REVIEW

The pathogenic role of the inflammasome in neurodegenerative diseases

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Abstract

The inflammasome is a large macromolecular complex that contains multiple copies of a receptor or sensor of pathogen-derived or damage-derived molecular patterns, pro-caspase-1, and an adaptor called ASC (apoptotic speck containing protein with a CARD), which results in caspase-1 maturation. Caspase-1 then mediates the release of pro-inflammatory cytokines such as IL-1β and IL-18. These cytokines play critical roles in mediating immune responses during inflammation and innate immunity. Broader studies of the inflammasome over the years have implicated their roles in the pathogenesis of a variety of inflammatory diseases. Recently, studies have shown that the inflammasome modulates neuroinflammatory cells and the initial stages of neuroinflammation. A secondary cascade of events associated with neuroinflammation (such as oxidative stress) has been shown to activate the inflammasome, making the inflammasome a promising therapeutic target in the modulation of neurodegenerative diseases. This review will focus on the pathogenic role that inflammasomes play in neurologic diseases such as Alzheimer's disease, traumatic brain injury, and multiple sclerosis.

Keywords: IL-1β, inflammasome, neurodegenerative diseases.

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Introduction to Inflammasomes and Neuroinflammation

Nucleotide-binding leucine-rich repeat containing (NLR), also known as NOD-like, receptors are a class of cytosolic sensors or receptors that respond to a variety of pathogen-associated molecular patterns (PAMPs) which are linked to various microbes as well as damage-associated molecular patterns (DAMPs) produced during tissue-based injury. There are more than 20 NLR genes in humans and more than 30 in mice. The structure of NLRs consists of a tripartite domain containing a variable N-terminal effector domain, a central nucleotide-binding domain, and a C-terminal domain consisting of variable leucine-rich repeats. NLRs are classified into various subgroups each with their own unique responses to PAMPs and DAMPS (Davis *et al.* 2011).

One of the most extensively studied classes of NLRs is the inflammasome-forming NLRs. Upon sensing DAMPs and PAMPs, these NLRs mediate the release of pro-inflammatory cytokines IL-1β and IL-18. These NLRs include NLRP1, NLRP3, NLRC4, NLRC5, NLRP6, NLRP7, NLRP12, as well as the non-NLR inflammasome receptor known as AIM2. The signal specificity and functional roles of NLRP1, NLRC4,

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Abbreviations used: AD, Alzheimer's disease; APP, amyloid precursor protein; ASC, apoptosis-associated speck-like protein containing a C-terminal caspase recruitment domain [CARD]; ATP, adenosine triphosphate; Aβ, amyloid-β; CSF, cerebrospinal fluid; DAMP, damage-associated molecular patterns; EAE, experimental autoimmune encephalomyelitis; GWAS, genome-wide association studies; MS, multiple sclerosis; NLR, nucleotide-binding leucine-rich repeat containing or NOD-like receptors; PAMP, pathogen-associated molecular patterns; PP, primary progressive; PR, progressive relapsing; PS1, presenilin-1; ROS, reactive oxygen species; RR, relapse remitting; SP, secondary progressive; TBI, traumatic brain injury.

AIM2, and in particular NLRP3, have been well characterized (Franchi *et al.* 2006; Martinon *et al.* 2006; Miao *et al.* 2006; Duncan *et al.* 2007; Dostert and Petrilli 2008; Fernandes-Alnemri *et al.* 2009; Hornung *et al.* 2009; Martinon 2010; He *et al.* 2012; Faustin and Reed 2013; Munoz-Planillo *et al.* 2013; Cirelli *et al.* 2014). The signal specificity and functional roles of other inflammasome-forming NLRs have yet to be fully elucidated. Upon sensing a PAMP or DAMP, an NLR forms a multimeric protein complex known as the inflammasome through the association of the adaptor protein ASC (apoptosis-associated speck-like protein containing a C-terminal caspase recruitment domain [CARD]). This initiates the cleavage of pro-caspase-1 into its active and mature form caspase-1. Recently, elegant biochemical, structural, electron microscopic, and functional analyses have shown that ASC serves as an enucleating template which associates with the other components of the inflammasome to form a fibril or prion-like multimeric, stacked structure (Cai *et al.* 2014; Lu *et al.* 2014). Active caspase-1 is then able to cleave the immature forms of IL-1β and IL-18 into their mature forms. In recent years, mouse model studies of inflammatory diseases and genome-wide association studies have implicated the inflammasome in the pathogenesis of various inflammatory diseases ranging from inflammatory bowel disease to asthma (Chaput *et al.* 2013; Davis *et al.* 2014).

