Journal of Ginseng Research 47 (2023) 615–621

Contents lists available at ScienceDirect

Journal of Ginseng Research

journal homepage: https://www.sciencedirect.com/journal/journal-of-ginseng-research

Review Article

The effects of Korean Red Ginseng on heme oxygenase-1 with a focus on mitochondrial function in pathophysiologic conditions

Chang-Hee Kim [a], Hahn Young Kim [b], Seung-Yeol Nah [c], Yoon Kyung Choi [d, *]

[a] Department of Otorhinolaryngology-Head and Neck Surgery, Konkuk University Medical Center, Konkuk University School of Medicine, Republic of Korea
[b] Department of Neurology, Research Institute of Medical Science, Konkuk University School of Medicine, Seoul, Republic of Korea
[c] Ginsentology Research Laboratory and Department of Physiology, College of Veterinary Medicine, Konkuk University, Seoul, Republic of Korea
[d] Bio/Molecular Informatics Center, Department of Bioscience and Biotechnology, Konkuk University, Seoul, Republic of Korea

ARTICLE INFO

Article history:
Received 14 November 2022
Received in revised form
21 March 2023
Accepted 7 April 2023
Available online 8 April 2023

Keywords:
Astrocytes
Ginseng
Heme oxygenase
Cell communication
Mitochondria

ABSTRACT

Korean Red Ginseng (KRG) plays a key role in heme oxygenase (HO)-1 induction under physical and moderate oxidative stress conditions. The transient and mild induction of HO-1 is beneficial for cell protection, mitochondrial function, regeneration, and intercellular communication. However, chronic HO-1 overexpression is detrimental in severely injured regions. Thus, in a chronic pathological state, diminishing HO-1-mediated ferroptosis is beneficial for a therapeutic approach. The molecular mechanisms by which KRG protects various cell types in the central nervous system have not yet been established, especially in terms of HO-1-mediated mitochondrial functions. Therefore, in this review, we discuss the multiple roles of KRG in the regulation of astrocytic HO-1 under pathophysiological conditions. More specifically, we discuss the role of the KRG-mediated astrocytic HO-1 pathway in regulating mitochondrial functions in acute and chronic neurodegenerative diseases as well as physiological conditions.

1. Introduction

Korean Red Ginseng (KRG) (*Panax ginseng Meyer*) contains a mixture of saponin monomers and ginsenosides [1]. KRG has been considered an herbal medicine for a long time. Heme oxygenase (HO) (i.e., HO-1 and HO-2) is an essential enzyme in heme catabolism that cleaves heme to produce carbon monoxide (CO), biliverdin (which is rapidly converted to bilirubin), and ferrous iron (Fe^{2+}); oxygen is required as a co-substrate in this step [2]. HO-1 is induced in numerous cells in response to hypoxia and stress and promotes neuroprotection and angiogenesis in mildly stressful environments [3]. In contrast, HO-2 is constitutively expressed in some cells, where it functions as an intrinsic protector [4].

Neurodegenerative diseases of the central nervous system can be divided into acute (e.g., traumatic brain injury (TBI) and stroke) and chronic (e.g., Alzheimer's disease) diseases. The molecular mechanisms involved in the acute and chronic phases may be different according to redox states, degree of neurovascular damage, and disruption of cell–cell communications [5–7]. Transient and acute expression of HO-1 may act as a protector, while chronic overexpression of HO-1 can be detrimental to cells due to the accumulation of excessive ferric iron (Fe^{3+}) in the mitochondria [8,9].

KRG may have dual effects: one by the stimulation of cytoprotection, partly through HO-1-mediated mitochondrial functions in the peri-injured region in acute neurodegenerative diseases and physiological conditions. In addition, KRG may diminish HO-1-mediated toxic effects, partly through excessive iron accumulation in the core region of acute and chronic neurodegenerative diseases. In this review, we have discussed KRG-induced HO-1-mediated functions in acute neurovascular diseases (in the penumbra vs. core region) and chronic neurodegenerative diseases, as well as physiological conditions.

2. Role of KRG in physiological conditions

Determining the prophylactic roles of KRG in the central nervous system is important because of the long-term use of KRG. KRG-mediated signaling molecules may be involved in HO-1 induction and interact with the HO metabolites [10,11]. In this

* Corresponding author. Bio/Molecular Informatics Center, Department of Bioscience and Biotechnology, Konkuk University, Seoul, 05029, Republic of Korea.
E-mail address: ykchoi@konkuk.ac.kr (Y.K. Choi).

https://doi.org/10.1016/j.jgr.2023.04.001

section, we will discuss the role of HO-1 with a focus on mitochondria in the presence of KRG under physiological conditions.

2.1. HO-1 induction in physiological conditions

KRG induces HO-1 in various cells such as endothelial cells and astrocytes under physiological conditions [12–14]. KRG-induced HO-1 may play key roles in anti-aging, neurovascular protection, angiogenesis, and mitochondrial biogenesis [12–14]. KRG has mitochondrial biogenic capacity, especially in astrocytes, through the HO-1-hypoxia-inducible factor-1α (HIF-1α) pathway under physiological conditions [13]. Moreover, astrocytes secrete HIF-1α-regulated vascular endothelial growth factor (VEGF), which induces the endothelial nitric oxide (eNOS)/NO pathway in endothelial cells and is related to vascular functions [15]. A clinical trial with healthy individuals revealed that KRG affects vascular tone [16]. Experiments using aged mice demonstrated that KRG restores impaired endothelial cell functions through the eNOS/NO pathway [17].

What are the factors important for HO-1 induction by KRG? Nuclear factor erythroid 2-related factor 2 (Nrf2) is an upstream factor and regulates HO-1 expression [12]. The Kelch-like ECH-associated protein 1 (Keap1)-Nrf2 pathway is important for HO-1 induction depending on the redox state. Under physiological conditions, Nrf2 binds to Keap1 in the cytoplasm, leading to the proteasomal degradation of Nrf2. Keap1 is inactivated by modification of reactive cysteine residues by oxidative stress, and then the stabilized Nrf2 translocates to the nucleus to activate the transcription of HO-1, superoxide dismutase (SOD), and glutathione (GSH) peroxidase 4 (GPX4) [18–20].

