

Novel single-cell RNA-sequencing data demonstrates upregulation of angiogenesis-associated gene pathways in a late-stage mouse model of Alzheimer's disease.



Developing a transcriptomic atlas of Alzheimer's disease progression in the Tg2576 mouse model using single-cell RNA-sequencing technology

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BACKGROUND

- Alzheimer's disease (AD) is a debilitating neurodegenerative condition characterized by cognitive decline, progressing to eventual dementia and death.
- Recent discoveries have challenged the traditional "amyloid hypothesis" of AD, warranting new approaches and prospective treatment agents in the AD therapeutic pipeline.
- Angiogenesis, the formation of new blood vessels, is characteristic of early AD. Early evidence suggests that modulating angiogenesis may repair damage in the AD brain, though transcriptomic data is currently lacking.
- AIM: to create the first single-cell view of AD progression in mice in order to identify alternative mechanisms of AD pathophysiology, and reveal putative molecular targets for AD drug therapy.

METHODS

- Single-cell RNA-sequencing (scRNA-seq) enables gene expression profiling from individual cells in a tissue to define the specific contribution of each cell to the function of the whole tissue.
- Tg2576 mice contain a mutation in the Amyloid Precursor Protein (APP) gene which results in a double amino-acid substitution. In Tg2576 mice, expression of mutated APP reliably causes overexpression of human APP in neurons, resulting in AD pathophysiology.

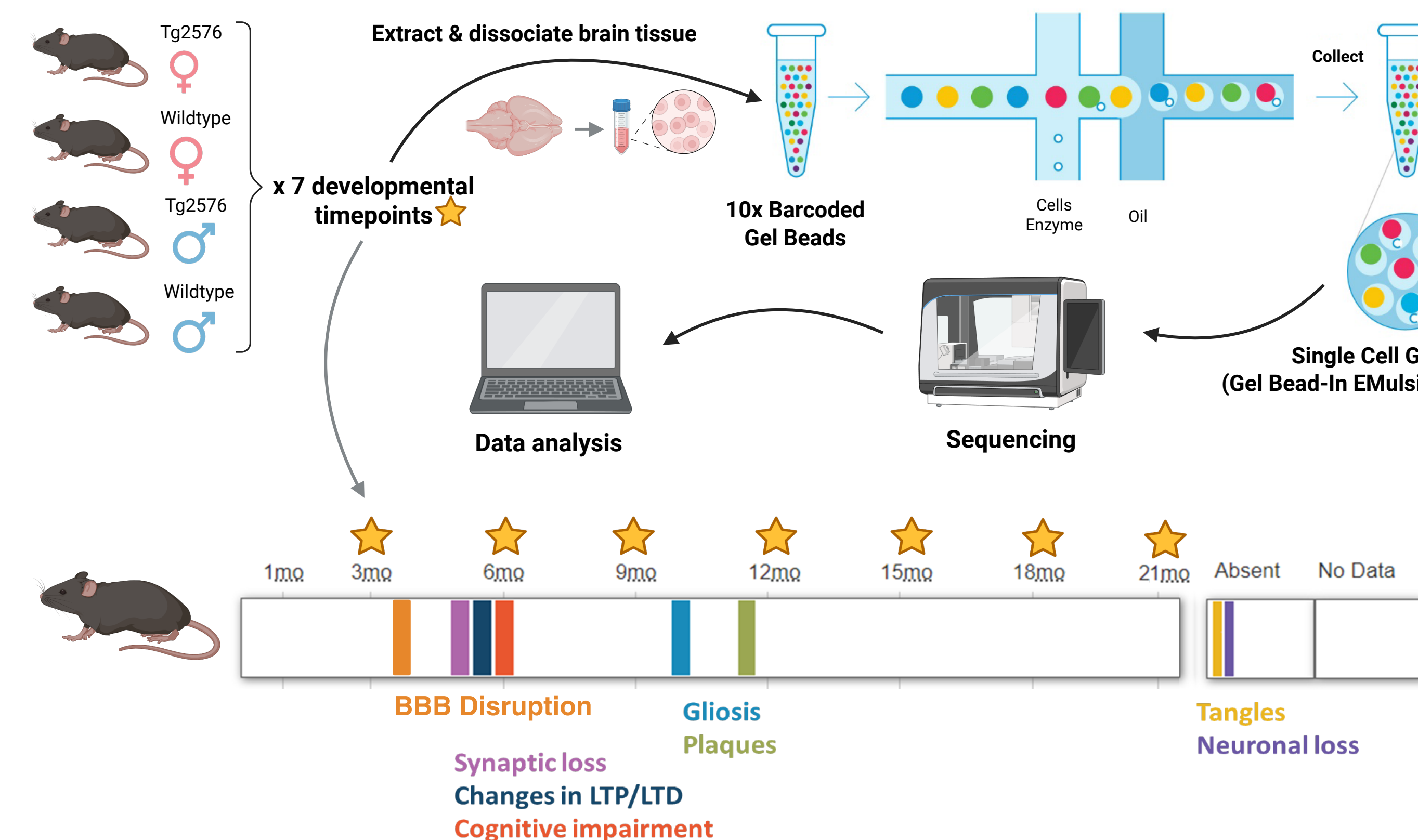


Figure 1: Experimental methods diagram.

