

Neuroprotection in Acute Ischemic Stroke: Pathological Mechanisms and Reflections on Translation from Bench to Bedside

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Abstract

Acute ischemic stroke is a leading cause of death and disability worldwide. Interruption of cerebral perfusion caused by arterial occlusion triggers a cascade of ischemic brain injury, imposing a substantial burden on public health systems. Intravenous thrombolysis and mechanical thrombectomy are currently the evidence-based reperfusion therapies; however, owing to constraints such as a narrow therapeutic time window, hemorrhagic transformation, and ischemia-reperfusion injury, many patients still suffer severe neurological deficits. Post-ischemic excitotoxicity, neuroinflammation, and oxidative stress are intertwined and collectively drive multiple forms of programmed cell death—including apoptosis, necroptosis, pyroptosis, and ferroptosis—thereby mediating progressive neuronal loss in the ischemic penumbra. Deciphering this pathological network is a core prerequisite for developing neuroprotective strategies. Although numerous preclinical studies have demonstrated that drugs, nanodelivery systems, stem cells/exosomes, and physical interventions targeting these pathways can exert neuroprotective effects, the vast majority of these strategies have not yet achieved clear, independent benefit in human clinical trials. With the widespread adoption of endovascular reperfusion therapy, neuroprotection is regarded as an important breakthrough in the management of acute ischemic stroke. This review systematically summarizes the key pathophysiological processes and various programmed cell death mechanisms after acute ischemic stroke, outlines the current status of interventions targeting different targets, and analyzes the practical obstacles to bench-to-bedside translation, aiming to provide a reference for the development of novel neuroprotective strategies.

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Keywords

Acute Ischemic Stroke, Neuroprotection, Programmed Cell Death, Excitotoxicity, Neuroinflammation, Oxidative Stress, Ischemia-Reperfusion Injury

1. Introduction

Acute ischaemic stroke arises from abrupt cerebral arterial occlusion and subsequent perfusion failure, ranking among the leading global sources of mortality and persistent long-term disability. Epidemiological data between 1990 and 2021 reflect a growing worldwide stroke burden: stroke incidence rose by 70.0%, mortality climbed by 44.0%, prevalent cases expanded by 86.0%, and disability-adjusted life-year metrics increased by 32% across this period [1]. Currently, the acute-phase treatments proven effective by evidence-based medicine mainly include intravenous thrombolysis and mechanical thrombectomy [2]. Intravenous thrombolysis, represented by alteplase and tenecteplase, is only applicable to patients within 4.5 h of symptom onset; even among those who receive thrombolysis, only about half achieve a favorable functional recovery [3]-[6]. Mechanical thrombectomy has become the standard treatment for ischemic stroke caused by large-vessel occlusion; however, reperfusion itself can induce secondary ischemia-reperfusion injury, which continues to limit overall patient prognosis. Neuronal injury in the infarct core is mostly irreversible, whereas progressive neuronal death within the ischemic penumbra is the key determinant of final infarct volume and neurological outcome. Excitotoxicity, oxidative stress, and neuroinflammation are relatively independent yet extensively interconnected, and can sequentially or concurrently activate multiple cell death modalities, including apoptosis, ferroptosis, pyroptosis, and necroptosis. Over the past decades, numerous preclinical studies have explored neuroprotection targeting these pathological processes; however, positive results from most animal experiments have been difficult to replicate in clinical trials, and there is still no generally accepted neuroprotective drug that can be used independently in clinical practice. As the reperfusion treatment system matures, combining neuroprotection with reperfusion has become an important research direction. This review focuses on neuroprotection in acute ischemic stroke, summarizing the pathophysiology of ischemic injury and the mechanisms of programmed cell death, reviewing advances in interventions targeting excitotoxicity, neuroinflammation, oxidative stress, and various cell death pathways, and analyzing the problems of existing strategies in clinical translation, so as to provide insights for the development of novel neuroprotective approaches.

Search strategy and study selection. This review was designed to systematically summarize the current research progress in neuroprotection for acute ischemic stroke. We searched the following electronic databases: PubMed, Embase,

Web of Science, and the Cochrane Library, from inception to March 2025. The search strategy combined database-specific subject headings and free-text keywords covering the following concepts: acute ischemic stroke/cerebral ischemia/cerebral ischemia-reperfusion injury; neuroprotection; excitotoxicity; neuroinflammation; oxidative stress; and programmed cell death (apoptosis, necroptosis, pyroptosis, and ferroptosis). Reference lists of relevant articles were also screened manually to identify additional eligible studies. Studies were included if they reported original clinical or experimental data on the pathophysiology of ischemic injury or on interventions with potential neuroprotective effects; conference abstracts, case reports, and studies unrelated to ischemic stroke were excluded, whereas review articles and guidelines were used only for background and conceptual framing. Titles and abstracts were screened independently by two reviewers, and full texts were assessed against the inclusion criteria, with disagreements resolved by discussion or by consultation with a third reviewer. In line with the systematic scope of this review, the evidence supporting each intervention was explicitly graded according to the study type from which the reported findings were derived, *i.e.*, clinical evidence (interventional studies in patients), human observational evidence (observational or epidemiological studies in humans), animal experimental evidence (*in vivo* studies using animal models of ischemic stroke), or *in vitro* experimental evidence (studies using cultured cells or isolated tissues). This grading is applied consistently in Section 2 to ensure that preclinical mechanistic findings are not misconstrued as clinically established therapeutic options.

2. Pathophysiological Mechanisms of Acute Ischemic Stroke

2.1. Excitotoxicity

The high demand of brain tissue for energy substrates determines its extreme sensitivity to interruption of blood perfusion. Maintenance of normal brain function depends on continuous blood circulation to deliver oxygen and nutrients; once a major cerebral artery becomes occluded, the supplied region enters a state of ischemia and hypoxia [7]. Insufficient perfusion disrupts oxidative phosphorylation, leading to impaired adenosine triphosphate (ATP) synthesis and rapid depletion of neuronal energy reserves. The loss of cellular energy directly impairs the function of critical ion pumps, such as Na^+/K^+ -ATPase and Ca^{2+} -ATPase. This failure disrupts the essential ion gradients across neuronal membranes, which in turn leads to widespread membrane depolarization [8]. In the early stage of ischemic injury, ion homeostasis imbalance and abnormal release of excitatory amino acids form a pathological cycle of mutual causation. Energy depletion causes failure of glutamate reuptake mechanisms, while membrane depolarization promotes massive glutamate release from presynaptic terminals, leading to abnormal accumulation of glutamate in the synaptic cleft and the extracellular space of neurons [9] [10].

Persistently raised glutamate concentrations within the extracellular space trig-

ger excessive activation of postsynaptic N-methyl-D-aspartate (NMDA) and α -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors. Overstimulated AMPA receptors chiefly drive robust sodium and chloride ion inflow, which raises intracellular osmotic pressure. Subsequent water influx into neuronal compartments gives rise to cytotoxic oedema, neuronal swelling and eventual cell rupture; these events constitute a major contributor to oedema formation in the acute phase of cerebral ischaemia [11] [12]. Beyond triggering neuronal necrosis and apoptotic cell death, excessive NMDA receptor activity disrupts physiological synaptic plasticity. Such dysregulation impairs cognitive capacities including learning and memory, representing one critical pathological driver for cognitive impairment following ischaemic stroke [13]. Furthermore, pore opening of NMDA receptor complexes permits substantial calcium entry into cytosol, producing the pathological state known as intracellular calcium overload [14]. Calcium overload is a key hub linking excitotoxicity to downstream neuronal death. Elevated cytosolic calcium concentration not only results from extracellular influx but can also trigger calcium release from intracellular stores such as mitochondria and the endoplasmic reticulum, further exacerbating calcium homeostasis dysregulation and forming a positive feedback loop, ultimately leading to severe impairment of mitochondrial structure and function [15]. Damaged mitochondria exhibit swelling, increased membrane permeability, and disintegration of membrane structure, thereby activating apoptotic pathways and inducing oxidative stress. Concurrently, the generation of large amounts of reactive oxygen species further aggravates lipid peroxidation and protein damage, forming a vicious cycle of “calcium overload-mitochondrial dysfunction-oxidative stress” [10] [16] [17].

2.2. Inflammatory Response

Current evidence indicates that the inflammatory response after acute ischemic stroke can be broadly divided into three phases [18]: the acute phase, occurring in the first few hours after stroke onset, during which microglia and macrophages clear necrotic cells and the first infiltration of immune cells, predominantly neutrophils, is observed; the subacute phase, occurring over the following days, during which the inflammatory response tends to resolve; and the late phase, during which inflammatory cells participate in astrocyte- and microglia-mediated repair processes. This temporal feature suggests that different immune cell subsets perform distinct functions during different time windows of injury and repair, providing a pathophysiological basis for neuroprotective interventions targeting inflammatory processes.

2.2.1. Microglia

Microglia are the predominant immune cells of the central nervous system and constitute the innate immune component that responds rapidly after ischemic injury [19]. Cerebral ischemia triggers the activation of microglia, which then proceed through a coordinated series of responses encompassing cytokine secretion, chemotaxis, phagocytosis, and proliferation. Once activated, microglia migrate to

ward the ischemic lesion, where they engulf and clear dead cells while simultaneously releasing neurotrophic factors that support neuronal survival [20]. Conversely, microglia also produce inflammatory cytokines and cytotoxic mediators, a response that may worsen the extent of ischemic injury [21]-[25]. In response to the dynamically changing post-ischemic microenvironment, microglia alter their phenotypes in response to changes in cerebral homeostasis, and each stimulus induces a unique reactive state. Although microglial activation was initially classified into two functional states—the classically activated M1 phenotype (pro-inflammatory and neurotoxic) and the alternatively activated M2 phenotype (tissue-reparative and immunosuppressive)—these labels are now recognized as simplified functional models rather than fixed cellular identities [26] [27]. Post-ischemic microglia instead exist in dynamic and overlapping activation states, the specific profile of which is determined by the time elapsed after stroke onset, the spatial location within the lesion, and the prevailing local signals. Under pathological conditions, pro-inflammatory polarization predominates during the acute inflammatory response and represents a key effector mechanism in early neuronal injury after ischemic stroke [28].

2.2.2. Astrocytes

Astrocytes exhibit dynamic and bidirectional responses in ischemic stroke. In the early stage of injury, they participate in blood-brain barrier (BBB) dysfunction and edema formation by releasing pro-inflammatory factors and activating matrix metalloproteinases (MMPs); subsequently, they shift toward a neuroprotective phenotype, supporting tissue recovery by clearing glutamate, promoting BBB repair, and secreting neurotrophic factors [29] [30]. After ischemic stroke, reactive astrogliosis can occur through morphological changes and upregulation of GFAP expression [31]. In animal experiments, LCN2 expression in astrocytes was significantly upregulated 24 h after stroke onset, and LCN2/24p3R mediated astrocyte pyroptosis after cerebral ischemia/reperfusion injury by activating the NLRP3 inflammasome [32].

2.2.3. Oligodendrocytes

Oligodendrocytes are indispensable for synthesizing and maintaining myelin in the central nervous system, and they therefore support the rapid, accurate conduction of neural signals. They also help stabilize the CNS microenvironment, participating in immune regulation, metabolic support, and neuroprotection [33]. Cerebral ischemic injury can trigger glutamate-mediated excitotoxicity in oligodendrocyte cells, which in turn induces excessive intracellular calcium accumulation and severely impairs mitochondrial function [34]. The disruption of intact myelin structures further promotes the release of antigenic components, with myelin basic protein (MBP) as a typical representative. Such substances can effectively activate microglia and recruit peripheral T cell infiltration in local lesions [35]. Additionally, ischemic stimulation significantly elevates the surface expression level of MHC class II molecules in oligodendrocytes. The acquired antigen-

presenting ability of these cells further intensifies neuroinflammatory responses in the central nervous system, thereby exacerbating secondary neuronal damage following ischemic stroke [36].

