

Inflammatory stimulus enhances synaptic material uptake by adult APP microglia in a microfluidic neuron–microglia co-culture model

Dolores Siedlecki-Wullich^{1,*}, Anne-Marie Ayrat¹, Lukas Iohan¹, Célia Lemeu¹, Valérie Buiche¹,
Karine Blary², Julien Chapuis¹, Fanny Eysert^{1,†}, Delphine Beury³, Myriam Delacre³, David Hot³,
Takahiro Masuda⁴, Klaus-Peter Knobloch⁵, Marco Prinz⁵⁻⁷, Jean-Charles Lambert^{1,‡}, Devrim
Kilinc^{1,‡,*}

¹ Université de Lille, Institut Pasteur de Lille, CHU Lille, INSERM U1167, LabEx DISTALZ, Lille
59019, France

² Univ. Lille, CNRS, Centrale Lille, Univ. Polytechnique Hauts-de-France, UMR 8520 - IEMN -
Institut d'Electronique de Microélectronique et de Nanotechnologie, F-59000 Lille, France

³ Univ. Lille, CNRS, Inserm, CHU Lille, Institut Pasteur de Lille, US 41 - UAR 2014 - PLBS, F-59000
Lille, France

⁴ Division of Molecular Neuroimmunology, Medical Institute of Bioregulation, Kyushu University,
Fukuoka, Japan

⁵ Institute of Neuropathology, Faculty of Medicine, University of Freiburg, Freiburg, Germany

⁶ Signalling Research Centres BIOSS and CIBSS, University of Freiburg, Freiburg, Germany

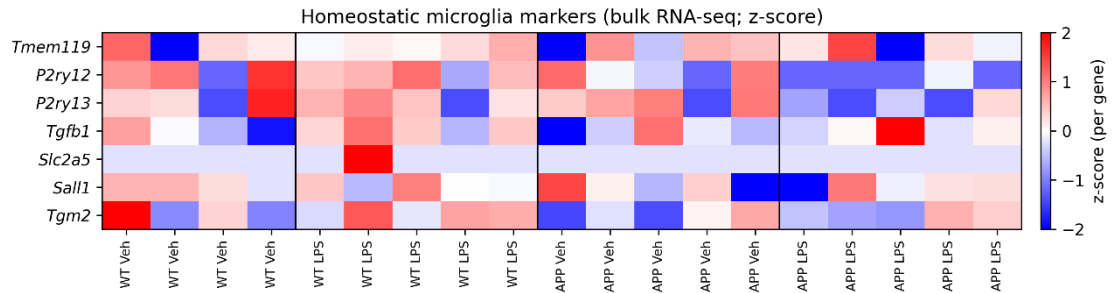
⁷ Center for Brain Research and Advancements In Neuroimmunology (BRAIN), Faculty of
Medicine, University of Freiburg, 79106 Freiburg, Germany

[†] Current address: Université Côte d'Azur, INSERM U1323, CNRS UMR7275, Institute of molecular
and cellular pharmacology, Laboratory of excellence DistALZ, 06560, Sophia-Antipolis, Valbonne,
France

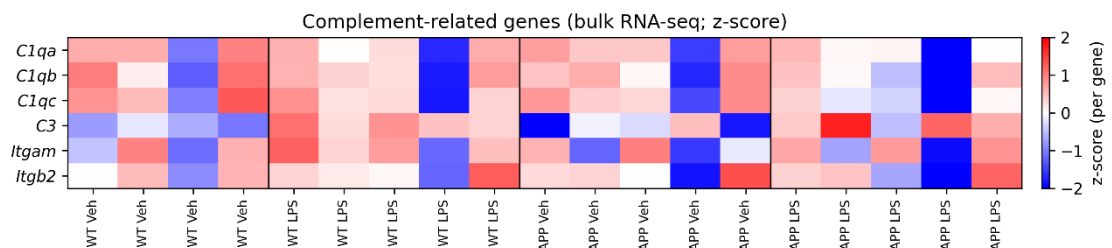
[‡] These authors share last authorship

* Address for correspondence: siedleckidolores@gmail.com ORCID: 0000-0002-9099-9406;
devrim.kilinc@pasteur-lille.fr ORCID: 0000-0003-3321-5203

SUPPLEMENTARY MATERIAL



Supplementary Figure 1. Expression of canonical microglial homeostatic markers: Heatmap showing normalized expression levels of canonical microglial homeostatic markers (including *Tmem119*, *P2ry12*, *P2ry13*, *Sall1*, *Tgfb1*, *Slc2a5*, and *Tgm2*) in adult WT and APP microglia cultured in vitro and treated with vehicle or LPS. Values are shown as z-score-normalized expression per gene across samples. While several homeostatic markers are reduced compared to in vivo reference states, consistent with long-term in vitro culture, their continued expression confirms preservation of microglial lineage identity across all conditions. This analysis is provided to contextualize the functional assays and is not intended as a comprehensive assessment of microglial state.



