Leveraging single-cell ATAC-seq to identify disease-critical fetal and adult brain cell types

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Alkes Price Group
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Kim et al. bioRxiv 2021
Disclosure Slide

Financial Disclosure for:
Samuel Kim

I have nothing to disclose
Outline

• Motivation

• Methods

• Results: disease-critical cell types using fetal brain data

• Results: disease-critical cell types using adult brain data
Outline

✓ Motivation

• Methods

• Results: disease-critical cell types using fetal brain data

• Results: disease-critical cell types using adult brain data
Disease-critical cell types $\rightarrow$ disease mechanisms

(Figure from Hekselman & Yeger-Lotem. Nat Rev Genet 2020)
Disease-critical cell types → disease mechanisms

<table>
<thead>
<tr>
<th>Mechanism</th>
<th>Affected tissue</th>
<th>Unaffected tissue</th>
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<tbody>
<tr>
<td><strong>a Expression-based mechanisms</strong></td>
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<tr>
<td>① Exclusive expression</td>
<td>![Image]</td>
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<tr>
<td>② Preferential expression</td>
<td>![Image]</td>
<td>![Image]</td>
</tr>
<tr>
<td><strong>b Regulatory mechanisms</strong></td>
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<tr>
<td>① Disrupted regulatory elements</td>
<td>![Image]</td>
<td>![Image]</td>
</tr>
<tr>
<td>② Effects of eQTLs</td>
<td>![Image]</td>
<td>![Image]</td>
</tr>
</tbody>
</table>

(Figure from Hekselman & Yeger-Lotem. Nat Rev Genet 2020, shown in part)
Identifying disease-critical cell types, leveraging the emergence of single-cell profiling of diverse cell types

**A human cell atlas of fetal gene expression**


Leveraging mouse chromatin data for heritability enrichment informs common disease architecture and reveals cortical layer contributions to schizophrenia

Paul W. Hook¹ and Andrew S. McCallion¹,2,3

**Scaling single-cell genomics from phenomenology to mechanism**

Amos Tanay & Aviv Regev

**HUMAN GENOMICS**

**A human cell atlas of fetal chromatin accessibility**


**Single-cell genomics identifies cell type-specific molecular changes in autism**

Dmitry Velmeshev¹,², Lucas Schirmer¹,²,⁴, Diane Jung¹,², Maximilian Haeussler², Yonatan Perez¹,², Simone Mayer¹,²,⁴, Aparna Bhaduri¹,²,³, Nitasha Goyal¹,²,⁷, David H. Rowitch¹,³,⁸,⁹, Arnold R. Kriegstein¹,²

**Single-cell epigenomic analyses implicate candidate causal variants at inherited risk loci for Alzheimer’s and Parkinson’s diseases**

M. Ryan Corces²,³, Anna Shcherbina²,³, Soumya Kundu⁴,⁵, Michael J. Gloudemans⁴,³, Laure Frésard¹, Jeffrey M. Granja⁴,⁵, Bryan H. Louie¹,³, Tiffany Eulalio³, Shadi Shams²,⁴, S. Tansu Bagdatli²,⁴, Maxwell R. Mumbach²,⁴, Boxiang Liu⁵,⁶, Kathleen S. Montine¹, William J. Greenleaf²,⁴,⁶,⁹, Anshul Kundaje⁴,³, Stephen B. Montgomery⁴,³, Howard Y. Chang²,⁴,¹⁰,¹¹ and Thomas J. Montine¹,¹²
Our goals

1. To identify disease-critical cell types using scATAC-seq data (and compare with scRNA-seq data)
Our goals

1. To identify disease-critical cell types using scATAC-seq data (and compare with scRNA-seq data)

2. To assess the impact on disease risk of cell types in different developmental stages of the brain (by comparing fetal vs. adult brain cell-type)
Outline

• Motivation

✓ Methods

• Results: disease-critical cell types using fetal brain data
• Results: disease-critical cell types using adult brain data
Analyzed **28 brain-related traits** (avg. N = 298K)

