Microglial lipid metabolism: a delicate balance

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General introduction

What are microglia?

The discovery of microglia evolved over 75 years, driven by the advancement of histology methods used to characterize brain tissue. They were first described under the term 'neuroglia' by Virchow in 1846[1,2]. Following the identification of astrocytes by Van Lenhossék in 1891, Cajal proposed the existence of a "third element" in 1913[3]. It was not until 1919 that Del Río-Hortega successfully distinguished microglia from oligodendrocytes[4]. Microglia were recognized for their unique phagocytic capacity, inferred from the presence of lipid-filled granules, as well as their remarkable structural plasticity. To this day, the fundamental link between microglial morphology and function continues to shape microglial research[5].

It took almost a century longer before the field agreed on the origin of microglia[6–10]. Unlike neurons and other glia, microglia originate from the myeloid lineage and are seeded from the yolk sac during early embryonic development. After infiltrating the developing central nervous system (CNS), microglia persist into adulthood and maintain their population through local proliferation, rather than replenishment by bone-marrow derived monocytes as is characteristic of other tissue-resident macrophages[11–13].

As the name suggests, microglia are the smallest of all neuroglia and generally exhibit a ramified morphology, characterized by a small soma and elongated processes that interact with synapses, dendrites, and debris[14] (**Figure 1.1**). Due to their uniform disposition throughout the CNS, with a higher density in grey compared to white matter, and their highly motile processes, microglia are swift to scan for and react to disruptions in the brain's microenvironment[1]. In pathological conditions, microglia retract their processes and mobilize their cell bodies, adopting an amoeboid morphology (**Figure 1.1**). These amoeboid microglia become increasingly prevalent in the aging brain, reflecting their role in responding to injury and neurodegeneration[15].

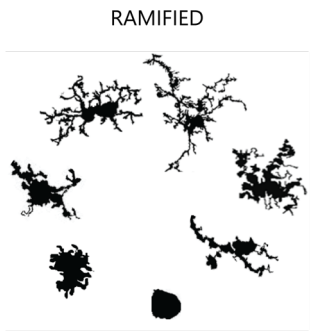

Figure 1.1 | Microglial morphology as presented in the human mature CNS. This morphology wheel displays snapshots of dynamic and interchangeable microglial shapes. The shapes are not in relative scale. Adapted from Karperien *et al.*[14].

What is the function of microglia?

Microglia are pivotal mediators that bridge the neural and immune system, facilitating crosstalk between these two complex networks[16]. By responding to signals from both systems, microglia help maintain homeostasis, coordinate immune responses within the CNS, and influence neural activity. Microglial properties that are critical for enabling these biological functions include surveillance, phagocytosis, and release of soluble factors, such as chemokines and cytokines[5].

Microglia are highly versatile in the receptors they express to regulate these effector functions, some of which are also commonly used as microglial markers (*). Examples include triggering receptors expressed on myeloid cells-2 (TREM2)*, ionized calcium-binding adapter molecule 1 (IBA1)*, CD11b that makes up complement receptor-3 (CR3) together with CD18, colony-stimulating factor 1 receptor (CSF1R), the fractalkine receptor CX3CR1*, purinoreceptor P2RY12*, and transmembrane receptor TMEM119*[5].

Only fairly recently, microglia were found to actively engulf pre- and postsynaptic components in the developing brain of mice[17,18]. This process, called synaptic pruning, is critical for neural circuit maturation and is monitored, among others, by CX3CR1 on microglia. Deficits in synaptic pruning by microglia may lead to altered brain circuits and behavior underlying neurodevelopmental disorders, particularly those that manifest in adolescence[19]. Patients with adult-onset leukoencephalopathy with axonal spheroids and pigmented glia (ALSP) challenge this concept[20]. ALSP is a microgliopathy arising from mutations in the *CSF1R* gene, causing white matter atrophy that affects behavior, cognition and motor function. Microglia viability and population maintenance in the brain are heavily dependent on CSF1R, as blockade of this receptor eliminates 99% of CNS microglia[21]. Yet, microglia-depleted mice do not show behavioral or cognitive abnormalities, nor do ALSP patients present with neurodevelopmental deficits, suggesting that microglia may be dispensable for healthy neurodevelopment. CSF1R is, however, expressed across the myeloid lineage, and its deficiency affects other monocytes and macrophage populations, including those in the CNS, which complicates the determination of specific microglia mediated defects[22,23].

