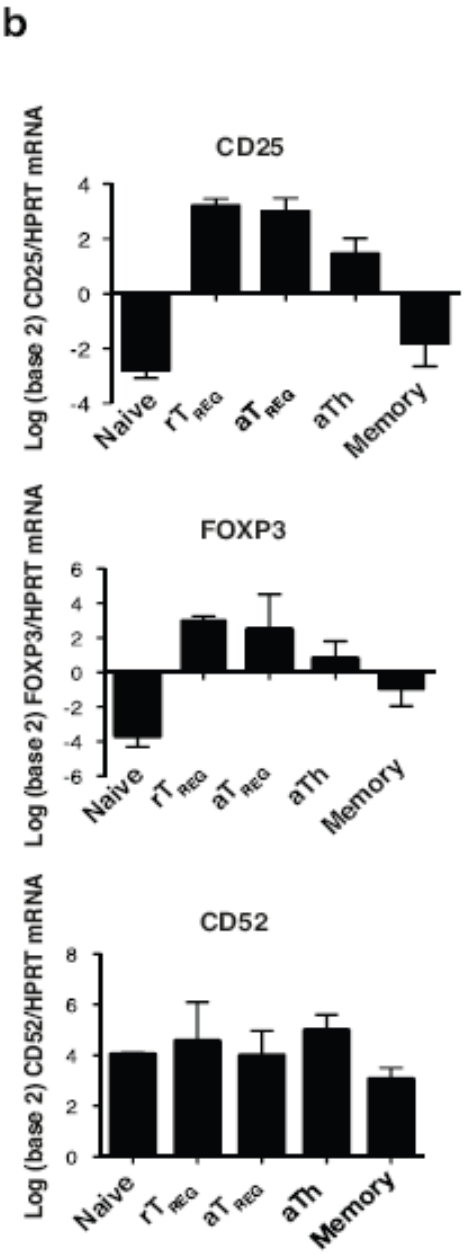
Fig. 1



Results

