

Review Article

A Protective Role of Glibenclamide in Inflammation-Associated Injury

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Glibenclamide is the most widely used sulfonylurea drug for the treatment of type 2 diabetes mellitus (DM). Recent studies have suggested that glibenclamide reduced adverse neuroinflammation and improved behavioral outcomes following central nervous system (CNS) injury. We reviewed glibenclamide's anti-inflammatory effects: abundant evidences have shown that glibenclamide exerted an anti-inflammatory effect in respiratory, digestive, urological, cardiological, and CNS diseases, as well as in ischemia-reperfusion injury. Glibenclamide might block K_{ATP} channel, Sur1-Trpm4 channel, and NOD-like receptor pyrin domain containing 3 (NLRP3) inflammasome activation, decrease the production of proinflammatory mediators (TNF- α , IL-1 β , and reactive oxygen species), and suppress the accumulation of inflammatory cells. Glibenclamide's anti-inflammation warrants further investigation.

1. Introduction

Glibenclamide, an oral hypoglycemic agent, belongs to the class of sulfonylureas, and its clinical utilization dates back to the 1960s [1]. Actually, sulfonylureas were discovered accidentally, as the antimicrobial sulfonamides caused hypoglycemia in animals. Thereafter, glibenclamide has been used widely in the type II diabetes mellitus (DM).

The mechanism of glibenclamide in DM treatment is due to its inhibition of ATP-sensitive potassium channel (K_{ATP}) (Sur1-Kir6.2) in pancreatic β islet cells, which leads to the depolarization of β cell plasma membrane and activation of voltage-gated calcium channels. Calcium influx triggers insulin release from β cells [2, 3]. Glibenclamide is an K_{ATP} channel blocker and broad-spectrum ATP-binding cassette transporter (ABC) inhibitor. K_{ATP} channels are widely distributed in muscle, pancreatic beta cells, and the brain. Their activity is regulated by adenine nucleotides, activated by falling ATP and rising ADP [4]. K_{ATP} channel is a heterooctamer consisting of four pore-forming subunits (Kir6.x) and four regulatory sulfonylurea receptor (Sur) subunits [5]

composed in a 1 : 1 stoichiometry as a tetramer (Sur-Kir6.X)₄. This complex conducts potassium ions across cell membranes and thereby couples cellular energy metabolism to membrane electrical activity. Three isoforms of Sur (Sur1, Sur2A, and Sur2B) have been cloned and are specified as pancreatic, cardiac, and (vascular) smooth muscle types, respectively [6]. The Kir subunit is either Kir6.1 or Kir6.2 [6]. Different combinations of Kir6.X and Sur isoforms in different cellular/tissue distributions result in functional diversity [5]: the classical β cell type and neuronal type with the combination of Kir6.2 and Sur1 (SUR₁-Kir6.2)₄; the cardiac and skeletal type with the combination of Kir6.2 and Sur2A (Sur2A-Kir6.2)₄; the smooth muscle type with the combination of Kir6.2 and Sur2B (Sur2B-Kir6.2)₄; or Kir6.1 and Sur2B (Sur2B-Kir6.1)₄ [5].

Besides its hypoglycemia effects, glibenclamide recently has been shown to play role in inflammation regulation. In addition to combining with the Kir6.X subunit to form the K_{ATP} channel, Sur1 also associates with an ATP- and calcium-sensitive nonselective cation channel to form Sur1-Trpm4 channels (previously called the Sur1-NCCa-ATP

channel). Interestingly, but not surprisingly, glibenclamide has been found to inhibit Sur1-Trpm4 channels by directly binding the Sur1 subunit to protect against inflammation-associated injury in the central nervous system (CNS) [7]. Activation of Sur1-Trpm4 channels depolarizes the cell membrane, which is associated with cell death and cerebral edema [8]. Glibenclamide reduces adverse neuroinflammation and behavioral outcomes in CNS injury [9]. Furthermore, glibenclamide displays a protective role in inflammation-induced injury in various systems, including respiration [10–12], digestion [13], urology [14–16], cardiology [17], CNS [18–21], some special conditions such as melioidosis [12, 22] and ischemia-reperfusion (IR) injury [23–26], and septic shock [27–29]. Here, we review anti-inflammatory effects of glibenclamide and its possible mechanisms.

2. Anti-Inflammatory Roles of Glibenclamide

2.1. Glibenclamide and Respiratory Diseases. K_{ATP} channels are expressed on the basolateral membrane of airway epithelial cells. K_{ATP} channel activation stimulates proliferation of fibroblasts, hepatocytes, and epithelial cells and induces migration of neutrophils and airway epithelial cells [30–32].

Bronchopulmonary dysplasia is a devastating lung complication in preterm infants. Inflammation plays a critical role in bronchopulmonary dysplasia development [33–35]. Recently, Liao et al. found that glibenclamide (5 μ M) protected neonatal mice from developing bronchopulmonary dysplasia, which was associated with inhibition of caspase-1 activation, reduction in interleukin-1 beta (IL-1 β) production, and suppression of neutrophils and macrophages influx [10].

Allergic asthma is characterized by excessive T-helper type 2 (Th2) immune response, large eosinophilic airway inflammation, airway hyperresponsiveness (AHR), and mucus hyperproduction, and subsequent airway remodeling [36]. In a mouse model of ovalbumin-induced asthma, glibenclamide significantly reduced eosinophils in bronchoalveolar lavage fluid and reduced Th2-associated cytokines like IL-5 and IL-13, therefore attenuated airway inflammation and AHR [11]. In vitro, glibenclamide also significantly downregulated ovalbumin-stimulated Th2 cytokine release, such as IL-5, IL-4, and IL-13 [11]. The authors attributed this anti-inflammatory effect to inhibition of IL-4/IL-13/p-STAT6/VCAM-1 signaling pathway.

Acute lung injury/acute respiratory distress syndrome (ALI/ARDS) is also associated with an overwhelming inflammatory response: proinflammatory cytokines IL-1 β and IL-18 levels are high in bronchoalveolar lavage fluid from patients with ALI/ARDS and correlate with mortality [37]. Melioidosis patients pretreated with glibenclamide showed lower accumulation of IL-1 β and IL-18 and better outcomes, which was mediated by the inhibition of NLRP3 inflammasome [12].

2.2. Glibenclamide and Severe Acute Pancreatitis (SAP). Acute pancreatitis (AP), characterized by parenchymal and peripancreatic fat necrosis, often displays a mild and self-limiting inflammatory process and has a good prognosis.

However, excessive inflammatory reactions result in severe acute pancreatitis (SAP), with a high mortality and morbidity [13]. Although the pathogenesis of SAP has not yet been clarified, inflammatory mediators such as IL-1 β , IL-6, IL-8, IL-10, tumor necrosis factor-alpha (TNF)- α , platelet-activating factor, and monocyte chemoattractant protein-1 (MCP-1) are considered vital for SAP development [38]. In vitro, glibenclamide significantly reduced IL-1 β release in lipopolysaccharide- (LPS-) induced peritoneal cells [13]. In a mouse model of cerulein-induced AP, York et al. showed that glibenclamide (500 mg/kg, intraperitoneal) dramatically reduced serum levels of IL-6, lipase, and amylase [13]. Furthermore, in NLRP3 knockout (Nlrp3 $^{-/-}$) mice, cerulein significantly provoked less pancreatic edema, leukocyte infiltration, and acinar cell apoptosis compared to wild-type mice [39], which indicates the importance of the NLRP3 inflammasome in SAP pathogenesis.

2.3. Glibenclamide and Urinary System Diseases. Inflammation plays a significant role in the progression of chronic kidney disease, both in animal experimental models [40] and in patients [41]. Expression of the NLRP3 inflammasome is enhanced in chronic kidney diseases [42]. Inflammatory markers, including TNF- α and NLRP3, were highly expressed in the kidneys from adenine diet-treated rats. Glibenclamide (10 mg/kg daily, 8 weeks) significantly reduced expression of TNF- α and NLRP3 [14, 15].

Bladder inflammation (cystitis) is a common and critical problem in urology. Hughes et al. indicated that NLRP3 played an important role in the pathogenesis of cyclophosphamide-induced cystitis, while glibenclamide could effectively block the inflammatory response [16]. In a cyclophosphamide-induced rat model of cystitis, glibenclamide treatment decreased caspase-1 activity along with levels of IL-1 β and IL-18 and subsequent bladder inflammation [16].