Since the development of *Csf1r*$^{\Delta FIRE/\Delta FIRE}$ mice, which affect CSF1R expression selectively in microglia and tissue-resident macrophage populations in the skin, heart, kidney and peritoneum, microglial deficiency defects could be discriminated from those of other CNS-resident macrophages[24]. Indeed, *Csf1r*$^{\Delta FIRE/\Delta FIRE}$ mice are healthy, fertile and show no (neuro)developmental issues[24,25], but a critical role for microglia becomes apparent later in life[26]. Myelination emerges as a key process influenced by microglia in an age-dependent manner. Up until 1 month of age, developmental myelination, maturation of myelinating oligodendrocytes, and axon myelination (in terms of numbers) were unaffected in *Csf1r*$^{\Delta FIRE/\Delta FIRE}$ mice. However, structural changes in myelin ensheathment began to appear at 1 month and worsened with age, ultimately impairing cognitive flexibility. In the absence

of microglia, myelin outfolding began at the age of 1 month, leading to hypermyelination at 3-4 months, which eventually progressed to demyelination from 4.5 to 6 months of age. This microglia-dependent regulation of myelin growth and integrity was also observed in the frontal white matter of ALSP patients, showing severe myelin outfoldings, altered thickness, and degeneration. The underlying mechanism involved a microglia-oligodendrocyte signaling axis dependent on TGF-β1 and implicated lipid metabolism in myelin growth and recycling, including cholesterol esters and triglycerides[26,27].

What is the role of microglia in neuroinflammation?

With the ever-growing life expectancy, degeneration of the human brain is becoming increasingly prevalent[28,29]. A common denominator across neurodegenerative diseases is the accumulation of macromolecules, either within cells or in the surrounding extracellular space. These accumulations indicate inefficient clearance mechanisms, such as phagocytosis, and result in (protein) dyshomeostasis. Examples include hyperphosphorylated tau and amyloid-β in Alzheimer's disease (AD), α-synuclein in Parkinson's disease, huntingtin in Huntington's disease and foamy microglia in Multiple Sclerosis (MS)[30,31]. Other factors commonly associated with neurodegeneration include CNS inflammation and a leaky blood-brain barrier, which makes the brain highly susceptible for further damage[32–34]. Tackling such diseases, however, present great challenges as they involve targeting the most complex organ of the human body.

As professional phagocytic immune cells, there is no doubt that microglia are active contributors to the progression of neurodegenerative diseases, highlighting their significant therapeutic potential[15]. Microglia govern the activation of an inflammatory response to eliminate harmful substances and prevent further damage[34]. In doing so, microglia excrete cytokines and chemokines that attract more glial cells and adaptive immune cells. However, if the response becomes chronic, prolonged inflammation can lead to neuroinflammation, a common hallmark of multiple neurodegenerative diseases. Given the established link between microglia and neurodegeneration, the perception of neuroinflammation has shifted from being merely an associated phenomenon to a central factor in the development of neurodegenerative diseases[34]. This is specifically underlined by the use of nonsteroidal anti-inflammatory drugs (NSAIDs) that reduce the risk of developing AD by 50%[35]. It is hypothesized that with ageing, microglia adopt a senescence-associated secretory phenotype perpetuating low-grade neuroinflammation and eventually contributing to neurodegeneration[36].