2.2.4. Neutrophils

Neutrophils are the first immune cells to accumulate at the ischemic lesion and to cross into the brain parenchyma after injury. Once recruited, they secrete elastase, which increases vascular permeability and allows plasma proteins and inflammatory mediators to leak into the cerebral interstitium [37]. Through NADPH oxidase, infiltrating neutrophils generate reactive oxygen species (ROS) and nitric oxide (NO), which damage lipids and DNA in neurons and glial cells [38]. Neutrophils can also release neutrophil extracellular traps (NETs)—complexes of histones, antimicrobial peptides, and proteases—that directly harm perilesional tissue and amplify local inflammation [39]. NET formation is tightly controlled by peptidylarginine deiminase 4 (PAD4); when PAD4 is activated, it accelerates the release of citrullinated histones, further strengthening inflammatory signaling in the ischemic brain [40].

2.2.5. Monocytes

Following ischemic stroke onset, circulating peripheral monocytes are recruited to injured cerebral tissues mainly through the CCL2/CCR2 chemokine signaling cascade [41]. Based on unique surface biomarker characteristics, monocytes are classified into three distinct subpopulations: classical (CD14⁺⁺CD16[−]), intermediate (CD14⁺⁺CD16⁺), and non-classical (CD14⁺CD16⁺⁺). The classical monocyte subset exhibits abundant CCR2 expression and robust chemotactic reactivity toward monocyte chemoattractant protein-1 (MCP-1) secreted in ischemic damaged areas [42]. After infiltrating the brain parenchyma, circulating monocytes differentiate into mature macrophages under the modulation of the local cerebral microenvironment. Macrophage activation has conventionally been classified into pro-inflammatory M1 and anti-inflammatory M2 phenotypes, induced respectively by stimuli such as IFN- γ and TNF- α , or by IL-4, IL-10, TGF- β and macrophage colony-stimulating factor (M-CSF); however, these labels represent simplified functional models rather than fixed cellular identities. Like other myeloid cells after stroke, macrophages exist in dynamic and overlapping activation states, the specific profile of which is determined by the time after stroke onset, the spatial location within the lesion, and the prevailing local signals. Pro-inflammatory phenotypes release inflammatory cytokines and nitric oxide (NO) and thereby exert antimicrobial effects, whereas anti-inflammatory phenotypes facilitate tissue restoration by producing extracellular matrix components and pro-angiogenic factors.

2.2.6. T Lymphocytes

T lymphocytes show complex, time-dependent behavior and dual functions during ischemic stroke [43]. Infiltration of CD4⁺ and CD8⁺ T cells into damaged tissue increases significantly after stroke, with corresponding shifts in the propor-

tions of T-cell subsets [44]. By day 14, although myeloid infiltration such as neutrophils has declined markedly, CD4+ and CD8+ T cells remain in the central nervous system; this persistence into the chronic phase may be linked to ongoing brain injury and secondary neurodegeneration [45].

2.3. Oxidative Stress

Obstruction of cerebral blood flow immediately disrupts energy supply to brain tissue, causing energy metabolic disturbance; subsequent vessel recanalization and reperfusion have also been confirmed as important triggers of oxidative stress-related injury. In fact, both ischemia itself and the more pronounced reperfusion process are accompanied by substantial generation of reactive oxygen species (ROS). Mitochondria have long been regarded as the main source of ROS during ischemia and play a key role in ROS production during secondary reperfusion injury. Under ischemic conditions, mitochondrial swelling and disintegration caused by mitochondrial dysfunction involve both initiation of pro-apoptotic cascades and release of free radicals, making mitochondria an important hub for amplification of injury signals [46]. Recent studies further show that under conditions of ischemia and NMDA receptor activation, the amount of superoxide anion produced by NADPH oxidase far exceeds that produced by mitochondria, suggesting that this enzyme may play a dominant role in ROS generation during ischemia [47]. When the production of ROS outruns the capacity of endogenous antioxidant defenses, the balance between oxidants and antioxidants collapses and oxidative damage spreads through the tissue.

Brain tissue is particularly vulnerable to this imbalance because its cells are rich in polyunsaturated fatty acids and are highly susceptible to oxidative attack; in addition, low levels of antioxidants, abundant pro-oxidants such as iron, and active oxidative metabolism in the brain collectively further amplify the intensity and extent of oxidative damage [48]. At the same time, the lack of glucose and oxygen during ischemia further increases intracellular ROS production, creating a vicious cycle in which oxidative stress and brain injury mutually aggravate each other [49]. Disruption of calcium homeostasis is an important initiating step in post-ischemic ROS generation. Acute ischemic stroke disrupts intracellular calcium homeostasis, prompting massive release of calcium ions in the brain and thereby activating signaling pathways that culminate in ROS production and oxidative damage [13]. Significantly elevated intracellular calcium concentration can activate calcium-dependent enzymes such as nitric oxide synthase (NOS), catalyzing the formation of highly reactive species such as peroxynitrite (ONOO⁻), thereby directly causing cellular injury [50].

2.4. Brain Cell Death

Neuronal death after ischemia is a complex cascade process in which multiple modes of programmed cell death, including apoptosis, necroptosis, ferroptosis, and pyroptosis, can be activated sequentially or concurrently. Extensive signaling

crosstalk exists among different cell death modalities, collectively mediating neuronal injury, blood-brain barrier disruption, and neuroinflammatory responses, promoting infarct expansion, and ultimately affecting disease prognosis [51].

2.4.1. Apoptosis

Apoptosis is the most common form of programmed cell death in multicellular organisms and a major route of neuronal loss after cerebral infarction. It can be engaged through either an intrinsic or an extrinsic pathway: the intrinsic arm is triggered by DNA damage or endoplasmic-reticulum stress, whereas the extrinsic arm depends on activation of death-receptor family members [52]. Intrinsic pathway: The intrinsic apoptotic pathway is a receptor-independent intracellular signaling cascade and an important regulatory mode of neuronal apoptosis in ischemic stroke, being abnormally activated mainly by intracellular stress induced by ischemia [53]. Hypoxia, ROS accumulation, and DNA damage caused by cerebral ischemia can all initiate this apoptotic program [54]. Ischemia can induce glutamate excitotoxicity, leading to intracellular Ca^{2+} overload and calcium homeostasis disturbance through activation of neuronal NMDARs [55]. Elevated calcium ions further activate calpain, mediating cleavage of BH3-interacting domain (BID) into its active form tBID [56]. tBID destabilizes the mitochondrial membrane and increases its permeability, promoting the release of cytochrome c, AIF, endonuclease G, and other mitochondrial components [57]. Cytosolic cytochrome c binds to Apaf-1 and activates caspase-9, further upregulating downstream caspase-3 activity, ultimately triggering neuronal DNA degradation and apoptosis [58] [59].

Extrinsic pathway: During inflammation, activated immune cells release multiple factors, including pro-inflammatory cytokines, that can drive neuronal death through this route [60] [61]. The decisive step is the binding of ligands to “death receptors” on the cell surface [62]. The relevant ligands include tumor necrosis factor- α (TNF- α), TNF-related apoptosis-inducing ligand (TRAIL), and FasL, while the receptors comprise apoptosis-related molecules (Fas/CD95), tumor necrosis factor receptor 1 (TNFR1), and TRAIL receptors (DR4 and DR5) [63] [64]. For example, under external stimuli, the extrinsic apoptotic pathway is triggered by ligand binding to death receptors of the tumor necrosis factor (TNF) family on the cell surface [62]. After receptor binding, the adaptor protein Fas-associated death domain protein (FADD) is recruited. FADD subsequently recruits and forms caspase-8 homodimers, triggering a caspase cascade that ultimately leads to increased mitochondrial membrane permeability, DNA fragmentation, and apoptosis in neuronal cells [65].

2.4.2. Necroptosis

Necroptosis represents one lytic subclass of programmed cell death with strong pro-inflammatory properties. Membrane disintegration during this process releases damage-associated molecular patterns (DAMPs) and provokes pronounced neuroinflammatory responses. This cell-death mechanism exerts notable biologi-

cal effects within the infarct core as well as ATP-deficient regions of the ischaemic penumbra [66]. Multiple death-sensing receptors, including TNFR1, TRAILR, Fas, IFNR and TLR3/4, can initiate necroptotic signalling cascades [67]. Upon receptor activation, the deubiquitinase CYLD catalyses deubiquitination of receptor-interacting protein kinase 1 (RIPK1), an essential step for downstream signal propagation [68]. RIPK1 then activates RIPK3 within a large cytoplasmic complex, the necrosome, and RIPK3 phosphorylates and activates mixed lineage kinase domain-like protein (MLKL), which assembles into homotrimeric necrosomes [69]. In this complex, oligomerization of RIPK3 and its subsequent phosphorylation trigger phosphorylation and activation of MLKL, thereby mediating necroptosis and promoting DAMP release; leakage of DAMPs can further exacerbate necroptosis and inflammation [70].

2.4.3. Ferroptosis

In ferroptosis induced by ischemic stroke, lipid peroxidation and iron accumulation occur simultaneously, accompanied by inhibition of intracellular anti-ferroptotic systems [71]. Lipid peroxidation is a key process in ferroptosis [72], and ferroptosis exhibits pronounced lipid peroxidative stress and cell membrane damage. Lipid peroxidation involves the generation of hydroperoxides from polyunsaturated fatty acids through enzymatic and iron-mediated reactions [73]–[75]. The resulting lipid peroxides can attack adjacent polyunsaturated fatty acids, initiating chain reactions and leading to ferroptosis [76]. Excessive iron accumulation is another defining feature, yet iron sources differ markedly across stroke subtypes [77]. Iron derived from damaged or senescent red blood cells represents a major contributor to brain iron overload predominantly in hemorrhagic stroke or hemorrhagic transformation, rather than in uncomplicated purely-ischemic lesions [77]. In ischemic stroke without hemorrhage, iron overload is largely driven by transferrin-mediated iron uptake and alterations in intracellular iron metabolism [78] [79]. Fe^{2+} released by red blood cell degradation can be oxidized to Fe^{3+} , which binds transferrin (TF) and is taken up by endocytosis via transferrin receptor 1 (TFR1) [78] [79]. After endocytosis, Fe^{3+} is released from TF and reduced back to Fe^{2+} by six-transmembrane epithelial antigen of prostate 3 (STEAP3). Free iron is readily taken up by neurons, producing intracellular iron overload [80]. Intracellular free iron can also catalyze the oxidation of phospholipids containing polyunsaturated fatty acids (PL-PUFAs) through the Fenton reaction and activation of lipoxygenases, thereby producing large amounts of lipid peroxides and further participating in the execution of ferroptosis [81].

