IL-1β, TNF-α, and C1q as Drivers of A1 Astrocyte Transformation in Alzheimer's Disease

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Abstract

Alzheimer's disease (AD) is the most common form of dementia, characterised by gradual cognitive decline and neurodegeneration. Traditionally, the research field has focused on amyloid-beta (Aβ) plaques and tau tangles as the primary pathological hallmarks of AD. However, growing evidence suggests that chronic neuroinflammation due to glial cell dysfunction plays a critical role in disease onset and progression. Among these glial cells, microglia and astrocytes are recognised as the main contributors to this neurotoxicity. Thus, this thesis focuses on the communication between microglia and astrocytes, particularly the microglia-derived cytokines IL-1β, TNF-α and C1q and how they contribute to astrocyte dysfunction to induce the neurotoxic A1 astrocyte phenotype in AD. The motivation of this essay stems from the need to better understand non-neuronal drivers of synaptic loss, BBB breakdown, and neuronal death, which contribute to the cognitive symptoms of AD. The thesis describes the physiological functions of astrocytes and microglia, highlighting their role in maintaining BBB integrity, immune surveillance and synaptic pruning. It then outlines how both cell types are dysregulated in AD, with microglia transitioning to a chronically activated state while astrocytes transform into the A1 phenotype due to the secretion of microglial cytokines. The role of AD-risk genes, more specifically TREM2, APOE, and CD33 is also discussed. Overall, this review concludes that the microglia-astrocyte communication axis via IL-1β, TNF-α, and C1q is a critical mechanism driving neurodegeneration in AD. Understanding the glial crosstalk offers potential in AD therapies, but targeting these pathways requires precision to avoid disrupting essential immune functions.

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1. Introduction

Alzheimer's disease (AD) is a progressive neurodegenerative disorder and the most prevalent form of dementia, affecting approximately 50 million people worldwide (Gustavsson et al., 2022; Swanson et al., 2021). The incidence of AD is predicted to rise dramatically as the population ages, thus making it a growing public health concern (Zhang et al., 2021). From a clinical point of view, AD is characterised by a gradual decline in memory (Tramutola et al., 2015), executive function and daily living skills, eventually resulting in complete cognitive and functional dependence (Tryphena et al., 2024). From a neuropathological standpoint, AD is defined by the presence of amyloid-beta (Aβ) plaques (Tramutola et al., 2015) and intracellular neurofibrillary tangles made of hyperphosphorylated tau proteins (Baik et al., 2019; Thal & Tomé, 2022). These proteinopathies disturb neuronal signalling pathways (Zhang et al., 2022) and ultimately cause synaptic failure, neurodegeneration and brain atrophy (Kocahan & Doğan, 2017).

While these key features remain crucial to AD diagnosis (Bomasang-Layno & Bronsther, 2021), they do not fully explain the complexity of the disease. More research indicates that chronic neuroinflammation, driven by non-neuronal cells, plays a pivotal role in the onset and progression of AD (Long et al., 2025). In recent years, the focus of the research has expanded to include glial cells, particularly microglia and astrocytes, which work together to initiate the brain's innate immune response (Olson & Miller, 2004).

Microglia are the resident immune cells of the central nervous system (CNS) (Olson & Miller, 2004; Baik et al., 2019). Under homeostatic conditions, they are responsible for surveying the brain microenvironment (Hu et al., 2019; Hu et al., 2023), phagocytosing debris (Cignarella et al., 2020) and secreting trophic factors to support the development of neurones (Song et al., 2019). However, in AD, constant exposure to Aβ and other damage-associated molecular patterns (DAMPs) encourages microglia to take a more reactive phenotype (Stewart et al., 2010). The activated microglia adopt a pro-inflammatory state, characterised by cytokines, chemokines and reactive oxygen species release (Kim & Lee, 2024). While this is protective at first, it becomes maladaptive in AD, thus contributing to chronic inflammation and ineffective clearance of Aβ (Kim & Lee, 2024). Notably, studies have linked several microglia-related genes, such as TREM2, CD33, and APOE, to an increased risk of AD (Griciuc et al., 2019; Belloy et al., 2023), highlighting the central role of microglia dysfunction in disease progression.

Astrocytes are the most abundant glial cells in the brain (Liddelow et al., 2017) and play equally important roles in maintaining neuronal health (Guttenplan et al., 2021). Astrocytes support synaptic function (Liddelow et al., 2017), regulate ion homeostasis and contribute to the integrity of the blood brain barrier (BBB) (Ye et al., 2024; Tóth et al., 2023). Under pathological conditions, astrocytes also become reactive (Escartin et al., 2021). Some forms of astrogliosis are protective (Verkhratsky et al., 2013), however, a distinct subtype, the A1 astrocytes have been shown to exhibit neurotoxic properties (Liddelow et al., 2017). These A1 astrocytes lose their homeostatic support functions and begin to secrete factors that induce apoptosis in neurones and oligodendrocytes (Clarke et al., 2018).

