High-Resolution Single-Cell RNA-seq Autoimmune Atlas for Cellular Characterization and Target Discovery in Rheumatoid Arthritis and Systemic Lupus Erythematosus

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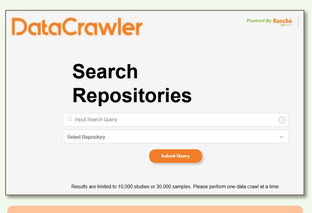
Background

Autoimmune diseases such as Rheumatoid Arthritis (RA) and Systemic Lupus Erythematosus (SLE) exhibit profound cellular heterogeneity across patients and tissues, complicating mechanistic insight and target discovery. In collaboration between **Rancho Biosciences** and **Nucleome Therapeutics**, we built a high-resolution single-cell RNA-seq (scRNA-seq) atlas that unifies curated Single Cell Data Science (**SCDS**) Consortium datasets with newly processed samples to enable consistent, cross-tissue comparisons in RA and SLE.

- **Scale and coverage:** The Autoimmune Atlas integrates **6 datasets** across **RA/SLE pathological states** (including **controls**), totaling **1 million cells** from **218 donors** and **290 samples**, spanning **synovial tissue, skin,** and **peripheral blood**.
- **Source datasets:** Accelerated development combines off-the-shelf analysis-ready datasets with newly re-analyzed datasets under a unified curation and processing strategy.
- **Annotation depth:** Employs a high-resolution cell-annotation procedure to resolve immune and stromal populations, with particular focus in **dendritic cells, memory B cells,** and **regulatory T cells** - cell types central to autoimmune pathogenesis.
- **Biological insight:** Reveals **shared** and **tissue-specific** transcriptional signatures across RA and SLE, supporting mechanism-of-disease hypotheses and prioritization of candidate targets.

1. Data tracking and collection

Query DataCrawler → Crawl Repositories → Annotate Datasets → Select “in scope” Datasets