How can Nrf2 be induced under physiological conditions? One hypothesis is that KRG boosts mitochondrial biogenesis, which may result in acute oxidative stress before SOD2 upregulation. Mitochondrial activity-mediated reactive oxygen species generation modifies Keap1 stability, enabling the translocation of Nrf2 into the nucleus. This process may result in detoxification and HO-1 induction. Another hypothesis is that HO-1-induced CO can dissociate Nrf2-Keap1 interactions, leading to the translocation of Nrf2 to the nucleus [21]. Under oxidative stress, biliverdin reductase A (BVR-A) acts as a transcription factor for HO-1 [22]. However, we could not determine the BVR-A–HO–1 axis in astrocytes under physiological conditions [23]. Collectively, Keap1 and BVR-A may have different susceptibilities to oxidative stress in KRG-treated physiological environments.

2.2. HO-1-mediated mitochondrial functions

KRG promotes the proliferation of astrocytes in the subventricular zone of murine brain tissues [13]. Under physiological conditions, KRG-induced HO-1 can induce HIF-1α protein expression in astrocytes [13]. HIF-1α is a key transcription factor regulating neurovascular remodeling through cell metabolism, angiogenesis, and neurogenesis [13,24]. KRG-induced HIF-1α increases the expression of mitochondrial functional proteins involved in processes including oxidative phosphorylation (OXPHOS) and ATP production [13]. KRG increases HIF-1α protein expression under normoxic conditions, partly due to increased KRG-mediated enhancement of mitochondrial mass and O_2 consumption [13,23].

One of the HO-1 metabolites is CO. CO can upregulate the translational activity and post-translational modification of HIF-1α, leading to HIF-1α stability in astrocytes [11]. The combination of CO and bilirubin enables Ca^{2+} influx and Ca^{2+}/calmodulin-dependent protein kinase β (CaMKKβ)-mediated AMP-activated protein kinase α (AMPKα) activation [25]. Inhibition of Ca^{2+} signaling or AMPKα decreases HIF-1α stability, which is related to the paired effects of CO and bilirubin [26]. The physiological roles of KRG in HIF-1α activation and the subsequent VEGF secretion may stem from HO-1 metabolites such as CO and biliverdin.

KRG-administered murine brain tissues show increased levels of BVR-A in hippocampal astrocytes [23]. Conversion of biliverdin to bilirubin by BVR-A upregulates SOD2, leading to the blocking of excessive reactive oxygen species production [23]. In addition, the KRG-mediated BVR-A/bilirubin pathway upregulates estrogen-related receptor α (ERRα) [23], which is an important transcription factor for VEGF, by binding to peroxisome proliferator-activated receptor γ coactivator 1α (PGC-1α) [27]. ERRα pathway is considered as a critical regulator of mitochondrial biogenesis-related functions [26,28]. A recent study suggested that KRG induces astrocytic mitochondrial activity and function through the liver kinase B1 (LKB1)-sirtuin 1 (SIRT1)-ERRα pathway under unstimulated conditions [23]. Activation of LKB1 is clearly detected by bilirubin but not by biliverdin, and KRG induces the expression of SIRT1 through LKB1, but not through CaMKKβ [23]. SIRT1 is expressed in cytosolic fractions and activates PGC-1α deacetylase [29]. Activation of PGC-1α through SIRT1 may be involved in ERRα-mediated mitochondrial functions and VEGF expression [27,30]. KRG-mediated SIRT5 expression, mainly expressed in the mitochondrial fraction, is regulated by CaMKKβ [23]. Diminished SIRT1 and SIRT5 expression almost completely blocks the expression of KRG-mediated mitochondrial functional proteins such as SOD2, cytochrome c, translocase of outer mitochondrial membrane 20 (TOM20), and TOM22 [23]. In addition to SIRT1 and SIRT5, there are other SIRTs (SIRT2, SIRT3, SIRT4, SIRT6, and SIRT7); however, KRG-mediated expression of SIRTs has not been well investigated. Therefore, further research is needed to determine the regenerative functions (e.g., mitochondrial biogenesis, angiogenesis, neurogenesis, and anti-aging) of KRG in relation to other SIRTs.

Taken together, KRG activates HIF-1α, ERRα, LKB1, CaMKKβ, SIRT1, and SIRT5 in normal astrocytes. These signaling molecules are regulated by HO-1 and its metabolites, such as CO and biliverdin. The conversion of bilirubin from biliverdin by BVR-A may be an important process for the KRG-mediated antioxidant effects through SOD2 and mitochondrial functions through the TOM complex (i.e., TOM20 and TOM22) and OXPHOS (Fig. 1).

3. Role of KRG in pathological conditions

In the penumbra region of TBI, HO-1 induction by KRG is much higher than that in physical murine brain tissues [31]. HO-1-induced astrocytes possess active mitochondrial functions [31,32]. Many researchers have tried to determine the functions of KRG, and its anti-inflammatory and regenerative roles have been evaluated under pathological conditions (reviewed in [1]). Therefore, we have discussed below the role of KRG in HO-1-mediated functions, especially mitochondrial functions, in acute and chronic diseases.

3.1. KRG in acute neurovascular diseases

We have discussed the effects of KRG in acute neurovascular brain regions, such as the peri-injured region (penumbra) and core-injured region separately, because the cellular morphology and degree of damage are different in the two regions [8]. We hypothesize that *Panax ginseng* may play dual roles by boosting regenerative potential in the peri-injured region and diminishing toxicity in the core-injured region.

3.1.1. KRG in the penumbra region of acute neurovascular diseases

In peri-injured mouse TBI brain tissues, Nrf2 and HO-1 expression is upregulated by KRG [32]. HO-1 induction by KRG is much higher in the peri-injured region than in physiological conditions