IL-1β is known to cause the proliferation of macrophages, and neuroinflammatory cells such as microglia and astrocytes (Feder and Laskin 1994). These cells are recruited to the site of injury or inflammation within the central nervous system. This represents one of the initial signature events during neuroinflammation and a hallmark of pathogenesis associated with various neurodegenerative diseases. Regulation of IL-1β and IL-18 may play a role in attenuating and or balancing the innate immune response during neuroinflammation. As the inflammasome mediates the release of IL-1β and IL-18 (both of which can trigger a cascade of secondary inflammatory events in neuroinflammation), the inflammasome represents a potential critical mediator of neuroinflammation and a potential therapeutic target of neurodegenerative diseases. Recent studies have begun to validate the pathogenic role of the inflammasome in neurodegenerative diseases. This review will focus primarily on emerging evidence suggesting that NLRP1 (NLR family, pyrin domain containing 1), NLRP3 (NLR family, pyrin domain containing 3), NLRC4 (NLR family, CARD domain containing 4), and AIM2 (absent in melanoma 2) may play a pathogenic role in neuroinflammatory diseases such as Alzheimer's disease (AD), traumatic brain injury (TBI), and multiple sclerosis (MS).

The inflammasome and Alzheimer's disease and disease models

AD is characterized as a neurodegenerative and progressive disease and is the leading cause of dementia. The disease typically affects people 65 years and older. Symptoms associated with the disease include a progressive decline in cognitive function and memory. The pathogenesis of this disease is believed to be the result of a continual accumulation of amyloid-β peptide deposits that form within senile plaques. These senile plaques lead to the disruption of synaptic activity and eventually neuronal death. Other pathogenic markers associated with this disease include the formation of neurofibrillary tangles (Weiner and Frenkel 2006). Although this is the recognized pathology of AD within the field, there still are unknown pathogenic factors that may contribute to the etiology of this disease. Over the years there has been evidence to suggest that the cytokines IL-1β and IL-18 may contribute to the pathogenesis of AD (Blum-Degen *et al.* 1995; Bossu *et al.* 2007; Ojala *et al.* 2009). Other evidence has suggested that microglia may play a key role in initiating AD pathology (Vehmas *et al.* 2003). Microglia are known to be recruited to the site of these senile plaques and secrete IL-1β (Griffin *et al.* 1989; Meyer-Luehmann *et al.* 2008). Figure 1 summarizes the proposed roles of the inflammasome in AD or AD disease models.

One of the initial studies looked to characterize the microglia at these senile plaques and assess if IL-1β secretion was inflammasome dependent (Halle *et al.* 2008). Using microglia that were incubated with fibrillar amyloid-β (Aβ) peptide, the investigators observed IL-1β release, caspase-1 activation, and the formation of ASC complexes. When fibrillar Aβ was injected into *Asc*$^{-/-}$ and *Casp1*$^{-/-}$ mice, there was a significant decrease in microglial accumulation in the brain compared to wild-type mice. These findings suggest that IL-1β and inflammasome-associated proteins influence microglial recruitment at the site of senile plaques in AD. In a subsequent study, it was shown that amyloid precursor protein (APP)/PS1/*Nlrp3*$^{-/-}$ mice showed less caspase-1 cleavage, and less amyloid-β deposits, and enhanced phagocytosis of amyloid-β compared to APP/PS1 mice. This study provided evidence that NLRP3 has an *in vivo* and exacerbating role in the pathogenesis of AD (Heneka *et al.* 2013).