DataCrawler

GEO, SRA, Synapse, dbGAP repositories

Single-cell and single-nuclei datasets

Catalog of 6,600 datasets

Client examines and picks datasets

id	shortname	title	authors	journal	pmid	abstract	organism	tissue	disease	number_cell	technique	repo_identifi	processed_d	in_scope
1	Absinta_2021_Nat	A lymphocyte	Martina Absi	Nature	34497421	Multiple sclei	Human	brain	Nervous Syst	66432	Chromium	GSE180759	TXT;CSV	in scope
2	Fernandes_2020_C	Single-Cell Tr	Hugo J.R. Fer	Cell Rep	33053338	The advent of	Human	mature B cell		15325	Chromium	E-MTAB-9154	ZIP;TXT	in scope
3	Pappalardo_2020_	Transcriptom	Jenna L. Pap	Sci Immunol	32948672	T cells provid	Human	brain;spine;t	Nervous Syst	6000	Chromium	GSE157277	CSV	in scope
4	Agarwal_2020_Nat	A single-cell	Devika Agarw	Nat Commun	32826893	We describe	Human	brain		18120	Chromium	GSE140231	TAR	in scope
5	Schafflick_2020_N	Integrated sir	David Schaffl	Nat Commun	31937773	Cerebrospina	Human	anatomical p		65326	Chromium	GSE138266	TSV;MTX	in scope
6	Schirmer_2019_Na	Neuronal vul	Lucas Schirm	Nature	31316211	Multiple sclei	Human	brain	Nervous Syst	48919	Chromium	PRJNA544731		in scope
7	Hong_2021_Front_I	Single-Cell Rl	Xiaoping Hor	Front Immun	33603736	Objective: Pri	Human	blood	Musculoskel	57288	Chromium	GSE157278	TSV;MTX	in scope
8	Caron_2020_Sci_Re	Single-cell ar	Maxime Caro	Sci Rep	32415257	Childhood ac	Human	bone marrow		39375	Chromium	GSE132509	MTX;TSV	in scope
9	Alivernini_2020_Na	Distinct syno	Stefano Alive	Nat Med	32601335	Immune-regu	Human	type B synovi	Musculoskel		Chromium	E-MTAB-8322	ZIP	in scope
10	Kim_2020_Nat_Mec	Targeted ther	Doyoung Kim	Nat Med	31959990	Drug-induce	Human	blood;skin	Mental Disor	54476	Chromium	GSE132802	H5	in scope
11	Aganezov_2021_Ge	Comprehensi	S. Aganezov,	Genome Res	32887686	Improved ide	Human		Neoplasms		Chromium	PRJNA476239		in scope
12	Bondoc_2021_Com	Identificatior	Alexander Bo	Commun Bio	34497364	Hepatoblastc	Human	liver;tumor	Neoplasms	26886	Chromium	GSE180665	XLSX;TAR;H5/	in scope
13	Sahu_2021_Oncog	Human cell tr	Biswajyoti Sa	Oncogene	34302118	Cancer is the	Human	mature B cell	Neoplasms	3500	Chromium	PRJEB31262		in scope
14	Jansky_2021_Nat_C	Single-cell tra	Selina Jansky	Nat Genet	33767450	Neuroblastor	Human	adrenal glan	Neoplasms		Chromium	GSE163431	TSV	in scope
15	Mercatelli_2021_Bi	Single-Cell Gr	Daniele Merc	Biomolecules	33525507	Neuroblastor	Human	mature B cell	Neoplasms	2067	Chromium	PRJNA668226		in scope
16	Luoma_2020_Cell	Molecular Pa	Adrienne M. I	Cell	32603654	Checkpoint b	Human	colon	Digestive Sys	51652	Chromium	GSE144469	TAR;CSV	in scope
17	Ji_2020_Cell	Multimodal A	Andrew L. Ji,	Cell	32579974	To define the	Human	integument	Neoplasms	50009	Chromium	GSE144240	JPG;TSV	in scope
18	Lu_2020_Cell	Complement	Yiwen Lu, Qi	Cell	32142650	Understandir	Human	regulation of	Neoplasms	998	Chromium	GSE135710	MTX;TSV	in scope
19	Laugney_2020_Nat	Regenerative	Ashley M. Lal	Nat Med	32042191	Development	Human	lung;tumor;n	Neoplasms	40505	Chromium	GSE123904	CSV	in scope
4850	Gideon_2022_Imm	Multimodal p	Gideon,Hann	Immunity	35483355	Mycobacteri	Monkey	granulocyte	Infections		Seq-Well	GSE200151		in scope
20	Rao_2020_Cold_Sp	Comparative	Rao M, Oh K,	Cold Spring H	32054662	Genomic ana	Human	breast	Neoplasms		Chromium	GSE140312		in scope
21	Wang_2020_Stem_(	Adult Human	Rong Wang, F	Stem Cell Rep	32004492	Radial glia (F	Human	brain;tumor	Neoplasms	12367	Chromium	GSE139448	CSV	in scope
22	Rendeiro_2020_Na	Chromatin m	André F. Ren	Nat Commun	31996669	The Bruton ty	Human		Neoplasms	43000	Chromium	GSE111015	TAR;MTX;TSV	in scope
4865	Hay_2023_Cancer_t	Low TCR Bind	Hay Zachary	Cancer Immu	36787375	T-cell recepto	Mouse	lymphocyte	Neoplasms		Chromium	GSE212980		in scope
23	Cillo_2020_Immun	Immune Lanc	Anthony R. Ci	Immunity	31924475	Head and nec	Human	neck	Neoplasms	131224	Chromium	GSE139324	MTX;TSV	in scope
24	Yost_2019_Nat_Me	Clonal replac	Kathryn E. Yo	Nat Med	31359002	Immunother	Human	regulation of	Neoplasms	79046	Chromium	GSE123813	TXT	in scope
4875	Hsu_2023_Inflamm	Responsive n	Hsu Paul, Ch	Inflamm Bow	37235748	Background:	Human	blood	Digestive Sys		CITE-Seq	GSE228165		in scope
25	Merino_2019_Nat_(	Barcoding re	D. Merino, T.	Nat Commun	30770823	Primary triple	Human		Neoplasms	25000	Chromium	GSE123926	MTX;TSV	in scope
26	Ben-David_2018_N	Genetic and t	Uri Ben-Davic	Nature	30089904	Human cance	Human	mature B cell	Neoplasms	26465	Chromium	GSE114462	CSV	in scope
27	Azizi_2018_Cell	Single-Cell M	Elham Azizi, /	Cell	29961579	Knowledge o	Human	regulation of	Neoplasms	72000	Chromium	GSE114727	CSV	in scope
28	Rodriguez_2022_Sc	Preneoplastic	Sara Rodrigu	Sci Adv	35044826	Normal cell c	Human	bone marrow	Neoplasms,	40169	Chromium	GSE185158	RDATA	in scope
29	Li_2022_J_Clin_Inve	Cardiomyocy	Shen Li, Tom	J Clin Invest	34813507	Various popu	Mouse	fibroblast of	Cardiovascul		Chromium	GSE185060	CSV	in scope
30	Guo_2022_JCI_Insig	Resolving the	Wei Guo, Cui	JCI Insight	34793335	Colorectal ca	Mouse	colon	Neoplasms	27927	Chromium	GSE188711	MTX;TSV	in scope