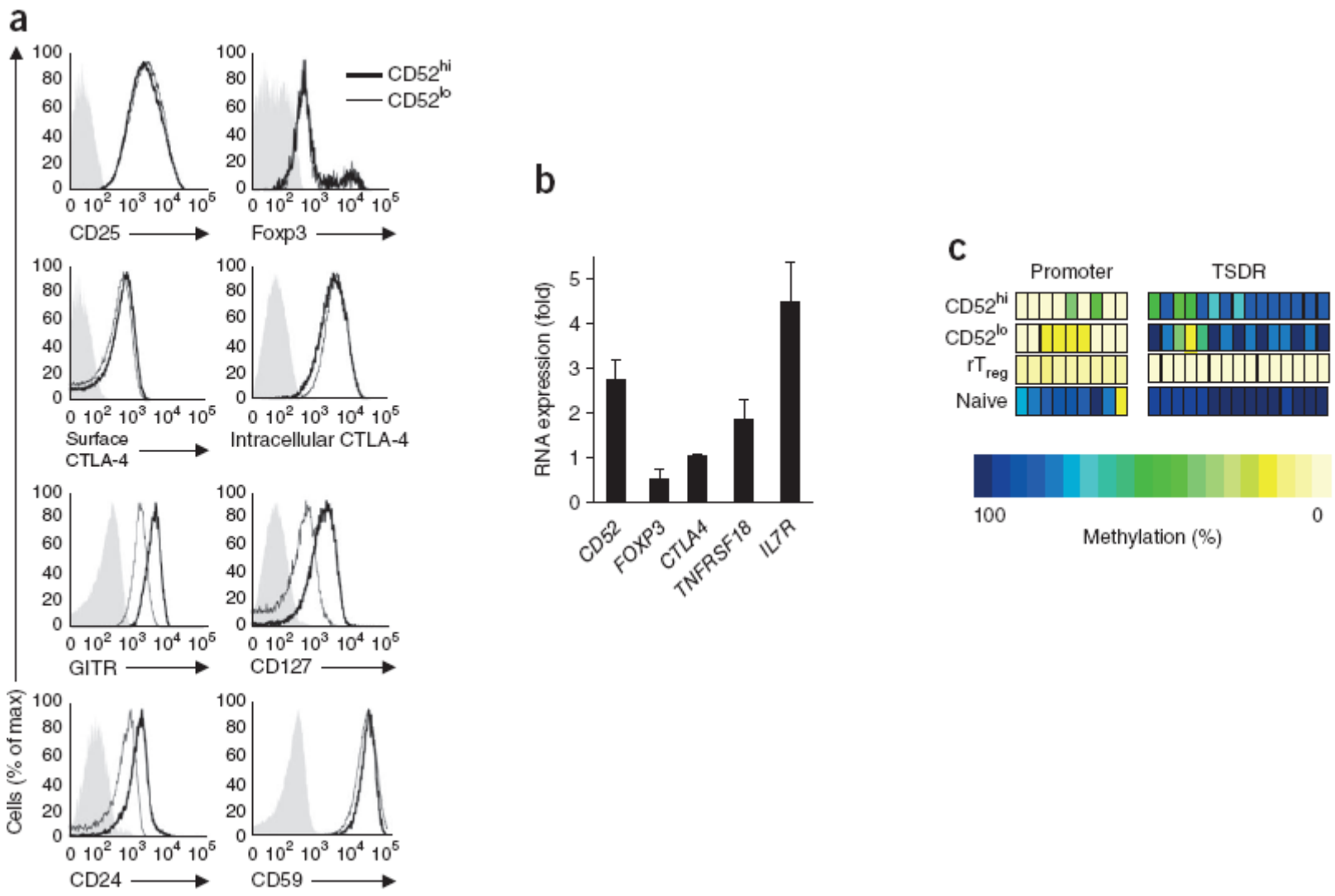
Sup. Fig. 2





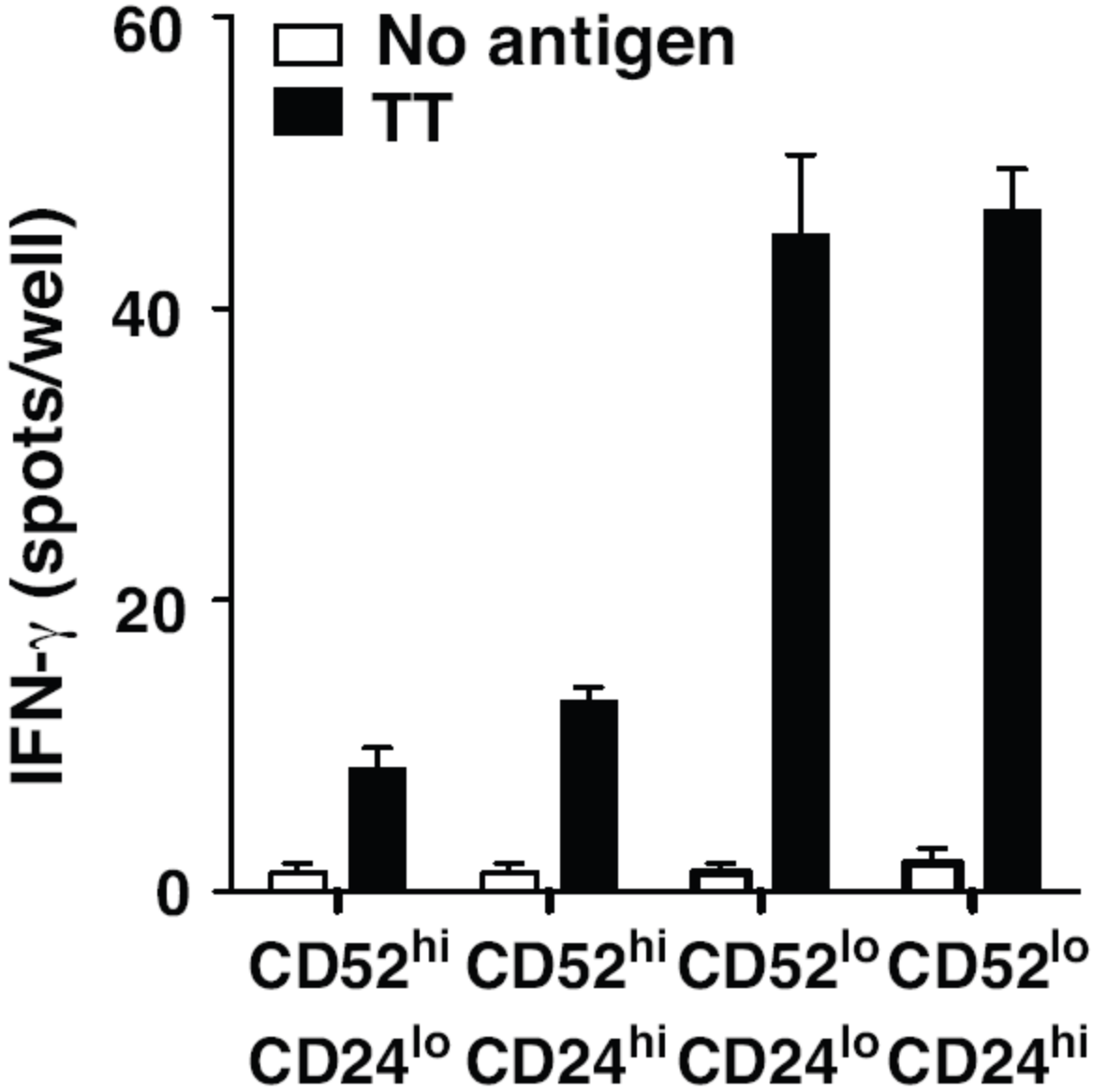
