NLRP3 Inflammasome Activation: A Therapeutic Target for Cerebral Ischemia–Reperfusion Injury

Lixia Wang1†, Wei Ren2†, Qingjuan Wu3, Tianzhu Liu2, Ying Wei1, Jiru Ding1, Chen Zhou2, Houping Xu4* and Sijin Yang2*

1 Hospital of Chengdu University of Traditional Chinese Medicine, Chengdu, China, 2 The Affiliated Traditional Chinese Medicine Hospital of Southwest Medical University, Luzhou, China, 3 Guang'anmen Hospital, China Academy of Chinese Medical Sciences, Beijing, China, 4 Preventive Treatment Center, The Affiliated Traditional Chinese Medicine Hospital of Southwest Medical University, Luzhou, China

Millions of patients are suffering from ischemic stroke, it is urgent to figure out the pathogenesis of cerebral ischemia–reperfusion (I/R) injury in order to find an effective cure. After I/R injury, pro-inflammatory cytokines especially interleukin-1β (IL-1β) upregulates in ischemic brain cells, such as microglia and neuron. To ameliorate the inflammation after cerebral I/R injury, nucleotide-binding oligomerization domain (NOD), leucine-rich repeat (LRR), and pyrin domain-containing protein 3 (NLRP3) inflammasome is well-investigated. NLRP3 inflammasomes are complicated protein complexes that are activated by endogenous and exogenous danger signals to participate in the inflammatory response. The assembly and activation of the NLRP3 inflammasome lead to the caspase-1-dependent release of pro-inflammatory cytokines, such as interleukin (IL)-1β and IL-18. Furthermore, pyroptosis is a pro-inflammatory cell death that occurs in a dependent manner on NLRP3 inflammasomes after cerebral I/R injury. In this review, we summarized the assembly and activation of NLRP3 inflammasome lead to the caspase-1-dependent release of pro-inflammatory cytokines, such as interleukin (IL)-1β and IL-18. Furthermore, pyroptosis is a pro-inflammatory cell death that occurs in a dependent manner on NLRP3 inflammasomes after cerebral I/R injury. In this review, we summarized the assembly and activation of NLRP3 inflammasome; moreover, we also concluded the pivotal role of NLRP3 inflammasome and inhibitors, targeting the NLRP3 inflammasome in cerebral I/R injury.

Keywords: NLRP3 inflammasome activation, ischemic stroke, pyroptosis, cerebral I/R injury, mitochondrion

INTRODUCTION

Ischemic stroke is the leading cause of disability in adults and has been a major health concern worldwide (Wang et al., 2017; Huang et al., 2022). Lack of understanding of the pathogenesis leads to limitations in the treatment of ischemic stroke. Currently, the immune response has shown both beneficial (Fernández-López et al., 2016) and detrimental effects (Jin et al., 2010) on the pathogenesis and prognosis of cerebral ischemia–reperfusion (I/R) injury. Oxidative stress, neuron death, and inflammation are involved in the pathogenesis of cerebral I/R injury (Pan et al., 2022a,b). Regarding neuroinflammation, the most definite mediator of inflammation after cerebral I/R injury is the cytokine interleukin-1 (IL-1) (Barrington et al., 2017). Nucleotide-binding oligomerization domain (NOD), leucine-rich repeat (LRR),
and pyrin domain-containing protein 3 (NLRP3) inflammasome act as the upstream of IL-1, which regulates the mature and secretion of proinflammatory cytokines. After transient middle cerebral artery occlusion (MCAO) in mice, NLRP3 is the major contributor among the inflammasomes (Franke et al., 2021). It is confirmed that NLRP3 inflammasome has been a therapeutic target for cerebral I/R injury. As a new detective signaling platform, the NLRP3 inflammasome could be activated by a range of microbial infections, such as coronavirus (Zheng et al., 2020), Staphylococcus aureus (Mariathasan et al., 2006), and candida albicans hyphae (Joly et al., 2009). As shown in Figure 1 and Table 1, the activation of NLRP3 inflammasome is involved with the onset and progression of various diseases, and the research for medicines that inhibit the activation of NLRP3 inflammasome could be therapeutically beneficial. Therefore, numerous studies are pursuing to figure out the physiological structure, assembly, and activation of NLRP3 inflammasome and detect the potential pathogenic mechanisms.

THE ASSEMBLY OF NLRP3 INFLAMMASOME

Inflammasome was first described by Fabio Martinon in 2002, which was the identification of a caspase-activating complex that consisted of a sensor (PRRs), an adaptor (ASC), and an effector (caspase1) (Martinon et al., 2002; Swanson et al., 2019). The PRRs are expressed in innate immune system cells (such as macrophages and neutrophils cells) (Abderrazak et al., 2015), and PRRs sense pathogen-associated molecular patterns (PAMPs) or damage/danger-associated molecular patterns (DAMPs) (Matzinger, 2002; Karasawa and Takahashi, 2017) for the innate immune system to defend against these "danger signals." PRRs can be divided into many subgroups, including members of Toll-like receptors (TLRs), C-type lectin receptors (CLRs), NOD-like receptors (NLRs), and absent-in-melanoma 2 receptors (ALRs) (Lamkanfi and Dixit, 2014; Kelley et al., 2019). NLRP3 inflammasome is the most well-characterized inflammasome and closely related to the cleavage of caspase-1 (Martinon et al., 2002). NLRP3 is composed of central NOD, a C-terminal LRR domain, and an N-terminal pyrin domain (PYD) (Menu and Vince, 2011; Alishahi et al., 2019). ASC (apoptosis speck protein) has a C-terminal pyrin domain (PYD) and a C-terminal caspase recruitment domain (CARD) (Alishahi et al., 2019). The CARD in ASC is homotypic with the CARD in pro-caspase-1. After the LRR domain is sensitized by stimuli, NLRP3 self-oligomerizes through the interaction of homotype NODs (Alishahi et al., 2019), then, the N-terminal PYD domain in oligomerized NLRP3 facilitates homotypic PYD–PYD interactions between NLRP and adapter protein ASC (Liepinsh et al., 2003), and assembled ASC recruits pro-caspase-1 via C-terminal CARD–CARD interactions (Srinivasula et al., 2002). The assembly and activation of NLRP3 inflammasome promote the cleavage of pro-caspase-1 to form active caspase-1, which leads to the maturation and cleaves of proinflammatory cytokines, such as IL-1β and IL-18.

THE ACTIVATION OF NLRP3 INFLAMMASOME UNDER I/R INJURY

Notably, the activation of NLRP3 inflammasome has been detected after I/R injury. It is confirmed that NLRP3 was upregulated significantly in 4 h after hypoxic-ischemic in rats, and the elevated levels of IL-1β were detected in 8 h after hypoxic-ischemic injury (Li et al., 2021b). A study of patients with acute ischemic stroke admitted <24 h showed that the level of serum concentration of NLRP3 was related to the increased risk of malignant brain edema (MBE) (Wang et al., 2021c). Therefore, elucidating the mechanism of NLRP3 activation and intervening early after the onset of ischemic stroke are of great importance for the treatment and prognosis of ischemic stroke. It is widely accepted that NLRP3 inflammasome activation is related to two signals: in signal I (priming), cytokines or PAMPs could lead to pro-IL-1β and NLRP3 upregulation in a nuclear factor-κ-gene binding (NF-κB)-dependent manner in response to the activation of proinflammatory cytokine receptors or transcription-modulating PRRs; in signal II (activation), various upstream DAMP and PAMP signaling events lead to the oligomerization of NLRP3 and the assembly of ASC and procaspase-1 to form NLRP3 inflammasome (Shao et al., 2015; Liu, Q. Y et al., 2018; Sho and Xu, 2019; Swanson et al., 2019). Recent studies demonstrate that there are mainly three models which activate the signal II in the activation of NLRP3 inflammasome, namely, ion flux, mitochondrial destabilization, and lysosomal damage (Gong et al., 2018a; Kelley et al., 2019). All these models are shown in Figure 2. In this study, we described the activation of NLRP3 inflammasome after cerebral I/R injury.

Ionic Flux

Intracellular ionic flux which includes K⁺ efflux, Ca²⁺ mobilization, and Cl⁻ efflux acts as an upstream regulation role in the activation of NLRP3 inflammasome (Gong et al., 2018a). K⁺ efflux is the most investigated target, the main reason for the decrease in intracellular K⁺ concentration is related to K⁺ channel opening (Di et al., 2018), membrane remodeling (Gianfrancesco et al., 2019), and membrane permeabilization change (Franchi et al., 2014); at the same time, many stimuli can trigger K⁺ effluxes such as adenosine triphosphate (ATP), nigericin, and monosodium urate (MSU) crystals (Nomura et al., 2015). The activation of NLRP3 could be blocked by inhibiting K⁺ efflux; therefore, the low intracellular K⁺ concentration is a key trigger for NLRP3 inflammasome activation (Pétrilli et al., 2007). It has demonstrated that K⁺-ATP channel pore-forming subunit Kir6.1 is a bona fide negative regulator of the NLRP3 inflammasome, and the suppression of Kir6.1 increases the accumulation of damaged mitochondria and production of reactive oxygen species (ROS) (Du et al., 2019; Hu et al., 2019). K⁺ efflux is a well-accepted upstream regulating signal for NLRP3 inflammasome, but some studies have reported that other signals could also affect NLRP3 inflammasome activation in a K⁺ efflux-independent manner. Imiquimod affects ROS production by regulating the quinone oxidoreductases, namely, NQO2 and mitochondrial Complex I to participate in the activation of the NLRP3 inflammasome, and the activation of the
NLRP3 inflammasome by imiquimod is K⁺ efflux-independent (Groß et al., 2016).