Supplementary Figure 2. Expression of complement-related genes in adult microglia: Heatmap showing expression of key complement pathway components implicated in synapse-associated phagocytosis, including *C1qa*, *C1qb*, *C1qc*, *C3*, *Itgam* (*CD11b*), and *Itgb2*, in WT and APP adult microglia under vehicle or LPS conditions. Values are shown as z-score-normalized expression per gene across samples. Complement-related transcripts are robustly expressed across conditions, with only modest and heterogeneous transcriptional modulation following LPS stimulation. These data support a functional engagement of complement signaling rather than large-scale transcriptional induction.

Supplementary Table 1. Top upregulated and downregulated genes following LPS stimulation in WT and APP adult microglia. Listing of the top genes ranked by log₂ fold-change in WT and APP microglia following LPS stimulation relative to vehicle controls.

Genotype	Direction	Gene	log2FC (log2CPM)	p (Welch t-test)	Mean CPM (LPS)	Mean CPM (Veh)
APP	Down (LPS<Veh)	Hpgd	-2.81	0.05	1.17	22.28
APP	Down (LPS<Veh)	Gpr34	-2.68	0.01	1.44	13.76
APP	Down (LPS<Veh)	Sparcl1	-2.48	0.05	4.93	15.40
APP	Down (LPS<Veh)	Cd83	-2.43	0.01	2.02	12.39
APP	Down (LPS<Veh)	Kmt5c	-2.42	0.01	1.62	8.61
APP	Down (LPS<Veh)	Rps27rt	-2.37	0.12	32.12	54.51
APP	Down (LPS<Veh)	Cnn1	-2.35	0.04	3.47	17.63
APP	Down (LPS<Veh)	Gm50388	-2.31	0.09	2.64	22.39
APP	Down (LPS<Veh)	Pik3r3	-2.27	0.02	3.29	16.31
APP	Down (LPS<Veh)	Mmab	-2.21	0.00	1.26	8.52
APP	Up (LPS>Veh)	Il1a	3.65	0.02	99.98	15.93
APP	Up (LPS>Veh)	Il1b	3.61	0.00	398.67	47.42
APP	Up (LPS>Veh)	Traf1	3.31	0.01	25.38	3.89
APP	Up (LPS>Veh)	Il12b	3.26	0.11	64.88	53.71
APP	Up (LPS>Veh)	Cxcl2	3.21	0.07	1147.82	387.37
APP	Up (LPS>Veh)	Ccl20	3.10	0.01	40.67	4.25
APP	Up (LPS>Veh)	Cxcl5	3.06	0.18	270.62	96.69
APP	Up (LPS>Veh)	Cxcl1	3.05	0.03	1396.59	262.99
APP	Up (LPS>Veh)	Wfdc21	3.00	0.02	27.52	1.90
APP	Up (LPS>Veh)	Cxcl3	2.99	0.09	51.55	23.77
WT	Down (LPS<Veh)	Ddias	-2.56	0.01	1.87	12.29
WT	Down (LPS<Veh)	Hpgd	-2.52	0.06	1.84	20.18
WT	Down (LPS<Veh)	Reps2	-2.34	0.03	2.51	21.45
WT	Down (LPS<Veh)	Inmt	-2.27	0.26	16.22	88.29
WT	Down (LPS<Veh)	Parp6	-2.20	0.00	1.64	10.67
WT	Down (LPS<Veh)	Pltp	-2.13	0.14	18.49	32.22
WT	Down (LPS<Veh)	Arhgap19	-2.11	0.06	7.05	24.47
WT	Down (LPS<Veh)	Aplp1	-2.06	0.01	1.69	8.69
WT	Down (LPS<Veh)	Slc46a3	-1.99	0.03	2.46	14.17
WT	Down (LPS<Veh)	Wwc1	-1.97	0.06	2.91	12.92
WT	Up (LPS>Veh)	Cxcl5	3.67	0.06	74.07	12.35
WT	Up (LPS>Veh)	Il1b	3.31	0.01	312.62	39.95
WT	Up (LPS>Veh)	Cxcl1	3.23	0.09	1078.65	297.06
WT	Up (LPS>Veh)	Il1a	2.81	0.04	105.62	21.62
WT	Up (LPS>Veh)	Ccl20	2.61	0.05	16.89	3.24
WT	Up (LPS>Veh)	Cxcl3	2.60	0.15	49.96	18.82
WT	Up (LPS>Veh)	Ccl5	2.52	0.12	895.63	289.58
WT	Up (LPS>Veh)	Cxcl2	2.51	0.18	1309.22	628.70
WT	Up (LPS>Veh)	Acod1	2.45	0.03	349.50	83.86
WT	Up (LPS>Veh)	Ifi206	2.42	0.09	26.91	8.26