2.4.4. Pyroptosis

During ischemic stroke, pyroptosis actively participates in inflammatory responses, can aggravate blood-brain barrier disruption, amplify neuroinflammatory cascades [82], and promote neuronal death. In the pathophysiological process of ischemic stroke, pyroptosis, as a pro-inflammatory inducing factor, mainly occurs in the ischemic penumbra of the brain [83]. Knockout of GSDMD substan-

tially reduces pyroptosis and infarct volume after ischemia/reperfusion by inhibiting microglial secretion of mature IL-18 and IL-1 β [84]. The gasdermin (GSDM) family is now widely recognized as a key regulator of pyroptosis; it comprises five proteins—GSDMA, GSDMB, GSDMC, GSDMD, and GSDME—each capable of mediating this process [85]. The central mechanism is the formation of pores in the cell membrane. These pores disturb the osmotic balance between the intracellular and extracellular compartments, so that the cell swells continuously until the membrane ruptures and intracellular DAMPs spill out, fueling further inflammation [86].

3. Current Status and Shortcomings of Neuroprotection in Acute Ischemic Stroke

In this section, the evidence level of each intervention is explicitly indicated according to the study type from which the reported findings were derived—clinical evidence, human observational evidence, animal experimental evidence, or *in vitro* experimental evidence—so that preclinical mechanistic findings are not misinterpreted as clinically established therapies.

3.1. Anti-Excitotoxicity

3.1.1. Glutamate

Excitotoxicity is one of the earliest discovered and widely recognized molecular mechanisms of post-ischemic stroke injury, in which glutamate and N-methyl-D-aspartate receptors (NMDARs) are important contributing factors [87] [88]. NMDAR is a glutamate-gated ion channel and is fundamental to physiological and pathological processes in the mammalian CNS [89]. NMDAR is a heterotetramer composed of GluN1 subunits and GluN2 subunits (GluN2A to GluN2D) [90]–[92], among which GluN2 subunits play a major role in activation of the NMDAR channel [93]. NMDARs composed of different subunits have different functions. It has been confirmed that NMDARs containing GluN2A (NR2A) are mainly present at synapses, whereas NMDARs containing GluN2B (NR2B) are mainly expressed extrasynaptically [94]. A recent study in a rat model of cerebral ischemia showed that SHPL-49, a structurally modified derivative of salidroside, exerts neuroprotective effects by activating the NR2A subunit of NMDA receptors, thereby upregulating cAMP response element-binding protein (CREB)-mediated neuronal survival pathways and Akt phosphorylation, and prolonging the therapeutic time window, making it a potential drug for the treatment of ischemic stroke (Evidence level: animal experimental evidence) [95].

3.1.2. Calcium Overload

During ischemic stroke, ischemia-induced calcium overload disturbs calcium homeostasis and sets off cytotoxic cascades through protease activation and free-radical generation, eventually producing irreversible neuronal injury [96]. In principle, therefore, blocking Ca²⁺ influx and limiting the degree of calcium overload should protect neurons, counteracting mitochondrial calcium overload, pre-

serving mitochondrial integrity, and improving outcome. Curcumin, a natural modulator, lowers mitochondrial calcium levels and prevents the mitochondrial swelling induced by calcium overload (Evidence level: *in vitro* experimental evidence) [97]. To improve targeting, nanotechnological delivery systems are under exploration; these include TAT peptides for membrane penetration and mitochondrial-enzyme targeting, and GYR peptides that bind transferrin receptors on diseased neurovascular units (NVUs) to achieve precise enrichment (Evidence level: not yet established; currently a preclinical delivery concept) [98]. Abnormal activation of voltage-gated calcium channels (VGCCs), including L-type and T-type channels, is another important source of calcium overload after ischemia [99] [100]. The L-type CCB nimodipine can alleviate vasoconstriction and improve cerebral blood flow by antagonizing CaV1.2 channels (Evidence level: preclinical experimental evidence) [101] [102]. However, traditional CCBs also have some limitations, such as insufficient target specificity (as exemplified by amlodipine) and low selectivity for neuronal calcium channels, which may limit their therapeutic efficacy [103] [104]. In addition, they are associated with hemodynamic risks—excessive blood pressure reduction may aggravate cerebral ischemia, especially in patients with hypoperfusion [102]. It is hoped that the efficacy and safety of these drugs can be further improved in the future [105].

3.2. Anti-Inflammatory Response

Neuroinflammation after stroke is a key process that drives neuronal injury and functional impairment, and many neuroprotective drugs act through inflammatory signaling pathways. Astragaloside IV (AS-IV), a major bioactive constituent of *Radix Astragali*, confers neuroprotection against acute cerebral ischemia-reperfusion injury. In a tMCAO rat model, AS-IV promoted neurogenesis, angiogenesis, and neurological functional recovery, likely via PPAR γ -dependent polarization of microglia/macrophages from the M1 toward the M2 phenotype (Evidence level: animal experimental evidence) [106]. Salvianolic acid C (SalC) markedly reduces post-stroke neuroinflammation and protects neurons from ischemia-reperfusion injury by blocking the TLR4-TREM1-NF- κ B pathway; it also improves neurological function by regulating microglial responses and limiting the release of pro-inflammatory factors, offering a new pharmacological option for neuroinflammation (Evidence level: animal experimental evidence) [107]. Studies have shown that both endogenous and exogenously induced Nrf1 activation can effectively protect nerves from injury (Evidence level: animal experimental evidence) [108] [109].

Despite these encouraging pre-clinical observations for natural-derived small-molecule agents, several translational caveats should be emphasized. Most of these results are derived from young, healthy rodents. Compounds such as polyphenols and herbal extracts frequently suffer from poor gastrointestinal absorption, rapid metabolic clearance and limited blood-brain barrier penetration, which compromise their bioavailability in the ischemic brain. Considerable inter-study variabil-

ity exists with respect to drug dosage, administration routes and intervention timing. Up to now, high-quality clinical evidence to support their routine application for acute ischemic stroke remains absent.

Alterations in drug-delivery strategies directly affect drug distribution and therapeutic efficacy within stroke-affected lesions. Curcumin-loaded polyamide-amine dendrimers can significantly reduce astrocyte reactivity, alleviate neuroinflammation, and promote functional recovery (Evidence level: animal experimental evidence) [110]. Second, taking FTY720 as an example, free drug administration hardly crosses the blood-brain barrier and carries cardiotoxicity; neutrophil membrane-camouflaged polymeric prodrug nanocarriers can actively migrate to ischemic brain regions and release the drug in situ when ROS levels rise, increasing drug concentration in the lesion by 15.2-fold compared with free drug while reducing cardiotoxicity and infection risk. This nanodelivery system also regulates NLRP3 inflammasome and CXCL2 secretion through Cebpb, driving a shift toward an anti-inflammatory microglial phenotype and amplifying the neuroprotective effects of FTY720, highlighting the value of delivery optimization in regulating stroke inflammation (Evidence level: animal experimental evidence) [111].

This therapeutic approach can be achieved through editing. Interferon-induced protein 35 (IFP35) is a 35-kDa protein belonging to a novel class of damage-associated molecular patterns (DAMPs). One study confirmed that IFP35 binds to the receptor TLR4 and activates NF- κ B and NLRP3 signaling pathways, thereby aggravating neuroinflammation and brain injury. Compared with traditional long-acting gene editing approaches, transient siRNA gene silencing is more suitable for the short-term intervention characteristics of acute stroke models (Evidence level: animal experimental evidence) [112]. Second, siRNA-mediated gene knock-down at appropriate concentrations remains a mature and effective strategy, especially suitable for short-term gene silencing in acute stroke models such as MCAO (Evidence level: animal experimental evidence) [113]-[115].

3.3. Anti-Oxidative Stress

Stem cells, as naturally occurring bioactive substances, have attracted considerable attention because they can be used to treat neurological diseases such as ischemic stroke by regulating mitochondrial bioenergetics. For example, human neural stem cells (hNSCs) are highly similar to neural cells in both morphology and function [116]. They can express neuron-specific membrane proteins on their surface, enabling them to cross the blood-brain barrier and deliver tissue-specific neurotrophic factors and differentiation-inducing factors to damaged brain regions, thereby promoting recovery of brain structure and neurological function after ischemic injury (Evidence level: preclinical experimental evidence) [117]. Studies have shown that exosomes derived from human neural stem cells (hNSC-Exos) can alleviate oxidative stress-induced neuronal injury by regulating the PINK1/Parkin pathway. This reveals a novel role of stem cell-derived mitochondria-

drial therapy in promoting neuroprotection and suggests its potential as a therapeutic approach for oxidative stress-related central nervous system diseases, including ischemic stroke (Evidence level: preclinical experimental evidence) [118].

Studies have confirmed that activation of the PI3K/Akt signaling pathway can effectively alleviate oxidative stress caused by ischemia-reperfusion (IS) (Evidence level: preclinical experimental evidence) [119]. Some commonly used natural drugs and their compound preparations can improve and treat ischemic stroke by targeting the PI3K/Akt signaling pathway. Panax notoginseng saponins (PNS) are one of the active components with rich pharmacological effects, capable of promoting mechanisms of neurological recovery and exerting important brain-protective effects after cerebral infarction (Evidence level: preclinical experimental evidence, from both *in vivo* and *in vitro* studies) [120]. They can activate Nrf2 antioxidant signaling through the PI3K/Akt signaling pathway, thereby protecting the blood-brain barrier from hypoxia/reoxygenation injury *in vitro* [121]. Xingnaojing injection (XNJI) is derived from An Gong Niu Huang Wan, and muscone is one of its main active components [122]. In ischemic stroke models, muscone is one of the active components of XNJI. Studies have also shown that it can significantly reduce neuronal apoptosis through the PI3K/Akt signaling pathway (Evidence level: preclinical experimental evidence) [123]. This suggests that XNJI may alleviate IS-induced oxidative stress and neuronal apoptosis through the PI3K/Akt signaling pathway. In addition, total flavonoids of Chujun (TFCJ) are flavonoid active components derived from Chujun, which can also alleviate oxidative stress in cerebral infarction models by activating the PI3K/Akt/mTOR signaling pathway (Evidence level: animal experimental evidence) [124]. In addition to pharmacological treatment, physical interventions can also exert neuroprotective effects by regulating oxidative stress pathways. Transcutaneous electrical acupoint stimulation (TEAS), as a non-invasive physical therapy, significantly reduces brain injury after ischemic stroke by inhibiting apoptosis, oxidative stress, and neuroinflammation and by regulating the SIRT1/FOXO3a and SIRT1/BRCC3/NLRP3 signaling pathways, and is expected to become a potential therapeutic approach for stroke patients (Evidence level: animal experimental evidence) [125]. TEAS does not need to cross the blood-brain barrier and has the advantages of being non-invasive and low-toxicity; however, current evidence is limited to animal models, and causal validation of pathways and human effects remain to be further confirmed. Second, currently common hyperbaric oxygen (HBO) therapy exerts antioxidant protective effects mainly by activating the Nrf2 signaling pathway: after entering the nucleus, Nrf2 upregulates downstream target genes such as HO-1, NQO1, and GCLC, thereby enhancing the activity of antioxidant enzymes such as SOD and promoting GSH synthesis, dually enhancing cellular antioxidant capacity and reducing oxidative damage markers such as MDA to achieve neuroprotection [126]-[129]. An animal study using a treatment regimen of 2.5 atmospheres, 1 h per session, twice daily for two consecutive days found that this treatment activated the Nrf2 signaling pathway in brain tissue of neonatal rats sub-

jected to hypoxic-ischemic injury, upregulated HO-1 and GST, strengthened endogenous antioxidant defense, effectively alleviated oxidative stress, reduced cerebral infarct volume, inhibited neuronal apoptosis, and improved neurological function (Evidence level: animal experimental evidence) [130].

