

Human microglia show unique transcriptional changes in Alzheimer's disease

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Prater, Green, et al. Supplemental Tables

Supplemental Table 1: Complete demographic information on the cohort

Sample	Study Designation	Coded AGE	SEX	Race	APOE Genotype	PMI	ADNC Score
1	AD	90+	F	White	3,3	5.08	3
2	AD	74	F	White	3,4	4.42	3
3	AD	90+	F	Mixed	3,3	5.08	2
4	AD	90+	F	White	3,3	3.75	3
5	AD	60	F	Unknown/Unreported	3,3	5.75	3
6	AD	86	F	White	3,3	8.07	2
7	AD	87	F	White	3,4	4.87	3
8	AD	90+	F	White	3,4	4.25	3
9	AD	77	F	White	4,4	3.33	3
10	AD		M	White	3,3	3.33	3
11	AD	83	M	White	3,4	3	3
12	AD	90+	M	White	3,4	4.58	3
13	Ctrl	90+	F	Hispanic / Latino	2,3	4.33	0
14	Ctrl	90+	F	White	3,3	3.75	1
15	Ctrl	90+	F	White	3,3	6.97	0
16	Ctrl	80	F	White	3,3	8.13	0
17	Ctrl	90+	F	White	3,3	7.72	1
18	Ctrl	70	F	White	3,4	7	1
19	Ctrl	74	M	White	2,3	4.83	0
20	Ctrl	84	M	White	3,3	3.92	1
21	Ctrl	90+	M	White	3,3	8.17	1
22	Ctrl	82	M	White	3,3	7.75	1

Ctrl = Control, AD = Alzheimer's Disease pathology, Coded Age = age at death in years. Ages greater than 90 are coded 90+ to maintain anonymity, F = Female, M = Male, Race = self-reported race, *APOE* Genotypes: *APOE* alleles $\epsilon 2/\epsilon 3$ (2/3), *APOE* alleles $\epsilon 3/\epsilon 3$ (3/3), *APOE* alleles $\epsilon 3/\epsilon 4$ (3/4), or *APOE* alleles $\epsilon 4/\epsilon 4$ (4/4), PMI = post-mortem interval in hours, ADNC = Alzheimer's Disease Neuropathic Change

Recipes for Nuclei Extraction and FANS Buffers:

Supplemental Table 2: Myelin gradient buffer (Store at 4°C)

Reagent	Volume for 1 Liter
NaH ₂ PO ₄ ·H ₂ O (Fisher Scientific, S369-500), adjust to pH 7.4 with 3.56g/L Na ₂ HPO ₄ ·H ₂ O (Fluka, 71643)	0.78 g
NaCl (Fisher Scientific, S271-3)	8.0 g
KCl (Fisher Scientific, P217-500)	0.4 g
Glucose (Sigma, G7021-1KG)	2.0 g
BSA (VWR, EM-2930)	0.2%

Supplemental Table 3: Nuclei buffer (NB) (Store at 4°C)

Reagent	Volume for 10mL
Nuclease-free water into a 15 ml conical tube. (Fisher Scientific, M46000)	9.85 mL
1 M Tris-HCl, pH 7.5 (ThermoFisher, 15567027)	100 µL
5 M NaCl (ThermoFisher AM9760G)	20 µL
1 M MgCl ₂ (ThermoFisher Scientific, AM9530G)	30 µL

Supplemental Table 4: Nuclei lysis buffer (NLB) (make same-day)

Reagent	Volume for 1 Sample
Nuclei Buffer	727 µL
10% NP-40 alternative (final concentration 0.1%)	10 µL
Protease inhibitors in DPBS (Sigma-Aldrich, 4693124001)	142.9 µL
1mM ATA in NB (make fresh evening prior)	112.5 µL
PMSF (Tocris Bioscience, 4486)	10 µL
Phosphatase inhibitors (Sigma-Aldrich, P5726-1ML)	5 µL
Protector RNase inhibitor (final concentration 1 U/µl). (Sigma, 3335402001)	28.25 µL
Total Volume	1,035.5 µL

Supplemental Table 5: Nuclei suspension solution (NSS) (make same-day)

Reagent	Volume for 1 Sample
DPBS (Sigma-Aldrich, D8537-500ML)	637 µL
10% BSA (final concentration 1%) (Sigma-Aldrich, A1595-50mL)	100 µL
1mM ATA in DPBS (make fresh evening prior)	100 µL
7x Protease inhibitors in DPBS (Sigma-Aldrich, 4693124001)	142.9 µL
1M PMSF (Tocris Bioscience, 4486)	10 µL
Protector RNase inhibitor (final concentration 1.0 U/µl) (Sigma, 3335402001)	5 µL
Phosphatase inhibitors (Sigma-Aldrich, P5726-1ML)	5 µL
Total Volume	1 mL

Supplemental Table 6: Percoll/myelin gradient buffer solution (PMB) (make same day)

Reagent	Volume for 1 Sample
Myelin gradient buffer	412 μ L
10x HBSS (Fisher Sci., 14185052)	30 μ L
1mM ATA in myelin gradient buffer (make fresh evening prior)	100 μ L
1.5M NaCl	25 μ L
Percoll (Fisher Sci., 17-089-101) (Do not use if crystals have precipitated out)	270 μ L
Protector RNase Inhibitor (final concentration 1.0 U/ μ l) (Sigma Aldrich, 3335402001)	5 μ L
Phosphatase inhibitors (Sigma-Aldrich, P5726-1ML)	5 μ L
7x Protease inhibitors in myelin gradient buffer (Sigma-Aldrich, 4693124001)	142.9 μ L
PMSF (Tocris Bioscience, 4486)	10 μ L
Total Volume	1 mL

Supplemental Table 7: FACS Media (FM) (store at 4°C)

Reagent	Volume for 1 sample
Nuclease free H ₂ O (Fisher Scientific, M46000)	7.7 mL
HEPES (Invitrogen, 15630080)	100 μ L
10x HBSS without Mg/Ca (Fisher Scientific, 14185052)	1 mL
FBS 10%	1 mL
Protease inhibitors (Sigma-Aldrich, 4693124001) (dissolve tablet in 9.8mL of the above media at room temp and chill on ice before adding the solutions below)	“~200 μ L”
PMSF (Tocris Bioscience, 4486)	100 μ L
Phosphatase inhibitors (Sigma-Aldrich, P5726-1ML)	50 μ L
Protector RNase Inhibitor (Sigma Aldrich, 3335402001)	50 μ L
100 μ M ATA in dPBS (Sigma-Aldrich, A1895) good at -20C for 1 month	1 mL
Total Volume	11.2 mL