Microglia and neuroinflammation: implications in neurodegenerative diseases

Kyoungho Suk

Department of Pharmacology, School of Medicine, Kyungpook National University, Daegu, 700-422 Korea

Abstract

Increasing evidence indicates that microglia-driven chronic inflammatory responses play a pathological role in the central nervous system. Activation of microglia is pivotal in the initiation and progression of neuroinflammation. Inhibition of the microglial activation may provide an effective therapeutic intervention that alleviates the progression of the neurodegenerative diseases. Anti-inflammatory agents may be a useful candidate for such a therapeutic approach. Continual investigation of the mechanisms underlying microglial activation and regulation of neuroinflammation by endogenous or exogenous factors would not only lead to the discovery of novel neuroprotective agents, but also help to understand complex pathophysiology of neurodegenerative diseases.

Introduction

In neuroinflammation, microglia and astrocytes play a critical role. Microglial cells are ubiquitously distributed in the central nervous system (CNS) and comprise up to 20% of the total glial cell population in brain [1, 2]. Although the ontogeny of microglial cells has long been debated, recent works using monoclonal antibodies specific for microglial cells indicated that these cells are closely related to monocytes and macrophages [3]. As the primary immune effector cells in the CNS, microglial cells migrate to the site of tissue injury or inflammation, where they respond to invading pathogens or other inflammatory signals [4, 5]. Like monocytes/macrophages, they also secrete inflammatory cytokines and toxic mediators which may amplify the neuroinflammatory responses [6, 7]. Astrocytes form an intimately connected network with neurons in the CNS, and they provide mechanical and metabolic support for neurons [8]. The critical role of these cells in ion buffering and clearance of neurotransmitters is also well established [9, 10]. Upon inflammatory stimulation, astrocytes proliferate and produce diverse intercellular mediators such as nitric oxide (NO) and tumor necrosis factor (TNF)-

α [11-13]. There is growing evidence that inflammatory mediators produced by activated astrocytes may be involved in the pathogenesis of various neurodegenerative diseases [10, 14]. Thus, the activation of astrocytes and ensuing production of toxic inflammatory mediators may need to be tightly regulated. Activation of inflammatory cells in CNS (microglia or astrocytes) may be intended to protect neurons at first. More frequently, however, activation of these neuroglial cells and inflammatory products derived from them have been implicated in neuronal destruction commonly observed in various neurodegenerative diseases [7]. Thus, our understanding of pathogenesis of neurodegenerative diseases may be enhanced by elucidation of the molecular mechanism underlying the regulation of neuroglial activation. Many endogenous or exogenous factors are thought to regulate neuroglial activation and resulting neuroinflammation [15-17].

Inflammation and tissue injury

Injury, trauma or infection induce a series of complex and interconnected reaction sequences, initiated at the site of tissue damage [18, 19]. This sequence of reaction serves to contain and destroy the infection or damaging agents, and to prevent continued tissue damage and initiate repair processes to restore normal function. This rapid response is known as acute inflammation [20]. The toxic reactions, which are employed to destroy infectious organisms or protect host, also paradoxically have the capacity to injure host tissues. If these toxic responses are not tightly regulated, tissue injury may predominate over tissue protection and repair, thereby leading to inflammatory diseases. The characteristics of the inflammatory response include localized changes within the damaged tissue such as the followings: 1) the release of preformed inflammatory mediators from intracellular stores; 2) the initiation of reaction cascade through the activation of soluble plasma components; 3) the new synthesis of inflammatory mediators such as eicosanoids and cytokines; and 4) resolution of the inflammatory response. The acute inflammatory response is beneficial to the organism in that it helps to deal with potentially dangerous microorganisms. However, inflammation does cause some degree of damage to surrounding tissues. Reactive oxygen species (ROS), reactive nitrogen species (RNS), prostanoids, leukotrienes, and hydrolytic enzymes produced by neutrophils, macrophages, and monocytes may all play a role in mediating inflammation. Persistence of infection or defective resolution of inflammatory reaction results in chronic inflammation where severe tissue damage may occur. Although inflammation is normally a self-limiting event and its benefit outweighs the minor tissue damage it causes, abnormal activation of the immune or

inflammatory system has the potential to provoke a devastating response [21]. In gout, for example, elevated concentration of uric acid in the blood leads to precipitation of sodium urate crystal within joints which triggers inflammation by a variety of mechanisms. Another striking consequence of abnormal inflammatory response is autoimmune diseases such as systemic lupus erythematosus, rheumatoid arthritis, autoimmune vasculitis, dermatomyositis, chronic autoimmune gastritis, and myasthenia gravis. Tissue-damaging chronic inflammatory response may also occur in CNS, where main inflammatory cells are microglia and astrocytes instead of monocytes/macrophages or neutrophils in periphery [22-24].