3.4. Targeting Regulated Cell Death

After ischemic stroke, key pathological processes such as neuronal death and neuroinflammation are strictly regulated by ubiquitin-modifying enzymes [131]. One study found that the deubiquitinase OTUB2 aggravates cerebral ischemic injury by promoting RIPK3-mediated neuronal necroptosis [132]. In addition, genetic knockout or pharmacological inhibition of OTUB2 significantly attenuates ischemic stroke injury, suggesting that inhibiting OTUB2 may become a potential therapeutic strategy for ischemic stroke. (Evidence level: animal experimental evidence) Furthermore, studies have shown that caspofungin can inhibit necroptosis by upregulating Pellino3 and promoting RIPK1 ubiquitination, thereby alleviating IS-induced injury (Evidence level: animal experimental evidence) [133]. Telaprevir also alleviates IS-induced brain injury by inhibiting necroptosis mediated by the RIPK1/RIPK3/MLKL pathway (Evidence level: animal experimental evidence) [134].

Various nanoscale strategies have been attempted for neuroprotection in ischemic stroke, although their carrier construction and targeted delivery approaches differ. The ZIF-8-based DZIF@HA nanosystem achieves active targeted delivery to inflammatory microglia via peptide and hyaluronic acid modification, and regulates microglial phenotype through the metabolic drug dimethyl malonate to alleviate neuroinflammation [135]. In contrast, MNPs@Mi uses cell membrane biomimetic camouflage to construct a NIR-II photothermal-responsive platform, synergistically regulating ion channel activity and microglial polarization, protecting neurons, and repairing the blood-brain barrier [136]. Unlike the above systems that require exogenous carrier scaffolds, quercetin-methylcobalamin self-assembled nanoparticles (Qu-Me) require no additional carrier or toxic organic solvents and improve the solubility and bioavailability of natural active molecules through molecular self-assembly; they can likewise inhibit excessive neuroinflammation after stroke and show favorable clinical translation potential [137]. Although the above studies employ markedly different nanoconstruction and drug delivery strategies, all can alleviate ischemic injury, reduce infarct volume, and improve neurological function in preclinical animal models; at the same time, they all face limitations, remain at the basic research stage, and require further clarification of therapeutic time windows, optimal dosing regimens, and synergistic effects with mainstream clinical approaches such as thrombolysis and mechanical thrombectomy (Evidence level: animal experimental evidence).

As a form of programmed cell death, pyroptosis has been fully confirmed in different cell types across multiple diseases. LARP7 regulates NLRP3 deacetyla-

tion by enhancing Sirt1 deacetylase activity, thereby inhibiting inflammasome assembly and neuronal pyroptosis after ischemic stroke. Targeting LARP7 may become a potential strategy for treating ischemic stroke (Evidence level: animal experimental evidence) [138]. HSPA8 lactylation attenuates neuronal pyroptosis after ischemic stroke through E3 ubiquitin ligase-mediated NLRP3 degradation (Evidence level: animal experimental evidence) [139]. Artemisinin can alleviate neuronal pyroptosis induced by ischemic stroke by inhibiting the ROS/TXNIP/NLRP3/caspase-1 signaling pathway (Evidence level: animal experimental evidence) [140]. STING mediates microglial pyroptosis in cerebral ischemic stroke by interacting with NLRP3 (Evidence level: animal experimental evidence) [141]. Ferroptosis is an iron-dependent form of programmed cell death driven by lipid peroxidation and has been confirmed as a key pathological mechanism exacerbating tissue injury during cerebral ischemia-reperfusion injury (IRI)—a stage that is critical in ischemic stroke. In one study, multimodal analysis revealed that cGAMP can alleviate oxidative stress and ferroptosis in ischemic stroke by activating the Nrf2/HO-1/GPX4 pathway, representing a novel dual regulator of redox homeostasis and ferroptosis (Evidence level: animal experimental evidence) [142]. Studies have shown that edaravone dextran can inhibit ferroptosis by activating the Nrf2/HO-1/GPX4 pathway (Evidence level: clinical evidence for its approved clinical use; the ferroptosis mechanism cited here is based on animal experimental evidence) [143]. In addition, activation of the PI3K/AKT signaling pathway can confer cellular resistance to IS-induced ferroptosis. Ginsenoside Rd alleviates ischemia-reperfusion-induced ferroptosis in vascular endothelial cells by activating the neuregulin 1/erb-B2 receptor tyrosine kinase 4/PI3K/Akt/mTOR signaling pathway (Evidence level: animal experimental evidence) [144]. Furthermore, another study revealed that vocarcarcin alleviates ischemia-reperfusion-induced ferroptosis by activating the PI3K-Akt-FoxO pathway (Evidence level: *in vitro* experimental evidence) [145].

4. Conclusion and Perspectives

In summary, brain injury in acute ischemic stroke is not mediated by a single pathway but is a complex cascade process involving crosstalk among excitotoxicity, oxidative stress, and neuroinflammation, with the participation of multiple programmed cell death pathways. In current preclinical studies, small-molecule drugs, natural active components, siRNA gene silencing, stem cell-derived exosomes, nanoscale targeted delivery, and physical interventions such as transcutaneous electrical acupoint stimulation and hyperbaric oxygen, targeting the glutamate-calcium overload axis, inflammatory signaling pathways, oxidative stress, ferroptosis, pyroptosis, and necroptosis, have all shown potential to reduce brain injury and improve neurological function in animal models. Edaravone dextran is one of the few neuroprotective agents in China that has completed clinical translation and is used in acute-phase stroke; however, more real-world studies are still needed to further verify its applicable population, optimal administration

timing, and the safety and efficacy of its combined use with thrombolysis and mechanical thrombectomy. Even so, it should be recognized that most neuroprotective candidates have not moved beyond pre-clinical investigation, leaving a considerable translational divide between laboratory findings and clinical practice. Multiple confounding factors account for such translational disappointments. Firstly, pre-clinical studies frequently adopt young, disease-free experimental animals, which poorly recapitulate the clinical reality of elderly patients complicated by hypertension, diabetes and other comorbid disorders. Secondly, drug-delivery time windows applied in animal models are often relatively permissive and inconsistent with the narrow therapeutic windows observed in real-world clinical settings. Thirdly, limited drug accumulation within damaged cerebral tissue arises from blood-brain-barrier restrictions. Furthermore, single-target therapeutic strategies cannot cope with the intricate pathological network triggered by cerebral ischaemia. Last but not least, few pre-clinical protocols incorporate combined therapeutic regimens alongside reperfusion therapies. Looking ahead, research and development of neuroprotection for acute ischemic stroke may focus on the following directions. To begin with, research should depart from conventional single-target frameworks and attach greater importance to signalling crosstalk across diverse injury cascades. Investigations ought to explore time-dependent multi-target combinatorial strategies, with therapeutic schemes tailored to distinct pathological characteristics between the infarct core and ischaemic penumbra. Additionally, advances in targeted delivery systems, including nanocarriers and cell-membrane biomimetic platforms, deserve further development, with the goal of elevating cerebral drug accumulation while minimizing unwanted systemic adverse reactions. Third, optimize preclinical experimental design, use more aged animal models with underlying comorbidities, and focus on combined intervention studies of neuroprotection with intravenous thrombolysis and mechanical thrombectomy, so as to better approximate real clinical scenarios. Fourth, rethink the bidirectional roles of glial cells, not merely inhibiting inflammation but placing greater emphasis on regulating the polarization of microglia and astrocytes toward reparative phenotypes, and on protecting the neurovascular unit as a whole. Fifth, standardize clinical trial design, strictly define inclusion and exclusion criteria, intervention time windows, and outcome evaluation indicators, and objectively assess benefits and risks. Overall, neuroprotection remains a highly promising research direction for acute ischemic stroke, but at present it cannot yet be used as an independent approach in routine clinical care. Only by fully bridging the gap between basic research and clinical reality and deciphering the complex pathological network of ischemic brain tissue can we truly achieve translation from the laboratory to the patient's bedside and reduce the disability burden caused by stroke.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

References

- [1] Feigin, V.L., Brainin, M., Norrving, B., Martins, S.O., Pandian, J., Lindsay, P., *et al.* (2025) World Stroke Organization: Global Stroke Fact Sheet 2025. *International Journal of Stroke*, **20**, 132-144. <https://doi.org/10.1177/17474930241308142>
- [2] Hankey, G.J. (2017) Stroke. *The Lancet*, **389**, 641-654. [https://doi.org/10.1016/s0140-6736\(16\)30962-x](https://doi.org/10.1016/s0140-6736(16)30962-x)
- [3] Dawson, J., Béjot, Y., Christensen, L.M., De Marchis, G.M., Dichgans, M., Hagberg, G., *et al.* (2022) European Stroke Organisation (ESO) Guideline on Pharmacological Interventions for Long-Term Secondary Prevention after Ischaemic Stroke or Transient Ischaemic Attack. *European Stroke Journal*, **7**, 1-41. <https://doi.org/10.1177/23969873221100032>
- [4] Warner, J.J., Harrington, R.A., Sacco, R.L. and Elkind, M.S.V. (2019) Guidelines for the Early Management of Patients with Acute Ischemic Stroke: 2019 Update to the 2018 Guidelines for the Early Management of Acute Ischemic Stroke. *Stroke*, **50**, 3331-3332. <https://doi.org/10.1161/strokeaha.119.027708>
- [5] Phipps, M.S. and Cronin, C.A. (2020) Management of Acute Ischemic Stroke. *British Medical Journal*, **368**, l6983. <https://doi.org/10.1136/bmj.l6983>
- [6] Rha, J.H. and Saver, J.L. (2007) The Impact of Recanalization on Ischemic Stroke Outcome. *Stroke*, **38**, 967-973. <https://doi.org/10.1161/01.str.0000258112.14918.24>
- [7] Fan, J.L., Brassard, P., Rickards, C.A., Nogueira, R.C., Nasr, N., McBryde, F.D., *et al.* (2021) Integrative Cerebral Blood Flow Regulation in Ischemic Stroke. *Journal of Cerebral Blood Flow & Metabolism*, **42**, 387-403. <https://doi.org/10.1177/0271678x211032029>
- [8] van Putten, M.J.A.M., Fahlke, C., Kafitz, K.W., Hofmeijer, J. and Rose, C.R. (2021) Dysregulation of Astrocyte Ion Homeostasis and Its Relevance for Stroke-Induced Brain Damage. *International Journal of Molecular Sciences*, **22**, Article 5679. <https://doi.org/10.3390/ijms22115679>
- [9] Rose, C.R., Ziemens, D. and Verkhratsky, A. (2020) On the Special Role of NCX in Astrocytes: Translating Na⁺-Transients into Intracellular Ca²⁺ Signals. *Cell Calcium*, **86**, Article 102154. <https://doi.org/10.1016/j.ceca.2019.102154>
- [10] Ludhiadch, A., Sharma, R., Muriki, A. and Munshi, A. (2022) Role of Calcium Homeostasis in Ischemic Stroke: A Review. *CNS & Neurological Disorders—Drug Targets*, **21**, 52-61. <https://doi.org/10.2174/1871527320666210212141232>
- [11] Belov Kirdajova, D., Kriska, J., Tureckova, J. and Anderova, M. (2020) Ischemia-Triggered Glutamate Excitotoxicity from the Perspective of Glial Cells. *Frontiers in Cellular Neuroscience*, **14**, Article 51. <https://doi.org/10.3389/fncel.2020.00051>
- [12] Sun, Y., Feng, X., Ding, Y., *et al.* (2019) Glutamate Receptor-Based Staged Therapeutic Strategy for Cerebral Ischemia. *Frontiers in Cellular Neuroscience*, **13**, Article 168.
- [13] Shen, Z., Xiang, M., Chen, C., Ding, F., Wang, Y., Shang, C., *et al.* (2022) Glutamate Excitotoxicity: Potential Therapeutic Target for Ischemic Stroke. *Biomedicine & Pharmacotherapy*, **151**, Article 113125. <https://doi.org/10.1016/j.biopha.2022.113125>
- [14] Jurcau, A. and Ardelean, A.I. (2022) Oxidative Stress in Ischemia/Reperfusion Injuries Following Acute Ischemic Stroke. *Biomedicine*, **10**, Article 574. <https://doi.org/10.3390/biomedicine10030574>
- [15] Suzuki, H., Kawakita, F. and Asada, R. (2022) Neuroelectric Mechanisms of Delayed Cerebral Ischemia after Aneurysmal Subarachnoid Hemorrhage. *International Journal of Molecular Sciences*, **23**, Article 3102.