In another study, it was shown through immunoprecipitation experiments of amyloid-β stimulated glial cultures, that there was an observed strong NLRP3 and ASC association. Conversely in these cultures, it was observed that NLRP10′s association with ASC decreased compared to untreated cultures in which NLRP10 showed a strong association with ASC (Murphy *et al.* 2014). NLRP10 is known to inhibit ASC and IL-1β in cell lines, although its function *in vivo* is unclear because of confounding issues with the background genetics of the gene deletion strain (Wang *et al.* 2004; Krishnaswamy *et al.* 2015). The decrease in NLRP10 was believed to be because of cathepsin-mediated degradation. The findings from this study suggest that NLRP10 may act as a negative regulator of NLRP3 inflammasome activation prior to sensing amyloid-β peptides. Studies of clean NLRP10-deficient mice are needed to verify these results *in vivo*.

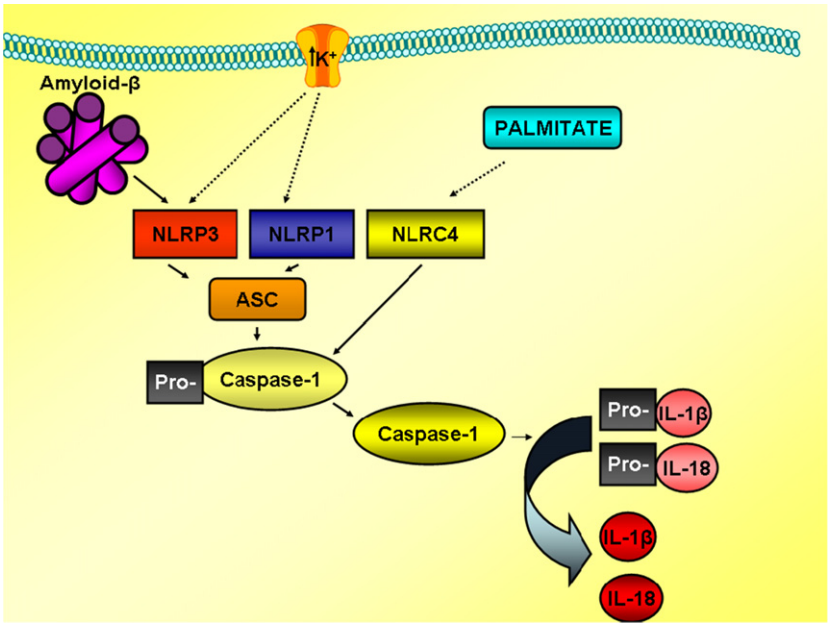

Fig. 1 The role of the inflammasome in Alzheimer's disease (AD). Amyloid-β is known to trigger the activation of NLRP3. The exact trigger for NLRP1 in AD is not known but it may be possible that potassium effluxes which trigger NLRP1 (as well as NLRP3) could occur during AD pathogenesis and in the process trigger NLRP1 activation. Recently it was suggested that palmitate a saturated fatty acid may play a role in activating NLRC4 during AD. These potential triggers may possibly activate NLRP3, NLRP1, and NLRC4 during AD but have yet to be confirmed (dashed lines). NLRC4 can associate with pro-caspase-1. NLRP1 and NLRP3 associate with the adaptor protein ASC. ASC initiates the cleavage of pro-caspase-1 into the mature form of caspase-1, which cleaves pro-IL-1β and pro-IL-18 into their mature forms of IL-1β and IL-18 which have been implicated in the pathogenesis of AD.

The pathogenic role of NLRP3 in AD is established and still developing, but this is not the only NLR that has been suggested to have a pathogenic role in AD. Mutations within the NLRP1 gene may contribute in combination with other known Alzheimer-related genes (such as APP and PS1) to the etiology of AD (Pontillo *et al.* 2012). NLRP1 is known to be highly expressed in the brain, specifically within neurons and oligodendrocytes (Kummer *et al.* 2007). A recent study showed that aged APPswe/PS1d1 mice displayed elevated NLRP1 expression. When NLRP1 si-RNA was injected into APPswe/PS1d1 mice, there was reduced caspase-1 activation, pyroptosis, and improved cognitive function compared to APPswe/PS1d1 mice injected with control si-RNA (Tan *et al.* 2014).