2.4. Glibenclamide and Melioidosis. Melioidosis, caused by the Gram-negative saprophytic bacillus *Burkholderia pseudomallei*, results in bacteremia, abscesses in many organ systems, pneumonia, and soft tissue infection [43, 44]. DM is an established risk factor for susceptibility to melioidosis [12, 45–47]. However, a cohort study reported that melioidosis patients with a preadmission diagnosis of diabetes treated with glibenclamide had decreased levels of TNF- α , IL-18-R1, and IL-8 and lower in-hospital mortality compared to patients without diabetes [12]. The same group confirmed their findings in a mouse diabetes model of streptozocin-induced melioidosis [22]. Glibenclamide-pretreated diabetic mice showed lower *Burkholderia pseudomallei* loads in the liver and spleen, reduced neutrophils and macrophages infiltration in lung, and decreased IL-1 β levels in the bronchoalveolar lavage fluid and lungs compared to control mice [22]. Whether glibenclamide displays a similar protective role against other infections is not known and needs further investigation.

Sepsis has been defined as a systemic inflammatory response to infection [48]. The newest definition for septic shock is “a subset of sepsis in which underlying circulatory, cellular, and metabolic abnormalities are associated with a

greater risk of mortality than sepsis alone” [49]. Contradictory results regarding glibenclamide’s administration in sepsis and septic shock have been reported. In a dog endotoxemia model, Landry and Oliver showed that glibenclamide (0.15 mg/kg, intravenous) effectively restored normotension [29]. Similarly, other studies showed that glibenclamide restored normotension in pig and rat endotoxemia models [27, 28]. However, sepsis or septic shock was also reported to be a negative effect of glibenclamide in experimental models and in human studies [27–29, 50–52]. In a ceecal ligation and puncture septic shock rat model, the administration of glibenclamide failed to improve cardiovascular and inflammatory parameters and actually increased mortality [50]. Similarly, in patients with septic shock, glibenclamide failed to improve hemodynamics or reduce noradrenaline requirements [51, 52]: in the randomized, double-blind, placebo-controlled crossover study, Warrillow et al. [51] found that glibenclamide (20 mg, enterally) treatment in patients with septic shock had no effect on hemodynamic variables, norepinephrine levels, or lactate concentrations but significantly decreased blood glucose concentration and increased dextrose utilization; Morelli et al. [52] performed another prospective, randomized, double-blind pilot study in patients with septic shock and showed that parenteral glibenclamide administration decreased blood glucose concentrations in a dose-dependent manner but failed to improve hemodynamics or reduce norepinephrine requirements. Possible explanations for this discrepancy might be related to species, pathogen, dose, and administration route (enteral versus intravenous) differences, which deserve further studies.

The proposed mechanisms of glibenclamide treatment of septic shock are reviewed as follows: first, glibenclamide could directly inhibit K_{ATP} channels in vascular smooth muscle (VSM). It is reported that K_{ATP} channels, especially Sur2B-Kir6.1 type, are abundantly expressed in vascular smooth muscle cells and have been suggested to regulate vascular tone in the presence of shock states and systemic inflammation [6, 53]. Under normal resting conditions, the K_{ATP} channel is closed, however, under increased tissue metabolism or tissue hypoxia conditions, and these channels are activated and opened [54]. A recent study by Shi and colleagues indicated that lipopolysaccharide upregulates expression of the K_{ATP} channel, especially the Kir6.1 and Sur2B subunits in smooth muscle cells dissociated from the mouse aorta [55]. Activation of K_{ATP} channels results in efflux of K^+ and hyperpolarization of the cell membrane, which reduces Ca^{2+} influx and subsequently blocks constriction of vascular smooth muscle cells.

Second, the protective effect of glibenclamide might be associated with anti-inflammation, as it effectively reduced the lipopolysaccharide-induced release of mediators like IL-1 β and TNF- α in vitro [56]. Many studies have shown that lipopolysaccharide-induced IL-1 β production was NLRP3 inflammasome activation dependent [15, 57], while this activation could be inhibited by glibenclamide [58].

2.5. Glibenclamide and Cardiovascular Diseases. Atherosclerosis is the primary cause of ischemic heart diseases, and

macrophages play a critical role in the pathogenesis of atherosclerosis and the formation of vulnerable plaque. The K_{ATP} subunits, especially Sur2A and Kir6.2, were upregulated on macrophages in vulnerable plaques, accompanied by the overproduction of TNF- α [17]. In vitro, glibenclamide (10 μ M) directly inhibited the lipopolysaccharide-induced TNF- α production by RAW264.7 cells [17]. In a mouse model of atherosclerosis, intragastric administration of glibenclamide at 2.5 mg/kg inhibited the formation/development of vulnerable plaque, diminished the vulnerability index (VI), and decreased the macrophage content in the plaque [17]. Furthermore, in a streptozocin-induced diabetic endotoxemia mouse model, pretreatment with glibenclamide (5 mg/kg i.p., 14 days) significantly attenuated myocardial injury via decreased production of IL- β and TNF- α , infiltration of macrophages, and apoptosis of cardiomyocytes [59]. In vitro, glibenclamide pretreatment significantly decreased peritoneal macrophage IL- β expression but had no effect on TNF- α expression. Additionally, glibenclamide inhibited NLRP3 and caspase-1 expression [59].

2.6. Glibenclamide and Central Nervous System Diseases. The inflammatory response, especially sterile inflammation, is involved in the pathogenesis of CNS diseases, such as multiple sclerosis (MS) [21], progression of neurogenesis [20], and brain injury after subarachnoid hemorrhage (SAH) [18, 19]. Sur1-regulated channels, including K_{ATP} (Sur1-Kir6.2) channels and Sur1-Trpm4 channels, are expressed in neurons, astrocytes, oligodendrocytes, endothelial cells, and microglial cells [7, 8, 60]. Glibenclamide’s inhibition of microglial K_{ATP} (Sur1-Kir6.2) channels and Sur1-Trpm4 channels significantly ameliorates neuroinflammation and improves neurological function.

Neural precursor cells are important in the progression of neurogenesis after brain injury. The K_{ATP} channels are expressed in microglial cells, where they are activated by cerebral ischemia and inflammatory stimuli [61, 62]. Ortega et al. reported that in lipopolysaccharide plus IFN- γ -stimulated microglial cells, glibenclamide treatment restored neural precursor cells activity by blocking K_{ATP} channels, inhibiting the microglia-induced suppression of neural precursor cell production, decreasing proinflammatory cytokines TNF- α and IL-6, and increasing MCP-1 [20]. Glibenclamide treatment enhanced MCP-1 production, which was partially associated with enhanced neural precursor cell differentiation [63, 64].

Multiple sclerosis is an autoimmune disease characterized by chronic inflammation, demyelination, and neurodegeneration of the CNS that causes neurological disability [65, 66]. Using an experimental of autoimmune encephalomyelitis induced by MOG35–55 peptides in wild-type mice, Ortega et al. demonstrated that Sur1-Trpm4 channels were upregulated mainly on astrocytes [20]. The lumbar spinal cords of wild-type/experimental autoimmune encephalomyelitis mice treated with glibenclamide showed significantly less inflammatory lesions in leukocytes and T cells and less proinflammatory cytokines, including TNF- α , IFN- γ , and IL-17, which correlated with better preservation of myelin, axons, and mature and precursor oligodendrocytes [20].

SAH is a major contributor to hemorrhage stroke [67]. The Sur1-Trpm4 channel is upregulated in cortical tissues in a rat model of SAH, induced either by puncture of the internal carotid arteries [18] or by blood injection into the entorhinal cortex [19]. Glibenclamide treatment significantly reduced expression of proinflammatory cytokines TNF- α and nuclear factor- κ B (NF- κ B), decreased permeability and neuroinflammation, and ameliorated impaired spatial learning memory by blocking the Sur1-Trpm4 channel [19].

A recent study performed by Kurland et al. suggested that glibenclamide inhibition of lipopolysaccharide-treated microglial Sur1-Trpm4 channels downregulated inducible nitric oxide synthase (iNOS) transcription and thus reduced the formation of peroxynitrite radicals, which partly attributed to proinflammatory and harmful effects of activated microglia in the central nervous system [60].

2.7. Glibenclamide and Ischemia-Reperfusion Injury. Ischemia-reperfusion (IR) injury usually results in the production of inflammatory mediators and reactive oxygen species induced by reperfusion. IR induces activation of K_{ATP} channels and cytokine production, such as IL-1 β , IL-6, IL-17, and TNF- α [68–70]. Glibenclamide reduced the production of inflammatory mediators via inhibiting Sur1 and the regulatory subunit of the K_{ATP} channel in the ischemic tissue [61]. Recruitment and activation of leukocytes and subsequent release of inflammatory mediators have been proposed to explain the inflammation-induced reperfusion-associated injury [71]. Glibenclamide was suggested to inhibit neutrophil migration during the acute inflammatory response via blocking K_{ATP} channels [30]. The role of glibenclamide in different IR models was investigated, including the CNS [8, 23, 72], kidney [25], intestine [24], and testis [26].