It is challenging to disentangle the beneficial and detrimental roles microglia may play in neuroinflammation and -degeneration. The rise of single-cell analysis of transcription or surface markers offers an unprecedented level of resolution providing tools to unravel these complex roles and identify key proteins associated with them as potential targets for

modulation[37–42]. However, the association of gene expression signatures with specific microglial roles does not necessarily indicate exclusive functions of these proteins. Therefore, it is important to understand the function of proteins across the full spectrum of microglial states.

The terminology used to describe microglial states is currently under debate and no general consensus has been reached[5]. It is important to note that in this thesis the term 'activated' is used to describe microglia that are treated with stimulant(s). This does not imply non-stimulated microglia are inactive, on the contrary, non-stimulated microglia also actively participate in cytokine secretion and phagocytosis. The term 'activation' is simply used to refer to the process in which microglia adopt a distinct functional state in response to the stimulant used relative to non-stimulated microglia.

Lipid metabolism in microglia

The role of microglia in the CNS typically centers on phagocytosis, whether of synapses, myelin, dead cells or pathogens. However, in certain diseases, like MS, inefficient clearing of lipids or lipid-rich structures gives rise to a third microglial/macrophage morphology: the foamy phenotype[30,43,44]. Foamy microglia resemble lipid-laden macrophages, a phenotype observed in various diseases, including Gaucher disease, atherosclerosis, and obesity. This morphology is commonly characterized by a dysregulated lipid metabolism[43]. Foamy microglia are a hallmark of MS[30,45] and are associated with increased disease severity[44], highlighting the role of microglial lipid metabolism as a key mediator.

Lipid metabolism not only provides energy or structural features to cells, but may also make up signaling networks[46]. The growing recognition of lipids as critical regulators of microglia functional states underscores the therapeutic potential of lipid signaling systems for neurodegenerative diseases[46–48]. The endocannabinoid system is such a sophisticated lipid signaling network that governs numerous biological processes, such as behavior, cognition, pain, but also neuroinflammation[49]. Its function is regulated by two lipids, *i.e.* endocannabinoids, 2-arachidonoylglycerol (2-AG)[50,51] and *N*-arachidonoylethanolamine (AEA)[52], that activate cannabinoid receptors CB_1 (CB1R) and CB_2 (CB2R). As most abundant endocannabinoid within the CNS[53], 2-AG forms the primary precursor for the production of inflammatory lipid mediators, thereby linking the endocannabinoid system to neuroinflammation[54–58]. The endocannabinoid system is compartmentalized across brain regions and cell types, with microglia expressing a specific set of endocannabinoid system proteins: CB2R, diacylglycerol lipase (DAGL) β, monoacylglycerol lipase (MAGL) and α,β-hydrolase-domain containing protein 12 (ABHD12)[57] (**Figure 1.2a,b**). DAGLβ hydrolyzes phospholipid-derived diacylglycerol (DAG) into 2-AG, that in turn can be cleaved by both MAGL and ABHD12 to form arachidonic acid (AA)[57,59]. AA is the predominant substrate for oxidation, producing a wide array of inflammatory mediators, *i.e.* oxylipins or eicosanoids[60].

Human mutations in ABHD12 are found to cause Polyneuropathy, Hearing loss, Ataxia, Retinitis pigmentosa, and Cataract (*i.e.* PHARC)[61–66]. PHARC patients suffer from neurodegeneration and ABHD12$^{-/-}$ mice display activated microglia, implicating ABHD12 is potentially crucial for microglial function[67]. In addition to 2-AG, ABHD12 hydrolyzes lysophospholipids[67,68] and oxidized phosphatidylserine[69], substrates that accumulate upon ABHD12 inactivation. To date, it remains unclear what mechanisms underpin microglia activation and ultimately PHARC symptoms as a consequence of ABHD12 inactivation.

