Supplemental Tables

Supplemental Table 1. Primers. *Primers used for round 1 and round 2 amplification of viral transcripts. Primers with TC1 and TC2 in the amplicon name indicate they were used only for those samples.*

Amplicon	Read	Round	Sequence (Ns indicate Illumina sample index)
All Viruses	1	1	CTACACGACGCTCTTCCGATCT
All Viruses	1	2	AATGATACGGCGACCACCGAGATCTACACTCTTTCCCTACACGACGCTCTTCCGAT
mNeonGreen TC1	2	1	TTCAAGGAGTGGCAAAAGGCCTTTACCGATGTGAT
mRuby2	2	1	CAACGGGAACATGCAGTTGCCAAGTTTGCTGG
mNeonGreen	2	1	TAACTATCTGAAGAACCAGCCGATGTAC
tdTomato TC2	2	1	AGGACTACACAATTGTCGAACAGTATGAG
tdTomato	2	1	ACAACGAGGACTACACCATCGTGG
mCherry	2	1	CATCGTGGAACAGTACGAACG
WPRE	2	2	CAAGCAGAAGACGGCATACGAGATNNNNNNNNGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCTAGACGAGTCGGATCTCCCT
mNeonGreen	2	2	CAAGCAGAAGACGGCATACGAGATNNNNNNNNGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCTTTCAAGGAGTGGCAAAAGGC
mRuby2	2	2	CAAGCAGAAGACGGCATACGAGATNNNNNNNNGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCTCAACGGGAACATGCAGTTGC
tdTomato TC2	2	2	CAAGCAGAAGACGGCATACGAGATNNNNNNNNGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCTGCATGGACGAGCTGTACAAG
tdTomato	2	2	CAAGCAGAAGACGGCATACGAGATNNNNNNNNGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCTCCTCTTTCTCTATGGGATGGATGA

mCherry	2	2	CAAGCAGAAGACGGCATACGAGATNNNNNNNNGT GACTGGAGTTCAGACGTGTGCTCTTCCGATCTCGGC ATGGACGAGCTGT

Supplemental Table 2. Marker Genes.

Cell Type	**Marker Gene(s)**
Astrocytes	Aldh1l1 (168), Sox9 (169)
Neurons	Rbfox3 (170)
Vascular Cells	Cldn5 (171)
Endothelial Cells	Slc2a1 (172)
Pericytes	Pdgfrb (173), Rgs5, Abcc9 (100)
Red Blood Cells	Hba-a1, Hba-a2 (174)
Vascular SMCs	Acta2, Myh11, Tagln (175)
Vascular LMCs	Fam180a, Slc6a13, Dcn, Ptgds (61)
Microglia	Cx3cr1, Tmem119 (109)
Leukocytes	Itgal, Gzma (75)
Perivascular Macropages	Mrc1 (109)
Oligodendrocytes	Olig2 (176)
OPCs	Pdgfra, Cspg4 (177)
Mature Oligos	Mog, Mbp (178)
Committed Oligos	Ptprz1, Bmp4, Nkx2-2, Vcan (61)

Supplemental Table 3. scVI Hyperparameter Tuning

Dispersion	**Latent Lib Size**	**# Latent**	**# Layers**	**# Hidden**	**Test KL Divergence**
Gene	False	10	1	128	5366.4

Gene-batch	True	10	1	128	5406.1
Gene	True	10	1	128	5391.0
Gene-batch	True	50	2	512	5362.6
Gene-batch	False	10	1	128	5378.7
Gene	False	25	1	128	5354.6
Gene	False	25	2	256	5337.8
Gene	False	20	2	256	5336.7
Gene	False	40	4	1024	5338.0

Supplemental Table 4. Sample Metadata. *Supplemental file contains the following fields.*

Field Name	***Description***
10X Version	Whether the sample was processed using 10X V2 or V3 chemistry
Animal ID	A unique animal identifier. Some animals provided multiple samples
Target # Cells	The target number of cells for extraction. 1.6X this number is loaded into the 10X Chromium instrument
Recovered Cells	The number of cells recovered, after debris and multiplet filtering
Typed Cells	The number of cells typed after neuronal and non-neuronal cell assignment
Cell Ranger # Cells	The number of cells as predicted by Cell Ranger
Predicted Multiplets	The number of predicted multiplets
Transcriptome Sequencing Depth	The number of reads
Transcriptome Reads/Cell	The number of reads divided by the number of recovered cells
Median UMIs/Cell	Of the recovered cells, the median total UMI count

Median Genes/Cell	Of the recovered cells, the median number of genes detected with at least one transcript
Variants Recovered	Which variants were recovered from this sample. Samples labeled “Cell Typing Only” were not used for tropism or immune analysis, but were included in the cell type classifier
Virus Sequencing Depth	The number of reads of the amplified viral transcripts across all templates
Virus Reads/Cell	The read depth of the amplified viral transcripts
Age at Extraction (Days)	The age of the animal at extraction time
Virus Incubation Time (Days)	How many days prior to extraction the animal was injected
Percent of Virus UMIs Determined	What percent of transcriptome reads that aligned to the virus gene were disambiguated from the amplified lookup table