The brain is very sensitive to ischemia; 5 minutes of ischemia can lead to irreversible neuronal cell death [23]. A review by Sun et al. indicated that K_{ATP} channel activation was important in the pathway of IR injury in the brain and was a promising target for protecting neurovascular function in stroke [70]. Another two studies performed by Simard et al. showed that glibenclamide treatment significantly reduced cerebral edema, infarct volume, and mortality in brain IR [8, 72]. Glibenclamide has been shown to ameliorate inflammatory mediators in the hippocampus in a rat IR model induced by occlusion of the bilateral carotid artery for 15 minutes followed by reperfusion for 60 minutes [23]. Glibenclamide reduced neutrophil infiltration, as well as TNF- α and prostaglandine E2 (PGE2), and boosted anti-inflammatory cytokine IL-10 expression [23].

Renal IR injury occurs in many clinical situations like organ transplantation, severe shock, and trauma. Both Kir6.2 and Sur1 are expressed in the normal renal epithelial cells [73] and upregulated in kidney IR [74]. In parallel with the increased expression of Kir and Sur, massive proinflammatory cytokines like IL-6, IL-17, and TNF- α increased [73]. Glibenclamide has generated contradictory results in kidney IR. Pompermayer et al. demonstrated that glibenclamide (20 mg/kg) was associated with amelioration of renal dysfunction in rat IR [25], characterized by decreased vascular permeability, neutrophil accumulation, NF- κ B translocation,

and cytokine production like TNF- α [25]. In contrast, Zhang et al. showed that glibenclamide enhanced IR injury in renal epithelial function as demonstrated by altered histology in the kidneys of newborn rats [73]. Possible explanations for these contradictory findings could be related to age of the animals, since age influences calcium-activated potassium channels and voltage-activated potassium channel expression [75].

Increased vascular permeability, accumulation of neutrophils, as well as elevation of IL-1 β , IL-6, and TNF- α was observed in a rat intestinal IR model. Histology showed marked intestinal destruction and inflammation, characterized by a striking loss of villi and crypts, and submucosal, hyperemia, and tissue edema [24]. Glibenclamide treatment significantly decreased intestinal IR injury in dose-dependent manner [24].

Testicular torsion, recognized as a urological emergency, can lead to testicular necrosis/apoptosis and decreased fertility in males, infants, and adolescents [76]. Reactive oxygen species produced in IR was responsible for the necrosis/apoptosis cascade [77]. Shimizu et al. showed that glibenclamide ameliorated the IR injury as demonstrated by a significant decrease in malondialdehyde concentration, myeloperoxidase activity, histological injury scores, and the number of TUNEL-positive germinal cells [26]. The binding of glibenclamide at the Sur1 subunit in the spermatogenic cells is a suggested mechanism since K_{ATP} channels are expressed on the sarcolemma, inner mitochondrial membrane, and nuclear membrane [7, 78, 79]. The role of glibenclamide in these inflammation-associated injury and disorders are summarized in Table 1.

3. Mechanisms of Glibenclamide Underlying Its Anti-Inflammatory Role

Glibenclamide exerts an anti-inflammatory effect involving inhibition of the NLRP3 inflammasome, decreased production of proinflammatory cytokines, reduced recruitment and migration of inflammatory cells, and production of nitric oxide (Figure 1).

3.1. Glibenclamide Might Inhibit the Activation of NLRP3 Inflammasome. In 2002, Martinon and coworkers introduced the term “inflammasomes” to describe a high-molecular-weight complex present in the cytosol of stimulated immune cells that mediates activation of inflammatory caspases [80]. The NLRP3 inflammasome (known as cryopyrin), like a number of NOD-like receptors (NLR), consists of the NLRP3 scaffold, the apoptosis-associated speck-like protein ASC/PYCARD adaptor and caspase-1, which is expressed predominantly in circulating monocytes and tissue macrophages [44]. It can be triggered by a plethora of pathogen-associated molecular patterns (PAMPs) and danger-associated molecular patterns (DAMPs) and leads to caspase-1 activation and cytokine secretion, including IL-18 and IL-1 β , which are associated with tissue damage and chronic inflammation. Inappropriate activation of NLRP3 inflammasome has been linked to the pathophysiologic processes of many inflammatory diseases, such as gout, pseudogout, silicosis, and asbestosis [81–84]. Glibenclamide inhibited NLRP3

TABLE 1: The anti-inflammatory effects of glibenclamide in previous studies.

Objects	Anti-inflammatory effects
Bronchopulmonary dysplasia model of mice [10]	Decreases in IL-1 β , neutrophils, and macrophages in bronchoalveolar lavage fluid
Allergic asthma model of mice [11]	Inhibitions in airway hyperresponsiveness, airway inflammation, and Th2 cytokines
Severe acute pancreatitis model of mice [13]	Decreases in serum levels of IL-6, IL-1 β , lipase, and amylase
Cystitis model of rats [16]	Decreases in bladder mucosa edema, neutrophils infiltration, and proinflammatory cytokines expression
Sepsis patients [12]	Attenuations of inflammatory responses and mortality
Sepsis model of mice [22]	Suppressions of inflammatory cells in the lung, bacterial dissemination, and IL-1 β secretion
Atherosclerosis model of mice [17]	Diminishment of vulnerability index and decrease in macrophages infiltration in plaque
Lipopolysaccharide-treated RAW264.7 cells [17]	Inhibition of TNF- α expression
Endotoxemia model of mice [59]	Attenuations of myocardial injury and macrophages infiltration and IL-1 β expression
Autoimmune encephalomyelitis model of mice [21]	Ameliorations of inflammatory cells and cytokines
Neural precursor cells treated with lipopolysaccharide and IFN- γ [20]	Restoration of lipopolysaccharide and IFN- γ induced decrease in neural precursor cell number
SAH model of rats [19]	Amelioration of brain-blood barrier permeability, decreased expression of proinflammatory cytokines like TNF- α , and suppression of neuron cell death
Brain IRI model of rats [23]	Amelioration of neutrophil infiltration, inhibitions in TNF- α , and PGE2 expressions, and upregulation of anti-inflammatory cytokine IL-10
Renal IRI model of rats [25]	Amelioration of vascular permeability and inhibitions in neutrophil accumulation and TNF- α production
Intestinal IRI model of rats [24]	Amelioration of vascular permeability, inhibition in neutrophil accumulation, and suppressive productions of cytokines like TNF- α , IL-1 β , and IL-6
Testis IRI model of rats [26]	Downregulations of malondialdehyde concentration, myeloperoxide activity, and histological score

SAH: subarachnoid hemorrhage; IRI: ischemia-reperfusion-induced injury.

inflammasome activation and subsequently protected against organ inflammation and tissue damage either in a cerulean-induced obese mouse model of SAP, or in an adenine-diet rat model of chronic kidney disease, or in a cyclophosphamide-induced rat model of bladder inflammation, or in a lipopolysaccharide-induced myocardial injury in streptozocin-induced diabetic rats [13, 14, 16, 59]. Since potassium release and low-intracellular potassium concentration have been reported to trigger NLRP3 activation by all the stimuli both in mouse peritoneal macrophages and in human macrophages/monocytes [85, 86], inhibition of K_{ATP} channels that prevent depletion of cytosolic potassium might be a very plausible mechanism of glibenclamide-mediated suppression of the NLRP3 inflammasome [16]. In contrast, Lamkanfi et al. reported that K_{ATP} channels were dispensable for inhibition of the NLRP3 inflammasome [58]. As evidence showed that another sulfonylurea drug glipizide also inhibited Sur1 containing K_{ATP} channels but failed to inhibit NLRP3 inflammasome activation. Glibenclamide abolished inflammasome activation in macrophages lacking K_{ATP} channel subunits as well [58]. Although the exact mechanisms remain unknown, glibenclamide could inhibit NLRP3 inflammasome activation via upstream inhibition of the inflammasome and downstream blockade of the P2X7 receptor, which then reduces NLRP3 inflammasome-mediated

capase-1 activation and inhibits secretion of the mature form of the IL-1 β protein [58].