Cohorts of four mice representing both AD and control genotypes (Tg2576 and wildtype) and both sexes (male and female) were humanely euthanized at seven different disease progression timepoints (N=28). Brains were dissected and dissociated into a single-cell suspension. Individual cells were captured, and cellular mRNA was sequenced and profiled at a single cell resolution. Single-cell expression data was analyzed using 10X Genomics software. Created with BioRender.com

PRELIMINARY RESULTS

- Pathway analysis demonstrated statistically significant enrichment in pathways related to: increased angiogenesis ($p = 0.0337$), vasculature development ($p = 0.0371$), and amyloid-beta ($A\beta$) formation ($p = 0.0149$).

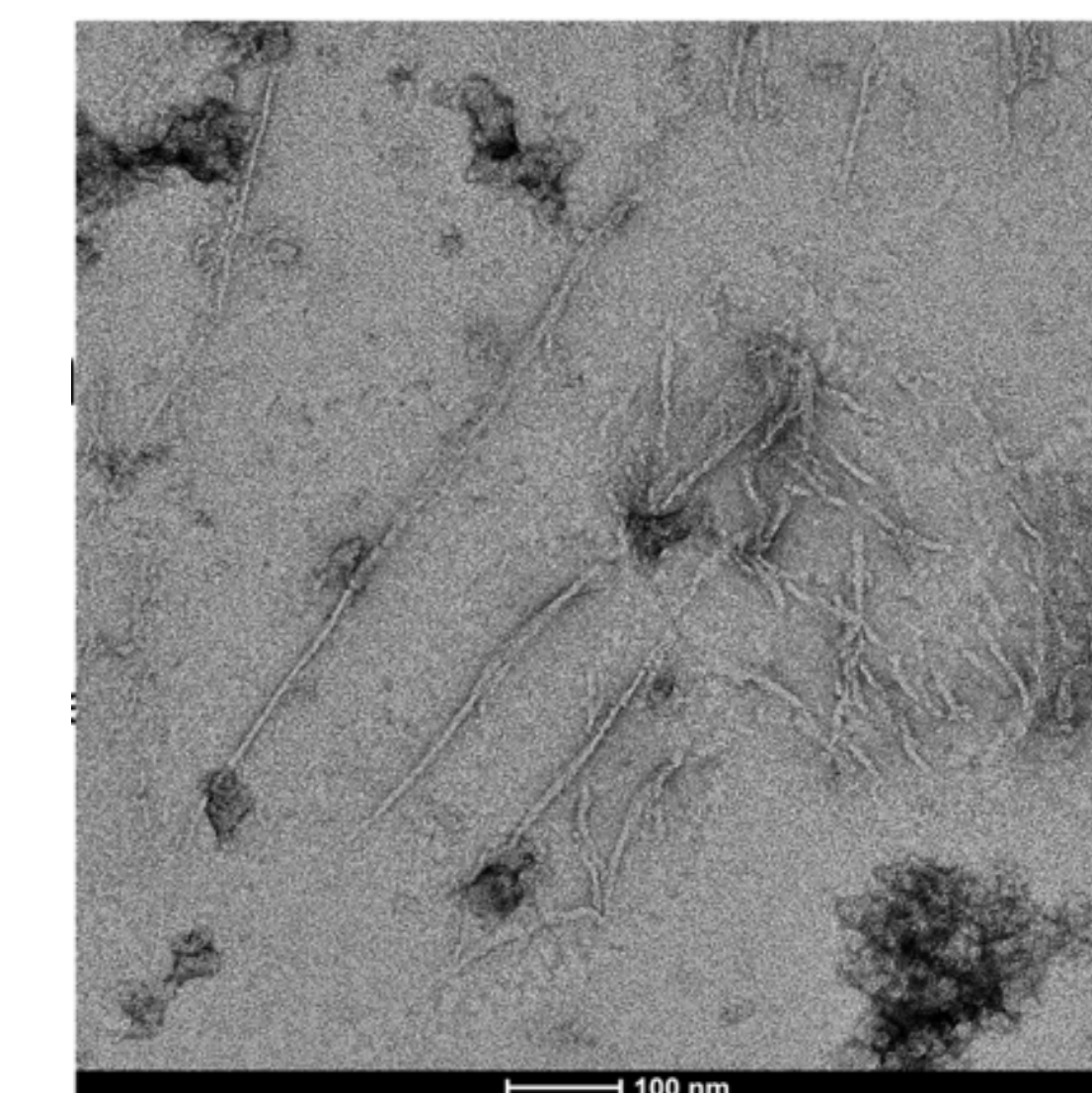


Figure 2. Negatively stained transmission electron micrograph of amyloid fibrils.