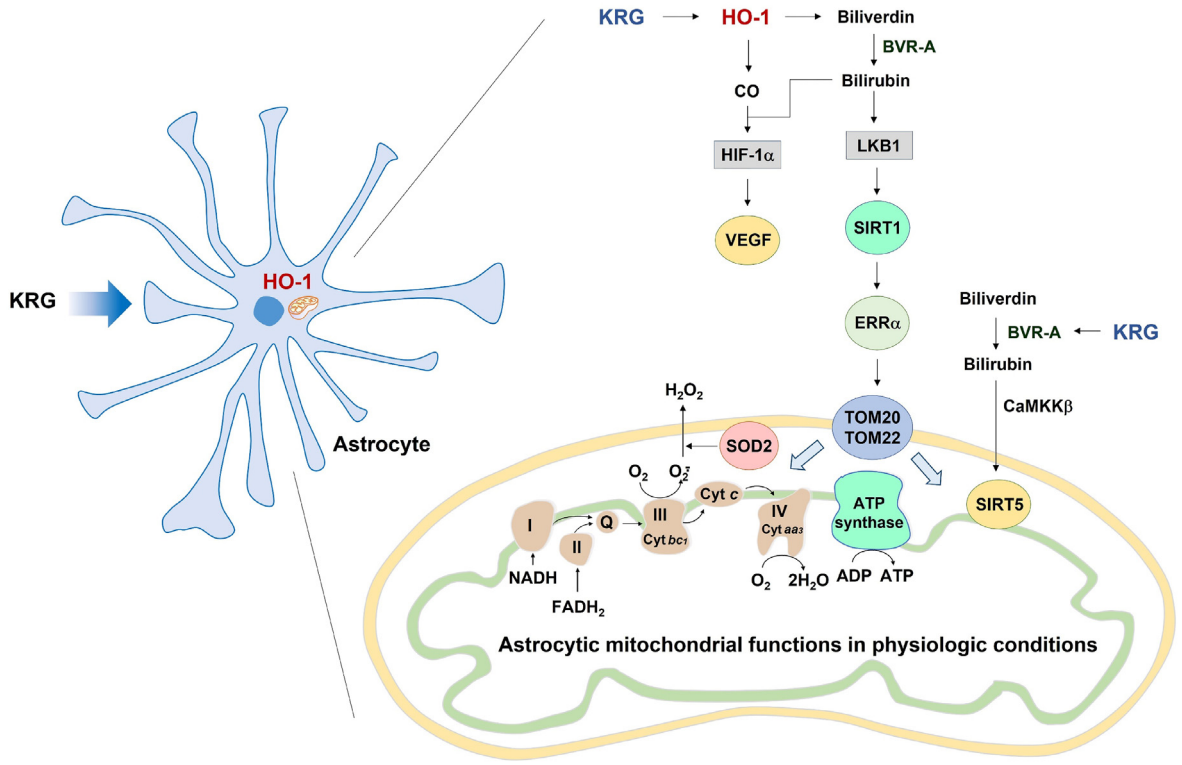

Fig. 1. KRG induces astrocytic mitochondrial functions through HO metabolites in physiologic conditions. *Abbreviations*. BVR-A, biliverdin reductase A; CaMKKβ, Ca^{2+}/calmodulin-dependent protein kinase kinase β; CO, carbon monoxide; Cyt, cytochrome; ERRα, estrogen-related receptor α; HIF-1α, hypoxia-inducible factor-1α; HO-1, heme oxygenase-1; I, oxidative phosphorylation (OXPHOS) complex I; KRG, Korean Red Ginseng; LKB1, liver kinase B1; SIRT1, sirtuin 1; SOD2, superoxide dismutase 2; TOM20, translocase of outer membrane 20; VEGF, vascular endothelial growth factor.

[32]. In TBI, after KRG administration for 3 days, astrocytes express HO-1 in the penumbra region, and the injection of Sn(IV) protoporphyrin IX dichloride (SnPP), an HO inhibitor, reduces KRG-mediated HO-1 expression [31]. Nicotinamide phosphoribosyltransferase (NAMPT) may be involved in many important biological processes, including metabolism, stress response, and aging [33]. Astrocytes in oxygen-glucose deprivation/recovery (OGD/R) in the presence of KRG upregulate Nrf2, NAMPT, and HO-1 expression [31]. Interestingly, KRG-induced NAMPT upregulates HO-1, and *vice versa* [31]. KRG-induced Nrf2 modulates HO-1 expression [32]. Taken together, in acute neurovascular injury, HO-1 expression can be induced in astrocytes by KRG in the penumbra region through NAMPT and Nrf2.

In astrocytes, KRG-induced NAMPT increases the NAD^+/NADH ratio, leading to increased levels of SIRT1, SIRT2, and SIRT3 [32]. Since knockdown of SIRT1, SIRT2, or SIRT3 does not reduce KRG-mediated HO-1 expression, two scenarios are possible: 1) it may be an upstream factor for HO-1 or 2) it is a downstream factor for HO-1 influencing increased NAD^+/NADH ratio-mediated increases in SIRT1, SIRT2, and SIRT3 levels. SIRTs (i.e., SIRT1, SIRT2 or SIRT3) play key roles in TOM20 expression in the outer membrane of mitochondria, consequently upregulating mitochondrial membrane potential, ATP production, and O_2 consumption [32]. In other pathways, KRG induces the HO-1-AMPKα axis, leading to PGC-1α and ERRα activation. AMPKα-PGC-1α-ERRα circuit upregulates OGD/R-mediated cytochrome c, OXPHOS, and cytochrome c oxidase subunit 2 (MTCO2) in *in vitro* astrocytes [31].

The *in vivo* TBI murine model administered KRG shows upregulation of HO-1, AMPKα phosphorylation, NAMPT, SIRT1, SIRT3, ERRα, PGC-1α, cytochrome c, MOTCO2, TOM20 and TOM22 [31,32]. These effects were blocked by injecting SnPP followed by KRG administration in a TBI model [31,32]. SnPP treatment diminishes the co-expression of HO-1 and glial fibrillary acidic protein (GFAP) in peri-injured TBI brain tissues [31], demonstrating astrocytic HO-1 inactivation in the penumbra region of TBI. SnPP-induced HO inactivation also results in reduced co-expression of GFAP and TOM20 [32]. Thus, astrocytic HO-1 plays a key role in mitochondrial function (Fig. 2).

The relationship between Ca^{2+} channels and *Panax ginseng* has previously been reported in neurons. During ischemia-related injury, KRG and ginsenoside (i.e., Rg1 and Rg3) protect neurons from excessive Ca^{2+} influx by inhibiting Ca^{2+} channels [34,35]. In contrast, during radiofrequency exposure, the levels of Ca^{2+}-binding proteins such as calbindin D28-k, parvalbumin, and calretinin are reduced in hippocampal neurons, which are recovered by KRG [36]. Taken together, ginseng and ginsenosides may play key roles in neuroprotection by regulating Ca^{2+} channels and Ca^{2+}-related proteins. The role of astrocytic Ca^{2+} via KRG in pathophysiological conditions has not yet been well established. Astrocytic Ca^{2+} is involved in various functions such as mitochondria biogenesis, neurotransmitter release via astrocyte-neuron communication, and vascular tone [7,25,37]. Among HO metabolites, CO and bilirubin stimulate Ca^{2+} influx through L-type Ca^{2+} channels in astrocytes [25]. Pretreatment with CO followed by recovery (CO/R) positively upregulates HO-1, which is blocked by Ca^{2+} chelators [25]. As KRG induces HO-1 expression in astrocytes, KRG-mediated Ca^{2+} signaling during pathophysiological conditions may need to be investigated to determine the molecular mechanisms of HO-1-induced mitochondrial functions.