3.2. Glibenclamide Inhibits Production of Non-NLRP3 Inflammasome-Mediated Proinflammatory Cytokines. Excessive production of proinflammatory cytokines and recruitment of neutrophils are involved in the pathophysiology of tissue damage in inflammatory disease [30, 87]. In addition to IL-1 β , glibenclamide also reduced NLRP3 noninflammasome-mediated proinflammatory cytokines, including TNF- α and INF- γ . Glibenclamide reduced lipopolysaccharide-induced TNF- α and INF- γ release and the corresponding mRNAs in monocytes by inhibiting the ATP/P2X7 receptor/calcium/AP-1 signaling [56], which was responsible for its protective role in lipopolysaccharide-induced sepsis shock [27–29]. Since cytosolic calcium was presumed to bind and activate transcription factors AP-1, which mediated the transcription of proinflammatory mediators in monocytes [88], the investigators ascribed the inhibitory potency of glibenclamide to its reduction of P2X7 receptor-induced calcium transients as well as the hypoxia-induced calcium elevations in monocytes [56]. ATP, released by injured erythrocytes, could activate P2X7 receptors on monocytes [89], which would provide a pore for calcium influx [56, 90]. P2X7 receptor activation depended on

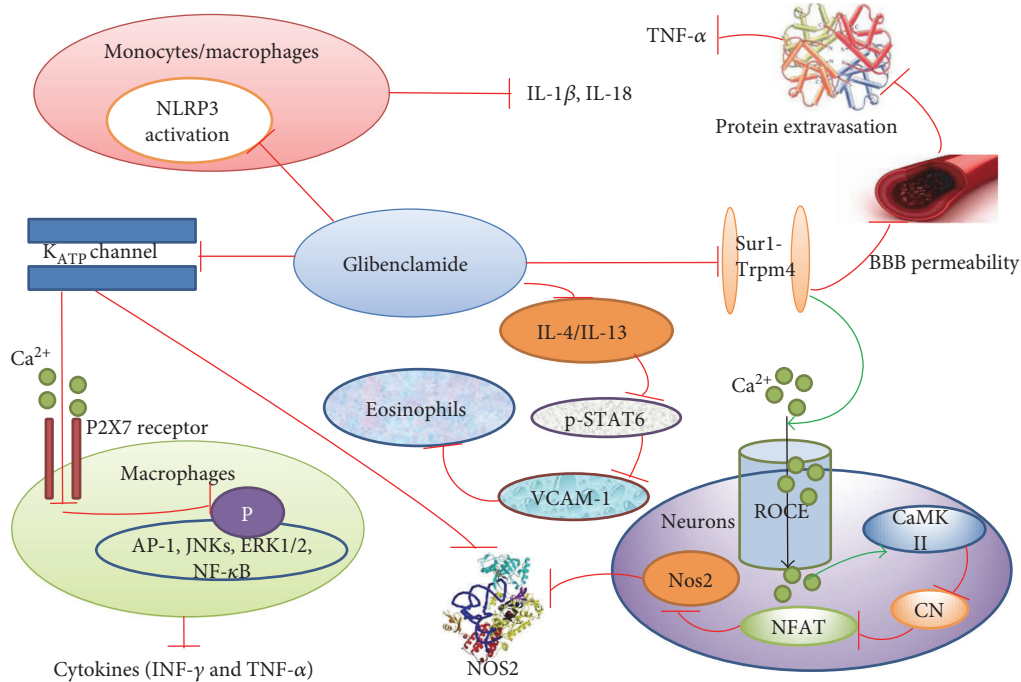


FIGURE 1: Anti-inflammatory mechanisms of glibenclamide. Glibenclamide blocks K_{ATP} channel to prevent K^+ efflux and changes the membrane potential, which inhibits P2X7 receptor-mediated Ca^{2+} influx, resulting in reducing inflammatory cytokines. Glibenclamide blocks the Sur1-Trpm4 channel to decrease the BBB permeability, which reduces extravasated protein-induced production of $TNF-\alpha$. Glibenclamide inhibition of Sur1-Trpm4 channel increases ROCE-mediated Ca^{2+} influx, which activates CaMKII and then inhibits CN/NFAT/Nos2 signaling. Glibenclamide can inhibit NLRP3 inflammasome-mediated production of $IL-1\beta$ and reduce $IL-13/4$, which blocks the migration of eosinophils. The inhibitory effect is indicated by red lines with bar ends; the excitatory effect is indicated by green lines with arrow ends. ROCE: receptor-operated Ca^{2+} entry channel; CaMKII: Ca^{2+} /calmodulin protein kinase II; CN: calcineurin; NFAT: nuclear of activated T cells; Nos2: inducible nitric oxide synthase gene; AP-1: activator protein-1; ERKs: extracellular signal-regulated kinases; JNKs: Jun N-terminal kinases; BBB: blood-brain barrier; p-STAT6: phosphorylated signal transducer and activator of transcription 6; VCAM-1: vascular cell adhesion molecular 1.

membrane potential, which could be influenced by Kir6.2 subunits of K_{ATP} channels expressed on monocytes [91]. Glibenclamide's effect on calcium was indirectly mediated by a change in membrane potential through inhibition of the Kir6.2 subunits [56].

Consistently, another study demonstrated that glibenclamide decreased $TNF-\alpha$ transcription by inhibiting the K_{ATP} channel as well. In the monocyte/macrophage RAW264.7 cell line, lipopolysaccharide significantly increased $TNF-\alpha$ transcript levels and robustly enhanced expression of K_{ATP} subunits, Sur1, Sur2A, Kir6.1, and Kir6.2 [17], which were depressed by glibenclamide. This study also provided strong evidence that the K_{ATP} channel was located upstream of $NF-\kappa B$ as well as mitogen-activated protein kinases (MAPKS) in lipopolysaccharide-TLR (toll-like receptors) signaling. By inhibiting the K_{ATP} channel, glibenclamide suppressed phosphorylation of $NF-\kappa B$, extracellular signal-regulated kinases (ERK) 1/2 and Jun N-terminal kinases (JNKs) in RAW264.7 cells and therefore reduced the release of $TNF-\alpha$, which rescued the progression of atherosclerosis in mice [17].

In both rat and human SAH models, glibenclamide could reduce the release of $TNF-\alpha$ and $NF-\kappa B$ via inhibition Sur1-Trpm4 channel [18, 19]. The upregulation of Sur1-Trpm4 channel in SAH model resulted in increased blood-brain

barrier permeability [18] and protein extravasation, which was related to the expression of $TNF-\alpha$ and $NF-\kappa B$ and promotes neuroinflammation [92]. Glibenclamide treatment could significantly reduce the extravasation of serum proteins by blocking Sur1-Trpm4 channel and subsequently reduce $TNF-\alpha$ and $NF-\kappa B$ expression.

3.3. Glibenclamide Inhibits Migration of Neutrophils and Eosinophils. Neutrophils are the main components of the innate immune system. Accumulation of neutrophils has been related to tissue damage due to their overwhelming release of cytotoxic and proinflammatory mediators, such as arachidonic acid metabolites, cytokines, superoxide anions, and nitric oxide [93] in infectious and ischemic diseases [23–25, 30]. Neutrophils have a high permeability to potassium, and their efflux occurs mainly through potassium channels [30]. Da Silva-Santos et al. showed that glibenclamide could effectively inhibit migration of neutrophils by blocking K_{ATP} channels [30], thereby reducing neutrophil accumulation and preventing further organ damage in ischemic brain, intestines, kidney, and lungs in a rat melioidosis model [22–25]. Additionally, glibenclamide's inhibitory effect on neutrophil migration is somewhat indirectly mediated by its inhibition of NLRP3 inflammasome assembly. NLRP3 inflammasome is the key for $IL-1\beta$ maturation, and

IL-1 β upregulates the expression of endothelial intercellular adhesion molecules essential for the recruitment of neutrophils to the area of inflammation [94–96].

Eosinophils are potent effector cells implicated in allergic inflammation and helminth infection, predominantly involved in the pathogenesis of allergic asthma, eosinophilic bronchitis, and eosinophilic colitis. Cui et al. showed that glibenclamide could effectively inhibit eosinophil infiltration and reduced airway inflammation in mouse model of asthma [11]. Eosinophil recruitments are dependent on specific eosinophil chemokines like eotaxins and endothelial adhesion molecules like VCAM-1 [97, 98]. The production of eotaxins and VCAM-1 is regulated by IL-4 and IL-13. These cytokines promote the development of Th2 lymphocytes and activating structural cells like bronchial fibroblasts and respiratory epithelial cells to produce eotaxins and promote the expression of VCAM-1 in STAT6-dependent manner. Glibenclamide has been reported to reduce the production of Th2-associated cytokines, IL-4 and IL-13. Therefore, glibenclamide blockade of eosinophilic migration might due to the inhibition of IL-4/IL-13/phosphorylated STAT6/VCAM-1 signaling pathway. The mechanism underlying glibenclamide in the suppression of Th2 cytokines needs to be clarified in the future.

