Investigating the Characteristics of Cutaneous T Cells in Systemic Sclerosis and Atopic Dermatitis Using Single-cell RNA Sequencing Data

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Background

- Single-cell RNA sequencing (scRNA-seq) has allowed for the high-resolution characterization of T cells, providing new insights into their gene expression patterns and plasticity.
- T cells, a key immune regulator, are a complex and heterogeneous population that can be classified into different subsets based on the expression of cell surface molecules, effector molecules, and transcription factors.
- T cells play an important role in the development of many different disorders, including systemic sclerosis (SSc).
- SSc is a systemic autoimmune disease with high morbidity and mortality and a paucity of good therapeutic options.
- Although there is some evidence supporting the role of a type 2-oriented response in SSc, it is not clear whether T cells in systemic sclerosis have the same characteristics as those found in classic type 2-driven diseases such as atopic dermatitis (AD).

Objective

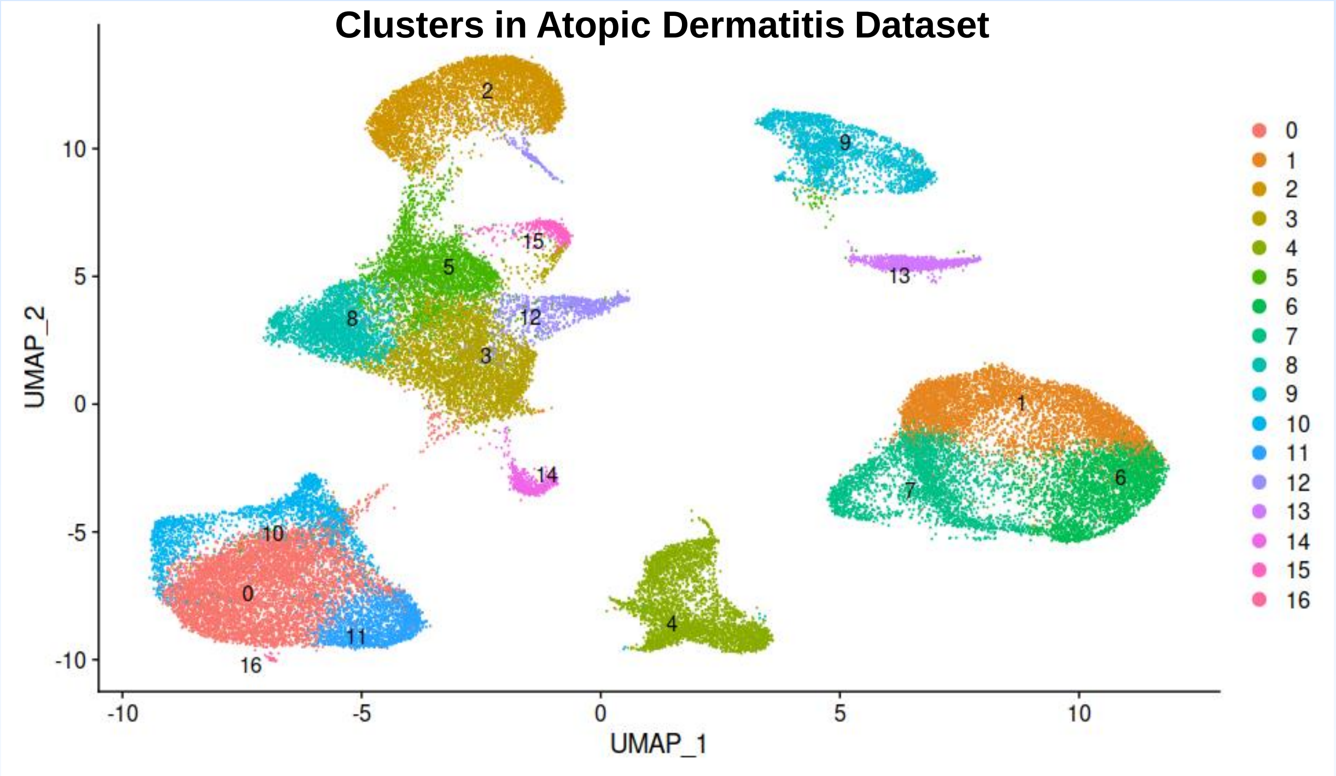
- Investigate the gene expression profile of T cells in systemic sclerosis as compared to atopic dermatitis using publicly available scRNA-seq data.