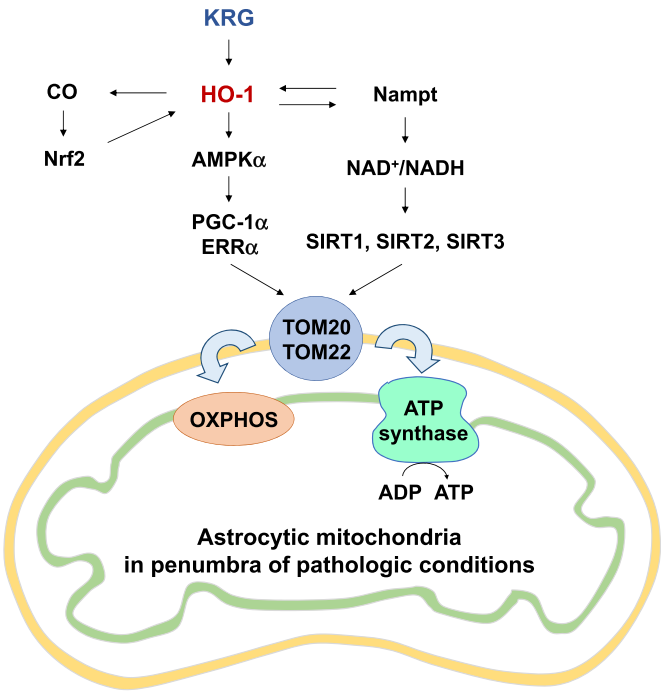

Fig. 2. KRG enhances astrocytic mitochondrial functions through HO-1-mediated signaling in the penumbra in pathological conditions. *Abbreviations.* AMPKα, AMP-activated protein kinase; CO, carbon monoxide; ERRα, estrogen-related receptor α; HO-1, heme oxygenase-1; KRG, Korean Red Ginseng; LKB1, liver kinase B1; NAMPT, nicotinamide phosphoribosyltransferase; Nrf2, nuclear factor erythroid-2-related factor 2; OXPHOS, oxidative phosphorylation; PGC-1α, peroxisome proliferator-activated receptor γ coactivator α; SIRT, sirtuin; TOM22, translocase of outer membrane 22.

3.1.2. KRG in the core region of acute neurovascular diseases

Excessive concentrations of HO metabolites (i.e., excessive intracellular labile iron) can be produced, enhancing the probability of ferroptosis [8,38–40]. Ferroptosis is a form of iron-dependent lipid peroxidation and causes brain cell death in the CNS that underlines the pathology of several neurodegenerative diseases [41]. Ferroptosis was detected in the core region of acute injury and widespread region in chronic neurodegenerative diseases [8,42,43]. A critical morphological feature of ferroptosis is mitochondrial dysfunction, characterized by smaller mitochondrial size and disruption of the mitochondrial outer membrane [38].

The neuroprotective effects of ginseng and ginsenosides in acute neuroinflammatory diseases have been reported [1]. Cerulein-mediated cell toxicity (pancreatitis) was ameliorated by the ginsenoside Rg3 [44]. Mice treated with Rg3 show suppressed cerulein-induced oxidative stress via the Nrf2-HO-1-GPX4 pathway [44], possibly inhibiting ferroptosis. GSH is converted into GSSG by GPX4 and superoxide is converted into hydrogen peroxide by SOD2 [45]. Since Nrf2 activation induces the expression of target genes, including *GPX4* and *SOD2* [45,46], KRG and ginsenosides may play important roles in the suppression of ferroptosis by Nrf2-dependent induction of antioxidant enzymes.

Blood–neural barrier disruption was observed in the core regions of acute neurovascular diseases [47]. Ischemia/reperfusion injury-mediated release of pro-inflammatory cytokines and other soluble mediators (e.g., excessive VEGF) triggers paracellular permeability and tight junction disruption [48–50]. Tight junctions are disrupted during neuroinflammatory diseases, which results in the infiltration of monocytes into the brain parenchyma, where they become activated macrophages [50,51]. Endothelial damage, pericyte apoptosis, reactive glial activation (gliosis), and inflammatory cytokines exacerbate CNS neurodegeneration by uncoupling normal cell–cell communication [47]. Pericyte loss can result in the depletion of neural stem cells in the core region in neurovascular diseases [47]. The HO metabolite CO can protect the blood–neural barrier, subsequently leading to intact surrounding tissues [47,52]. Ginseng-mediated preservation of the blood–neural barrier structure and function during neuroinflammatory disease is partly due to its anti-inflammatory effects, thereby preventing detrimental signaling cascades [1].

3.2. KRG in chronic neurodegenerative diseases

In a case report, oxidative stress resulted in increased endothelial cell injury in a 6-year-old boy with HO-1 deficiency [53]. Malfunctional endothelial cells trigger hypoperfusion (chronic hypoxia) and reduce the drainage of aggregated proteins and molecules [6]. HO-1 has iron-reutilizing functions [40]. HO-1 expression results in the rapid expression of ATPase pumps [54]. In addition, mild HO-1 upregulation and activity increases the levels of free iron and the subsequent expression of ferritin [9,55,56], which actively removes intracellular iron, thereby exerting cytoprotective effects. In contrast, ferroptosis induction by excessive HO-1 overexpression may be associated with the reduced free iron-binding ability of ferritin induced by oxidative and metabolic stress [57,58]. Sustained overexpression of HO-1 in astrocytes leads to abnormal iron deposition and mitochondrial dysfunction in the brain, resulting in neuronal loss and declined cognitive ability [8,9,59,60]. Brain iron accumulation may be associated with cognitive decline in patients with Alzheimer's disease [61] (Fig. 3).