3.4. Glibenclamide Downregulates Generation of Reactive Oxygen Species. Excessive reactive oxygen species (ROS) triggers oxidative damage to biomolecules, resulting in the development of a variety of diseases, including ischemia-reperfusion injury, septic shock, and neuroinflammation [23, 26, 60]. There are various types of ROS, including oxygen-derived free radicals (superoxide, hydroxyl radicals, and nitric oxide) and nonradical oxygen derivatives of high reactivity (hydrogen peroxide, peroxynitrite, and hypochlorite) [99]. The generation of reactive oxygen species is dependent on specialized enzymes, such as NADPH-oxidase, myeloperoxidase, and nitric oxide synthase (NOS). Three isoforms of NOS have been identified: endothelial (eNOS), neuronal (nNOS) (both constitutively expressed), and inducible (iNOS) (produced primarily after stimulation by cytokines and endotoxin).

Shimizu et al. reported that glibenclamide ameliorated the rat testis IR by reducing the myeloperoxidase, and thus reducing the oxidative stress [26]. But the underlying mechanism of inhibition myeloperoxidase by glibenclamide is not clear. The authors suggested that this might be mediated by blockade membrane K_{ATP} channel, since the openers of membrane K_{ATP} channel do not reduced injury. In addition to myeloperoxidase, glibenclamide could inhibit NOS2 expression. Overproduction of nitric oxide due to NOS2 activation has been found in septic shock and is suggested to be one of the mechanisms underlying hypotension and vascular hyporeactivity to vasoconstrictors [100–102]. Wu et al. showed that glibenclamide inhibited NOS2 induction caused by LPS in cultured macrophages and in the anaesthetized rat [103]. This explains the beneficial effects of glibenclamide in endotoxin-induced shock. The inflammatory cytokine TNF- α has been reported to upregulate NOS2,

thereby mediated a generalized production of the potent vasodilator, nitric oxide [104]. Since glibenclamide effectively reduced the production of TNF- α in septic shock [56], it might be possible that glibenclamide downregulates NOS2 by inhibiting TNF- α . This mechanism needs to be investigated.

Furthermore, a recent study indicated that glibenclamide reduced *Nos2* mRNA dose dependently in activated microglia by blocking Sur1-Trpm4 channels [60]. NOS2 expression in activated microglia is regulated by NFATc1 (a kind of NFAT, nuclear factor of activated T cells) [105], which is phosphorylated and sequenced in the cytoplasm. Nuclear translocation occurs following dephosphorylation of NFATc1 by the Ca^{2+} -sensitive phosphatase, calcineurin (CN) [106]. Sur1-Trpm4 channel in microglia has been shown to be significantly upregulated by LPS stimulation in vivo and in vitro. The activation of Sur1-Trpm4 channels in activated microglia depolarizes the cell membrane, which decreases the inward driving force for Ca^{2+} [7]. While glibenclamide inhibition of Sur1-Trpm4 leads to increase in intracellular concentration of Ca^{2+} ($[Ca^{2+}]_i$), which causes the activation of CaMKII (Ca^{2+} /calmodulin protein kinase II) [107]. Activated CaMKII inhibits the phosphatase activity of CN through phosphorylation [108], thus fails to dephosphorylate NFATc1 and inhibits the nuclear translocation [107, 108], which results in induction of *Nos2* mRNA induction. The nitrite production by NOS2 in activated microglia is attributable to neuroinflammation and injury in a growing list of CNS diseases. The beneficial effects of glibenclamide in multiple sclerosis, ischemic stroke, and hemorrhage stroke might be mediated by the inhibition of Sur1-Trpm4- $[Ca^{2+}]_i$ -CN/NFAT-*Nos2* signaling. Glibenclamide could also inhibit oxidative stress by elevating activity of antioxidant enzyme, such as glutathione peroxidase, superoxide dismutase, and catalase [23, 109].

Hypoglycemia, the most common side effect of glibenclamide, can result in large releases of norepinephrine, epinephrine, and steroids, which can inhibit macrophage migration and production of TNF- α [110, 111]. Studies involved in our review showed that patients who were not pretreated had no obvious changes in blood glucose levels [10–12, 17–19, 22–26, 59]. This finding indicates that the anti-inflammatory effect of glibenclamide is independent of its hypoglycemic effect.

4. Conclusions

Glibenclamide exerts a protective role in inflammation-related injury and disorders. With the blockades of NLRP3 inflammasome/IL-1 β signaling and non-NLRP3 mechanisms like Sur1-Trpm4/TNF- α signaling and Sur1-Trpm4/*Nos2*/ROS signaling, glibenclamide downregulates proinflammatory cytokines and reactive oxygen species and suppresses migration of inflammatory cells. Glibenclamide might be a promising agent for inflammation-associated disorders. Further investigations could focus on translating the anti-inflammatory potential of glibenclamide to clinical application in inflammatory diseases.

Conflicts of Interest

The authors declare they have no competing interests.

Authors' Contributions

Gensheng Zhang, Xiuhui Lin, and Shufang Zhang contributed equally to this work.

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References

- [1] A. Marble, "Glibenclamide, a new sulphonylurea: whither oral hypoglycaemic agents?" *Drugs*, vol. 1, no. 2, pp. 109–115, 1971.
- [2] F. M. Ashcroft, "K(ATP) channels and insulin secretion: a key role in health and disease," *Biochemical Society Transactions*, vol. 34, Part 2, pp. 243–246, 2006.
- [3] C. G. Nichols, "KATP channels as molecular sensors of cellular metabolism," *Nature*, vol. 440, no. 7083, pp. 470–476, 2006.
- [4] M. Benadiba and Y. Maor, "Importance of ABC transporters in drug development," *Current Pharmaceutical Design*, vol. 22, no. 38, pp. 5817–5829, 2016.
- [5] A. P. Babenko, L. Aguilar-Bryan, and J. Bryan, "A view of sur1/KIR6.X, KATP channels," *Annual Review of Physiology*, vol. 60, no. 1, pp. 667–687, 1998.
- [6] H. Yokoshiki, M. Sunagawa, T. Seki, and N. Sperelakis, "ATP-sensitive K⁺ channels in pancreatic, cardiac, and vascular smooth muscle cells," *The American Journal of Physiology*, vol. 274, no. 1 Part 1, pp. C25–C37, 1998.
- [7] J. M. Simard, S. K. Woo, G. T. Schwartzbauer, and V. Gerzanich, "Sulphonylurea receptor 1 in central nervous system injury: a focused review," *Journal of Cerebral Blood Flow and Metabolism*, vol. 32, no. 9, pp. 1699–1717, 2012.
- [8] J. M. Simard, M. Chen, K. V. Tarasov et al., "Newly expressed SUR1-regulated NC(Ca-ATP) channel mediates cerebral edema after ischemic stroke," *Nature Medicine*, vol. 12, no. 4, pp. 433–440, 2006.
- [9] N. Caffes, D. B. Kurland, V. Gerzanich, and J. M. Simard, "Glibenclamide for the treatment of ischemic and hemorrhagic stroke," *International Journal of Molecular Sciences*, vol. 16, no. 3, pp. 4973–4984, 2015.
- [10] J. Liao, V. S. Kapadia, L. S. Brown et al., "The NLRP3 inflammasome is critically involved in the development of bronchopulmonary dysplasia," *Nature Communications*, vol. 6, p. 8977, 2015.
- [11] W. Cui, S. Zhang, Z. Cai et al., "The antidiabetic agent glibenclamide protects airway hyperresponsiveness and inflammation in mice," *Inflammation*, vol. 38, no. 2, pp. 835–845, 2015.
- [12] G. C. Koh, R. R. Maude, M. F. Schreiber et al., "Glyburide is anti-inflammatory and associated with reduced mortality in melioidosis," *Clinical Infectious Diseases*, vol. 52, no. 6, pp. 717–725, 2011.
- [13] J. M. York, K. J. Castellanos, R. J. Cabay, and G. Fantuzzi, "Inhibition of the nucleotide-binding domain, leucine-rich containing family, pyrin-domain containing 3 inflammasome reduces the severity of experimentally induced acute pancreatitis in obese mice," *Translational Research*, vol. 164, no. 4, pp. 259–269, 2014.
- [14] M. J. Mulla, K. Myrtolli, J. Potter et al., "Uric acid induces trophoblast IL-1 β production via the inflammasome: implications for the pathogenesis of preeclampsia," *American Journal of Reproductive Immunology*, vol. 65, no. 6, pp. 542–548, 2011.
- [15] D. De Nardo and E. Latz, "NLRP3 inflammasomes link inflammation and metabolic disease," *Trends in Immunology*, vol. 32, no. 8, pp. 373–379, 2011.
- [16] F. M. Hughes Jr., N. P. Vivar, J. G. Kennis et al., "Inflammasomes are important mediators of cyclophosphamide-induced bladder inflammation," *American Journal of Physiology. Renal Physiology*, vol. 306, no. 3, pp. F299–F308, 2014.
- [17] M. Y. Ling, Z. Y. Ma, Y. Y. Wang et al., "Up-regulated ATP-sensitive potassium channels play a role in increased inflammation and plaque vulnerability in macrophages," *Atherosclerosis*, vol. 226, no. 2, pp. 348–355, 2013.
- [18] J. M. Simard, Z. Geng, S. K. Woo et al., "Glibenclamide reduces inflammation, vasogenic edema, and caspase-3 activation after subarachnoid hemorrhage," *Journal of Cerebral Blood Flow and Metabolism*, vol. 29, no. 2, pp. 317–330, 2009.
- [19] C. Tosun, D. B. Kurland, R. Mehta et al., "Inhibition of the Sur1-Trpm4 channel reduces neuroinflammation and cognitive impairment in subarachnoid hemorrhage," *Stroke*, vol. 44, no. 12, pp. 3522–3528, 2013.
- [20] F. J. Ortega, J. Vukovic, M. J. Rodriguez, and P. F. Bartlett, "Blockade of microglial KATP channel abrogates suppression of inflammatory-mediated inhibition of neural precursor cells," *Glia*, vol. 62, no. 2, pp. 247–258, 2014.
- [21] T. K. Makar, V. Gerzanich, V. K. Nimmagadda et al., "Silencing of Abcc8 or inhibition of newly upregulated Sur1-Trpm4 reduce inflammation and disease progression in experimental autoimmune encephalomyelitis," *Journal of Neuroinflammation*, vol. 12, no. 1, p. 210, 2015.
- [22] G. C. Koh, T. A. Weehuizen, K. Breitbach et al., "Glyburide reduces bacterial dissemination in a mouse model of melioidosis," *PLoS Neglected Tropical Diseases*, vol. 7, no. 10, article e2500, 2013.
- [23] D. M. Abdallah, N. N. Nassar, and R. M. Abd-El-Salam, "Glibenclamide ameliorates ischemia-reperfusion injury via modulating oxidative stress and inflammatory mediators in the rat hippocampus," *Brain Research*, vol. 1385, pp. 257–262, 2011.
- [24] K. Pompermayer, F. A. Amaral, C. T. Fagundes et al., "Effects of the treatment with glibenclamide, an ATP-sensitive potassium channel blocker, on intestinal ischemia and reperfusion injury," *European Journal of Pharmacology*, vol. 556, no. 1–3, pp. 215–222, 2007.
- [25] K. Pompermayer, D. G. Souza, G. G. Lara et al., "The ATP-sensitive potassium channel blocker glibenclamide prevents renal ischemia/reperfusion injury in rats," *Kidney International*, vol. 67, no. 5, pp. 1785–1796, 2005.
- [26] S. Shimizu, R. Oikawa, P. Tsounapi et al., "Blocking of the ATP sensitive potassium channel ameliorates the ischaemia-reperfusion injury in the rat testis," *Andrology*, vol. 2, no. 3, pp. 458–465, 2014.