- <https://doi.org/10.3390/ijms23063102>
- [16] Wen, B., Xu, K., Huang, R., *et al.* (2022) Inhibition of GRP75 Maintains Mitochondrial Function and Ameliorates Neuronal Injury Caused by Ischemic Stroke. *Molecular Medicine Reports*, **25**, 1-11.
 - [17] Feske, S.K. (2021) Ischemic Stroke. *The American Journal of Medicine*, **134**, 1457-1464. <https://doi.org/10.1016/j.amjmed.2021.07.027>
 - [18] Drieu, A., Levard, D., Vivien, D. and Rubio, M. (2018) Anti-Inflammatory Treatments for Stroke: From Bench to Bedside. *Therapeutic Advances in Neurological Disorders*, **11**, 1-15. <https://doi.org/10.1177/1756286418789854>
 - [19] Iadecola, C. and Anrather, J. (2011) The Immunology of Stroke: From Mechanisms to Translation. *Nature Medicine*, **17**, 796-808. <https://doi.org/10.1038/nm.2399>
 - [20] Yusuying, S., Yusuyin, S. and Cheng, X. (2022) Translocator Protein Regulate Polarization Phenotype Transformation of Microglia after Cerebral Ischemia-Reperfusion Injury. *Neuroscience*, **480**, 203-216. <https://doi.org/10.1016/j.neuroscience.2021.09.024>
 - [21] Cherry, J.D., Olschowka, J.A. and O'Banion, M.K. (2014) Neuroinflammation and M2 Microglia: The Good, the Bad, and the Inflamed. *Journal of Neuroinflammation*, **11**, Article No. 98. <https://doi.org/10.1186/1742-2094-11-98>
 - [22] Works, M.G., Koenig, J.B. and Sapolsky, R.M. (2013) Soluble TNF Receptor 1-Secreting *ex Vivo*-Derived Dendritic Cells Reduce Injury after Stroke. *Journal of Cerebral Blood Flow & Metabolism*, **33**, 1376-1385. <https://doi.org/10.1038/jcbfm.2013.100>
 - [23] Jin, L., Zhu, Z., Hong, L., Qian, Z., Wang, F. and Mao, Z. (2023) ROS-Responsive 18 β -Glycyrrhetic Acid-Conjugated Polymeric Nanoparticles Mediate Neuroprotection in Ischemic Stroke through HMGB1 Inhibition and Microglia Polarization Regulation. *Bioactive Materials*, **19**, 38-49. <https://doi.org/10.1016/j.bioactmat.2022.03.040>
 - [24] Richard, S.A., Sackey, M., Su, Z. and Xu, H. (2017) Pivotal Neuroinflammatory and Therapeutic Role of High Mobility Group Box 1 in Ischemic Stroke. *Bioscience Reports*, **37**, BSR20171104. <https://doi.org/10.1042/bsr20171104>
 - [25] Li, J., Wang, Z., Li, J., Zhao, H. and Ma, Q. (2024) HMGB1: A New Target for Ischemic Stroke and Hemorrhagic Transformation. *Translational Stroke Research*, **16**, 990-1015. <https://doi.org/10.1007/s12975-024-01258-5>
 - [26] Perego, C., Fumagalli, S. and De Simoni, M.G. (2011) Temporal Pattern of Expression and Colocalization of Microglia/Macrophage Phenotype Markers Following Brain Ischemic Injury in Mice. *Journal of Neuroinflammation*, **8**, Article No. 174. <https://doi.org/10.1186/1742-2094-8-174>
 - [27] Yang, J., Ding, S., Huang, W., Hu, J., Huang, S., Zhang, Y., *et al.* (2016) Interleukin-4 Ameliorates the Functional Recovery of Intracerebral Hemorrhage through the Alternative Activation of Microglia/Macrophage. *Frontiers in Neuroscience*, **10**, Article 61. <https://doi.org/10.3389/fnins.2016.00061>
 - [28] Zhao, S.C., Ma, L.S., Chu, Z.H., *et al.* (2017) Regulation of Microglial Activation in Stroke. *Acta Pharmacologica Sinica*, **38**, 445-458. <https://doi.org/10.1038/aps.2016.162>
 - [29] Xie, X. and Liu, J. (2023) New Role of Astrocytes in Neuroprotective Mechanisms after Ischemic Stroke. *Arquivos de Neuro-Psiquiatria*, **81**, 748-755. <https://doi.org/10.1055/s-0043-1770352>
 - [30] Liu, J., Guo, Y., Zhang, Y., Zhao, X., Fu, R., Hua, S., *et al.* (2024) Astrocytes in Is-

- chemic Stroke: Crosstalk in Central Nervous System and Therapeutic Potential. *Neuropathology*, **44**, 3-20. <https://doi.org/10.1111/neup.12928>
- [31] Choudhury, G.R. and Ding, S. (2016) Reactive Astrocytes and Therapeutic Potential in Focal Ischemic Stroke. *Neurobiology of Disease*, **85**, 234-244. <https://doi.org/10.1016/j.nbd.2015.05.003>
- [32] Li, J., Xu, P., Hong, Y., Xie, Y., Peng, M., Sun, R., *et al.* (2023) Lipocalin-2-Mediated Astrocyte Pyroptosis Promotes Neuroinflammatory Injury via NLRP3 Inflammasome Activation in Cerebral Ischemia/Reperfusion Injury. *Journal of Neuroinflammation*, **20**, Article No. 148. <https://doi.org/10.1186/s12974-023-02819-5>
- [33] Menn, B., Garcia-Verdugo, J.M., Yaschine, C., Gonzalez-Perez, O., Rowitch, D. and Alvarez-Buylla, A. (2006) Origin of Oligodendrocytes in the Subventricular Zone of the Adult Brain. *The Journal of Neuroscience*, **26**, 7907-7918. <https://doi.org/10.1523/jneurosci.1299-06.2006>
- [34] Mifsud, G., Zammit, C., Muscat, R., Di Giovanni, G. and Valentino, M. (2014) Oligodendrocyte Pathophysiology and Treatment Strategies in Cerebral Ischemia. *CNS Neuroscience & Therapeutics*, **20**, 603-612. <https://doi.org/10.1111/cns.12263>
- [35] Michalski, D., Keck, A.L., Grosche, J., Martens, H. and Härtig, W. (2018) Immunosignals of Oligodendrocyte Markers and Myelin-Associated Proteins Are Critically Affected after Experimental Stroke in Wild-Type and Alzheimer Modeling Mice of Different Ages. *Frontiers in Cellular Neuroscience*, **12**, Article 23. <https://doi.org/10.3389/fncel.2018.00023>
- [36] Falcão, A.M., van Bruggen, D., Marques, S., Meijer, M., Jäkel, S., Agirre, E., *et al.* (2018) Disease-Specific Oligodendrocyte Lineage Cells Arise in Multiple Sclerosis. *Nature Medicine*, **24**, 1837-1844. <https://doi.org/10.1038/s41591-018-0236-y>
- [37] Kim, E. and Cho, S. (2021) CNS and Peripheral Immunity in Cerebral Ischemia: Partition and Interaction. *Experimental Neurology*, **335**, Article 113508. <https://doi.org/10.1016/j.expneurol.2020.113508>
- [38] Ansari, J., Vital, S.A., Yadav, S. and Gavins, F.N.E. (2023) Regulating Neutrophil PAD4/NOX-Dependent Cerebrovascular Thromboinflammation. *International Journal of Biological Sciences*, **19**, 852-864. <https://doi.org/10.7150/ijbs.77434>
- [39] Denorme, F., Portier, I., Rustad, J.L., Cody, M.J., de Araujo, C.V., Hoki, C., *et al.* (2022) Neutrophil Extracellular Traps Regulate Ischemic Stroke Brain Injury. *Journal of Clinical Investigation*, **132**, e154225. <https://doi.org/10.1172/jci154225>
- [40] Li, C., Xing, Y., Zhang, Y., Hua, Y., Hu, J. and Bai, Y. (2021) Neutrophil Extracellular Traps Exacerbate Ischemic Brain Damage. *Molecular Neurobiology*, **59**, 643-656. <https://doi.org/10.1007/s12035-021-02635-z>
- [41] Bai, M., Sun, R., Cao, B., Feng, J. and Wang, J. (2023) Monocyte-Related Cytokines/Chemokines in Cerebral Ischemic Stroke. *CNS Neuroscience & Therapeutics*, **29**, 3693-3712. <https://doi.org/10.1111/cns.14368>
- [42] Hume, D.A., Ross, I.L., Himes, S.R., Sasmono, R.T., Wells, C.A. and Ravasi, T. (2002) The Mononuclear Phagocyte System Revisited. *Journal of Leukocyte Biology*, **72**, 621-627. <https://doi.org/10.1189/jlb.72.4.621>
- [43] Yilmaz, G., Arumugam, T.V., Stokes, K.Y. and Granger, D.N. (2006) Role of T Lymphocytes and Interferon- γ in Ischemic Stroke. *Circulation*, **113**, 2105-2112. <https://doi.org/10.1161/circulationaha.105.593046>
- [44] Jones, K.A., Maltby, S., Plank, M.W., Kluge, M., Nilsson, M., Foster, P.S., *et al.* (2018) Peripheral Immune Cells Infiltrate into Sites of Secondary Neurodegeneration after