The protective role of ginseng in chronic neurodegenerative diseases has been previously reported [62,63]. Proper levels of HO-1 induced by Nrf2 protect cells from iron-dependent toxicity [9,64], partly through the Nrf2-GPX4 pathway. If ginseng provides an Nrf2-mediated antioxidant milieu during chronic neurodegenerative diseases, it may diminish sustained HO-1/free iron-mediated neurovascular damage. Balanced interactions between the nucleus and mitochondria may help to slow down the processes of age-related neurodegenerative diseases [65]. Nrf2-mediated transcription of genes can maintain the mitochondrial function by upregulating proteins associated with OXPHOS, the TOM complex, and ATP production through HO-1 [32].

In addition, the cells exhibit heterogeneous characteristics. There are various types of astrocytes, such as unstimulated, reactive, and autophagy-dysregulated types [7,66]. These various types of astrocytes exhibit alterations in transcription, cell hypertrophy, and morphology [67,68]. The repair effects of ginseng can be expected because ginseng boosts the antioxidant and regenerative capacity of astrocytes through Nrf2, NAMPT, and HO-1 [31,32]. In the neurovascular unit, there are multiple cells and heterogeneity of the single-cell type. To determine the exact repair mechanisms, further studies are necessary, especially on the HO-1 pathway in various cell types (i.e., pericytes, endothelial cells, neurons, oligodendrocytes, and microglia) of the neurovascular unit.

3.3. Cell–cell interactions via KRG

In the CNS, cell–cell interactions amplify signals [47,68,69]. Repair systems can be facilitated by healthy cellular interactions [70]. Repaired neurovascular systems may further recover cognitive function by repairing the neural circuits. In this step, the brain cells may require an antioxidant and energy-enriched milieu.

In pathophysiologic conditions, KRG-treated astrocytes exhibit upregulated mitochondrial mass and energy production [13,31,32]. Treatment of neural stem cells with conditioned media from KRG-treated astrocytes promotes O_2 consumption and TOM20

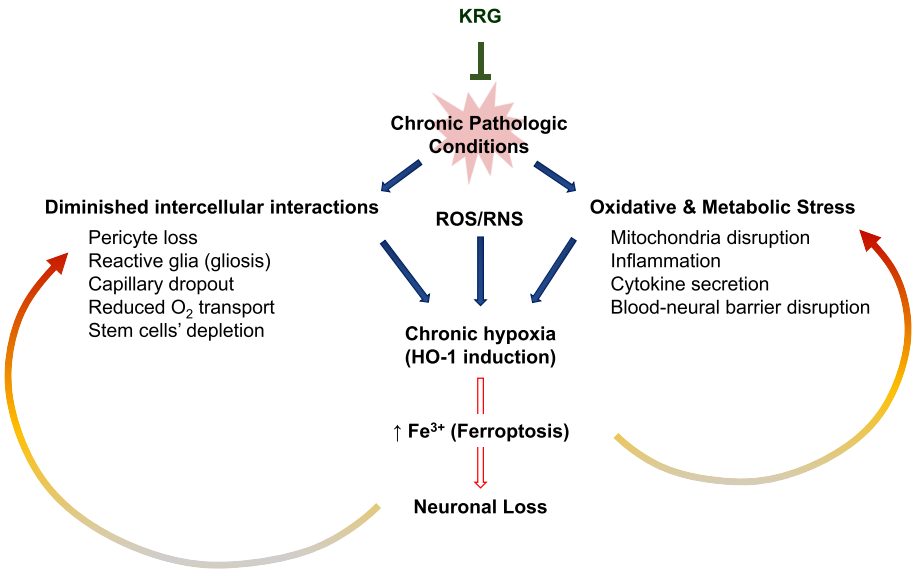

Fig. 3. KRG diminishes the processes of chronic pathological conditions by promoting intercellular interactions and reducing oxidative and metabolic stress. *Abbreviations*. KRG, Korean Red Ginseng; ROS, reactive oxygen species; RNS, reactive nitrogen species.

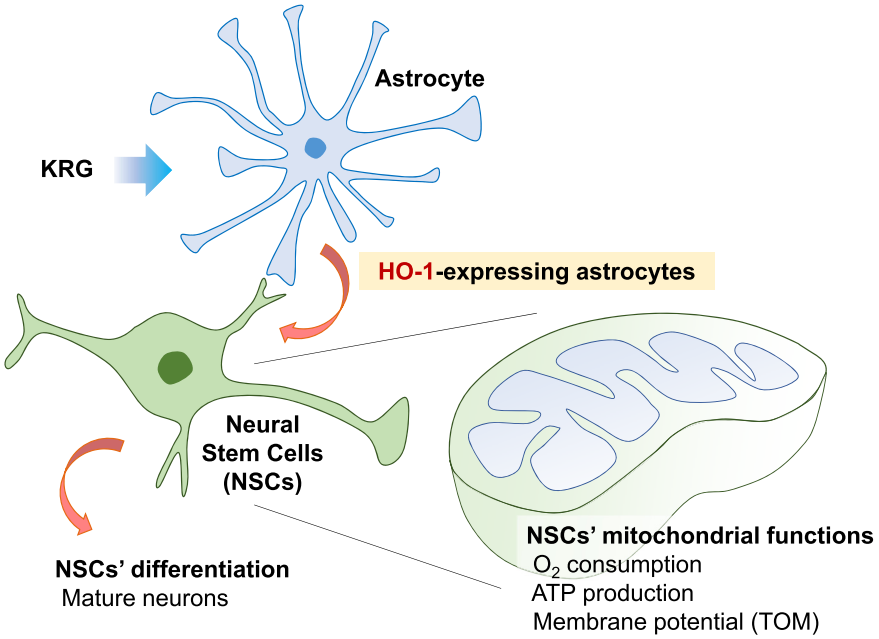

Fig. 4. KRG boosts astrocyte-to-neural stem cell communication through astrocytic HO-1. CO, carbon monoxide; KRG, Korean Red Ginseng.

expression, possibly through astrocytic HO-1 [32]. Moreover, energy-enriched astrocytes may affect neural stem cell differentiation into mature neurons via astrocytic HO-1 [13,32] (Fig. 4). Damaged astrocytes via chronic overexpression of HO-1 may be detrimental to stem cell capacity [8,9,71].