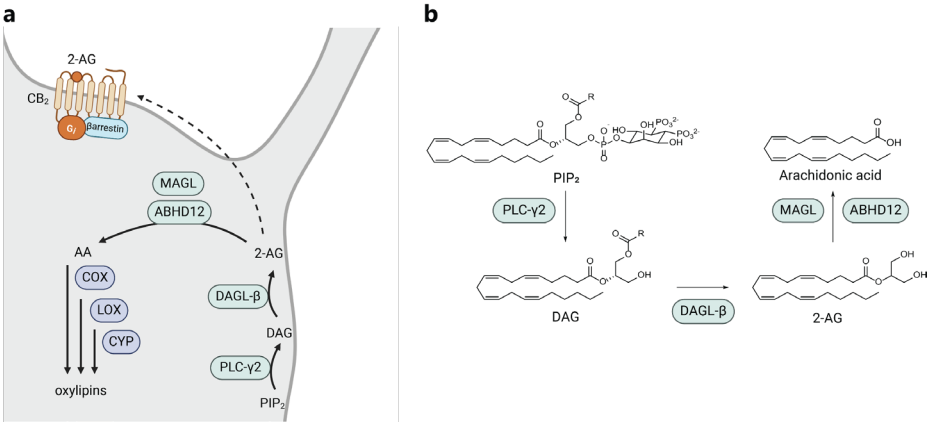

Figure 1.2 | Endocannabinoid system in microglia. (a) Membrane phospholipid PIP_2 is hydrolyzed by PLCγ2 into DAG, that in turn is degraded by DAGLβ to generate the endocannabinoid 2-AG. 2-AG can activate CB_2 receptors on microglia or inactivated via hydrolysis by MAGL or ABHD12 into AA. The oxidation of AA by enzymes from the COX, LOX or CYP family leads to the formation of oxylipins. **(b)** Synthesis route described in (**a**). 2-AG: 2-arachidonoylglycerol. AA: arachidonic acid. ABHD12: α, β-hydrolase domain-containing protein 12. CB_2: cannabinoid receptor type 2. COX: cyclooxygenase. CYP: cytochrome P450. DAG: diacylglycerol. DAGLβ: diacylglycerol lipase β. LOX: lipoxygenase. PIP_2: phosphatidylinositol 4,5-bisphosphate. PLCγ2: phospholipase Cγ2.

Aim and outline

The aim of this thesis is to investigate the role of lipid metabolism in microglial functions.

Chapter 2 describes the novel application of activity-based protein profiling (ABPP) to characterize lipid hydrolase activities across microglial activation states. Using various pro- and anti-inflammatory stimuli on N9 microglia, a lipid hydrolase activity map was generated. Cluster analysis highlighted endocannabinoid enzymes DAGLβ and ABHD12 as key players in microglial activation. Targeted lipidomics further established a link between the endocannabinoid system and the modulation of microglial activation. Inhibiting DAGLβ or ABHD12 activity demonstrated anti-inflammatory effects by reducing LPS-induced cytokine secretion.

Chapter 3 further elaborates on DAGLβ's role in microglial cytokine secretion. As a 2-AG producing enzyme, DAGLβ contributes to the formation of downstream lipid arachidonic acid (AA) and pro-inflammatory oxylipins. Genetic inactivation of DAGLβ reduced cellular 2-AG and AA, but only suppressed *Il6* mRNA at low LPS doses, likely due to compensatory upregulation of other 2-AG producing enzymes DAGLα and ABHD6. Pharmacological inhibition of DAGLβ confirmed anti-inflammatory effects by reducing IL-6 secretion.

Chapter 4 explores ABHD12's role in microglial cytokine secretion. ABHD12 was shown to regulate 2-AG and AA levels, linking ABHD12 activity to oxylipin-driven inflammation. ABHD12 inhibition reduced TNF-α and IL-6 secretion in LPS-stimulated microglia, an effect attributed to CB2R activation and reduced PGE2 levels. In primary microglia, ABHD12 inactivation reduced IL-6 levels, which was linked to lowered PGE2 concentrations.