In another study, the inhibition of IPAF (also known as NLRC4) expression in palmitate-treated astrocytes led to decreased IL-1β secretion as well as the reduction in amyloid-β_{42} in primary neurons that were incubated with conditioned media from palmitate-treated astrocytes. In this study, it was also observed that there was elevated expression levels of IPAF and ASC in the post-mortem brain tissue of patients with sporadic AD (Liu and Chan 2014).The authors of this study suggest that palmitate, a saturated fatty acid that is potentially linked to AD pathogenesis, may stimulate IPAF/NLRC4 expression in astrocytes (Geekiyanage and Chan 2011; Geekiyanage *et al.* 2013). They suggested that NLRC4 may be linked to AD pathogenesis.

There has been accumulating evidence to suggest that NLRP3 plays a role in AD and that it responds to the molecules associated with AD such as fibrillar amyloid-β (Halle *et al.* 2008; Heneka *et al.* 2013, 2015). NLRP1 and NLRC4 have also been suggested to play a role in AD as well, but in order to fully elucidate their functional role, NLRC4- and NLRP1-deficient mice will need to be tested in AD mouse models (L. Liu and Chan 2014; Pontillo *et al.* 2012; Tan *et al.* 2014). These aforementioned studies suggest that inflammasome activation may be linked to severity in AD, however, inflammasome activation is not always linked to more disease severity, as one study has found that mice with a transgenic *Il1b* gene displayed chronic IL-1β expression and increased immune cell infiltration in the hippocampus, but this was not accompanied by enhanced neurodegeneration (Shaftel *et al.* 2007). Another study showed that sustained IL-1β over-expression actually resulted in reduced amyloid plaques, but increased tau phosphorylation (Ghosh *et al.* 2013). This underscores the complexity of targeting neuro-immune interactions for therapies.

The inflammasome and traumatic brain injury

TBI can be characterized as physical force to the brain resulting in injury that exceeds the protective capabilities of the brain. Symptoms associated with TBI include dizziness, cognitive deficits, and most commonly headaches. Post-TBI symptoms include memory impairments and behavioral changes (Riggio 2011). Some symptoms may be temporary and resolve, however, some may last for years. TBI can range from being mild (such as a concussion) to severe depending on the extent of physical damage the brain experiences. Physical damage to the brain in TBI initiates a primary insult followed by a secondary cascade of events. The primary insult results in direct neuronal loss and necrotic death (Lozano *et al.* 2015). The secondary cascade of events that occurs after the primary insult can be characterized as neuroinflammatory responses such as the recruitment of microglia and astrocytes to the site of injury, oxidative stress, mitochondrial dysfunction, blood–brain barrier disruption, and the elevation of cytokines (Mbye *et al.* 2008; Chodobski *et al.* 2011; Dasuri *et al.* 2013). Figure 2 shows the proposed roles of the inflammasome in TBI or TBI-related disease models.

As with other neurological diseases, IL-1β and IL-18 have been associated with the pathogenesis of TBI (McClain *et al.* 1987; Yatsiv *et al.* 2002). In one study, mice treated with an anti-ASC antibody immediately after TBI showed a decrease in contusion volume compared to vehicle-treated mice (de Rivero Vaccari *et al.* 2009). Clinical relevance toward the pathogenic role of NLRP1 in TBI, was provided in which there were higher levels of expression of NLRP1 in the cerebrospinal fluid (CSF) of TBI patients that were clinically diagnosed as having a poor or unfavorable outcome while lower expression of these proteins was associated with patients that had a favorable outcome (Adamczak *et al.* 2012). NLRP1 is expressed in neuronal tissue and represents an ideal inflammasome to study in TBI. NLRP3 may also serve as an ideal candidate therapeutic target in TBI.

