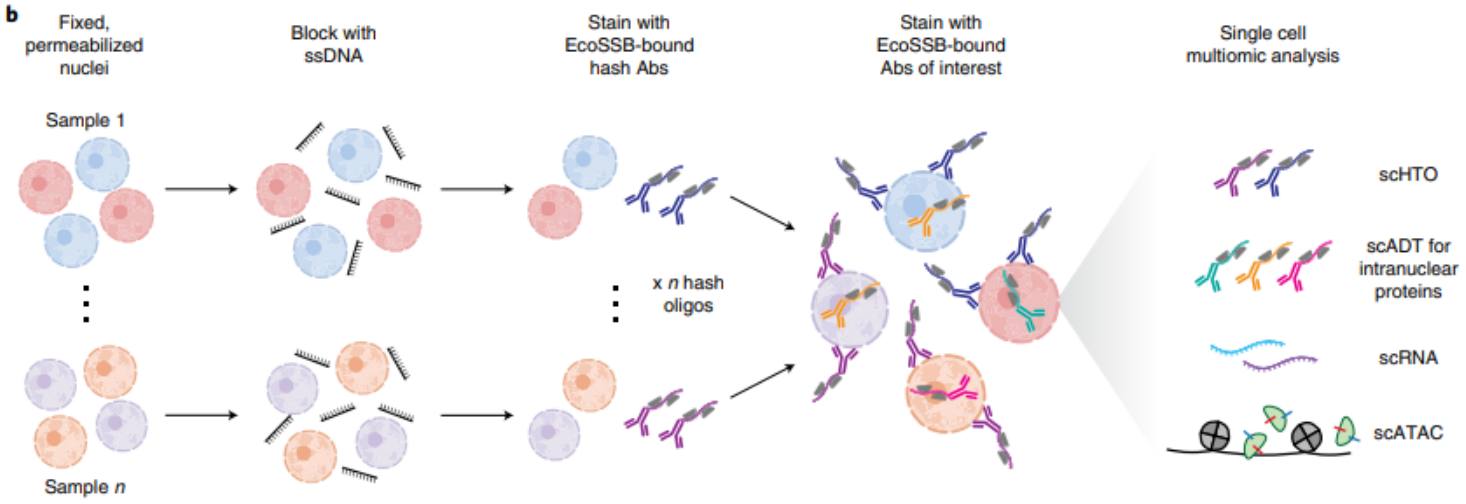
Single Cell Transcriptomics

Newsletter May 2022

Paper 1

Chen, AF *et al.* [NEAT-seq: simultaneous profiling of intra-nuclear proteins, chromatin accessibility and gene expression in single cells], *Nature Methods*, 2022

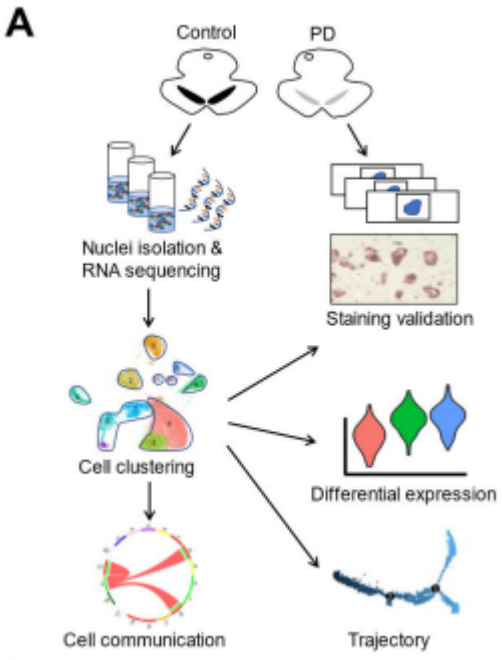
This month's first paper regards a novel method, sequencing of nuclear protein epitope abundance, chromatin accessibility and the transcriptome in single cells (NEAT-seq). NEAT-seq allows for investigation of regulatory elements, in this case transcription factors (TFs), through integration of data on transcription, translation and regulation of chromatin binding. Specifically, the authors use an *E. coli* ssDNA binding protein to decrease the signal from nonspecific binding of oligo-antibody complexes in the nucleus. As an example, they use NEAT-seq to profile primary human CD4 memory T cells composed of distinct T cell subsets driven by known master TFs. Doing so, they identify global changes in GATA3 motif accessibility consistent with posttranscriptional regulatory mechanisms restricting GATA3 protein expression in memory T cells, which could only be uncovered with the addition of protein quantification incorporated in NEAT-seq.