To assemble a catalog of RA and SLE studies, we surveyed expression repositories, including GEO, SRA, Synapse, and dbGaP, for single-cell or single-nuclei datasets. Our catalog includes almost 8,000 publications from the last 10 years and is annotated for species, disease, tissue, and assay types.
Rancho have processed > 1000 datasets through Single Cell Data Science Consortium (SCDS), a member-led initiative to create resources which advance therapeutic discovery. After 1 year, SCDS datasets are commercialized and available as a high-quality product that enhances our client's research.

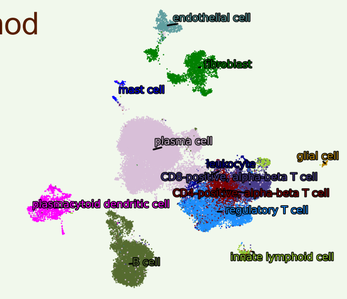
2. QC, data ingestion, and curated metadata import

- Rancho's **QC** and **ingestion pipeline** was applied to ensure high-quality data while preserving the underlying biological signal.
- Our data integrity team collaborated with study authors to resolve discrepancies, link donor information with sample identifiers, and enrich metadata for downstream use.
- Any non-SCDS datasets were processed using the same principles and standardized procedures, ensuring consistency across the entire atlas.

3. Automated cell-type annotation workflow

CZ CELL×GENE · scGPT · Azimuth · CellTypist · scArches

For Cell-type annotation we used a comprehensive, multi-method strategy leveraging CELLxGENE Census reference data.



- Transfer/label-transfer approaches (scArches, Azimuth)
- Supervised classification (CellTypist)
- Representation learning (scGPT)
- Reference-backed annotation (CELLxGENE Census)

Predictions were reconciled into **consensus labels** and validated against canonical marker-gene signatures with targeted manual review. Cell-type categories were further organized into parent classes to enable intuitive browsing and cross-dataset comparisons.

4. Data Integration and atlas creation

Integration & Feature Selection

- Integration model: scVI (variational autoencoder; scvi-tools) used as the batch-correction/latent-representation method of choice.
- Tissue-aware markers: Curated canonical cell-type markers for synovium, skin, and peripheral blood from CellMarker 2.0.
- Variable features: Aggregated union of highly variable genes (HVGs) across datasets (a gene counted if HVG in ≥1 dataset).
- Final integration genes: Intersection of the marker panel and the HVG union → ~2,000 genes used to train scVI.