Influential work by Liddelow et al. (2017) identified critical microglia-astrocyte interaction which drives this A1 transformation of the astrocytes (Liddelow et al., 2017). Specifically, a combination of three microglia-derived cytokines was found to induce the A1 phenotype. These included interleukin-1 beta (IL-1β), tumour necrosis factor-alpha (TNF-α) and complement component 1q (C1q) (Liddelow et al., 2017; Guttenplan et al., 2021). These cytokines are upregulated in the AD brain, and their detection correlates with an increased number of A1 astrocytes in the vicinity of Aβ plaques (Hu et al., 1998). Importantly, this phenotypic change of astrocytes appears to be irreversible and contributes to synapse loss and neurodegeneration (Clarke et al., 2018). IL-1β and TNF-α are classical pro-inflammatory mediators, while C1q is part of the complement cascade that also acts as a signalling molecule in glial communication (Asano et al., 2020). Together, these cytokines initiate a transcriptional program in astrocytes that alters their role from neuronal support to neurotoxicity (Hyvärinen et al., 2019).

Despite the growing research on glial involvement in AD, the precise mechanism by which these cytokines drive astrocyte dysfunction remains unclear. Furthermore, it is still incompletely understood at what stage and region these signals act (Johnson et al., 2020). Clarifying these processes is essential as it may offer novel therapeutic strategies aimed at modulating the glial responses or preventing the A1 transformation altogether. This essay aims to examine the current understanding of how IL-1β, TNF-α and C1q released by microglia mediate astrocyte phenotype change into A1 astrocytes in the context of AD. Understanding this glial crosstalk can provide important insights into the non-neuronal drivers of neurodegeneration and help the development of targeted interventions aimed at restoring glial homeostasis.

2.1 Astrocyte Function

Astrocytes are the most populous glial cells in the CNS, accounting for almost half of all brain cells. Astrocytes are essential for the homeostasis of the CNS through a wide range of functions (Liddelow et al., 2017). One of the crucial roles of these cells is to maintain the integrity of the BBB (Tóth et al., 2023). The BBB is a highly selective barrier formed by endothelial cells, pericytes and astrocytic endfeet, which together regulate the passage of substances between the bloodstream and the brain (Knox et al., 2022).

Astrocytes contribute to BBB maintenance by releasing several signalling molecules, such as angiopoietin-1, glial-derived neurotrophic factor and sonic hedgehog, which improve tight junction integrity and endothelial cell survival (Knox et al., 2022). The astrocytes' endfeet cover over 90% of the brain's capillary surface, allowing them to influence barrier properties (Wilhelm et al., 2016). By regulating BBB permeability, astrocytes play a defensive role in protecting neurones from toxins, pathogens and peripheral immune cells (Ye et al., 2024). Beyond their structural support, astrocytes also regulate cerebral blood flow through interactions with vascular smooth muscle cells and pericytes. They respond to synaptic activity with intracellular calcium signals that can lead to the release of vasoactive substances such as prostaglandins and nitric oxide, hence coupling neuronal demand with blood supply (MacVicar & Newman, 2015). Disruption of these astrocyte-mediated functions can result in impaired nutrient delivery, contributing to neurodegenerative processes (Clarke et al., 2018). Furthermore, astrocytes support endothelial cells by regulating ion and water homeostasis through the expression of channels such as aquaporin-4 and potassium channels, which are found in high abundance in the endfeet of the astrocytes (Zhou et al., 2022). These mechanisms are essential for maintaining the osmotic balance and electrochemical stability in the brain microenvironment (Zhou et al., 2022). Astrocytes also play a role in the immune response at the BBB by modulating the expression of adhesion molecules and cytokines, which can either stimulate or limit leukocyte infiltration (Figure 1). Their ability to sense and respond to inflammatory cues makes them active participants in the CNS immunity and surveillance (Cekanaviciute & Buckwalter, 2016).

Overall, astrocytes serve as indispensable regulators of BBB integrity, function and signalling. Their disruption has been associated with a range of neurological disorders, including AD, where BBB breakdown is a contributing factor to disease progression (Wang et al., 2022).

Understanding astrocyte involvement in BBB regulation is therefore fundamental to comprehending their role in both health and disease.

2.2 Astrocyte in AD

In AD, astrocytes display several changes in structure and function (Liddelow et al., 2017). These changes result in reactive astrogliosis, which represents a range of responses that can vary from protective to detrimental (Ferrer, 2017). In early diseased stages, astrocytes act to contain damage by isolating plaques and secreting anti-inflammatory molecules. However, in later stages, they often contribute to pathology via the loss of supportive functions and acquiring a more toxic phenotype (Ferrer, 2017).

One of the concerning phenotypic shifts in AD is the transformation of astrocytes into the A1 subtype. First described by Liddelow et al., 2017, A1 astrocytes are induced by microglia-derived cytokines and are characterised by the upregulation of genes associated with the classical component cascade, such as C3 (Liddelow et al., 2017). These astrocytes lose their ability to promote neuronal survival, synapse formation and phagocytic clearance, and instead secrete neurotoxic factors that damage neurones and oligodendrocytes (Liddelow et al., 2017). A1 astrocytes are identified by transcriptomic markers, including Serping1, Ggta1 and Amigo2, and are significantly abundant near Aβ plaques in both AD mouse models and postmortem tissue (Matusova et al., 2023). Their presence correlates with regions of synaptic loss, suggesting a causal role in neurodegeneration (Matusova et al., 2023). Astrocytes in AD also display impaired glutamate uptake, increased oxidative stress and altered calcium signalling (Rothstein et al., 1996). Moreover, reactive astrocytes can compromise the BBB integrity by altering their interaction with endothelial cells and pericytes, potentially increasing vascular permeability and immune cell infiltration, thus causing more damage through an immune response (Figure 1) (Cekanaviciute & Buckwalter, 2016). Despite their harmful potential, not all reactive astrocytes are A1. Some may adopt an A2 phenotype, which is thought to promote tissue repair and release neurotrophic factors (Chang et al., 2023). However, the conditions under which astrocytes shift between these states remain poorly understood, and whether these phenotypes are fixed endpoints or dynamic transition states is an ongoing area of research.