DAMPs such as oxidative stress and ATP are known to be released during the secondary cascade of events in TBI (Cristofori *et al.* 2005). These DAMPs may trigger NLRP3 activation. This was supported by a study in which there were elevated levels of NLRP3, ASC, and IL-1β release in TBI- induced rat brains (Liu *et al.* 2013). Besides the generation of reactive oxygen species in TBI, other studies suggested that there may be a correlation between elevated circulating plasma DNA and severe TBI (Campello Yurgel *et al.* 2007). AIM2 is a cytosolic sensor of dsDNA (Hornung *et al.* 2009). AIM2 may contribute to the pathogenesis of

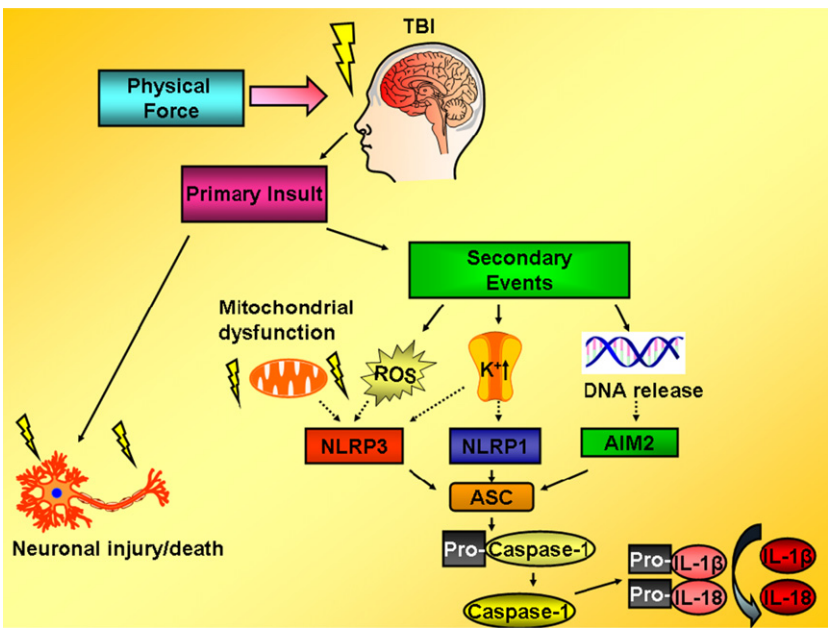

Fig. 2 The role of the inflammasome in traumatic brain injury (TBI). TBI is initiated by physical force exerted to head, this results in a primary insult which results in the immediate injury and death of neurons. The primary insult is followed by a secondary cascade of events following neuroinflammation such as mitochondrial dysfunction, the production of reactive oxygen species (ROS), potassium effluxes and the release of circulating DNA. These events can potentially trigger the activation of NLRP3, NLRP1, and AIM2 during TBI but have yet to be confirmed (dashed lines).NLRP3, NLRP1, and AIM2 associate with the adaptor protein ASC, which initiates the cleavage of pro-caspase-1 to the mature form of caspase-1 which cleaves pro-IL-1β and pro-IL-18 into their mature forms of IL-1β and IL-18. NLRP3, NLRP1, AIM2, IL-1β, and IL-18 have been implicated in the pathogenesis of TBI.