Results

Fig. 2



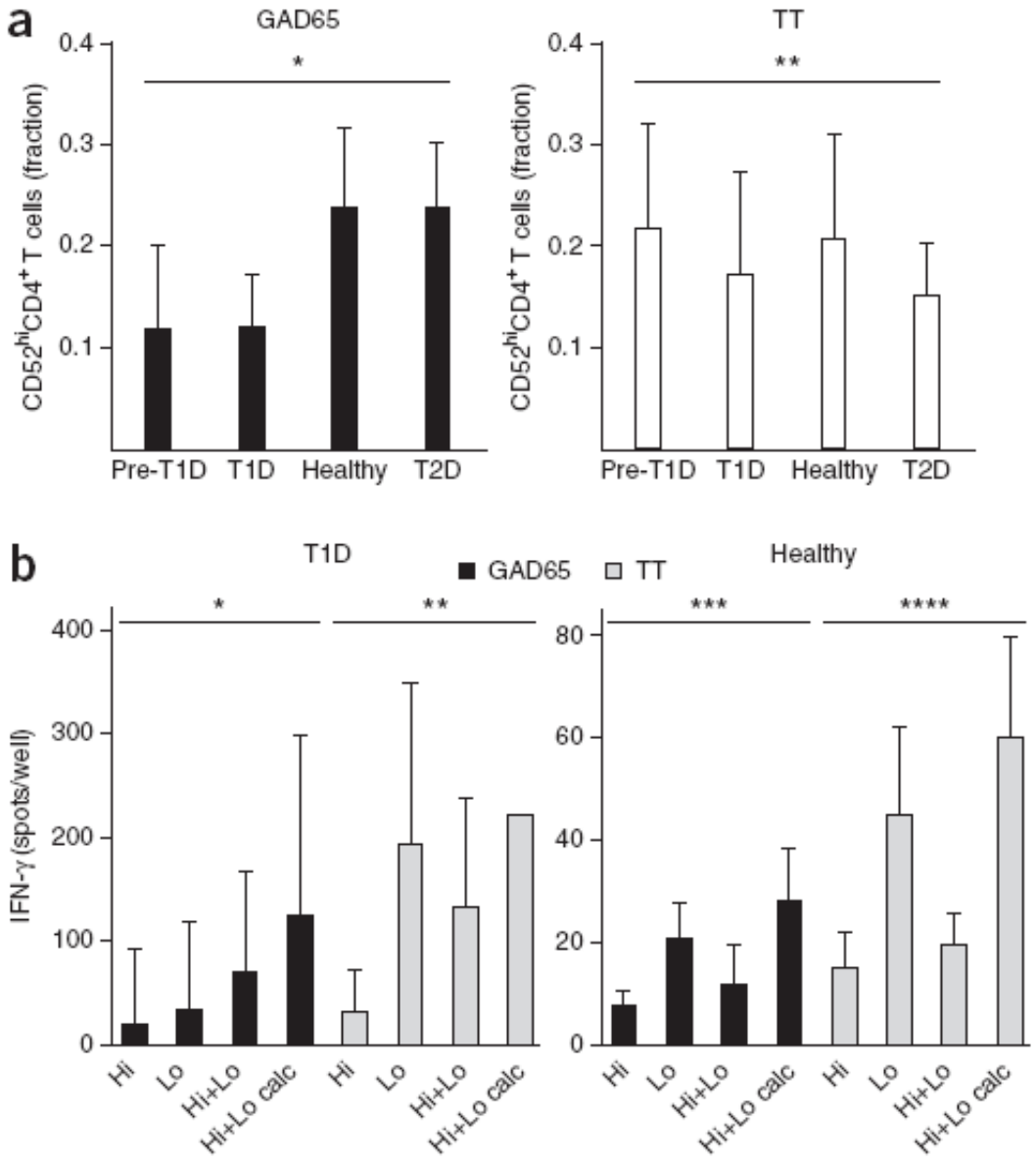
Results