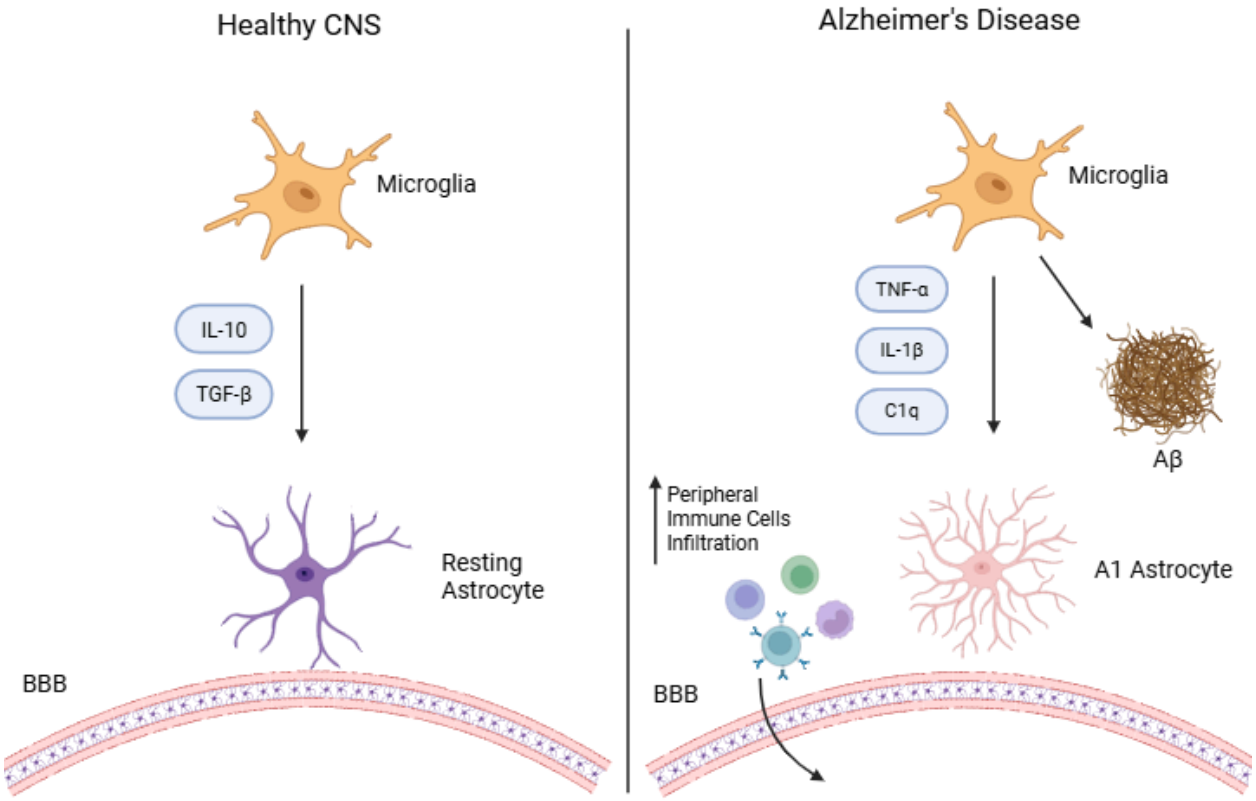

Figure 1: Microglia-Astrocyte Crosstalk in Health and in Alzheimer's Disease. In a healthy CNS, microglia secrete anti-inflammatory cytokines such as IL-10 and TGF-β to maintain astrocytes in a resting, supportive state and preserve the integrity of the BBB. In AD, microglia become activated by Aβ plaques and release pro-inflammatory cytokines TNF-α, IL-1β, and C1q. These cytokines induce the transformation of resting astrocytes into A1 astrocytes, which are neurotoxic. The inflammatory environment also compromises BBB integrity, allowing peripheral immune cells to infiltrate the CNS, exacerbating neurodegeneration. CNS: central nervous system, IL-10: interleukin-10, IL-1β: interleukin-1 beta, TNF-α: tumor necrosis factor-alpha, TGF-β: transforming growth factor-beta, C1q: complement component 1q, Aβ: amyloid-beta, BBB: blood–brain barrier.

2.3 Microglia Function

Microglia originate from yolk sac progenitors during early embryogenesis and are distinct from peripheral immune cells (Askew & Gomez-Nicola, 2018). In the healthy brain, microglia perform a constant surveillance function for signs of infection, injury or cellular debris (Hu et al., 2023). They are essential for brain development, synaptic pruning, and immune defence (Cignarella et al., 2020).