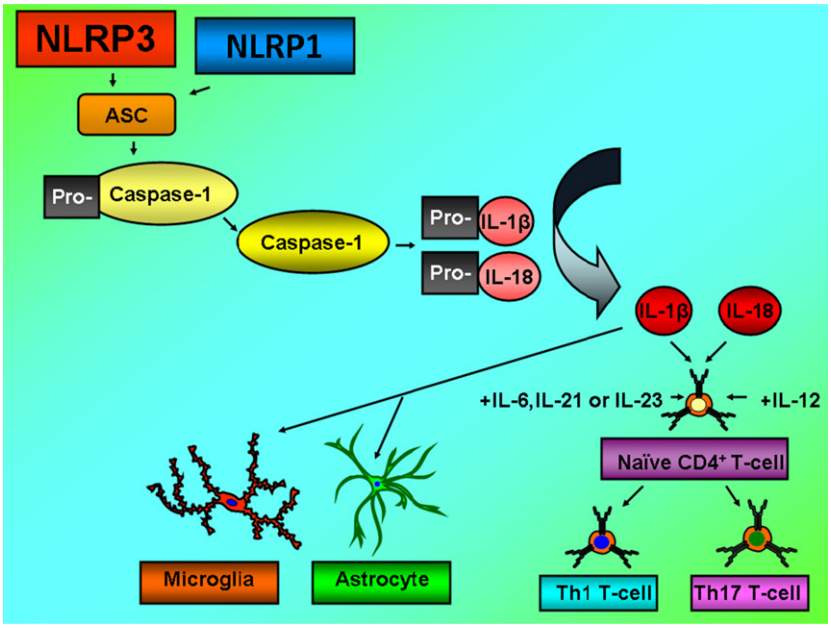

Fig. 3 The role of the inflammasome in multiple sclerosis (MS). NLRP3 and NLRP1 have been implicated in the pathogenesis of MS. NLRP3 and NLRP1 associate with the adaptor protein ASC. ASC initiates the cleavage of pro-caspase-1 into the mature form of caspase-1 which cleaves pro-IL-1β and pro- IL-18 into their mature forms of IL-1β and IL-18. IL-1β is known to drive the proliferation of microglia and astrocytes which are cells that represent hallmarks of neuroinflammation (such as astrogliosis and microglial accumulation). IL-1β in combination with cytokines such as IL-6, IL-21, or IL-23 drive the differentiation of naïve $CD4^+$ T cells into Th17 T cells. IL-18 (also known as the IFN-γ inducing factor) in combination with IL-12 drives the differentiation of naïve $CD4^+$ T cells into Th1 T cells. Microglia, astrocytes, Th1, and Th17 T cells have all been implicated in the pathogenesis of MS.

TBI. In a study, poly (deoxyadenylic-deoxythimidylic acid sodium salt (poly(dA:DT)-stimulated embryonic cortical neurons led to neuronal pyroptosis. It was also observed that neurons that were co-cultured with CSF from TBI patients showed significantly elevated levels of AIM2 and cleaved caspase-1 compared to neurons cultured with CSF from control patients (Adamczak *et al.* 2014). Whether increased AIM2 expression was a result of inflammatory activators such as dead cell debris in the former, or a factor that impacts disease outcome is unclear.

In a recent study, NLRP1 was found to be contained within the exosomes derived from the CSF of spinal cord injury and TBI patients (de Rivero Vaccari *et al.* 2015). In this study, administration of ASC-targeting si-RNA in exosomes to primary rat cortical neurons resulted in decreased ASC expression. Although the study examined protein expression and saw a correlation between elevated inflammasome protein expression and unfavorable TBI pathology, the potential functional roles that inflammasome components may play in TBI remain to be directly elucidated.

Inflammasome and multiple sclerosis

MS is a neuroinflammatory demyelinating disease that affects 1.5 million people worldwide (Bhat and Steinman 2009). MS is a heterogeneous disease in which the progression, pathology, and onset of the disease can vary from patient to patient depending on multiple forms of MS such as relapse remitting, primary progressive, secondary progressive, and progressive relapsing (Denic *et al.* 2011). Although previous studies have suggested a T cell-based pathology, additional hallmarks of MS include blood–brain barrier disruption, demyelination and oligodendrocytic and neuronal defects (Steinman 2008). Although the etiology of the disease remains unknown, clinical studies have suggested that elevated expression of caspase-1, IL-1β, and IL-18 may be associated with the susceptibility, progression, and severity of MS patients (Balashov *et al.* 1999; Losy and Niezgoda 2001; Karni *et al.* 2002; Mann *et al.* 2002). IL-1β is known to promote the differentiation of naïve $CD4^+$ T cells into a subset of Th17 T cells (Sato *et al.* 2011; Shaw *et al.* 2011). IL-18 was originally identified as the IFNγ inducing factor and is able to act in a synergistic fashion with IL-12 to promote the differentiation of naïve $CD4^+$ T cells into Th1 T cells (Shaw *et al.* 2011). Both Th1 and Th17 T cells have been implicated in the pathology of MS.

Previous studies using MS animal models have shown that the presence of inflammasome-associated proteins such as ASC, caspase-1, IL-β, and IL-18 may play an exacerbating role in the pathogenesis of MS (Furlan *et al.* 1999; Shaw

et al. 2010; Lalor *et al.* 2011). Within recent years, growing evidence has suggested that NLRP3 may contribute to the pathogenesis of MS by accelerating demyelination in the cuprizone model which is a T cell-independent model of demyelination, but also in the classical T cell-dependent model by enhancing Th1 and Th17 responses, and inducing the migration of T cells into the CNS which has been shown by Gris and also confirmed in later studies (Gris *et al.* 2010; Jha *et al.* 2010; Inoue *et al.* 2012a).