Question

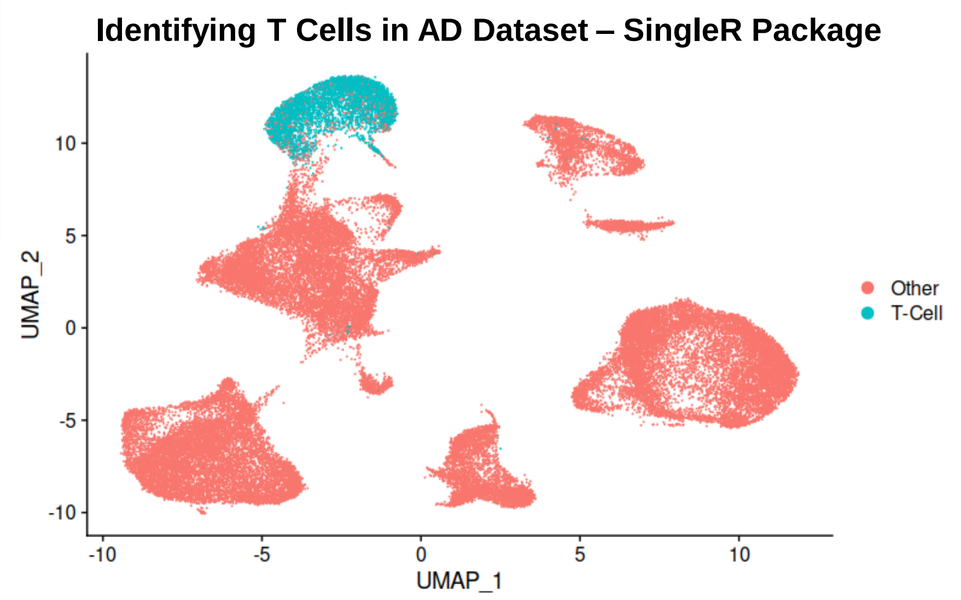
- Are there similarities in the characteristics of T cells in SSc and AD that could support shared therapeutic strategies specific for T cell-mediated inflammation and tissue damage?
- For example, dupilumab, a monoclonal antibody which blocks the binding of the type 2 cytokines IL-4 and IL-13 to their receptor, is currently used to treat AD.

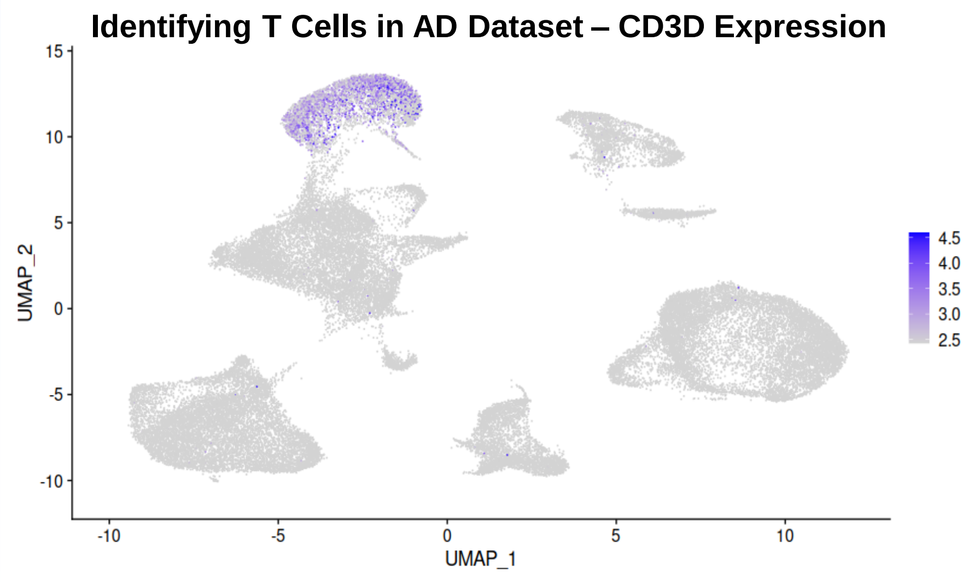
Methods

- scRNA-seq analysis can be used to investigate cellular heterogeneity and transcriptional similarities and differences within a population of cells at the single cell level.
- This project used publicly available scRNA-seq datasets, one for atopic dermatitis (5 patients with AD, 7 healthy controls) and one for SSc (55 patients with SSc, 21 healthy controls), to compare T cell profiles in skin samples.
- The datasets were analyzed using R-based platforms, including the Seurat package.
- For each dataset, a quality control workflow was used to remove low-quality cells. The data was normalized and scaled to allow for cell clustering based on transcriptomic profiles.
- Due to differences between the AD and SSc experimental datasets, these datasets were analyzed separately in R.