Chapter 5 expands on the role for ABHD12 in microglial effector functions, specifically phagocytosis of myelin debris. TGF-β1 induced ABHD12 activity, which was concomitant with enhanced myelin uptake, while ABHD12 inhibition impaired lysosomal function, elevating lysosomal pH and causing myelin buildup. This led to lysosomal stress, as demonstrated by the induction of lysosomal stress marker GPNMB, highlighting a critical role for ABHD12 in lysosomal health and myelin recycling.

Chapter 6 examines ABHD12's global effects on microglial metabolism by applying global proteomics and targeted lipidomics. ABHD12 inactivation disrupted phospholipid homeostasis, increased ER stress, and impaired lysosomal and mitochondrial function. Enhanced ROS metabolism resulted in heightened susceptibility of microglia to ferroptosis, emphasizing ABHD12's role in maintaining cellular homeostasis, particularly that of the ER.

Chapter 7 investigates the TREM2-PLCγ2-MAGL axis in human iPSC-derived microglia. TREM2 activation with small molecule SM 3969 enhanced PLCγ2 activity, 2-AG metabolism, and anti-inflammatory transcriptomic responses. The AD protective variant PLCγ2^{P522R} reduced inflammation while preserving the transcriptomic response to TREM2, highlighting a potential therapeutic strategy of targeting TREM2 in AD.

Chapter 8 summarizes the work described in this thesis and discusses future directions.