Microglia are highly plastic cells that adapt to environmental signals (Olson & Miller, 2004). They express a variety of pattern recognition receptors (PRRs), including Toll-like receptors, which

allow them to detect DAMPs and PAMPs (Olson & Miller, 2004). Upon activation, microglia change morphology, upregulate MHC molecules and produce a range of bioactive factors (Zhang et al., 2023). In early life, microglia contribute to synaptic refinement by engulfing unnecessary synapses, a process known as synaptic pruning (Hong et al., 2016). This process is mediated by complement proteins such as C1q and C3, which bind and tag the synapses for phagocytosis (Hong et al., 2016). Microglia also secrete growth factors such as BDNF and IGF-1, supporting neuronal survival and plasticity (Hu et al., 2019). Recent single-cell RNA sequencing has challenged the M1/M2 paradigm, revealing a spectrum of microglial activation states (Sun et al., 2023). Among these, the disease-associated microglia (DAM) phenotype has gained attention in AD (Sun et al., 2023). DAMs are characterised by increased expression of Trem2, Apoe and CD33, and reduced expression of homeostatic markers (Sun et al., 2023). DAMs appear around Aβ plaques and may initially aid in clearance, but their prolonged activation can contribute to inflammation and tissue damage (Sun et al., 2023).

2.4 Microglia in AD

Microglia are the primary immune cells of the CNS, where they play crucial roles in immune surveillance, synaptic remodelling and tissue repair (Baik et al., 2019; Hu et al., 2023). In the healthy brain, microglia maintain homeostasis by constantly monitoring their environment, clearing apoptotic cells and debris, as well as shaping neuronal networks through synaptic pruning (Hong et al., 2016; Cignarella et al., 2020). However, in AD, microglial responses become dysregulated and change from protective to pathogenic, making them key players in disease progression (Stewart et al., 2010; Kim & Lee, 2024).

One of the earliest detectable pathological events in AD is the extracellular accumulation of Aβ peptides, which aggregate into insoluble plaques (Zhang et al., 2022). Microglia are highly sensitive to these aggregates and are among the first cells to react. In the initial stages of AD, microglia surround Aβ plaques and attempt to clear them via phagocytosis (Figure 1). This early response may be neuroprotective, as it helps contain Aβ toxicity and reduces its spread within the brain parenchyma (Kim & Lee, 2024). Microglia also secrete anti-inflammatory cytokines like IL-10 and TGF-β (Figure 1) and neurotrophic factors that support neurons and glial health in the early phases of AD (Hu et al., 2019).

However, with persistent exposure to Aβ and age-related decline in immune efficiency, microglial function begins to diminish in effectiveness. Over time, their phagocytic capacity

declines, and they shift into a chronically activated state (Gao et al., 2023). This dysfunctional microglial phenotype is characterised by prolonged secretion of pro-inflammatory cytokines, oxidative stress mediators, and reduced debris-clearing ability (Gao et al., 2023). These changes establish a neurotoxic environment and lead to a feed-forward cycle of chronic inflammation, tissue damage, and further Aβ accumulation.

Activated microglia in AD produce several pro-inflammatory cytokines, among which the most influential are IL-1β, TNF-α, and C1q, as well as chemokines like CCL2 (Liddelow et al., 2017). These cytokines have several effects on the CNS and other glial cells. They alter neuronal signalling, worsen synaptic loss through pruning, and induce surrounding astrocytes to shift into the neurotoxic A1 phenotype. The transformation of astrocytes is especially detrimental, as A1 astrocytes lose their supportive roles and begin releasing factors that promote neuronal apoptosis, impair synaptic function, and weaken the integrity of the BBB (Figure 1). All these effects of the reactive astrocyte phenotype contribute to the neurotoxicity, causing the symptoms of memory loss and decline in cognitive function seen in AD. In addition to cytokine production, microglia generate reactive oxygen species (ROS) and reactive nitrogen species (RNS), such as nitric oxide and superoxide. While these molecules can help eliminate pathogens or damaged cells, their excessive accumulation in the AD brain leads to oxidative damage to DNA, lipids, and proteins. This oxidative stress further disrupts mitochondrial function in neurons and glial cells, contributing to energy deficits, neuronal death, and cognitive impairment (Kim & Lee, 2024).

Another pathological function of activated microglia in AD is the dysregulated pruning of functional synapses. Normally, microglial synaptic pruning is a tightly regulated process important for brain development and plasticity (Hong et al., 2016). However, in AD, complement proteins like C1q and C3 mark active synapses for removal, even in the absence of injury. Microglia then phagocytose these tagged synapses, leading to the loss of critical neural connections, particularly in regions like the hippocampus and prefrontal cortex that are essential for memory and learning. This aberrant synapse loss contributes directly to the hallmark cognitive symptoms of AD (Clarke et al., 2018). Importantly, microglia also influence the permeability and integrity of the BBB. In their activated state, they release signals that can disrupt tight junction proteins in endothelial cells, increasing vascular permeability. This allows peripheral immune cells to infiltrate the CNS, further amplifying the pro-inflammatory response. Increased BBB permeability is a common feature of AD and correlates with disease severity. The extent of microglial contribution to AD pathology is further underscored by genetic studies.

Mutations in TREM2 impair microglial lipid sensing and phagocytosis, leading to reduced plaque compaction and increased neurotoxicity (Krasemann et al., 2017). CD33 is another AD risk gene that negatively regulates phagocytosis and promotes inflammatory signalling (Griciuc et al., 2019). APOE4, the strongest genetic risk factor for sporadic AD, influences microglial response to injury and modulates cytokine release (Krasemann et al., 2017). Moreover, ageing reduces the efficiency of microglial homeostatic programs, and genetic vulnerabilities build up upon this decline, contributing to a less effective immune response in the ageing brain. This impaired clearance is one of the key reasons why AD is considered a disease of ageing.