- Ischemic Stroke. *Brain, Behavior, and Immunity*, **67**, 299-307.
<https://doi.org/10.1016/j.bbi.2017.09.006>
- [45] Abadier, M., Haghayegh Jahromi, N., Cardoso Alves, L., Boscacci, R., Vestweber, D., Barnum, S., *et al.* (2015) Cell Surface Levels of Endothelial ICAM-1 Influence the Transcellular or Paracellular T-Cell Diapedesis across the Blood-Brain Barrier. *European Journal of Immunology*, **45**, 1043-1058.
<https://doi.org/10.1002/eji.201445125>
- [46] Wu, M.Y., Yang, G.T., Liao, W.T., *et al.* (2018) Current Mechanistic Concepts in Ischemia and Reperfusion Injury. *Cellular Physiology and Biochemistry*, **46**, 1650-1667. <https://doi.org/10.1159/000489241>
- [47] Andrabi, S.S., Parvez, S. and Tabassum, H. (2020) Ischemic Stroke and Mitochondria: Mechanisms and Targets. *Protoplasma*, **257**, 335-343.
<https://doi.org/10.1007/s00709-019-01439-2>
- [48] Pawluk, H., Kołodziejska, R., Grzešk, G., Woźniak, A., Kozakiewicz, M., Kosinska, A., *et al.* (2022) Increased Oxidative Stress Markers in Acute Ischemic Stroke Patients Treated with Thrombolytics. *International Journal of Molecular Sciences*, **23**, Article 15625. <https://doi.org/10.3390/ijms232415625>
- [49] Cheng, Z., Wang, L., Qu, M., Liang, H., Li, W., Li, Y., *et al.* (2018) Mesenchymal Stem Cells Attenuate Blood-Brain Barrier Leakage after Cerebral Ischemia in Mice. *Journal of Neuroinflammation*, **15**, Article No. 135.
<https://doi.org/10.1186/s12974-018-1153-1>
- [50] Sims, N.R. and Muyderman, H. (2010) Mitochondria, Oxidative Metabolism and Cell Death in Stroke. *Biochimica et Biophysica Acta—Molecular Basis of Disease*, **1802**, 80-91. <https://doi.org/10.1016/j.bbadis.2009.09.003>
- [51] Mao, R., Zong, N., Hu, Y., Chen, Y. and Xu, Y. (2022) Neuronal Death Mechanisms and Therapeutic Strategy in Ischemic Stroke. *Neuroscience Bulletin*, **38**, 1229-1247.
<https://doi.org/10.1007/s12264-022-00859-0>
- [52] Davis, S.M. and Pennypacker, K.R. (2017) Targeting Antioxidant Enzyme Expression as a Therapeutic Strategy for Ischemic Stroke. *Neurochemistry International*, **107**, 23-32. <https://doi.org/10.1016/j.neuint.2016.12.007>
- [53] Ouyang, L., Shi, Z., Zhao, S., *et al.* (2012) Programmed Cell Death Pathways in Cancer: A Review of Apoptosis, Autophagy and Programmed Necrosis. *Cell Proliferation*, **45**, 487-498. <https://doi.org/10.1111/j.1365-2184.2012.00845.x>
- [54] Sarmah, D., Kaur, H., Saraf, J., Vats, K., Pravalika, K., Wanve, M., *et al.* (2018) Mitochondrial Dysfunction in Stroke: Implications of Stem Cell Therapy. *Translational Stroke Research*, **10**, 121-136.
<https://doi.org/10.1007/s12975-018-0642-y>
- [55] Lüscher, C. and Malenka, R.C. (2012) NMDA Receptor-Dependent Long-Term Potentiation and Long-Term Depression (LTP/LTD). *Cold Spring Harbor Perspectives in Biology*, **4**, a005710. <https://doi.org/10.1101/cshperspect.a005710>
- [56] Edlich, F. (2018) BCL-2 Proteins and Apoptosis: Recent Insights and Unknowns. *Biochemical and Biophysical Research Communications*, **500**, 26-34.
<https://doi.org/10.1016/j.bbrc.2017.06.190>
- [57] Yin, X.M. (2000) Signal Transduction Mediated by Bid, a Pro-Death Bcl-2 Family Proteins, Connects the Death Receptor and Mitochondria Apoptosis Pathways. *Cell Research*, **10**, 161-167. <https://doi.org/10.1038/sj.cr.7290045>
- [58] Sugawara, T., Fujimura, M., Noshita, N., Kim, G.W., Saito, A., Hayashi, T., *et al.* (2004) Neuronal Death/Survival Signaling Pathways in Cerebral Ischemia. *NeuroRX*,

- 1, 17-25. <https://doi.org/10.1602/neurorx.1.1.17>
- [59] Polster, B.M. and Fiskum, G. (2004) Mitochondrial Mechanisms of Neural Cell Apoptosis. *Journal of Neurochemistry*, **90**, 1281-1289. <https://doi.org/10.1111/j.1471-4159.2004.02572.x>
- [60] Martin-Villalba, A., Herr, I., Jeremias, I., Hahne, M., Brandt, R., Vogel, J., *et al.* (1999) CD95 Ligand (Fas-L/APO-1l) and Tumor Necrosis Factor-Related Apoptosis-Inducing Ligand Mediate Ischemia-Induced Apoptosis in Neurons. *The Journal of Neuroscience*, **19**, 3809-3817. <https://doi.org/10.1523/jneurosci.19-10-03809.1999>
- [61] Samary, C.S., Ramos, A.B., Maia, L.A., Rocha, N.N., Santos, C.L., Magalhães, R.F., *et al.* (2018) Focal Ischemic Stroke Leads to Lung Injury and Reduces Alveolar Macrophage Phagocytic Capability in Rats. *Critical Care*, **22**, Article No. 249. <https://doi.org/10.1186/s13054-018-2164-0>
- [62] Elmore, S. (2007) Apoptosis: A Review of Programmed Cell Death. *Toxicologic Pathology*, **35**, 495-516. <https://doi.org/10.1080/01926230701320337>
- [63] Nakka, V.P., Gusain, A., Mehta, S.L. and Raghuram, R. (2007) Molecular Mechanisms of Apoptosis in Cerebral Ischemia: Multiple Neuroprotective Opportunities. *Molecular Neurobiology*, **37**, 7-38. <https://doi.org/10.1007/s12035-007-8013-9>
- [64] Ashkenazi, A. (2002) Targeting Death and Decoy Receptors of the Tumour-Necrosis Factor Superfamily. *Nature Reviews Cancer*, **2**, 420-430. <https://doi.org/10.1038/nrc821>
- [65] Gilbert, L.C., Rubin, J. and Nanes, M.S. (2005) The p55 TNF Receptor Mediates TNF Inhibition of Osteoblast Differentiation Independently of Apoptosis. *American Journal of Physiology-Endocrinology and Metabolism*, **288**, E1011-E1018. <https://doi.org/10.1152/ajpendo.00534.2004>
- [66] Kaczmarek, A., Vandenabeele, P. and Krysko, D.V. (2013) Necroptosis: The Release of Damage-Associated Molecular Patterns and Its Physiological Relevance. *Immunity*, **38**, 209-223. <https://doi.org/10.1016/j.immuni.2013.02.003>
- [67] Frank, D. and Vince, J.E. (2019) Pyroptosis versus Necroptosis: Similarities, Differences, and Crosstalk. *Cell Death & Differentiation*, **26**, 99-114. <https://doi.org/10.1038/s41418-018-0212-6>
- [68] Yuan, J.Y., Amin, P. and Ofengeim, D. (2019) Necroptosis and RIPK1-Mediated Neuroinflammation in CNS Diseases. *Nature Reviews Neuroscience*, **20**, 19-33. <https://doi.org/10.1038/s41583-018-0093-1>
- [69] Galluzzi, L., Kepp, O., Krautwald, S., Kroemer, G. and Linkermann, A. (2014) Molecular Mechanisms of Regulated Necrosis. *Seminars in Cell & Developmental Biology*, **35**, 24-32. <https://doi.org/10.1016/j.semcdb.2014.02.006>
- [70] Weber, K., Roelandt, R., Bruggeman, I., Estornes, Y. and Vandenabeele, P. (2018) Nuclear RIPK3 and MLKL Contribute to Cytosolic Necrosome Formation and Necroptosis. *Communications Biology*, **1**, Article No. 6. <https://doi.org/10.1038/s42003-017-0007-1>
- [71] Zhu, K., Zhu, X., Liu, S., Yu, J., Wu, S. and Hei, M. (2022) Glycyrrhizin Attenuates Hypoxic-Ischemic Brain Damage by Inhibiting Ferroptosis and Neuroinflammation in Neonatal Rats via the HMGB1/GPX4 Pathway. *Oxidative Medicine and Cellular Longevity*, **2022**, Article 8438528. <https://doi.org/10.1155/2022/8438528>
- [72] Magtanong, L., Ko, P.J. and Dixon, S.J. (2016) Emerging Roles for Lipids in Non-Apoptotic Cell Death. *Cell Death & Differentiation*, **23**, 1099-1109. <https://doi.org/10.1038/cdd.2016.25>
- [73] Das, U.N. (2019) Saturated Fatty Acids, MUFAs and PUFAs Regulate Ferroptosis.

- Cell Chemical Biology*, **26**, 309-311. <https://doi.org/10.1016/j.chembiol.2019.03.001>
- [74] Bu, Z.Q., Yu, H.Y., Wang, J., *et al.* (2021) Emerging Role of Ferroptosis in the Pathogenesis of Ischemic Stroke: A New Therapeutic Target? *ASN Neuro*, **13**, Article 17590914211037505. <https://doi.org/10.1177/17590914211037505>
- [75] Zou, Y., Li, H., Graham, E.T., Deik, A.A., Eaton, J.K., Wang, W., *et al.* (2020) Cytochrome P450 Oxidoreductase Contributes to Phospholipid Peroxidation in Ferroptosis. *Nature Chemical Biology*, **16**, 302-309. <https://doi.org/10.1038/s41589-020-0472-6>
- [76] Yang, W.S., Kim, K.J., Gaschler, M.M., Patel, M., Shchepinov, M.S. and Stockwell, B.R. (2016) Peroxidation of Polyunsaturated Fatty Acids by Lipoxygenases Drives Ferroptosis. *Proceedings of the National Academy of Sciences*, **113**, E4966 E4975. <https://doi.org/10.1073/pnas.1603244113>
- [77] Zheng, Y., Tan, X., Wang, X., Mao, R. and Guo, J. (2025) Targeting Ferroptosis with Natural Products in Stroke: Therapeutic Mechanisms and Translational Opportunities. *Frontiers in Pharmacology*, **16**, Article 1586345. <https://doi.org/10.3389/fphar.2025.1586345>
- [78] Dixon, S.J. and Stockwell, B.R. (2014) The Role of Iron and Reactive Oxygen Species in Cell Death. *Nature Chemical Biology*, **10**, 9-17. <https://doi.org/10.1038/nchembio.1416>
- [79] Muckenthaler, M.U., Galy, B. and Hentze, M.W. (2008) Systemic Iron Homeostasis and the Iron-Responsive Element/Iron-Regulatory Protein (IRE/IRP) Regulatory Network. *Annual Review of Nutrition*, **28**, 197-213. <https://doi.org/10.1146/annurev.nutr.28.061807.155521>
- [80] Palmer, C., Menzies, S.L., Roberts, R.L., Pavlick, G. and Connor, J.R. (1999) Changes in Iron Histochemistry after Hypoxic-Ischemic Brain Injury in the Neonatal Rat. *Journal of Neuroscience Research*, **56**, 60-71. [https://doi.org/10.1002/\(sici\)1097-4547\(19990401\)56:1<60::aid-jnr8>3.0.co;2-a](https://doi.org/10.1002/(sici)1097-4547(19990401)56:1<60::aid-jnr8>3.0.co;2-a)
- [81] Seiwert, N., Heylmann, D., Hasselwander, S. and Fahrner, J. (2020) Mechanism of Colorectal Carcinogenesis Triggered by Heme Iron from Red Meat. *Biochimica et Biophysica Acta—Reviews on Cancer*, **1873**, Article 188334. <https://doi.org/10.1016/j.bbcan.2019.188334>
- [82] 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 Murine Stroke. *Cell Death & Disease*, **13**, Article No. 20. <https://doi.org/10.1038/s41419-021-04379-z>
- [83] Dong, Z., Pan, K., Pan, J., Peng, Q. and Wang, Y. (2018) The Possibility and Molecular Mechanisms of Cell Pyroptosis after Cerebral Ischemia. *Neuroscience Bulletin*, **34**, 1131-1136. <https://doi.org/10.1007/s12264-018-0294-7>
- [84] Wang, K.K., Sun, Z.Z., Ru, J.N., *et al.* (2020) Ablation of GSDMD Improves Outcome of Ischemic Stroke through Blocking Canonical and Non-Canonical Inflammasomes Dependent Pyroptosis in Microglia. *Frontiers in Neurology*, **11**, Article 577927. <https://doi.org/10.3389/fneur.2020.577927>
- [85] Feng, Y., Li, M., Yangzhong, X., Zhang, X., Zu, A., Hou, Y., *et al.* (2022) Pyroptosis in Inflammation-Related Respiratory Disease. *Journal of Physiology and Biochemistry*, **78**, 721-737. <https://doi.org/10.1007/s13105-022-00909-1>
- [86] Xia, S., Hollingsworth, L.R. and Wu, H. (2020) Mechanism and Regulation of Gasdermin-Mediated Cell Death. *Cold Spring Harbor Perspectives in Biology*, **12**, a036400. <https://doi.org/10.1101/cshperspect.a036400>
- [87] Lucas, D.R. and Newhouse, J.P. (1957) The Toxic Effect of Sodium L-Glutamate on