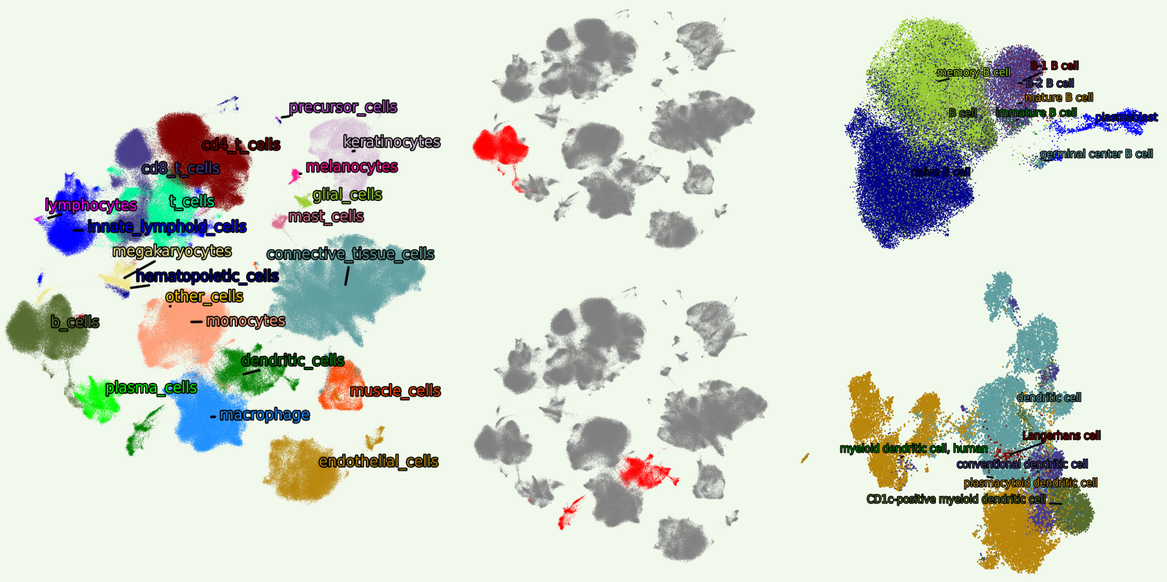
Latent-Space QC & Refinement

- Neighborhood-concordance filtering: In the scVI latent space, a cell is retained if ≥55% of its neighbors share the same parent cell class (reduces ambiguous/outlier assignments).
- Minimum category size: Parent cell classes with <40 cells (total) were removed to stabilize downstream analyses.

Clustering & Visualization

- Graph-based clustering and UMAP dimensionality reduction were performed on the scVI-corrected embedding.

5. Atlas-level Queries: Zooming into Biological Signal



- Our robust annotation workflow enables high-confidence cell identities across tissues and studies, supporting hierarchical exploration from major lineages to fine-grained subsets.
- This makes the atlas query-ready: users can “zoom” from refined parent classes to disease-relevant components (B cells, dendritic cells) across RA/SLE and tissues.
- Downstream analyses can resolve intra-cell-type diversity (e.g., memory B-cell states, dendritic-cell subtypes), providing a principled path to interrogate underlying biology and generate hypotheses.

Conclusions

The Nucleome-Rancho Autuimmune atlas uncovers both shared and tissue-specific cellular signatures in RA and SLE, offering a powerful foundation for understanding disease pathogenesis. The atlas scale and resolution creates a unique opportunity to query disease-relevant cell states, identify candidate biomarkers, and guide target discovery in future analyses. Beyond this, the atlas establishes a scalable and reproducible framework for disease-focused single-cell analysis, enabling the construction of reference datasets that support hypothesis generation and biomedical research.

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