To repair the neurovascular unit, new blood vessels are required for the supply of O_2 and nutrients. KRG induces angiogenesis by upregulating VEGF and eNOS, which may be related to glucocorticoid receptors [15]. KRG induces eNOS expression through the HO-1-mediated nuclear factor kappa-light-chain-enhancer of activated B cells-miRNA-155-5p axis in endothelial cells [14]. Angiogenesis via astrocyte–endothelial cell network may be facilitated by KRG because KRG induces VEGF through HIF-1α and ERRα [13,23]. Since VEGF is involved in neurogenesis, angiogenesis, and cell survival [72], KRG-induced astrocytic VEGF may regenerate the neurovascular unit.

4. Conclusion

Using ginseng, clinical trials in human study has been shown in pathophysiological conditions [1]. However, the molecular mechanisms by which ginseng contributes to the anti-aging and regeneration through mitochondrial functions have not been well revealed. In this review, we suggest that the prophylactic effects of KRG are important for preventing aging by improving mitochondrial functions. Among the preventive effects of KRG after an injury,

mitochondrial functions enhanced by KRG play key roles in anti-aging, anti-inflammatory, and regenerative processes. Here, we specifically discussed the KRG-mediated transient HO-1 expression as a beneficial effect on mitochondrial functions in physical and pathological conditions. Uncovering HO-1-related molecular mechanisms by which KRG stimulates regeneration by boosting the energetic potential in glial cells is important for establishing therapeutic strategies.

Funding

This paper was supported by Konkuk University Premier Research Fund in 2021.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