Overall, microglia in AD undergo a functional shift from protective to harmful. They fail to clear Aβ efficiently, contribute to synapse loss, promote oxidative stress, and induce reactive astrocytes, all of which synergistically help neurodegeneration (Sun et al., 2023). Their central role in glial crosstalk and maintaining inflammation highlights them as an important factor in AD pathology. Thus, understanding the molecular signals that govern microglial activation and communication with astrocytes is essential for elucidating their role in AD and for developing targeted therapies aimed at restoring glial balance in the AD brain.

2.5 Glial Genes Associated with AD

Recent genetic analysis identified several genes that are involved in modulating disease risk and progression in AD. Among the most important are TREM2, APOE and CD33, which play a role in microglial phenotype, phagocytosis ability and inflammatory response (Sun et al., 2023; Krasemann et al., 2017; Griciuc et al., 2019).

TREM2 is a transmembrane receptor expressed on microglia, which plays a vital role in lipid sensing, phagocytosis and immune regulation (Cignarella et al., 2020). It is a member of the immunoglobulin superfamily and forms a complex with the adaptor protein DAP12 to initiate downstream signalling cascades for inflammatory modulation, proliferation and cytoskeletal reorganisation (Cignarella et al., 2020). In AD, TREM2 facilitates microglial response to amyloid plaques, thus promoting plaque removal and preventing neuronal damage by increasing microglial clustering around plaques, promoting phagocytosis and helping the transition of the microglia to a DAM phenotype (Sayed et al., 2021).

These crucial processes are impaired by TREM2 mutations, which lead to a loss of function (Sayed et al., 2021). Microglia that are deficient in TREM2 display lower migration and surveillance ability as well as lower expression of DAM-associated genes, thus they are not able

to clear the Aβ plaques as efficiently through phagocytosis, resulting in a more neurotoxic amyloid environment and neurodegeneration (Sayed et al., 2021; Qin et al., 2021) (Figure 2). Moreover, the loss of TREM2 leads to an increased release of pro-inflammatory cytokines (Figure 2) and oxidative stress mediators, creating a harmful neuroinflammatory context (Sayed et al., 2021; Qin et al., 2021). These impairments contribute to an increased risk of developing AD and suggest that TREM2 is a key regulator of microglial homeostasis in the AD brain (Qin et al., 2021).

APOE is another critical gene that regulates microglial function in AD (Krasemann et al., 2017; Shi et al., 2019). Under homeostatic conditions, APOE is abundantly expressed by astrocytes. However, its expression is significantly upregulated in microglia in the context of neurodegeneration (Shi et al., 2019). This gene encodes apolipoprotein E, which is a protein involved in lipid transport, cholesterol redistribution and repair of neuronal membranes (Belloy et al., 2023). The APOE gene has three major isoforms, namely APOE2, APOE3 and APOE4. The APOE4 allele is the strongest genetic risk factor for late-onset sporadic AD (Shi et al., 2019). In microglia, APOE4 contributes to a pro-inflammatory and dysfunctional phenotype (Krasemann et al., 2017). It impairs the efflux of cholesterol via the ABCA1 and ABCG1 transporters, causing intracellular lipid accumulation (Raulin et al., 2022). This lipid dysregulation leads to the formation of lipid droplets, disrupts mitochondrial function, and makes microglia metabolically stressed, hence causing them to adopt an inflammatory state (Zhang et al., 2025). Moreover, microglia that express APOE4 exhibit an increased expression of inflammatory cytokines, such as TNF-α and IL-6, as well as a lower capacity to phagocytose Aβ plaques (Zhang et al., 2025) (Figure 2). In contrast, microglia expressing APOE2 or APOE3 are more efficient at Aβ uptake and less predisposed to chronic inflammatory activation (Wang et al., 2022).

Furthermore, APOE interacts with TREM2 in lipid sensing and phagocytosis (Nguyen et al., 2020). Krasemann et al., 2017, suggest that APOE acts as a ligand for TREM2, and that this APOE-TREM2 axis may regulate the switch from homeostatic to a DAM state (Krasemann et al., 2017). The dysregulation of this axis in cells that express APOE4 could impair the beneficial aspects of the DAM activation and push the microlgia towards a harmful, neurodegenerative phenotype (Krasemann et al., 2017). Overall, APOE4 promotes a feed-forward loop regarding inflammation, lipid dysregulation, and impaired clearance of amyloid plaques, aggravating neuronal injury and astrocyte reactivity in AD (Shi et al., 2019).