**Mitochondrial Destabilization**

After being stimulated, the products derived from mitochondria and other mitochondrial signaling molecules contribute to the NLRP3 inflammasome activation, such as mitochondrial ROS (mROS), mitochondrial DNA (mtDNA), and cardiolipin (Gong et al., 2018b; Zhong et al., 2018; Dagvadorj et al., 2021). All these details are shown in Figure 3. It is well-agreed that inducing ROS is the common characteristic of most NLRP3 activators (Hornung and Latz, 2010), and the main source of cellular ROS is the mitochondria (Dan Dunn et al., 2015). mROS is critical for NLRP3 inflammasome activation (Tschopp and Schroder, 2010; Gurung et al., 2015; Minutoli et al., 2016; Yu and Lee, 2016). The overexpression of mROS would activate the NLRP3 inflammasome through mainly two-signal models, namely, NF-kB pathway and mitochondria/thioredoxin-interacting protein (TXNIP), which activates the NLRP3 inflammasome (Sho and Xu, 2019). Accumulating studies have shown that the inhibitors of mtDNA synthesis and mROS can alleviate diseases by suppressing NLRP3 inflammasome activation (Zhong et al., 2016; Guo et al., 2017; Lee et al., 2019). Cardiolipin could activate the NLRP3 inflammasome in a ROS-independent way during mitochondrial destabilization. In addition to mROS, mtDNA and cardiolipin also serve as the ultimate NLRP3 ligand for the activation of NLRP3 inflammasome (Shimada et al., 2012; Iyer et al., 2013).

What is more, mitochondrial antiviral signaling protein (MAVS) is a critical regulator in the recruitment of NLRP3 to mitochondria, promoting the production of IL-1β and the pathophysiological activity of the NLRP3 inflammasome (Subramanian et al., 2013). Park et al. further discovered that MAVS not only accelerated the recruitment of NLRP3 to the mitochondria and brought it close to mtROS to improve its activation but also was involved in the assembly of NLRP3 inflammasome (Park et al., 2013). During mitochondrial destabilization, the recruitment of dynamin-related protein 1 (Drp1) on mitochondria leads to excessive mitochondrial fission, ultimately activating the NLRP3 inflammasome. It was conducted that AMP-activated protein kinase (AMPK) activation inhibited mitochondrial fission by upregulating Drp1 phosphorylation at serine637 (Ser637) in an AMPK-dependent manner to protect mitochondrial integrity and then, suppressed ER stress to inhibit the activation of NLRP3 inflammasome (Li et al., 2015, 2016; Guo et al., 2018b).
What is more, it was also shown that receptor-interacting serine/threonine kinase (RIP) I/RIp3 (RIP1-RIp3) complex could phosphorylate Drp1 at serine616 (Ser616) to activate and promote Drp1 translocation to mitochondria and ultimately contributed to the activation of NLRP3 inflammasome via (RIP1–RIp3)—Drp1 pathway (Wang et al., 2014). It has also proven that RIp3 regulates potassium efflux-dependent NLRP3 inflammasome activation via mixed-lineage kinase domain-like protein (MLKL)-induced pores and can be inhibited by supplementing extracellular potassium (Conos et al., 2017).

**Lysosomal Damage**

Pyrin domain-containing protein 3 activators induce lysosomal damage, which is indirectly sensed by the NLRP3 inflammasome (Hornung and Latz, 2010). Several reports attribute the activation of the NLRP3 inflammasome to the leakage of lysosomal contents into the cytosol following phagocytosis of particulate stimuli that could damage their integrity (Hornung et al., 2008; Shimada et al., 2012). Lysosomal rupture can release cathepsins and ROS, which also significantly impacted mitochondria membrane integrity and lead to membrane permeabilization (Shimada et al., 2012). Leu-Leu-O-methyl ester (LLME) is a lysosomedamaging compound when LLME is transported to the lysosome, resulting in lysosomal membrane permeability (LMP) (Hornung and Latz, 2010). It is proved that low-dose LLME causes mild LMP and strongly activates the inflammasome (Schilling, 2016). What is more, lysosomes are emerging as intracellular Ca\(^{2+}\) stores (Zhong et al., 2017), and the activity of lysosomal ion channels and transporters maintains concentration gradients of K\(^+\), Ca\(^{2+}\), Na\(^{+}\), and Cl\(^−\) across the lysosomal membrane. Once the lysosomal damage occurs, it would contribute to the Ca\(^{2+}\) overloading and the disorder of lysosomal ion channel activity, which stimulates the activation of the NLRP3 inflammasome (Kendall and Hollian, 2021). It has been proven that the restoration of lysosomal dysfunction could augment neuroprotection against ischemic stroke in neurons (Zhang et al., 2022).

**NLRP3 INFLAMMASOME IN CEREBRAL I/R INJURY**

The main pathogenesis of ischemic stroke includes inflammation, oxidative stress, and programmed cell death (PCD) (Jin et al., 2010; Ren et al., 2021). With the involvement of the innate immune system, the activation and expression site of NLRP3 inflammasome act as a critical role in the development of ischemic stroke. In this study, we explained in detail the involvement of NLRP3 inflammasome in the pathogenesis of cerebral I/R injury, and the details are shown in Figure 4.

**Inflammation**

After cerebral I/R injury, various cellular responses are aroused, such as the activation of inflammatory cytokines and the accumulation of ROS and other oxygen free radicals (Fann et al., 2013; Guo et al., 2016), which exacerbate I/R injury by promoting brain oxidative stress, inflammation, and cerebral infarction volume (Jiang et al., 2019; Franke et al., 2021; Joaquim et al., 2021). The activation of the NLRP3 inflammasome plays an important role in the development of inflammation after cerebral I/R injury. NLRP3 inflammasome is first expressed in microglia and then in microvascular endothelial cells and neurons, but finally mainly in neurons at 24 h (Gong et al., 2018b).

**Microglia**

After ischemic I/R injury, the circulating macrophages were recruited to the ischemic tissue to be involved in cerebral I/R injury (Cai et al., 2018). As brain resident macrophages, microglia could be activated first after I/R injury, and macrophages/microglia change their M1 or M2 phenotype depending on the microenvironment of the central nervous system (CNS) (Hanisch and Kettenmann, 2007; Dong et al., 2021). M1 phenotype recognizes harmful stimuli and consequently generates inflammatory cytokines such as IL-1β, IL-6, and tumor necrosis factor-α (TNF-α) (Cherry et al., 2014). M2 phenotype shifts into an anti-inflammatory state where extracellular matrix deposition, debris clearance, and angiogenesis are promoted (Varin and Gordon, 2009). Regulating the polarization of macrophage/microglia could alleviate brain damage after ischemic stroke (Ye et al., 2019). Thirty minutes after modeling MCAO in mice, activated microglia were detected in the ischemic lesions (Rupalla et al., 1998). After ischemic stroke, DAMPs and PAMPs such as ischemia, hypoxia, and inflammatory factors could activate microglia (Gülke et al., 2018) and facilitate the proinflammatory role via hypoxia-inducible factor 1α (HIF-1α) in microglia (Yang et al., 2014). Additionally, anti-inflammatory factors, such as IL-10 and IL-4, could induce the M2 phenotype of microglia to protect against ischemic stroke (Xiong et al., 2015). As the monitoring of the microenvironment, immune system response after ischemic stroke can also affect the polarization of microglia, such as interferon regulatory factor (IRF) 4/5 signaling (Al Mamun et al., 2018). IRF5 is required for the M1 phenotype in microglia, and IRF4 was identified as a key transcription factor for M2 polarization in microglia (Al Mamun et al., 2018). In the mouse model of MCAO, the inhibition of NLRP3 inflammasome activation in activated microglia significantly improved functional neurological deficits (Sapkota and Choi, 2021). Additionally, the polarization of M2 microglia could protect against cerebral ischemic injury via the NF-E2–related factor 2 (Nrf2)/heme oxygenase-1 (HO-1)/NLRP3 pathway (Wang et al., 2021c). Therefore, it is therapeutic in ischemic stroke to inhibit inflammatory response via inhibiting phenotype switch of microglia and NLRP3 inflammasome activation.

Indoleamine 2,3-dioxygenase 1 (IDO-1) is an immunosuppressive metabolic enzyme and elicits neuroprotective effects on ischemic injury (Park et al., 2020). IDO-1 is mainly expressed in the macrophage/microglia of the perivascular but not the parenchymal microglia of the brain (Ji R. et al., 2021). As the downstream enzyme of the NLRP3 inflammasome, the inhibition of IDO-1 with curcumin decreased NLRP3 expression (Zhang W.-Y et al., 2019); in contrast, the inhibition of IDO-1 reduced NLRP3 expression, and inhibiting NLRP3 also increased IDO-1 expression,
| Disease | NLRP3 inflammasome inhibitor | Model | Animals and cells | Signaling | Effects | References |
|---------|-------------------------------|-------|-------------------|-----------|---------|------------|
| Rheumatoid arthritis | MCC950 | MCAO/R and OGD/R | Mice and SH-SY-SY cells | The mitochondrial translocation of Drp1 | Mitochondrial Function, ER Stress | Guo et al., 2018a |
| Endothelial inflammation and atherosclerosis | NLRP3 shRNA | Diabetes mellitus model | Diabetes patients and diabetic Apolipoprotein E−/− mice and human umbilical vein endothelial cells (HUVECs) | NLRP3 inflammasome signaling | Endothelial inflammation | Wan et al., 2019 |
| Inflammatory bowel diseases | Glyburide | IL-10 mice | C57BL/6 mice and Patients with diagnosis of CD | NLRP3 inflammasome signaling | Inflammation | Liu et al., 2017 |
| Renal fibrosis | MCC950 | Renal fibrosis model | C57BL/6 mice | NLRP3 inflammasome signaling | Oxidative stress, inflammation, renal dysfunction, histological injury, and interstitial fibrosis | Li et al., 2019 |
| Renal ischemia/reperfusion injury | Hydroxychloroquine | Renal I/R injury model | C57BL/6 male mice and HK-2 cell | NF-κB signaling | Renal inflammation | Tang et al., 2018 |
| Renal inflammation | B-cell lymphoma 6 (BCL6) | Spontaneously hypertensive rats (SHR) and Inflammation models | SHR, Wistar-Kyoto rats (WKY), and HK-2 cell | NLRP3 transcription | Inflammation in the renal cortex | Chen et al., 2017 |
| Chronic renal dysfunction | Phloretin | Hyperuricemia model | C57BL/6 male mice and HK-2 cell | NLRP3 pathway | Inflammation | Cui et al., 2020 |
| Traumatic brain injury (TBI) | NIMA-related kinase 7 (NEK7)-shRNA | Controlled Cortical Impact (CCI) Model | C57BL/6 male mice and Primary Cortical Neurons | NEK7-NLRP3 signaling | Neuroinflammation and pyroptosis | Chen et al., 2019b |
| Acute pancreatitis (AP) | INF-39 | Severe acute pancreatitis (SAP) Model | NLRP3−/− C57BL/6 mice | NLRP3 inflammasome signaling | Inflammatory cascade and neutrophil infiltration | Fu et al., 2018 |
| Systemic lupus erythematosus (SLE) | Methylprednisolone | SLE patients | – | NEK7-NLRP3 inflammasome signaling pathway | Inflammation | Ma et al., 2018 |
| Ischemic stroke | Ketogenic Diet | MCAO Model and OGD/R | C57BL/6 mice and SH-SY-SY cells | Mitochondrial translocation of Drp1 | ER stress, apoptosis and inflammation, Immunocompetent cell death | Guo et al., 2018b |
| Breast cancer | MiRNA-233-3p | Breast cancer cell lines | C57BL/6 mice and SH-SY-SY cells, HMEC, MDA-MB231, MCF-7, and SKBR3 cell lines | Mitochondrial translocation of Drp1 | Mitophagy | Xue et al., 2019 |
| Myocardial infarction | Colchicine | Myocardial Infarction Mouse Model | C57BL/6J mice | NLRP3 inflammasome signaling | Cardiomyocyte pyroptosis | Qiu et al., 2017 |
| Diabetic cardiomyopathy | Empagliflozin | Diabetic db/db mice | Mice | aGlu-cAMP-PKG pathway | Pyroptotic cell death | Chen et al., 2019a |
| Myocardial Ischemia/Reperfusion (I/R) Injury | BAY11-7082/MCC950 | Myocardial ischemia/reperfusion (MiR) injury and H/R Injury model | Sprague-Dawley rats and H9C2 Cell lines | NLRP3 inflammasome signaling | Mitochondrial dysfunction | Qiu et al., 2017 |
| Diabetic nephropathy (DN) | Optineurin | High glucose culture | DN patient and Murine primary renal tubular epithelial cells (RTECs) | Mitophagy | Mitochondrial dysfunction | Chen et al., 2019a |
| Diabetic retinopathy | Fenofibrate | Diabetes model | C57BL/6 mice | Nrf2 signaling | Retinal leukostasis and vascular leakage | Liu Q. P. et al., 2018 |