[1] Kim M, Mok H, Yeo WS, Ahn JH, Choi YK. Role of ginseng in the neurovascular unit of neuroinflammatory diseases focused on the blood-brain barrier. J Ginseng Res 2021;45(5):599–609.
[2] Prabhakar NR, Semenza GL. Oxygen sensing and homeostasis. Physiology (Bethesda) 2015;30(5):340–8.
[3] Imuta N, Hori O, Kitao Y, Tabata Y, Yoshimoto T, Matsuyama T, Ogawa S. Hypoxia-mediated induction of heme oxygenase type I and carbon monoxide release from astrocytes protects nearby cerebral neurons from hypoxia-mediated apoptosis. Antioxid Redox Signal 2007;9(5):543–52.
[4] Dore S, Takahashi M, Ferris CD, Zakhary R, Hester LD, Guastella D, Snyder SH. Bilirubin, formed by activation of heme oxygenase-2, protects neurons against oxidative stress injury. Proc Natl Acad Sci U S A 1999;96(5):2445–50.
[5] Barisano G, Montagne A, Kisler K, Schneider JA, Wardlaw JM, Zlokovic BV. Blood-brain barrier link to human cognitive impairment and Alzheimer's Disease. Nat Cardiovasc Res 2022;1(2):108–15.
[6] Feng T, Yamashita T, Zhai Y, Shang J, Nakano Y, Morihara R, Fukui Y, Hishikawa N, Ohta Y, Abe K. Chronic cerebral hypoperfusion accelerates Alzheimer's disease pathology with the change of mitochondrial fission and fusion proteins expression in a novel mouse model. Brain Res 2018;1696:63–70.
[7] Kim Y, Park J, Choi YK. The role of astrocytes in the central nervous system focused on BK channel and heme oxygenase metabolites: a review. Antioxidants (Basel) 2019;8(5):121.
[8] Choi YK, Kim YM. Beneficial and detrimental roles of heme oxygenase-1 in the neurovascular system. Int J Mol Sci 2022;23(13):7041.
[9] Schipper HM, Song W, Tavitian A, Cressatti M. The sinister face of heme oxygenase-1 in brain aging and disease. Prog Neurobiol 2019;172:40–70.
[10] Choi YK, Kim YM. Regulation of endothelial and vascular functions by carbon monoxide via crosstalk with nitric oxide. Front Cardiovasc Med 2021;8:649630.
[11] Choi YK, Kim CK, Lee H, Jeoung D, Ha KS, Kwon YG, Kim KW, Kim YM. Carbon monoxide promotes VEGF expression by increasing HIF-1alpha protein level via two distinct mechanisms, translational activation and stabilization of HIF-1alpha protein. J Biol Chem 2010;285(42):32116–25.
[12] Yang H, Lee SE, Jeong SI, Park CS, Jin YH, Park YS. Up-regulation of heme oxygenase-1 by Korean red ginseng water extract as a cytoprotective effect in human endothelial cells. J Ginseng Res 2011;35(3):352–9.
[13] Park J, Lee M, Kim M, Moon S, Kim S, Kim S, Koh SH, Kim YM, Choi YK. Prophylactic role of Korean Red Ginseng in astrocytic mitochondrial biogenesis through HIF-1alpha. J Ginseng Res 2022;46(3):408–17.
[14] Kim TH, Kim JY, Bae J, Kim YM, Won MH, Ha KS, Kwon YG, Kim YM. Korean Red ginseng prevents endothelial senescence by downregulating the HO-1/NF-kappaB/miRNA-155-5p/eNOS pathway. J Ginseng Res 2021;45(2):344–53.
[15] Sung WN, Kwok HH, Rhee MH, Yue PY, Wong RN. Korean Red Ginseng extract induces angiogenesis through activation of glucocorticoid receptor. J Ginseng Res 2017;41(4):477–86.
[16] Jovanovski E, Peeva V, Sievenpiper JL, Jenkins AL, Desouza L, Rahelic D, Sung MK, Vuksan V. Modulation of endothelial function by Korean red ginseng (Panax ginseng C.A. Meyer) and its components in healthy individuals: a randomized controlled trial. Cardiovasc Ther 2014;32(4):163–9.
[17] Choi K, Yoon J, Lim HK, Ryoo S. Korean red ginseng water extract restores impaired endothelial function by inhibiting arginase activity in aged mice. Korean J Phys Pharm 2014;18(2):95–101.
[18] Bellezza I, Giambanco I, Minelli A, Donato R. Nrf2-Keap1 signaling in oxidative and reductive stress. Biochim Biophys Acta Mol Cell Res 2018;1865(5):721–33.
[19] Loboda A, Damulewicz M, Pyza E, Jozkowicz A, Dulak J. Role of Nrf2/HO-1 system in development, oxidative stress response and diseases: an evolutionarily conserved mechanism. Cell Mol Life Sci 2016;73(17):3221–47.
[20] Dodson M, Castro-Portuguez R, Zhang DD. NRF2 plays a critical role in mitigating lipid peroxidation and ferroptosis. Redox Biol 2019;23:101107.
[21] Wang B, Cao W, Biswal S, Dore S. Carbon monoxide-activated Nrf2 pathway leads to protection against permanent focal cerebral ischemia. Stroke; J Cereb Circ 2011;42(9):2605–10.
[22] Tudor C, Lerner-Marmarosh N, Engelborghs Y, Gibbs PE, Maines MD. Biliverdin reductase is a transporter of haem into the nucleus and is essential for regulation of HO-1 gene expression by haematin. Biochem J 2008;413(3):405–16.
[23] Moon S, Kim CH, Park J, Kim M, Jeon HS, Kim YM, Choi YK. Induction of BVR-A expression by Korean red ginseng in murine hippocampal astrocytes: role of bilirubin in mitochondrial function via the LKB1-SIRT1-ERRalpha Axis. Antioxidants (Basel) 2022;11(9):1742.
[24] Kim S, Lee M, Choi YK. The role of a neurovascular signaling pathway involving hypoxia-inducible factor and notch in the function of the central nervous system. Biomol Ther (Seoul) 2020;28(1):45–57.
[25] Choi YK, Kim JH, Lee DK, Lee KS, Won MH, Jeoung D, Lee H, Ha KS, Kwon YG, Kim YM. Carbon monoxide potentiation of L-type Ca2+ channel activity increases HIF-1alpha-Independent VEGF expression via an AMPKalpha/SIRT1-mediated PGC-1alpha/ERRalpha Axis. Antioxid Redox Signal 2017;27(1):21–36.
[26] Choi YK, Park JH, Yun JA, Cha JH, Kim Y, Won MH, Kim KW, Ha KS, Kwon YG, Kim YM. Heme oxygenase metabolites improve astrocytic mitochondrial function via a Ca2+-dependent HIF-1alpha/ERRalpha circuit. PLoS One 2018;13(8):e0202039.
[27] Arany Z, Foo SY, Ma Y, Ruas JL, Bommi-Reddy A, Girnun G, Cooper M, Laznik D, Chinsomboon J, Rangwala SM, et al. HIF-independent regulation of VEGF and angiogenesis by the transcriptional coactivator PGC-1alpha. Nature 2008;451(7181):1008–12.
[28] Choi YK, Park JH, Baek YY, Won MH, Jeoung D, Lee H, Ha KS, Kwon YG, Kim YM. Carbon monoxide stimulates astrocytic mitochondrial biogenesis via L-type Ca2+ channel-mediated PGC-1alpha/ERRalpha activation. Biochem Biophys Res Commun 2016;479(2):297–304.
[29] Gurd BJ. Deacetylation of PGC-1alpha by SIRT1: importance for skeletal muscle function and exercise-induced mitochondrial biogenesis. Appl Physiol Nutr Metab 2011;36(5):589–97.
[30] Canto C, Auwerx J. PGC-1alpha, SIRT1 and AMPK, an energy sensing network that controls energy expenditure. Curr Opin Lipidol 2009;20(2):98–105.
[31] Kim M, Kim J, Moon S, Choi BY, Kim S, Jeon HS, Suh SW, Kim YM, Choi YK. Korean red ginseng improves astrocytic mitochondrial function by upregulating HO-1-Mediated AMPKalpha-PGC-1alpha-ERRalpha circuit after traumatic brain injury. Int J Mol Sci 2021;22(23):13081.
[32] Kim M, Moon S, Jeon HS, Kim S, Koh SH, Chang MS, Kim YM, Choi YK. Dual effects of Korean red ginseng on astrocytes and neural stem cells in traumatic brain injury: the HO-1-Tom20 Axis as a putative target for mitochondrial function. Cells 2022;11(5):892.
[33] Ma C, Pi C, Yang Y, Lin L, Shi Y, Li Y, Li Y, He X. Nampt expression decreases age-related senescence in rat bone marrow mesenchymal stem cells by targeting Sirt1. PLoS One 2017;12(1):e0170930.
[34] Lee JH, Jeong SM, Kim JH, Lee BH, Yoon IS, Lee JH, Choi SH, Lee SM, Park YS, Lee JH, et al. Effects of ginsenosides and their metabolites on voltage-dependent Ca(2+) channel subtypes. Mol Cells 2006;21(1):52–62.
[35] Zhang YF, Fan XJ, Li X, Peng LL, Wang GH, Ke KF, Jiang ZL. Ginsenoside Rg1 protects neurons from hypoxic-ischemic injury possibly by inhibiting Ca2+ influx through NMDA receptors and L-type voltage-dependent Ca2+ channels. Eur J Pharm 2008;586(1–3):90–9.
[36] Maskey D, Lee JK, Kim HR, Kim HG. Neuroprotective effect of ginseng against alteration of calcium binding proteins immunoreactivity in the mice hippocampus after radiofrequency exposure. Biomed Res Int 2013;2013:812641.
[37] Girouard H, Bonev AD, Hannah RM, Meredith A, Aldrich RW, Nelson MT. Astrocytic endfoot Ca2+ and BK channels determine both arteriolar dilation and constriction. Proc Natl Acad Sci U S A 2010;107(8):3811–6.
[38] Xie Y, Hou W, Song X, Yu Y, Huang J, Sun X, Kang R, Tang D. Ferroptosis: process and function. Cell Death Differ 2016;23(3):369–79.
[39] Bao WD, Pang P, Zhou XT, Hu F, Xiong W, Chen K, Wang J, Wang F, Xie D, Hu YZ, et al. Loss of ferroportin induces memory impairment by promoting ferroptosis in Alzheimer's disease. Cell Death Differ 2021;28(5):1548–62.
[40] Poss KD, Tonegawa S. Heme oxygenase 1 is required for mammalian iron reutilization. Proc Natl Acad Sci U S A 1997;94(20):10919–24.
[41] Gutteridge JM. Iron promoters of the Fenton reaction and lipid peroxidation can be released from haemoglobin by peroxides. FEBS Lett 1986;201(2):291–5.
[42] Daglas M, Adlard PA. The involvement of iron in traumatic brain injury and neurodegenerative disease. Front Neurosci 2018;12:981.
[43] Liu JL, Fan YG, Yang ZS, Wang ZY, Guo C. Iron and alzheimer's disease: from pathogenesis to therapeutic implications. Front Neurosci 2018;12:632.
[44] Shan Y, Li J, Zhu A, Kong W, Ying R, Zhu W. Ginsenoside Rg3 ameliorates acute pancreatitis by activating the NRF2/HO1mediated ferroptosis pathway. Int J Mol Med 2022;50(1):89.
[45] Chen QM. Nrf2 for cardiac protection: pharmacological options against oxidative stress. Trends Pharmacol Sci 2021;42(9):729–44.