Neuroglia (microglia, astrocytes), neuroinflammation, and neurodegeneration

Microglia and astrocytes are essential for ensuring proper functioning of neurons. They are quick to intervene when neurons become injured or stressed. As they are sentinels of neuron well-being, pathological impairment of microglia or astrocytes could have devastating consequences for brain function. Nevertheless, there is still a debate over neuroprotective and neurotoxic functions of these neuroglial cells [22, 25]. It is assumed that neuroglial activation is largely determined by neuronal signals. Acute injury causes neurons to generate signals that inform neuroglia about the neuronal status. Depending on how severe a degree of neuronal injury, neuroglia will either nurse the injured neurons into regeneration or kill them if they are not viable. These types of neuroglial responses are considered to represent normal physiological and neuroprotective responses. In contrast, some processes that are chronic in nature persistently activate neuroglia eventually causing a failure in their physiological ability to maintain homeostasis. This could have detrimental consequences and may lead to bystander damage due to neuroglial dysfunction. In this scenario, neuroglia exert neurotoxic effects through the secretion of a variety of toxic inflammatory mediators. Thus, although activation of neuroglial cells may be intended to protect neurons, inflammatory products derived from activated neuroglia may also be implicated in neuronal injury, potentially leading to neurodegenerative diseases [7]. These deleterious effects of neuroglial activation may be exacerbated by the failure of auto-regulatory mechanisms of neuroglia. Recently, activated macrophages, whose functions are closely related to microglia, have been shown to undergo apoptosis [26-28]. It has been suggested that the apoptosis of activated macrophages is one mechanism whereby an organism may regulate immune and inflammatory responses involving macrophages [28]. It has been recently demonstrated that a similar regulatory mechanism exists for microglial cells [29, 30] and astrocytes [31]

as well. Microglial cells and astrocytes underwent apoptosis upon inflammatory activation in a manner similar to activation-induced cell death (AICD) of lymphocytes [30, 31]. AICD is an active process. T and B lymphocytes undergo AICD as an auto-regulatory mechanism for the body to remove unwanted activated cells after making appropriate use of them [32, 33]. Compared to lymphocytes, neuroglial cells in CNS are not well studied in this respect. Now, as results in this and other laboratories indicated that neuroglial cells might be under the control of a similar regulatory mechanism [29-31, 34-37], further investigation is warranted to better understand the molecular mechanism(s) of neuroglial AICD and its physiological significance. Nevertheless, it has been shown that, in contrast to AICD of T lymphocytes where Fas-FasL interaction plays a central role, neither Fas-FasL interaction nor TNFα is important in AICD of microglial cells [30]. Instead, NO produced by activated neuroglial cells themselves was the major cytotoxic mediator [30, 31]. However, the presence of NO-independent cytotoxic mechanism has been also suggested [38, 39].

Elimination of activated neuroglial cells by apoptosis could be an important mechanism whereby undesirable effects of long-term neuroglial activation can be minimized. Inflammatory mediators that are produced by activated neuroglia in CNS may have harmful effects on neurons or other neuroglial cells that they originally intended to protect [6, 7]. Thus, in various neurodegenerative diseases involving chronic neuroglial activation, neuroglial functions seem to play a more significant role in mediating diseases than in the protection of neurons. According to the model of activation-induced apoptosis of neuroglial cells, inflammatory signals that activate neuroglia may also initiate internal death program [38, 40]. One interesting question that can be raised then is how neuroglial cells could survive the inflammatory activation. It should be kept in mind that neuroglial cells *in vivo* are heterogeneous and interact with other neuroglial cells as well as neurons. There is also growing evidence that activated neuroglial cells proliferate *in vivo* as one way of replenishment [1]. Thus, not all neuroglial cells may respond to the inflammatory signals in the same fashion. Upon inflammatory activation, individual neuroglial cells in heterogeneous population may either undergo AICD or return to the resting state via other regulatory mechanisms depending on the specific microenvironment under which they react to the signals. Although many of activated neuroglial cells may be eliminated, some would survive to be deactivated. Whatever the mechanism of down-regulation is, this may be an excellent auto-regulatory system for the neuroglial activation. One can easily imagine pathological situations where this type of auto-regulatory mechanism goes wrong. Failure of the auto-regulation of 'over-activated' neuroglial cells may result in pathological