Supplemental Table 5. Variant Barcodes

Variant	**Cargo**	**Barcodes**
AAV-PHP.eB	pAAV:CAG-NLS-mNeonGreen	
AAV-PHP.V1	pAAV:CAG-NLS-mRuby2	
AAV-PHP.eB	pAAV:CAG-NLS-mNeonGreen	CCTGACA, GGACAGA, GCACAGA, CGAGAGA
AAV-PHP.V1	pAAV:CAG-tdTomato	
AAV-PHP.V1	pAAV:CAG-NLS-mNeonGreen	CAGTGTC, GAGAGTG, GTGTGAG
AAV-CAP-B10	pAAV:CAG-NLS-mNeonGreen	
AAV-PHP.eB	pAAV:CAG-NLS-tdTomato	
AAV-CAP-B10	pAAV:CAG-NLS-tdTomato	

AAV9	UBC-mCherry-AAV-cap-in-cis	CGTCTCAGCTATAACTTCCAA CGAGGTCGTAAGGTCGGCATT TGATTATCATGCCTGCTCAGG
AAV-PHP.B	UBC-mCherry-AAV-cap-in-cis	TATACCCAACCACTCAGTCCC CGGTTTTAGCACGGCCATAGA AAGCGATGTCTCTACACGATA
AAV-PHP.eB	UBC-mCherry-AAV-cap-in-cis	TACAGCTTTTTGACTGGAGGT CTGGCATTAATACGCGGGTCA TACAGGTCCTAGACAGGTGAT
AAV-CAP-B10	UBC-mCherry-AAV-cap-in-cis	GCTGGGCGTTAAAGTACTCGC GCAACTGGGATAATCGTAGTC AACGGAGTGAACGGACCCTAG
AAV-PHP.V1	UBC-mCherry-AAV-cap-in-cis	GTGGCGGGTTTCCGAAAAAGT TCGTCGGCACTCTCTTAGAGC CATGTGATAGTGAAGCACGCC
AAV-PHP.C1	UBC-mCherry-AAV-cap-in-cis	TCTGTGCTGCTCTTCTAACAA TCTGACGGCGGGTAAACACTG TGGCCACCCGCAGAGTATACT
AAV-PHP.C2	UBC-mCherry-AAV-cap-in-cis	GACTAGGGTAAGTGAGCTATG CGAATTTCTTCCATACCTCCT TAGTGCCAACAACGGAGAAGA

Supplemental Table 6. Cell Metadata. *Supplemental file contains one row per cell, with the following fields.*

Field Name	**Description**
Cell ID	The cell barcode plus a sample index
Cell Type	The final cell type of this cell
AAV	The number of UMIs in this cell that align to the custom AAV reference gene

CCN202105041	The predicted output of this cell from the droplet classifier – neurons vs non-neurons
p_CCN202105041	The probability of classification of this droplet type
CCN202105050	The predicted neuronal subtype of this cell, if it is a neuron
p_CCN202105050	The probability of classification of this neuron subtype
CCN202105051	The named non-neuronal subtype of this cell, if it is a non-neuron
c_CCN202105051	The cluster this cell was assigned to in the non-neuronal model
X_CCN202105051	The x-coordinate of a t-SNE projection of this cell in the non-neuronal model
Y_CCN202105051	The y-coordinate of a t-SNE projection of this cell in the non-neuronal model
CCN202105070	The cluster assignment of this cell in the joint neuronal and non-neuronal model
X_CCN202105070	The x-coordinate of a t-SNE projection of this cell in the joint neuronal and non-neuronal model
Y_CCN202105070	The y-coordinate of a t-SNE projection of this cell in the joint neuronal and non-neuronal model
Cell Set	The origin cell set of this cell
[Variant]	The number of UMIs in this cell that are associated with the given barcoded AAV variant

Supplemental Table 7. Transduction Rates by Cell Type. *Supplemental file contains one row per unique variant in each cell set.*

Field Name	**Description**
Entry Id	{Cell Set}-{Variant}
Cell Set	The source cell set
Virus	Which variant this row describes. Note: “AAV” means the transduction rate of any AAV present in the sample, as determined by a read’s alignment to the

	custom AAV gene. All other variants originate from disambiguated counts.
[Cell Type] Transduction Rate	The estimated transduction rate of this variant in this cell type
[Cell Type] Num Cells	The number of total cells of this type in this cell set
Field Name	Description

Supplemental Table 8. Differentially Expressed Genes Across Time Points. *Supplemental file contains one tab per cell type, with the following fields.*

Field Name	**Description**
Gene ID	The Ensembl Gene ID
Gene name	The canonical gene name
Mean expression	The mean expression of this gene in this group
L2FC	The log fold change of this gene
L2FC SE	The standard error of the L2FC
Stat	The stat, as reported by DESeq2
P-value	The unadjusted P-value
Adjusted P-value	The adjusted P-value

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