References

1. Tremblay, M.-È., Lecours, C., Samson, L., Sánchez-Zafra, V. & Sierra, A. From the Cajal alumni Achúcarro and Río-Hortega to the rediscovery of never-resting microglia. *Front. Neuroanat.* **9**, (2015).
2. Rezaie, P. & Hanisch, U.-K. Historical Context. in *Microglia in Health and Disease* (eds. Tremblay, M.-È. & Sierra, A.) 7–46 (Springer, New York, NY, 2014). doi:10.1007/978-1-4939-1429-6_2.
3. García-Marín, V., García-López, P. & Freire, M. Cajal's contributions to glia research. *Trends Neurosci.* **30**, 479–487 (2007).
4. Río-Hortega, P. del. *Estudios Sobre La Neuroglia. La Microglía Y Su Transformación En Células En Bastoncito Y Cuerpos Gránulo-Adiposos*. (1920).
5. Paolicelli, R. C. *et al.* Microglia states and nomenclature: A field at its crossroads. *Neuron* **110**, 3458–3483 (2022).
6. Alliot, F., Godin, I. & Pessac, B. Microglia derive from progenitors, originating from the yolk sac, and which proliferate in the brain. *Dev. Brain Res.* **117**, 145–152 (1999).
7. Schulz, C. *et al.* A Lineage of Myeloid Cells Independent of Myb and Hematopoietic Stem Cells. *Science* **336**, 86–90 (2012).
8. Kierdorf, K. *et al.* Microglia emerge from erythromyeloid precursors via Pu.1- and Irf8-dependent pathways. *Nat. Neurosci.* **16**, 273–280 (2013).
9. Ginhoux, F. *et al.* Fate Mapping Analysis Reveals That Adult Microglia Derive from Primitive Macrophages. *Science* **330**, 841–845 (2010).
10. Stremmel, C. *et al.* Yolk sac macrophage progenitors traffic to the embryo during defined stages of development. *Nat. Commun.* **9**, 75 (2018).
11. Lei, T. *et al.* Defining newly formed and tissue-resident bone marrow-derived macrophages in adult mice based on lysozyme expression. *Cell. Mol. Immunol.* **19**, 1333–1346 (2022).
12. Scott, C. L. *et al.* Bone marrow-derived monocytes give rise to self-renewing and fully differentiated Kupffer cells. *Nat. Commun.* **7**, 10321 (2016).
13. Wynn, T. A. & Vannella, K. M. Macrophages in Tissue Repair, Regeneration, and Fibrosis. *Immunity* **44**, 450–462 (2016).
14. Karperien, A., Ahammer, H. & Jelinek, H. Quantitating the subtleties of microglial morphology with fractal analysis. *Front. Cell. Neurosci.* **7**, (2013).
15. Gao, C., Jiang, J., Tan, Y. & Chen, S. Microglia in neurodegenerative diseases: mechanism and potential therapeutic targets. *Signal Transduct. Target. Ther.* **8**, 1–37 (2023).
16. Colonna, M. & Butovsky, O. Microglia Function in the Central Nervous System During Health and Neurodegeneration. *Annu. Rev. Immunol.* **35**, 441–468 (2017).
17. Paolicelli, R. C. *et al.* Synaptic Pruning by Microglia Is Necessary for Normal Brain Development. *Science* **333**, 1456–1458 (2011).
18. Pereira-Iglesias, M. *et al.* Microglia as hunters or gatherers of brain synapses. *Nat. Neurosci.* 1–9 (2024) doi:10.1038/s41593-024-01818-w.
19. Mordelt, A. & de Witte, L. D. Microglia-mediated synaptic pruning as a key deficit in neurodevelopmental disorders: Hype or hope? *Curr. Opin. Neurobiol.* **79**, 102674 (2023).
20. Rademakers, R. *et al.* Mutations in the colony stimulating factor 1 receptor (CSF1R) gene cause hereditary diffuse leukoencephalopathy with spheroids. *Nat. Genet.* **44**, 200–205 (2012).
21. Elmore, M. R. P. *et al.* Colony-Stimulating Factor 1 Receptor Signaling Is Necessary for Microglia Viability, Unmasking a Microglia Progenitor Cell in the Adult Brain. *Neuron* **82**, 380–397 (2014).
22. Dai, X.-M. *et al.* Targeted disruption of the mouse colony-stimulating factor 1 receptor gene results in osteopetrosis, mononuclear phagocyte deficiency, increased primitive progenitor cell frequencies, and reproductive defects. *Blood* **99**, 111–120 (2002).
23. Pridans, C. *et al.* Pleiotropic Impacts of Macrophage and Microglial Deficiency on Development in Rats with Targeted Mutation of the Csf1r Locus. *J. Immunol.* **201**, 2683–2699 (2018).
24. Rojo, R. *et al.* Deletion of a Csf1r enhancer selectively impacts CSF1R expression and development of tissue macrophage populations. *Nat. Commun.* **10**, 3215 (2019).
25. O'Keeffe, M. *et al.* Typical development of synaptic and neuronal properties can proceed without microglia in the cortex and thalamus. *Nat. Neurosci.* 1–12 (2025) doi:10.1038/s41593-024-01833-x.
26. McNamara, N. B. *et al.* Microglia regulate central nervous system myelin growth and integrity. *Nature* **613**, 120–129 (2023).
27. Berghoff, S. A. *et al.* Microglia facilitate repair of demyelinated lesions via post-squalene sterol synthesis. *Nat. Neurosci.* **24**, 47–60 (2021).
28. Wyss-Coray, T. Ageing, neurodegeneration and brain rejuvenation. *Nature* **539**, 180–186 (2016).
29. Hou, Y. *et al.* Ageing as a risk factor for neurodegenerative disease. *Nat. Rev. Neurol.* **15**, 565–581 (2019).