- the Inner Layers of the Retina. *Archives of Ophthalmology*, **58**, 193-201. <https://doi.org/10.1001/archopht.1957.00940010205006>
- [88] Olney, J.W. (1969) Brain Lesions, Obesity, and Other Disturbances in Mice Treated with Monosodium Glutamate. *Science*, **164**, 719-721. <https://doi.org/10.1126/science.164.3880.719>
- [89] Paoletti, P., Bellone, C. and Zhou, Q. (2013) NMDA Receptor Subunit Diversity: Impact on Receptor Properties, Synaptic Plasticity and Disease. *Nature Reviews Neuroscience*, **14**, 383-400. <https://doi.org/10.1038/nrn3504>
- [90] Jewett, B.E. and Thapa, B. (2021) Physiology, NMDA Receptor. StatPearls Publishing.
- [91] Cull-Candy, S., Brickley, S. and Farrant, M. (2001) NMDA Receptor Subunits: Diversity, Development and Disease. *Current Opinion in Neurobiology*, **11**, 327-335. [https://doi.org/10.1016/s0959-4388\(00\)00215-4](https://doi.org/10.1016/s0959-4388(00)00215-4)
- [92] Yashiro, K. and Philpot, B.D. (2008) Regulation of NMDA Receptor Subunit Expression and Its Implications for LTD, LTP, and Metaplasticity. *Neuropharmacology*, **55**, 1081-1094. <https://doi.org/10.1016/j.neuropharm.2008.07.046>
- [93] Tu, Y.C. and Kuo, C.C. (2015) The Differential Contribution of GluN1 and GluN2 to the Gating Operation of the NMDA Receptor Channel. *Pflügers Archiv—European Journal of Physiology*, **467**, 1899-1917. <https://doi.org/10.1007/s00424-014-1630-z>
- [94] Thomas, C.G., Miller, A.J. and Westbrook, G.L. (2006) Synaptic and Extrasynaptic NMDA Receptor NR2 Subunits in Cultured Hippocampal Neurons. *Journal of Neurophysiology*, **95**, 1727-1734. <https://doi.org/10.1152/jn.00771.2005>
- [95] Xie, D., Zhang, P., You, S., Shen, Y., Xu, W., Zhan, C., *et al.* (2024) Salidroside Derivative SHPL-49 Attenuates Glutamate Excitotoxicity in Acute Ischemic Stroke via Promoting NR2A-CAMKII α -Akt/CREB Pathway. *Phytomedicine*, **134**, Article 155583. <https://doi.org/10.1016/j.phymed.2024.155583>
- [96] Zhang, Y., Zhang, G. and Chen, X. (2024) Elevated Calcium after Acute Ischemic Stroke Predicts Severity and Prognosis. *Molecular Neurobiology*, **61**, 266-275. <https://doi.org/10.1007/s12035-023-03581-8>
- [97] Jara, C., Torres, A.K., Park-Kang, H.S., Sandoval, L., Retamal, C., Gonzalez, A., *et al.* (2025) Curcumin Improves Hippocampal Cell Bioenergetics, Redox and Inflammatory Markers, and Synaptic Proteins, Regulating Mitochondrial Calcium Homeostasis. *Neurotoxicity Research*, **43**, Article No. 3. <https://doi.org/10.1007/s12640-024-00726-y>
- [98] Novorolsky, R.J., Kasheke, G.D.S., Hakim, A., Foldvari, M., Dorighello, G.G., Sekler, I., *et al.* (2023) Preserving and Enhancing Mitochondrial Function after Stroke to Protect and Repair the Neurovascular Unit: Novel Opportunities for Nanoparticle-Based Drug Delivery. *Frontiers in Cellular Neuroscience*, **17**, Article 1226630. <https://doi.org/10.3389/fncel.2023.1226630>
- [99] Rahi, V. and Kaundal, R.K. (2024) Exploring the Intricacies of Calcium Dysregulation in Ischemic Stroke: Insights into Neuronal Cell Death and Therapeutic Strategies. *Life Sciences*, **347**, Article 122651. <https://doi.org/10.1016/j.lfs.2024.122651>
- [100] Pichardo-Rojas, D., Pichardo-Rojas, P.S., Cornejo-Bravo, J.M. and Serrano-Medina, A. (2023) Memantine as a Neuroprotective Agent in Ischemic Stroke: Preclinical and Clinical Analysis. *Frontiers in Neuroscience*, **17**, Article 1096372. <https://doi.org/10.3389/fnins.2023.1096372>
- [101] Poore, C.P., Yang, J., Wei, S., Fhu, C.K., Bichler, Z., Wang, J., *et al.* (2024) Enhanced Isradipine Sensitivity in Vascular Smooth Muscle Cells Due to Hypoxia-Induced Ca v 1.2 Splicing and RbFox1/Fox2 Downregulation. *The FEBS Journal*, **291**, 4265-4285.

- <https://doi.org/10.1111/febs.17159>
- [102] Kecskés, S., Menyhárt, Á., Bari, F. and Farkas, E. (2023) Nimodipine Augments Cerebrovascular Reactivity in Aging but Runs the Risk of Local Perfusion Reduction in Acute Cerebral Ischemia. *Frontiers in Aging Neuroscience*, **15**, Article 1175281. <https://doi.org/10.3389/fnagi.2023.1175281>
 - [103] Bernard, P.J., Bellili, D. and Ismaili, L. (2024) Calcium Channel Blockers' Contribution to Overcoming Current Drug Discovery Challenges in Alzheimer's Disease. *Expert Opinion on Drug Discovery*, **19**, 21-32. <https://doi.org/10.1080/17460441.2023.2266994>
 - [104] Tian, S., Ma, C., Zhu, Y., Xu, Q., Wu, J., Qiu, Y., *et al.* (2025) A Light-Addressable Potentiometric Sensor-Based Extracellular Calcium Dynamic Monitoring and Imaging Platform for Cellular Calcium Channel Drug Evaluation. *Biosensors and Bioelectronics*, **267**, Article 116814. <https://doi.org/10.1016/j.bios.2024.116814>
 - [105] Zhou, R., Lin, K., Leng, C., Zhou, M., Zhang, J., Li, Y., *et al.* (2022) Chebulic Acid Prevents Hypoxia Insult via Nrf2/ARE Pathway in Ischemic Stroke. *Nutrients*, **14**, Article 5390. <https://doi.org/10.3390/nu14245390>
 - [106] Li, L., Gan, H., Jin, H., Fang, Y., Yang, Y., Zhang, J., *et al.* (2021) Astragaloside IV Promotes Microglia/Macrophages M2 Polarization and Enhances Neurogenesis and Angiogenesis through PPAR γ Pathway after Cerebral Ischemia/Reperfusion Injury in Rats. *International Immunopharmacology*, **92**, Article 107335. <https://doi.org/10.1016/j.intimp.2020.107335>
 - [107] Guo, W., Xu, X., Xiao, Y., Zhang, J., Shen, P., Lu, X., *et al.* (2024) Salvianolic Acid C Attenuates Cerebral Ischemic Injury through Inhibiting Neuroinflammation via the TLR4-TREM1-NF- κ B Pathway. *Chinese Medicine*, **19**, Article No. 46. <https://doi.org/10.1186/s13020-024-00914-0>
 - [108] Fan, H., Ding, R., Liu, W., Zhang, X., Li, R., Wei, B., *et al.* (2021) Heat Shock Protein 22 Modulates NRF1/TFAM-Dependent Mitochondrial Biogenesis and DRP1-Sparked Mitochondrial Apoptosis through AMPK-PGC1 α Signaling Pathway to Alleviate the Early Brain Injury of Subarachnoid Hemorrhage in Rats. *Redox Biology*, **40**, Article 101856. <https://doi.org/10.1016/j.redox.2021.101856>
 - [109] Yang, M., Shen, Y., Zhao, S., Zhang, R., Dong, W. and Lei, X. (2023) Protective Effect of Resveratrol on Mitochondrial Biogenesis during Hyperoxia-Induced Brain Injury in Neonatal Pups. *BMC Neuroscience*, **24**, Article No. 27. <https://doi.org/10.1186/s12868-023-00797-1>
 - [110] Stadler, J., Garmo, L.G., Doyle, D., Cheng, C., Richardson, G., Waheed, Z., *et al.* (2025) Curcumin Encapsulated in PAMAM Dendrimers for the Therapeutic Treatment of Ischemic Stroke in Rats. *Frontiers in Cell and Developmental Biology*, **12**, Article 1467417. <https://doi.org/10.3389/fcell.2024.1467417>
 - [111] Zhao, Y., Li, Q., Niu, J., Guo, E., Zhao, C., Zhang, J., *et al.* (2024) Neutrophil Membrane-Camouflaged Polyprodrug Nanomedicine for Inflammation Suppression in Ischemic Stroke Therapy. *Advanced Materials*, **36**, e2311803. <https://doi.org/10.1002/adma.202311803>
 - [112] Zhang, M., Guo, B., Zhang, X., Han, D., Lv, L., Yan, X., *et al.* (2025) IFP35, a Novel DAMP, Aggravates Neuroinflammation Following Acute Ischemic Stroke via TLR4/NF- κ B/NLRP3 Signaling. *Journal of Neuroinflammation*, **22**, Article No. 164. <https://doi.org/10.1186/s12974-025-03492-6>
 - [113] Burnett, J.C., Rossi, J.J. and Tiemann, K. (2011) siRNA Treatment: "A Sword-in-the-Stone" for Acute Brain Injuries. *Journal of Neuroimmune Pharmacology*, **6**, 360-375.

