Foreign and autoreactive CD4 conventional and regulatory T cells in T1D and healthy subjects

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Dysfunction in regulatory T cells (Tregs) is linked to Type 1 Diabetes (T1D), as Tregs prevent autoreactive conventional T cells (Tconvs) from damaging insulin-producing pancreatic Islet β cells. The moderation mechanism of Islet antigen-reactive (IAR) Tconvs by Tregs is unclear. Previously, we found limited TCR sharing between IAR Tconvs and Tregs. To look for this finding in foreign AR T cells, we compared CD4 Tconv and Treg cells reactive to a microbial peptide pool (CEFX) to Tconv and Treg cells reactive to islet peptides in 3 HC and 3 T1D subjects. We ran sc-seq of 6 subjects using 10X GEM-X for RNA-seq, TCR-seq, and CITE-seq antibodies (ab). Fresh blood was processed to PBMCs and stimulated with islet peptides, CEFX, or anti-CD3/anti-CD28 (polyclonal stim.) for 20 h. Post-stim., cells were stained with a unique hashtag ab, combined, enriched for CD154+ and CD137+ cells, stained with CITE-seq and flow abs, and sorted for CD154+ or CD137+ cells. We added polyclonal cells to the AR cells to enable analysis against the CD4 landscape. IAR and polyclonal cells showed similar transcriptomic signatures and recovery, while CEFX cells displayed a distinct profile and higher recovery. Treg and Tconv cells were distinguishable by scRNA-seq and CITE-seq across stimulations. Approximately 85% of recovered cells contained TCR pairs. Ongoing study aims to determine if autoreactive T cells differ from foreign-reactive T cells by investigating the TCR repertoire and transcript phenotypes of CD4 Tconv and Treg cells.