Paper 2

Wang, Q *et al.* [Single-cell transcriptomic atlas of the human substantia nigra in Parkinson's disease](), *bioRxiv*, 2022

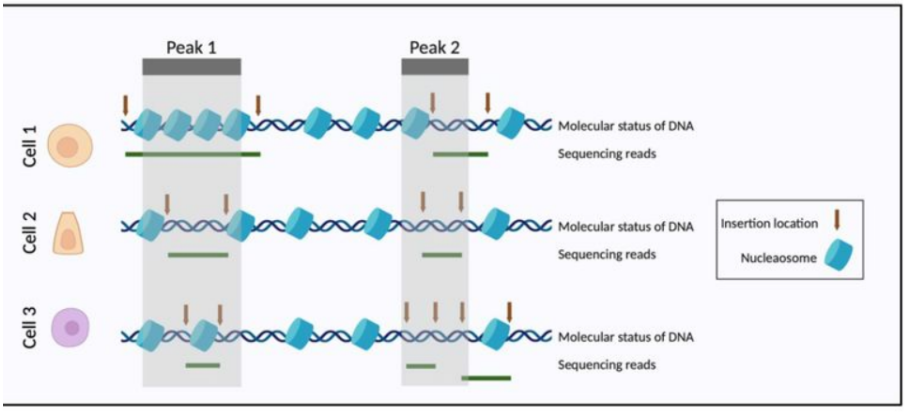
The second paper is a preprint on snRNA-seq of the substantia nigra in Parkinson's disease (PD) patients and healthy controls. Impressively, using 32 samples the authors were able to quantify transcriptomes for more than 300,000 nuclei from this area of the brain crucial to the development of PD. Through clustering analyses and follow-up immunohistochemical (IHC) validation, the authors identify three dopaminergic neuron subtypes that were all degenerated in the patient group. In contrast, the composition of non-neuronal cell clusters including major glial types showed little change. Specifically, the authors identify *RIT2*, a PD susceptible gene, as a marker for a neuronal cluster in their dataset. Through IHC validation experiments in two independent cohorts, they found neuromelanin-expressing (NM+) neurons of the substantia nigra to be divided by high expression of either *RIT2* or the gene coding for tyrosine hydroxylase (*TH*), an enzyme necessary for dopamine synthesis. Hence, NM+ neurons may lose the ability to produce TH in exchange for high expression of *RIT2*. Subsequently, Wang *et al.* performed trajectory analyses to show how DA neurons exhibit trajectories that correlate with PD progression. All in all, this study provides novel insights into PD pathology.



Paper 3

Miao, Z. & Kim, J. [Is single nucleus ATAC-seq accessibility a qualitative or quantitative measurement?](), *bioRxiv*, 2022

The last paper is a preprint touching upon an interesting discussion regarding snATAC-seq. Existing snATAC-seq analysis methods create chromosomal domain features either by arbitrarily dividing the entire genome into fixed-width segments (bins), or estimating discrete domains by peak-calling (peaks). This is followed by assigning feature counts based on the number of fragments that overlap with a region, or based on the number of insertions within the region. Miao & Kim argue how this is problematic since the configuration of fragment/insertion positions around the peak/bin interval can create different quantifications dependent on whether one uses fragments or insertions (figure below). To overcome this, the authors propose a simple consistent counting strategy called Paired-Insertion-Counting (PIC). With PIC, for a given chromosome interval, if an ATAC-seq fragment's pair of insertions are both within the interval, counted as one (pair); if only one insertion is within the interval also count one (pair). In summary, Miao & Kim propose a new counting method that is consistent with the molecular basis of the assays.



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	Fragment-based counting		Insertion-base counting	
	Peak 1	Peak 2	Peak 1	Peak
Cell 1	1	1	0	1
Cell 2	1	1	2	2
Cell 3	1	2	2	3

Next Single Cell Seminar

Date: 30th May 2022, Maersk Tower top floor, 7.15.92

9:00 – 10:00

Jenny Brown, postdoc, CBMR, Pers lab

Brain Glucose Sensing

10:00 – 11:00

Rasmus Rydbirk, postdoc, BRIC, Khodosevich lab

Neurodegeneration of the lentiform nucleus in parkinsonian diseases at the single cell level

If you would like to announce anything single cell related, being it job announcement, event, your published paper, technology development etc., please contact us.

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