New data have emerged suggesting that NLRP3 may play a role in current MS therapeutics such as IFN-β. IFN-β represents one of the first lines of therapeutics used to treat RRMS. Its efficacy in the treatment of patients is limited however. This may in part be because of the heterogeneity of MS. The mechanism by which IFN-β works as a therapeutic has not been fully elucidated. Recent evidence has suggested that IFN-β may be able to provide therapeutic benefits by dampening the NLRP3 and NLRP1 inflammasome pathways and by inhibiting IL-1β production (Guarda *et al.* 2011). More recently, it has been suggested that the efficacy of IFN-β therapy in the experimental autoimmune encephalomyelitis model is NLRP3 dependent, and mice without NLRP3 do not benefit from IFN-β treatment (Inoue *et al.* 2012b). Recent clinical evidence was reported suggesting that responsive MS patients treated with IFN-β therapy had elevated NLRP3 and IL-1β expression, while non-responsive patients did not. This suggested that NLRP3 may play a role in the efficacy of IFN-β therapy in MS patients, although the precise mechanism of this involvement is unclear (Malhotra *et al.* 2015). In a subsequent study, a small molecule, MCC950, was identified as an NLRP3 antagonist of IL-1β secretion. MCC950 was found to inhibit NLRP3 activity and attenuate experimental autoimmune encephalomyelitis activity at the same time (Coll *et al.* 2015). To date amongst the inflammasomes, NLRP3 appears to play the strongest pathogenic role in MS. Other inflammasome-forming NLRs such as NLRP1 may play a role in MS but their functional role will need to be further elucidated. Figure 3 shows the proposed roles of the inflammasome in MS disease or MS-related disease models.