Clusters in Atopic Dermatitis Dataset

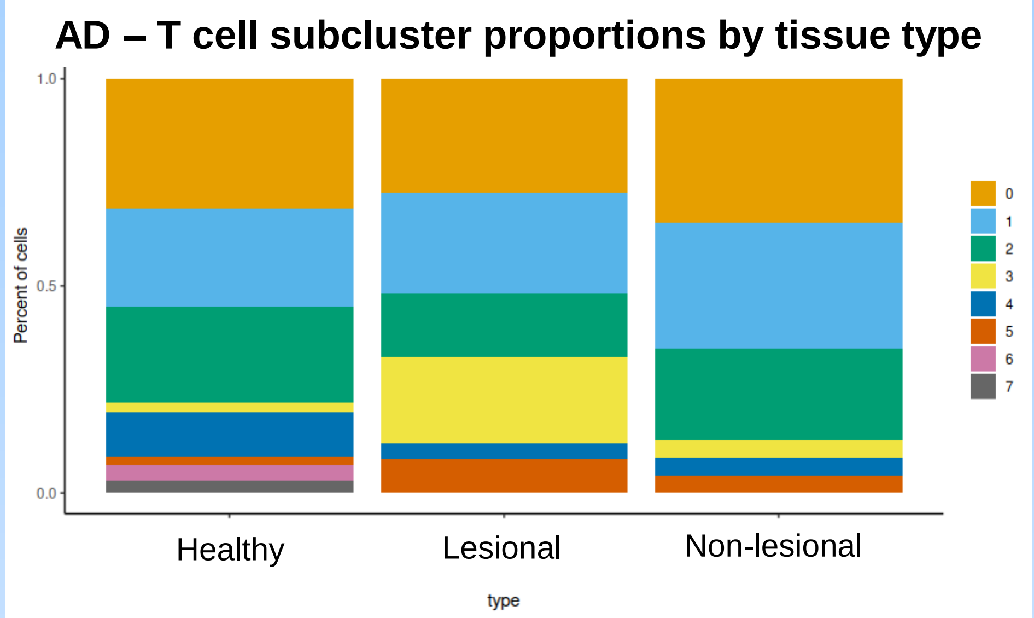
- Biased and unbiased approaches were used to examine and identify clusters.