CD33 is a sialic acid-binding immunoglobulin-like lectin (Siglec-3) expressed on the surface of microglia, and its primary function is that of an inhibitory receptor (Eskandari-Sedighi et al., 2022; Griciuc et al., 2020). It contains an intracellular immunoreceptor tyrosine-based inhibitory motif that propagates signals inside the cell to dampen the immune cell activity (Eskandari-Sedighi et al., 2022; Griciuc et al., 2020). In the CNS, CD33 regulates microglial activation, phagocytosis and inflammatory responses (Griciuc et al., 2020). CD33 is associated with increased susceptibility to late-onset AD (Naj et al., 2011). In microglia, CD33 plays a suppressive role in Aβ removal (Griciuc et al., 2020). Elevated CD33 expression inhibits the phagocytosis of Aβ plaques (Figure 2) by downregulating pathways required for lysosomal activity and cytoskeletal reorganisation (Griciuc et al., 2020). More specifically, CD33 interferes with the TREM2-DAP12 signalling pathways and reduces the expression of genes involved in endocytosis (Griciuc et al., 2019). Therefore, this impaired clearance contributes to the accumulation of extracellular Aβ plaques. Furthermore, CD33 signalling promotes a pro-inflammatory microglial phenotype (Eskandari-Sedighi et al., 2022; Griciuc et al., 2020) (Figure 2). CD33 activation causes the recruitment of SHP-1 and SHP-2 phosphatases, which suppress signalling cascades downstream of activating receptors. This mechanism dampens the protective response while triggering the secretion of inflammatory cytokines, thus contributing to chronic neuroinflammation (Eskandari-Sedighi et al., 2022). Additionally, CD33 seems to act antagonistically to TREM2. While TREM2 enhances microglial activation and Aβ clearance, CD33 dampens these processes (Griciuc et al., 2019). This relationship suggests that microglial responses in AD could be brought back in balance by targeting CD33, possibly in combination with TREM2 modulation. Genetic knockout of CD33 in mouse models showed enhanced microglial Aβ phagocytosis and lowered plaque burden, making CD33 a promising immunomodulatory target in AD (Griciuc et al., 2020).

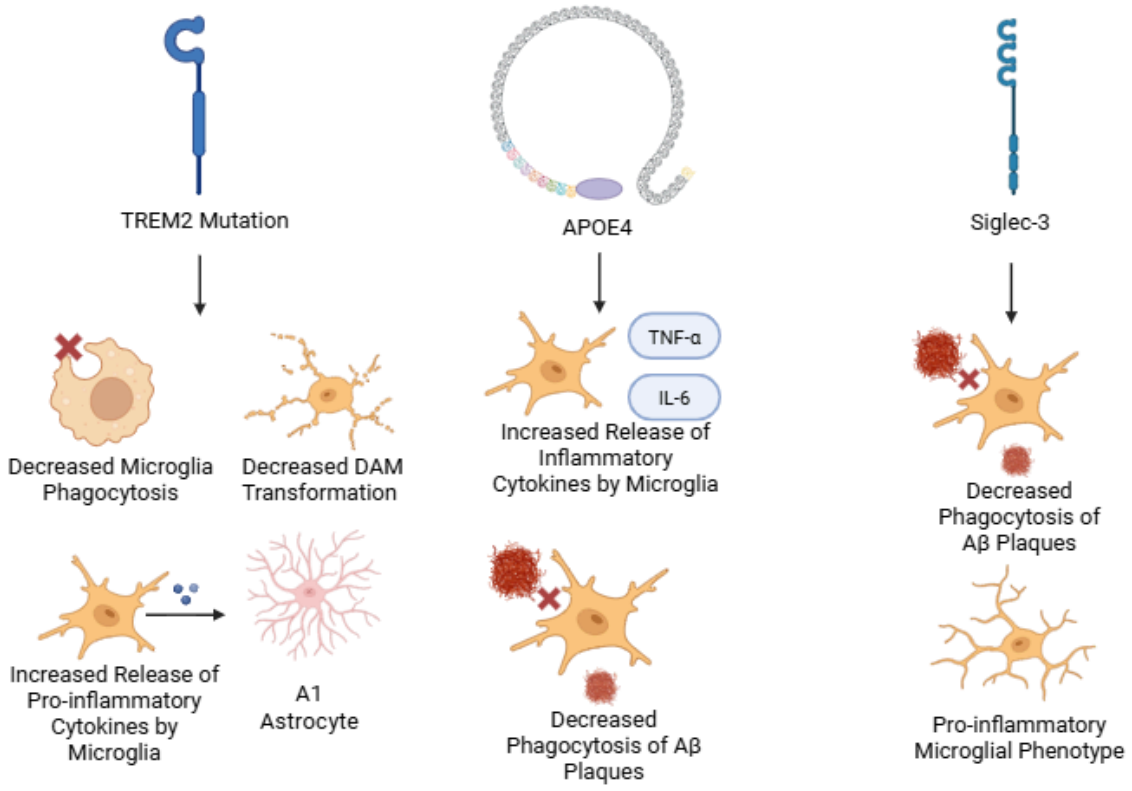

Figure 2: Genes Correlated with Changes in Microglia that Promote AD. Mutations in key AD risk genes alter microglial behaviour and contribute to disease pathology. TREM2 mutations reduce microglial phagocytosis and impair transition to the DAM phenotype, while increasing the release of pro-inflammatory cytokines that can induce the A1 astrocyte transformation. APOE4 expression in microglia promotes the release of inflammatory cytokines such as TNF-α and IL-6 and impairs Aβ plaque clearance. Siglec-3 (CD33) suppresses Aβ endocytosis and promotes a pro-inflammatory phenotype in microglia. Thus, these genetic changes contribute to chronic neuroinflammation and reduced amyloid clearance in AD. TREM2: triggering receptor expressed on myeloid cells 2, DAM: disease-associated microglia, APOE4: apolipoprotein E ε4 isoform, IL-6: interleukin-6, TNF-α: tumour necrosis factor-alpha, Aβ: amyloid-beta, Siglec-3: CD33.