- [27] G. Vanelli, S. N. Hussain, and G. Aguggini, "Glibenclamide, a blocker of ATP-sensitive potassium channels, reverses endotoxin-induced hypotension in pig," *Experimental Physiology*, vol. 80, no. 1, pp. 167–170, 1995.
- [28] S. M. Gardiner, P. A. Kemp, J. E. March, and T. Bennett, "Regional haemodynamic responses to infusion of lipopolysaccharide in conscious rats: effects of pre- or post-treatment with glibenclamide," *British Journal of Pharmacology*, vol. 128, no. 8, pp. 1772–1778, 1999.
- [29] D. W. Landry and J. A. Oliver, "The ATP-sensitive K⁺ channel mediates hypotension in endotoxemia and hypoxic lactic acidosis in dog," *The Journal of Clinical Investigation*, vol. 89, no. 6, pp. 2071–2074, 1992.
- [30] J. E. Da Silva-Santos, M. C. Santos-Silva, Q. Cunha Fde, and J. Assreuy, "The role of ATP-sensitive potassium channels in neutrophil migration and plasma exudation," *The Journal of Pharmacology Experimental Therapeutics*, vol. 300, no. 3, pp. 946–951, 2002.
- [31] N. T. Trinh, A. Prive, L. Kheir et al., "Involvement of KATP and KvLQT1 K⁺ channels in EGF-stimulated alveolar epithelial cell repair processes," *American Journal of Physiology. Lung Cellular and Molecular Physiology*, vol. 293, no. 4, pp. L870–L882, 2007.
- [32] N. T. Trinh, A. Prive, E. Maille, J. Noel, and E. Brochiero, "EGF and K⁺ channel activity control normal and cystic fibrosis bronchial epithelia repair," *American Journal of Physiology. Lung Cellular and Molecular Physiology*, vol. 295, no. 5, pp. L866–L880, 2008.
- [33] R. M. Ryan, Q. Ahmed, and S. Lakshminrusimha, "Inflammatory mediators in the immunobiology of bronchopulmonary dysplasia," *Clinical Reviews in Allergy & Immunology*, vol. 34, no. 2, pp. 174–190, 2008.
- [34] F. Mosca, M. Colnaghi, and M. Fumagalli, "BPD: old and new problems," *The Journal of Maternal-Fetal & Neonatal Medicine*, vol. 24, no. Supplement 1, pp. 80–82, 2011.
- [35] S. Benko, D. J. Philpott, and S. E. Girardin, "The microbial and danger signals that activate Nod-like receptors," *Cytokine*, vol. 43, no. 3, pp. 368–373, 2008.
- [36] D. T. Umetsu, J. J. McIntire, O. Akbari, C. Macaubas, and R. H. DeKruyff, "Asthma: an epidemic of dysregulated immunity," *Nature Immunology*, vol. 3, no. 8, pp. 715–720, 2002.
- [37] G. U. Meduri, S. Headley, G. Kohler et al., "Persistent elevation of inflammatory cytokines predicts a poor outcome in ARDS. Plasma IL-1 beta and IL-6 levels are consistent and efficient predictors of outcome over time," *Chest*, vol. 107, no. 4, pp. 1062–1073, 1995.
- [38] J. Li, Y. Wu, S. Zhang et al., "Baicalein protect pancreatic injury in rats with severe acute pancreatitis by inhibiting pro-inflammatory cytokines expression," *Biochemical and Biophysical Research Communications*, vol. 466, no. 4, pp. 664–669, 2015.
- [39] R. Hoque, M. Sohail, A. Malik et al., "TLR9 and the NLRP3 inflammasome link acinar cell death with inflammation in acute pancreatitis," *Gastroenterology*, vol. 141, no. 1, pp. 358–369, 2011.
- [40] C. Zoja, P. B. Garcia, and G. Remuzzi, "The role of chemokines in progressive renal disease," *Frontiers Bioscience*, vol. 14, pp. 1815–1822, 2009.
- [41] K. Turkmen, H. Z. Tonbul, A. Toker et al., "The relationship between oxidative stress, inflammation, and atherosclerosis in renal transplant and end-stage renal disease patients," *Renal Failure*, vol. 34, no. 10, pp. 1229–1237, 2012.
- [42] S. Granata, V. Masola, E. Zoratti et al., "NLRP3 inflammasome activation in dialyzed chronic kidney disease patients," *PLoS One*, vol. 10, no. 3, article e0122272, 2015.
- [43] B. J. Currie and S. P. Jacups, "Intensity of rainfall and severity of melioidosis, Australia," *Emerging Infectious Diseases*, vol. 9, no. 12, pp. 1538–1542, 2003.
- [44] B. J. Currie, D. A. Fisher, D. M. Howard et al., "Endemic melioidosis in tropical northern Australia: a 10-year prospective study and review of the literature," *Clinical Infectious Diseases*, vol. 31, no. 4, pp. 981–986, 2000.
- [45] S. Pongcharoen, P. N. Ritvirool, D. Sanguanserm Sri et al., "Reduced interleukin-17 expression of Burkholderia pseudomallei-infected peripheral blood mononuclear cells of diabetic patients," *Asian Pacific Journal of Allergy and Immunology*, vol. 26, no. 1, pp. 63–69, 2008.
- [46] C. Y. Chin, D. M. Monack, and S. Nathan, "Delayed activation of host innate immune pathways in streptozotocin-induced diabetic hosts leads to more severe disease during infection with Burkholderia pseudomallei," *Immunology*, vol. 135, no. 4, pp. 312–332, 2012.
- [47] K. A. Hodgson, J. L. Morris, M. L. Feterl, B. L. Govan, and N. Ketheesan, "Altered macrophage function is associated with severe Burkholderia pseudomallei infection in a murine model of type 2 diabetes," *Microbes and Infection*, vol. 13, no. 14–15, pp. 1177–1184, 2011.
- [48] R. C. Bone, W. J. Sibbald, and C. L. Sprung, "The ACCP-SCCM consensus conference on sepsis and organ failure," *Chest*, vol. 101, no. 6, pp. 1481–1483, 1992.
- [49] M. Shankar-Hari, G. S. Phillips, M. L. Levy et al., "Developing a new definition and assessing new clinical criteria for septic shock: for the Third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3)," *Jama*, vol. 315, no. 8, pp. 775–787, 2016.
- [50] R. Sordi, D. Fernandes, B. T. Heckert, and J. Assreuy, "Early potassium channel blockade improves sepsis-induced organ damage and cardiovascular dysfunction," *British Journal of Pharmacology*, vol. 163, no. 6, pp. 1289–1301, 2011.
- [51] S. Warrillow, M. Egi, and R. Bellomo, "Randomized, double-blind, placebo-controlled crossover pilot study of a potassium channel blocker in patients with septic shock," *Critical Care Medicine*, vol. 34, no. 4, pp. 980–985, 2006.
- [52] A. Morelli, M. Lange, C. Ertmer et al., "Glibenclamide dose response in patients with septic shock: effects on noradrenaline requirements, cardiopulmonary performance, and global oxygen transport," *Shock*, vol. 28, no. 5, pp. 530–535, 2007.
- [53] J. F. Buckley, M. Singer, and L. H. Clapp, "Role of KATP channels in sepsis," *Cardiovascular Research*, vol. 72, no. 2, pp. 220–230, 2006.
- [54] J. A. Oliver and D. W. Landry, "Potassium channels and septic shock," *Critical Care Medicine*, vol. 34, no. 4, pp. 1255–1257, 2006.
- [55] W. Shi, N. Cui, Z. Wu et al., "Lipopolysaccharides up-regulate Kir6.1/SUR2B channel expression and enhance vascular KATP channel activity via NF-kappaB-dependent signaling," *The Journal of Biological Chemistry*, vol. 285, no. 5, pp. 3021–3029, 2010.
- [56] D. Schmid, M. Svoboda, A. Sorgner et al., "Glibenclamide reduces proinflammatory cytokines in an ex vivo model of human endotoxaemia under hypoxaemic conditions," *Life Sciences*, vol. 89, no. 19–20, pp. 725–734, 2011.