(Continued)
indicating that the relationship between NLRP3 and IDO-1 is bidirectional, and the direction depends on the activation status of macrophages/microglia (Ji R. et al., 2021). It is indicated that IDO-1 may decrease the expression of NLRP3 in macrophage/microglia of the perivascular space to inhibit inflammation and protect the integrity of the blood-brain barrier (BBB) against cerebral I/R injury.

**Microvascular Endothelial Cell**

As the important structure of BBB, microvascular endothelial cells play a vital role in cerebral I/R injury. With regard to I/R inflammation, the adhesion of neutrophils to vascular endothelial cells is fundamental to the development of I/R inflammation (Dong et al., 2019). It has been studied that endothelial cells can secrete inflammatory factors such as vascular cell adhesion molecule-1 (VCAM-1) to recruit neutrophils and/or lymphocytes, leading to the infiltration of inflammatory cells in the ischemic region (Gao et al., 2021). Lysophosphatidylcholine (LPC) is the main active component of oxidized low-density lipoproteins (ox-LDLs), and it has been proven that LPC could facilitate inflammatory response in brain microvascular endothelial cells (BMECs) via G protein-coupled receptor 4
(GPR4)-mediated activation of NLRP3 inflammasomes (Liu et al., 2021a). The expression inhibition of NLRP3 inflammasome in endothelial cells could improve the integrity of BBB and behavioral outcomes (Cao et al., 2016). The functional integrity of BMECs has shown a significant protective effect against brain I/R injury (Wang et al., 2021a).

**Neuron**

After the onset of cerebral I/R injury, cellular damage is mainly triggered by excitotoxicity, mitochondrial disturbances, dysfunction of the endoplasmic reticulum, ROS production, calcium toxicity, nitric oxide toxicity, zinc toxicity, and PCD (Lo et al., 2005; Hossmann, 2006). After the ischemic injury, neuron death occurs within minutes (Amantea et al., 2009), and the neuronal cell death after cerebral I/R injury included apoptosis, autophagy, pyroptosis, ferroptosis, parthanatos, phagoptosis, and necroptosis (Tuo et al., 2022). With the time prolongation, the neuron cell and nuclear membrane in the center of the ischemic region ruptured, and the cells were lysed (Gou et al., 2021). As mentioned earlier, the cellular damage could contribute to the activation of the NLRP3 inflammasome, ultimately exacerbating the inflammatory response. Based on the fact that NLRP3 inflammasome is ultimately mainly expressed in neurons after ischemic stroke, drugs that inhibit NLRP3 inflammasome to alleviate cerebral I/R injury and reduce inflammatory response must be studied.

**Pyroptosis**

Pyroptosis is one kind of pro-inflammatory PCD, which has been shown to be involved in cerebral I/R injury (Voet et al., 2019). Different from other PCD, pyroptosis is characterized by the rupture of the plasma membrane to form pores, increases in cell permeability, and the release of inflammatory cytokines (Fink and Cookson, 2005). The occurrence of pyroptosis relies on caspase-1/4/5/11, and caspase-1 and caspase-11 are the main activated inflammatory caspases during cerebral ischemia (Gou et al., 2021). The oxidative stress and inflammatory response activate the NLRP3 inflammasome and consequently induce the cleave of pro-caspase-1 to form active caspase-1, which ultimately promotes pyroptosis via the canonical inflammasome pathway (Broz, 2015). Caspase-1 cleaves pro-IL-1β and pro-IL-18 into a mature form of IL-1β and IL-18. Caspase-1 could also cleave Gasdermin D (GSDMD), expose Asp280 amino acid sites, and promote the recruitment of the N-domain of GSDMD to the cell membrane, leading to the cell membrane pore formation and pyroptosis (Zhang et al., 2019; Ji et al., 2021), thereby releasing IL-1β and IL-18, and leading to cascade inflammatory response and inflammatory cell recruitment (Vande Walle and Lamkanfi, 2016).

Pyroptosis has been shown to be expressed in various CNS cells, such as microglia, neuron, oligodendrocytes, and astrocytes (McKenzie et al., 2018; Zhou et al., 2019; Hu et al., 2021; Zhao et al., 2021). Additionally, it has been proven that the pyroptosis of endothelial cells participates in the pathogenesis of cerebral I/R injury (Wang et al., 2021d). Regarding the critical downstream effector of pyroptosis (Shi et al., 2015), the expression of GSDMD was found to increase after I/R and peak at 3–5 days in mice (Lu et al., 2021) and so aggravates I/R-induced cerebral infarction and brain injury. The ablation of GSDMD exerts a neuroprotective effect by inhibiting microglia pyroptosis in mice after cerebral I/R injury (Wang et al., 2020a). Moreover, the inhibition of NLRP3 inflammasome-dependent pyroptosis could reduce neuronal injury and cerebral infarct after I/R injury (Kang et al., 2021; Shi et al., 2022). Regarding the upstream of pyroptosis, the suppression of NLRP3 inflammasome is a critical target for the treatment of cerebral I/R injury.

**THERAPEUTIC APPROACHES TARGETING NLRP3 INFLAMMASOME FOR CEREBRAL I/R INJURY**

As a neurological disease with a high disability rate, ischemic stroke still lacks efficient therapeutic treatment in the clinic. As previously mentioned, NLRP3 inflammasome plays a major role in cerebral I/R injury, and many drugs that inhibit NLRP3 inflammasome activation have been studied for the treatment of ischemic stroke. In this study, we summarized the therapeutic approaches involving NLRP3 inflammasome and classified these drugs as follows: clinical treatment, herbal/natural component, and novel inhibitor.

**Novel Inhibitor**

To find out the potential pathogenesis and therapeutic drugs for cerebral I/R injury application, many novel inhibitors have been found to mitigate cerebral I/R injury by suppressing the activation of the NLRP3 inflammasome. The most well-studied inhibitor is MCC950, which is proven specificity in inhibiting NLRP3 inflammasome targeting for the assembly of NLRP3 inflammasome (Wu et al., 2020). MCC950 can modify the active conformation of NLRP3, and it prevents NLRP3 oligomerization in response to external stimulation (Tapia-Abellán et al., 2019). Experimental studies have found that the treatment of MCC950 could reduce the infarction and edema and improved neurological deficits and BBB integrity via inhibiting inflammatory cytokines, pyroptosis, and brain oxidative stress in the ischemic region after MCAO (Ismael et al., 2018; Bellut et al., 2021; Joaquim et al., 2021). CY-09 could inhibit ATPase activity and block NLRP3 oligomerization to inhibit the activation of NLRP3 inflammasome (Jiang et al., 2017). It has shown the therapeutic effect of CY-09 in cerebral I/R injury via inhibiting NLRP3 inflammasome-induced inflammation and pyroptosis (Sun et al., 2020; Franke et al., 2021). Oridonin could prevent NLRP3 inflammasome complex assembly against NLRP3 inflammasome activation (He et al., 2018). The study found oridonin prevented oxidative stress-induced endothelial injury via promoting the Nrf2 pathway and thereby repaired BBB integrity, alleviated neuroinflammation, and infarct volume after ischemic stroke (Li et al., 2021a).

**Clinical Treatment**

Although the pathogenesis of ischemic stroke remains unclear, many conventional medicines revealed the therapeutic effect on
ischemic stroke function by inhibiting the activation of NLRP3 inflammasome. As an oxygen radical scavenger, Edaravone reduced neurotoxicity, oxidative stress, and inflammatory response after cerebral I/R injury (Xu et al., 2021a). Indobufen and Aspirin and their Combinations with Clopidogrel or Ticagrelor (IACT) could alleviate pyroptosis via NF-κB/NLRP3 pathway after cerebral I/R injury, which indicated that the combination of antiplatelet drugs is a promising strategy for the curation of cerebral I/R injury (Li et al., 2021c). In addition, idebenone, as a mitochondrial protectant, was found to decrease ROS and cytosolic oxidized mtDNA, suppress uncontrolled NLRP3 activation, and consequently alleviate cerebral inflammatory response after cerebral I/R injury (Peng et al., 2020). Hypothermia is used for clinical neuroprotective purposes after ischemic stroke. It was suggested that hypothermia downregulated the expression of NLRP3 and attenuated I/R-induced pyroptosis partially via the phosphatidylinositol-3-kinase (PI3K)/Akt/Glycogen synthase kinase-3β (GSK-3β) pathway (Diao et al., 2020). Electroacupuncture (EA) is commonly used to relieve chronic pain and stroke rehabilitation, and some studies have uncovered that EA pretreatment protects against transient cerebral I/R injury (Wang et al., 2012). EA stimulus has a neuroprotective effect through α7nAChR by modulating the inhibition of NLRP3 inflammasome-associated...
inflammatory response and cellular apoptosis (Jiang et al., 2019).