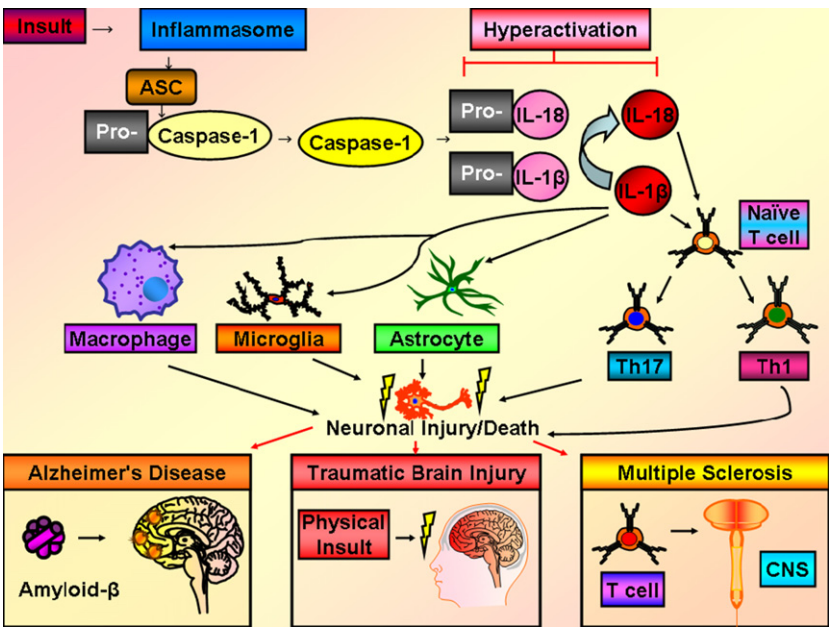

Fig. 4 Regulation of the release of pro-inflammatory cytokines IL-1β and IL-18 by the inflammasome may impact Alzheimer's disease (AD), traumatic brain injury (TBI), and multiple sclerosis (MS) pathology. As mentioned earlier, upon sensing insults (such as ROS, oxidative stress, or released DNA) during neuroinflammation, the inflammasome complex may form consisting of an NLR or non-NLR protein which results in the release of IL-1β and IL-18. IL-1β is known to drive the recruitment and proliferation of macrophages, microglia, and astrocytes during neuroinflammation. IL-18 and IL-1β are known to work in combination with other cytokines to drive the differentiation of naïve $CD4^+$ T cells into Th17 and Th1 T cells which can cause the demyelination and death of neurons during MS. The hyperactivation of IL-1β and IL-18 may lead to the overproduction of these cytokines which will result in excessive proliferation and recruitment of these cell types which eventually will lead to neuronal injury and or death which can exacerbate (as indicated by the red arrows) the various pathologies of neurodegenerative diseases such as AD (a disease in which the accumulation of amyloid-β results in the formation of senile plaques resulting in dementia), TBI (a neurodegenerative disease triggered by physical injury resulting in cognitive deficits), and MS (a T cell-mediated demyelinating autoimmune disease resulting in paralysis).

Concluding Remarks

Each neurodegenerative disease comes with its unique pathogenesis and etiologies, although the inflammasome has been implicated in several neurodegenerative diseases. This review focused on AD, TBI, and MS, although increasing evidence suggests roles for the inflammasome in cerebral ischemic stroke and Parkinson's disease which are not covered here. AD is a progressive neurodegenerative disease in which amyloid-β accumulation leads to the formation of senile plaques and eventually dementia. TBI is a neurodegenerative disease in which physical force results in trauma that exceeds the protective capabilities of the brain. The initial trauma results in immediate neuronal injury and death but triggers a secondary cascade of events such as reactive oxygen species production, and oxidative stress, which further exacerbate neuroinflammation. MS is considered to be a T cell-mediated demyelinating disease which results in the demyelination and death of neurons. Although the progression and pathology associated with these diseases vary, these diseases share commonalities in that their pathologies are heterogeneous and initiated by innate immunology. The cytokines IL-1β and IL-18 have been implicated in the pathogenesis of these different diseases or disease models. These cytokines are critical in the proliferation of neuro-immunoreactive cell types such as microglia and astrocytes which are cell types that respond immediately to neuronal injury and death which occur in all three disease states. The hyper-activation of IL-1β and IL-18 by an inflammasome-forming protein complex can lead to excess production of IL-1β and IL-18 cytokine production which can impact AD, MS, and TBI pathology. Figure 4 summarizes the importance of regulating IL-1β and IL-18 and the impact it may have on AD, TBI, and MS pathology. Excess IL-1β production can lead to an over accumulation of microglia at the site of senile plaques in AD resulting in the eventual neuronal injury and death which contribute to cognitive deficits and dementia associated with AD. Depending on the extent of TBI, the neuroinflammation may resolve on its own, however excess or unresolved neuroinflammation such as the production of cytokines such as IL-1β may lead to the proliferation of microglia, macrophages, and astrocytes at the site of TBI. These neuroinflammatory cells can cause further neuronal injury and death besides the neuronal injury that was caused by the initial primary insult. IL-1β and IL-18 drive the differentiation of naïve $CD4^+$ T cells into Th17 and Th1 T cells which can attack and cause the demyelination and death of neurons in MS. Inflammasome-forming proteins such as AIM2, NLRP1, NLRP3, and NLRC4 play a critical role in mediating the release of the cytokines IL-1β and IL-18 and thus represent potential ideal therapeutic targets. These studies have provided insight into the pathogenic role that NLRP3, NLRC4, NLRP1, and AIM2 may play in neurodegenerative diseases. There are more than nine known inflammasome-forming proteins, but only these four have been well characterized. It is likely that other inflammasome-forming proteins also play important roles during the initial neuroinflammatory stages of these diseases. There is also an added complexity in that the pathogenesis of these diseases may involve more than one inflammasome-forming protein in response to a multitude of signals (such as amyloid plaques, oxidative stress and DNA release), which occur concurrently during neuroinflammation. The confirmation that inflammasomes functionally contribute to the neurodegenerative process, the identification of other inflammasome-forming NLRs that are involved in neurodegenerative pathogenesis and the design of therapeutics targeting these molecules should be of great interest to researchers in neurology and immunology.

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All experiments were conducted in compliance with the ARRIVE guidelines.

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