30. Grajchen, E., Hendriks, J. J. A. & Bogie, J. F. J. The physiology of foamy phagocytes in multiple sclerosis. *Acta Neuropathol. Commun.* **6**, 124 (2018).
31. Ross, C. A. & Poirier, M. A. Protein aggregation and neurodegenerative disease. *Nat. Med.* **10**, S10–S17 (2004).
32. Bruno, M. *et al.* Blood–brain barrier permeability is associated with different neuroinflammatory profiles in Alzheimer's disease. *Eur. J. Neurol.* **31**, e16095 (2024).
33. Brandl, S. & Reindl, M. Blood–Brain Barrier Breakdown in Neuroinflammation: Current In Vitro Models. *Int. J. Mol. Sci.* **24**, 12699 (2023).
34. Zhang, W., Xiao, D., Mao, Q. & Xia, H. Role of neuroinflammation in neurodegeneration development. *Signal Transduct. Target. Ther.* **8**, 1–32 (2023).
35. Kinney, J. W. *et al.* Inflammation as a central mechanism in Alzheimer's disease. *Alzheimers Dement. Transl. Res. Clin. Interv.* **4**, 575–590 (2018).
36. Matsudaira, T. *et al.* Cellular senescence in white matter microglia is induced during ageing in mice and exacerbates the neuroinflammatory phenotype. *Commun. Biol.* **6**, 1–13 (2023).
37. Pettas, S. *et al.* Profiling Microglia through Single-Cell RNA Sequencing over the Course of Development, Aging, and Disease. *Cells* **11**, 2383 (2022).
38. Absinta, M. *et al.* A lymphocyte–microglia–astrocyte axis in chronic active multiple sclerosis. *Nature* **597**, 709–714 (2021).
39. Masuda, T. *et al.* Spatial and temporal heterogeneity of mouse and human microglia at single-cell resolution. *Nature* **566**, 388–392 (2019).
40. Böttcher, C. *et al.* Single-cell mass cytometry reveals complex myeloid cell composition in active lesions of progressive multiple sclerosis. *Acta Neuropathol. Commun.* **8**, 136 (2020).
41. Böttcher, C. *et al.* Human microglia regional heterogeneity and phenotypes determined by multiplexed single-cell mass cytometry. *Nat. Neurosci.* **22**, 78–90 (2019).
42. Sankowski, R. *et al.* Mapping microglia states in the human brain through the integration of high-dimensional techniques. *Nat. Neurosci.* **22**, 2098–2110 (2019).
43. van Eijk, M. & Aerts, J. M. F. G. The Unique Phenotype of Lipid-Laden Macrophages. *Int. J. Mol. Sci.* **22**, 4039 (2021).
44. Van Der Vliet, D. *et al.* Foamy microglia link oxylipins to disease progression in multiple sclerosis. Preprint at https://doi.org/10.1101/2024.10.18.619040 (2024).
45. Kuhlmann, T. *et al.* An updated histological classification system for multiple sclerosis lesions. *Acta Neuropathol. (Berl.)* **133**, 13–24 (2017).
46. Prakash, P., Randolph, C. E., Walker, K. A. & Chopra, G. Lipids: Emerging Players of Microglial Biology. *Glia* **73**, (2024).
47. Vassiliou, E. & Farias-Pereira, R. Impact of Lipid Metabolism on Macrophage Polarization: Implications for Inflammation and Tumor Immunity. *Int. J. Mol. Sci.* **24**, 12032 (2023).
48. Chausse, B., Kakimoto, P. A. & Kann, O. Microglia and lipids: how metabolism controls brain innate immunity. *Semin. Cell Dev. Biol.* **112**, 137–144 (2021).
49. Egmond, N. van, Straub, V. M. & Stelt, M. van der. Targeting Endocannabinoid Signaling: FAAH and MAG Lipase Inhibitors. *Annu. Rev. Pharmacol. Toxicol.* **61**, 441–463 (2021).
50. Sugiura, T. *et al.* 2-Arachidonoylglycerol: A Possible Endogenous Cannabinoid Receptor Ligand in Brain. *Biochem. Biophys. Res. Commun.* **215**, 89–97 (1995).
51. Mechoulam, R. *et al.* Identification of an endogenous 2-monoglyceride, present in canine gut, that binds to cannabinoid receptors. *Biochem. Pharmacol.* **50**, 83–90 (1995).
52. Devane, W. A. *et al.* Isolation and Structure of a Brain Constituent That Binds to the Cannabinoid Receptor. *Science* **258**, 1946–1949 (1992).
53. Stella, N., Schweitzer, P. & Piomelli, D. A second endogenous cannabinoid that modulates long-term potentiation. *Nature* **388**, 773–778 (1997).
54. Nomura, D. K. *et al.* Endocannabinoid Hydrolysis Generates Brain Prostaglandins That Promote Neuroinflammation. *Science* **334**, 809–813 (2011).
55. Pihlaja, R. *et al.* Monoacylglycerol lipase inhibitor JZL184 reduces neuroinflammatory response in APdE9 mice and in adult mouse glial cells. *J. Neuroinflammation* **12**, 81 (2015).
56. Chen, R. *et al.* Monoacylglycerol Lipase Is a Therapeutic Target for Alzheimer's Disease. *Cell Rep.* **2**, 1329–1339 (2012).
57. Viader, A. *et al.* A chemical proteomic atlas of brain serine hydrolases identifies cell type-specific pathways regulating neuroinflammation. *eLife* **5**, e12345 (2016).
58. Piro, J. R. *et al.* A Dysregulated Endocannabinoid-Eicosanoid Network Supports Pathogenesis in a Mouse Model of Alzheimer's Disease. *Cell Rep.* **1**, 617–623 (2012).
59. Jing, H. *et al.* Phospholipase Cγ2 regulates endocannabinoid and eicosanoid networks in innate immune cells. *Proc. Natl. Acad. Sci.* **118**, e2112971118 (2021).