**Herbal/Natural Component**

Herbs and their extracts have been an important source of approach for the treatment of ischemic stroke, many herbal medicines have shown neuroprotection after cerebral I/R injury via inhibiting NLRP3 inflammasome activation, and the detailed information is summarized in Table 2. **Tongxinluo** (TXL) is a common Chinese patent drug used clinically in the treatment of stroke, and TXL could protect ischemic brain tissues against pyroptosis in astrocytic by inactivating caspase-11/GSDMD (Wang et al., 2021b). **Xingnaojing injection** (XNJ) is isolated from famous traditional Chinese medicine prescriptions named *An-Gong-Niu-Huang Wan*, which is well-accepted in the clinic due to its significant therapeutic effect (Lai et al., 2017). XNJ ameliorated neurological deficits and BBB disruption following I/R injury in a manner of NLRP3 inflammasome suppression (Qu et al., 2019). The effective compounds extracted from *Buyang Huanwu Decoction* (BYHWD) can alleviate neuronal damage and inhibit NLRP3 inflammasome-mediated neuronal pyroptosis (She et al., 2019). Hispidulin is a component widely existing in traditional Chinese medicine and could inhibit I/R-induced pyroptosis in the ischemic cortex by modulating AMPK/GSK3β signaling (An et al., 2019). Resveratrol, anthocyanin, melodinhenine B, and 6-Gingerol could alleviate cerebral I/R injury by inhibiting NLRP3 inflammasome activation (He et al., 2017; Cui et al., 2018;
Li et al., 2020a; Luo et al., 2021). Icariin, a flavonol glycoside extracted from *Epimedium brevicornum* Maxim (Berberidaceae), has shown an anti-inflammatory effect against Oxygen and Glucose Deprivation/Reoxygenation (OGD/R) through the inositol-requiring enzyme-1 (IRE1)/X-box binding protein 1 (XBP1) pathway in microglia (Mo et al., 2021). Moreover, the pretreatment of sulforaphane (SFN) in the ischemic stroke model could protect neurovascular and alleviate neurological deficits and BBB disruption via the Nr1-2/NF-κB defense pathway (Alfieri et al., 2013; Warpsinski et al., 2020). Herbal medicines especially traditional Chinese medicines have been an essential approach for the recovery of ischemic stroke, identifying natural herbs which ameliorate I/R injury via NLRP3 inflammasome that has prospective value.

**Others**

Intriguingly, non-coding RNA (ncRNA), including long non-coding RNA and microRNA, is involved in the pyroptosis, inflammatory response, oxidative stress, apoptosis, and BBB permeability in a manner of NLRP3 inflammasome after cerebral I/R injury (Ghafouri-Fard et al., 2020), and all the information is shown in Table 3. It is of great importance to develop drugs that inhibit NLRP3 by targeting ncRNAs.

Bone marrow mesenchymal stem cell-derived exosomes (BMSC-Exos) can attenuate the activation of NLRP3 inflammasome and NLRP3 inflammasome-mediated pyroptosis via promoting AMPK-dependent autophagic flux in OGD/R injury (Zeng et al., 2020), by a mechanism that switches microglial phenotypes from M1 to M2, so as to ameliorate cerebral I/R injury (Liu et al., 2021b). Moreover, intermittent fasting can attenuate the inflammation and neuronal damage following cerebral I/R injury through the suppression of NLRP3 inflammasome activation (Fann et al., 2014). Additionally, an enriched environment (EE) could rescue neurological deficits after I/R injury via inhibiting the activities of NLRP3 inflammasome and attenuating neuronal pyroptosis (Liu et al., 2021c).

The continuous studies for NLRP3 inflammasome will promote the new drug research and development for cerebral I/R injury clinical treatment, but further research is needed for the clinical application of new compounds.
### TABLE 2 | Herbal drugs that target NLRP3 inflammasome after cerebral ischemia–reperfusion (I/R) injury.

| Drugs            | Type               | Source                                      | Target                  | Signaling                      | Animals and cells | Models                                   | Current clinical trial                          | References                          |
|------------------|--------------------|---------------------------------------------|-------------------------|--------------------------------|--------------------|------------------------------------------|-----------------------------------------------|-----------------------------------|
| Icariin (ICA)    | Glycoside          | Epimedium brevicornum Maxim (Berberidaceae) | NLRP3 and caspase-1     | IRE1/XBP1’s pathway            | Microglia          | Oxygen-glucose deprivation (OGD/R)       | –                              | Mo et al., 2021                     |
| Tongxintuo       | TCM prescription   | Jiang Xiang, Ru Xiang, Bing Plan, Ren Shen, Chi Sao, Suan Zao Ren, Tari Xiang, Can Tu, Shui Zhi, Tu Bie Chong, Quan Xie, Wu Gong | Inhibited Astrocytic Pyroptosis | Caspase-11/GSDMD         | Sprague–Dawley rats | Middle cerebral artery occlusion/reperfusion (MCAO/R) | Cardiogenic brovascular diseases of blood stasis syndrome | Wang et al., 2021b |
| 6-Gingerol       | Phenolic compound  | Ginger                                      | NLRP3 and caspase-1     | TRPV1/FAF1 complex             | Sprague–Dawley rats | MCAO                                     | –                              | Luo et al., 2021                     |
| Bakuchiol (BAK)  | Prenylated phenolic mono-terpene | the seeds of psoralea corylifolia          | NLRP3 and caspase-1     | Nrf2 signaling                | Mice and BV-2 cells | MCAO and OGD/R                            | –                              | Xu et al., 2021b                    |
| Gastrodin (GAS)  | Versatile compound | Traditional Chinese herb Tianma              | NLRP3 and caspase-1     | LncRNA NEAT1/miR-22-3p/NLRP3   | Sprague–Dawley rats | MCAO                                     | –                              | Zhang et al., 2021                  |
| Oridinon (Ori)   | Diterpenoid isolated | Rabdosia rubescens                         | NLRP3 and caspase-1     | NF-κB signaling               | CS7BL/6 mice and BV-2 cells | MCAO and OGD/R                            | –                              | Jia et al., 2021                    |
| D- Carvone       | D- carvone dietary monoterpenes | Seed variety caraway essential oil          | NLRP3                   | TRL4/NLRP3 signaling pathway  | Sprague–Dawley rats | MCAO                                     | –                              | Dai et al., 2020                    |
| Cepharanthine (CEP) | Bibenzyliso quinoline (BBI) aka-loids | Stephania cepharantha                      | NLRP3                   | 12/15-LOX signaling           | Mice and BV-2 cells | MCAO and OGD/R                            | –                              | Zhao et al., 2020                   |
| Tetrandrine      | Alkaloid           | Radix Stephania tetrandra                  | NLRP3                   | Sirt-1                         | Mice               | MCAO                                     | –                              | Wang et al., 2020b                  |
| Astilbin         | Dihydroflavonol derivative | Rhinoma Smilacis glabrae (RSG)             | NLRP3                   | MAPK pathway and PI3K/AKT pathway | PC12 cell          | OGD/R                                    | –                              | Li et al., 2020b                    |
| Melodinhenine B  | Eburnean-vindoline-type bisindole alkaloid | M. henryi                               | NLRP3                   | BBB integrity                  | Sprague–Dawley rats | MCAO                                     | –                              | Li et al., 2020a                    |
| XingNaoJing      | TCM prescription named An-Gong-Nu-Huang pill | Moschus, Radix Curcumae, borneol and Fructus gardeniae | NLRP3                   | BBB integrity                  | Sprague–Dawley rats | MCAO                                     | the treatment of stroke             | Qu et al., 2019                     |
| Glycosides       | Astragaloside IV, paeoniflorin, and amygdalin | Buyang Huanwu Decoction                  | NLRP3                   | Classical pyroptosis pathway  | Sprague–Dawley rats | MCAO                                     | Prevent and treat cerebral ischemia     | She et al., 2019                   |
| Hispidulin       | Flavonoid          | Chinese herbal medicines                   | NLRP3                   | AMPK/GSK3β signaling pathway  | Sprague–Dawley rats and primary cerebral astrocytes | MCAO and OGD/R | –                              | An et al., 2019                     |

(Continued)
**TABLE 2 | Continued**

| Drugs | Type | Source | Target | Signaling | Animals and cells | Models | Current clinical trial | References |
|-------|------|--------|--------|-----------|-------------------|--------|------------------------|------------|
| Anthocyanin | Phenolics or polyphenolics | Myrica rubra | NLRP3 | TLR4/NF-κB and NLRP3 Pathways | ICR mice | MCAO | – | Cui et al., 2018 |
| Resveratrol (RSV) | Poly-phenolic compound | Veratrum grandiflorum | NLRP3 | Sirt1-dependent autophagy induction | Sprague–Dawley rats | MCAO | – | He et al., 2017 |
| Sulforaphane (SFN) | Isothiocyanate | Cruciferous vegetables | NLRP3 | The activation of NLRP3 inflammasome | Sprague–Dawley rats | MCAO | – | Yu et al., 2017 |

**TABLE 3 | Selected noncoding RNAs involving NLRP3 inflammasome in cerebral I/R injury.**

| ncRNA | TUG1 | NEAT1 | MiR-139 | MiR-668 |
|-------|------|-------|---------|---------|
| Target Expression | miR-200a-3p | miR-22-3p | c-Jun | Mitochondrial function |
| Species | Adult C57BL/6 mice | Male Sprague–Dawley rats | Human neuroblastoma cells and mouse microglia cells |
| Model Treatment Pathway | MCAO Knockdown of TET2 TUG1/miR-200a-3p/NLRP3 | Gastrodin | NEAT1/miR-22-3p Axis | OGD/R |
| Therapeutic effect | Attenuate I/R-induced inflammatory response and brain injuries | Improve the neurological scores of rats, reduce the area of cerebral infarction, and inhibit pyroptosis | c-Jun/NLRP3 inflammasome-mediated pyroptosis and inflammatory response |
| References | Yin et al., 2021 | Zhang et al., 2021 | Wang Q.-S et al., 2020 | He and Zhang, 2020 |