### 28 Brain traits
- Major depressive disorder
- Ischemic stroke
- Anorexia
- Alzheimer’s disease
- Autism spectrum disorder
- Schizophrenia
- Bipolar disorder
- ADHD
- Schizophrenia vs. Bipolar
- BMI
- Years of education
- Smoking status
- Intelligence
- Neuroticism
- Morning person
- Age at menarche
- Worry
- Reaction time
- # Children ever born
- Smoking initiation
- Sleep duration
- General risk tolerance
- Insomnia
- Drinks per week
- Medication use
- Cigarettes per day
- Smoking cessation
- Age of drinking initiation

### 6 Control traits
- Coronary artery disease
- Bone mineral density
- Rheumatoid arthritis
- Type 2 diabetes
- Sunburn occasion
- Breast cancer
Analyzed 4 single-cell atlases of human brain

**Fetal**

**A human cell atlas of fetal gene expression**


N (donors) = 28; 2M cells; 34 brain cell types

**HUMAN GENOMICS**

**A human cell atlas of fetal chromatin accessibility**


N = 26; 720k cells; 14 brain cell types

**Adult**

**Single-cell genomics identifies cell type-specific molecular changes in autism**

Dmitry Velmeshev2,3, Lucas Schirrer3,4,4, Diane Jung1,3, Maximilian Haussler5, Yonatan Perez2,4, Simone Mayer2,4, Aparna Bhaduri1,4, Nitasha Goyal1,3,7, David H. Rowitch1,4,8,9, Arnold R. Kriegstein1,3,7

N = 31; 104K cells; 17 brain cell types

**Single-cell epigenomic analyses implicate candidate causal variants at inherited risk loci for Alzheimer’s and Parkinson’s diseases**

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N = 10; 70K cells; 18 brain cell types
Overview of methods: building and assessing cell-type annotations

Specifically expressed genes

<table>
<thead>
<tr>
<th>Genes</th>
<th>FDR for cell type A</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gene_1</td>
<td>0.02</td>
</tr>
<tr>
<td>...</td>
<td>...</td>
</tr>
<tr>
<td>Gene_{20k}</td>
<td>0.005</td>
</tr>
</tbody>
</table>

scRNA-seq cell-type annotations

- Brain enhancer-gene links (Roadmap U ABC)

Chromatin accessible regions

<table>
<thead>
<tr>
<th>Peak regions</th>
<th>FDR for cell type A</th>
</tr>
</thead>
<tbody>
<tr>
<td>Region_1</td>
<td>0.01</td>
</tr>
<tr>
<td>...</td>
<td>...</td>
</tr>
<tr>
<td>Region_N</td>
<td>0.02</td>
</tr>
</tbody>
</table>

scATAC-seq cell-type annotations

- SNPs under accessible peaks

S-LDSC: Disease heritability enrichment for disease X, conditioning on other annotations

To evaluate disease heritability enrichment, applied stratified LD score regression (**S-LDSC**)

**Summary statistics**

**Reference panel**

**Cell-type annotations**

**Input**

1. **Enrichment** = Prop. $h_2g$ / Prop. SNPs
2. **Standardized effect size** ($\tau^*$) = $M\tau \cdot \text{csd}(c)$ / $h_2g$

That is, proportionate change in per-SNP heritability associated to a one $sd(annot)$ increase, conditional on 53 annotations (baseline model) and background annotation.

(Finucane et al. Nat. Genet. 2015)
Assessed $\tau^*$ after additionally conditioning on background annotation.

- Additionally conditioned on the background annotation as a conservative metric to ensure cell-type specificity.
  - scATAC-seq data: union of per-dataset open chromatin regions.
  - scRNA-seq data: union of brain enhancer-gene links across all genes analyzed.

(Finucane et al. Nat. Genet. 2015)
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• Motivation

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✔ Results: disease-critical cell types using fetal brain data

• Results: disease-critical cell types using adult brain data
Identified more significant disease-cell type pairs from scATAC-seq*

For 13 cell types appearing in both data

<table>
<thead>
<tr>
<th></th>
<th>Fetal scATAC</th>
<th>Fetal scRNA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brain cell types</td>
<td>14</td>
<td>34</td>
</tr>
<tr>
<td>Total disease-cell type pairs</td>
<td>392</td>
<td>952</td>
</tr>
<tr>
<td>Significant disease-cell-type pairs</td>
<td>152</td>
<td>9</td>
</tr>
<tr>
<td>Significant diseases (out of 28)</td>
<td>22</td>
<td>8</td>
</tr>
</tbody>
</table>

- Identified no significant enrichments for control traits

*for the datasets we have analyzed
Confirming associations: SCZ/MDD/ADHD – excitatory neurons

(Finucane et al. NG 2018, Bryois et al. NG 2020, Corces et al. NG 2020; Moriguchi et al. Mol. Psychiatry 2019)
Interesting associations: insomnia - photoreceptor cells

Interesting associations: insomnia - photoreceptor cells

Photoreceptor cells convert light into signals to the brain and thus play an essential role in circadian rhythms.