Sup. Fig. 3



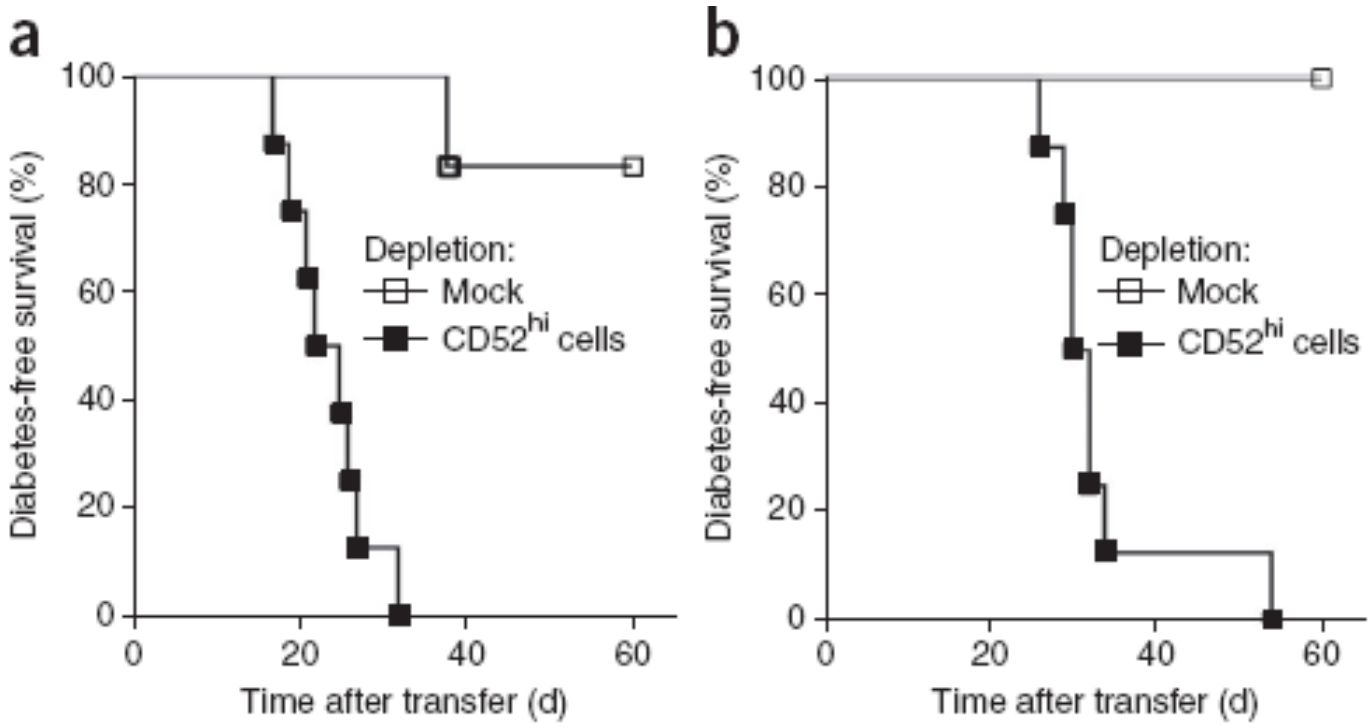
Results

Fig. 3



Results