**DISCUSSION**

Despite efforts to understand the pathophysiology of ischemic stroke, no effective neuroprotective drugs have been identified to modulate brain damage following ischemic stroke in human due to the complexity of ischemic stroke. With a fall of cerebral blood flow in ischemic stroke, the ischemic region contains two aspects, namely, ischemic core and penumbra, the damage to the brain in the penumbra is reversible based on the ionic homeostasis and transmembrane electrical potentials (Astrup et al., 1981), and how to restore blood flow in penumbra is a therapeutic target for the ischemic stroke clinic treatment (Ramos-Cabrera et al., 2011). Due to the sudden and rapid onset, ischemic stroke is often treated out of the therapeutic time window and, therefore, produces irreversible neuronal death (Sommer, 2017). As the most well-characterized inflammasome, NLRP3 inflammasome is closely related to the inflammatory response and pyroptosis; therefore, the blocking of NLRP3 inflammasome becomes a significant therapeutic target for ischemic stroke. Therefore, it is meaningful to downregulate the expression of NLRP3 inflammasome after cerebral I/R injury. The abnormal expression of NLRP3 inflammasome is not only detected in lab experiments but also is exactly confirmed in burgeoning clinical evidence (Chen et al., 2013; Zheng et al., 2013). It is shown that the neuronal upregulation of NLRP3 is an early event within the first 24 h of cerebral I/R injury which corresponds to the hyperacute and acute phase of human stroke (Franke et al., 2021). After the onset of ischemic stroke, the inhibition of NLRP3 inflammasome according to its expression time sequence could be considered as a future therapeutic target. Within 24 h of the occurrence of ischemic stroke, the early activation of NLRP3 inflammasome in microglia and subsequent activation in neurons should be effectively targeted according to the cell type, thereby, reducing cerebral I/R injury (Chumboatong et al., 2022; Wang et al., 2022), but the exact time needs further experimental studies. The NLRP3 inflammasome is a critical part of the innate immune system which regulates the cleaves and secretion of proinflammatory cytokines, such as IL-1β and IL-18 in response to the DAMP and PAMP signaling. Inflammatory cytokines are involved with the secondary brain injury in cerebral I/R, especially IL-1β and IL-18. It is influential for the treatment of cerebral I/R to regulate the expression of inflammatory cytokines under control, thus the involvement with inflammatory response makes NLRP3 inflammasome extensively investigated for assembly and activation. In addition to inflammation, the activation of NLRP3 inflammasome in ischemic brain tissue promotes the activities of pyroptosis and eventually aggravates brain injury. According to the activation and assembly of the NLRP3 inflammasome, molecules and compounds interfering with NLRP3 activation can alleviate cerebral I/R injury by...
downregulating the expression of the NLRP3 inflammasome in animal experiments. Importantly, these inhibitors have not been conducted in clinical trials due to their limited pharmacokinetic profiles and safety. Moreover, it is proven that the impact of the unspecific medications (SFN, Genipin) on the inflammasomes besides NLRP3 is negligible in the treatment of ischemic stroke (Franke et al., 2021). Therefore, highly specific and efficient inhibitors that focus on inhibiting the NLRP3 inflammasome and can effectively permeate the cell membrane and BBB will be an important topic for their research and clinical application.

Interestingly, it is a hot spot where mitochondria are closely related to NLRP3 inflammasome activation. Mitochondria are the central hub in innate and adaptive immune cells (Breda et al., 2019), and at the same time, mitochondria also participate in pyroptosis, a kind of cell death, which is related to cerebral I/R injury (Gurung et al., 2015). NLRP3 inflammasome triggers caspase-1-dependent mitochondrial damage. Caspase-1 activates multiple pathways to precipitate mitochondrial disassembly, which leads to the mROS production and dissipation of mitochondrial membrane potential, mitochondrial permeabilization, and mitochondrial network fragmentation (Yu et al., 2014). The molecules derived from mitochondria such as mROS, mtDNA, and cardiolipin are important regulators for the activation of the inflammasome, and mtDNA and cardiolipin are found to bind to NLRP3 and serve as a ligand for the activation of NLRP3 inflammasome. Now that the proven studies conducted that mitochondria are essential for NLRP3 inflammasome activation, the further studies are kindly suggested to focus on the specific mechanism by which mitochondria regulate the activation of NLRP3 inflammasomes.

CONCLUSION AND FUTURE PERSPECTIVE

The role of NLRP3 inflammasome in cerebral I/R injury is mainly concentrated on NLRP3 inflammasome-dependent cytokine release and pyroptosis, which makes NLRP3 inflammasome a target therapeutic protein in cerebral I/R injury. Although the essential of NLRP3 inflammasome activation in ischemic stroke has been proved, its specific role needs to be further explored. It is expected that the more effective pharmacokinetic profiles and safe medicines for NLRP3 inflammasome could be used for cerebral I/R injury treatments in clinical.

AUTHOR CONTRIBUTIONS

This study was finished under the guidance of SY. LW and WR proposed the idea of this manuscript. LW wrote the original draft. WR critically edited and revised the manuscript. QW, HX, and TL are responsible for the editing. YW, JD, and CZ investigated the literature. All authors contributed to the article and approved the submitted version.

FUNDING

This study was supported by the Science and Technology Department of Sichuan Province (Grant No. 2019YFS0543); Sichuan Administration of Traditional Chinese Medicine (20202ZD002, 2021ZD015, and 2020JC0150); the strategic cooperation project between Luzhou Municipal People’s Government (2019LZXN1DC02); China Postdoctoral Science Foundation (Grant No. 2020M683365); and the National Natural Science Foundation of China (Grant No. 82074378).

ACKNOWLEDGMENTS

We acknowledge the support of supervisor SY and appreciate the support of the National Natural Science Foundation of China, Science and Technology Department of Sichuan Province. The authors are sincerely thankful to all individuals who were involved in this study.

REFERENCES

Abderrazak, A., Syrovets, T., Couchie, D., El Hadri, K., Friguet, B., Simmet, T., et al. (2015). NLRP3 inflammasome: from a danger signal sensor to a regulatory node of oxidative stress and inflammatory diseases. Redox Biol. 4, 296–307. doi: 10.1016/j.redox.2015.01.008
Al Mamun, A., Chauhan, A., Yu, H., Xu, Y., Sharmeen, R., and Liu, F. (2018). Interferon regulatory factor 4/5 signaling impacts on microglial activation after ischemic stroke in mice. Eur. J. Neurosci. 47, 140–149. doi: 10.1111/ejn.13778
Allieri, A., Srivastava, S., Snow, R. C. M., Cash, D., Modo, M., Duchen, M. R., et al. (2013). Sulforaphane preconditioning of the Nrf2/ HO-1 defense pathway protects the cerebral vasculature against blood-brain barrier disruption and neurological deficits in stroke. Free Radic. Biol. Med. 65, 1012–1022. doi: 10.1016/j.freeradbiomed.2013.08.190
Alishahi, M., Farzaneh, M., Ghaedrahmati, F., Nejatbadoust, A., Sarkaki, A., and Khoshnam, S. E. (2019). NLRP3 inflammasome in ischemic stroke: as possible therapeutic target. Int. J. Stroke. 14, 574–591. doi: 10.11171/1747493019841242
Amantea, D., Nappi, G., Bernardi, G., Bagetta, G., and Corasaniti, M. T. (2009). Post-ischemic brain damage: pathophysiology and role of inflammatory mediators. FEBS J. 276, 13–26. doi: 10.1111/j.1742-4658.2008.06766.x
An, P., Xie, J., Qiu, S., Liu, Y., Wang, J., Xiu, X., et al. (2019). Hispidulin exhibits neuroprotective activities against cerebral ischemia reperfusion injury through suppressing NLRP3-mediated pyroptosis. Life Sci. 232, 116599. doi: 10.1016/j.lfs.2019.116599
Astrup, J., Siesjö, B. K., and Symon, L. (1981). Thresholds in cerebral ischemia - the ischemic penumbra. Stroke 12, 723–725. doi: 10.1161/01.STR.12.6.723
Barrington, J., Lemarchand, E., and Allan, S. M. (2017). A brain in flame; do inflammasomes and pyroptosis influence stroke pathology? Brain Pathol. 27, 205–212. doi: 10.1111/bpa.12478
Bellut, M., Papp, L., Bieber, M., Kraft, P., Stoll, G., and Schuhmann, M. K. (2021). NLPR3 inflammasome inhibition alleviates hypoxic endothelial cell death in vitro and protects blood-brain barrier integrity in marine stroke. Cell Death Dis. 13, 20. doi: 10.1038/s41419-021-04379-z
Breda, C. N. D. S., Davanzo, G. G., Basso, P. J., Saraiva Câmara, N. O., and Moraes-Vieira, P. M. M. (2019). Mitochondria as central hub of the immune system. Redox Biol. 26, 101255. doi: 10.1016/j.redox.2019.101255
Brez, P. (2015). Immunology: Caspase target drives pyroptosis. Nature 526, 642–643. doi: 10.1038/nature15632
Cai, W., Liu, S., Hu, M., Sun, X., Qiu, W., Zheng, S., et al. (2018). NLRP3 inflammasome inhibition alleviates hypoxic endothelial cell death in vitro and protects blood-brain barrier integrity in marine stroke. Cell Death Dis. 13, 20. doi: 10.1038/s41419-021-04379-z
Franke et al. (2021). NLRP3 inflammasome inhibition alleviates hypoxic endothelial cell death in vitro and protects blood-brain barrier integrity in marine stroke. Cell Death Dis. 13, 20. doi: 10.1038/s41419-021-04379-z
Breda, C. N. D. S., Davanzo, G. G., Basso, P. J., Saraiva Câmara, N. O., and Moraes-Vieira, P. M. M. (2019). Mitochondria as central hub of the immune system. Redox Biol. 26, 101255. doi: 10.1016/j.redox.2019.101255
Broz, P. (2015). Immunology: Caspase target drives pyroptosis. Nature 526, 642–643. doi: 10.1038/nature15632
Cai, W., Liu, S., Hu, M., Sun, X., Qiu, W., Zheng, S., et al. (2018). Post-stroke DHA treatment protects against acute ischemic brain injury by skewing...
macrophage polarity toward the M2 phenotype. *Transl. Stroke Res.* 9, 669–680. doi: 10.1007/s12795-018-0662-7