destruction of bystander cells (neurons and other neuroglial cells) exposed to toxic mediators produced by activated neuroglia. Recently, up-regulated Bcl-x_L expression has been detected in reactive microglia of patients with neurodegenerative diseases [41]. Authors proposed that high level of Bcl-x_L protein might render microglia more resistant to cytotoxic environment such as areas of neurodegeneration. Expression of anti-apoptotic Bcl-2 protein has been also associated with aged brain and neurodegenerative diseases [42]. An importance of physiological regulation of neuroglial activation by AICD is supported by these previous reports.

Recent studies focused on the possible role of neuroglia in causing neurodegeneration. Convincing evidence from *in vitro* studies pointed to the neurotoxic role of neuroglia during traumatic or ischemic brain injury [4] and AD pathogenesis [43]. Supernatants obtained from neuroglial cell cultures kill cultured neurons. Such supernatants contain various neurotoxic substances which include glutamate, NO, ROS, inflammatory cytokines, as well as yet unidentified neurotoxins [44, 45]. Production of these neurotoxins by neuroglia is enhanced by treatment with inflammatory stimuli such as lipopolysaccharide (LPS) and/or interferon (IFN)-γ. Paradoxically, other investigators have shown that neuroglia-conditioned media promote neuronal survival [46]. Thus, the balance of neurotoxic and neurotrophic effects of neuroglia appears to depend on the nature of the experimental paradigm used. In traumatic brain injury where neuronal regeneration may occur, neuroglial secretory products might help to promote regenerative efforts by injured, but surviving, neurons. However, a situation may be different in neurodegenerative diseases such as Alzheimer's disease (AD) or human immunodeficiency virus (HIV)-associated dementia, where functionally compromised neuroglia may produce neurotoxins, thereby resulting in neuronal damage. There is considerable evidence from post-mortem examinations of AD brains that auto-destructive mechanisms are at work, which could in part be responsible for the neurodegeneration [47, 48].

Neuroglia as a target of pharmacological intervention

Considering neuroglial activation as a common feature in many neuropathologies, and keeping in mind that over-activation of neuroglia can have neurotoxic outcomes, it is reasonable to assume that manipulation of neuroglial activation could serve future clinical approaches [49]. Although treatment of the primary events in neurodegenerative diseases would still be the preferred intervention, this may not always be possible. Brain or spinal cord injury is sudden event that is followed by secondary cascades of destruction.

Invading macrophages and intrinsic neuroglia in brain may carry a significant portion of these cascades of reaction. Now, there is growing evidence that toxic mediators produced by activated neuroglial cells might be involved in the pathogenesis of various neurodegenerative diseases such as Parkinson's disease (PD), AD, and HIV-associated dementia [6, 7, 47]. Thus, it is of great interest to find a means to modulate neuroglial activation and CNS inflammatory responses for the therapeutic interventions against these neurodegenerative diseases. Based on understanding of intracellular signaling pathways that are specific for activated neuroglia, a temporary inhibition of signaling molecules or protein-protein interaction associated with signaling pathways would probably allow for a rather selective effect on activated neuroglia, while respective functions in other cell types are unaffected. Elucidation of the intracellular key events that drive neuroglial activation could provide new routes for drug development [49]. Alternatively, potentially harmful products of neuroglia could be neutralized to limit undesired consequences for CNS cells and tissues. Whether it is a direct inhibition of neuroglial activation or indirect suppression of neuroglia-derived toxic inflammatory mediators, a better understanding of neuroglial biology and selective manipulation of neuroglial activation processes represent a promising goal for developing novel neuroprotective strategies.

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