2.6 Microglia-Astrocyte Communication in AD

In the healthy CNS, microglia and astrocytes engage in dynamic, two-way communication that supports tissue homeostasis (Garland et al., 2022). This relationship helps regulate synaptic function, vascular integrity, and immune responses. Under non-pathological conditions, microglia promote astrocytic homeostasis through the release of anti-inflammatory cytokines such as IL-10 and TGF-β, which keep astrocytes in a quiescent, neuroprotective state (Butovsky et al., 2014; Garland et al., 2022). In turn, resting astrocytes support microglial surveillance

functions, buffer ions and neurotransmitters, and contribute to BBB stability (Liddelow et al., 2017; Tóth et al., 2023).

However, in AD, this glial communication becomes disrupted. The accumulation of Aβ plaques and other DAMPs activates microglia, which undergo a phenotypic shift toward a pro-inflammatory state. As a result, the nature of microglia–astrocyte communication changes drastically towards a neurodegenerative phenotype. One of the most influential consequences of this shift is the induction of a neurotoxic astrocyte phenotype known as A1 (Figure 1) (Liddelow et al., 2017). Unlike reactive astrocytes that aim to protect and repair the CNS, A1 astrocytes lose their supportive functions and gain the ability to secrete factors that are toxic to neurons and oligodendrocytes. The transformation into the A1 phenotype is instigated by three key cytokines secreted by activated microglia: IL-1β, TNF-α, and C1q. These cytokines act together to reprogram astrocytes at the transcriptional level, triggering a loss of trophic support and the acquisition of a pro-degenerative profile (Liddelow et al., 2017).

On a molecular level, IL-1β binds to the IL-1 receptor on astrocytes, activating the NF-κB signalling pathway. This results in the transcription of a host of pro-inflammatory genes and downregulation of neurotrophic factors that normally promote neuronal survival and synaptogenesis (Guo et al., 2024). TNF-α further amplifies the NF-κB response, but also signals through the TNF receptor 1, which can initiate apoptotic cascades in both astrocytes and neurons (Zhang et al., 2018). C1q, a component of the classical complement pathway, binds to specific receptors on astrocytes and initiates a signalling cascade that enhances local inflammation and primes the astrocyte for further complement activation (Clarke et al., 2018). The result of this cytokine-driven transformation is a transcriptional profile marked by the upregulation of genes such as C3, Amigo2, and Serping1, and the downregulation of genes associated with synaptic support and neuroprotection. A1 astrocytes produce C3, which binds to neuronal synapses and marks them for microglial-mediated pruning, contributing to synapse loss in the hippocampus (Clarke et al., 2018). In addition to synaptic degeneration, A1 astrocytes may impair glutamate uptake, promote oxidative stress, and reduce the integrity og the BBB. These functional deficits worsen the hostile brain microenvironment in the AD brain, exacerbating the neuronal damage initiated by amyloid plaques and tau pathology. Furthermore, the conversion to A1 astrocytes appears to be a relatively stable and possibly cause irreversible damage, making early intervention critical (Liddelow et al., 2017). Animal studies have demonstrated that blocking any one of the three influential cytokines can prevent the conversion of astrocytes into the A1 phenotype. In models where these cytokines were genetically deleted

or pharmacologically inhibited, the number of A1 astrocytes was significantly reduced, and markers of neuronal health and synaptic density were preserved (Liddelow et al., 2017; Carpanini et al., 2019). These findings underscore the therapeutic potential of targeting glial crosstalk as a means of slowing or halting AD progression.

Overall, the microglia astrocyte communication is a crucial axis in AD pathology. The release of IL-1β, TNF-α, and C1q by activated microglia drives astrocytes into a neurotoxic A1 state, contributing to synaptic loss, BBB dysfunction, and neuronal death. The dysregulation of this signalling represents a key mechanism in AD, leading the way for a potential target in future targeted therapies.

3. Discussion

Alzheimer's disease is fundamentally a neurodegenerative disorder of failed clearance of Aβ plaques or an increased production of these misfolded proteins (Hampel et al., 2021). As humans age, the brain's immune system, which is primarily governed by astrocytes and microglia, becomes less efficient (Gao et al., 2023). This immunosenescence contributes to the ineffective removal of Aβ plaques, which begin to accumulate before the patients experience their first clinical symptoms (Zhang et al., 2021). At the same time, the production of Aβ plaques increases, either due to genetic predisposition or altered neuronal activity (Hampel et al., 2021). These imbalances cause the gradual buildup of plaques and trigger a chronic inflammatory response, which further impairs cellular function and structural integrity in the brain, ultimately affecting memory and learning, leading to dementia (Hampel et al., 2021).

Microglia, as the primary immune cells of the brain, are pivotal to both protective and pathological processes in AD. They play a crucial role in immune surveillance, debris clearance and synaptic pruning, a process by which diseased or unnecessary synaptic connections are eliminated to refine neural circuits, especially during learning and memory consolidation. However, in AD, these mechanisms become dysregulated. Activated microglia begin to prune functional synapses excessively. This is particularly obvious in the hippocampus and prefrontal cortex, regions of the brain which are heavily involved in memory and cognition (Cornell et al., 2022). Therefore, this will result in a loss of synaptic density, which contributes directly to cognitive symptoms like memory loss, confusion, and impaired decision-making (Cornell et al., 2022).