Fig. 4



Results

Sup. Fig. 4

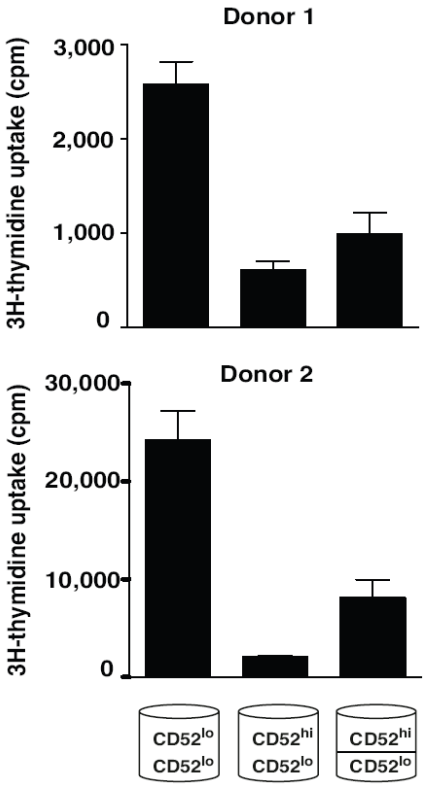
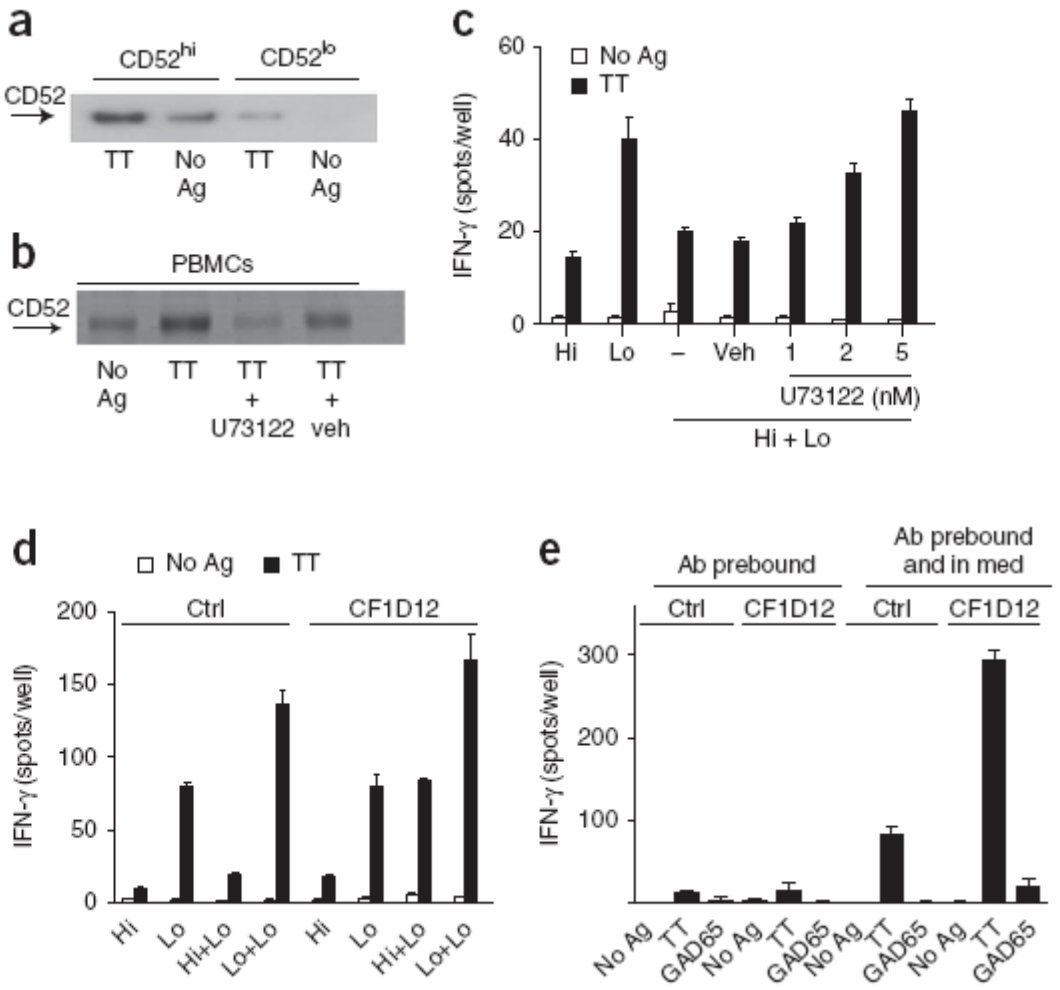
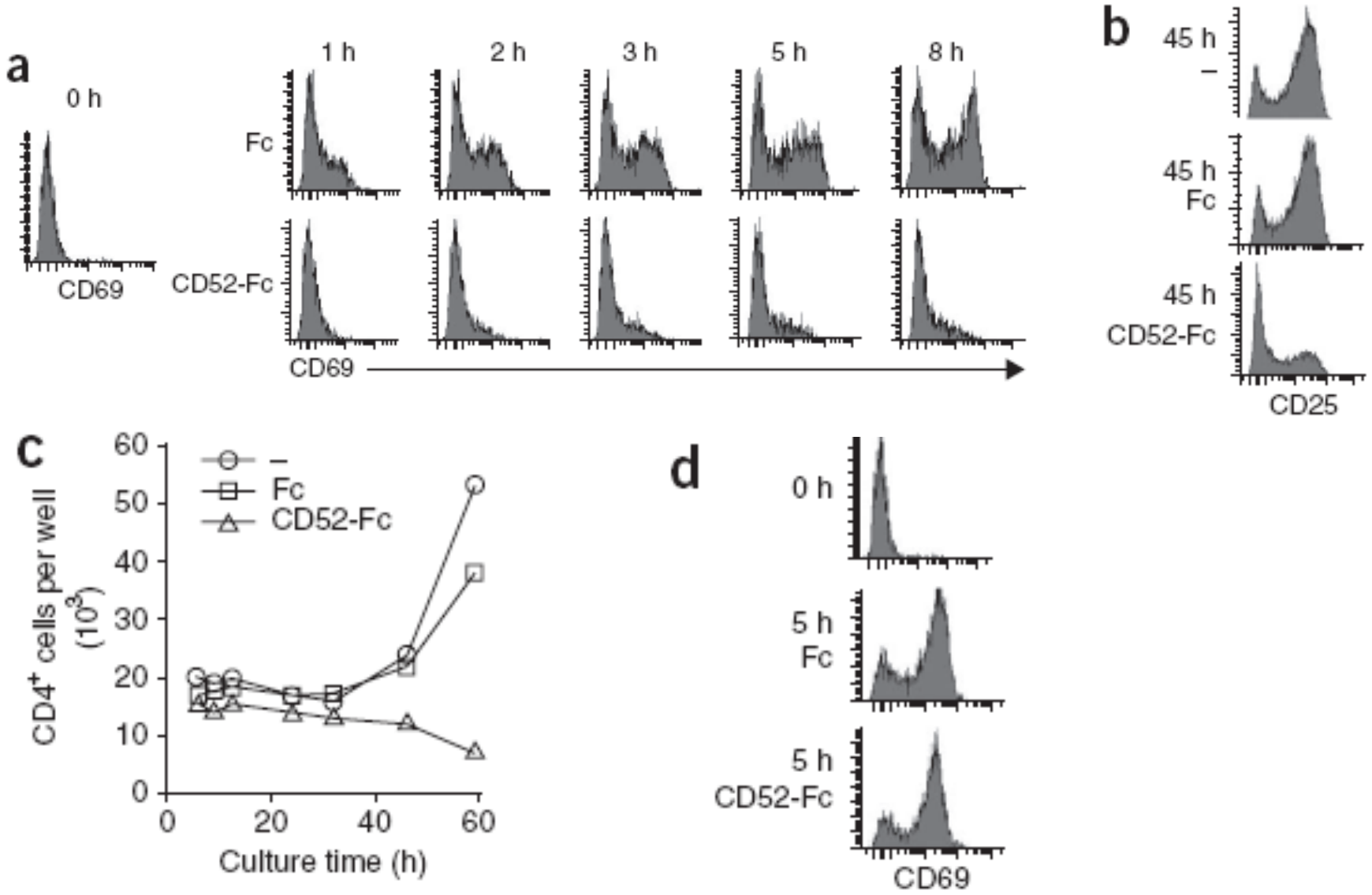