- [114] Kim, I., Shin, J., Kim, S., Choi, S., Ahn, J., Han, P., *et al.* (2012) Intranasal Delivery of HMGB1 Sirna Confers Target Gene Knockdown and Robust Neuroprotection in the Postischemic Brain. *Molecular Therapy*, **20**, 829-839. <https://doi.org/10.1038/mt.2011.291>
- [115] Chelluboina, B., Kim, T., Mehta, S.L., Kim, J., Bathula, S. and Vemuganti, R. (2020) Impact of Age and Sex on α -Syn (α -Synuclein) Knockdown-Mediated Poststroke Recovery. *Stroke*, **51**, 3138-3141. <https://doi.org/10.1161/strokeaha.120.028978>
- [116] Chen, L., Zhang, G., Gu, Y. and Guo, X. (2016) Meta-Analysis and Systematic Review of Neural Stem Cells Therapy for Experimental Ischemia Stroke in Preclinical Studies. *Scientific Reports*, **6**, Article No. 32291. <https://doi.org/10.1038/srep32291>
- [117] Jiao, Y., Liu, Y.W., Chen, W.G., *et al.* (2021) Neuroregeneration and Functional Recovery after Stroke: Advancing Neural Stem Cell Therapy toward Clinical Application. *Neural Regeneration Research*, **16**, 80-92. <https://doi.org/10.4103/1673-5374.286955>
- [118] Zhao, M., Wang, J., Zhu, S., Zhang, S., Han, C., Tan, C., *et al.* (2025) Human Neural Stem Cell-Derived Exosomes Activate PINK1/Parkin Pathway to Protect against Oxidative Stress-Induced Neuronal Injury in Ischemic Stroke. *Journal of Translational Medicine*, **23**, Article No. 402. <https://doi.org/10.1186/s12967-025-06283-y>
- [119] Samakova, A., Gazova, A., Sabova, N., Valaskova, S., Jurikova, M. and Kyselovic, J. (2019) The PI3K/Akt Pathway Is Associated with Angiogenesis, Oxidative Stress and Survival of Mesenchymal Stem Cells in Pathophysiologic Condition in Ischemia. *Physiological Research*, **68**, S131-S138. <https://doi.org/10.33549/physiolres.934345>
- [120] Liu, L., Zhu, L., Zou, Y., Liu, W., Zhang, X., Wei, X., *et al.* (2014) Panax Notoginseng Saponins Promotes Stroke Recovery by Influencing Expression of Nogo-A, NgR and p75NGF, *in Vitro* and *in Vivo*. *Biological and Pharmaceutical Bulletin*, **37**, 560-568. <https://doi.org/10.1248/bpb.b13-00770>
- [121] Hu, S., Wu, Y., Zhao, B., Hu, H., Zhu, B., Sun, Z., *et al.* (2018) Panax Notoginseng Saponins Protect Cerebral Microvascular Endothelial Cells against Oxygen-Glucose Deprivation/Reperfusion-Induced Barrier Dysfunction via Activation of PI3K/Akt/Nrf2 Antioxidant Signaling Pathway. *Molecules*, **23**, Article 2781. <https://doi.org/10.3390/molecules23112781>
- [122] Song, Y.F., Chu, Y., Ma, X., Zheng, H., Bai, X., Zhou, S., *et al.* (2017) GC-MS/MS Method for the Determination and Pharmacokinetic Analysis of Borneol and Muscone in Rat after the Intravenous Administration of Xingnaojing Injection. *Journal of Separation Science*, **40**, 4264-4271. <https://doi.org/10.1002/jssc.201700341>
- [123] Liu, Z., Li, H., Ma, W. and Pan, S. (2021) Network Pharmacology to Investigate the Pharmacological Mechanisms of Muscone in Xingnaojing Injections for the Treatment of Severe Traumatic Brain Injury. *PeerJ*, **9**, e11696. <https://doi.org/10.7717/peerj.11696>
- [124] Wang, C., Chen, H., Jiang, H., Mao, B. and Yu, H. (2021) Total Flavonoids of Chuju Decrease Oxidative Stress and Cell Apoptosis in Ischemic Stroke Rats: Network and Experimental Analyses. *Frontiers in Neuroscience*, **15**, Article 772401. <https://doi.org/10.3389/fnins.2021.772401>
- [125] Tan, Z., Dong, F., Wu, L., Xu, G. and Zhang, F. (2024) Transcutaneous Electrical Acupoint Stimulation Attenuated Neuroinflammation and Oxidative Stress by Activating SIRT1-Induced Signaling Pathway in MCAO/R Rat Models. *Experimental Neurology*, **373**, Article 114658. <https://doi.org/10.1016/j.expneurol.2023.114658>
- [126] Adelusi, T.I., Du, L., Hao, M., Zhou, X., Xuan, Q., Apu, C., *et al.* (2020) Keap1/Nrf2/ARE Signaling Unfolds Therapeutic Targets for Redox Imbalanced-Mediated Dis-

- eases and Diabetic Nephropathy. *Biomedicine & Pharmacotherapy*, **123**, Article 109732. <https://doi.org/10.1016/j.biopha.2019.109732>
- [127] Liu, X., Liang, F., Song, W., Diao, X., Zhu, W. and Yang, J. (2021) Effect of Nrf2 Signaling Pathway on the Improvement of Intestinal Epithelial Barrier Dysfunction by Hyperbaric Oxygen Treatment after Spinal Cord Injury. *Cell Stress and Chaperones*, **26**, 433-441. <https://doi.org/10.1007/s12192-020-01190-1>
- [128] Li, C., Wu, Z., Chen, F., Dai, C., Yang, X., Ye, S., *et al.* (2025) Regulation of Nrf2/GPX4 Signaling Pathway by Hyperbaric Oxygen Protects against Depressive Behavior and Cognitive Impairment in a Spinal Cord Injury Rat Model. *CNS Neuroscience & Therapeutics*, **31**, e70421. <https://doi.org/10.1111/cns.70421>
- [129] Lindenmann, J., Kamolz, L., Graier, W., *et al.* (2022) Hyperbaric Oxygen Therapy and Tissue Regeneration: A Literature Survey. *Biomedicines*, **10**, Article 3145. <https://doi.org/10.3390/biomedicines10123145>
- [130] Zhai, X., Lin, H., Chen, Y., Chen, X., Shi, J., Chen, O., *et al.* (2016) Hyperbaric Oxygen Preconditioning Ameliorates Hypoxia-Ischemia Brain Damage by Activating Nrf2 Expression *in Vivo* and *in Vitro*. *Free Radical Research*, **50**, 454-466. <https://doi.org/10.3109/10715762.2015.1136411>
- [131] Huang, J., Zhu, Z., Schlüter, D., Lambertsen, K.L., Song, W. and Wang, X. (2024) Ubiquitous Regulation of Cerebrovascular Diseases by Ubiquitin-Modifying Enzymes. *Clinical and Translational Medicine*, **14**, e1719. <https://doi.org/10.1002/ctm2.1719>
- [132] Mei, F., Deng, D., Cao, Z., Lou, L., Chen, K., Hu, M., *et al.* (2025) Deubiquitination of RIPK3 by OTUB2 Potentiates Neuronal Necroptosis after Ischemic Stroke. *EMBO Molecular Medicine*, **17**, 679-695. <https://doi.org/10.1038/s44321-025-00206-6>
- [133] Zhang, Y.Y., Tian, J., Peng, Z.M., *et al.* (2023) Caspofungin Suppresses Brain Cell Necroptosis in Ischemic Stroke Rats via Up-Regulation of Pellino3. *Cardiovascular Drugs and Therapy*, **37**, 9-23. <https://doi.org/10.1007/s10557-021-07231-w>
- [134] Zhang, Y.Y., Peng, J.J., Chen, D., *et al.* (2023) Telaprevir Improves Memory and Cognition in Mice Suffering Ischemic Stroke via Targeting Malt1-Mediated Calcium Overload and Necroptosis. *ACS Chemical Neuroscience*, **14**, 3113-3124. <https://doi.org/10.1021/acscchemneuro.3c00250>
- [135] Li, X., Tao, T., Feng, Y., Zhao, Y., Chai, J. and Zhang, B. (2026) Targeted Delivery of Dimethyl Malonate via a ZIF-8-Based Nanocarrier for Ischemic Stroke Treatment. *ACS Applied Nano Materials*, **9**, 9652-9665. <https://doi.org/10.1021/acsnm.6c00714>
- [136] Liu, X., Wu, Z., Lv, H., Li, B., Li, C., Zhao, J., *et al.* (2026) NIR-II-Responsive Biomimetic NanoplatforM for Precise Inhibition of Ischemic Stroke via Dual Modulation of Ion Channels and Microglia. *ACS Applied Materials & Interfaces*, **18**, 9497-9511. <https://doi.org/10.1021/acsam.5c23577>
- [137] Zhang, T., Fan, S., Ji, H., Liu, W., Chen, X. and Fu, D. (2026) Self-Assembled Quercetin-Mecobalamin Nanoparticles Mitigate Ischemic Stroke Injury via Enhanced Neuroprotection and Microglial Modulation. *ACS Biomaterials Science & Engineering*, **12**, 2606-2618. <https://doi.org/10.1021/acsbomaterials.5c01855>
- [138] Tang, H., Wen, J., Yang, Q., Wang, L., Qin, T., Ren, Y., *et al.* (2025) LARP7 Protects against Ischemic Brain Injury by Modulating NLRP3 Deacetylation to Suppress Neuronal Pyroptosis. *Journal of Neuroinflammation*, **23**, Article No. 13. <https://doi.org/10.1186/s12974-025-03649-3>
- [139] Fu, J., Yu, L., Yu, N., Ni, P., He, C., Chen, Q., *et al.* (2026) HSPA8 Lactylation Attenuates Neuronal Pyroptosis via E3 Ligase-Mediated NLRP3 Degradation after Ischemic Stroke. *Apoptosis*, **31**, Article No. 37.

- <https://doi.org/10.1007/s10495-025-02226-6>
- [140] Wang, Y., Yuan, H., Shen, D., Liu, S., Kong, W., Zheng, K., *et al.* (2024) Artemisinin Attenuated Ischemic Stroke Induced Pyroptosis by Inhibiting ROS/TXNIP/NLRP3/Caspase-1 Signaling Pathway. *Biomedicine & Pharmacotherapy*, **177**, Article 116894. <https://doi.org/10.1016/j.biopha.2024.116894>
- [141] Li, W., Shen, N., Kong, L., Huang, H., Wang, X., Zhang, Y., *et al.* (2023) STING Mediates Microglial Pyroptosis via Interaction with NLRP3 in Cerebral Ischaemic Stroke. *Stroke and Vascular Neurology*, **9**, 153-164. <https://doi.org/10.1136/svn-2023-002320>
- [142] Li, X., Qian, S., Hou, Z., Yang, G., Ma, Y., Gao, Y., *et al.* (2025) Activation of the Nrf2/HO-1/GPX4 Pathway by cGAMP Mitigates Oxidative Stress and Ferroptosis in Ischaemic Stroke. *Biochemical Pharmacology*, **242**, Article 117334. <https://doi.org/10.1016/j.bcp.2025.117334>
- [143] Xiao, P., Huang, H., Zhao, H., Liu, R., Sun, Z., Liu, Y., *et al.* (2024) Edaravone Dexborneol Protects against Cerebral Ischemia/Reperfusion-Induced Blood-Brain Barrier Damage by Inhibiting Ferroptosis via Activation of Nrf-2/HO-1/GPX4 Signaling. *Free Radical Biology and Medicine*, **217**, 116-125. <https://doi.org/10.1016/j.freeradbiomed.2024.03.019>
- [144] Hu, S., Fei, Y., Jin, C., Yao, J., Ding, H., Wang, J., *et al.* (2024) Ginsenoside Rd Enhances Blood-Brain Barrier Integrity after Cerebral Ischemia/Reperfusion by Alleviating Endothelial Cells Ferroptosis via Activation of NRG1/ErbB4-Mediated PI3K/Akt/mTOR Signaling Pathway. *Neuropharmacology*, **251**, Article 109929. <https://doi.org/10.1016/j.neuropharm.2024.109929>
- [145] Li, Y., Sun, Y., Wang, J., Wang, X. and Yang, W. (2024) Voacangine Protects Hippocampal Neuronal Cells against Oxygen-Glucose Deprivation/Reoxygenation-Caused Oxidative Stress and Ferroptosis by Activating the PI3K-Akt-FoxO Signaling. *Journal of Applied Toxicology*, **44**, 1246-1256. <https://doi.org/10.1002/jat.4615>