Interesting associations: BMI - ganglion cells

Interesting associations: BMI - ganglion cells

- Ganglion cells relay information from bipolar and amacrine cells to the brain
- Patents with morbid obesity display significant differences in retinal ganglion cells

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✓ Results: disease-critical cell types using adult brain data
Identified more significant disease-cell type pairs from scATAC-seq*

<table>
<thead>
<tr>
<th></th>
<th>Adult scATAC</th>
<th>Adult scRNA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brain cell types</td>
<td>18</td>
<td>17</td>
</tr>
<tr>
<td>Total disease-cell type pairs</td>
<td>504</td>
<td>476</td>
</tr>
<tr>
<td>Significant disease-cell-type pairs</td>
<td>168</td>
<td>64</td>
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<tr>
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- Identified no significant enrichments for control traits

*for the datasets we have analyzed
Confirming associations: SCZ/BP – excitatory neurons

(Finucane et al. NG 2018, Hook et al. GR 2020, Corces et al. NG 2020, Ziffra et al. biorxiv 2020)
Interesting associations: SCZ/MDD - BDNF excitatory neurons

Interesting associations: SCZ/MDD - BDNF excitatory neurons

<table>
<thead>
<tr>
<th>Adult brain scATAC-seq</th>
<th>Adult brain scRNA-seq</th>
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<tbody>
<tr>
<td>ADHD</td>
<td></td>
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<tr>
<td>SCZ</td>
<td>! * * * *</td>
</tr>
<tr>
<td>Bipolar disorder</td>
<td>! * * * *</td>
</tr>
<tr>
<td>SCZ vs bipolar</td>
<td>! * *</td>
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<tr>
<td>MDD</td>
<td>! * *</td>
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<tr>
<td>Insomnia</td>
<td>! * !</td>
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<tr>
<td>Reaction time</td>
<td>! * ! * ! !</td>
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<tr>
<td>Age at menarche</td>
<td>! * * ! * !</td>
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<tr>
<td>BMI</td>
<td>! * * * ! *</td>
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<tr>
<td>Intelligence</td>
<td>! * * * *</td>
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</tbody>
</table>

- Involved in supporting survival of existing neurons and differentiating new neurons
- Decreased BDNF levels have been observed in untreated MDD, BP, and SCZ cases

Interesting associations: Bipolar/SCZ – parvalbumin interneurons

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Interesting associations: Bipolar/SCZ – parvalbumin interneurons

**• Decreased expression and diminished function of parvalbumin interneurons in regulating balance of E/I have been observed in BP and SCZ cases**

### Table: Adult brain scATAC-seq

<table>
<thead>
<tr>
<th>Condition</th>
<th>ADC</th>
<th>SCZ</th>
<th>Bipolar disorder</th>
<th>SCZ vs bipolar</th>
<th>MDD</th>
<th>Insomnia</th>
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<td>somatostatin interneurons</td>
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<td>Inhibitory neurons</td>
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<td>VGLUT2 excitatory spiny neurons</td>
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<td>Dopaminergic neurons</td>
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### Table: Adult brain scRNA-seq

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</table>

Interesting associations: intelligence – corticofugal projection neuron

Interesting associations: intelligence – corticofugal projection neuron

• CPN connects neocortex and the subcortical regions and transmits axons from the cortex
• Imbalance in neuronal activity has been hypothesized to lead to deficits in learning

Conclusions

• Identified significant trait-cell type pairs that can confirm previously associated critical cell types for disease and suggest distinct associations

• Determined that cell-type annotations derived from scATAC-seq were particularly powerful in the data that we analyzed

• Highlight the benefits of analyzing data from different sequencing platforms and different developmental stages to identify disease-critical cell types
Thank you!

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Twitter: samsungilkim
LinkedIn: samuel-s-kim

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