Fig. 5



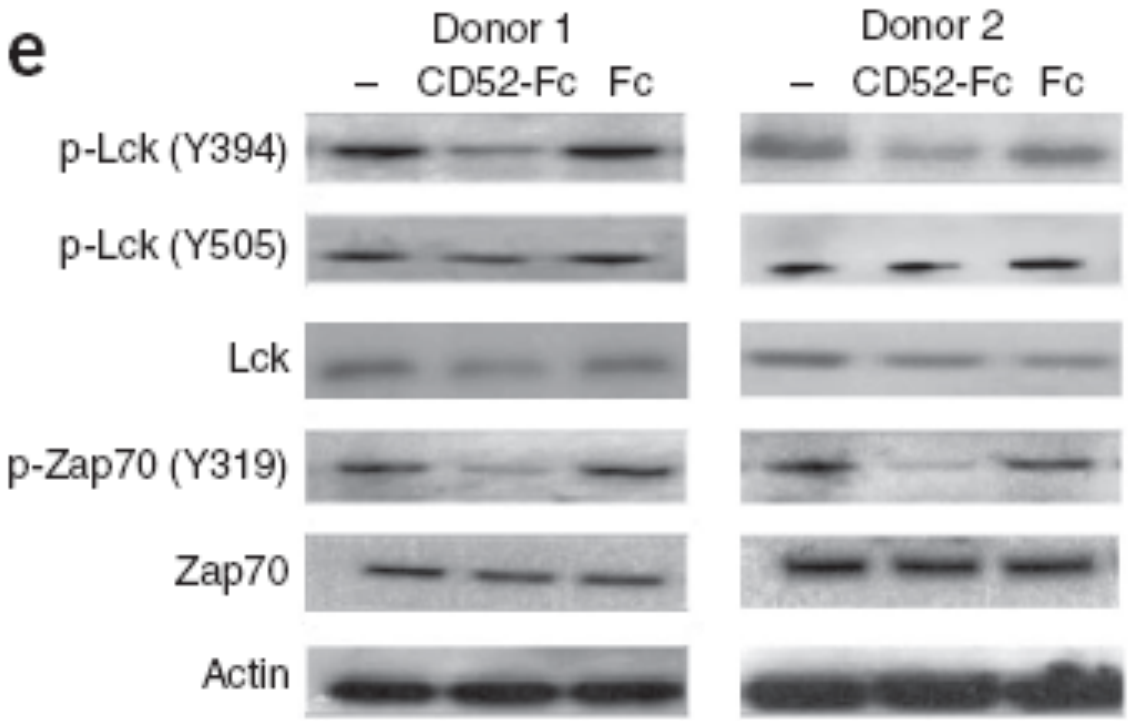
Results

Fig. 6



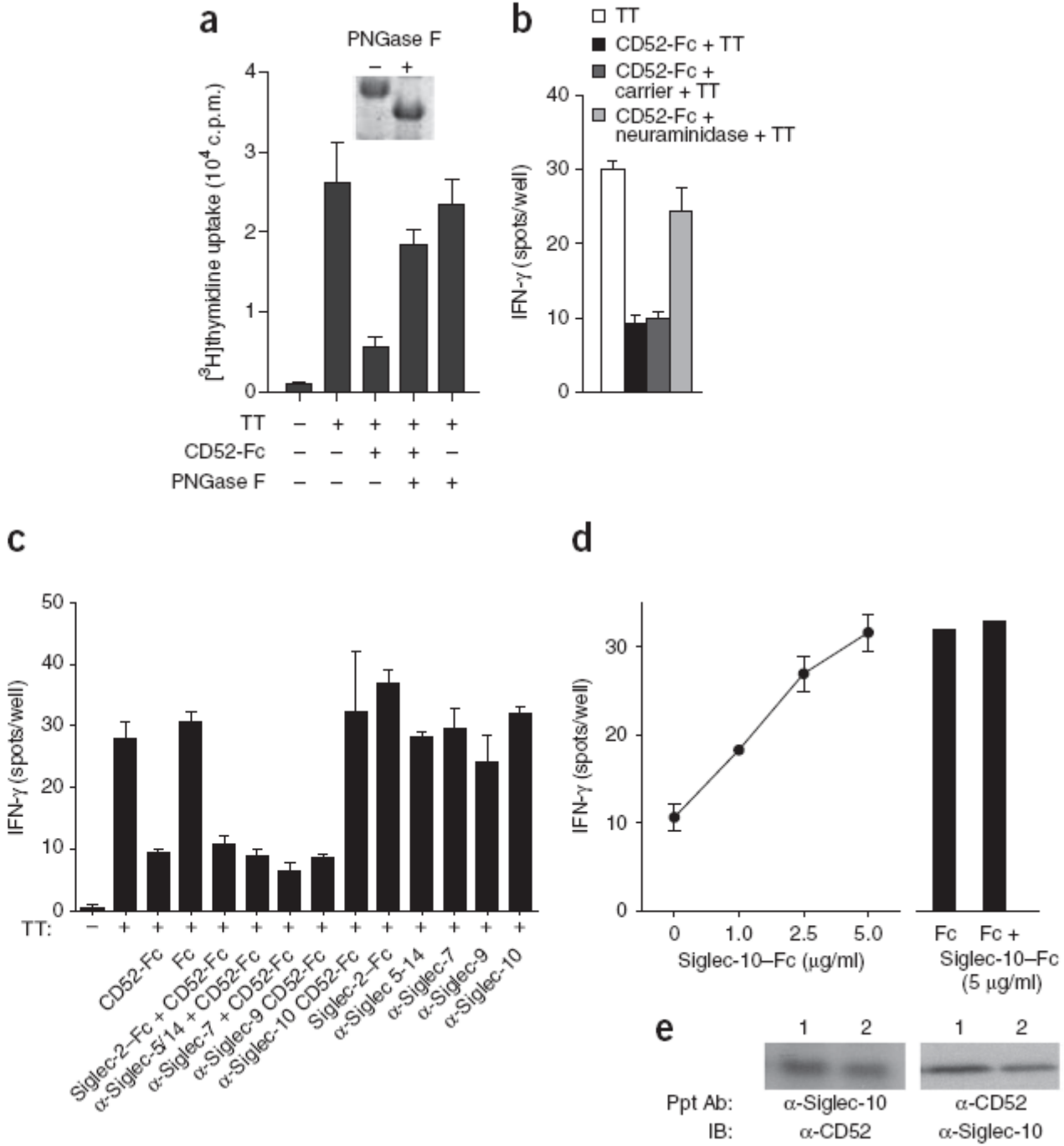
Results

Fig. 6



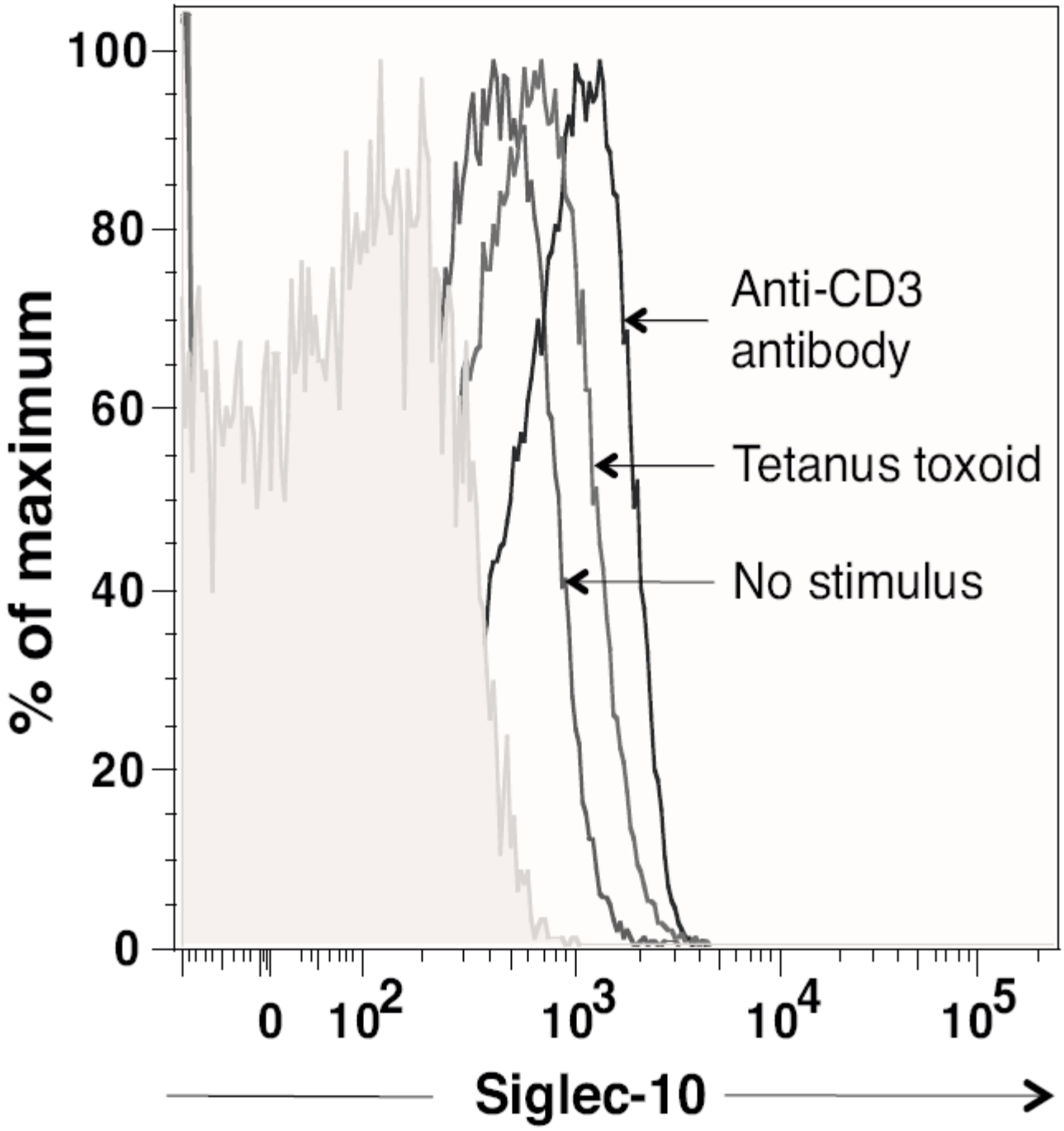
Results

Fig. 7



Results

Sup. Fig. 6



Discussion/Conclusions

- New T cell subset with suppressor activity.
- CD52 – Siglec-10 new receptor – ligand pair

Open questions

- How strong is Siglec-10 expressed on T cells?
- Interaction with cell bound CD52?
- CD8 suppressor cells?