- [57] K. Schroder, R. Zhou, and J. Tschopp, "The NLRP3 inflammasome: a sensor for metabolic danger?" *Science*, vol. 327, no. 5963, pp. 296–300, 2010.
- [58] M. Lamkanfi, J. L. Mueller, A. C. Vitari et al., "Glyburide inhibits the Cryopyrin/Nalp3 inflammasome," *The Journal of Cell Biology*, vol. 187, no. 1, pp. 61–70, 2009.
- [59] J. Cai, S. Lu, Z. Yao et al., "Glibenclamide attenuates myocardial injury by lipopolysaccharides in streptozotocin-induced diabetic mice," *Cardiovascular Diabetology*, vol. 13, no. 1, p. 106, 2014.
- [60] D. B. Kurland, V. Gerzanich, J. K. Karimy et al., "The Sur1-Trpm4 channel regulates NOS2 transcription in TLR4-activated microglia," *Journal of Neuroinflammation*, vol. 13, no. 1, p. 130, 2016.
- [61] F. J. Ortega, J. Gimeno-Bayon, J. F. Espinosa-Parrilla et al., "ATP-dependent potassium channel blockade strengthens microglial neuroprotection after hypoxia-ischemia in rats," *Experimental Neurology*, vol. 235, no. 1, pp. 282–296, 2012.
- [62] F. J. Ortega, J. Jolkkonen, N. Mahy, and M. J. Rodriguez, "Glibenclamide enhances neurogenesis and improves long-term functional recovery after transient focal cerebral ischemia," *Journal of Cerebral Blood Flow and Metabolism*, vol. 33, no. 3, pp. 356–364, 2013.
- [63] X. S. Liu, Z. G. Zhang, R. L. Zhang et al., "Chemokine ligand 2 (CCL2) induces migration and differentiation of subventricular zone cells after stroke," *Journal of Neuroscience Research*, vol. 85, no. 10, pp. 2120–2125, 2007.
- [64] A. Turbic, S. Y. Leong, and A. M. Turnley, "Chemokines and inflammatory mediators interact to regulate adult murine neural precursor cell proliferation, survival and differentiation," *PLoS One*, vol. 6, no. 9, p. e25406, 2011.
- [65] R. Dutta and B. D. Trapp, "Mechanisms of neuronal dysfunction and degeneration in multiple sclerosis," *Progress in Neurobiology*, vol. 93, no. 1, pp. 1–12, 2011.
- [66] International Multiple Sclerosis Genetics Consortium, Wellcome Trust Case Control Consortium 2, S. Sawcer et al., "Genetic risk and a primary role for cell-mediated immune mechanisms in multiple sclerosis," *Nature*, vol. 476, no. 7359, pp. 214–219, 2011.
- [67] J. I. Suarez, R. W. Tarr, and W. R. Selman, "Aneurysmal subarachnoid hemorrhage," *The New England Journal of Medicine*, vol. 354, no. 4, pp. 387–396, 2006.
- [68] M. Rahgozar, D. A. Willgoss, G. C. Gobe, and Z. H. Endre, "ATP-dependent K⁺ channels in renal ischemia reperfusion injury," *Renal Failure*, vol. 25, no. 6, pp. 885–896, 2003.
- [69] B. C. Perry, D. Soltys, A. H. Toledo, and L. H. Toledo-Pereyra, "Tumor necrosis factor-alpha in liver ischemia/reperfusion injury," *Journal of Investigative Surgery*, vol. 24, no. 4, pp. 178–188, 2011.
- [70] X. L. Sun and G. Hu, "ATP-sensitive potassium channels: a promising target for protecting neurovascular unit function in stroke," *Clinical and Experimental Pharmacology & Physiology*, vol. 37, no. 2, pp. 243–252, 2010.
- [71] J. T. Willerson, "Pharmacologic approaches to reperfusion injury," *Advances in Pharmacology*, vol. 39, pp. 291–312, 1997.
- [72] J. M. Simard, V. Yurovsky, N. Tsybalyuk, L. Melnichenko, S. Ivanova, and V. Gerzanich, "Protective effect of delayed treatment with low-dose glibenclamide in three models of ischemic stroke," *Stroke*, vol. 40, no. 2, pp. 604–609, 2009.
- [73] Y. J. Zhang, A. Q. Zhang, X. X. Zhao, Z. L. Tian, and L. Yao, "Nicorandil protects against ischaemia-reperfusion injury in newborn rat kidney," *Pharmacology*, vol. 92, no. 5–6, pp. 245–256, 2013.
- [74] F. Sgard, C. Faure, C. Drieu la Rochelle et al., "Regulation of ATP-sensitive potassium channel mRNA expression in rat kidney following ischemic injury," *Biochemical and Biophysical Research Communications*, vol. 269, no. 2, pp. 618–622, 2000.
- [75] J. Cheng, L. Mao, J. Wen et al., "Different effects of hypertension and age on the function of large conductance calcium- and voltage-activated potassium channels in human mesentery artery smooth muscle cells," *Journal of American Heart Association*, vol. 5, no. 9, article e003913, 2016.
- [76] R. M. Heindel, R. E. Pakyz, L. N. Reinking, and M. J. Cosentino, "The effect of various degrees of unilateral spermatic cord torsion on fertility in the rat," *The Journal of Urology*, vol. 144, no. 2 Part 1, pp. 366–369, 1990.
- [77] M. Al-Maghrebi, E. O. Kehinde, and J. T. Anim, "Long term testicular ischemia-reperfusion injury-induced apoptosis: involvement of survivin down-regulation," *Biochemical and Biophysical Research Communications*, vol. 395, no. 3, pp. 342–347, 2010.
- [78] A. J. Kowaltowski, S. Seetharaman, P. Pauczek, and K. D. Garlid, "Bioenergetic consequences of opening the ATP-sensitive K⁽⁺⁾ channel of heart mitochondria," *The American Journal of Physiology - Heart and Circulatory Physiology*, vol. 280, no. 2, pp. H649–H657, 2001.
- [79] M. L. Zhuo, Y. Huang, D. P. Liu, and C. C. Liang, "KATP channel: relation with cell metabolism and role in the cardiovascular system," *The International Journal of Biochemistry & Cell Biology*, vol. 37, no. 4, pp. 751–764, 2005.
- [80] F. Martinon, K. Burns, and J. Tschopp, "The inflammasome: a molecular platform triggering activation of inflammatory caspases and processing of proIL-beta," *Molecular Cell*, vol. 10, no. 2, pp. 417–426, 2002.
- [81] F. Martinon, V. Petrilli, A. Mayor, A. Tardivel, and J. Tschopp, "Gout-associated uric acid crystals activate the NALP3 inflammasome," *Nature*, vol. 440, no. 7081, pp. 237–241, 2006.
- [82] S. L. Cassel, S. C. Eisenbarth, S. S. Iyer et al., "The Nalp3 inflammasome is essential for the development of silicosis," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 26, pp. 9035–9040, 2008.
- [83] C. Dostert, V. Petrilli, R. Van Bruggen, C. Steele, B. T. Mossman, and J. Tschopp, "Innate immune activation through Nalp3 inflammasome sensing of asbestos and silica," *Science*, vol. 320, no. 5876, pp. 674–677, 2008.
- [84] V. Hornung, F. Bauernfeind, A. Halle et al., "Silica crystals and aluminum salts activate the NALP3 inflammasome through phagosomal destabilization," *Nature Immunology*, vol. 9, no. 8, pp. 847–856, 2008.
- [85] R. Munoz-Planillo, P. Kuffa, G. Martinez-Colon, B. L. Smith, T. M. Rajendiran, and G. Nunez, "K⁽⁺⁾ efflux is the common trigger of NLRP3 inflammasome activation by bacterial toxins and particulate matter," *Immunity*, vol. 38, no. 6, pp. 1142–1153, 2013.
- [86] V. Petrilli, S. Papin, C. Dostert, A. Mayor, F. Martinon, and J. Tschopp, "Activation of the NALP3 inflammasome is triggered by low intracellular potassium concentration," *Cell Death Differentiation*, vol. 14, no. 9, pp. 1583–1589, 2007.