Identifying T Cells in AD Dataset – SingleR Package

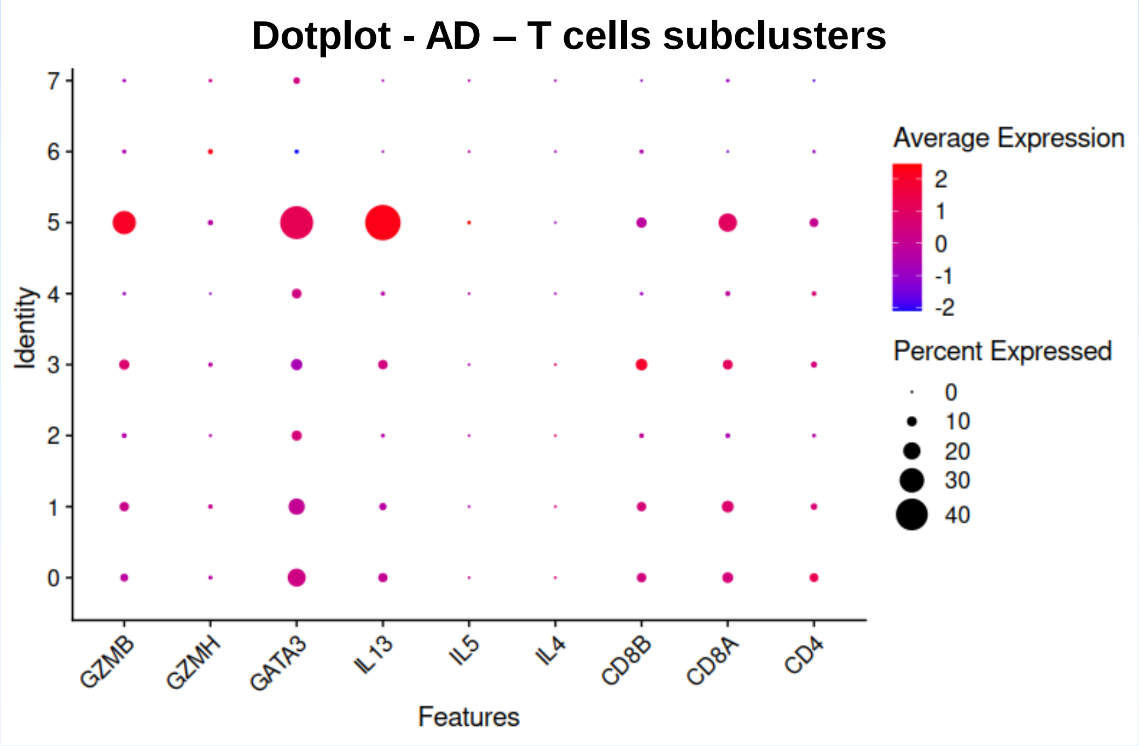

Identifying T Cells in AD Dataset – CD3D Expression

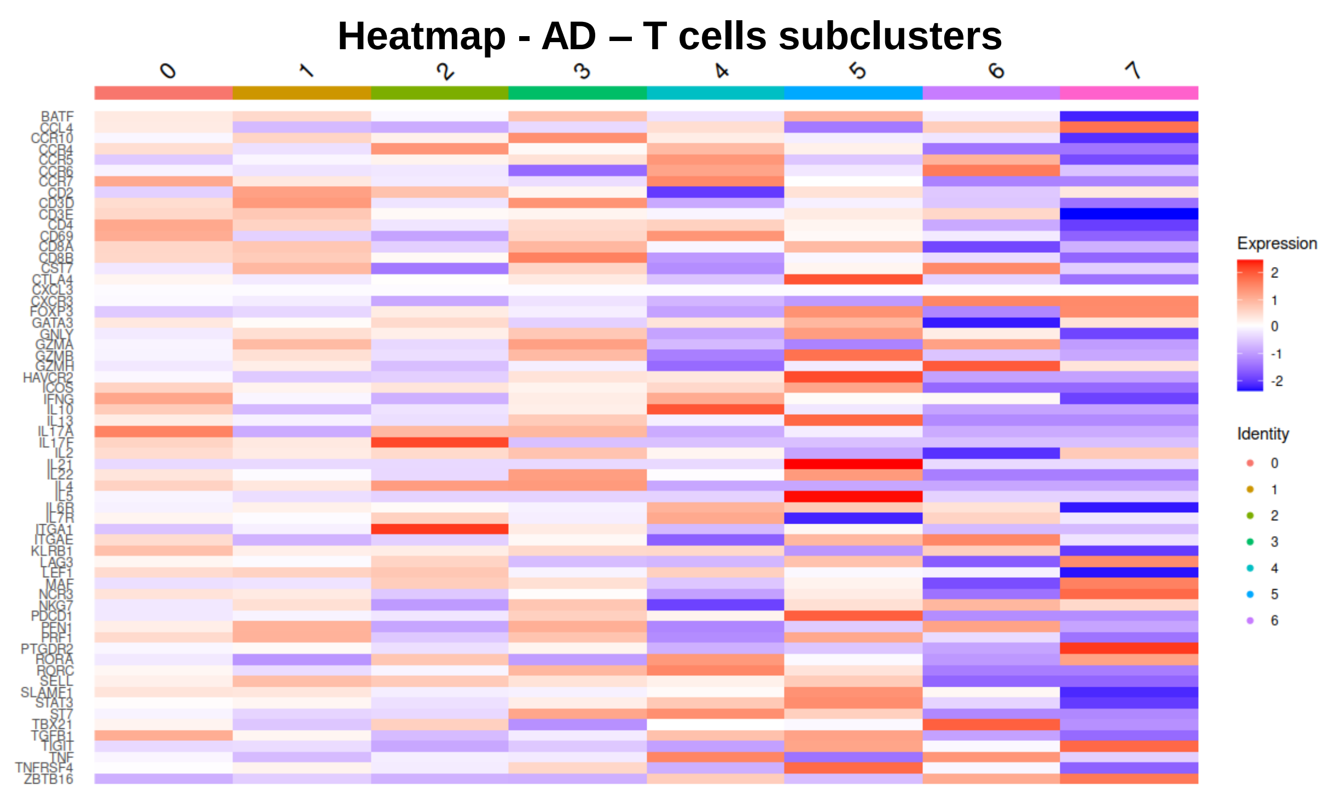
Results

- In the AD data, there were two T cell clusters that showed notable expansion in lesional atopic dermatitis samples as compared to healthy samples.


AD – T cell subcluster proportions by tissue type

- Both clusters had increased expression of *CD8*, type 2 cytokines, and cytotoxic molecules, although each cluster had a slightly different profile.


Dotplot - AD – T cells subclusters


Heatmap - AD – T cells subclusters

Results (continued)

- In the SSc data, the cluster with the highest expression of *CD8* was found in similar percentages in both controls and in patients with SSc. This cluster expressed relatively high levels of cytotoxic molecules such as *GZMB* but not type 2 cytokines such as *IL13*. Also in the SSc dataset, a T cell cluster with comparatively high levels of *CD4* and the type 2 response master regulator *GATA3* modestly expanded in SSc as compared to healthy controls.

Limitations

- This was a limited analysis given the following:
  - Small sample sizes in the AD data
  - Inherent differences between the atopic dermatitis and SSc experiments/datasets
  - Due to the differences between the SSc and AD datasets, the two datasets were analyzed separately in R, therefore allowing for only a qualitative comparison of the two diseases.

Conclusions

- Based on the above limited analysis, lesional CD8+ T cells in AD appear somewhat different than those in SSc.
- In comparison to AD, CD8+ T cells with *IL13* expression in SSc skin appear to be less frequent. However, given the above limitations, further research will be needed to see if this conclusion holds in future studies.

References

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