Cao, G., Jiang, N., Hu, Y., Zhang, Y., Wang, G., Yin, M., et al. (2016). Ruscogenin attenuates cerebral ischemia-induced blood-brain barrier dysfunction by suppressing TNF/NLRP3 inflammasome activation and the MAPK pathway. *Int. J. Mol. Sci.* 17, 1418. doi: 10.3390/ijms17091418

Chen, D., Xiong, X. Q., Zang, Y. H., Tong, Y., Zhou, B., Chen, Q., et al. (2017). BCL6 attenuates renal inflammation via negative regulation of NLRP3 transcription. *Cell Death Dis.* 8, e3156. doi: 10.1038/cddis.2017.567

Chen, F., Jiang, G. W., Liu, H., Li, Z. M., Pei, Y. X., Wang, H., et al. (2020). Melatonin alleviates intervertebral disk degeneration by disrupting the IL-1β/NF-κB-NLRP3 inflammasome positive feedback loop. *Bone Res.* 8, 10. doi: 10.1088/2044-0596/ab9895

Chen, K. H., Feng, L., Hu, W., Chen, J., Wang, X. Y., Wang, L., et al. (2019a). Optineurin inhibits NLRP3 inflammasome activation by enhancing mitophagy of renal tubular cells in diabetic nephropathy. *FASEB J.* 33, 4571–85. doi: 10.1096/fj.201801749R

Chen, S., Ma, Q., Krafft, P. R., Hu, Q., Rolland, W., Sherchan, P., et al. (2013). P2X7R/cryopyrin inflammasome axis inhibition reduces neuroinflammation after SAH. *Neurobiol. Dis.* 58, 296–307. doi: 10.1016/j.nbd.2013.06.011

Chen, Y. H., Meng, J., Bi, F. F., Li, H., Chang, C. C., Ji, C., et al. (2019b). EK7 regulates NLRP3 inflammasome activation and neuroinflammation post-traumatic brain injury. *Front Mol. Neurosci.* 12, 202. doi: 10.3389/fnmol.2019.00202

Cherry, J. D., Olschowka, J. A., and O’Banion, M. K. (2014). Neuroinflammation and M2 microglia: the good, the bad, and the inflamed. *J. Neuroinflammation.* 11, 98. doi: 10.1186/1742-2094-11-98

Chumboatong, W., Khamchai, S., Tocharus, C., Govitrapong, P., and Tocharus, J. (2022). Agomelatine exerts an anti-inflammatory effect by inhibiting microglial activation through TLR4/NLRP3 pathway in pMCAO Rats. *Neurotox. Res.* 40, 259–266. doi: 10.1007/s12974-021-00447-6

Conos, S. A., Chen, K. W., De Nardo, D., Hara, H., Whitehead, L., Núñez, G., et al. (2017). Active MLKL triggers the NLRP3 inflammasome in a cell-intrinsic manner. *Proc. Natl. Acad. Sci. U.S.A.* 114, E961–E969. doi: 10.1073/pnas.1613305114

Cui, D. L, Liu, S. Y, Tang, M. H, Lu, Y. Z, Zhao, M., Mao, R. W., et al. (2020). CBL6 alleviates renal inflammation via negative regulation of NLRP3 inflammasome activation by ion fluxes. *Front. Mol. Neurosci.* 13, 187. doi: 10.3389/fnmol.2019.01873

Dong, R., Huang, R., Wang, J., Liu, H., and Xu, Z. (2021). Effects of microglial activation and polarization on brain injury after stroke. *Front. Neurol.* 12, 620948. doi: 10.3389/fneur.2021.620948

Dong, X., Gao, J., Zhang, C. Y., Hayworth, C., Frank, M., and Wang, Z. (2019). Neutrophil membrane-derived nanovesicles alleviate inflammation to protect mouse brain injury from ischemic stroke. *ACS Nano.* 13, 1272–1283. doi: 10.1021/acsnano.8b06572

Du, R. H., Lu, M., Wang, C., Ding, J. H., Wu, G., and Hu, G. (2019). The pore-forming subunit Kir6.1 of the K-ATP channel negatively regulates the NLRP3 inflammasome to control insulin resistance by interacting with NLRP3. *Exp. Mol. Med.* 51, 1–13. doi: 10.1038/s41226-019-0291-6

Du, R. H., Tan, J., Sun, X. Y., Lu, M., Ding, J. H., and Hu, G. (2016). Fluoxetine inhibits NLRP3 inflammasome activation: implication in depression. *Int. J. Neuropsychopharmacol.* 19, pyw037. doi: 10.1038/ijnp.2015.103

Eberwein, J., Shin, H. J., Van Landuyt, S., and O’Neill, L. A. (2014). NLRP1 inflammasome activity in cerebral ischemia is mediated by neural death in ischemic stroke. *Cell Death Dis.* 4, e790. doi: 10.1038/cddis.2013.326

Fang, D. Y.-W., Santoro, T., Manzanero, S., Widiapradja, A., Cheng, Y.-L., Lee, S.-Y., et al. (2014). Intervenous immunoglobulin suppresses NLRP1 and NLRP3 inflammasome-mediated neuronal death in ischemic stroke. *Cell Death Dis.* 5, e120. doi: 10.1038/cddis.2014.105

Fink, S. L., and Cookson, B. T. (2005). Apoptosis, pyroptosis, and necrosis: mechanistic description of death and dying eukaryotic cells. *Infect. Immun.* 73, 1907–1916. doi: 10.1128/IAI.73.7.1907-1916.2005

Fronch, L., Eigenbrod, T., Muñoz-Planillo, R., Oszkurede, U., Kim, Y. G., Franke, M., Bieber, M., Kraft, P., Weber, A. N. R., Stoll, G., and Schuhmann, M. K. (2021). The NLRP3 inflammasome drives inflammation in ischemia/reperfusion injury after transient middle cerebral artery occlusion in mice. *Brain Behav. Immun.* 92, 223–233. doi: 10.1016/j.bbi.2020.12.009

Fu, Q., Zhai, Z. S., Wang, Y. Z., Xu, L. X., Jia, P. C., Xia, P., et al. (2018). NLRP3 deficiency alleviates severe acute pancreatitis and pancreatitis-associated lung injury in a mouse model. *Biomed. Res. Int.* 2018, 1294951. doi: 10.1155/2018/1294951

Fujise, K., Sugamura, K., Kurokawa, H., Matsuura, J., Ishii, M., Izuimiy, Y., et al. (2017). Colchicine improves survival, left ventricular remodeling, and chronic cardiac function after acute myocardial infarction. *Circ. J.* 81, 1174–1182. doi: 10.1253/circj.CJ-16-0949

Gao, C., Jia, W., Xu, W., Wu, Q., and Wu, J. (2021). Downregulation of CD151 restricts VCAM-1 mediated leukocyte infiltration to reduce neurobiological injuries after experimental stroke. *J. Neuroinflammation.* 18, 118. doi: 10.1186/s12974-021-02171-6

Ghafouri-Fard, S., Shoorei, H., and Taheri, M. (2020). Non-coding RNAs participate in the ischemia-reperfusion injury. *Biomed. Pharmacother.* 129, 110419. doi: 10.1016/j.biopha.2020.110419

Gianfrancesco, M. A., Dehaen, L., L’Homme, L., Herinxcx, G., Esser, N., Jansen, O., et al. (2019). Saturated fatty acids induce NLRP3 activation in human macrophages through K+ efflux resulting from phospholipid saturation and Na+-K-ATPase disruption. *Biochim. Biophys. Acta Mol. Cell. Biol. Lipids* 1864, 1017–1030. doi: 10.1016/j.jbpl.2019.04.001

Goldberg, E. L., Asher, J. L., Molony, R. D., Shaw, A. C., Zeiss, C. J., Wang, C., et al. (2017). β-Hydroxybutyrate deactivates neutrophil NLRP3 inflammasome to relieve gout flares. *Cell Rep.* 18, 2077–2087. doi: 10.1016/j.celrep.2017.02.004

Gong, T., Yang, Y. Q., Jin, T. C., Jiang, W., and Zhou, R. B. (2018a). Orchestration of NLRP3 inflammasome activation by ion fluxes. *Trends Immunol.* 39, 393–406. doi: 10.1016/j.tim.2018.01.009

Gong, Z., Pan, J., Shen, Q. F., Li, S., and Peng, Y. (2018b). Mitochondrial ROS induction of NLRP3 inflammasome activation during cerebral ischemia/reperfusion injury. *J. Neuroinflammation.* 15, 242. doi: 10.1186/s12974-018-1282-6
Gou, X., Xu, D., Li, F., Hou, K., Fang, W., and Li, Y. (2021). Pyroptosis in stroke—new insights into disease mechanisms and therapeutic strategies. *J. Physiol. Biochem.* 77, 511–529. doi: 10.1007/s13110-021-00817-w

Groß, C. J., Mishra, R., Schneider, K. S., Mérédard, G., Wettenhausen, J., Dittler, D. C., et al. (2016). K efflux-independent NLRP3 inflammasome activation by small molecules targeting mitochondria. *Immunity* 45, 761–773. doi: 10.1016/j.immuni.2016.08.010

Gülkе, E., Gelderblom, M., and Magnus, T. (2018). Danger signals in stroke and their role on microglia activation after ischemia. *Ther. Adv. Neurol. Disord.* 11, 86. doi: 10.3389/fnmol.2018.00086

Guo, W. J., Liu, W., Chen, Z., Gu, Y. H., Peng, S., Shen, L. H., et al. (2021). NLRP3 is involved in the regulation of the NLRP3 inflammasome activation through Sirt1-dependent mitochondrial fission and endoplasmic reticulum stress. *Front. Mol. Neurosci.* 11, 86. doi: 10.3389/fnmol.2020.00086