The reviewed literature highlights the crucial role of microglia-derived cytokines IL-1β, TNF-α, and C1q in initiating astrocyte dysfunction and neurotoxicity (Liddelow et al., 2017). These cytokines act in concert to convert astrocytes from supportive, homeostatic cells into the neurotic phenotype A1. This transformation can be identified by the upregulation of genes such as Serping1, Ggta1 and Amigo2, which leads to the loss of astrocytic functions that are critical to neuronal health, including synapse support, neurotransmitter recycling, and BBB maintenance (Matusova et al., 2023). A1 astrocytes actively secrete factors that can induce apoptosis in neurones and oligodendrocytes. Moreover, A1 astrocytes have been identified in the vicinity of amyloid plaques in both transgenic mouse models and postmortem AD patient brains, suggesting a direct contribution to local neurodegeneration (Matusova et al., 2023). Furthermore, the C3 released by A1 astrocytes can bind synaptic terminals, thus marking them

for phagocytosis by microglia (Chen et al., 2024). This feedback loop, where microglia activate astrocytes to become neurotoxic, which in turn tag synapses for elimination, could explain how inflammation contributes not only to neuron loss but to the widespread damage to the neuronal networks underlying cognition.

From a genetic perspective, mutations in microglial genes such as TREM2, APOE, and CD33 further influence these processes. TREM2 is needed for the microglial transition to a phagocytotic DAM state that helps clear Aβ plaques and limit their spread. Mutations in TREM2 impair this transformation, resulting in lower plaque compaction and increased neurotoxicity. Similarly, APOE4 is one of the most common genetic risk factors for sporadic AD, and it is responsible for modifying microglial lipid uptake and enhancing pro-inflammatory responses, thus worsening microglial dysfunction and astrocyte activation. On the other hand, CD33 suppresses phagocytosis and promotes a chronic inflammatory state. These gene mutations not only increase disease risk but also affect the glial cells in ways that support the pathological feedback loop of inflammation, plaque accumulation and synapse loss.

This pathological glial interaction model allows for possible targets for therapeutic interventions. Preclinical studies showed that knocking out IL-1β, TNF-α, or C1q prevents A1 astrocyte formation and reduces neuronal damage, thus suggesting that regulating these cytokines could mitigate disease progression (Liddelow et al., 2017). However, IL-1β and TNF-α are also crucial for normal immune responses and synaptic plasticity. Therefore, long-term inhibition in the CNS may cause side effects and an increased risk of infection susceptibility and impaired neuroplasticity. In addition, targeting proteins in the complement cascade, for example, C1q or C3, has shown promising results in reducing aberrant synaptic pruning (Zelek & Tenner, 2025). However, complement proteins are essential for immunity and early neuronal development (Zelek & Tenner, 2025). These potential side effects raise the therapeutic challenge of timing and specificity of targeting these pathways to slow AD progression without consequences. One of the key difficulties in this therapeutic potential is glial heterogeneity. Not all astrocytes in the AD brain adopt the A1 phenotype, and microglia also exhibit a continuum of states beyond the old-fashioned activated or homeostatic phenotypes (Sun et al., 2023; Chang et al., 2023). Some microglia keep their neuroprotective roles even in disease, while others become maladaptive. Similarly, the A2 astrocyte phenotype has been proposed as neuroprotective, promoting tissue repair and survival. A more in depth understanding of how glial phenotypes are regulated is needed to avoid interventions that could suppress the protective functions alongside the neurotoxic ones. Another open question is whether the A1 astrocytes can be reprogrammed. If

the neurotoxic phenotype is not terminal, then therapies aimed at restoring the homeostasis state rather than eliminating A1 astrocytes may be more beneficial. Recent research has explored the transcriptional reprogramming, metabolic intervention, and anti-inflammatory conditioning strategies to shift the astrocyte state (Tyzack et al., 2017; Calì et al., 2024). Similarly, enhancing TREM2 signalling or inhibiting CD33 could restore microglial phagocytic function and reduce inflammation overall.

Methodological limitations must also be considered. Much of the literature research stems from mouse models, which are informative, but do not fully mimic the human AD pathology (Humpel & Foidl, 2020). Advancements in human stem cell-derived glial models and single cell transcriptomics show potential but also reveal additional complexity, such as sex-specific responses and brain region-specific vulnerabilities (Nunes et al., 2022; Su et al., 2022). Future studies must incorporate these aspects to develop therapies that can be translated into the clinic.

Beyond IL-1β, TNF-α, and C1q, many other molecules contribute to the inflammatory microenvironment in AD. ATP, released from dying neurones, acts on P2X and P2Y receptors to activate both astrocytes and microglia (Franke et al., 2007). Fractalkine signalling, extracellular vesicles, and reactive oxygen species further modulate glial responses (Paul & Basavan, 2022). These factors likely interact in a broader, complex signalling network that governs glial behaviour and may amplify or modulate cytokine-driven pathways. In conclusion, the microglia astrocyte interaction is a central mechanism in AD, particularly the induction of A1 astrocytes by IL-1β, TNF-α, and C1q. This cytokine-driven glial reprogramming promotes synapse pruning, neuronal death and neuronal network dysfunction, hence directly contributing to the hallmark cognitive decline in AD. These processes are exacerbated by genetic risk factors and immunosenescence due to ageing, making them mechanistically important and clinically relevant. While glial signalling opens the door for promising therapeutic targets, the researchers must be careful when exploring the spectrum of roles of these cells. An effective intervention will require precise, context-dependent modulation of glial behaviour, ideally before irreversible damage occurs. Ongoing research into the molecular and temporal specificity of glial phenotypes will be essential to take advantage of their potential in combating AD.

4. Reference List

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