[46] Ma CS, Lv QM, Zhang KR, Tang YB, Zhang YF, Shen Y, Lei HM, Zhu L. NRF2-GPX4/SOD2 axis imparts resistance to EGFR-tyrosine kinase inhibitors in non-small-cell lung cancer cells. Acta Pharmacol Sin 2021;42(4):613–23.
[47] Choi YK, Maki T, Mandeville ET, Koh SH, Hayakawa K, Arai K, Kim YM, Whalen MJ, Xing C, Wang X, et al. Dual effects of carbon monoxide on pericytes and neurogenesis in traumatic brain injury. Nat Med 2016;22(11):1335–41.
[48] Bates DO, Harper SJ. Regulation of vascular permeability by vascular endothelial growth factors. Vascul Pharmacol 2002;39(4–5):225–37.
[49] Sandoval KE, Witt KA. Blood-brain barrier tight junction permeability and ischemic stroke. Neurobiol Dis 2008;32(2):200–19.
[50] Knowland D, Arac A, Sekiguchi KJ, Hsu M, Lutz SE, Perrino J, Steinberg GK, Barres BA, Nimmerjahn A, Agalliu D. Stepwise recruitment of transcellular and paracellular pathways underlies blood-brain barrier breakdown in stroke. Neuron 2014;82(3):603–17.
[51] Desai BS, Monahan AJ, Carvey PM, Hendey B. Blood-brain barrier pathology in Alzheimer's and Parkinson's disease: implications for drug therapy. Cell Transplant 2007;16(3):285–99.
[52] Wang J, Zhang D, Fu X, Yu L, Lu Z, Gao Y, Liu X, Man J, Li S, Li N, et al. Carbon monoxide-releasing molecule-3 protects against ischemic stroke by suppressing neuroinflammation and alleviating blood-brain barrier disruption. J Neuroinflammation 2018;15(1):188.
[53] Yachie A, Niida Y, Wada T, Igarashi N, Kaneda H, Toma T, Ohta K, Kasahara Y, Koizumi S. Oxidative stress causes enhanced endothelial cell injury in human heme oxygenase-1 deficiency. J Clin Invest 1999;103(1):129–35.
[54] Baranano DE, Wolosker H, Bae BI, Barrow RK, Snyder SH, Ferris CD. A mammalian iron ATPase induced by iron. J Biol Chem 2000;275(20):15166–73.
[55] Yu X, Song N, Guo X, Jiang H, Zhang H, Xie J. Differences in vulnerability of neurons and astrocytes to heme oxygenase-1 modulation: implications for mitochondrial ferritin. Sci Rep 2016;6:24200.
[56] Ferris CD, Jaffrey SR, Sawa A, Takahashi M, Brady SD, Barrow RK, Tysoe SA, Wolosker H, Baranano DE, Dore S, et al. Haem oxygenase-1 prevents cell death by regulating cellular iron. Nat Cell Biol 1999;1(3):152–7.
[57] Kwon MY, Park E, Lee SJ, Chung SW. Heme oxygenase-1 accelerates erastin-induced ferroptotic cell death. Oncotarget 2015;6(27):24393–403.
[58] Chiang SK, Chen SE, Chang LC. A dual role of heme oxygenase-1 in cancer cells. Int J Mol Sci 2018;20(1):39.
[59] Schipper HM. Glial HO-1 expression, iron deposition and oxidative stress in neurodegenerative diseases. Neurotox Res 1999;1(1):57–70.
[60] Schipper HM, Song W, Tavitian A, Cressatti M. The sinister face of heme oxygenase-1 in brain aging and disease. Prog Neurobiol 2019;172:40–70.
[61] Ayton S, Wang Y, Diouf I, Schneider JA, Brockman J, Morris MC, Bush AI. Brain iron is associated with accelerated cognitive decline in people with Alzheimer pathology. Mol Psychiatr 2020;25(11):2932–41.
[62] Kim KH, Lee D, Lee HL, Kim CE, Jung K, Kang KS. Beneficial effects of Panax ginseng for the treatment and prevention of neurodegenerative diseases: past findings and future directions. J Ginseng Res 2018;42(3):239–47.
[63] Razgonova MP, Veselov VV, Zakharenko AM, Golokhvast KS, Nosyrev AE, Cravotto G, Tsatsakis A, Spandidos DA. Panax ginseng components and the pathogenesis of Alzheimer's disease (Review). Mol Med Rep 2019;19(4):2975–98.
[64] Nitti M, Piras S, Brondolo L, Marinari UM, Pronzato MA, Furfaro AL. Heme oxygenase 1 in the nervous system: does it favor neuronal cell survival or induce neurodegeneration? Int J Mol Sci 2018;19(8):2260.
[65] Campello L, Singh N, Advani J, Mondal AK, Corso-Diaz X, Swaroop A. Aging of the retina: molecular and metabolic turbulences and potential interventions. Annu Rev Vis Sci 2021;7:633–64.
[66] Anderson MA, Ao Y, Sofroniew MV. Heterogeneity of reactive astrocytes. Neurosci Lett 2014;565:23–9.
[67] Zamanian JL, Xu L, Foo LC, Nouri N, Zhou L, Giffard RG, Barres BA. Genomic analysis of reactive astrogliosis. J Neurosci 2012;32(18):6391–410.
[68] Liddelow SA, Guttenplan KA, Clarke LE, Bennett FC, Bohlen CJ, Schirmer L, Bennett ML, Munch AE, Chung WS, Peterson TC, et al. Neurotoxic reactive astrocytes are induced by activated microglia. Nature 2017;541(7638):481–7.
[69] Eroglu C, Barres BA. Regulation of synaptic connectivity by glia. Nature 2010;468(7321):223–31.
[70] Lee H, Choi YK. Regenerative effects of heme oxygenase metabolites on neuroinflammatory diseases. Int J Mol Sci 2018;20(1):78.
[71] Chun H, Lee CJ. Reactive astrocytes in Alzheimer's disease: a double-edged sword. Neurosci Res 2018;126:44–52.
[72] Moon S, Chang MS, Koh SH, Choi YK. Repair mechanisms of the neurovascular unit after ischemic stroke with a focus on VEGF. Int J Mol Sci 2021;22(16):8543.