Guo, Z., Yu, S., Chen, X., Ye, R., Zhu, W., and Liu, X. (2016). NLRP3 is involved in the regulation of the NLRP3 inflammasome activation via ANT1-dependent mitochondrial homeostasis. *Immunity* 45, 36–47. doi: 10.1016/j.immuni.2016.08.010

Hossmann, K.-A. (2006). Pathophysiology and therapy of experimental stroke. *J. Neurosci.* 26, 1073–1083. doi: 10.1523/JNEUROSCI.0616-06.2006

Hornung, V., and Latz, E. (2010). Critical functions of priming and activation of the NLRP3 inflammasome. *J. Immunol.* 185, 1387–1394. doi: 10.4049/jimmunol.0901323

Jia, Y., Tong, Y., Min, L., Li, Y., and Cheng, Y. (2021). Protective effects of oridonin on brain ischemia/reperfusion injury by inhibiting the NLRP3 inflammasome activation. *Tissue Cell* 71, 101514. doi: 10.1016/j.tice.2021.101514

Jiang, H., He, H., Chen, Y., Huang, W., Cheng, J., Ye, J., et al. (2017). Identification of a selective and direct NLRP3 inhibitor to treat inflammatory disorders. *J. Exp. Med.* 214, 3218–3236. doi: 10.1084/jem.20171419

Jiang, T., Wu, M., Zhang, Z., Yan, C., Ma, Z., He, S., et al. (2019). Electroacupuncture attenuated cerebral ischemic injury and neuroinflammation through α7nACHR-mediated inhibition of NLRP3 inflammasome in stroke rats. *Mol. Med.* 25, 22. doi: 10.1186/s10295-019-0091-4

Jin, R., Yang, G., and Li, G. (2010). Inflammatory mechanisms in ischemic stroke: role of inflammatory cells. *J. Leukoc. Biol.* 87, 779–789. doi: 10.1189/jlb.1109766

Jia, J., Li, Z., Zhang, Z., Wang, X., and Sun, R. (2018). Mitochondria: diversity of a selective and direct NLRP3 inhibitor to treat inflammatory disorders. *J. Exp. Med.* 214, 3218–3236. doi: 10.1084/jem.20171419

Joly, S., Ma, N., Sadler, J. J., Soll, D. R., Cassel, S. L., and Sutterwala, F. S. (2009). Cutting edge: *Candida albicans* hyphae formation triggers activation of the Nlrp3 inflammasome. *J. Immunol.* 183, 3578–3581. doi: 10.4049/jimmunol.0901323

Kelley, N., Jeltema, D., Duan, Y., and He, Y. (2019). The NLRP3 inflammasome: an overview of mechanisms of activation and regulation. *Int. J. Mol. Sci.* 20, 3328. doi: 10.3390/ijms20133328

Kendall, R. L., and Holian, A. (2021). The role of lysosomal ion channels in lysosome dysfunction. *Inhal. Toxicol.* 33, 41–54. doi: 10.1080/08985837.2021.1876168

Lee, H. E., Yang, G., Park, Y. B., Kang, H. C., Cho, Y. Y., Lee, H. S., et al. (2021). Epigallocatechin-3-gallate prevents acute gout by suppressing NLRP3 inflammasome activation and mitochondrial fission and endoplasmic reticulum stress after transient global cerebral ischemia in rats. *Int. J. Neurosci.* 20, 1–14. doi: 10.1080/0275507X.2019.1622402

Lamkanfi, M., and Dixit, V. M. (2014). Mechanisms and functions of NLRP3 inflammasomes in atherosclerosis. *J. Atheroscler. Thromb.* 24, 443–451. doi: 10.5551/jat.RV17001

Lee, H. E., Yang, G., Park, Y. B., Kang, H. C., Cho, Y. Y., Lee, H. S., et al. (2021). Epigallocatechin-3-gallate prevents acute gout by suppressing NLRP3 inflammasome activation and mitochondrial fission and endoplasmic reticulum stress after transient global cerebral ischemia in rats. *Int. J. Neurosci.* 20, 1–14. doi: 10.1080/0275507X.2019.1622402

Lee, H. E., Yang, G., Park, Y. B., Kang, H. C., Cho, Y. Y., Lee, H. S., et al. (2021). Epigallocatechin-3-gallate prevents acute gout by suppressing NLRP3 inflammasome activation and mitochondrial fission and endoplasmic reticulum stress after transient global cerebral ischemia in rats. *Int. J. Neurosci.* 20, 1–14. doi: 10.1080/0275507X.2019.1622402

Lee, H. E., Yang, G., Park, Y. B., Kang, H. C., Cho, Y. Y., Lee, H. S., et al. (2021). Epigallocatechin-3-gallate prevents acute gout by suppressing NLRP3 inflammasome activation and mitochondrial fission and endoplasmic reticulum stress after transient global cerebral ischemia in rats. *Int. J. Neurosci.* 20, 1–14. doi: 10.1080/0275507X.2019.1622402
promoting Nrf-2 pathway in ischaemic stroke. J. Cell. Mol. Med. 25, 9753–9766. doi: 10.1111/jcmm.19623

Li, N., Liu, C., Wang, C., Chen, R., Li, X., Wang, Y., et al. (2021b). Early changes of NLRP3 inflammasome activation after hypoxic-ischemic brain injury in neonatal rats. Int. J. Clin. Exp. Pathol. 14, 209–220.

Li, S., Lin, Q. S., Shao, X. H., Mou, S., Gui, L. Y., Wang, L., et al. (2019). NLRP3 inflammasome inhibition attenuates cisplatin-induced renal fibrosis by decreasing oxidative stress and inflammation. Exp. Cell Res. 383, 111488. doi: 10.1016/j.yexcr.2019.07.001

Li, W., Cheng, H., Xu, Z., Lv, Y., Zhao, Z., Zhang, J., et al. (2020a). Melodihenine B attenuates NLRP3 expression in a cerebral ischemia/reperfusion-induced neuronal injury rat model. Folia Neuropathol. 58, 30–37. doi: 10.5114/fn.2020.94004

Li, Y., Wang, R., Xue, L., Yang, Y., and Zhi, F. (2020b). Astilbin protects against cerebral ischemia/reperfusion injury by inhibiting cellular apoptosis and ROS-NLRP3 inflammasome axis activation. Int. Immunopharmacol. 84, 106571. doi: 10.1016/j.intimp.2020.106571

Liepinsh, E., Barbals, R., Dahl, E., Sharipo, A., Staub, E., and Otting, G. (2001). The inflammasome: a molecular platform triggering activation of inflammatory caspases and processing of pro-IL-beta. Mol. Cell. 10, 417–426. doi: 10.1016/S1097-2765(02)05993-9

Matsinger, P. (2002). The danger model: a renewed sense of self. Science 296, 301–305. doi: 10.1126/science.1071039

McKenzie, B. A., Mamik, M. K., Saito, L. B., Boghobian, R., Monaco, M. C., Major, E. O., et al. (2018). Caspase-1 inhibition prevents glial inflammasome activation and pyroptosis in models of multiple sclerosis. Proc. Nat. Acad. Sci. U.S.A. 115, E6065. doi: 10.1073/pnas.1722041115

Menu, P., and Vince, J. E. (2011). The NLRP3 inflammasome in health and disease: the good, the bad and the ugly. Clin. Exp. Immunol. 166, 1–15. doi: 10.1111/j.1365-2249.2011.04440.x

Minutoli, L., Puzzolo, D., Rinaldi, M., Irrera, N., Marini, H., Arcoraci, V., et al. (2016). ROS-Mediated NLRP3 inflammasome activation in brain, heart, kidney, and testis ischemia/reperfusion injury. Oxid. Med. Cell. Longev. 2016, 2183026. doi: 10.1155/2016/2183026

Mo, Z.-T., Zheng, J., and Liao, Y.-L. (2021). Icariin inhibits the expression of IL-1β-IL-6 and TNF-α induced by OGD/R through the IRE1/XBP1 pathway in microglia. Pharm. Biol. 59, 1473–1479. doi: 10.1080/13880981.2021.1991595

Mridha, A. R., Wree, A., Robertson, A. B. A., Yeh, M. M., Chen, C. D., Van Rooyen, G. M., et al. (2017). NLRP3 inflammasome blockade reduces liver inflammation and fibrosis in experimental NASH in mice. J. Hepatol. 66, 1037–1046. doi: 10.1016/j.jhep.2017.01.022

Nomura, J., So, A., Tamura, M., and Busso, N. (2015). Intracellular ATP decrease mediates NLRP3 inflammasome activation upon nigericin and crystal stimulation. J. Immunol. 195, 5718–5724. doi: 10.4049/jimmunol.1402512

Pan, R., Xie, Y., Fang, W., Liu, Y., and Zhang, Y. (2022a). USP20 mitigates ischemic stroke in mice by suppressing neuroinflammation and neuron death via regulating PTEN signal. Int. Immunopharmacol. 103, 107840. doi: 10.1016/j.intimp.2021.107840

Pan, X., Fan, J., Peng, F., Xiao, L., and Yang, Z. (2022b). SET domain containing 7 promotes oxygen-glucose deprivation/reoxygenation-induced PC12 cell inflammation and oxidative stress by regulating Keap1/Nrf2/ARE and NF-kB pathways. Bioengineered 13, 7253–7261. doi: 10.1080/21655979.2022.2054830

Park, J. H., Kim, D. W., Shin, M. J., Park, J., Han, K. H., Lee, K. W., et al. (2020). Tat-indoleamine 2,3-dioxygenase 1 elicits neuroprotective effects on ischemic injury. RMB Rep. 53, 582–587. doi: 10.5483/MBRRep.2020.53.11.114

Park, S., Juliana, C., Hong, S., Datta, P., Hwang, I., Fernandes-Alnemri, T., et al. (2013). The mitochondrial antiviral protein MAVS associates with NLRP3 and regulates its inflammasome activity. J. Immunol. 191, 4358–4366. doi: 10.4049/jimmunol.1301170

Peng, J., Wang, H., Gong, Z., Li, X., He, L., Shen, Q., et al. (2020). Idebenone attenuates cerebral inflammatory injury in ischemia and reperfusion via dampening NLRP3 inflammasome activity. Mol. Immunol. 123, 74–87. doi: 10.1016/j.molimm.2020.04.013