Representative microscopy visualizing characteristic amyloid fibrils in brain tissue from the Tg2576 mouse model of AD. Image source: Dr. Razieh Kamali-Jamil, unpublished

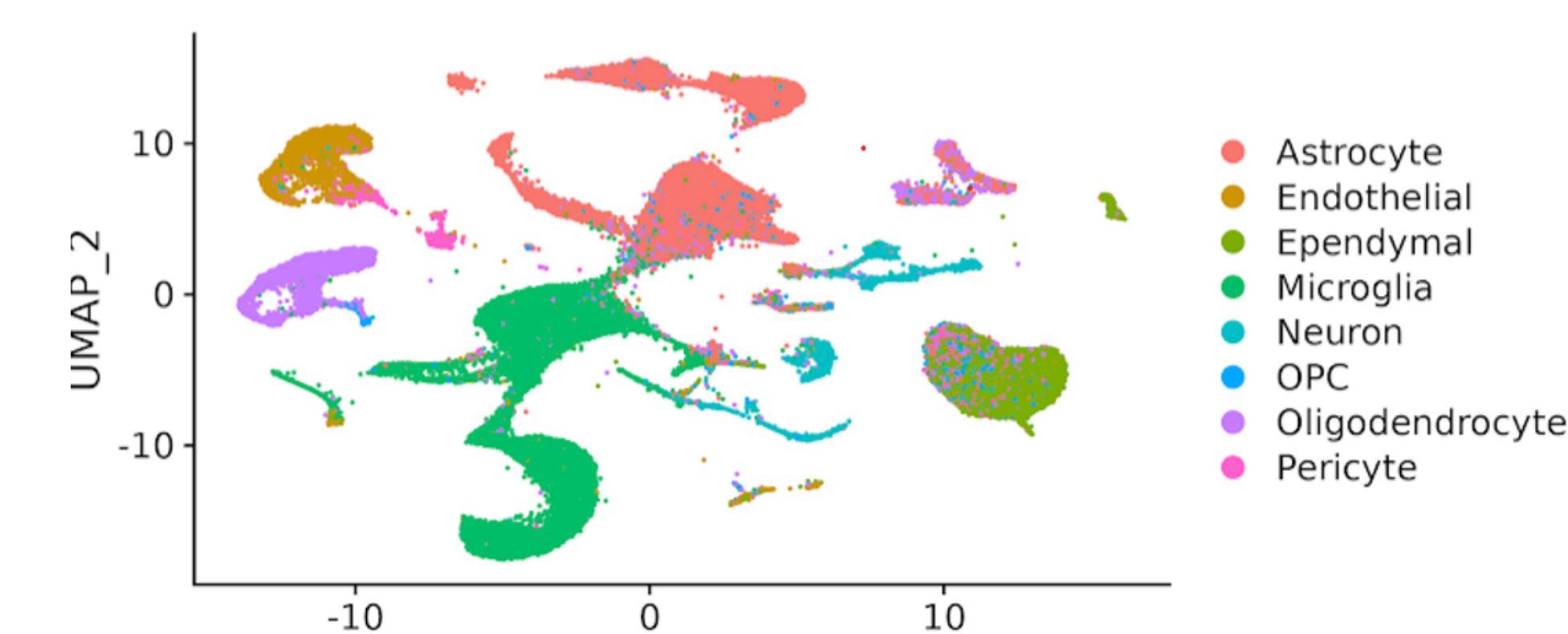


Figure 3: Cluster diagram for merged AD and wt samples. During the data analysis process, clusters are formed by grouping cells with similar transcriptomic profiles. (N=28)

CONCLUSION

- This study offers a unique cellular-level view of transcriptional alterations associated with AD pathology in mouse and human tissue. Our methods will capture:
 - Cell-type-specific and shared gene-expression perturbations
 - Disease-associated cellular subpopulations
 - Sex-biased transcriptional responses.
- Preliminary findings support the neurovascular theory of AD
 - Supports growing evidence that neoangiogenesis-induced disruptions to blood-brain-barrier integrity lead to the increase in $A\beta$ accumulation and subsequent AD pathology seen in Tg2576 mice and AD patients.
- Next steps:
 - Long-read and deep sequencing on mouse brain samples to identify precise splice variants differentially expressed in AD cells.
 - Knock-out studies in rodent and neural organoid models of AD to probe differentially-expressed genes of interest.
 - Preclinical drug repurposing of established medications with angiogenesis-modifying properties for treatment and prevention of AD using scRNA-seq as a readout/endpoint.