60. Wang, B. *et al.* Metabolism pathways of arachidonic acids: mechanisms and potential therapeutic targets. *Signal Transduct. Target. Ther.* **6**, 1–30 (2021).
61. Fiskerstrand, T. *et al.* Mutations in *ABHD12* Cause the Neurodegenerative Disease PHARC: An Inborn Error of Endocannabinoid Metabolism. *Am. J. Hum. Genet.* **87**, 410–417 (2010).
62. Chen, D.-H. *et al.* Two Novel Mutations in ABHD12: Expansion of the Mutation Spectrum in PHARC and Assessment of Their Functional Effects. *Hum. Mutat.* **34**, 1672–1678 (2013).
63. Nishiguchi, K. M. *et al.* Exome Sequencing Extends the Phenotypic Spectrum for ABHD12 Mutations: From Syndromic to Nonsyndromic Retinal Degeneration. *Ophthalmology* **121**, 1620–1627 (2014).
64. Lerat, J. *et al.* A complex homozygous mutation in ABHD12 responsible for PHARC syndrome discovered with NGS and review of the literature. *J. Peripher. Nerv. Syst.* **22**, 77–84 (2017).
65. Tingaud-Sequeira, A. *et al.* Functional validation of *ABHD12* mutations in the neurodegenerative disease PHARC. *Neurobiol. Dis.* **98**, 36–51 (2017).
66. Thimm, A. *et al.* Genotype-phenotype correlation in a novel ABHD12 mutation underlying PHARC syndrome. *J. Peripher. Nerv. Syst.* **25**, 112–116 (2020).
67. Blankman, J. L., Long, J. Z., Trauger, S. A., Siuzdak, G. & Cravatt, B. F. ABHD12 controls brain lysophosphatidylserine pathways that are deregulated in a murine model of the neurodegenerative disease PHARC. *Proc. Natl. Acad. Sci.* **110**, 1500–1505 (2013).
68. Ogasawara, D. *et al.* Selective blockade of the lyso-PS lipase ABHD12 stimulates immune responses in vivo. *Nat. Chem. Biol.* **14**, 1099–1108 (2018).
69. Kelkar, D. S. *et al.* A chemical–genetic screen identifies ABHD12 as an oxidized-phosphatidylserine lipase. *Nat. Chem. Biol.* **15**, 169–178 (2019).