Petrielli, V., Papan, S., Dostert, C., Mayor, A., Martinon, F., and Tschopp, J. (2007). Activation of the NALP3 inflammasome is triggered by low intracellular potassium concentration. Cell Death Differ. 14, 1583–1589. doi: 10.1038/sj.cdd.4402195

Qiu, Z., Lei, S., Zhao, B., Wu, Y., Su, W., Liu, M., et al. (2017). NLRP3 inflammasome activation-mediated pyroptosis aggravates myocardial ischemia/reperfusion injury in diabetic rats. Oxid. Med. Cell. Longev. 2017, 9743280. doi: 10.1155/2017/9743280

Qu, X.-Y., Zhang, Y.-M., Tao, L.-N., Gao, H., Zhai, J.-H., Sun, J.-M., et al. (2019). Xinongao injections protect against cerebral ischemia/reperfusion injury and alleviate blood-brain barrier disruption in rats, through an underlying mechanism of NLRP3 inflammasomes suppression. Chin. J. Nat. Med. 17, 498–505. doi: 10.1007/s11875-5364(14)30071-8

Ramos-Cabrera, P., Campos, F., Sobrino, T., and Castillo, J. (2011). Targeting the ischemic penumbra. Stroke. 42 (1 Suppl), 57–11. doi: 10.1161/STROKEAHA.110.596684

Ren, J.-X., Li, C., Yan, X.-L., Qu, Y., Yang, Y., and Guo, Z.-N. (2021). Crosstalk between oxidative stress and ferroptosis/oxidization in ischemic stroke: possible targets and molecular mechanisms. Oxid. Med. Cell. Longev. 2021, 6643382. doi: 10.1155/2021/6643382

Rupalla, K., Allegrini, P. R., Sauer, D., and Wiessner, C. (1998). Time course of microglial activation and apoptosis in various brain regions after permanent focal cerebral ischemia in mice. Acta Neuropathol. 96, 172–178. doi: 10.1007/s004010050878
Wree, A., Eguchi, A., McGeough, M. D., Pena, C. A., Johnson, C. D., Canbay, A., et al. (2014). NLRP3 inflammasome activation results in hepatocyte pyroptosis, liver inflammation, and fibrosis in mice. Hepatology 59, 898–910. doi: 10.1002/hep.26592

Wu, D., Chen, Y., Sun, Y., Gao, Q., Li, H., Yang, Z., et al. (2020). Target of MCC950 in inhibition of NLRP3 inflammasome activation: a literature review. Inflammation 43, 17–23. doi: 10.1007/s10753-019-01098-8

Xiong, X., Xu, L., Wei, L., White, R. E., Ouyang, Y.-B., and Giffard, R. G. (2015). IL-4 is required for sex differences in vulnerability to focal ischemia in mice. Stroke 46, 2271–2276. doi: 10.1161/STROKEAHA.115.008897

Xu, J., Wang, A., Meng, X., Yalkun, G., Xu, A., Gao, Z., et al. (2021a). Edaravone versus edaravone alone for the treatment of acute ischemic stroke: a phase III, randomized, double-blind, comparative trial. Stroke 52, 772–780. doi: 10.1161/STROKEAHA.115.006877

Xu, L., Shen, L. Y., Yu, X. L., Li, P., Wang, Q., and Li, C. Q. (2020). Effects of irisin on osteoblast apoptosis and osteoporosis in postmenopausal osteoporosis rats through upregulating Nrf2 and inhibiting NLRP3 inflammasome. Exp. Ther. Med. 19, 1084–1090. doi: 10.3892/etm.2019.8313

Ye, Y., Gao, X., Wang, L., Yang, M., and Xie, R. (2021b). Bukcholizol ameliorates cerebral ischemia-reperfusion injury by modulating NLRP3 inflammasome and Nrf2 signaling. Respir. Physiol. Neurobiol. 292, 103707. doi: 10.1016/j.resp.2021.103707

Xue, M., Li, T., Wang, Y., Chang, Y. P., Cheng, Y., Lu, Y. H., et al. (2019). Empagliflozin protects cardiovascular function in mice. Proc. Natl. Acad. Sci. USA 116, 1705–1707. doi: 10.1073/pnas.1909588116

Yang, Z., Zhao, T.-Z., Zou, Y.-J., Zhang, J. H., and Feng, H. (2014). Hypoxia Induces autophagic cell death through hypoxia-inducible factor 1α in microglia. PLoS ONE 9, e96509. doi: 10.1371/journal.pone.0096509

Yao, S., Li, L. J., Sun, X., Hua, J., Zhang, K. Q., Hao, L., et al. (2019). FTY720 inhibits MPP-induced microglial activation by affecting NLRP3 Inflammasome activation. J. Neuroimmune Pharmacol. 14, 478–492. doi: 10.1007/s11481-019-09843-4

Ye, Y., Jin, T., Zhang, X., Zeng, Z., Ye, B., Wang, J., et al. (2019). Mesoindigo protects against focal cerebral ischemia-reperfusion injury by inhibiting NLRP3 inflammasome activation and regulating microglia/macrophage polarization via Toll-like receptor 4/NF-κB signaling pathway. Front. Cell. Neurosci. 13, 553. doi: 10.3389/fncel.2019.00553

Yin, M., Chen, W.-P., Yin, X.-P., Tu, J.-H., Hu, N., and Li, Z.-Y. (2021). lncRNA TUG1 demethylated by TET2 promotes NLRP3 expression, contributes to cerebral ischemia/reperfusion inflammatory injury. ASN. Neuro. 13, 1759909142110002347. doi: 10.1177/1759909142110002347

Yu, C., He, Q., Zheng, J., Li, L. Y., Hou, Y. H., and Song, F. Z. (2017). Sulforaphane improves outcomes and slows cerebral ischemia/reperfusion injury via inhibition of NLRP3 inflammasome activation in rats. Int. Immunopharmacol. 45, 74–78. doi: 10.1016/j.intimp.2017.01.034

Yu, J., Nagasu, H., Murakami, T., Hoang, H., Broderick, L., Hoffman, H. M., et al. (2014). Inflammasome activation leads to Caspase-1-dependent mitochondrial damage and block of mitophagy. Proc. Natl. Acad. Sci. U.S.A. 111, 15514–15519. doi: 10.1073/pnas.1411489

Yu, J. W., and Lee, M. S. (2016). Mitochondria and the NLRP3 inflammasome: physiological and pathological relevance. Arch. Pharm. Res. 39, 1503–1518. doi: 10.1007/s12272-016-0827-4

Zeng, Q., Zhou, Y., Liang, D., He, H., Liu, X., Zhu, R., et al. (2020). Exosomes secreted from bone marrow mesenchymal stem cells attenuate oxygen-glucose deprivation/reoxygenation-induced pyroptosis in PC12 cells by promoting AMPK-dependent autophagic flux. Front. Cell. Neurosci. 14, 182. doi: 10.3389/fncel.2020.00182

Zhang, D., Qian, J., Zhang, P., Li, H., Shen, H., Li, X., et al. (2019). Gasdermin D serves as a key effector of pyroptosis in experimental cerebral ischemia and reperfusion model both in vivo and in vitro. J. Neurosci. Res. 97, 645–660. doi: 10.1002/jnr.24385

Zhang, H.-S., Ouyang, B., Ji, X.-Y., and Liu, M.-F. (2021). Gastrodin alleviates cerebral ischemia/reperfusion injury by inhibiting pyroptosis by regulating the lncRNA NEAT1/miR-22-3p axis. Neurochem. Res. 46, 1747–1758. doi: 10.1007/s11064-021-03285-2

Zhao, B., Fei, Y., Zhu, J., Yin, Q., Fang, W., and Li, Y. (2021). PAF receptor inhibition attenuates neuronal pyroptosis in cerebral ischemia/reperfusion injury. Mol. Neurobiol. 58, 6520–6539. doi: 10.1007/s12031-021-05237-0

Zhao, J., Piao, X., Wu, Y., Liang, S., Han, F., Liang, Q., et al. (2020). Cerepharanthines attenuates cerebral ischemia/reperfusion injury by reducing NLRP3 inflammasome-induced inflammation and oxidative stress via inhibiting 12/15-LOX signaling. Biomed. Pharmacother. 127, 110151. doi: 10.1016/j.biopha.2020.110151

Zheng, X., Feng, S., Gong, Z., and Xing, Q. (2013). NLRP3 inflammasomes show high expression in aorta of patients with atherosclerosis. Heart Lung Circ. 22, 746–750. doi: 10.1016/j.hlnc.2013.01.012

Zheng, M., Williams, E. P., Malireddi, R. K. S., Karki, R., Banoth, B., Burton, A., et al. (2020). Impaired NLRP3 inflammasome activation/pyroptosis leads to robust inflammatory cell death via caspase-8/RIPK3 during coronavirus infection. J. Biol. Chem. 295, 14040–14052. doi: 10.1074/jbc.RA120.015036

Zhong, Z. Y., Umemura, A., Sanchez-Lopez, E., Liang, S., Shalapour, S., Wong, J., et al. (2017). Methods for monitoring Ca²⁺ and ion channels in the lysosome. Cell Calcium. 64, 20–28. doi: 10.1016/j.cca.2016.12.001

Zhong, Z., Liang, S., Sanchez-Lopez, E., He, F., Shalapour, S., Lin, X.-J., et al. (2018). New mitochondrial DNA synthesis enables NLRP3 inflammasome activation. Nature 560, 198–203. doi: 10.1038/s41586-018-0372-2

Zhang, Z. Y., Umemura, A., Sanchez-Lopez, E., Liang, S., Shalapour, S., Wong, J., et al. (2016). NF-κB restricts inflammasome activation via elimination of damaged mitochondria. Cell 164, 896–910. doi: 10.1016/j.cell.2015.12.057

Zhou, Y., Gu, Y., and Liu, J. (2019). BRD4 suppression alleviates cerebral ischemia-induced brain injury by blocking gial activation via the inhibition of inflammatory response and pyroptosis. Biochem. Biophys. Res. Commun. 519, 481–488. doi: 10.1016/j.jbc.2019.07.097

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Copyright © 2022 Wang, Ren, Wu, Liu, Wei, Ding, Zhou, Xu and Yang. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.