- [87] Y. Hamon, M. F. Luciani, F. Becq, B. Verrier, A. Rubartelli, and G. Chimini, "Interleukin-1beta secretion is impaired by inhibitors of the Atp binding cassette transporter, ABC1," *Blood*, vol. 90, no. 8, pp. 2911–2915, 1997.
- [88] D. M. Brown, K. Donaldson, P. J. Borm et al., "Calcium and ROS-mediated activation of transcription factors and TNF-alpha cytokine gene expression in macrophages exposed to ultrafine particles," *American Journal of Physiology. Lung Cellular and Molecular Physiology*, vol. 286, no. 2, pp. L344–L353, 2004.
- [89] C. B. Grahames, A. D. Michel, I. P. Chessell, and P. P. Humphrey, "Pharmacological characterization of ATP- and LPS-induced IL-1beta release in human monocytes," *British Journal of Pharmacology*, vol. 127, no. 8, pp. 1915–1921, 1999.
- [90] R. A. North, "Molecular physiology of P2X receptors," *Physiological Reviews*, vol. 82, no. 4, pp. 1013–1067, 2002.
- [91] D. Schmid, D. L. Staudacher, R. Bueno, P. G. Spieckermann, and T. Moeslinger, "ATP-sensitive potassium channels expressed by human monocytes play a role in stasis-induced thrombogenesis via tissue factor pathway," *Life Sciences*, vol. 80, no. 11, pp. 989–998, 2007.
- [92] K. R. Wagner, C. Dean, S. Beiler et al., "Plasma infusions into porcine cerebral white matter induce early edema, oxidative stress, pro-inflammatory cytokine gene expression and DNA fragmentation: implications for white matter injury with increased blood-brain-barrier permeability," *Current Neurovascular Research*, vol. 2, no. 2, pp. 149–155, 2005.
- [93] E. R. Chilvers, K. A. Cadwallader, B. J. Reed, J. F. White, and A. M. Condliffe, "The function and fate of neutrophils at the inflamed site: prospects for therapeutic intervention," *Journal of the Royal College of Physicians of London*, vol. 34, no. 1, pp. 68–74, 2000.
- [94] L. Yang, R. M. Froio, T. E. Sciuto, A. M. Dvorak, R. Alon, and F.W. Lusinskas, "ICAM-1 regulates neutrophil adhesion and transcellular migration of TNF-alpha-activated vascular endothelium under flow," *Blood*, vol. 106, no. 2, pp. 584–592, 2005.
- [95] B. Pugin, B. Ricou, K. P. Steinberg, P. M. Suter, and T. R. Martin, "Proinflammatory activity in bronchoalveolar lavage fluids from patients with ARDS, a prominent role for interleukin-1," *American Journal of Respiratory and Critical Care Medicine*, vol. 153, no. 6, Part 1, pp. 1850–1856, 1996.
- [96] J. A. Leff, J. W. Baer, M. E. Bodman et al., "Interleukin-1-induced lung neutrophil accumulation and oxygen metabolite-mediated lung leak in rats," *The American Journal of Physiology*, vol. 266, no. 1, Part 1, pp. L2–L8, 1994.
- [97] D. Voehringer, K. Shinkai, and R. M. Locksley, "Type 2 immunity reflects orchestrated recruitment of cells committed to IL-4 production," *Immunity*, vol. 20, no. 3, pp. 267–277, 2004.
- [98] H. Yusuf-Makagiansar, M. E. Anderson, T. V. Yakovleva, J. S. Murray, and T. J. Siahaan, "Inhibition of LFA-1/ICAM-1 and VLA-4/VCAM-1 as a therapeutic approach to inflammation and autoimmune diseases," *Medicinal Research Reviews*, vol. 22, no. 2, pp. 146–167, 2002.
- [99] A. Weidinger and A. V. Kozlov, "Biological activities of reactive oxygen and nitrogen species: oxidative stress versus signal transduction," *Biomolecules*, vol. 5, no. 2, pp. 472–484, 2015.
- [100] C. Thiemermann, "Nitric oxide and septic shock," *General Pharmacology*, vol. 29, no. 2, pp. 159–166, 1997.
- [101] P. P. Wolkow, "Involvement and dual effects of nitric oxide in septic shock," *Inflammation Research*, vol. 47, no. 4, pp. 152–166, 1998.
- [102] J. B. Ochoa, A. O. Udekwo, T. R. Billiar et al., "Nitrogen oxide levels in patients after trauma and during sepsis," *Annals of Surgery*, vol. 214, no. 5, pp. 621–626, 1991.
- [103] C. C. Wu, C. Thiemermann, and J. R. Vane, "Glibenclamide-induced inhibition of the expression of inducible nitric oxide synthase in cultured macrophages and in the anaesthetized rat," *British Journal of Pharmacology*, vol. 114, no. 6, pp. 1273–1281, 1995.
- [104] S. Lazarov, M. Balutsov, and E. Ianev, "The role of bacterial endotoxins, receptors and cytokines in the pathogenesis of septic (endotoxin) shock," *Vutreshni Bolesti*, vol. 32, no. 4, pp. 33–40, 2000.
- [105] K. Nagamoto-Combs and C. K. Combs, "Microglial phenotype is regulated by activity of the transcription factor, NFAT (nuclear factor of activated T cells)," *The Journal of Neuroscience*, vol. 30, no. 28, pp. 9641–9646, 2010.
- [106] P. G. Hogan, L. Chen, J. Nardone, and A. Rao, "Transcriptional regulation by calcium, calcineurin, and NFAT," *Genes & Development*, vol. 17, no. 18, pp. 2205–2232, 2003.
- [107] R. J. Jones, D. Jourdeheuil, J. C. Salerno, S. M. Smith, and H. A. Singer, "iNOS regulation by calcium/calmodulin-dependent protein kinase II in vascular smooth muscle," *The American Journal of Physiology - Heart and Circulatory Physiology*, vol. 292, no. 6, pp. H2634–H2642, 2007.
- [108] S. M. MacDonnell, J. Weisser-Thomas, H. Kubo et al., "CaMKII negatively regulates calcineurin-NFAT signaling in cardiac myocytes," *Circulation Research*, vol. 105, no. 4, pp. 316–325, 2009.
- [109] N. K. Nazaroglu, A. Sepici-Dincel, and N. Altan, "The effects of sulfonylurea glyburide on superoxide dismutase, catalase, and glutathione peroxidase activities in the brain tissue of streptozotocin-induced diabetic rat," *Journal of Diabetes and Its Complications*, vol. 23, no. 3, pp. 209–213, 2009.
- [110] F. Xiu, M. Stanojic, and M. G. Jeschke, "Norepinephrine inhibits macrophage migration by decreasing CCR2 expression," *PloS One*, vol. 8, no. 7, article e69167, 2013.
- [111] J. Deng, K. Muthu, R. Gamelli, R. Shankar, and S. B. Jones, "Adrenergic modulation of splenic macrophage cytokine release in polymicrobial sepsis," *American Journal of Physiology. Cell Physiology*, vol. 287, no. 3, pp. C730–C736, 2004.